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Industrial-Scale Manufacture of Oleosin 30G for Use as Contrast Agent in Echocardiography

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Abstract
In ultrasound sonography, microbubbles are used as contrasting agents to improve the effectiveness of ultrasound imaging. Monodisperse microbubbles are required to achieve the optimal image quality. In order to achieve a uniform size distribution, microbubbles are stabilized with surfactant molecules. One such molecule is Oleosin, an amphiphilic structural protein found in vascular plant oil bodies that contains one hydrophobic and two hydrophilic sections. Controlling the functionalization of microbubbles is a comprehensive and versatile process using recombinant technology to produce a genetically engineered form of Oleosin called Oleosin 30G. With the control of a microfluidic device, uniformly-sized and resonant microbubbles can be readily produced and stored in stable conditions up to one month. Currently, Oleosin microbubbles are limited to the lab-scale; however, through development of an integrated batch bioprocessing model, the overall product yield of Oleosin 30G can be increased to 7.39 kg/year to meet needs on the industrial-scale. An Oleosin-stabilized microbubble suspension as a contrast agent is in a strong position to take a competitive share of the current market, capitalizing on needs unmet by current market leader, Definity®. Based on market dynamics and process logistics, scaled-up production of Oleosin 30G for use as a contrast agent is expected to be both a useful and profitable venture.

Disciplines
Biochemical and Biomolecular Engineering | Chemical Engineering | Engineering

Industrial-Scale Manufacture of Oleosin 30G For Use as Contrast Agent in Echocardiography

April 12, 2016

Steve Casey, Hailey Edelstein, Rebecca Michelson, Nikita Rao
Department of Chemical and Biomolecular Engineering, University of Pennsylvania

April 12, 2016
Professor Leonard Fabiano
University of Pennsylvania
School of Engineering and Applied Science Department of Chemical and Biomolecular Engineering 220 S. 33rd Street
Philadelphia, PA 19104

Dear Professor Fabiano,

Enclosed is a carefully considered process design to manufacture Oleosin 30G on an industrial scale. Oleosin 30G is a genetically engineered recombinant protein that functions as a surfactant to stabilize uniformly-sized, monodisperse microbubbles for use as a contrast agent in ultrasound. This process focuses specifically on echocardiograms and assumes successful FDA approval to allow for a full product launch on the market.

The process utilizes *E. coli* cells grown up in a bioreactor in the presence of LB-Kanamycin medium with glucose as the carbon source for cell growth. Upstream cell culture is then transferred to the separations phase of processing. First, centrifugation is used, followed by high pressure homogenization to lyse cells and release expressed protein. Next, a cobalt affinity chromatography column is used for further protein purification. Final downstream purification is then used, where Oleosin 30G in solution is sent through an ultra/diafiltration (UFDF) membrane. Lastly, the product is fully purified via endotoxin removal using a bulk microfiltration membrane. The final step in the process is packaging the microbubble suspension, using eight microfluidic device molds to produce uniformly sized microbubbles with Oleosin 30G at a concentration of 1 mg/mL. Glass vials, each holding 10 mL of solution containing single doses of Oleosin 30G for IV injection, are stable up to one month when maintained at 4 ºC, and will be shipped to order using pharmaceutical-grade shipping within one week of production.

Initial profitability analysis of the process shows favorable results with an overall annual production goal at 7.39 kg of Oleosin 30G in microbubble suspensions, representing a 100% market saturation of current echocardiograms with use of a contrast agent. With an internal rate of return (IRR) of 72.43%, a net present value (NPV) of $201,670,700 and a return on investment (ROI) of 72.34%, the process assumes a 15-year facility life span and requires a $41.5 million capital investment.

All calculations performed use either primary laboratory data, data acquired from Dr. Hammer's lab at the University of Pennsylvania, or references from literature. Please feel free to contact us with any questions you may have concerning our process below.

Best,

Steve Casey Hailey Edelstein Rebecca Michelson Nikita Rao

Industrial-Scale Manufacture of Oleosin 30G for Use as Contrast Agent in Ultrasound

Department of Chemical and Biomolecular Engineering,
University of Pennsylvania

April 2016

Steve Casey, Hailey Edelstein, Rebecca Michelson, Nikita Rao

Project Advisor: Dr. Miriam Wattenbarger

Project Recommended by: Dr. Daniel Hammer and Dr. Miriam Wattenbarger

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1. Abstract

In ultrasound sonography, microbubbles are used as contrasting agents to improve the effectiveness of ultrasound imaging. Monodisperse microbubbles are required to achieve the optimal image quality. In order to achieve a uniform size distribution, microbubbles are stabilized with surfactant molecules. One such molecule is Oleosin, an amphiphilic structural protein found in vascular plant oil bodies that contains one hydrophobic and two hydrophilic sections. Controlling the functionalization of microbubbles is a comprehensive and versatile process using recombinant technology to produce a genetically engineered form of Oleosin called Oleosin 30G. With the control of a microfluidic device, uniformly-sized and resonant microbubbles can be readily produced and stored in stable conditions up to one month. Currently, Oleosin microbubbles are limited to the lab-scale; however, through development of an integrated batch bioprocessing model, the overall product yield of Oleosin 30G can be increased to 7.39 kg/year to meet needs on the industrial-scale. An Oleosin-stabilized microbubble suspension as a contrast agent is in a strong position to take a competitive share of the current market, capitalizing on needs unmet by current market leader, Definity®. Based on market dynamics and process logistics, scaled-up production of Oleosin 30G for use as a contrast agent is expected to be both a useful and profitable venture.

2. Introduction

2.1. Project Background

The widespread nature of ultrasound imaging as a diagnostic technique in medicine has long dominated as an affordable and safe method. While ultrasound on its own is highly effective, imaging can be visually enhanced with the use of microbubbles to boost resonance and signal image. Small gaseous bubbles covered with a surfactant molecule, such as Oleosin 30G, can enhance ultrasound wave scattering by alternatingly expanding and compressing. It is critical for these microbubbles to be uniform in size and monodisperse in solution to ensure an even distribution of motion across the acoustic waves[1]. The uniform size distribution in a monodisperse suspension promotes bubble attenuation within a narrow frequency range, producing better image resolution. Surfactants like Oleosin 30G provide this necessary stability and uniformity in bubble size, resulting in clearer ultrasound images that provide superior results

in scans[2]. These higher-resolution images can lead to a reduction in repeat scans, providing a crystal clear picture of the targeted area and an elevation in patient care.

The application in question for this project is echocardiography, more simply known as a sonogram of the heart. It is the most common and routinely used diagnostic method for patients with heart conditions, and can provide physicians with crucial information about heart function, blood pumping capacity, and the precise location of a problem. One condition that is readily identified using an echocardiogram is cardiomyopathy, or heart muscle disease. As a disease that led to 443,000 deaths in 2013,[3] cardiomyopathy is one of the biggest existing disease areas and could immensely benefit from higher-quality imaging. Contrast agents used to enhance echocardiography signals are extremely promising solutions.

In the United States, there is a defined but relatively underdeveloped market for contrast agents for use in ultrasound. Contrast agents, in the form of small gas bubbles coated with a shell layer, enhance the signal of ultrasound by acting as resonators to increase scan resolution and efficacy. The history of contrast agents receiving FDA approval goes back to only the early 1990s, where the first successful agents utilized a human albumin shell coating to stabilize bubbles (Albunex®). Further developments in technology saw the rise of agents using a phospholipid coating (Optison®, Definity®) but suffered from shell rigidity and varying bubble size. Currently, there are nearly 700,000 echocardiograms performed each year in the US that use a contrast agent for increased resolution[4]. These scans would greatly benefit from an agent with stable, monodisperse bubbles.

Oleosin is a naturally occurring protein found in vascular plant oil bodies. Through extensive research and manipulation, Dr. Daniel Hammer's lab at the University of Pennsylvania has developed Oleosin 30, a genetically engineered recombinant form of Oleosin, altering the hydrophobic domain of the protein by removing 57 amino acids to prevent the forming of a secondary structure in order to create a random coil surfactant capable of self-assembly.[5] The recombinant form is also given a 6-histidine tag on the C-terminus to aid in purification. Oleosin 30G is a variant of Oleosin 30, with the addition of five glycine amino acid groups in the hydrophobic domain to increase flexibility. This

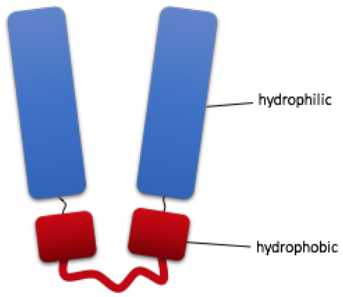


Figure 2.1.1: Oleosin 30G Protein. Oleosin 30G is an amphiphilic protein, with two hydrophilic sections and one hydrophobic section.

functionality has been studied in stabilizing the surface of microbubbles that are formed using a microfluidic device[5], and is found to be extremely promising in producing stable, uniformly sized bubbles that are monodisperse. Oleosin 30G, combined with a pluronic block copolymer, shows potential to fill a need unmet in the current market[6].

Using a lab-scale protocol from Dr. Hammer's lab as a starting point, a scaled-up process to manufacture Oleosin 30G on an industrial scale was designed. The process will produce, package, and distribute Oleosin 30G-coated microbubbles for use in echocardiograms around the country. *E. coli* cells are first grown up in LB-Kanamycin media with glucose in a bioreactor. The cells are then centrifuged and homogenized to release the intracellular protein. Thorough pharmaceutical-grade downstream purification is then conducted, with the protein first sent through a cobalt affinity chromatography column, then further purified using ultra/diafiltration (UFDF). Lastly, a bulk nanofiltration is used to remove endotoxins below an FDA-approved threshold. Complete quality assurance and lab validation checks will be performed on the final product to ensure safety and efficacy.

Highly purified Oleosin 30G will be sent to final packaging where eight microfluidic molds made using a master copy will process the protein solution to produce uniformly sized, stable microbubbles in buffer solution. Oleosin will be kept at a concentration of 1 mg/mL with stability up to one month in solution. Final suspensions will be packaged in individual doses and stored in sterile glass vials. Vials will be shipped to order around the country using pharmaceutical-grade shipping.

An initial economic analysis of the process was performed and shows significant promise in profitability metrics. With consistent market growth and unmet needs on the consumer end, there looks to be a good opportunity for Oleosin to enter the market.

2.2. Project Charter

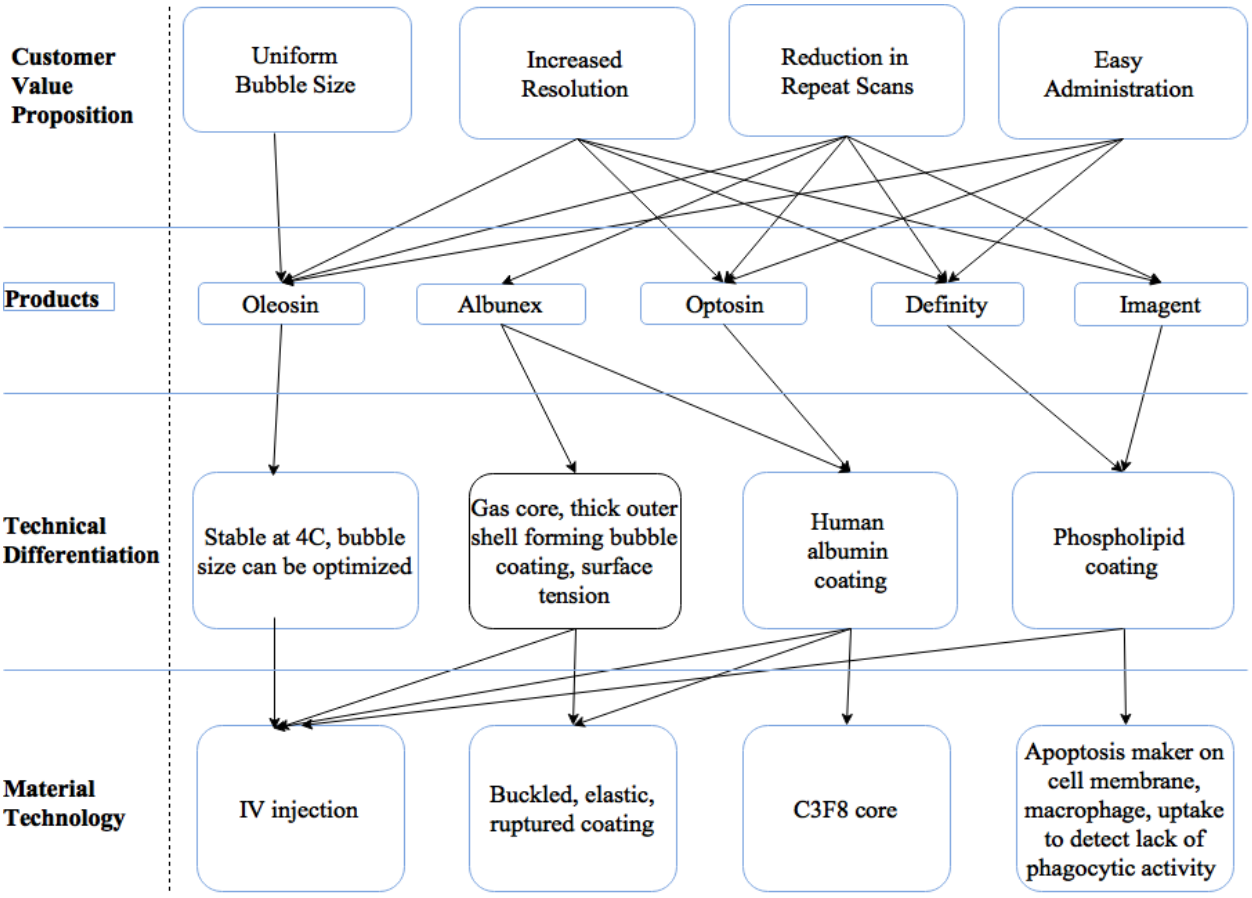
Project Name	Industrial-Scale Manufacture of Oleosin 30G for Use as Contrast Agent in Echocardiography
Leaders	Steve Casey, Hailey Edelstein, Rebecca Michelson, Nikita Rao
Project Scope	In Scope: • Manufacturing process for 7.39 kg/year production of Oleosin 30G beginning with inoculum preparation and concluding with large-scale fermentation • Post-manufacturing purification steps, beginning with cell lysis and affinity chromatography and concluding with filtration steps to purify the protein product. • Additional tests to verify the product, determine purity, and test for endotoxins • Production of microbubble suspension using a custom-designed microfluidic device • Preparation of microbubble suspension for packaging and shipment • Adherence to current health and safety regulations for intravenous compounds in the medical industry • Observance of process integrity and compliance by adhering to GMP (good manufacturing processes) • Costs of FDA approval and clinical trials for a drug candidate • Pilot plant study to analyze cell growth rates and optimal conditions

	Out of Scope:
	• Development of Recombinant Oleosin 30G (gifted from Dr. Daniel Hammer)
	• Research and development (performed in laboratory)
	  • For general characterization of Oleosin 30G
	  • Design of microfluidic device for microbubble production
	  • Identification of Oleosin concentration per microbubble
	  • Efficacy testing of microbubbles
	• FDA approval for use as an ultrasound contrast agent
	• Success of clinical trials

Deliverables	Business opportunity assessment • What is the current market for contrast agents? • How does the size distribution of the Oleosin-stabilized contrast agent compare to the distribution of a currently employed contrast agent? Technical feasibility assessment • Is it feasible to produce 7.39 kg/year of Oleosin to address consumer needs? Manufacturing capability assessment • Will this facility require significant capital investment to produce Oleosin? • Will the facility require significant capital investment to produce and store microbubbles with a custom microfluidic device? • Will the process satisfy FDA requirements?
Timeline	Facility, process design and economic analysis will be completed in 5 months.

2.3. Innovation Map

As seen in Figure 2.3.1 below, contrast agents for ultrasound hold customer value based on their technologies in application. The variety of contrast agents available on the market in the United States today fall into two main categories: those that have a bubble coating made of human albumin, and those with a phospholipid coating. Albunex®[7] and Optison®, earlier entrants to the market, use human albumin to stabilize the bubble surface, forming a thick outer shell with significant surface tension. This rigidity has negative implications for ultrasound due to the restriction of bubbles to vibrate effectively. Definity® and Imagent® by contrast, using a phospholipid coating, have a thinner and more flexible shell resulting in greater movement. All agents mentioned utilize a C_3F_8 core, whereas Oleosin 30G microbubbles use nitrogen gas, which is safer and more stable inside the bubble[4]. In terms of administration, Oleosin 30G bubbles are administered via intravenous (IV) injection, as are the competitors. Oleosin 30G utilizes a pluronic, the concentration of which can be manipulated to optimize bubble size for a particular application or therapeutic area.



3. Concept Stage

The current U.S. marketplace for contrast agents with applications for ultrasounds is narrow and relatively underdeveloped. While merits of ultrasound contrast agents were initially studied going back to the 1960s, it was not until within the last 20 years that any contrast agent successfully received FDA approval after proceeding through clinical trials. The market is in its youth compared to other pharmaceutical areas.

There are currently two main types of contrast agents available in the US: those utilizing a human albumin coating on the surface of the gas microbubble (Albunex®, 1994 and Optison®, 1997), and those with a phospholipid coating (Definity®, 2001 and Imagent®, 2002). Toxicity concerns were addressed during the FDA approval processes for these products but these commercially available products have faced issues with efficacy in the years since their approval. The ultrasound contrast agents with albumin coatings have previously been treated as a solid elastic shell and therefore experience small oscillations in bubble size. Those with phospholipid coatings have been treated as surfactants with a thinner shell; however, these bubbles have been seen to only respond with compression and no corresponding expansion in conducted studies[12].

Oleosin 30G, the genetically engineered recombinant form of naturally occurring Oleosin protein that this process is designed to manufacture, functions as a surfactant on the surface of the microbubble, forming a thin layer with Oleosin 30G comprising approximately 10% of the bubble's surface. The resulting bubbles are highly uniform in size, with corresponding variability at a minimum to ensure greater efficacy in ultrasound scans. Oleosin 30G can replace current commercially available contrast agents, ensuring increased resolution and accuracy on scans, ultimately reducing the need for repeat scans and contributing to generally better patient care.

The current targeted market is sized at 700,000 contrast agent injections performed each year, specifically in the area of cardiology. This process aims to produce Oleosin 30G for injection as a contrast agent in echocardiograms across the country.

3.1. Market and Competitive Analysis

While Oleosin microbubbles have the potential to increase contrast in many different types of ultrasound, this analysis and subsequent production level will focus on data for contrast-enhanced echocardiograms. The use of contrast agents in echocardiography in the United States

is well established and proven to be profitable, with multiple agents approved for use in the US including Albunex®, Definity®, and Optison®.

The amount of echocardiograms that incorporated contrast agents has remained rather constant between 2010 and 2015, with 700,000 injections per year in 2010 and 600,000 injections per year in 2015[13]. Definity®, with 90% of the market share in 2015,[14] reported a revenue of $106,000,000, which would be the main competitor to an Oleosin 30G-based contrast agent[16]. We expect this product to be as quickly adopted in medical practice over Definity® as Definity® was adopted over Optison®. In Oleosin 30G-stabilized microbubbles, the echogenicity and therapeutic functionality can be optimized to each type of echocardiography procedure simply by changing the relative concentrations of Oleosin and pluronic fed to the microfluidic device used to form the microbubbles. This should lead to increased resolution in scans as compared to scans performed with Definity®. Definity® is also sold as a solid that must be mixed in a proprietary VIALMIX® mixer. Oleosin 30G-stabilized microbubbles will be shipped as a finished product ready to be injected. This increased ease of use will decrease market inertia to adopt our product, as no additional training or knowledge is needed to prepare the microbubbles before injection.

Contrast agents are used in a small percentage of overall echocardiogram procedures, as less than 10% of echocardiograms use contrast agents. Contrast agents are mainly used only in a follow-up procedure, after the first procedure is inconclusive. It has been shown that contrast agents increase the percentage of adequate scans from 58% to 70% for harmonic imaging, and from 38% to 80% for left ventricular ejection fraction imaging (LVEF). Average cost savings, due to a reduction in the need for repeat scans, were calculated to be $2.17 per patient for each percent increase in quality for harmonic wall imaging and $4.23 per patient for each percent increase for LVEF. This translates to a savings of $26.04 for harmonic wall imaging and $177.66 for LVEF[17]. Since the use of contrast agents in all echocardiograms has been shown to reduce the overall cost per patient, it can be expected that with proper marketing the percentage of procedures that use contrast agents can greatly increase in the future, so there is much room for the market to grow.

3.2. Customer Requirements

Recombinant Oleosin 30G protein used as a surfactant in a microbubble suspension is targeted for use as a contrast agent in echocardiogram ultrasounds. As a result, the identified customers are patients requiring echocardiograms. The product will be directly sold to hospitals, radiology offices, and other medical facilities at which echocardiograms are conducted. It is expected that customers will purchase the product either out of pocket or through their insurance companies. Requirements that must be fulfilled for the product's success include appropriate dosage, product efficacy, packaging for easy administration, and distribution to facilities quickly.

An important requirement for the customers is contrast agent dosage, which must abide by FDA regulations. The current on-the-market contrast agent is Albunex®, which uses doses that range from 3.4 to 10mL, containing $4x10^8$ microbubbles per mL, with bubbles of 4 μm in diameter on average[18]. The optimal microbubble suspension dose is between 0.033 and 0.5 mL/kg. It is expected that the suspension of microbubbles made with Oleosin 30G will begin with a similar concentration of microbubbles, and will be optimized for patients through clinical trials. Each dose will be produced to have an Oleosin 30G concentration of 1 mg/mL in bubble solution for optimal microbubble stabilization.

Similarly, product efficacy and advantages over the current market contrast agents will need to be proved in a clinical trial. The microbubbles in the suspension must be consistently-sized at the resonant diameter for echocardiogram ultrasound, remain stable in both storage and administration, and produce molecular images with better resolution and contrast so that it can lead to more definitive diagnoses for patients.

Another relevant requirement is in the product packaging to enhance ease of use of the product. After the recombinant protein has been isolated, purified, and packaged into the appropriate concentration per dose, microfluidic generation of the microbubbles will also be conducted. This is necessary to ensure that all medical facilities can administer the contrast agent without purchasing a custom microfluidic device to create the suspensions. Doses will be packaged in sterile vials of the microbubble suspension, which will be ready for injection upon arrival to the patient at a medical facility.

The shelf life of the prepared microbubble suspension is 30 days, so orders for the product will be placed when the echocardiogram is scheduled. The suspensions will be prepared seven days before use and shipped overnight to the medical facility location. This will allow

enough time for facilities to receive and store the product use in echocardiograms. It is recommended that the injection be used within 7 days because the microbubbles have a uniform size distribution within this time frame.

3.3.1. Process and Facility Requirements

The new facility for production of Oleosin will be built in Medford, MA. The Boston area has become a hub for biotechnology innovation. Medford is located less than five miles from Cambridge, MA. The area provides access to the world's top universities including MIT, Harvard, Tufts, Mass General, etc. Purchasing land is a savings of almost 50% as compared with Cambridge: roughly $31 vs $60 per square foot[19].

The 5053 square foot Cummings Properties' lab-ready suite in Medford features already-existing equipment which eliminates some construction costs. It has available rental space for modification and construction for portions of our design facility that need to be modified to fit production needs. The building features an in-house design and construction team that can be employed to make necessary modifications before the production begins with major build-out financing available. Also, heating, cooling and electrical utility charges can be included in a facility's base rent if defined as below the threshold amount in the contract. Some properties already feature "R&D" style design with previously designated "clean-room" areas in the floor-plan[20].

The site will be modified to match the design needs for Oleosin 30G production on an industry-scale. Particularly, a large requirement for the success of the company depends on the production of microfluidic devices to produce microbubbles; these bubbles are ultimately injected into the patient for contrast-enhanced ultrasounds. While many currently existing microfluidics companies offer services to create custom devices according to a design blueprint, none have the capacities to use the device to produce microbubbles in compliance FDA regulations for a drug product. Therefore, the facility design will also incorporate a process and packaging facility to produce microbubbles stabilized with Oleosin 30G using a custom-made microfluidic device. Additionally, the facility will have equipment to test and ensure product quality. The company will also be responsible for packaging, storage and overnight shipment of these devices in accordance with FDA regulations.

3.3.2. Plant Layout

The floor plan in Figure 3.3.2.1 is based on a base floor plan of our facility provided by Cummings Properties. The largest lab will be used for inoculation, protein production and cell growth. This is separated from the purification & packaging facilities in order to prevent bacterial contamination of the drug product and cross-contamination between batches. The facility also includes “air locks” to prevent contamination between different areas of the facility as per cGMP requirements.

The two large, pre-existing lab facilities are enough to accommodate our process as our tanks/bioreactors are “benchtop” size. The property also features a “construction-ready” zone with HVAC (used for clean, filtered air) and other lab capabilities as indicated on the floor plan diagram. This area will allow for the addition of additional office space and storage of our product. Also, a “cold room” is constructed in one of these construction-ready areas as all protein purification processes must be conducted at 4°C[20].

The property has an autoclave room to sterilize lab equipment used in each batch of protein production. Lastly, the rental agreement includes a “cleaning” program that will take care of any bio-waste and chemical waste that needs to be removed from the premises after heat inactivation and neutralization.

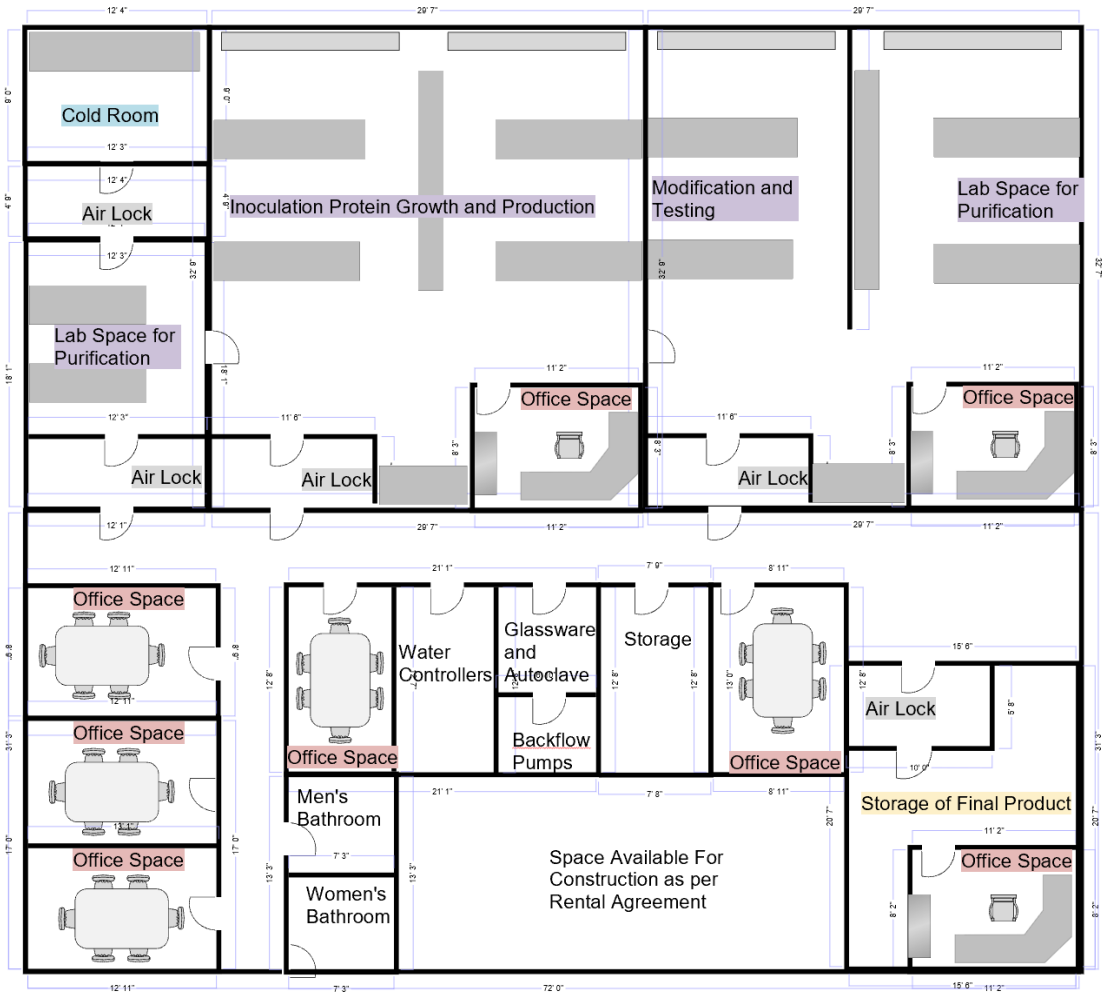


Figure 3.3.2.1 Floor Plan of Facility. Separate spaces are divided off for cell culture, purifications, testing and validation, and final product storage. A clean room is included to be kept at 4 ºC. Office spaces and air locks are included as well as bathrooms, controllers, and equipment storage spaces.

3.4. Overall Process Diagram

The overall process flow diagram in Figure 3.4.1 outlines the designed production process macroscopically. Initially, recombinant *E. coli* will be grown to a desired concentration. At the desired cell concentration, an inducer will be used to initiate and drive protein expression. After five hours of protein production, the protein will be removed from the cells using cell lysis and the sludge mixture will go through a series of purification steps to isolate the Oleosin 30G protein. These purification techniques include affinity chromatography, ultra/diafiltration, and positively-charged bulk microfiltration. Quality control tests will be conducted before microbubble suspension assembly and packaging.

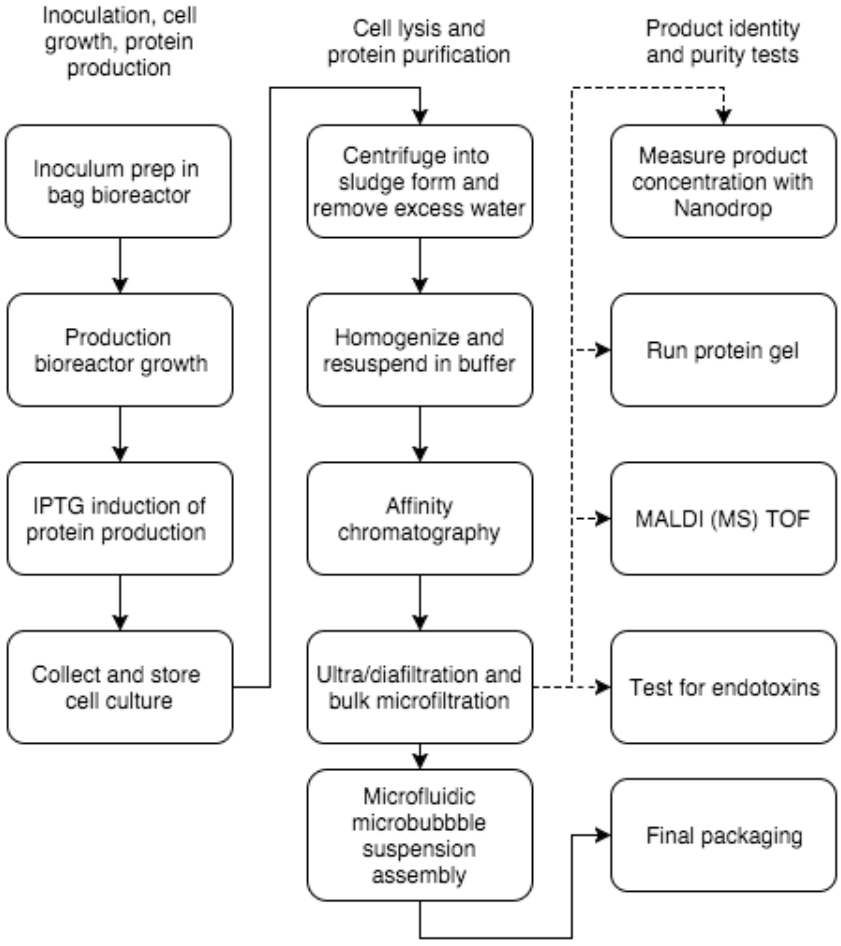


Figure 3.4.1: Oleosin Overall Process Flow Diagram.

4. Process Flow Diagrams

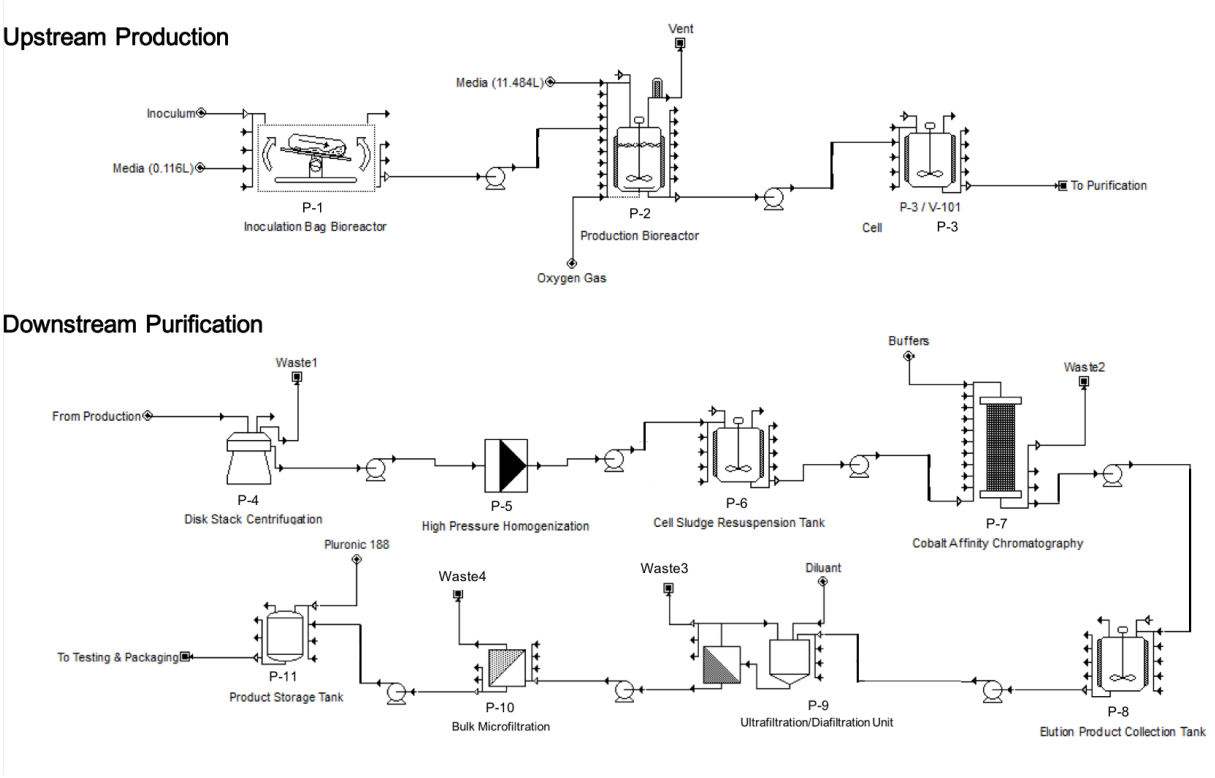


Figure 4.1: Upstream and downstream process flow diagrams.

5. Production Process Description

5.1. Inoculum Preparation

The inoculum for the production bioreactor is prepared in a 0.2L disposable bag bioreactor with a 0.12L working volume. The unused volume will allow for rocking without overflow. The working volume will be inoculated with 1.2 mL of a frozen stock of *Escherichia coli* (*E. coli*) at a cell concentration of 7.6×10^8 cells/mL. The culture will grow overnight while rocking at 220 rpm and 37°C for 12 hours to reach a saturated cell culture concentration of 1×10^9 cells/mL. This cell concentration corresponds to an OD600 value of 1. Medium in the bag bioreactor will contain 10 g tryptone/L, 10 g NaCl/L, 5 g yeast extract/L, 1 g glucose/L, and 50 mg kanamycin/L[22]. After the 12-hour growth period, the inoculum culture will be transferred to a disposable storage bag and refrigerated to prevent further growth until it is time to inoculate the production bioreactor.

5.2. Production Bioreactor Preparation

The production bioreactor will be inoculated with the culture from the inoculation bag bioreactor. First, 11.48 L of prepared and sterilized media will be transferred into the 19.5 L production bioreactor via a peristaltic pump. Immediately after, the 0.12 L of inoculum culture will be transferred in through a peristaltic pump to fill the working volume to 11.6 L. The culture will be stirred continuously at 220 rpm to keep the culture well-mixed. The pH will be maintained at 7.0, the temperature will be held at 37°C via a heated jacket, and sterile oxygen will also be sparged to maintain a dissolved oxygen concentration of 40% throughout the full growth period. All conditions will be stabilized using PID feedback controls. Growth will continue for 7.04 hours to reach a cell concentration of $1x10^9$ cells/mL, an OD600 value of 1. Media in the production bioreactor will contain 10 g tryptone/L, 10 g NaCl/L, 5 g yeast extract/L, 1 g glucose/L, and 100 mg kanamycin/L[22].

When the desired OD600 value is reached, isopropyl β-D-1-thiogalactopyrranoside (IPTG) will be added to the culture to induce protein production. IPTG will be transferred in to reach a final concentration in the working volume of 1 mM. Protein production will continue for five hours, with the same conditions specified for cell growth maintained. Assuming that the cell concentration remains approximately constant throughout the protein production period, with a cell productivity of 70 pg Oleosin 30G/cell/day,[23] 169 g Oleosin 30G will be produced per batch. After the 5-hour protein production period, the full culture will be transferred via peristaltic pump to a disposable bag holding tank that will keep the culture at 37°C via a heated jacket in order to clear out the production bioreactor so that CIP/SIP procedures can start immediately after transfer out. The disposable storage bag will also be rocked to avoid cell settling and refrigerated to prevent further cell growth. Due to some losses accounted for in transport, an 85.7% recovery in the full upstream production process is assumed, so 73.9 g Oleosin 30G will be recovered per batch to move on to downstream purification.

5.3. Cell Growth Model for Batch Bioreactor

Cell growth times were optimized based on the desired final cell concentration using Monod growth kinetics analyses, included in Appendix A. A specific growth rate of 0.5931 hr^{-1}, a glucose yield coefficient of 0.5 g cells/g glucose[24], and an oxygen yield coefficient of 1 g cells/g oxygen were assumed[25]. A study using a 3.7 L bioreactor in the Department of Chemical

and Biomolecular Engineering was conducted to validate cell growth on a larger scale of the recombinant *E. coli* line. These results are included in Appendix A.

6. Purification Process Description

6.1. Holding Tank

Upon completion of upstream protein production, the spent media and culture (11.6 L) with the product will be transferred via a peristaltic pump to a 20 L disposable holding bag. The holding bag will be rocked at 220 rpm and 4 °C to prevent settling of the cells and to minimize loss during transfer out of the holding tank via peristaltic pump to centrifugation.

6.2. Centrifugation

The holding bag contents will be transferred into a disk stack centrifuge by a peristaltic pump. The unit will separate the solid parts of the culture (including cells, product, and debris) from the spent liquid media. Spent media will be directed to a waste tank for safe disposal while solids will remain in a sludge that is 60% by mass water and 40% by mass solids. The centrifuge will run with a flow rate of 100 L/hr at a temperature of 25 °C. It is assumed that 5% of Oleosin 30G will be lost via the liquid waste stream, leading to a product yield of 95%. Centrifugation, with CIP and SIP procedures, will require 2.8 hours.

6.3. Buffer Preparation

Both chromatography and resuspension steps after centrifugation will require buffer solutions. These buffers will be made by dissolving appropriate amounts of solids in water-for-injection (WFI). For chromatography, the equilibration, wash, elution, and regeneration buffers will require various concentrations of imidazole, sodium chloride, sodium phosphate, and urea, all of which will be purchased as solids. All buffers will be prepared in separate disposable containers at 25°C to minimize contamination and cleaning and sterilization procedures. The pH of each buffer will be verified and controlled to ensure proper preparation for each. The concentrations of components for each buffer are listed in Table 6.3.1.

Table 6.3.1: Component breakdown of buffers for affinity chromatography. Equilibration buffer will be used to bind protein initially, wash buffer will be used to wash away any cell debris, elution buffer will be used to collect bound protein from the column, and finally regeneration buffer will be used to regenerate the resin for another column run.

	Equilibration (pH 7.4)	**Wash** (pH 7.4)	**Elution** (pH 7.4)	**Regeneration** (pH 5.0)
Sodium phosphate	20 mM	20 mM	20 mM	0 mM
Sodium chloride	300 mM	300 mM	300 mM	0.1 M
Imidazole	5 mM	10 mM	150 mM	0 mM
Urea	8 M	8 M	8 M	0 M
Ethanol	0%	0%	0%	20%
2-ethanesulfonic acid	0 mM	0 mM	0 mM	20 mM

6.4. High Pressure Homogenization

The cell sludge from centrifugation will be sent through a high-pressure microfluidizer processor for cell homogenization. Bacterial protein extraction agent (B-PER) and DNAse will also be added to the sludge as it goes through homogenization. The unit will pump the sludge through a pressure change of up to 20,000 psi in fixed-geometry microchannels to generate high velocity and shear rate. The process will require 3 passes to create a uniform emulsion. The total homogenization process including cleaning and sterilization will require 3.8 hours. Recovery of product in this step is 95%.

6.5. Cobalt Affinity Chromatography

After complete cell lysis and resuspension in equilibration buffer, cobalt ion affinity chromatography will be the first step to separate the Oleosin 30G protein product from the rest of the cell debris. The total chromatography column size is 21 L (refer to Appendix D for separation sizing). All procedures in chromatography will be conducted at 25 °C and the batch will require 13 L of resin purchased from Thermo Scientific[26]. The linear velocity of all parts of chromatography will be maintained at 150 cm/hr. The chromatography separation, including set up and resin regeneration, will require 5 hours.

The column must first be run with 10 column volumes of equilibration buffer. The resuspended solution of cell debris and product with equilibration buffer will then be applied to the column to allow for protein binding to the resin. The column will be washed with 10 volumes worth of wash buffer to eliminate any debris that may bind to the resin. Lastly, 10 column volumes of elution buffer will be used to collect all bound protein. In total, approximately 11.1 mg of

Oleosin 30G will be recovered per mL of resin and purity is guaranteed to be at least 90%. Recovery rate of the product is calculated to be 63.4%.

Regeneration of the column resin will requires washing the column with 10 column volumes of regeneration buffer. The resin can be regenerated a maximum of 25 times without decreasing the purity of the protein product below 90%.

6.6. Ultra/Diafiltration

The second step in separating the Oleosin 30G protein from all other proteins will require concentration and the addition of clean buffer, using a batch ultra/diafiltration process. The filters used will have a 10 kD molecular weight cut off (MWCO) and will be run in tangential flow. This filter size will allow Oleosin to remain in the retentate while buffer and smaller particles flow through to the permeate. The elution buffer with Oleosin 30G from the chromatography column will flow through the ultrafiltration system, with the concentrated retentate flowing back into the original elution buffer storage tank, and the permeate containing small particles and buffer will be discarded. While this process is occurring, new buffer solution will be pumped into the elution buffer storage tank. Over time, as new buffer flows in and buffer is pulled through the filter to the permeate, the concentrated Oleosin 30G solution will contain only new buffer.

It is assumed that 95% of the product will be recovered from ultrafiltration and 5% will lost in the permeate. This filtration step will require 2 hours. The filters contain Ultracel membranes and 1.14 m^2 surface area and can be run with a flowrate of 4 L/min. The product will be concentrated to a final concentration of 1 mg Oleosin 30G/mL. Recovery of product in this step is 95%.

6.7. Sterile, Bulk Microfiltration

Sterile filtration is used as a final purification step to remove endotoxins from the final product solution to meet FDA standards[27]. The filters will be positively-charged Durapore PVDF membranes with 0.22-micron pore size[28]. The filters and holder unit together are disposable and one will be used per batch. The filters have 100 cm^2 surface area and will run with a flowrate of 11 L/min. Recovery of product in this step is 95%. This unit will be run twice to ensure that FDA requirements for endotoxin removal are met, where 99% of endotoxins are removed per filtration.

6.8. Microfluidic Processing

From the final product holding tank, the solution with 1 mg/mL Oleosin 30G will be pumped through eight microfluidic devices in series to produce 5934, 10 mL-doses with $4x10^8$ nitrogen gas bubbles/mL. The suspensions will be stored in pharmaceutical-grade glass vials. These vials will be stored at 4 °C until shipment. Recovery of product in this step is 99%.

7. Major Unit Operation Specifications

7.1. Common Units

7.1.1. Pumps

Peristaltic pumps will be used to transfer all fluids that contain cell mass or protein product. Peristaltic pumps are best for fluids that are viscous and can show better performance for fluid transport than centrifugal pumps. Twenty small Masterflex[29], 600 rpm pumps will be purchased from Cole-Parmer for $2,000 per pump. The flowrate through each pump ranges from 0.006 to 3400 mL/min. In addition, two large peristaltic pumps will be purchased from Watson-Marlow[30]. The flowrate through each pump ranges from 653 to 8,140 L/h. Each pump will operate at room temperature. Sterile tubing will also be purchased to guide fluids through the process.

7.1.2. Digital Control Units

Digital control units will be purchased to conduct proportional integral derivative (PID) feedback control. In the bioreactor, this unit will control agitation speed and work to maintain constant temperature at 37°C, constant pH at 7.0, and constant percent oxygen at 40%, and constant liquid level to avoid excessive foaming (see Appendix J for an overall bioreactor control diagram). Separate units will be purchased to control each of these parameters. A total of 20 units will be needed, including back-ups. The units will be purchased at a price of $300 per unit.

7.2. Inoculum Preparation Section

7.2.1. Bag Bioreactor (P-1)

In each batch, a new disposable and sterile bag will be used to hold the inoculation culture for overnight incubation. This helps to reduce opportunities for cross-contamination between

batches and saves time, energy, and labor for CIP and SIP procedures. The control tower will be purchased from Sartorius Stedim Biotechnology Group for $49,000. A wave bag rocker will be purchased from Sartorius Stedim Biotechnology Group for $9,300[33]. The base will rock at 220 rpm. The bags will be purchased from HyClone for $174 per 0.2L bag[34]. The unit will keep the bag and its contents will be kept at 37 °C, 1 bar, pH of 7.0, and a percent oxygen of 40%. This unit will operate overnight for 12 hours.

7.3. Production Bioreactor Section

7.3.1. Production Bioreactor (P-2)

The BioFlo bioreactor will be purchased from Eppendorf for $104,000[35]. It has a total volume of 19.5 L and a working volume of 11.6 L, or 59.5% working capacity. The bioreactor is stainless steel with dimensions of 134.6 cm in height, 66.0 cm in diameter, and 63.5 cm in width. It will also have a stainless steel agitator. The reactor will be jacketed and a digital PID control unit will be used to maintain the temperature at 37°C. Control units will be used to maintain pH, percent oxygen, and agitator speed (see section 7.1.2.). The bioreactor will operate at 1 bar. The bioreactor will also have a collection tube for sampling to record the optical density over time until the desired OD600 value of 1 is met, corresponding to a cell concentration of $1x10^9$ cells/mL.

The cells will grow for 7.04 hours to reach this cell concentration, after which IPTG will be added to induce Oleosin 30G production. The temperature, pH and percent oxygen will still be held at the same values used for the cell growth period. It is assumed that negligible cell growth will occur after the addition of IPTG because IPTG puts stress on the cells to overexpress the recombinant protein. The protein production will continue for 5 hours until 169 g of Oleosin 30G has been produced.

7.3.2. Storage/Mixing Tank (P-3)

As soon as the desired cell concentration is reached in the production bioreactor, all culture contents will be transferred to a holding tank at 4°C. This will allow for CIP and SIP procedures for the bioreactor to begin as soon as possible. The holding tank will be stainless steel and have an agitator rotating at 220 rpm to prevent cells from settling at the bottom of the tank. The 20L disposable bag will be purchased from HyClone for $200.

7.4. Purification Section

7.4.1. Disk Stack Centrifuge (P-4)

The purpose of the centrifuge is to separate the solid product (the cells and their produced protein) from the spent liquid media. The media will be directed to a waste holding tank (see section 8.1) while the solid product will move downstream through purification. A disk stack centrifuge will be purchased from Alfa Laval for $46,500 per unit[36]. The solid product will be in the form of a sludge that is 40% solids and 60% water by volume. The flow rate through the centrifuge is 100 L/hr. This unit will operate at 25°C and its process time including CIP and SIP is 2.8 hours.

7.4.2. High Pressure Homogenizer (P-5)

The cell sludge from centrifugation will be sent through a high pressure microfluidizer processor to conduct homogenization. The unit will pump the sludge through a pressure change of up to 20,000 psi in fixed-geometry microchannels to generate high velocity and shear rate. The process will require 3 passes to create a uniform emulsion. The total homogenization process including cleaning and sterilization will require 3.8 hours.

7.4.3. Homogenizer Product Resuspension Tank (P-6)

After centrifugation and homogenization, the full volume of cell sludge will be resuspended in an equal volume of equilibration buffer. Additional equilibration buffer can be added until the pH reaches that of the equilibration buffer, 7.4. The equilibration buffer will first be transferred in at a volume equal to that of the cell sludge, and immediately after, the cell sludge will be transferred in. The resuspension tank will mix its contents at 220 rpm and hold a temperature of 25°C. Additional equilibration buffer will be added in to reach the desired pH of 7.4, based on PID feedback controls.

7.4.4. Cobalt Affinity Chromatography Column (P-7)

Cobalt affinity chromatography will be used to separate the Oleosin 30G protein from the rest of the cell debris that comes out of homogenization. The column will be purchased from Pall Corporation for $190,000. It will be made of stainless steel and have a total volume of 21 L. The

column will be packed with HisPur Cobalt Superflow Agarose resin[26] purchased from Thermo Scientific for $5,680 per L. The resin allows for a linear velocity of 150 cm/hr. The binding capacity of the resin is 11.11 mg Oleosin 30G/mL. The column will be located in the cold room section of the workspace, so the chromatography will operate at 4°C and 1 bar.

7.4.5. Affinity Chromatography Collection Tank (P-8)

All column flow-through will be directed to the waste tank for heat inactivation for disposal. A separate holding bag will be used to collect the eluted protein from the column during the elution step. This disposable bag will be purchased from Hyclone[34] for $500 and will have a total volume of 500 L and working volume of 210 L. The bag will also be stored in the cold room to keep the proteins at 4°C.

7.4.6. Ultra/Diafiltration Unit (P-9)

The ultrafiltration base unit will be used to conduct tangential flow filtration (TFF) and will be purchased from EMD Millipore[37] for $1,000. The unit has a base that holds disposable filters. Filters will cost $200 each and one filter will be required for each batch. The filters contain a membrane with a Molecular Weight Cutoff (MWCO) of 10kD and is also from EMD Millipore as part of the Pellicon 3 cassettes with Ultracel membranes. The effective filtration surface area is 1.14 m^2 and the fluid will flow at a flow rate of 4 L/min. Process time including SIP procedures for the base will be 5 hours. Further recirculation is not required.

7.4.7. Bulk Microfiltration (P-10)

This positively-charged filtration unit will be used as a final filtration step to remove endotoxins, nucleic acids, and viruses. Since endotoxins, nucleic acids, and viruses are all negatively-charged, this type of positively-charged Durapore filter[38] will catch 99% of them, along with other negatively-charged functional groups. This filtration will be run twice, with a final endotoxin level 0.874 Endotoxin Units (EU) per dose (87.4 pg per dose), satisfying FDA requirements for maximum endotoxin levels per 0.5 kg of body weight. The filters with 2.2 μm pores will be purchased from EMD Millipore for $177 per filter. The filters and the housing unit for each will be disposable[32].

7.4.8. Final Product Holding Tank (P-11)

A 200 L disposable holding bag will collect the 169 L of final product (169 g Oleosin 30G in 169 L). This bag will keep it sterile at 4°C while testing is being done before microfluidic packaging. Assuming that testing is successful, solid pluronic P-188 will be added to a final concentration of 10 mg/mL[2].

7.5. Microbubble Suspension Assembly

7.5.1. Microfluidic Device Master Fabrication

An SU-8 developer is used to thin a negative photoresist SU-8 2010 to a 3:1 ratio. The photoresist is then spin-coated onto silicon wafer to a thickness of 5 μm. Using a Karl Suss MA4 Mask Aligner, it is then patterned through a transparency photomask to UV light. Sylgard 184 PDMS is mixed with cross-linker in a ratio of 12:1. In order to degas the mixture, it is kept in a desiccator to allow escape of trapped air-bubbles. It is then poured onto a photoresist pattern and cured for an hour at 65 °C. The curing process transforms the PDMS into a flexible solid and makes the membrane highly compliant. The PDMS replica are peeled off the designed wafer and bonded to a membrane via spin-coating PDMS on a glass slide[39].

7.5.2. Microbubble Production

A solution containing 1mg/mL Oleosin 30G protein is mixed with 10 mg/mL triblock copolymer (pluronic P-188) to reach a pH of 7.2. This mixture is introduced into the device using a syringe pump at flow rates between 500 and 1000 μL h^{-1}. A pressure regulator supplies 99.999% pure nitrogen gas to the device at pressures between 15 and 20 psi. To produce microbubbles, initially, a small pressure between 2 and 5 psi is first applied to the gas inlet at the desired flow rate. The pressure is increased slowly until bubble generation reaches steady state.

7.6. Product Packaging

The produced suspension will be separated into 10 mL aliquots in pharmaceutical-grade vials. The concentration of bubbles will be $4x10^8$ bubbles per mL of solution, with 1mg/mL of Oleosin 30G. About 10% of the bubbles' surface area will be covered by Oleosin 30G after an

equilibrium is reached and the rest of the protein will remain in solution. These vials will last up to 1 month, stored at 4°C.

8. Additional Equipment Description

There are a number of units that will need to be purchased for product completion that are not shown on the process flow diagram. These pieces of equipment are required with sterile cell culture and storage, sterile buffer preparation, proper waste disposal, and product verification steps.

8.1. Biosafety Cabinet

A biosafety cabinet will be necessary for use as a sterile cell culture space. The cabinet provides special sterile air circulation that minimizes exposure to airborne pathogens. This cabinet will be used to prepare cell stocks for inoculation and to maintain a constant amount of frozen stock available for inoculation. The frozen stock will have a cell concentration of $7.6x10^8$ cells/mL and aliquots will be prepared in 1 mL tubes. The rented lab space provides a biosafety cabinet, so its price is included in payment towards rent in annual expenses.

8.2. Refrigeration

A -80°C freezer will be necessary to maintain the frozen cell stock and certain reagents for long term storage. This ultra-cold freezer will be purchased from Thermo Scientific for $30,000. Additionally, a -20°C freezer will be necessary for storage of some reagents. This freezer will also be purchased from Thermo Scientific for $8,700. Product storage will require four 4°C refrigerators. These will be purchased from Thermo Scientific for $3,400 per unit. Utilities to run each of these refrigeration units is included in the annual expenses[40].

8.3. Waste Holding Tank

A stainless steel tank with 2,000 L volume will be purchased from Sharpsville Container for $500[41]. All waste that has been in contact with the cell culture or debris will be directed to this tank for proper heat inactivation before disposal. This tank will be jacketed so that it can reach and hold a temperature of 130°C for 2 minutes via a digital PID control unit to complete at least an 18-

log reduction of bacteria. Neutralization will be conducted to adjust the cell-free waste pH to 7.0. After neutralization, this waste will be discarded by the laboratory facility.

8.4. Water-for-Injection Generator

A still will be used to purify water for use in preparing media, preparing all buffers, and conducting CIP procedures. FDA standards require that the injectable product does not touch any water other than WFI to ensure sterility and reduce contamination risks. The still will be purchased from Paul Mueller Company for $20,000[42].

8.5. Clean Steam Generator

Clean steam will be necessary to conduct SIP procedures for many process units. The generator will produce clean steam from WFI, supplied by the WFI generator. The generator will be purchased from BMT USA for $60,000[43].

8.6. Buffer Transfer Bags

Disposable bags will be used to prepare the equilibration, wash, elution, and regeneration buffers for the affinity chromatography column. The bags, along with holders, will be purchased from HyClone[34]. A total of 6 holders will be needed and each holder must have a sterile bag for each batch.

8.7. Filter Integrity Test

A filter integrity test will be necessary to ensure that all filters are working properly before each batch because all filters used are disposable and will be replaced after each batch. The test can identify if a filter is torn or has some sort of blockage before the intermediate products of the process are sent through. This will help to minimize any loss due to filter manufacturing error. The test will be purchased from Millipore for $3,500[44].

8.8. Purified Air Generator

A purified air generator for all space in the labs will be purchased from Domnick Hunter for $2,500[45].

8.9. Quality Control Lab Equipment

After the final bulk filtration step of the product, there are product verification tests that must be passed for the product to go on to the microfluidics section and to final packaging. In total, less than 10μg of Oleosin 30G will be needed to complete these tests.

8.9.1 MALDI-TOF Mass Spectrometry Unit

Matrix Assisted Laser Desorption/Ionization—Time of Flight Mass Spectrometry is used to verify the molecular weight of the isolated protein based on how quickly the protein flies a fixed distance within the unit. The protein is ionized so that it can be pulled from one side to another. The MALDI-TOF MS unit will be purchased from Shimadzu for $288,000[46].

8.9.2. Protein Gel Materials

A protein gel will also need to be run for each batch to confirm the identity of the isolated protein. The materials needed include hydrochloric acid, 30% acrylamide, SDS ammonium persulfate, TEMED, glycine, EDTA, 50% glycerol, mercaptoethanol, bromophenol blue, and pre-stained molecular weight markers. These materials will be purchased from BioRad for $1,540 per batch[47].

8.9.3. NanoDrop

The NanoDrop is necessary to determine the product concentration in the final solution. Once known, the concentration will be used to determine appropriate volumes of buffer to add to for dilution before the microfluidics assembly. The NanoDrop 2000 UV-Vis Spectrophotometer will be purchased from Thermo Scientific for $9,100[48].

8.9.4. Endotoxin Test Kit

A test kit will be purchased to ensure that all batches have an endotoxin level below the FDA standard. If a batch does not pass this test, it will need to be discarded. The kit will be purchased from Lonza[49] for $1,000 per kit. Each kit can be used 192 times.

9. Unit Specification Sheets

9.1 Small Peristaltic Pumps

Description and Function	Peristaltic pumps are used for all downstream separation and purification steps from centrifugation onwards in the process. The consistent flow ensures accuracy in dispensing flow from unit to unit. A brushless motor allows for speed control of the pump.	
Vendor	Cole Parmer	
Operation	Batch	
Characteristics		
	Model:	Masterflex L/S Digital Drive
	Material Construction:	Stainless Steel
	Flowrate:	3.4 L/min
	Maximum Pressure Drop:	2.07 bar
	Graphical LCD display to show operating modes, continuous runs, timed dispense, and volume dispense. Anti-drop function to ensure accurate dispensing.	
Operating Conditions	Temperature:	4-37ºC
	Pressure:	1 bar
Purchase Cost	$2,037/pump	

9.2 Large Peristaltic Pumps

Description and Function	Peristaltic pumps are used for all downstream separation and purification steps from centrifugation onwards in the process. Large pumps will be used to transport larger volumes quickly, specifically with the transport of buffers to and from the chromatography column. The consistent flow ensures accuracy in dispensing flow from unit to unit. A brushless motor allows for speed control of the pump.	
Vendor	Watson-Marlow	
Operation	Batch	
Characteristics	Model:	840 Series Hygienic Pump
	Material Construction:	Stainless Steel
	Flowrate:	653-8,140 L/hr
	Maximum Pressure Drop:	2 bar
	Graphical LCD display to show operating modes, continuous runs, timed dispense, and volume dispense. Anti-drop function to ensure accurate dispensing.	
Operating Conditions	Temperature:	4ºC in cold room
	Pressure:	1 bar
Purchase Cost	$25,000/pump	

9.3 Bag Bioreactor (P-1)

Description and Function

Inoculum for the production bioreactor will be prepared and grown overnight in a 0.2L disposable bag bioreactor. The bag will be placed in a rocker to prevent cell settling. The purchased unit will also include a control tower to monitor aeration, pH, temperature, and rocking speed. The bags are made of plastic and sterilized.

Vendor

Sartorius Stedim Biotechnology Group

Operation

Batch

Materials Handled

	Input (kg/batch)	Output (kg/batch)
Cells	1.67×10^{-9}	1.96×10^{-4}
Nutrients	4.00×10^{-4}	0
Water	1.16×10^{-1}	1.10×10^{-1}
Kanamycin	5.80×10^{-6}	5.51×10^{-6}
Endotoxin	0	5.44×10^{-11}
Waste	0	6.09×10^{-3}

Characteristics

Model:	BIOSTAT RM
Material Construction:	Sterile plastic bag, stainless steel unit
Percent Yield:	95%
Sterilization:	Disposable bag, CIP/SIP for base
Rocking speed:	220 rpm
Volume:	0.2 L

Operating Conditions

Temperature:	37ºC
Pressure:	1 bar
pH:	7.0
pO_2:	40%
Growth time:	12 hours

Purchase Cost

Control tower:	$49,000
Rocker:	$9,300
Bags:	$200/bag

9.4 Production Bioreactor (P-2)

Description and Function: The production bioreactor will be used for cell growth to a desired cell concentration and protein (Oleosin 30G) production using induction by IPTG. The total reactor volume is 19.5L and both the vessel and agitator are made from stainless steel. Control units will be purchased separately to monitor and control the pH, temperature, pO_2, and agitation rate.

Vendor: Eppendorf

Operation: Batch

Materials Handled

	Input (kg/batch)	Output (kg/batch)
Cells	1.96×10^{-4}	1.10×10^{-2}
Nutrients	2.25×10^{-2}	0
Water	11.6	11.1
Kanamycin	5.80×10^{-4}	5.51×10^{-4}
Endotoxin	5.44×10^{-11}	5.19×10^{-6}
Waste	0	5.92×10^{-1}
IPTG	2.96×10^{-3}	2.81×10^{-3}
Oleosin	0	1.45×10^{-1}

Characteristics

Model:	BioFlo 415 Fermentation System
Material Construction:	Stainless steel unit
Finish:	Electro-polished
Dimensions:	63.5cm x 66.0cm x 134.6cm
Percent Yield:	95%
Sterilization:	CIP/SIP
Agitation speed:	220 rpm
Volume:	19.5 L

Operating Conditions

Temperature:	37ºC
Pressure:	1 bar
pH:	7.0
pO_2:	40%
Growth Time:	7.04 hours
Protein Production Time:	5 hours

Purchase Cost: $104,000

9.5 Cell Culture Storage Tank (P-3)

Description and Function	The disposable cell culture storage bag will be used to clear out the production bioreactor quickly upon completion of the fermentation and protein production periods. This will enable CIP and SIP procedures to begin promptly.	
Vendor	Hyclone	
Operation	Batch	
Characteristics	Material Construction:	Sterile plastic
	Volume:	20 L
	Sterilization:	Disposable
Operating Conditions	Temperature:	25ºC
	Pressure:	1 bar
Purchase Cost	$100/bag	

9.6 Disk Stack Centrifuge (P-4)

Description and Function: The centrifuge will be used to separate the cells and product (solids) from the spent media (liquid).

Vendor: Alfa-Laval

Operation: Batch

Materials Handled

	Input (kg/batch)	Output (kg/batch)
Cells	1.10×10^{-2}	1.05×10^{-2}
Water	11.02	3.70×10^{-1}
IPTG	2.81×10^{-3}	2.67×10^{-3}
Kanamycin	5.51×10^{-4}	5.23×10^{-4}
Endotoxin	4.96×10^{-9}	4.71×10^{-9}
Oleosin	1.45×10^{-1}	1.38×10^{-1}
Waste	0	10.7

Characteristics

Model:	Culturefuge 100
Centrifuge Type:	Disk Stack
Material Construction:	Stainless steel
Finish:	Electro-polished
Flowrate:	2,000 L/hr capacity
Percent Yield:	95%
Sterilization:	CIP/SIP

Operating Conditions

Temperature:	25ºC
Pressure:	1 bar
Flowrate:	100 L/hr

Purchase Cost: $46,500/unit

9.7 High Pressure Homogenizer (P-5)

Description and Function

The high pressure homogenizer is used to lyse cells after centrifugation to release expressed Oleosin along with cell debris and other excess proteins. The device utilizes an extremely high pressure pump with a downstream valve for homogenization. The quick pressure drop results in extreme turbulence in flow, resulting in grinding of cells and release of the product.

Vendor

IKA Process Technology

Operation

Batch

Materials Handled

	Input (kg/batch)	**Output (kg/batch)**
Cells	1.05×10^{-2}	9.95×10^{-3}
Water	3.70×10^{-1}	3.52×10^{-1}
IPTG	2.67×10^{-3}	2.54×10^{-3}
Kanamycin	5.23×10^{-4}	4.98×10^{-4}
Endotoxin	4.71×10^{-9}	4.47×10^{-9}
Oleosin	1.38×10^{-1}	1.31×10^{-1}
B-PER	3.14×10^{-1}	0
Waste	0	3.40×10^{-1}

Characteristics

Model:	M700 Series Microfluidizer
Material Construction:	Stainless steel
Flowrate:	3 L/hr capacity
Maximum Pressure:	2000 bar
Percent Yield:	95%

Operating Conditions

Temperature:	25ºC
Pressure:	2000 bar

Purchase Cost

$8,000/unit

9.8 Cell Sludge Resuspension Tank (P-6)

Description and Function	The cell sludge resuspension bag will be used to resuspend the sludge product from the microfluidic homogenizer in equilibration buffer to prepare for binding in the cobalt affinity chromatography column. A control unit will be used to monitor the pH until the solution reaches the desired pH of 7.4 and buffer will continue to be added. The total volume of the bag will be 500L and at least 10-column volumes of equilibration buffer will be added. The bag will be rocked at 220 rpm to ensure a homogeneous mixture.	
Vendor	Hyclone	
Operation	Batch	
Characteristics	Material Construction:	Sterile plastic
	Volume:	500 L
	Sterilization:	Disposable
Operating Conditions	Temperature:	4ºC in cold room
	Pressure:	1 bar
Purchase Cost	$300/bag	

9.9 Cobalt Affinity Chromatography Column (P-7)

Description and Function: The cobalt affinity chromatography column will be used to bind the Oleosin 30G protein (along with other proteins produced by the cell) and separate it from most of the cell debris. It will also help to separate the product from negatively-charged DNA and endotoxins.

Vendor: Pall Corporation

Operation: Batch

Materials Handled

	Input (kg/batch)	Output (kg/batch)
Cells	9.95×10^{-3}	0
Buffers	902	302
Endotoxin	4.47×10^{-9}	2.84×10^{-9}
Oleosin	1.31×10^{-1}	8.28×10^{-2}
Waste	0	600

Characteristics

Model:	Resolute Chromatography Column
Material Construction:	Stainless steel
Volume:	21 L
Diameter:	0.4 m
Height:	0.16 m
Max Linear Flowrate:	1200 cm/hr
Percent Yield:	63.4%
Sterilization:	CIP/SIP
Regeneration:	25 times
Purity:	≥ 90%
Resin:	HisPur Cobalt Superflow Agarose

Operating Conditions

Temperature:	4ºC in cold room
Pressure:	1 bar
Linear Flowrate:	150 cm/hr

Purchase Cost

Column:	$190,000

9.10 Affinity Chromatography Product Collection Tank (P-8)

Description and Function	The product from affinity chromatography that is eluted from the column in the elution buffer will be directed to a disposable holding bag. All other column flow-through will be directed to the waste tank.	
Vendor	Hyclone	
Operation	Batch	
Characteristics	Material Construction: Volume: Sterilization:	Sterile plastic 500 L Disposable
Operating Conditions	Temperature: Pressure:	4ºC in cold room 1 bar
Purchase Cost	$300/bag	

9.11 Ultrafiltration-Diafiltration Unit (P-9)

Description and Function

The first step in purification after the chromatography column will be an ultrafiltration-diafiltration (UFDF) step using casette filters, stabilized with a cassette holder. The filter will have a 10 kDa MWCO to allow for Oleosin to stay in the retentate (15 kDa in size) and allowing smaller particles to filter through. It is assumed that 95% of Oleosin will be recovered in this step, with a 5% loss associated with filtration through the cassette membrane.

Vendor

EMD Millipore

Operation

Batch

Materials Handled

	Input (kg/batch)	**Output (kg/batch)**
Buffer	302	12.5
Oleosin 30G	8.28×10^{-2}	7.87×10^{-2}
Endotoxin	2.84×10^{-9}	2.69×10^{-9}
Waste	0	289

Characteristics

Model:	Pellicon 3 cassettes
Membrane Filter Material:	Regenerated cellulose, polyethylene
Filtration Area:	$1.14 m^2$
Maximum Pressure Drop:	2 bar
Sterilization:	Disposable cassettes, SIP for stainless steel holder
Percent Yield:	95%

Operating Conditions

Temperature:	4 ºC in cold room
Pressure:	1 bar

Purchase Cost

Stainless steel cassette holder:	$995
100 pack of Ultracel membranes:	$479.08/pack

9.12 Bulk Filtration (Microfiltration) (P-10)

Description and Function

The final step in purification after ultrafiltration-diafiltration (UFDF) will be a microfiltration to dispose of endotoxins. Disposable Durapore filter membranes will be used with pore size of 0.22 μm. These filters are ideal for sterilization and clarification of protein solutions, allowing other small proteins to pass through to waste, with Oleosin left in the retentate. The filters are housed in a disposable Millipak holder unit as well.

Vendor

EMD Millipore

Operation

Batch

Materials Handled

	Input (kg/batch)	Output (kg/batch)
Buffer	12.5	11.9
Oleosin 30G	7.87×10^{-2}	7.48×10^{-2}
Endotoxin	2.69×10^{-9}	2.69×10^{-9}
Waste	0	6.29×10^{-1}

Characteristics

Model:	Millipak® Disposable Units, 0.22um
Membrane Filter Material:	Durapore PVDF
Filtration Area:	$500 cm^2$
Maximum Pressure Drop:	4.1 bar
Sterilization:	Disposable
Percent Yield:	95%

Operating Conditions

Temperature:	4 ºC in cold room
Pressure:	1 bar

Purchase Cost

Pack of 3 disposable membranes and holders:	$530/pack

9.13 Product Storage Tank (P-11)

Description and Function	A final 200 L disposable bag will be used to collect the product solution from bulk microfiltration. Solid pluronic P-188 will be mixed into the solution (using a wave rocker moving at 220 rpm) to prepare the final solution for microfluidic packaging.	
Vendor	Hyclone	
Operation	Batch	
Characteristics	Material Construction:	Sterile plastic
	Volume:	200 L
	Sterilization:	Disposable
Operating Conditions	Temperature:	4ºC in cold room
	Pressure:	1 bar
Purchase Cost	$200/bag	

9.14 Waste Holding Tank

Description and Function	All waste that contacts the cell culture or the protein product at any point in the process will be directed to a stainless steel 2,000 L holding tank. This waste includes all spent media, water, buffers, and cell debris. Heat inactivation and neutralization will be conducted before disposal is taken care of by the rented facility.	
Vendor	Sharpsville Container	
Operation	Batch	
Characteristics	Material Construction: Volume: Sterilization:	Stainless Steel 2,000 L CIP/SIP
Operating Conditions	Temperature: Pressure:	25-121ºC 1 bar
Purchase Cost	$500	

9.15 PID Control Units

Description and Function	Control units will be purchased to regulate conditions such as temperature, pH, pO_2, and agitation rates. They will be used for all units that are not already equipped with control units, such as the buffer mixing tanks and production bioreactor.	
Vendor	Omega	
Operation	Batch	
Characteristics	Control Type:	PID with autotuning capabilities
Purchase Cost	$300/unit	

10. Scheduling

10.1 Batch Scheduling

In order to produce the desired 100 batches per year, the designed biopharmaceutical plant will only be required to operate for 16 hours per day during business days, requiring two shifts per day, and storing the product and intermediates during the hours the plant is closed. The average number of operation days per year with viable batches was determined to be 191, starting with 251 business days per year, then accounting for 15% down time and 10% batch failure.

10.2 Gantt Chart

The designed plant must produce an average of 2.67 batches per week to meet the current market demand. With 8 hours of down time each day, as there is no third shift, no bottlenecks occur in the process, and the intermediates are stored in storage tanks overnight or over the weekend. The product is believed to be stable enough to be stored over these time frames (shown in black) without any measureable degradation or yield loss. Each batch will take 84 hours to complete, with 24 hours between batches. The standard 3 batches per week schedule is shown below. One third of the time, the plant will be run at a rate of only two batches per week so as to not overproduce product by eliminating the third batch shown in the chart below.

This schedule allows for a maximum scale up of 5 batches per 5-day week if the market allows, providing flexibility without modifying the overall process. There will be two employees per department per shift (growth and production, purification, testing, and storage), giving 16 full time employees working 40 hour weeks at the current production rate.

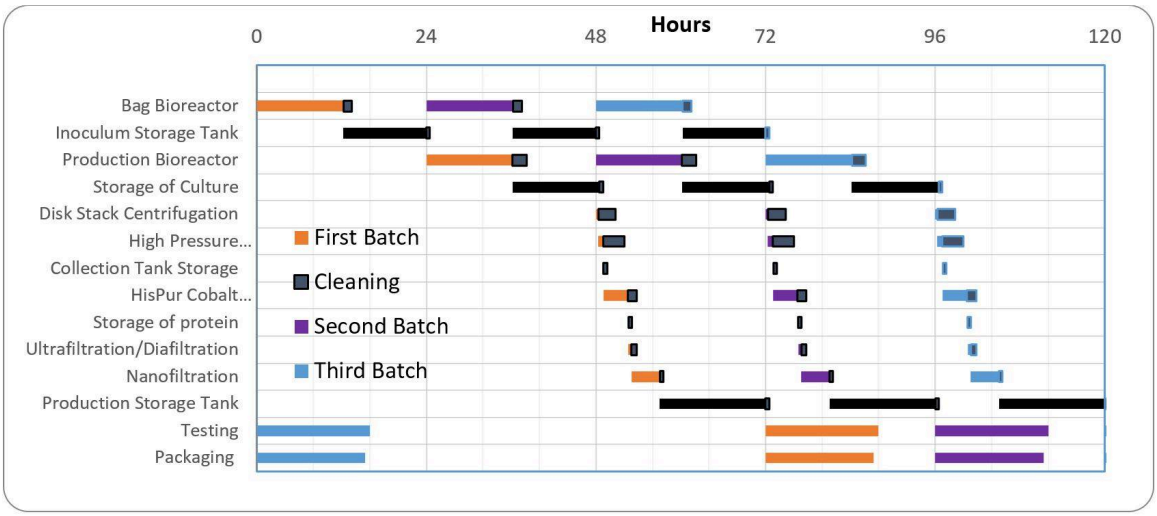


Figure 10.2.1. Gantt chart of process. Detailed scheduling is mapped out showing the overlap of batches. Cleaning times are accounted for in between batches.

The CIP/SIP times for the equipment were estimated using the manufacturer's protocol and the dimensions of the unit, as detailed in the Appendix F. It was assumed that testing and packaging will occur simultaneously, as the time number of failed batches is expected to be low, so the time lost waiting to pass all test requirements for every batch is more valuable than the time and material wasted packaging a bad batch. The run times for the bag bioreactor and production bioreactor are detailed in Appendix A, and the chromatography column timing is detailed in Appendix D. Centrifugation time was determined by the volume in the production bioreactor and the flow rate data specified in the Culturfuge100 vendor sheet, and the ultrafiltration time was determined by the volume of elution buffer from the chromatography column (Appendix D) and the maximum flow rate through the filters, as stated in its vendor sheet.

11. Other Considerations

11.1 Steam-in-Place (SIP) Requirements

Each piece of equipment will be steam sterilized in place (SIP) after CIP unless otherwise noted below to ensure complete destruction of *E. coli* and other biological contaminants between batches. The process will require 50 psi (138°C) saturated steam for 40 minutes, ensuring that every part of the equipment reaches 121°C for 20 minutes as required by the FDA[51]. After

sterilization, the equipment will be cooled by flowing room temperature air through the tubing. These cooling times were estimated for each piece of equipment based on the size or based upon the vendor sheets. SIP procedures that deviate from this are listed in Appendix F.

11.2 Clean-in-Place (CIP) Requirements

It is of the utmost importance to follow FDA regulations at every step in the process. The process is designed in accordance with Current Good Manufacturing Practice (cGMP) protocols. All equipment and process machinery that comes in contact with live cell culture must be treated as bio-waste and either disposed of or cleaned according to FDA stipulations. Clean-in-Place (CIP) and Steam-in-Place (SIP) procedures will be followed from batch to batch.

The equipment will be cleaned according to CIP protocols, with proper validation implemented after each cleaning step. Cleaning will take place in each non-disposable unit between each batch, and is accounted for in the scheduling of operations. Cleaning will first consist of a rinse using Water for Injection (WFI), followed by a detergent flush. A second circulation of detergent will be flushed, followed by an intermediate WFI rinse. Thirdly, an acid solution will be used to clear remaining cell and protein residue. A final WFI rinse will be used to clear remaining detergent from the system, with an air blow to remove moisture as the final step[52]. The amounts of these reagents used on an annual basis is considered negligible to the significance of other factors in the profitability analysis, and therefore without precise time and volume measurements for necessary cleaning reagents, the costs associated are not substantial.

11.3 Waste Treatment and Environmental Concerns

All biowaste will be heated to 80 ºC to inactivate live cultures. In order to ensure complete death of all *E. coli* cells, the live cultures will be heated to 121°C for 1 minute, with temperature control maintained, then returned to 80 ºC for the remainder of heating. As opposed to installing the necessary equipment for these procedures onsite, a costly startup venture, disposal of biowaste will be contracted out to a waste disposal service for a per gallon fee. This fee is rounded up to account for extra charges including waste shipment and transportation. Equipment that comes in contact with live cultures over the course of the process must be autoclaved to ensure thorough sterilization. All other waste is sent to a neutralization tank, where the pH is adjusted to 7.0. The neutralized waste can then travel to the sewer system. Foregoing

the pricey impact of onsite installation of a proper neutralization tank, this service is as well contracted out to an offsite party.

In order to minimize the environmental impact of the process, a limited amount of landfill waste is used to reduce the carbon footprint resulting from the production of Oleosin 30G. No hazardous chemicals or toxins are produced here, therefore disposal of toxic materials is not a concern. In addition to following CIP and SIP procedures as mentioned above, the plant will be reviewed by a quality management team. Hygienic conditions will be met for personnel as well as equipment, with regular quality ventilation and materials. Further quality assessments will be performed on the product at various checkpoints to ensure thorough uniformity of batches. Backup storage tanks are also implemented after each major step in the process to serve as potential holders for failed batches or materials, in order to minimize effects further downstream and reduce possible downtime of operation. A final quality check will be performed on the product before being sent to the packaging operation, where cGMP protocols will continue to be implemented, including proper clear and concise labeling and dosage information. Explosion-proof electrical equipment will be used in all steps involving solvents.

11.4 FDA Regulations for Contrast Agents

Since contrast agents are administered intravenously to systemic circulation, there are tight FDA regulations. There have been cases of cardiopulmonary reactions after injection, including acute myocardial infarction, acute coronary syndromes, worsening or decompensated heart failure, arrhythmias, respiratory failure, emphysema, pulmonary emboli, and other conditions causing pulmonary hypertension[53]. As a result, the FDA requires a “black box” warning (previously a contraindication) after 11 patient deaths between 2001 and 2007[54], advising patients of the risk for “severe cardiopulmonary reactions” and a blanket 30-minute monitoring period after the injection of the contrast agent for all patients with pulmonary hypertension and unstable cardiopulmonary conditions. These regulations apply to contrast agents sold as Definity® (Perflutren Lipid Microsphere) Injectable Suspension and Optison® (Perflutren Protein-Type a Microspheres for Injection)[54].

As Oleosin 30G is produced from a bacterial host, there are various requirements on the endotoxin levels in the product; failure to comply with these specifications can lead to “loss of product to market”. LifeASSURE™ PLA, which shows the highest level of endotoxin reduction,

can be used to meet the endotoxin challenge concentration of 0.25 EU/mL. This is generally a percentage reduction of 99.98%. The Limulus Amebocyte Lysate (LAL) gel tot method will be used to quantify the endotoxins present. The samples collected during endotoxin testing will meet the chemical requirements of the WFI Monographs in USP 23[55].

Lastly, Poloxamer 188 (P-188), also known as Pluronic F-68, will be used to make the microbubble suspensions. The FDA approved P-188 as a therapeutic agent to reduce viscosity in the blood before transfusions and has passed all required immunotoxicity tests[56].

11.5 Water for Injection (WFI)

When the WFI system is installed, the system will include the presence of a backflow valve which will serve to protect the source water. Sanitary clamped piping, valves and instruments will be constructed with 316L stainless steel will be used to transport water in the distribution and storage systems. Piping will be supported, labelled and sloped in order for the water to drain completely. The instruments will be maintained such that they will always pass integrity testing.

The temperature of the WFI distribution during peak load will be maintained at 80 ºC. In order to comply with regulations, daily microbiological monitoring will be conducted every day at the WFI still. For 20 working days, samples from various portions of the supply loop will be tested for compliance with the specifications. The tests will be completed over the course of a year to monitor for any changes over time, with sample sizes of 100- 300 ml per test[57].

Total heterotrophic plate count is a membrane filtration technique used to test the quality of the water that was described in Section 9215 of the Standard Methods for the Examination of Water and Wastewater. Plate Count Agar will be used to perform testing for microbiological agents present in the water. The colonies that are present on the filter will be identified and tested further for harmful effects. Though there are some deviations permitted for facilities that are located in “field environments,” because this is a lab facility, few microbial species should be present in the water systems. According to the government specifications, plate counts must be less than 10 CFU/ml for all samples[57].

Tests for residual chlorine, pH and conductivity are performed year round. Also, all media must pass through growth promotion testing.

11.6 Intellectual Property Concerns

When analyzing other aspects of the process required to successfully launch Oleosin 30G on the market, it is important to consider intellectual property concerns. It is assumed for the purposes of this project that a New Drug Application (NDA) would be submitted to the FDA during the approval process along with a patent filing for the Oleosin 30G formulation in question. With protection lasting 20 years under a US Patent Office patent holding, at the time of NDA approval exclusivity would also be granted[58]. Oleosin 30G would be classified as a New Chemical Exclusivity (NCE) with a five-year protection period, preventing any other competitors from filing an Abbreviated New Drug Application (ANDA) during this period. Records with the FDA must also be kept up to date in order to include the patent holding in the FDA's Orange Book[55]. This would ensure full protection and give extended exclusivity for the life of the patent.

11.7 Alternative Applications

Additionally, it is strategic to keep other potential applications of Oleosin in mind. This recombinant form of the protein can be used for other kinds of ultrasound imaging beyond echocardiograms. Alternative recombinant forms of Oleosin have promising applications in drug delivery and the food industry. Scale-up procedures may look similar to this process but require slight alterations.

12. Packaging and Shipping Specifications

12.1 Microfluidics, Packaging and Shipping Information

The final product of Oleosin in solution after the final bulk filtration will be sent to the microfluidics arm of operations for final packaging. Oleosin will be shipped in its final form to customers in sterile glass vials for injection, at a stable concentration of 1 mg/mL. Each vial will contain slightly more than the one dose, or 12.45 mg of Oleosin required per scan held in 10 mL aliquots, to account for administrator error.

Oleosin in buffer solution will first be transferred through sterile tubing using a pump to the microfluidic device. The device in question is a disposable mold made from a master copy, equipped to handle flow rates of solution up to 1 L/hr. This flow rate has been achieved in Dr. Daeyeon Lee's research at the University of Pennsylvania[59]. For realistic purposes, the maximum flow rate expected to be achieved in this process is set to 0.75 L/hr. Eight simultaneous disposable molds will be used, with the product evenly split between the eight devices. Two

master copies will be ordered for purchase, as each mold requires one day to set from the master. Using an estimate from Dr. Lee, the master copies will each be purchased for $2,000. The disposable molds will be purchased for $100 each, with the cost of molds coming to $8,000 per year, accounting for eight molds per each of 100 batches. Adding in $4,000 for purchasing the master copies, the cost of microfluidics equipment comes to $12,000 per year.

Oleosin bubbles will be formed using the devices mentioned above, where the final mass of Oleosin taken from downstream purification is suspended in buffer solution including a stable and nontoxic pluronic, F87. A stabilization time of 30 minutes is required to reach steady state bubble production, with another 30 minutes required for transfer of bubbles to stable storage at the end of production. Each mold requires 13.32 hours to produce a total of 5,934 10 mL glass vials of solution over the eight molds. These molds will be operated in parallel, overseen by two microfluidics operators. Molds will be disposed of between batches and the area properly sterilized to maintain sterile operating conditions. Each mold will have its own accompanying microscope in order to measure bubble consistency and validate uniformity upon reaching steady state.

The resulting 5,934 vials will be stored in standard tube racks at 4 ºC in a sterile refrigerator. Bubbles are stable up to one month in solution, but will be shipped to order within one week of production. UPS Pharmaceutical Overnight shipping will be used to ensure minimal bubble disturbance, with temperature stabilization and package immobilization aiding in safe and effective delivery of the product to its final destination for patient administration[60]. Using mass data from supplier Sigma Aldrich, sterile glass vials comprise 0.05 kg, with Oleosin and solution comprising 0.01 g. With each vial at a mass of 0.11 lbs, total batch weight comes to 654 lbs. At a standard shipping price of $22.95 per package up to 70 lbs, final associated shipping costs will be a function of orders received, dependent on weight required per shipment dispatched. Individual vials should be examined upon delivery to verify product viability.

13. Cost Summaries

13.1 Major Unit Operation Costs

Table 13.2.1. Major Unit Operation Costs

Type	Number	Units	Size	Vendor	Purchase Cost (per unit)
Bag Bioreactor Control Tower	P-1	1	0.2 L	Sartorius Stedim	$49,000
Large Peristaltic Pumps		2		Watson-Marlow	$25,000
Bioreactor	P-2	1	19.5L	Eppendorf	$104,000
Disk Stack Centrifuge	P-4	1		Alfa Laval	$46,500
High Pressure Homogenizer	P-5	1		Microfluidics	$8,000
Wave Bag Rocker	P-1	2	20L	Sartorius Stedim	$9,300
Chromatography Column	P-7	1	21L	Pall Corporation	$190,000
Small Peristaltic Pumps		20		Cole-Parmer	$2,000
UFDF Base Unit	P-9	2		EMD Millipore	$1,000
Microfluidic Master Device		8			$5,000
Waste Tank		1	2,000L	Sharpsville Container	$500

The total purchase cost for major unit operation equipment is $548,600. With a bare module factor of 3.21, the bare module cost for major unit operation equipment is $1,760,000. The most expensive units are the stainless steel 19.5L bioreactor and the 21L chromatography column. The 20 purchased small peristaltic pumps include four extra pumps to replace any damaged pumps in order to keep the process moving while new pumps are purchased.

13.2. Additional Equipment Costs

Table 13.2.1. Additional Equipment Purchase Cost

Type	Units	Size	Vendor	Purchase Cost (total)
UV-Vis Spectrophotometer	1		Nanodrop	$9,100
MALDI-TOF Mass Spectrometer	1		Shimadzu	$288,000
Clean Steam Generator	1		BMT USA	$60,000
Refrigerator (4°C)	4	556L	Thermo Scientific	$3,400
Freezer (-20°C)	2	481L	Thermo Scientific	$8,700
Freezer (-80°C)	1		Thermo Scientific	$30,000
Control Units (PID)	20		Omega	$300
Water for Injection Supplier	1		Paul Mueller Co.	$20,000
Filter Integrity Tester	1		EMD Millipore	$3,500
Clean Air Generator	1		Domnick Hunter	$2,500
Incubator Shaker	1		Sheldon Manufacturing, Inc.	$4,500
Microplate Reader	1		Tecan Group	$3,500
Buffer Storage Bags	6	500L	HyClone	$22,500

The total purchase cost for additional equipment is $593,000. With a bare module factor of 3.21, the bare module cost for major unit operation equipment is $1,904,000. The most expensive unit is the MALDI-TOF mass spectrometer. The 20 purchased control units include extra to replace any damaged units in order to keep the process moving while new units are purchased.

13.3 Materials and Reagent Costs

Table 13.3.1. Materials and Reagent Costs.

Type	Units	Required Ratio		Cost of Raw Material:	
Media	L	1.62E-04	L per mg of Oleosin 30G	$100	per L
IPTG	Kg	3.35E-09	kg per mg of Oleosin 30G	$54200	per kg
Resin	mL	0.007	mL per mg of Oleosin 30G	$5.68	per mL
Urea	kg	4.09E-03	kg per mg of Oleosin 30G	$63.60	per kg
B-PER	L	4.69E-06	L per mg of Oleosin 30G	$590	per L
DNase	mg	4.69E-06	mg per mg of Oleosin 30G	$143000	per mg
P-188	kg	8.03E-04	kg per mg of Oleosin 30G	$88.50	per kg
Imidazole	kg	3.29E-05	kg per mg of Oleosin 30G	$298.80	per kg
MES Buffer	kg	1.09E-05	kg per mg of Oleosin 30G	$324.80	per kg
Na_3PO_4	kg	2.79E-05	kg per mg of Oleosin 30G	$67.80	per kg

The purchase cost of raw materials per mg of Oleosin 30G is approximately $0.41. This cost estimate was determined from price quotes provided by vendors and consultants.

LB media[61] with kanamycin for growth of *E. coli* strain BL21 DE3 in a required ratio of 1.62E-04 L per mg of Oleosin 30G is purchased at a cost of $100 per L. LB media is a typical growth media for *E. coli* strains and kanamycin is an antibiotic that is used to prevent contamination of the culture from other bacterial strains present in the environment. IPTG (Isopropyl β-D-1-thiogalactopyranoside)[62] is purchased for induction of the protein production in the bacterial culture at a required ratio of 3.35E-09 at $54200 per kg. The compound is a mimic of allolactose that triggers the transcription of the lac operon. Both inoculation prep raw materials are purchased from Sigma Aldrich.

Following Oleosin production, the protein must be purified using immobilized metal affinity chromatography (IMAC). For histidine tag purification, either nickel or cobalt is immobilized onto a solid chromatography resin. Resin[63] for cobalt chromatography is purchased from Aquarion Water at a ratio of 0.007 mL per mg of Oleosin 30G a cost of $5.680 per mL.

IMAC resins work by sparking charge interactions with the nitrogen atoms on the histidine amino acid side chain to bind and immobilize the histidine-tagged protein from a cell lysate.

The resin is washed with denaturing wash buffer which consists of urea, imidazole, sodium chloride and sodium phosphate. Urea[64], the denaturing agent, is purchased at a required ratio of 4.09E-03 kg per mg of Oleosin 30G at a cost of $63.60 per kg. The imidazole[68] is purchased at a ratio of 3.3E-04 kg per mg Oleosin 30G at a cost of $298.00 per kg. In cobalt chromatography, the imidazole is used to disrupt the charge attractions between the immobilized metal affinity chromatography resin and the histidine- tagged protein. Also, trace amounts are included in the protein binding and wash steps to prevent binding of multiple histidines. Na_3PO_4 is purchased at a ratio of 2.79E-05 kg per mg Oleosin 30G at a cost of $67.80 per kg[69]. Finally, B-Per protein extraction reagent is also purchased to isolate the protein at a ratio of 4.69E-06 L per mg of Oleosin 30G at a cost of $590.00 per L[65].

13.4 Utilities Requirements

The cost of utilities per mg of Oleosin 30G is approximately $0.03 per mg of Oleosin 30G. This utility calculation includes low pressure steam, water for injection, electricity and refrigeration costs.

Low pressure steam is used in the process plant to sterilize equipment. The steam is used to kill any live mass remaining in the bio-waste tank; cultures are heated to 121°C for 20 minutes and then cooled to 80°C. The steam is priced at $0.013 per kg and the process requires 1.00E-04 kg per mg of Oleosin 30G[69].

Because the facility is built to be pharmaceutical grade, water for injection (WFI) must be for all production processes. As defined by the FDA, WFI is high-quality water, used for the production of parenterals and water-based ophthalmic products. Water for Injection will be purchased in bulk from Sigma Aldrich at a rate of $7.120 per L, with 0.013 L of water per mg of Oleosin 30G. For more information on the FDA requirements for WFI, refer to Section 14. The costs for Water for Injection constitute the most significant component of the utility costs at 77.5%.

Refrigeration costs for the facility are largely derived from costs of running the “cold room.” This cold room (9 by 12 feet) is maintained at 4 ºC during all protein purification processes. Heat load calculations were based on heat conduction through insulated walls, ceiling

and floor. R-values for wall/ceiling materials were provided by North Carolina University for general cold room facility floor and ceiling materials. Service loads were also accounted for in the refrigeration costs; these loads are derived from lights, equipment, people and most air entering through the door or cracks and account for 10 percent of the heat conduction. As determined by these parameters, the total heat load is 733 Btu/hr. Per mg of Oleosin 30G, this refrigeration cost is 0.001 KWh. The utility costs in Boston, MA, the location of the production facility, is $0.20 per KWh per mg of Oleosin 30G. Costs of the four storage refrigerators were assumed to be negligible in comparison[70].

Electricity costs were based on values in an article titled "Energy Benchmarking in the Pharmaceutical Industry." This costing was applied to the required kW-hr necessary for the process.

Table 13.4.1. Summary of requirements and associated costs for utilities.

Utility	**Unit**	**Required Ratio**		**Utility Cost**	
Low Pressure Steam	kg	1.00E-04	kg per mg of Oleosin 30G	$0.013	per kg
Water for Injection	L	0.013	L per mg of Oleosin 30G	$7.12	per L
Electricity	kWh	3.00E-01	kWh per mg of Oleosin 30G	$0.07	per kWh
Refrigeration	KWh	0.0013	KWh per mg of Oleosin 30G	$0.20	per KWh
	Total Weighted Average:			$0.12	per mg of Oleosin 30G

14. Economic Analysis

14.1 Market Analysis

The set selling price and market share in our profitability analysis were based on the known market share, revenue, and dose price data for the current echocardiography contrast agent Definity® from 2010 to 2016[71]. Our price/mg in our first year sold is set to be equal to the current price of Definity®. In our listed selling price data there is a 75% multiplier to account for shipping costs, as we assume 25% of the revenue for our product will be lost to shipping. The increase in selling price per year mirrors that of Definity®, which matched Optison®'s price until

gaining the majority of the market increasing 5% per year, then increasing 10% per year after cornering the market[72]. Since no data is available after 2016 for pricing, we assumed a conservative 5% price increase per year.

Years Sold	Selling Price/mg	Year Over year Increase	Year (Definity)	Definity Price/Dose	Year over Year Increase	Market Share
1	$11.92	0.00%	2010	$125.00	0.00%	63%
2	$12.51	4.97%	2011	$130.00	4.00%	69.82%
3	$13.14	5.00%	2012	$136.00	4.62%	70.39%
4	$13.80	5.00%	2013	$142.75	4.96%	77.56%
5	$15.17	10.00%	2014	$157.00	9.98%	87.47%
6	$16.69	10.00%	2015	$172.75	10.03%	91.05%
7	$17.53	5.00%	2016	$198.50	14.91%	unknown
8	$18.40	5.00%				
9	$19.32	5.00%				
10	$20.29	5.00%				
11	$21.30	5.00%				
12	$22.37	5.00%				
13	$23.49	5.00%				
14	$24.66	5.00%				
15	$25.90	5.00%				

Figure 14.1.1 Selling price and yearly increases of Definity® market share. The trend of increasing market share at a higher selling point can be seen for Definity®, the strongest competitor currently on the market.[1]

The number of echocardiograms that incorporated contrast agents has remained relatively constant between 2010 and 2015, with 700,000 injections per year in 2010 and 600,000 injections per year in 2015[73]. We use a conservative estimate of a market of 600,000 doses per year, although there is a chance for increase in this market, as contrast agents are used in less than 10% of echocardiograms, but shown to reduce the overall cost per patient. With proper marketing, the market can further expand into the larger echocardiogram market, rather than the contrast enhanced echocardiogram market. As shown in Scheduling, we are currently producing three batches per week, which can be easily increased to a maximum 5 batches per week without any equipment overlap. This allows for flexibility in the production rate if the market changes.

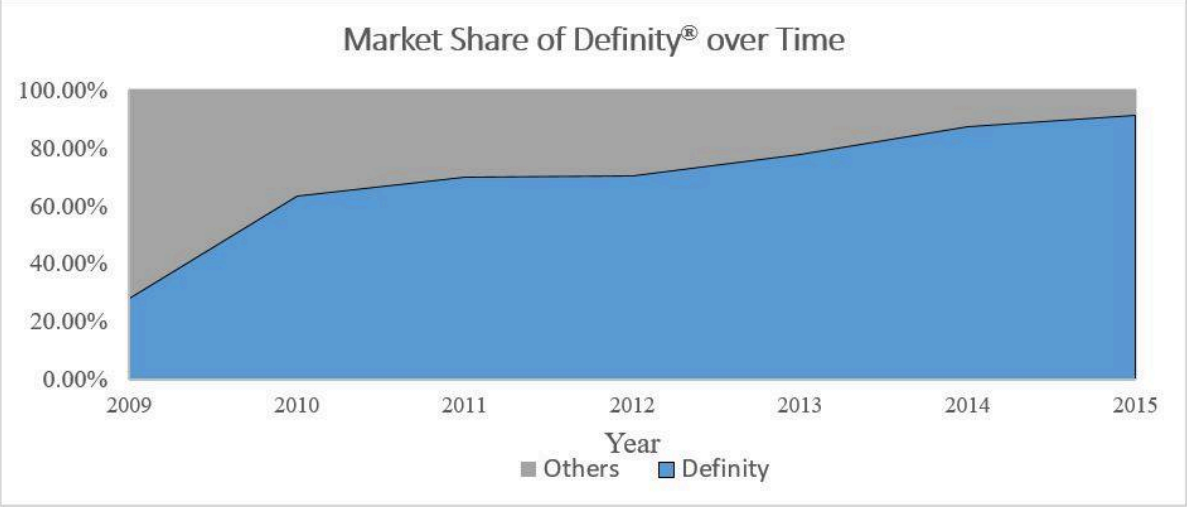


Figure 14.1.2. Market share of Definity® over time. The steady increase in Definity®'s market share can be seen from 2009 through 2015, starting from under 40% and growing to nearly 100% last year.[2]

The expected market share of our product also mirrors that of Definity® after 2010. During 2001 to 2009, Definity® received a black box warning from the FDA[74], which severely limited its adoption, which is shown below. In late 2009, it was shown that the fatalities due to Definity® were so few as to not be statistically significant, and removed the warning label. Without the warning label, market share quickly increased to 63% in 2010, then steadily increased to about 90% in 2014. We assume that our product will have no serious adverse reactions upon administration, so our expected market share mirrors that of Definity® starting in 2010, with 3 years to reach a steady market share of 90%, which is 90% of the plants current production capacity at 3 batches per week[75].

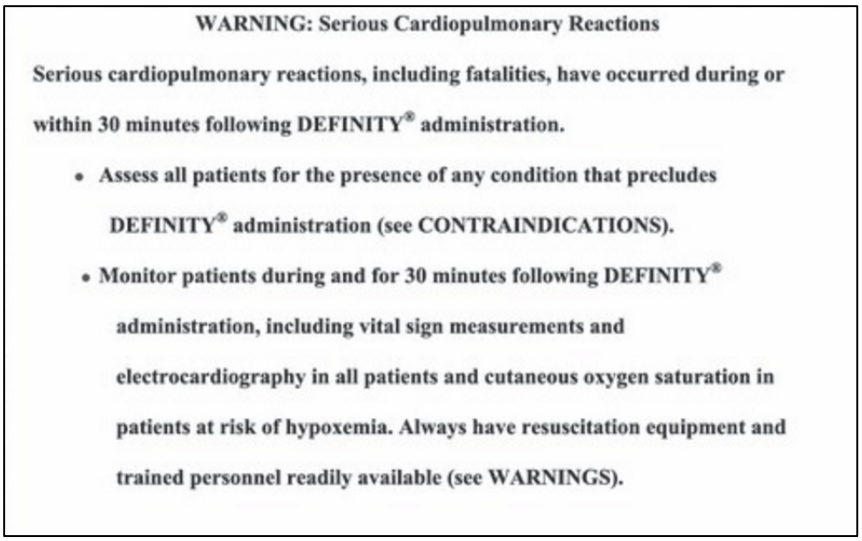


Figure 14.1.3. Black box warning for Definity®. Due to potential cardiopulmonary risks, Definity® has been mandated by the FDA to include a black box warning on their product.[3]

14.2 Profitability Analysis

The process designed for scaled up production of Oleosin 30G for application as a contrast agent for ultrasound poses great promise as a profitable venture. The market is ready for the addition of a new agent with consistent, uniform bubble size and promising revolutionary aspects like variability of bubbles for therapeutic area.

The Internal Rate of Return (IRR) of the plant is 77.45% with a Net Present Value of $214,124,800.00. The Return on Investment (ROI) is 78.57%, coming from a sales figure of $75,731,510.00 annually. This figure is based on the production of 10 kilograms of Oleosin product per year, corresponding to a 100% market saturation of current echocardiograms with use of a contrast agent. Further market growth predicts an increasing trend in the number of scans utilizing contrast agents and therefore Oleosin is posed to reap even higher sales than this figure in years down the road.

The cost of development, or FDA approval process including clinical trials, was included in the analysis of this plant's profitability. The high cost associated with this approval process is standard in the pharmaceutical industry, as a company must make back losses associated with other projects in the pipeline. It is estimated that, based on preexisting proven efficacy for similar contrast agents, the bulk of clinical trial focus will be on toxicity in Phase I. This corresponds to an estimated timeframe of two to five years between preclinical studies and FDA approval. While the cost associated with these trials is included in the profitability analysis, this timeframe is considered out of scope, where the facility discussed will be constructed and fully operational

as of the day FDA approval is given. The first full year of operations is considered to be the first year that the facility reaches 100% of its production capacity.

Profitability Measures

The Internal Rate of Return (IRR) for this project is	72.43%
The Net Present Value (NPV) of this project in 2016 is	$ 201,670,700

ROI Analysis (Third Production Year)

Annual Sales	**75,731,510**
Annual Costs	**(25,438,087)**
Depreciation	**(2,628,491)**
Income Tax	**(17,636,025)**
Net Earnings	**30,028,907**
Total Capital Investment	**41,511,886**
ROI	**72.34%**

Figure 14.2.1 Profitability Measures for the process. Using the profitability analysis spreadsheet to generate metrics for the process, an Internal Rate of Return (IRR), Net Present Value (NPV), and Return on Investment (ROI) were calculated. Annual sales, costs, and resulting total capital investment required based on equipment purchases and installations were tabulated.

14.2.1 Price of Oleosin 30G

The first year's selling price of Oleosin 30G is set at $11.92 per mg. This is competitively priced with those currently on the market, Definity® and Optison®. The selling price is set to be increased by 5% over each of the first three years of operation, rising to $13.80 per mg, with a 10% increase setting in the fourth year up, and returning to a 5% increase in the sixth year of production. This is to account for natural market growth during this period, with the number of scans performed both with and without contrast agents growing at a consistent pace. Agent prices are appropriately raised to adjust for this growth. This growth trend will correspond to a doubling in selling price by the year 2030.

14.2.2 Facility Lifespan

The plant has an operational life expectancy of about 15 years. This anticipated lifetime accounts for the lifespan of purchased equipment, the market's receptiveness to Oleosin as a contrast agent for ultrasounds, and the value of the facility itself. If the facility were to shut down before the end of this 15-year timeline, there could be some remaining salvage value in the plant; however, it is considered to be most profitable for the facility to maintain a 15-year occupancy at full production capacity.

14.2.3 Input Costs and Summaries

Variable Cost Summary

Variable Costs at 100% Capacity:

General Expenses

Selling / Transfer Expenses:		$	2,641,950
Direct Research:		$	4,227,121
Allocated Research:		$	440,325
Administrative Expense:		$	1,761,300
Management Incentive Compensation:		$	1,100,813
Total General Expenses		$	10,171,509
Raw Materials	$0.472792 per mg of Oleosin 30G		$3,493,930
Byproducts	$0.000000 per mg of Oleosin 30G		$0
Utilities	$0.027462 per mg of Oleosin 30G		$202,947
Total Variable Costs		**$**	**13,868,386**

Fixed Cost Summary

Operations

Direct Wages and Benefits	$	4,571,008
Direct Salaries and Benefits	$	685,651
Operating Supplies and Services	$	274,260
Technical Assistance to Manufacturing	$	-
Control Laboratory	$	-
Total Operations	**$**	**5,530,920**

Maintenance

Wages and Benefits	$	1,320,113
Salaries and Benefits	$	330,028
Materials and Services	$	1,320,113
Maintenance Overhead	$	66,006
Total Maintenance	**$**	**3,036,259**

Operating Overhead		
General Plant Overhead:	$	490,383
Mechanical Department Services:	$	165,763
Employee Relations Department:	$	407,501
Business Services:	$	511,103
Total Operating Overhead	**$**	**1,574,750**
Property Taxes and Insurance		
Property Taxes and Insurance:	$	586,717
Other Annual Expenses		
Rental Fees (Office and Laboratory Space):	$	3,550,000
Licensing Fees:	$	-
Miscellaneous:	$	342,100
Total Other Annual Expenses	**$**	**3,892,100**
Total Fixed Costs	**$**	**14,620,746**

Investment Summary

Installed Equipment Costs:			
Total Direct Materials and Labor Costs	$	100,500	
Miscellaneous Installation Costs	$	2,500,300	
Material and Labor G&A Overhead and Contractor Fees	$	-	
Contractor Engineering Costs	$	-	
Indirect Costs	$	20,000,000	
Total:			**$ 22,600,800**

Direct Permanent Investment

Cost of Site Preparations:	$	1,130,040	
Cost of Service Facilities:	$	1,130,040	
Allocated Costs for utility plants and related facilities:	$	-	
Direct Permanent Investment			**$ 24,860,880**

Total Depreciable Capital

Cost of Contingencies & Contractor Fees	$	4,474,958	
Total Depreciable Capital			**$ 29,335,838**

Total Permanent Investment

Cost of Land:	$	586,717	
Cost of Royalties:	$	-	
Cost of Plant Start-Up:	$	2,933,584	
Total Permanent Investment - Unadjusted			$ 32,856,139
Site Factor			1.00
Total Permanent Investment			**$ 32,856,139**

Working Capital

	2017	2018	2019
Accounts Receivable	$ 3,908,639	$ 868,586	$ 868,586
Cash Reserves	$ 485,183	$ 107,818	$ 107,818
Accounts Payable	$ (164,081)	$ (36,462)	$ (36,462)
Oleosin 30G Inventory	$ 912,016	$ 202,670	$ 202,670
Raw Materials	$ 51,691	$ 11,487	$ 11,487
Total	**$ 5,193,448**	**$ 1,154,100**	**$ 1,154,100**
Present Value at 15%	*$ 4,516,042*	*$ 872,665*	*$ 758,839*

Total Capital Investment **$ 39,003,685**

Figure 14.2.3.1 Summary of variable costs for the process. Based on metrics preset in the profitability analysis spreadsheet, raw materials and utilities figures were used to determine the variable costs per year.

14.2.4 Fixed Cost Summary

The plant will be run 16 hours per day, 5 days per week for a total of 100 batches per year. This amounts to 191 days per year of operation, with downtime scheduled for necessary maintenance, repairs, and scheduled upkeep of the facility. Eight operators will be scheduled to work on each shift: two on upstream cell culture and bioreactor management, two on downstream separations and purifications, two in the lab handling quality assurance and validation testing, and two in the final packaging and shipping wing of the plant to handle the microfluidic devices to produce final bubble suspensions. Operators will be paid direct wages and benefits equaling $27 per operator hour.

Recurring annual operating expenses also include the purchase of disposable equipment not included in the capital investment analysis. Among these are a clean steam generator, Water for Injection (WFI) generator, disposable storage bags and filters, and CIP and SIP supplies.

Fixed Costs

Operations

Operators per Shift:	8	(assuming 10
Direct Wages and Benefits:	$27	/operator hour
Direct Salaries and Benefits:	15%	of Direct Wages and Be
Operating Supplies and Services:	6%	of Direct Wages and Be
Technical Assistance to Manufacturing:		per year, for each Opera
Control Laboratory:		per year, for each Opera

Maintenance

Wages and Benefits:	4.50%	of Total Depreciable Ca
Salaries and Benefits:	25.00%	of Maintenance Wages
Materials and Services:	100.00%	of Maintenance Wages
Maintenance Overhead:	5.00%	of Maintenance Wages

Operating Overhead

General Plant Overhead:	7.10%	of Maintenance and Ope
Mechanical Department Services:	2.40%	of Maintenance and Ope
Employee Relations Department	5.90%	of Maintenance and Ope
Business Services	7.40%	of Maintenance and Ope

Property Taxes and Insurance

Property Taxes and Insurance:	2.00%	of Total Depreciable Ca

Straight Line Depreciation

Direct Plant:	8.00%	of Total Depreciable Capital, less		1.18
Allocated Plant:	6.00%	of	1.18	times the Allocated Cost

Other Annual Expenses

Rental Fees (Office and Laboratory Space):	$3,550,000
Licensing Fees:	$0
Miscellaneous:	$342,100

Depletion Allowance

Annual Depletion Allowance:	$0

Figure 14.2.4.1 Fixed cost analysis. Ooperations and maintenance figures were determined to be factored into fixed costs per year, with operating overhead and property taxes and insurance factoring in as well.

14.2.5 Other Variable Costs and Working Capital

It was assumed that the cost of sales and other transfer expenses would be approximately 3% of annual sales, with seven days of inventory kept in stock. While the final product is stable up to one month in solution, in order to ensure full efficacy and reliability of the product it is

assured that product will be shipped within one week of packaging. Ten days of raw material inventory will be kept onsite, with regular material purchases made in accordance with supply and demand dynamics.

Other Variable Costs

General Expenses

Selling / Transfer Expenses:	3.00% of Sales
Direct Research:	4.80% of Sales
Allocated Research:	0.50% of Sales
Administrative Expense:	2.00% of Sales
Management Incentive Compensation:	1.25% of Sales

Working Capital

Accounts Receivable	⇨	**30**
Cash Reserves (excluding Raw Materials)	⇨	**30**
Accounts Payable	⇨	**30**
Oleosin 30G Inventory	⇨	**7**
Raw Materials	⇨	**10**

Figure 14.2.5.1 Other variable costs and working capital. Using a one week inventory of final product and 10 day inventory of raw materials, working capital and general expenses were set with appropriate scaling factors.

14.2.6 Cost of FDA Approval

As mentioned above, the cost of clinical trials and FDA approval was included in the profitability analysis for the process. Per a quote from Dr. Scott Diamond (February 23, 2016, Chemical and Biomolecular Engineering, University of Pennsylvania) an estimate of $20,000,000 was used to calculate profitability measures. This cost associated with development is included in miscellaneous operating costs and is a one-time required cost upon startup. In a realistic industry setting, this high price point is appropriate when considering the entire pipeline of a pharmaceutical company. The return in sales must be high enough not only to recover this initial development investment, but to also recover losses from the many other unsuccessful products in the pipeline.

14.3 Sensitivity Analysis

To examine key factors in the process' profitability measures, a sensitivity analysis was performed to determine important variables in the plant's success. Product price, plant capacity, and lifespan of the facility were studied to determine impact on final metrics.

14.3.1 Product Price

Product price was studied in relation to variable costs to determine the effect on Internal Rate of Return (IRR). Product price was varied in increments of plus or minus 50% of the set product price value. Based on the initial price point set of $11.92/mg Oleosin, this price is equivalent to Definity®'s current price point. Therefore, Oleosin poses great opportunity to price at a higher point after initial market success. Because of slight inertia in within the industry and resistance to change, it is required to initially price equal to Definity® to gain market share before raising the price once superior efficacy is established.

Product prices studied vary up to $17.88/mg Oleosin, at which point if variable cost is held constant, the IRR of the plant increases from 72.43% to 107.48%, a sizable change. The change in variable cost does not seem to have a large impact on IRR across similar product prices and therefore is determined not to be a key factor in plant profitability.

Product Price	Variable Costs										
	$6,934,193	$8,321,032	$9,707,870	$11,094,709	$12,481,548	***$13,868,386***	$15,255,225	$16,642,063	$18,028,902	$19,415,741	$20,802,579
$5.96	37.68%	36.42%	35.17%	33.93%	32.68%	31.44%	30.21%	28.98%	27.75%	26.52%	25.30%
$7.15	46.60%	45.37%	44.15%	42.93%	41.72%	40.51%	39.31%	38.11%	36.91%	35.72%	34.53%
$8.34	55.03%	53.82%	52.62%	51.42%	50.22%	49.03%	47.84%	46.65%	45.47%	44.30%	43.13%
$9.53	63.10%	61.91%	60.71%	59.53%	58.34%	57.16%	55.98%	54.80%	53.63%	52.47%	51.30%
$10.73	70.89%	69.70%	68.52%	67.34%	66.17%	64.99%	63.82%	62.66%	61.49%	60.33%	59.18%
$11.92	78.43%	77.25%	76.08%	74.91%	73.74%	72.58%	71.42%	70.26%	69.10%	67.95%	66.80%
$13.11	85.75%	84.59%	83.43%	82.27%	81.11%	79.95%	78.80%	77.65%	76.50%	75.35%	74.21%
$14.30	92.88%	91.73%	90.58%	89.43%	88.28%	87.13%	85.98%	84.84%	83.70%	82.56%	81.42%
$15.49	99.84%	98.69%	97.55%	96.41%	95.27%	94.13%	92.99%	91.86%	90.72%	89.59%	88.46%
$16.68	106.63%	105.49%	104.36%	103.22%	102.09%	100.96%	99.83%	98.71%	97.58%	96.46%	95.34%
$17.88	113.26%	112.13%	111.01%	109.89%	108.76%	107.64%	106.52%	105.40%	104.29%	103.17%	102.06%

Figure 14.3.1.1 IRR as a function of product selling price and variable costs. IRR was calculated for a range of product prices at plus or minus 50% increments above and below the set point. Variable costs were appropriately scaled based on a 3% sales factor.

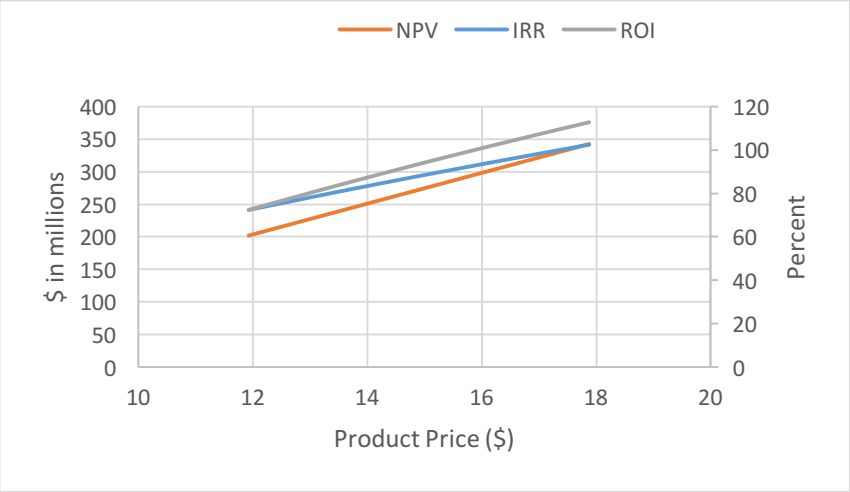


Figure 14.3.1.2 IRR, NPV, and ROI sensitivity to product price. A sensitivity analysis was done on IRR, NPV, and ROI where product price was increased up to 200% of the set point. The red series represent NPV in millions of dollars, with the gray and blue series representing percentages of IRR and ROI.

Cash Flow Summary

Year	Percentage of Design Capacity	Product Unit Price	Sales	Capital Costs	Working Capital	Var Costs	Fixed Costs	Depreciation	Depletion Allowance	Taxible Income	Taxes	Net Earnings	Cash Flow	Cumulative Net Present Value at 15%
2016	0%		-	-	-	-	-	-	-	-	-	-	-	-
2017	0%		-	(32,856,100)	(5,188,300)	-	-	-	-	-	-	-	(38,044,500)	(33,082,200)
2018	54%	$11.92	47,555,100	-	(1,153,000)	(7,488,900)	(14,505,600)	(5,867,200)	-	19,693,400	(7,286,600)	12,406,800	17,121,000	(20,136,200)
2019	66%	$12.51	61,029,100	-	(1,153,000)	(9,153,100)	(14,505,600)	(9,387,500)	-	27,982,800	(10,353,700)	17,629,200	25,863,700	(3,130,400)
2020	78%	$13.14	75,731,500	-	(1,153,000)	(10,817,300)	(14,505,600)	(5,632,500)	-	44,776,100	(16,567,100)	28,208,900	32,688,400	15,559,300
2021	90%	$13.80	91,751,600	-	-	(12,481,500)	(14,505,600)	(3,379,500)	-	61,385,000	(22,712,400)	38,672,500	42,052,000	36,466,600
2022	90%	$15.17	100,926,800	-	-	(12,481,500)	(14,505,600)	(3,379,500)	-	70,560,200	(26,107,300)	44,452,900	47,832,400	57,145,900
2023	90%	$16.69	111,019,500	-	-	(12,481,500)	(14,505,600)	(1,689,700)	-	82,342,600	(30,466,800)	51,875,800	53,565,600	77,283,200
2024	90%	$17.53	116,570,500	-	-	(12,481,500)	(14,505,600)	-	-	89,583,300	(33,145,800)	56,437,500	56,437,500	95,732,700
2025	90%	$18.40	122,399,000	-	-	(12,481,500)	(14,505,600)	-	-	95,411,800	(35,302,400)	60,109,400	60,109,400	112,819,500
2026	90%	$19.32	128,518,900	-	-	(12,481,500)	(14,505,600)	-	-	101,531,800	(37,566,800)	63,965,000	63,965,000	128,630,700
2027	90%	$20.29	134,944,900	-	-	(12,481,500)	(14,505,600)	-	-	107,957,700	(39,944,400)	68,013,400	68,013,400	143,249,700
2028	90%	$21.30	141,692,100	-	-	(12,481,500)	(14,505,600)	-	-	114,705,000	(42,440,800)	72,264,100	72,264,100	156,756,400
2029	90%	$22.37	148,776,700	-	-	(12,481,500)	(14,505,600)	-	-	121,789,600	(45,062,100)	76,727,400	76,727,400	169,226,800
2030	90%	$23.49	156,215,600	-	-	(12,481,500)	(14,505,600)	-	-	129,228,400	(47,814,500)	81,413,900	81,413,900	180,732,900
2031	90%	$24.66	164,026,300	-	-	(12,481,500)	(14,505,600)	-	-	137,039,200	(50,704,500)	86,334,700	86,334,700	191,342,900
2032	90%	$25.90	172,227,700	-	8,647,200	(12,481,500)	(14,505,600)	-	-	145,240,500	(53,739,000)	91,501,500	100,148,700	202,045,300

Figure 14.3.1.3. Cash flow summaries for 15 year forecast. Cash flow figures including sales, working capital, variable and fixed costs, and depreciation were calculated for a 15 year forecasting from the start of operation. It can be seen that the process becomes profitable in the third year of operation.

14.3.2 Operating Capacity

Initially, the production capacity was set to 90% of design capacity to account for issues such as failed batches, unexpected maintenance requirements, and regulatory checks. If production capacity is increased to 95% of design capacity, corresponding IRR increases from 72.34% to 75.77%. Return on Investment (ROI) increases from 72.43% to 76.92% with a Net Present Value (NPV) of $216,701,900. While this is a slight increase in profitability from a process-wide perspective, it is important to realize the significance of increasing production capacity by an extra 5%. It is possible that this increase is not realistically achievable in the face of unexpected problems in operation, and therefore should not be treated as a key metric in profitability.

Profitability Measures

The Internal Rate of Return (IRR) for this project is	75.77%
The Net Present Value (NPV) of this project in 2016 is	$ 216,701,900

ROI Analysis (Third Production Year)

Annual Sales	79,938,816
Annual Costs	(26,039,051)
Depreciation	(2,628,491)
Income Tax	(18,970,371)
Net Earnings	32,300,903
Total Capital Investment	41,992,761
ROI	76.92%

Figure 14.3.2.1 Profitability measures for increased operating capacity. A sensitivity analysis was conducted to see the effect of increasing operating capacity from 90% to 95%. Very small impact was seen on the profitability metrics. In reality, this increase in capacity is most likely unfeasible due to practical constraints.

14.3.3 Operator Wages

A large portion of operating expenses is devoted to labor and associated costs. An analysis was performed to see effect on IRR, NPV, and ROI given an increase in direct salary per operator hour. Values were determined for wages at $20, $27, $35, $40, and $50. As expected, all three metrics dramatically decline with increasing operator wages. For the purposes of the plant's profitability, wages were set at $27 per operator hour, corresponding to the IRR, NPV, and ROI values mentioned above.

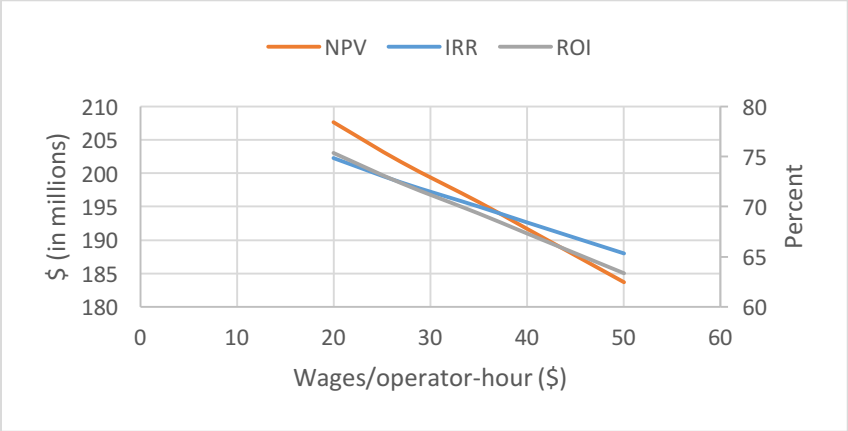


Figure 14.3.3.1. IRR, NPV, and ROI sensitivity to operator wages. A sensitivity analysis was done on IRR, NPV, and ROI where operator wages were increased up to 200% of the set point. The red series represent NPV in millions of dollars, with the gray and blue series representing percentages of IRR and ROI.

14.3.4 Number of Doses Produced

While the original process is set to produce 7.39 kg of Oleosin 30G annually, there is room to grow in the contrast agent market as market dynamics will allow. Sensitivity was determined using the number of doses produced as a key factor in profitability metrics. IRR, NPV, and ROI all saw striking increases as production increased, from the starting point of 7.39 kg representing a market saturation of 100% and 593,000 doses up to the maximum capacity of the current plant schedule. This maximum capacity takes operator scheduling and a five-day-per-week schedule into account, resulting in 1.088 million doses of Oleosin 30G, nearly a doubling of the current opportunity. With an IRR of 118%, NPV of $427,951,000, and ROI of 133.55%, this maximum capacity production shows great promise if growth continues.

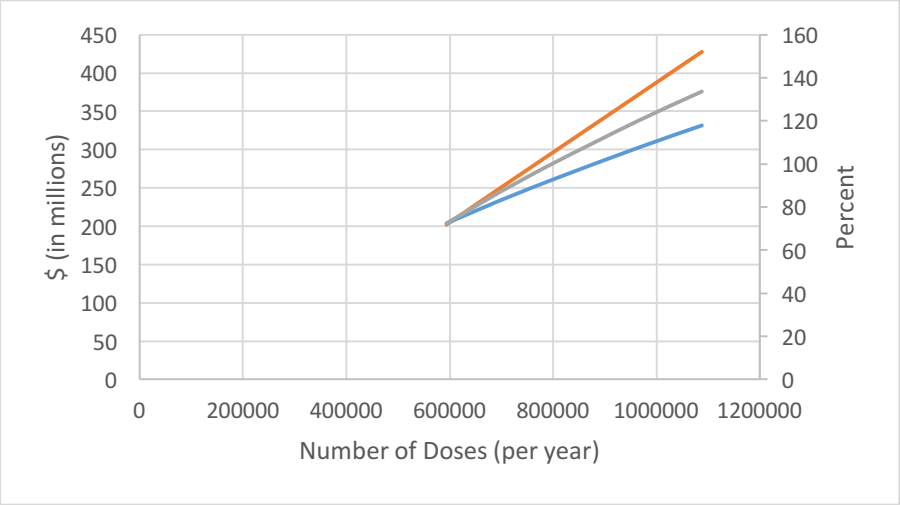


Figure 14.3.4.1. IRR, NPV, and ROI sensitivity to number of doses. A sensitivity analysis was done on IRR, NPV, and ROI where number of doses was increased up to 200% of the set point. The red series represent NPV in millions of dollars, with the gray and blue series representing percentages of IRR and ROI.

15. Conclusions and Recommendations

Ultimately it has been determined that Oleocor should maintain a manufacturing capacity of 7.39 kg of Oleosin 30G per year to accommodate current market needs. This figure accounts for an overall process percent yield of 46.1%. This profitable production goal represents a 100% market saturation of current echocardiograms performed with contrast agents. Additionally, a sensitivity analysis shows that production remains profitable under a variety of conditions. Based on analysis of Definity's controlling stake in the market with continued, increased market share over the inferior product Optison, there looks to be a significant opportunity for Oleocor's superior product to enter the market and gain over 90% market share. There is also a sizable opportunity for Oleosin 30G-stabilized microbubbles to claim a stake in alternative markets, both within echocardiography and in other forms of ultrasound. As outlined in this report, the designed plant can accommodate significant growth in production.

This opportunity is supported by the extremely attractive qualities Oleosin 30G possesses as a surfactant in a contrast agent. With the assured production of uniformly sized, monodisperse microbubbles in solution with concentration of 1 mg/mL, individual doses administered through IV injection have the potential to greatly increase resolution and efficacy of scans by stabilizing microbubbles at a variety of resonant sizes. With bubble stability holding for over one month after final suspension, Oleosin 30G can readily accommodate custom echocardiography needs all over the country. The facility should take on the nuanced task of microfluidic assembly to ensure ease of use for all consumers to guarantee the most comprehensive administration as possible on site.

While initial clinical testing must be done to confirm safety and efficacy of the product as well as acquire FDA approval to enter the market, due to a favorable profitability analysis it is highly recommended that Oleocor pursue this opportunity to capitalize on a significant opening in the market.

16. Acknowledgements

We would like to acknowledge our advisors, Dr. Hammer and Dr. Wattenbarger for their input throughout the course of this project. We would like to thank Professor Fabiano for his guidance this semester. We would also like to express our sincere gratitude to Dr. Huff for her support and help during our two-day fermentation experiment. We extend thanks to Dr. Lee for his advice concerning microfluidics and Dr. Diamond for his help with understanding the FDA approval process. We could not have successfully scaled up our process from the lab protocol without the input and assistance of Chen Gao, and we are extremely appreciative of her patience and direction throughout the course of the project.

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All of the feedback and help we have received from our advisors, professors, and consultants has been invaluable to our project and to our understanding of what it takes to be a bioprocess engineer. We truly appreciate all of the time you have all devoted to our group and the impact you have made on our final project. We thank each and every one of you from the bottom of our hearts.

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Appendix A: Bioreactor Reactor Analysis

Appendix A.1: Pilot Plant Cell Growth Study

To support cell growth calculations and oxygen consumption, a pilot plant study was conducted in the Chemical Engineering Laboratory at The University of Pennsylvania. Recombinant *E. coli* generated by the Hammer lab were obtained and used to run a 5L bioreactor with 3L of working volume of LB Miller media with 1 g/L glucose and 50 μg/mL kanamycin. The goal of the experiment was to obtain a more accurate oxygen consumption and pH profile as well as a cell growth curve for the Oleosin-producing, recombinant *E. coli* cell line. All goals were met and this data was used to inform growth calculations in this report as well as to set the most ideal bioreactor conditions for optimal growth.

Pilot Plant Study Procedure:

1. Inoculate two sterile 50mL flasks (containing 30mL of LB media with added 1 g/L glucose and 50 μg/mL kanamycin) with 300μL of frozen cell stock.
2. Allow for overnight growth in shaking incubator at 37°C at 220rpm. The concentration after growth was found to be saturated (OD650 ≥ 1).
3. Sterilize 2,970L of LB media in the bioreactor by autoclaving. Then, add 1 g/L glucose and 50 μg/mL kanamycin.
4. Allow bioreactor to reach equilibrium at 37°C, pH = 6.8, pO_2 = 40% (with air sparging).
5. Inoculate bioreactor media with 30mL of overnight growth culture.
6. Since this bioreactor does not have a control system for pH and pO_2, monitor pH and add 0.5M NaOH as needed to keep pH between 6.2 and 7.0. Also monitor pO_2 and switch to oxygen flow when cell metabolism rate is high enough to deplete the oxygen supplied by air. Adjust oxygen flow rate to keep pO_2 fairly stable between 30-50% for the remainder of the fermentation until the system becomes oxygen limited.
    a. Collect 10mL samples of culture every 20 minutes to measure optical density (OD650) and dry mass per volume.
    b. Analyze optical density measurements using a generated standard curve of serial dilutions of the second flask of overnight growth culture.

The following figures outline the collected data, which will be used in Appendix A.2 to complete the cell growth calculations. Figure A.A.1 is the standard curve for cell concentration as a function of optical density, generated using serial dilutions of the saturated overnight growth culture. Figure A.A.2 shows the optical density and dry mass data (both converted to cell concentration values) plotted over time. As shown, the growth is exponential as expected through the fermentation. The system became oxygen limited before growth could reach the stationary phase.

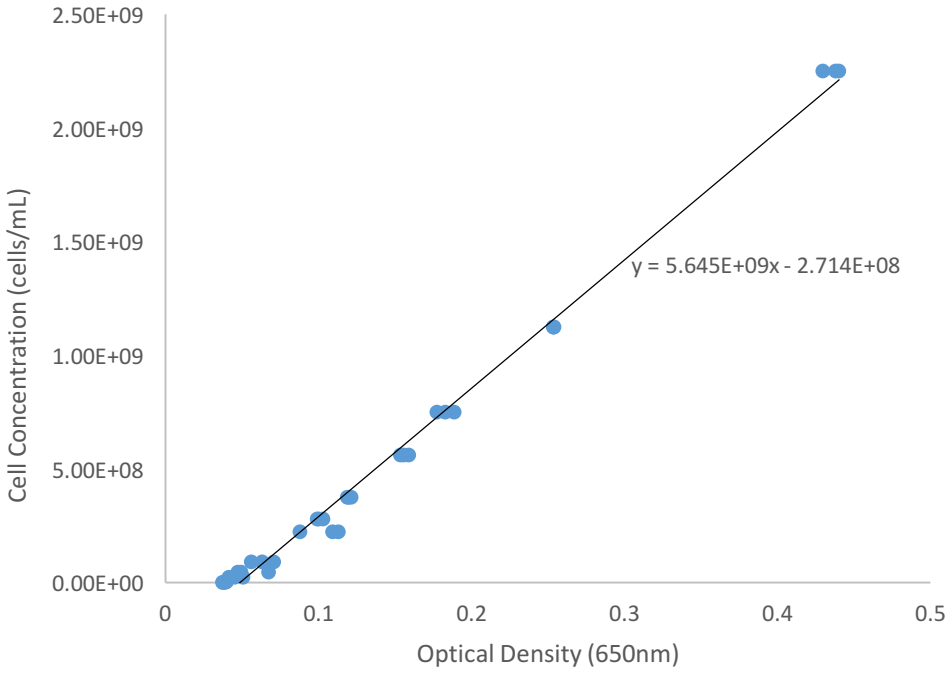

Figure A.A.1: Cell concentration as a function of optical density standard curve. Serial dilutions of the overnight growth culture (found to be at a saturated concentration of 2.25×10^9 cells/mL) were made to generate a standard curve for conversion of optical density measurements from the bioreactor culture over time to cell concentrations.

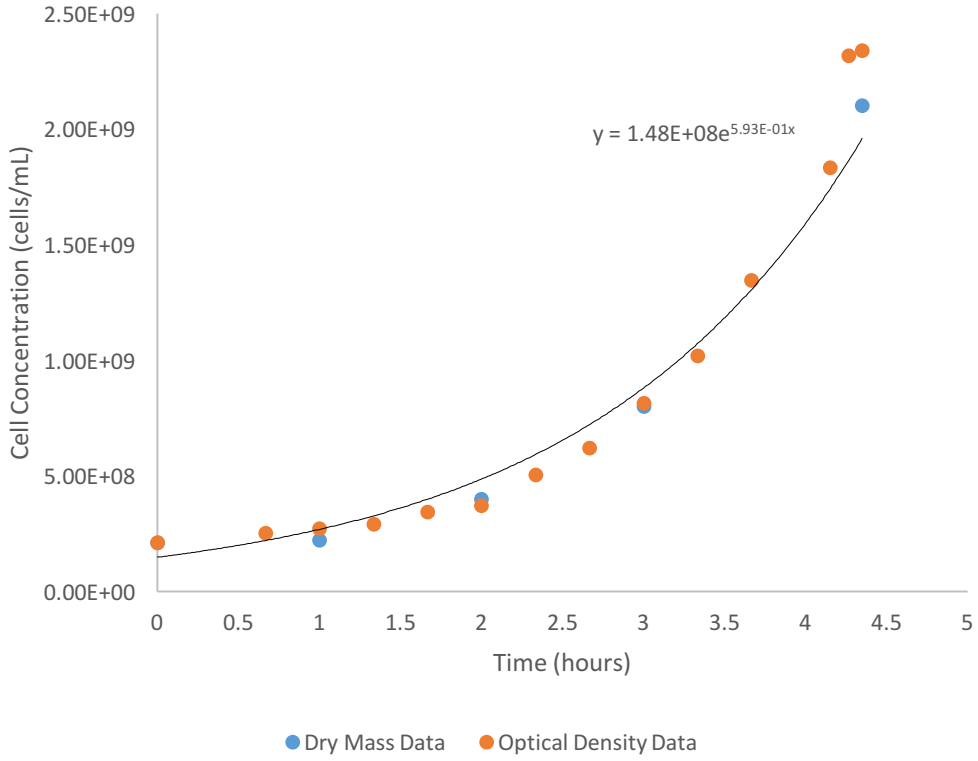

Figure A.A.2: Cell concentration over time based on collected dry mass and optical density data. Cell concentration was measured using both optical density and dry mass/volume measurements. The blue data points correspond to dry mass data and the orange data points correspond to optical density data points. The exponential fit was used to calculate cell concentrations for the process. The fit suggests a growth rate of 0.5931 hr^{-1} and an initial concentration of $1.48x10^8$ cells/mL.

Based on the reported pO_2 values from the computerized measurement system, it was evident that the *E. coli* consumed oxygen. A percent dissolved oxygen probe was calibrated and used to measure the dissolved oxygen throughout the full fermentation. From the time of inoculation (t = 0 hr) to 2.14 hours after inoculation, sterile air was sparged through the system at 129.5 mL/sec. The concentration of dissolved oxygen was stable for approximately the first half hour before it began to drop down to 1 mg/L after a total of 2.14 hours. The valve was then switched to flow pure oxygen gas through the bioreactor and the flow rate was adjusted to maintain conditions at 40% dissolved oxygen. As a result, the scaled-up process has been designed to operate only with oxygen gas in order to avoid oxygen limitations.

Looking more closely at the first 2.14 hours of the fermentation, comparison of the expected oxygen transfer rate (OTR) and the actual decrease in dissolved oxygen concentration leads to a sound estimation of the oxygen uptake rate (OUR) by the cells. The OTR is based on

the difference between the maximum solubility of oxygen in LB media and the dissolved oxygen concentration (measured by the probe) at a given time. Figure A.1.3 shows both the measured dissolved oxygen concentration and the calculated oxygen uptake rates from inoculation through the first 2.14 hours of the fermentation. The OTR and OUR were calculated as follows with equations (A.1.1) through (A.1.3):

C = measured dissolved oxygen concentration (mg/L)
C^* = solubility of oxygen in LB media at 37°C = 6.038 mg/L
k_La = liquid phase (LB media) oxygen mass transfer coefficient = 15.6 hr^{-1}

$$OTR = k_L a(C^* - C) \tag{A.1.1}$$

$$OTR + OUR = \frac{dC}{dt} \tag{A.1.2}$$

$$OUR = \frac{dC}{dt} - k_L a(C^* - C) \tag{A.1.3}$$

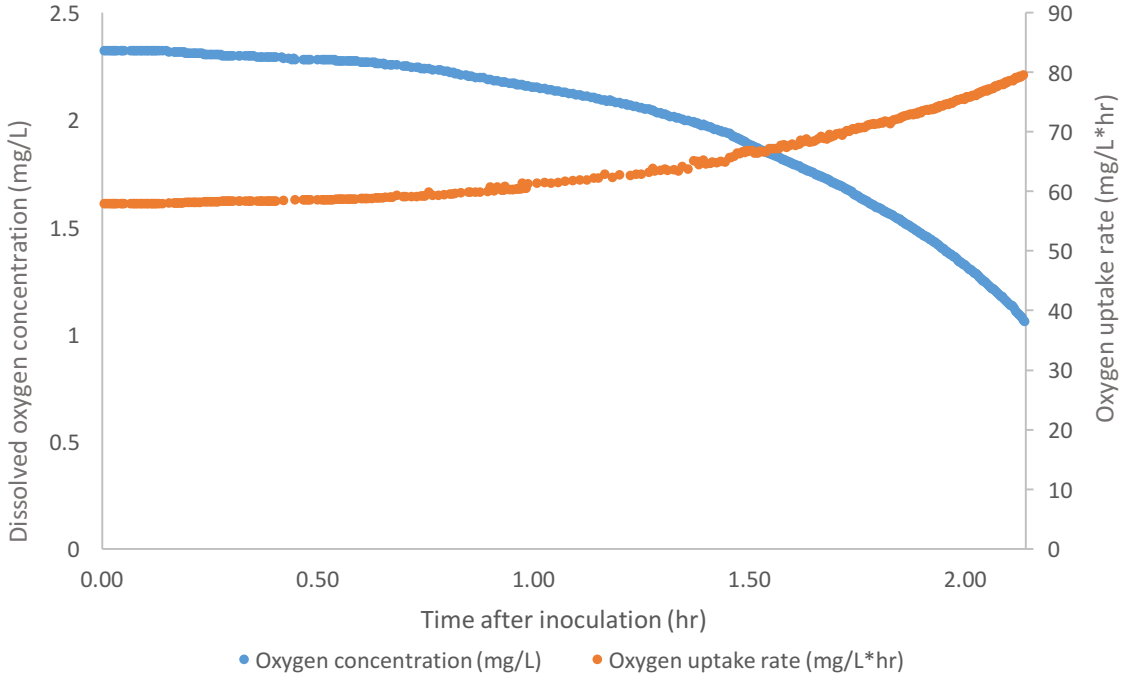

Figure A.A.1.3: Dissolved oxygen concentration and oxygen uptake rate over time. The figure shows the measured dissolved oxygen concentration and the calculated oxygen uptake rate from inoculation (t = 0 hr) to 2.14 hours into the fermentation. The decrease in dissolved oxygen concentration measured by the bioreactor probe confirms the consumption of oxygen by the present cells. The flowrate of sterile air was not changed during this period, so cell metabolism is the only source of the decrease in oxygen concentration. Oxygen concentration was measured by the bioreactor probe and oxygen uptake rate was calculated based on equations (A.1.1-A.1.3)

Also, a probe in the bioreactor monitored the pH over time without adding additional acid or base. At the time of inoculation, the system began with a pH of 6.8. Over time, the pH dropped by approximately 0.1 every 0.2 hours, although the probe only recorded values to the tenth decimal place so there is some error associated with this rate of change. After the pH reached 6.2, additional 0.5 M NaOH was added to help the pH stabilize at 6.8 for the remainder of the fermentation to promote cell growth. It is expected that the pH drops over time, becoming more acidic during the course of the fermentation because more metabolism in the bioreactor leads to more metabolic by-products that can be very acidic, such as lactic acid. Figure A.1.4 shows both the pH and the cell concentration over time. As the cell concentration in the bioreactor rises, the pH drops.

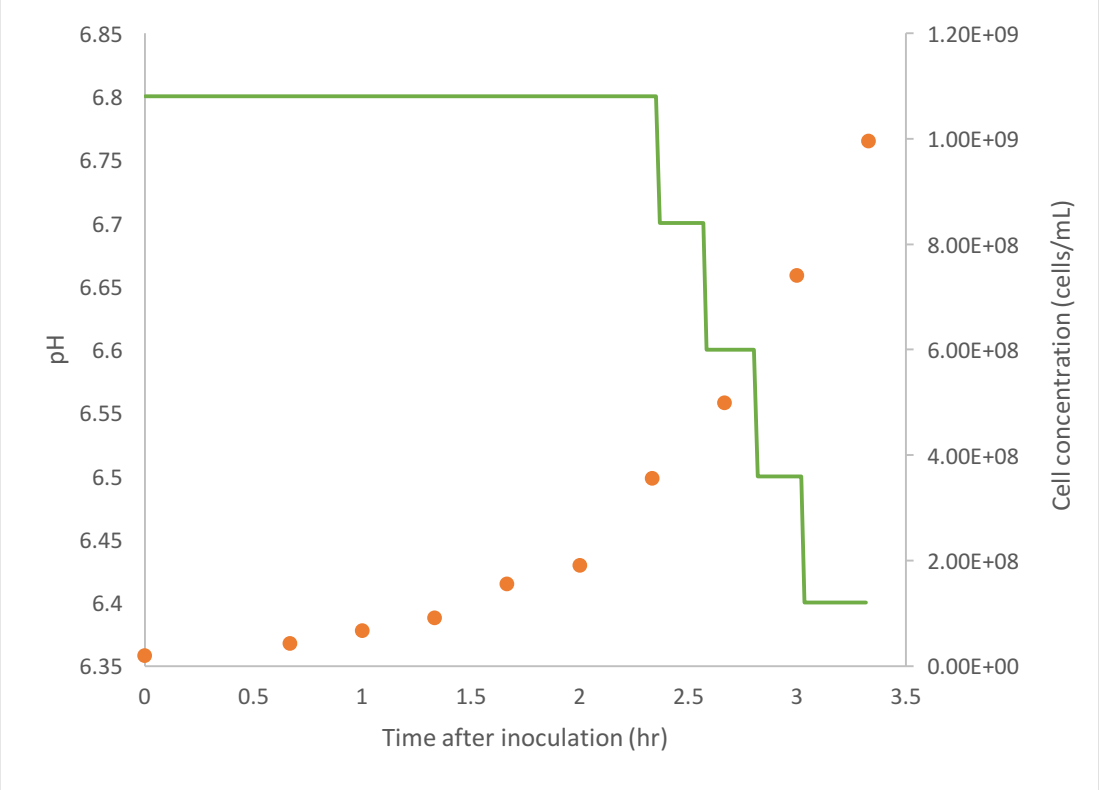

Figure A.A.1.4: pH and cell concentration over time. The figure shows the measured pH and the calculated cell concentration from inoculation (t = 0 hr) to 3.33 hours into the fermentation. The decrease in pH measured by the bioreactor probe confirms the metabolism of the present cells. As cell concentration (and metabolism) increases, the pH is expected to become more acidic due to a buildup of metabolic by-products, such as lactic acid. The pH probe in the bioreactor only had the capability to measure pH to the tenth decimal place.

Appendix A.2: Cell Growth Analysis

Assumptions:

1. Prepared media begins with a glucose concentration of 1 g/L in both the production bioreactor and the inoculation shake flask.
2. In the exponential growth phase, $\mu_{net} = \mu_g = 0.5931\ hr^{-1}$ because there is negligible cell death.
3. All glucose consumption is due to metabolic consumption by the cells.
4. The batch bioreactor is well-mixed at all times and the cell, nutrient, and oxygen concentrations are assumed to be consistent throughout the reactor.
5. Optical density testing of both the inoculum and product culture result in negligible cell loss. The only cell loss that is not negligible is accounted for in the mass balance during on transfer out procedures.
6. Oxygen level is held constant in the production bioreactor to ensure aerobic cellular respiration throughout the growth period.
7. Oxygen level in the shake flask is in equilibrium with the atmosphere at the start of the inoculum growth period and the only loss of oxygen in the shake flask is due to metabolic consumption by the cells.

<u>Cell Growth</u>

t = time (day)

X = cell concentration at time, t (cells/mL)

X_0 = initial cell concentration at (cells/mL)

μ_{net} = specific growth rate for the cell culture (hr^{-1})

$$\frac{dX}{dt} = \mu_{net} X$$

Rearrangement to solve for final cell concentration, X:

$$X = X_0 e^{\mu_{net} t}$$

For the inoculation step in 0.12L of culture, with $X_0 = 1.44 \times 10^6 \ \frac{cells}{mL}$, $\mu_{net} = 0.59\ hr^{-1}$, and t = 12 hours, the final cell concentration in the inoculation flask is $X = 1.78 \times 10^9$ cells/mL.

Rearrangement to solve for growth time, t:

$$t = \frac{1}{\mu_{net}} \ln\left(\frac{X}{X_0}\right)$$

For the production bioreactor in 11.6L of culture, with $X_0 = 1.54 \times 10^7$ cells/mL, final cell concentration $X = 1 \times 10^9$ cells/mL, and $\mu_{net} = 0.59\ hr^{-1}$, the growth time requires is t = 7.04 hours.

Appendix B: Bio-waste system design

Assumptions:

1. The final cell concentration in the production culture is $1x10^9$ cells/mL and this is the only source of bacteria that must be inactivated in the waste tank.
2. Inactivation kinetics are first order.
3. The decimal reduction time for *E. coli* at 121°C is $2.3x10^{-13}$ minutes.

For the assumed final cell concentration, the waste tank will contain:

$$1\times10^9\,\frac{cells}{mL}\times11.6\,\frac{L}{batch}\times1000\,\frac{mL}{L}=1.16\times10^{13}\,\frac{cells\ to\ inactivate}{batch}$$

First order inactivation kinetics:

$$\ln\left(\frac{N}{N_0}\right)=-kt$$

$$\log\left(\frac{N}{N_0}\right)=-\frac{t}{D}$$

Substitution:

$$k=\frac{2.303}{D}$$

where N = microbial population at any time, t

N_0 = initial microbial population

D = decimal reduction time; time required for a 1-log (or 10-fold) cycle reduction in the microbial population

For an 18-log reduction in the microbial population:

$$N=1\times10^{-9}cells/mL$$

This means that on average, there is a $1x10^{-9}$ chance that a single recombinant organism will be active after heat inactivation in each batch.

$$-18 = -\frac{t}{2.3\times10^{-13}}$$

$$t = 4.14\times10^{-12}\ minutes$$

Therefore, heating the bio-waste tank up to 121°C and holding at the temperature for one minute is sufficient to inactivate bacteria.

Appendix C: Batch Sizing

Assumptions:

1. The process is designed to run 100 batches per year, with each batch requiring five, 16-hour days broken up into eight-hour shifts.
2. The ultimate goal is to produce 7.39 kg Oleosin 30G per year in order to reach 100% market saturation.
3. Based on the bioreactor experiment (see Appendix A.1), the specific growth rate for this strain of BL21 recombinant *E. coli* is $0.5931 hr^{-1}$.
4. Frozen stock of this strain of BL21 *E. coli* will have a consistent concentration of $7.6x10^8$ cells/mL, half of a saturated *E. coli* culture after overnight growth ($1.52x10^9$ cells/mL). And cell viability for freeze-thaw is 65% (meaning that 65% of the frozen cells will survive thawing to be able to go on and divide).
5. Oxygen limitations will not be a concern for the growth periods and will be held constant using PID control at 40% O_2.
6. Recovery from upstream production is 85.74%, recovery from downstream purification is 51.12%, and overall recovery of the protein product is 43.83%.

Table A.C.1: Inoculum batch sizing details. Cell concentrations were calculated using the growth rate found in the pilot plant study, known initial concentration, and desired growth time.

Inoculum		
Working volume	0.116	L
Initial cell concentration	1.44E+06	cells/mL
Specific growth rate	0.5931	1/hr
Growth time	12	hr
Final cell concentration	1.78E+09	cells/mL

Table A.C.2: Production bioreactor batch sizing details. Cell concentrations were calculated using the growth rate found in the pilot plant study, known initial concentration, and desired growth time.

Production Bioreactor		
Working volume	11.6	L
Initial cell concentration	1.54E+07	cells/mL
Specific growth rate	0.5931	1/hr
Target cell concentration	1.00E+09	cells/mL
Growth time	7.04	hr
Protein production rate	70	pg/cell*day
Protein production time	5	hr
Protein produced	0.169	kg

Table A.C.3: Overall production statistics. Batch sizing was conducted to meet 100% market saturation. The plant has the capabilities required to produce more or less, as needed.

Annual Production		
Protein produced per batch	0.169	kg/batch
Production percent yield	85.7	%
Protein recovered in production	0.145	kg/batch
Purification percent yield	51.1	%
Protein recovered in purification	0.074	kg/batch
Overall protein percent yield	43.8	%
Batches per year	100	batches/yr
Days/batch	5	days/batch
Batches in parallel	2	batches
Active plant days per year	250	days/yr
Protein recovered per year	7.39	kg/yr

Appendix D: Separation Sizing and Timing

The chromatography column size and volumes of solution needed were calculated according to the protocol for the HisPur® cobalt affinity resin. The volume of resin required per batch was calculated using the given binding capacity, and the mass of Oleosin 30G per batch after protein production given in Appendix A. The amount of resin needed per batch was calculated assuming that all Oleosin 30G run through the column would bind to the resin, even though the actual recovery from this chromatography column with monoclonal has been shown to be 68%. This was done to ensure that our recovery is not limited by the binding capacity of the resin. Since no column packing data was available, random packing was assumed with a void fraction of 0.39.

$$\frac{Mass\ oleosin\ into\ the\ column\ (g)}{batch} * \frac{L\ resin}{g\ protein} = Resin\ Volume \frac{L}{batch}$$

$$7.087 \frac{g}{batch} * \frac{1\ L\ resin}{11.11\ g\ protein} = .628 L$$

$$Resin\ Volume * \frac{1}{1 - Void\ Fraction} = Column\ Volume$$

$$.628\ L * \frac{1}{1 - .39} = 1.03 L$$

The data sheet for the column also shows no observable recovery or purity changes through 24 regenerations, so the resin will be used 25 times before it is discarded. Also we use a conservative approach in calculating the batch time and amount of buffer needed, using the maximum of their ranges, since no exact values can be known without further experimental separation data using Oleosin.

$$\frac{Total\ Volume\ of\ resin\ required}{year} = \frac{Resin}{Batch} * \frac{Batches}{year} * \frac{1}{Number\ of\ Uses\ before\ Discarding}$$

$$7.03 \frac{L\ Resin}{year} = .628\ \frac{L\ Resin}{Batch} * 276 \frac{Batches}{Year} * \frac{1}{25\ Uses}$$

Table A.D.1: Calculated material requirements for chromatography buffers. Compositions and volumes required were obtained from the HisPur® Superflow Agarose resin vendor sheet. Masses required were calculated using the maximum protocol volumes listed.

Buffer Composition	Protocol Volumes	Composition	Amount per Batch	
Equilibration	3x-10x column volume	5mM Imidazole 20mM Sodium Phosphate 300mM sodium chloride	Imidazole	119g
Wash	6x-10x column volume	15mM Imidazole 20mM Sodium Phosphate 300mM sodium chloride	Sodium Phosphate	101g
Elution	4x-10x column volume	150mM Imidazole 20mM Sodium Phosphate 300mM sodium chloride	Sodium Chloride	541g
Regeneration	10x column volume	20mM MES	MES	39.5g
CIP	2x column volume 5x column volume water	6M guanidine HCl 1% nonionic detergent	Gunidine HCl	1180g
			Detergent	20.6g
			Ultrapure Water	48.4L

$$Column\ \ Run\ time = Volume\ of\ water * \frac{1}{linear\ flow\ rate\ * column\ cross\ section}$$

$$8.34hrs = 48{,}400mL * \frac{1}{150\,\frac{cm}{hr} * (3.5cm)^2 * \pi} = 6.54\ hrs\ run\ tme + 1.78\ hrs\ regeneration + CIP$$

Appendix E: Mass per Dose Calculation

The mass of Oleosin 30G required in a single dose was calculated from flow rate and solution composition data collected during the creation of the microbubbles with a microfluidic device. The flow rates of nitrogen gas and of the solution used to create the microbubbles are known, as is the concentration of Oleosin 30G in solution. It was assumed that all nitrogen gas flowed through the device was encapsulated in microbubbles. The total gas volume enclosed in one dose of microbubbles was based upon that of the current contrast agent, Albunex. A calculation based on liquid and gas flow rate was used as opposed to using a surface area calculation, because the ratio of Oleosin 30G that remains in solution to that which forms a microbubble surface is not known. The calculation is shown below.

$$average\ gas\ volume\ in\ albunex\ dose = dose\ volume\ (ml) * \left(\frac{number\ of\ bubbles}{ml}\right) * volume\ of\ one\ bubble\ (\mu l)$$

$$771.8\ \mu l = 7.2ml\left(4 * 10^8 \frac{bubbles}{ml}\right)\left(\frac{4}{3}\ \pi(.004mm)^3\right)$$

$$\frac{(average\ gas\ volume\ in\ albunex\ dose\ (\mu l))}{(gas\ flow\ rate\ in\ micofluidic\ device\ (\frac{\mu l}{hour}))}\left(liquid\ flow\ rate(\frac{\mu l}{hour}\right) * [oleosin](\frac{\mu g}{\mu l}) = \frac{oleosin}{dose}$$

$$\frac{771.8\ \frac{\mu l}{dose}}{62\ \frac{\mu l}{hr}} * 1000\frac{\mu l}{hr} * 1\frac{\mu g\ oleosin}{\mu l} = 12{,}450\frac{\mu g\ Oleosin}{dose} = 80{,}300\frac{doses}{kg\ oleosin}$$

Appendix F: Steam-in-Place Calculations

Below is a sample cooling time estimation for the piping in the disk stack centrifuge. The diameter, length, air flow rate, and mass of the bioreactor vessel are specified by the Culturfuge100. Temperature in the bioreactor vessel itself can be controlled by cooling water running through the reactor shell, which cools the reactor by 1^{o}C per minute. It is assumed that there are 15m of piping in the system, the pipe thickness is 4mm, and the flow rate through the system is the max air flow rate through the bioreactor in the utilities requirement of 52L/min.

$$Re = \frac{QD}{vA} = \frac{8.67 * 10^{-4}\frac{m^3}{s} * .019m}{1.568 * \frac{10^{-5}\,m^2}{s} * \frac{(.019)^2}{2}\,\pi\,m^2} = 3704$$

.

$$Nu_{average} = \frac{h_{average}D}{k} = 3.66 + \frac{0.065\,\mathrm{Re\,Pr}\,\frac{D}{L}}{1+0.04\left(\mathrm{Re\,Pr}\,\frac{D}{L}\right)^{2/3}}$$

$$h_{average=}\ 3.86 * \frac{0.0257\,\frac{\mathrm{W}}{\mathrm{mK}}}{.019m} = 5.221\,\frac{W}{m^2K}$$

$$mC_p dt = hA(T_{air} - T_{bioreactor})$$

$$.00197m^3 * 7850\,\frac{kg}{m^3} * 510\,\frac{J}{kg\,K}\frac{dT}{dt} = 5.221\,\frac{W}{K\,m^2} * 1.19m^2(25 - T_{bioreactor})$$

T(0)=138^{O}C

$$T(t) = 113e^{-.000788t} + 25$$

T(cooling time)=37 ^{O}C, cooling time =2,895sec= 48.25min

Chromatography Column SIP

Because the resin is regenerated after the chromatography column is run, the column can only be steamed in place after 25 uses, when the resin is switched out. Therefore, the CIP/SIP procedure which is detailed in the HisPur® resin user manual will be followed: after washing

with the elution buffer, the column will be run with 2 column volumes (42L) of 6M guanidine HCl with 1% nonionic detergent for 20 minutes, then the column will be flushed with 5 column volumes (105L) of water for injection. The column will then be regenerated using 10 column volumes (210L) of regeneration buffer.

Appendix G: Utility Requirements for the Refrigerated Room

Assumptions:

1. Source for R-values are provided by North Carolina University in an article titled "Design of Cooling Facilities: Structural & Energy Requirements."
2. Field heat is equal to 10 percent of the heat given off through conduction.
3. 21 ^{O}C (room temperature) assumed to be the "outside temperature" as the cold room is located in the interior of the building.
4. Costs for refrigerators in the lab for storage of final materials are included in the electricity requirements calculations.

Table A.G.1: Building Heating Cost Calculation

Heat Conduction	
Building Dimensions	
Wall Width (ft.)	9
Wall Length (ft.)	12
Wall Height (ft.)	7
Temperature Difference	
Outside (C)	21
Inside (C)	4
R-values	
Walls	16 sq ft/F Btu
Ceiling	20 sq ft/F Btu
Floor	11 sq ft/F Btu
Heat Conduction	
Walls	408 Btu/hr
Ceiling	91.8 Btu/hr
Floor	166.90 Btu/hr
Total	666.70 Btu/hr
Service Load	
Total	66.67 Btu/hr
Total Heat	733.38 Btu/hr
Price Calculations	
Price of Utilities	19.7 cents per kilowatt hr
Price per year (cents)	37071. 13 cents/year
Price per year (dollars)	370.7113 dollars/year

Appendix H: Microfluidics Calculations

Assumptions

1. Microfluidic device operates at a consistent flow rate with reliable output
2. Stabilization time will be constant and equal for each mold
3. Bubble volume and concentration data is sourced from Albunex® and modeled after a similar suspension
4. All Oleosin recovered from bulk filtration is sent directly for packaging with negligible loss
5. Cost of pharmaceutical grade shipping is higher than standard Priority Mail and accounted for in sales figure (revenue = 75% of selling price)
6. Uniform bubble concentration ($4x10^8$ bubbles/mL) is maintained throughout the solution

Table A.H.1: Calculations for Microfluidics packaging operations. Using the output per batch of Oleosin 30G. bubble production times were calculated per hour using a flow rate estimation courtesy of Dr. Lee. It was determined that eight masters would be used to produce eight molds for production of final bubble suspensions. Associated shipping costs were calculated based on the number of 10 mL glass vials produced per batch, with each vial containing just over one dose of Oleosin 30G in solution.

Output for Packaging (kg/Batch oleosin)	$7.39x10^{-2}$
Concentration desired (mg/mL)	1
Volume of output for packaging (mL/batch)	$7.39x10^4$
Flow rate of microfluidic device (L/hr)	0.75
Bubble production time/batch (hr)	98.5
Stabilization time (min)	30
Clearing time after process (min)	30
Time per batch (1 device) (hrs)	99.5
Number of disposables	8
Bubble production time per batch (8 devices)	12.3
Stabilization time (min)	30
Clearing time after process (min)	30
Time per batch (8 devices) (hr)	13.3
Cost/disposable ($)	100
Cost of disposables ($)	1600
Cost of master ($)	2000
Total cost ($)	3600
Volume/Bubble (μL)	$2.68x10^{-7}$
Number of bubbles/mL	$4x10^8$
Total volume of gas per mL (mL)	0.107

average dose (mL)	7.2
gas/dose	0.772
Gas flow rate (μL/h)	62
time for one dose (hr)	12.5
Liquid flow rate (μL/h)	1000
liquid per dose (mL)	12.5
mass Oleosin/dose (mg)	12.5
doses/batch	5934
vials/ batch	5934
vials/yr	5.934×10^5
market penetration (%)	100
Shipping Price	$22.95/70 lbs
10mL glass vial (kg)	0.05
Mass of product per vial (kg)	0.00001
Mass per finished vial (lb)	0.11
Mass vials/batch (lb)	654
Price UPS overnight/batch ($)	215
Price shipping/year ($)	5.74×10^4

Appendix I: Endotoxin Level Calculation

Endotoxins are lipopolysaccharides (LPS) on the surface of E. coli cells that causes a pyrogenic (fever producing) response in humans if present in the bloodstream at significant concentrations. The endotoxin limit for intravenous injections is set by the FDA to be 0.5 endotoxin units (EU) per kg body mass, where an endotoxin unit equals about 100pg of *E. coli* lipopolysaccharides. In order to ensure our product is well below this threshold, we first calculated the mass of endotoxins produced per batch, assuming a high estimate of $4.94*10^{-19}$ kg endotoxin per *E. coli* cell[27]. The final concentration of 10^9 cell/ml and volume in the bioreactor in mL are taken from Appendix A.

$$4.94 * 10^{-19} \frac{kg\ LPS}{cell} * 10^9 \frac{cells}{ml} * 10.5 * 10^3 ml = 5.187 * 10^{-6}\ \frac{kg\ LPS}{batch}$$

The amount of endotoxin in EU after two bulk filtration steps was then calculated to determine if our final product is within the FDA guidelines of 0.5 EU/kg of body mass, assuming a 99% reduction of endotoxin mass per bulk filtration. The calculated value of .874 EU/dose is within the FDA guidelines for any patient above 1.7kg, and well within the limits for the average patient.

$$5.19 * 10^{-6} \frac{kg\ LPS}{batch} * 1\%\ * 1\%\ * \frac{10^{15}\ pg}{kg} * \frac{1\ EU}{100\ pg} * \frac{1\ batch}{5934\ doses} = 0.874 \frac{EU}{dose}$$

Appendix J: Bioreactor Control Diagram

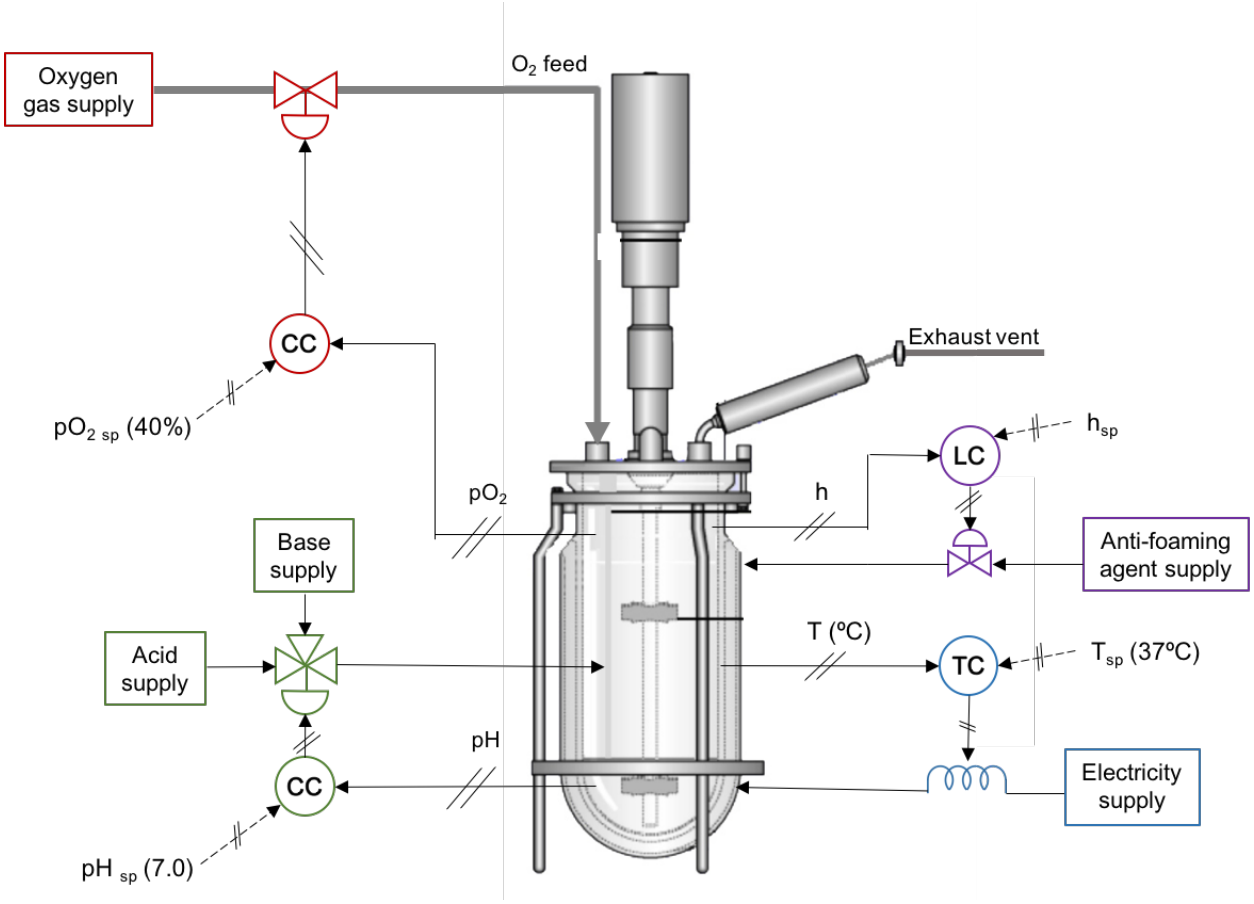

Figure A.J.1: Bioreactor Control Diagram. The figure shows the four control systems that will be used to keep the bioreactor conditions stable. PID control will be used for all. These conditions include temperature (T), pH, percent dissolved oxygen (pO_2), and foam level (h).

BIOSTAT® RM

Introduction
The BIOSTAT® RM is a single use, wave-mixed bioreactor for culture volumes from 100 mL to 300 L. The system consists of a rocker unit with bag holder, a digital controller and a disposable bag. The bag, which forms the cultivation chamber, is mounted on the rocker platform and contains the medium and the cells. Due to the wave in the bag, generated by the moving platform, the media surface is renewed constantly, providing bubble-free aeration with low sheer.

Applications
- Cultivation with or without pH and DO feedback control
- Cultivation of mammalian, insect and plant cells
- Cultivation of stem cells
- Seed bioreactor
- Cost efficient cell mass, protein, Mab & vaccine production

BIOSTAT® RM Product family description
The RM product family comprises four different bioreactor sizes, 20 L, 50 L, 200 L and 600 L in different configurations. For applications where advanced control is not required instruments are available without pH and DO measurement (basic systems). For more complex processes optical systems with sophisticated feedback control for all process parameters including pH and DO are available as well as perfusion systems for fully automated continuous cultivations. BIOSTAT® RM 20 and 50 share the identical rocker unit and differ in size of the bag holder, which can be exchanged from 20 L to 50 L and vice versa.

Basic systems
Basic systems are designed for stand alone bench top use and allow controlling rocking rate, angle, and temperature. An internal gassing module can be added for aeration with air and CO_2 to work with a fixed CO_2 concentration of 0–15% in the process gas. The digital controller is directly integrated into the rocker unit and operated with an easy to use colour touch screen directly on the rocker.

Features of the BIOSTAT® RM basic include:
- Setting of rocking rate and angle
- Individual temperature control of two bags (2 L, 10 L) or one bag (20 L, 50 L)
- Independent gassing of two bags (0–500 ml/min) or one bag (0–1000 ml/min)
- Setting of the bag configuration: will automatically select the right gassing and temperature control parameters of the system
- Integrated Air | CO_2 mixing by optional gassing module
- Air supply, switchable between internal air pump or process gas
- Positioning of the platform for harvest and sampling
- 2 Filter heaters made of PC, plug directly into rocker base
- Color coded plugs and socket for easy operation
- Tube and cable organizer at the sides of the bag holder
- Security function, check plug in of filter heater when gassing is switched on
- Alarm display
- 3 different user level (Administrator, User, Locked)
- Trend display for data visualization
- Time and date display
- Selection of control mode: Local or DCU
- Potential free alarm contact
- RS232 serial interface for communication with PC running
- Optional Ethernet interface with communication protocol for connection to third party software
- Optional ProfiBus DP interface with communication protocol for connection to third party software
- Service Interval Display

Optical systems
The BIOSTAT® RM Optical provides full process automation with sophisticated feed back control. In addition to the rocker unit, it comprises a BIOSTAT® DCU (digital control unit) tower. The control tower is connected to the rocking unit for monitoring and controlling the culture, including pO_2, pH, agitation, and temperature in batch and fed batch mode of operation. Pre-calibrated, single-use optical sensors are included in the bag for the measurement of DO and pH.

Perfusion systems
The BIOSTAT® RM Perfusion systems allow fully automated, continuous processes. The single-use bag is equipped with optical pH and DO probes. It contains an internal perfusion membrane for efficient cell retention. The feed and harvest pumps are controlled by gravimetric flow controllers, which monitor the weight of the feed and harvest containers to ensure precise flow rates.

Different perfusion configurations are available depending on the working volume, the required perfusion rate and the maximum feed and harvest container weight.

Order Code	Description	Perfusion Rate (L/day)	Type of Pump	Weighing Capacity Balances (kg)	Readability Balances (g)
DH-----PRM11	Perfusion Option 1 – 120 VAC	2–55	int. WM102	60	1
DH-----PRM12	Perfusion Option 1 – 230 VAC	2–55	int. WM102	60	1
DH-----PRM21	Perfusion Option 2 – 120 VAC	2–55	int. WM102	300	10
DH-----PRM22	Perfusion Option 2 – 230 VAC	2–55	int. WM102	300	10
DH-----PRM31	Perfusion Option 3 – 120 VAC	23–1100	ext. WM323	300	10
DH-----PRM32	Perfusion Option 3 – 230 VAC	23–1100	ext. WM323	300	10
DH-----PRM41	Perfusion Option 4 – 120 VAC	23–1100	ext. WM323	600	20
DH-----PRM42	Perfusion Option 4 – 230 VAC	23–1100	ext. WM323	600	20
DH-----PRM51	Perfusion Option 5 – 120 VAC	23–1100	ext. WM323	1500	20
DH-----PRM52	Perfusion Option 5 – 230 VAC	23–1100	ext. WM323	1500	20

BIOSTAT® RM 200, single-use bioreactor

BIOSTAT® RM 600 optical,
large scale single-use bioreactor

Twin systems
The BIOSTAT® RM 20, RM 50 and RM 200 systems are available as Single and Twin systems. One controller can independently control the temperature, gas flow, pH and DO of two bags. The bags can be mounted on two different rockers (Twin Rocker) or on the same rocker (Twin Controller). The BIOSTAT® RM 20 and RM 50 are available either in Twin Rocker or Twin Controller configuration, also as mixed RM 20 | RM 50 Twin variants. The BIOSTAT® RM200 always comes as Twin Controller model.

Model	max. working volume (L)	basic	system type optical	perfusion	Twin availability Twin rocker	Twin controller	temperature control* heating only	heating|cooling
20	10	×	×	×	×*	×	×	×
50	25	×	×	×	×*	×	×	×
200	100		×			×	×	×**
600	300		×				×	×**

* only for optical and perfusion
** on request

BIOSTAT® RM digital control unit (DCU)
- Cabinet contains measurement & control hardware, pumps & gassing system
- Separate from rocker unit (RM 20, RM 50, RM 600) or installed on a same skid (RM 200)
- Graphical user interface with colour display and touch screen operation
- Integrated amplifiers for temperature, pressure, single use DO and pH sensors
- Integrated control loops for temperature, DO, pH, rocker speed, rocker angle, gas flow and substrate
- Fully automated perfusion control using gravimetric flow controllers (Perfusion systems)
- Multi-channel DO cascade control
- Calibration of DO and pH sensors
- In-process DO and pH recalibration
- Time-controlled profile function for all process parameters
- Optional password and logbook function
- Trend display for up to 6 process values

Gassing module
- Outlet to overlay aeration
- 4-fold gas mixing of air, N_2, O_2 and CO_2
- One mass flow min for total flow (see technical data for flow rates)
- One separate mass flow controller for CO_2 (see technical data for flow rates)
- Rotameters for air, N_2, O_2, and CO_2
- Control via pH/DO controller
- Measurement of bag pressure (RM 20, RM 50, RM 200). Control of bag pressure (RM 600)
- Double safety feature: Electronic shut off plus mechanical pressure release valve to protect from overpressure
- Filter Heater on exhaust filter to prevent formation of condensate and avoid filter blockage

Pumps
- Two integrated digital peristaltic pumps
- Two additional integrated or external analogue feed pumps available on request (standard with perfusion configuration)

Temperature control
Choice between heating only and heating | cooling by optional thermostat unit

Heating Only
- Electrical heating blankets on bag holder
- Simultaneous, independent control of two bags on one platform (RM 20, RM 50, RM 200) or one bag (RM 600)
- Temperature range: ambient to 40°C
- Automatic safety shutdown for prevention of overheating

Heating | Cooling
- Stainless steel temperature coil mounted on bag holder.
- Thermostat for heating
- Cooling water to be connected to cooling water supply or external chiller
- Circulation pump
- Quick coupling connectors
- Temperature range: 8°C above cooling water to 40°C
- Automatic safety shutdown for prevention of overheating

Agitation System & Bag Holder
- Bag holder mounted on rocking platform
- Clamping rails to hold down bag
- Material stainless steel or ABS
- Detachable or swivelling top for easy access probes, ports and sample lines

Sensors
- Disposable optical chemical sensors DO are installed in every optical and perfusion bag
- Sensors are pre-calibrated and supplied with calibration parameters
- Range: pH: 6.5 – 8.5
  DO: 0 – 100%
- PT100 reusable sensors for temperature measurement

SCADA Software BioPAT® MFC S/DA
- Part of every bioreactor package
- Plug and play configuration
- Online data acquisition
- Sample data management
- Enhanced plotting
- Export functions
- Easy to use programming interface
- Upgrade to advanced BioPAT® MFCS/Win control software possible

Features & Benefits
- Single use Bioreactor with very low operation costs
- Based on proven rocking motion ("wave induced motion") principle
- Basic systems provide flexible, autonomous stand-alone systems for simple cultivations
- Optical and Perfusion systems are designed for high end applications
- Large working volume range in one bag
- Flexible gassing system
- Gas flow adjustments via rotameters and mass flow controllers
- Double pressure safety control to avoid overpressure in bag
- Advanced cascaded DO control
- Intuitive touch screen interface for easy operation
- Easy bag installation
- Supervisory Process Control software (BioPAT® MFCS/DA) included

BIOSTAT® RM 20 basic, with mounted lid

BIOSTAT® RM 20 basic, without lid

BIOSTAT® RM 20




Technical Specifications

	BIOSTAT® RM 20 | 50 basic	BIOSTAT® RM 20 | 50 optical & perfusion	BIOSTAT® RM 200 optical & perfusion	BIOSTAT® RM 600 optical
Volume				
Total volume	up to 20 L (RM50:50 L)	up to 20 L (RM50:50 L)	up to 200 L	600 L
Minimum working volume (bags with sensors may require higher minimum volumes)	0.1 L (RM50:5 L)	0.1 L (RM50:5 L)	10 L	60 L
Maximum working volume	10 L (RM50:25 L)	10 L (RM50:25 L)	100 L	300 L
Bag Holder				
ABS	×	×		
Stainless steel			×	×
Clamping rails for bag fixation	×	×	×	×
Pressure sensor with gassing safety shut off	×	×	×	×
Proportional valve to maintain bag pressure at constant level	N/A	N/A	N/A	×
Redundant overpressure relieve valve	–	×	×	×
Sensor clamps for secure fixation of glass fiber cables	N/A	2	4	2
Filter heater	2	2	2	2
Controller				
Integrated into rocker	×			
DCU control tower	N/A	×	×	×
Potential free alarm contact	× [max 0.5 A]	(×)	(×)	(×)
Color touch screen	×	×	×	×
Different user level log in	×	(×)	(×)	(×)
Logbook function	N/A	(×)	(×)	(×)
Temperature Control				
Temperature modes	Heating Only	Only Heating or Heating|Cooling	Only Heating or (Heating|Cooling)	Only Heating or (Heating|Cooling)
Temperature range, heating only	RT –40°C	RT –40°C	RT –40°C	RT –40°C
Temperature range heating | cooling	N/A	8°C above cooling water –40°C	8°C above cooling water –40°C	8°C above cooling water –40°C
pT 100 probes and temperature amplifiers	2	2	2	1
Heating power, heating only	2 × 140 W (48 VAC)	2 × 140 W (48 VAC)	2 × 650 W	1 × 1500 W
Heating power, heating | cooling	N/A	1 × 1000 W	2 × 1000 W	2 × 1000 W
Overtemperature protection	×	×	×	×
Gassing module basic rocker				
Maximum total flow (ml/min)	(0–1000, or 2 × 0–500), ± 5%	N/A	N/A	N/A
Fixed CO_2 gassing (%)	(0–15)	N/A	N/A	N/A
Internal air pump	(×)	N/A	N/A	N/A




	BIOSTAT® RM 20 | 50 basic	BIOSTAT® RM 20 | 50 optical & perfusion	BIOSTAT® RM 200 optical & perfusion	BIOSTAT® RM 600 optical
Gassing module optical systems				
Rotameter				
– O_2	N/A	×	×	×
– N_2	N/A	×	×	×
– CO_2	N/A	×	×	×
– Air	N/A	×	×	×
Mass flow controllers for:				
– CO_2	N/A	0–500 mL/min	×	×
– total flow (Air, O_2, N_2)	N/A	RM20: 0.02–1 L/min RM50: 0.2–10 L/min	×	×
Multi-channel DO cascade control	N/A	×	×	×
Agitation				
Rocker speed (rpm), electronic adjustment	8–42 ± 1	8–42 ± 1	6–20 ± 1	2–16 ± 1
Rocker angle (°), electronic adjustment	4–10°, ± 0,3°,	4–10°, ± 0,3°,	4–10° ± 0.3°	4–10° ± 0.3°
DO and pH Measurement				
pH range	N/A	6.5–8.5	6.5–8.5	6.5–8.5
DO range	N/A	0–100%	0–100%	0–100%
Amplier for optical single-use DO sensor	N/A	1	2	2
Amplier for optical single-use pH sensor	N/A	1	2	2
Recalibration function for:				
– Disposable DO sensor	N/A	×	×	×
– Disposable pH sensor	N/A	×	×	×
Interface:				
– RS232	1	2	2	2
– Ethernet	(1)	1	1	1
- Profibus DB	(1)	N/A	N/A	N/A
– Analogue IN	N/A	2	2	2
– Analogue OUT	N/A	2	2	2
BioPAT® MFCS/DA	×	×	×	×
Pumps & Balances				
Digital pumps WM102	N/A	3		
Analogue pumps WM313D	N/A	(2)		
Analogue pumps (via analogue OUT)	N/A	(up to 2)		
External balances	N/A	(up to 2)		
Measurement of media weight (integrated balance)	(×)	(×)	(×)	(×)




	BIOSTAT® RM 20 | 50 basic	BIOSTAT® RM 20 | 50 optical & perfusion	BIOSTAT® RM 200 optical & perfusion	BIOSTAT® RM 600 optical
Power Supply				
Rocker 120 V–230 VAC | 1-phase | 6.3 A	×	×	N/A	N/A
Control tower: 120 VAC | 1-phase | 16 A	N/A	country specific	N/A	N/A
Control tower: 230 VAC | 1-phase | 16 A	N/A	country specific	N/A	N/A
Complete System: 208 VAC | 3-phase TN-S | 32 A	N/A	N/A	country specific	country specific
Complete system: 400 VAC | 3-phase TN-S | 32 A	N/A	N/A	country specific	country specific
Laboratory Supply				
Process gasses pressure (barg)	1.0–1–5	1.3–1.5	1.3–1.5	1.3–1.5
Gas specifications according to ISO 8573-1				
– Particle size: < 0.1 mm amount: max. 0.1 mg/m³ (class 1)	×	×	×	×
– Condensate: dew point < ×3°C (Class 4)	×	×	×	×
– Oil: < 0.01 mg/m³ (Class 1)	×	×	×	×
– Germs Class 0	×	×	×	×
Quick Couling for gas tubes, Festo Type (OD 4 mm)	×			
Hose barb for gas tubing, ID 6 mm	N/A	×	×	x
Cooling water (for heating cooling system only)	N/A	(×)	(×)	(×)
Hardware Dimensions & Weight				
W × H × D (mm)	RM20: 765 × 600 × 400 mm RM50: 1085 × 600 × 450 mm	BIOSTAT® RM Control Tower 320 × 735 × 565 mm plus size of basic rocker	1998 × 1241 × 830 mm	1790 × 1470 × 1330 cm
Weight	RM 20: 30 kg RM 50: 32.2 kg	BIOSTAT® RM Control Tower 50 kg plus weight of basic rocker	272 kg	340 kg

* Available from Q1/2012
(×) optional, needs to be ordered separately

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Sterilizable-in-place convenience
Why struggle carrying heavy vessels to and from the autoclave? Now you can sterilize your vessel, inlet and exhaust lines on your lab bench — with no external steam supply needed.

> Sterilization sequences are fully automated, easily initiated and configurable to match a wide variety of process requirements
> Rapid heat-up and cool-down

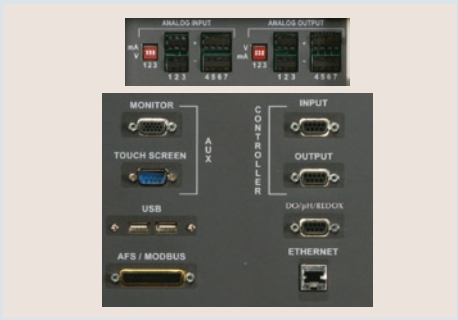
Powerful controller with large touchscreen display
We've seamlessly blended power & simplicity into one easy-to-use control station.

> Controls up to 32 process loops
> Easily integrates multiple external devices including scales, analyzers or sensors for optimized yields
> Saves up to 10 of your recipes for repeat usage

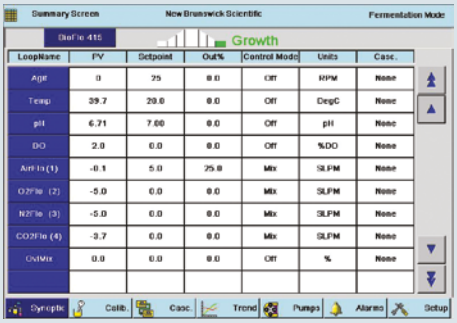
Pre-configured or customizable to fit your process needs
Simplify ordering by choosing one of our pre-configured packages, or select from a wide array of options to customize to your process needs.

> Interchangeable 7, 14 & 19.5 L stainless-steel vessels; there's no hard piping, so you can interchange another vessel of any size, at any time
> 1 Thermal Mass Flow Controller (TMFC) is standard
> Multiple TMFCs optional
> Multiple impeller options available
> Optional sensors, addition kits and BioCommand® supervisory software can be added. Validation and training packages are also available

Sparger and exhaust condenser with integral heating pad eliminate clogging during fermentation



Multiple connections are provided for integrating ancillary equipment & BioCommand® supervisory software



Summary screen lets you conveniently view setpoints, current values, cascade loops and more

Headplate Design lets you incorporate optional redundant sensors and multiple septums

Safety Features: a sanitary rupture disk in the vessel and an ASME safety release valve on the drain jacket are standard

Adjustable-Angle, User-Friendly 15 in (38 cm) Touchscreen Interface

Connections for Gasses and Vessel Components are easily accessible

Front-Mounted On-Off Switch is easily accessed

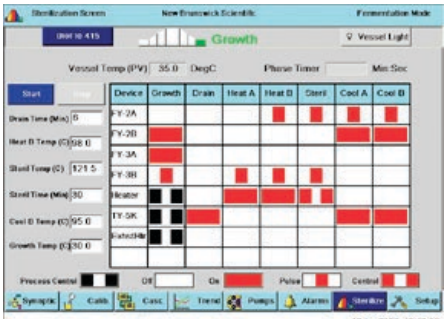
Quick Connects allow utilities to be added in seconds

Three Built-in, Assignable, Peristaltic Pumps

ASME and CE Certified vessel

Optional Drain Valve is easily accessed through an opening at the bottom of the vessel

The trend graph screen makes it simple to track and export data on up to eight process variables over a six day span



Enter and view sterilization parameters and valve sequences from the sterilization screen

The cascade screen provides sophisticated process control

eppendorf

BioFlo® 415 Fermentor Specifications*

Vessel Volume	**Total Capacity**	**7.0 Liters**	**14.0 Liters**	**19.5 Liters**
	Working Volume	2.0 - 5.0 Liters	4.0 - 10.5 Liters	5.0 - 15.5 Liters
Vessel Construction	Aspect Ratio	2:1	2:1	3:1
	Fabrication	ASME/CE certified. 316L stainless steel. 20 CLA (0.5 µ) Ra internal finish and 35 CLA (0.875 µ) Ra external finish		
Ports	Headplate	(2) 6.35 mm	(2) 6.35 mm	(2) 6.35 mm
		(9) 12 mm	(10) 12 mm	(10) 12 mm
		(1) 19 mm	(1) 19 mm	(1) 19 mm
		(2) PG 13.5	(2) PG 13.5	(2) PG 13.5
	Upper Side Wall	2 in Tri-clamp (1.5 in round sight glass)		
	Bottom	0.75 in NA-Connect®		
Net Weight	Control Station	40 kg (88 lbs.) including 6.8 kg (15 lbs.) touchscreen		
	Vessel	21 kg (47 lbs.)	27 kg (60 lbs.)	36 kg (80 lbs.)
Dimensions (W X D X H)	cm	63.5 x 66.0 x 97.8	63.5 x 66.0 x 114.3	63.5 x 66.0 x 134.6
	Inches	25 x 26 x 38.5	25 x 26 x 45	25 x 26 x 53
Controller	Control Station	Controls 1 vessel with 32 control loops. Stores 10 recipes and 8 process variables for trend graphing. Includes an industrial touchscreen monitor/user interface, 3 built-in pumps, and connections for all utilities and communications signals		
	Touchscreen Interface / Display	38 cm (15 in) Industrial touchscreen interface/display		
Temperature	Heat and Sterilization†	Electric heaters and automatic sterilization control, capable of achieving temperature rises of ~ 1 °C/min.		
	Range and Control◊	Culture temperature 5 °C to 80 °C, displayed in 0.1 °C increments using Platinum RTD sensor		
Agitation	Drive	Top magnetic drive with single mechanical seal. Digital display in 1 rpm increments		
	Range and Control	50 - 1000 rpm, ±1 at 100 rpm ; ± 2 at 500 rpm ; ± 5 at 1000 rpm		
	Impellers	Two six-bladed Rushton impellers on 7.0 and 14 L systems; Three impellers on 19.5 L systems		
	Baffles	Four 316L removable, stainless steel baffles		
Exhaust	Condenser and Filter	Stainless-steel exhaust condenser on headplate. 1.2 µ disposable depth filter; 0.2 µ absolute option		
Aeration	Gas System	Standard: 1 Thermal Mass Flow Controller (TMFC) with 0.5 to 25 SLPM flow rate and built in four-gas control (4 solenoid valves). Optional: Rotameter or 2nd, 3rd or 4th TMFCs for individual gas control		
	Gas Inlet	Ring sparger is provided with 0.2µ absolute disposable filter for use as a sparger or overlay		
pH	Sensor	Option of one or two Gel-filled pH sensor with digital display in 0.01 increments		
	Range and Control	2 - 12 pH via PI control. Cascade to pumps, gases and/or loops from external devices		
DO	Sensor	Option of one or two Polaragraphic DO sensor with digital display in 0.1 % increments		
	Range and Control	0 - 200 % via PI control. Cascade to agitation, gases, pumps and/or loops from external devices		
Other Sensors	Foam/Level	Two foam/level sensor provided		
	Optional Sensors	Redox or 2nd pH sensor or 2nd DO sensor available		
Pumps	Standard, Options and Control	Three built-in, assignable, peristaltic pumps are standard. External pumps can be added. Control modes: Off, Prime, Base, Acid, Foam, Level 2 Wet, Level 2 Dry		
	Speed	Pumps 1 and 2: 12 rpm Fixed speed duty cycle, ability to view total pump flow rates Pump 3: 100 rpm Fixed speed duty cycle, ability to view total pump flow rates		
Utility Requirements and Connections	Process Air and Oxygen	20 PSIG (1.38 barg) each, with push on connection. (No requirement for instrument air)		
	Water Return	Maximum backpressure 5 PSIG (0.34 barg), accessed via Quick Connects		
	Facility Water	2 GPM (9.1 LPM) must be regulated to 10 PSIG (0.69 barg), accessed via Quick Connects		
	Electric Service	208 - 230 VAC, 50/60 Hz. Single phase, 15 Amps. (Fluctuations not to exceed ± 10 %)		
Input / Output Connections and Comm Ports (Built Into The Back Panel Of Master Control Station)	External Devices	Seven analog inputs and seven analog outputs for your external devices such as analyzers, sensors, external pumps, etc. (Reduce by 1 input and output for each additional TMFC added)		
	2 USB Ports	Import firmware/software upgrades and export trend data. Connect optional 8-port serial box		
	Communications Port	For optional BioCommand®/SCADA software		
Regulatory Compliance		c ETL US CE	CAN/CSA-C22.2 Nos. 1010.1 and 1010.2.010 UL Standard UL-61010A-1 and 61010A-2-010	

* Specifications are subject to change without notice. As shown, for operation as a fermentor. Optional impellers and accessories enable use as a cell culture system. Ask your Eppendorf sales representative for details. † In 14 & 19.5 L vessels, temperature rises are longer. ◊ Ambient operating conditions of 10 to 30 °C, up to 80 % relative humidity, non-condensing.

Your local distributor: www.eppendorf.com/contact
Eppendorf AG • 22331 Hamburg • Germany
newbrunswick@eppendorf.com

www.eppendorf.com

Culturefuge 100

Solids ejecting centrifuge

Many new biological products are derived from fragile organisms. Although relatively easy to separate the trick is accomplishing the separation in a gentle manner without destroying the shear sensitive cell wall membranes that isolate the complex intracellular proteins from the extracellular liquid. If this can be avoided, downstream purification of the target proteins becomes much easier.

Fig. 1 Culturefuge 100 centrifuge

Applications

The machine is designed for clarification duty. Especially when clarifying liquids from shear sensitive particles. Applications that requires low oxygen pick-up can also take advantage of the hermetic features this machine offers.

Standard design

The machine consists of a frame that has a horizontal drive shaft, worm gear, lubricating oil bath and hollow vertical bowl spindle in the lower part. The bowl is mounted on top of the spindle, inside the space formed by the upper part of the frame, the ring solids cover, the collecting cover, and the frame hood. The liquid discharge system also rests on this structure. All parts in contact with the process liquid are made of stainless steel. The bowl is of the solids-ejecting disc type with an automatic hydraulic operating system for discharging. It is a so-called timer triggered partial discharge system, meaning that only part of the bowl content is emptied during pre-set discharge intervals. The discharge takes place at full speed without any interruption of the feed. The centrifuge is available with main connections as sanitary flanges and all other utility connections clamp type. The electric motor is of standard type and has a built-in variable frequency drive. The design conforms with a number of EC directives, and machine is made in accordance with the general directives for machinery. Finally, the centrifuge is equipped with nozzles for flushing of the bowl top, the bowl bottom and the cyclone.

Standard equipment

Each Culturefuge 100 centrifuge comes with control unit, electric motor, in- and outlet connections, spare parts kit and set of tools.

Material data

Bowl body, hood and lock ring	s.s. 1.4462 UNS S31803
Solids cover and frame hood	s.s. ASME SA-240 UNS 31603
Cyclone	s.s. ASME SA-240 UNS 31603
Bottom frame	Cast grey iron
In and outlet	s.s. mostly 1.4401 UNS 31600
Gaskets and O-rings	EPDM rubber (FDA approved)

Operating principles

The feed is introduced to the rotating centrifuge bowl (fig 2) from the bottom through a hollow spindle (1), and is accelerated in a distributor (2) before entering the disc stack (3), where the separation takes place. The separated liquid phase leaves through the liquid outlet (4) at the top of the bowl. The collected solids in the solid space (5) are intermittently discharged from the periphery of the bowl. During normal production the operating water keeps the sliding bowl bottom (6) closed against the bowl hood (7). During discharge the sliding bowl bottom drops for a short time (less than a second) and the solids are ejected through the discharge ports (8). The high velocity of the ejected solids is reduced in the cyclone.

Available models

The Culturefuge 100 centrifuge is available in pressure vessel designs according to ASME or to PED. In addition, different surface finish executions are available:

Bowl spindle	Ra 0.8
Bowl spindle	Ra 0.5 and electropolished
Machine top part	Inside: Ra 0.8, Outlet cover: Ra 0.5 and electropolished
Machine top part	Inside: Ra 0.8, Outlet cover: Ra 0.8
Machine top part	Inside: Ra 1.2, Outlet cover: Ra 1.2
Separator bowl	Inside: Ra 0.5 and electropolished, Outside: Ra 0.8
Separator bowl	Inside: Ra 0.8, Outside: Ra 0.8
Separator bowl	Inside: Ra 1.2, Outside: Ra 1.2

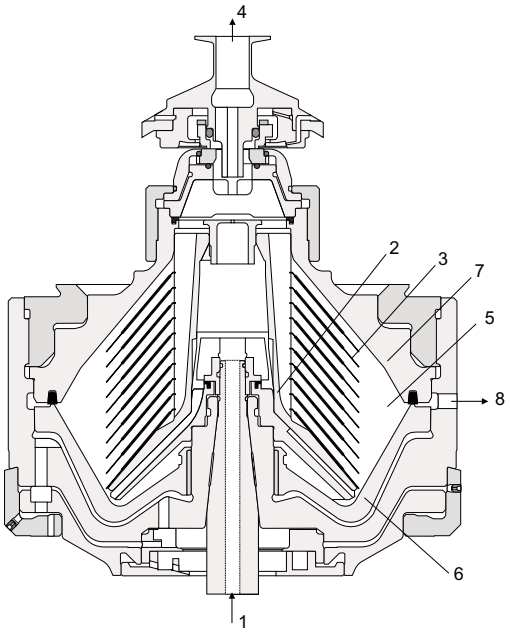


Fig. 2 Typical bowl for a hermetic solids-ejecting centrifuge. The details illustrated do not necessarily correspond to the centrifuge

Technical specification

Hydraulic capacity	max. 2.0 m^3/h [1]
G-force	max. 12200 g
Bowl speed	max. 9650 rpm
Motor power installed	7.5 kW
Sound pressure	74 dB(A) [2]
Overhead hoist lifting capacity	min. 100 kg

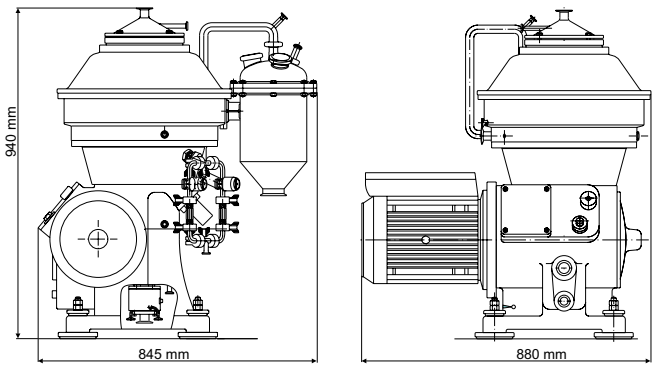
[1] Actual capacity depends on feed material and separation demands

[2] In compliance to EN ISO 4871

Utilities consumption

Electric power	5.5 kW
Operating water	0.3 l/discharge
Cyclone flush	0 - 8 l/discharge
Cooling for seals	max. 300 l/h
Flushing above the bowl	0 - 1 l/discharge
Flushing under the bowl	0 - 1 l/discharge
Steam per sterilization cycle	5 - 10 kg

Dimensions (approximate)



Shipping data (approximate)

Centrifuge with bowl with motor	450 kg
Gross weight	600 kg
Volume	1.0 m^3

How to contact Alfa Laval

Contact details for all countries are continually updated on our website. Please visit www.alfalaval.com to access the information direct.

INSTRUCTIONS

HisPur™ Cobalt Superflow Agarose

25228 25229 25230 25231

2407.0

Number	Description
25228	**HisPur Cobalt Superflow Agarose,** 10mL settled resin
25229	**HisPur Cobalt Superflow Agarose,** 50mL settled resin
25230	**HisPur Cobalt Superflow Agarose,** 250mL settled resin
25231	**HisPur Cobalt Superflow Agarose,** 1000mL settled resin

Binding Capacity: ≥ 30mg/mL of settled resin for a 27kDa 6xHis-tagged protein from a bacterial source

Resin: Highly crosslinked 6% Superflow agarose

Supplied: 50% slurry in a 20% ethanol solution

Storage: Upon receipt store at 4°C. Product shipped at ambient temperature.

Table of Contents

Introduction

The Thermo Scientific HisPur Cobalt Superflow Agarose enables high-yield, high-purity purification of polyhistidine-tagged proteins. This immobilized metal affinity chromatography (IMAC) purification support consists of a cobalt-charged tetradentate chelator immobilized onto Superflow 6% agarose, which exhibits minimal metal leaching and is compatible with a wide range of chemicals and pH values. The highly crosslinked Superflow Support has a high dynamic binding capacity across a wide range of flow rates and is stable to multiple reuses making it ideal for large-scale FPLC purifications.

The HisPur Cobalt Superflow Agarose protocol uses gentle wash and elution conditions to typically produce > 90% pure target protein after purification. Protein purities achieved with the Cobalt Superflow Resin are generally higher than those achieved with nickel IMAC resins, resulting in the Cobalt Superflow Resin being a valuable tool for users interested in one-step purifications.

Table 1. Properties of Thermo Scientific HisPur Cobalt Superflow Agarose.

Support:	Superflow 6 Resin, 6% highly crosslinked agarose
Bead Size:	60-160µm
Recommended Linear Flow	≤ 150cm/hr (for binding, washing and elution steps)
Maximum Linear Flow Rate†:	1200cm/hr
Metal Loading:	≥ 11µMol Co^{2+}/mL resin
Binding Capacity:	Typical Static Capacity: ~30mg 6xHis-GFP/mL resin Typical Dynamic Capacity*: ~20mg 6xHis-GFP/mL resin
Chemical Compatibility:	1M acetic acid, pH 2; 1% SDS, pH 7; 6M guanidine HCl; 70% ethanol: ≥ 1 week at 37°C 8M urea, 10mM DTT, 5mM TCEP: ≥ 2 hours at 22°C
pH Limits:	pH 3-9 (≥1 week at 4°C) pH 2-3 or 9-12 (≥ 2 hours at 22°C)
Storage Solution:	20% ethanol
Reuse:	Up to 25 times

†Maximum linear flow-rate conditions:
Column dimensions (w × h): 13mm × 38mm (5mL resin)
Ultrapure water at room temperature
Linear flow rate = volumetric flow rate (mL/min) × 60 (min/hr)/cross sectional area (cm^2)

*Dynamic binding conditions (10% breakthrough):
Sample: 1mg/mL 6xHis-GFP (27kDa) pure protein in 20mM NaH_2PO_4, 300mM NaCl, 5mM imidazole
Column dimensions (w × h): 5mm × 50mm (1mL resin)
Flow rate: 1mL/min

Important Product Information

- Protein yield and purity are dependent upon the expression level, conformation and solubility characteristics of the recombinant fusion protein, as well as the buffer conditions and flow rates used. Therefore, it is important to optimize these parameters before attempting a large-scale purification. For best results, perform a small-scale test to estimate the expression level and determine the solubility of each His-tagged protein. Decreasing the flow rate during the sample load will increase binding capacity.
- To avoid sample loss, try not to exceed the maximum resin binding capacity for the target protein for the purification conditions used. Volumes will vary based on the protein and expression efficiency and will have to be determined and optimized for each over-expressed protein. Typically over-expressed proteins represent 1-30% of the final sample protein concentration. Adjust resin or sample volume as appropriate.
- Optimization of the lysis procedure is critical for maximizing protein yield. Some methods for protein extraction include using commercially available detergent-based reagents, such as Thermo Scientific B-PER Bacterial Protein Extraction Reagent with Enzymes (Product No. 90078), and mechanical methods, such as freeze/thaw cycles, sonication or French press. Add EDTA-free protease inhibitors, such as Thermo Scientific Halt Protease Inhibitor Cocktail, EDTA-free (Product No. 78437), to protect proteins from degradation.
- Overexpressed proteins can be sequestered in inclusion bodies. The Cobalt Superflow Resin is compatible with purification under native or denaturing conditions. Inclusion bodies containing His-tagged proteins can be solubilized in 8M urea, 6M guanidine or Thermo Scientific Inclusion Body Solubilization Reagent (Product No. 78115) and purified with the Cobalt Superflow Resin, but a denaturant must be added to buffers so the protein remains soluble throughout the procedure.
- For liquid chromatography (LC) applications, use highly pure buffer components and ultrapure water. Use low-absorbance imidazole (Fisher Scientific, Product No. BP 305-50) to avoid UV interference. Degas or filter buffers through a 0.45µm filter before use.
- Avoid using chelators such as EDTA, which will disrupt the function of the cobalt resin and potentially strip cobalt from the resin.
- Reducing agents, such as 10mM DTT and 5mM TCEP, have been tested and do not affect function of the resin; however, avoid using higher concentrations of these reducing agents.

Recommended Buffers

Note: For some specific proteins or expression systems, adjustments to the imidazole concentration may be required to decrease nonspecific binding or increase yield.

For native conditions:

- Equilibration Buffer: 20mM sodium phosphate, 300mM sodium chloride, 5mM imidazole; pH 7.4
- Wash Buffer: 20mM sodium phosphate, 300mM sodium chloride, 10-15mM imidazole; pH 7.4
- Elution Buffer: 20mM sodium phosphate, 300mM sodium chloride, 150mM imidazole; pH 7.4

For denaturing conditions:

- Equilibration Buffer: 20mM sodium phosphate, 300mM sodium chloride, 6M guanidine•HCl, 5mM imidazole; pH 7.4
- Wash Buffer: 20mM sodium phosphate, 300mM sodium chloride, 6M guanidine•HCl, 10-15mM imidazole; pH 7.4
- Elution Buffer: 20mM sodium phosphate, 300mM sodium chloride, 6M guanidine•HCl, 150mM imidazole; pH 7.4

For regeneration:

- MES Buffer: 20mM 2-(*N*-morpholine)-ethanesulfonic acid, 0.1M sodium chloride; pH 5.0
- Ultrapure water
- 20% ethanol in ultrapure water

For clean-in-place:

- 6M guanidine•HCl with 1% non-ionic detergent
- Ultrapure water

Procedure for Purification of His-tagged Proteins Using an LC System

Note: Monitor and collect all fractions during a purification to avoid accidental loss of target protein. User can adjust sample collection based on their needs and comfort level with the purification methods used. Maximum flow rates will be dependent on application and equipment used. The procedure may be performed at room temperature or 4°C.

Additional Materials Required

- Suitable LC system
- Empty column for resin packing (follow column manufacturer's protocol for packing)
- Recommended buffers (see Recommended Buffers Section) and volumes (see below)

1. Pack an appropriate-sized column with resin according to column manufacturer's protocol. Ensure the packing flow rate is at least 20% faster than the flow rate to be used during purification.
2. Equilibrate the column and all buffers to working temperature. Perform purifications at room temperature or at 4°C. Ensure that all solutions are degassed.
3. Prepare the LC system by washing pumps and filling tubing with buffer. To avoid introducing air into the system, allow a few drops of buffer to flow from the tubing into the column top. Connect column to the tubing.
4. Equilibrate the column with 5-10 column volumes of the Equilibration Buffer at a flow rate of 300cm/hr or less (150cm/hr recommended).

5. Apply any sample volume that does not exceed column-binding capacity for target protein at a flow rate of 300cm/hr or less (150cm/hr recommended).

   Note: Binding capacity is flow rate- and protein-dependent. Decreasing the flow rate during the sample load will increase binding capacity. Higher flow rates will decrease production time but may result in losing a small portion of the target protein in the flow-through fraction.

   Note: For maximum binding, prepare sample by mixing protein extract 1:1 with Equilibration Buffer (to adjust the sample to the ionic strength and pH of the Equilibration Buffer) before applying to the column.

   Note: If the sample contains insoluble matter, centrifuge or filter (0.45µm filter) before use.

6. Wash the resin at a flow rate of 300cm/hr or less (150cm/hr recommended) with 10-15 column volumes of Wash Buffer or until the absorbance approaches baseline.

   Note: Due to the gentle binding characteristics of cobalt IMAC resin, excessive washing can elute target protein from the column.

7. Elute at a flow rate of 300cm/hr or less (150cm/hr recommended) with approximately 5-10 column volumes of Elution Buffer and collect fractions.

   Note: Monitor protein elution by measuring the absorbance of the fractions at 280nm. The eluted protein can be directly analyzed by SDS-PAGE. To remove excess imidazole for downstream applications, use gel filtration or dialysis (e.g., Thermo Scientific Zeba Spin Desalting Columns or Slide-A-Lyzer Dialysis Cassettes; see the Thermo Scientific Related Products Section).

8. Regenerate column by washing with 10 column volumes of Regeneration Buffer, followed by 10 column volumes of ultrapure water at a flow rate of 300cm/hr or less (150cm/hr recommended). The column is now ready for reuse (return to step 1), storage (proceed to step 9) or routine clean-in-place procedures (see the Procedure for Resin Cleaning-In-Place).

9. For storage, equilibrate the column with 5 column volumes of 20% ethanol. Seal column and store at 4°C.

Procedure for Purification of His-tagged Proteins by Batch Method

Note: The procedure may be performed at room temperature or 4°C. The Cobalt Superflow Resin allows for customization of your purification strategy. Purification conditions can be scaled as needed and performed in several formats. A batch method based on centrifugation is included below. Alternatively, methods based on vacuum filtration or gravity flow can be used to collect flow-through, wash and elution fractions.

A. Additional Materials Required

- Sample-handling containers, such as centrifugation bottle or spin filters/columns
- Recommended buffers (see Recommended Buffers Section) and volumes (see below)
- End-over-end rotary mixer or equivalent mixing apparatus

B. Procedure

1. Add the required amount of Cobalt Superflow Resin to a container with 3-4X greater volume. Centrifuge for 2 minutes at 700 × *g* and carefully remove and discard the supernatant.
2. Add two resin-bed volumes of Equilibration Buffer and mix until the resin is fully suspended.
3. Centrifuge for 2 minutes at 700 × *g* and carefully remove and discard buffer.
4. Prepare sample by mixing the protein extract with Equilibration Buffer to a volume greater than or equal to the resin bed volume.
5. Add the prepared protein extract to the tube and mix slowly for 30 minutes ensuring the resin remains suspended. For best results, use an end-over-end rotary mixer.
6. Centrifuge for 2 minutes at 700 × *g* and carefully remove supernatant. If desired, save supernatant for downstream analysis.

7. Wash the resin with two resin-bed volumes of Wash Buffer. Centrifuge for 2 minutes at 700 × *g* and carefully remove supernatant. If desired, save supernatant for downstream analysis.
8. Repeat wash step three times.
9. Elute bound His-tagged proteins by resuspending resin bed in one resin-bed volume of Elution Buffer. Mix slowly for 10 minutes ensuring that resin remains suspended.
10. Centrifuge for 2 minutes at 700 × *g*. Carefully remove and save the supernatant.
11. Repeat elution steps 9-10 two to four times, saving each supernatant fraction in a separate tube.
12. Monitor protein elution by measuring the absorbance of the fractions at 280nm or by Thermo Scientific Coomassie Plus (Bradford) Assay Reagent (Product No. 23238) or Pierce 660nm Protein Assay (Product No. 22660). Dilute the samples at least 1:2 to decrease the imidazole concentration before performing the protein assay to avoid interference with the assay. The eluted protein can be directly analyzed by SDS-PAGE. To remove excess imidazole for other downstream applications, use gel filtration or dialysis (e.g., Thermo Scientific Zeba Spin Desalting Columns or Slide-A-Lyzer Dialysis Cassettes).
13. Regenerate the resin by resuspending resin with 10 resin-bed volumes of Regeneration Buffer. Centrifuge tube for 2 minutes at 700 × *g* and discard the supernatant. Repeat once.
14. Resuspend resin with 10 resin-bed volumes of ultrapure water. Centrifuge tube for 2 minutes at 700 × *g*. Discard supernatant. Repeat once. The column is now ready for reuse (return to step 1), storage (proceed to step 15) or routine clean-in-place procedures (see the Procedure for Resin Cleaning-in-Place).
15. For storage, re-suspend resin with 1 column volume of 20% ethanol. Seal column and store at 4°C.

Procedure for Resin Cleaning-in-Place

Note: The Cobalt Superflow Resin can be used multiple times without affecting protein yield or purity. To prevent cross contamination of samples, designate a given column to one specific fusion protein. If an increase in backpressure is observed, the following cleaning procedures can be followed.

1. To remove precipitated or denatured proteins and hydrophobic substances, wash resin with 2 volumes of 6M guanidine•HCl plus 1% nonionic detergent (e.g., Thermo Scientific Triton X-100 Surfact-Amps Detergent Solution, Product No. 28314) with 10 minutes of contact time, followed by 5 volumes of ultrapure water at a flow rate of less than 300cm/hr (150cm/hr recommended).
2. Store resin in 20% ethanol at 4°C.

Troubleshooting

Problem	Possible Cause	Solution
Low protein yield	Poor expression of soluble protein	Optimize expression conditions (e.g., lower temperature during induction, vary induction time, optimize codon usage for expression system)
	His-tagged protein formed inclusion bodies	Alter growth conditions to minimize inclusion body formation and maximize soluble protein yield; alternatively, solubilize inclusion bodies and perform the purification with a compatible denaturant (e.g., Thermo Scientific Inclusion Body Solubilization Reagent, Product No. 78115)
	Insufficient cell lysis and extraction	Optimize cell lysis protocol
	Fusion protein did not bind to the column	Verify the sequence
		Perform an ELISA or Western blot using an antibody against the His tag to confirm the His tag is present
	Flow rate was too fast	Decrease flow rate during binding to allow for greater residence time and increased binding of fusion protein
	Column washing was too extensive	Reduce imidizole concentration in wash buffer
		Reduce amount of wash buffer used
Poor protein purity	Insufficiently washed	Increase duration of wash
		Modify imidazole concentration and pH of the Equilibration or Wash Buffer
	Column was dirty	Follow clean-in-place procedure to remove nonspecifically bound proteins
Slow column flow	Column was overloaded	Apply less protein extract onto the column
	Extract was too viscous or highly particulate	Dilute lysate with Equilibration Buffer to decrease viscosity
		Centrifuge lysate at higher speed to remove particulate

Related Thermo Scientific Products

25214-7 **HisPur Ni-NTA Superflow Agarose**

25236-9 **Pierce™ Glutathione Superflow Agarose**

89896-8 **Pierce Centrifuge Columns**

87785 **Halt™ Protease Inhibitor Cocktail (100X), EDTA-free**

88661 **Pierce Protease Inhibitor Tablets, EDTA-free**

88270 **Pierce High Capacity Endotoxin Removal Resin**

90078 **B-PER™ Bacterial Protein Extraction Reagent with Enzymes**

88282 **Pierce LAL Chromogenic Endotoxin Quantitation Kit**

23238 **Coomassie Plus™ (Bradford) Assay Reagent**

22660 **Pierce 660nm Protein Assay Reagent**

78115 **Inclusion Body Solubilization Reagent**

89891-4 **Zeba Spin Desalting Columns, 7K MWCO**

87730-8 **Slide-A-Lyzer™ G2 Dialysis Cassettes, 10K MWCO**

28313-4 **Triton™ X-100 Surfact Amps™ Detergent Solution**

Triton is a trademark of The Dow Chemical Company.

This product ("Product") is warranted to operate or perform substantially in conformance with published Product specifications in effect at the time of sale, as set forth in the Product documentation, specifications and/or accompanying package inserts ("Documentation") and to be free from defects in material and workmanship. Unless otherwise expressly authorized in writing, Products are supplied for research use only. No claim of suitability for use in applications regulated by FDA is made. The warranty provided herein is valid only when used by properly trained individuals. Unless otherwise stated in the Documentation, this warranty is limited to one year from date of shipment when the Product is subjected to normal, proper and intended usage. This warranty does not extend to anyone other than the original purchaser of the Product ("Buyer").

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Current product instructions are available at *www.thermoscientific.com/pierce*. For a faxed copy, call 800-874-3723 or contact your local distributor.

Pierce Biotechnology 3747 N. Meridian Road | PO Box 117 Rockford, IL 61105 USA | (815) 968-0747 (815) 968-7316 fax | www.thermoscientific.com/pierce

840 series hygienic pump

800 series
Watson-Marlow Pumps

FEATURES

- Flow rates from 653 L/h to 8,140 L/h (1.33 L/rev)
- Clean-in-place and steam-in-place sterilisation at full velocity with no bypass required
- Bioprene tube to USP Class VI and hygienic stainless steel connectors certificated to 3.1.B BS/EN 10204.
- Bioprene tubing available in two hardnesses for pressures up to 2 bar and 3.5 bar.
- Pumpheads to accept B5 output flange-mounted gear motors in a range of single and three phase fixed and electronic variable speeds
- Hinged door with only two captive bolts makes tube inspection and change extremely simple and safe

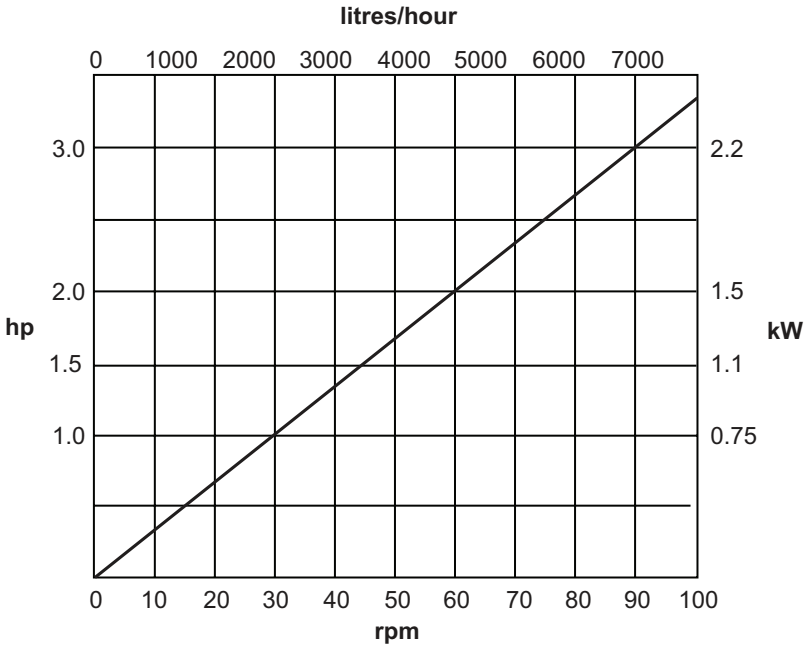
2 warranty

24 hour duty

Watson-Marlow...Innovation in Full Flow

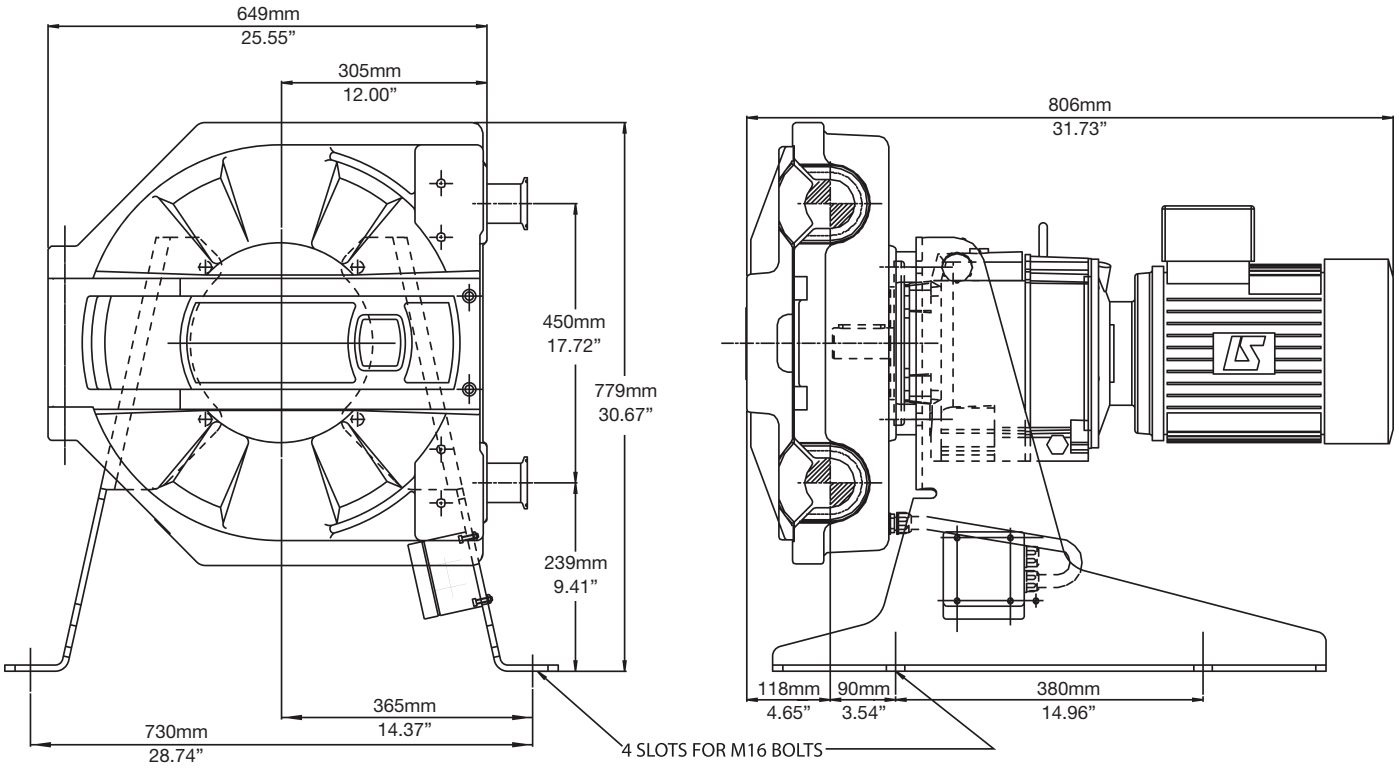
PERFORMANCE

Speed (rpm)	Flow rate (L/hr)	Fitted motor power (kW)	Speed (rpm)	Flow rate (L/hr)	Fitted motor power (kW)
8	653	0.55	42	3352	1.1
10	790	0.55	47	3751	1.5
13	1045	0.55	52	4150	1.5
16	1277	0.55	58	4628	1.5
22	1756	0.75	65	5187	1.5
25	1995	0.75	73	5825	2.2
29	2314	1.1	79	6304	2.2
33	2649	1.1	89	7102	2.2
36	2905	1.1	102	8140	3



For variable speed units and other build options, please contact your local representative, or the Watson-Marlow aApplications Engineering department.

TECHNICAL DRAWINGS



TECHNICAL SPECIFICATIONS

Environment temperature	5C to 40C (40F to 104F)
Fluid temperature	0C to 80C (-32F to 175F)
Max pressure	3.5 bar
Pump weight	Dependant upon drive. Nominally 130kgs
Noise	<75dB(A) at 1m

MATERIALS OF CONSTRUCTION

Component	Material
Pumphead body	Aluminum alloy with epoxy polyester powder coat finish
Pumphead door	Aluminum alloy with epoxy polyester powder coat finish
Pumphead rotor hub	Aluminum alloy with epoxy polyester powder coat finish
Rotor rollers	Stainless steel 316
Support frame	Stainless steel 304
Door fixings	Stainless steel
Motor fixings	Zinc plated high tensile steel bolts, stainless steel nuts and washers
Frame fixings	Stainless steel

All flow rates shown were obtained pumping water at 20C (68F) with zero suction and delivery heads. Watson-Marlow, Bioprene and Marprene are trademarks of Watson-Marlow Limited. Disclaimer: The information contained in this document is believed to be correct but Watson-Marlow Limited accepts no liability for any errors it contains, and reserves the right to alter specifications without notice. LoadSure is a trademark of Watson-Marlow Limited.
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Please state the product code when ordering pumps and tubing.

WATSON MARLOW Pumps
wmpg.co.uk
info@wmpg.co.uk
+44 (0) 1326 370370

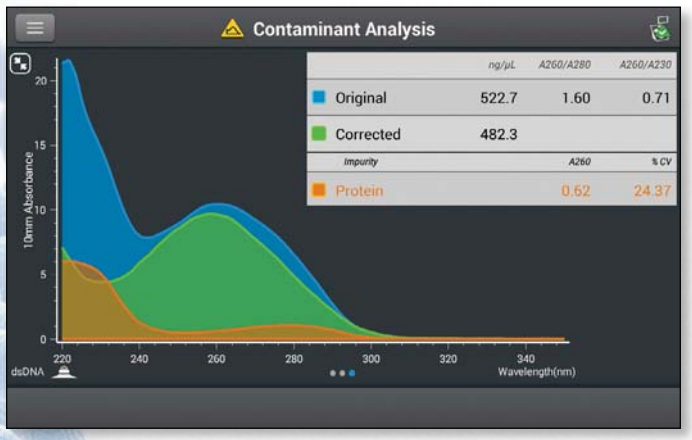
Move one step closer to success with NanoDrop One

Quantify and qualify DNA, RNA, and protein samples with only 1–2 µL in seconds with no dilutions using the Thermo Scientific™ NanoDrop™ One microvolume UV-Vis spectrophotometers. Gain a more complete understanding of sample quality before using samples in downstream applications with Thermo Scientific™ Acclaro™ Sample Intelligence technology built into every NanoDrop One instrument.

Know more with Acclaro Sample Intelligence technology

Save days of troubleshooting experiments when you make informed decisions on sample suitability for your application. Acclaro technology offers enhanced sample analysis with:

- Contaminant identification and corrected results
- Instant feedback about sample quality with on-demand technical support and guided troubleshooting
- Embedded sensor and digital image analysis that ensures measurement integrity



Acclaro technology detects dsDNA sample contaminated with protein. The absorbance contribution from the protein (orange) is subtracted from the original result (blue) to obtain the corrected dsDNA concentration (green).

Pipette. Measure. Know.

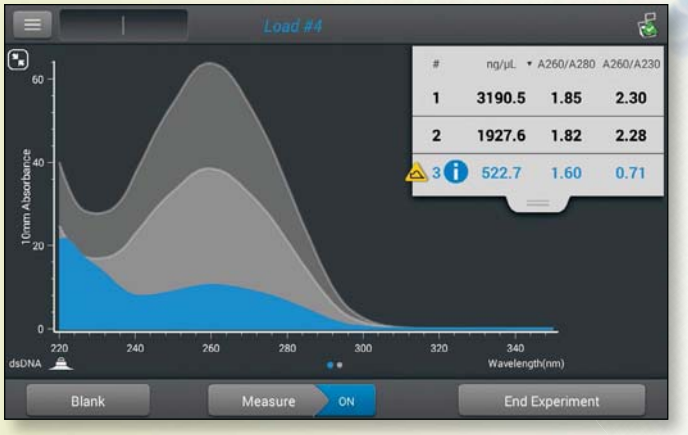
- **Fast and easy evaluation** of nucleic acids and proteins with Auto-Measure and pre-programmed applications
- **Modern standalone design** with local control and high-resolution touchscreen interface saves bench space
- **Accurate measurements up to 27,500 ng/µL** (dsDNA) with extended auto-range pathlength technology
- **No consumables needed** – pipette directly onto the pedestal sample-retention system
- **Enhanced connectivity with data transfer via** USB, Ethernet, Bluetooth® and Wi-Fi options; PC software available for data management
- **Optional cuvette position** for measuring dilute solutions and performing temperature sensitive experiments

Thermo SCIENTIFIC

Qualify nucleic acid samples

Accurate concentration and purity evaluation of RNA and DNA samples is critical to the success of your downstream experiments. Choose from a selection of pre-programmed applications that include:

- Direct A260 quantitation; custom factor option
- A260/A280 purity ratios
- A260/A230 purity ratios
- Labeled Nucleic Acids quantitation
- Oligo Calculator

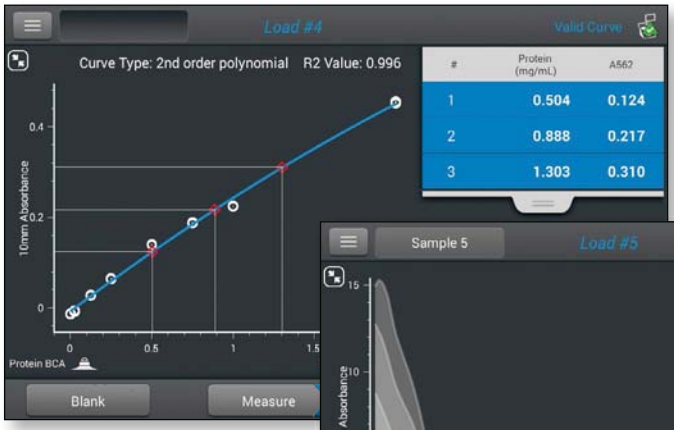


View multiple samples at once on your measurement screen. Here Acclaro has flagged dsDNA sample #3 for the presence of a contaminant.

Evaluate protein samples effectively

Quantify protein samples accurately and reproducibly. A selection of pre-programmed protein applications guides you to high-quality results.

- Direct A280 quantitation with Protein Editor
- A205 peptide bond absorbance
- Quantitation of labeled proteins
- Colorimetric protein assays: Bradford, BCA, Lowry and Thermo Scientific™ Pierce™ 660 nm Protein Assay



Total protein concentration for three samples shown as red squares on a BCA assay standard curve.

Four different purified protein samples were measured using the Protein A280 application.

FREE Trial Program

Request a trial instrument to evaluate in your lab at: **www.thermoscientific.com/nanodrop**

Ordering Information

Instruments	Product Number
NanoDrop One spectrophotometer *(Pedestal position only)*	ND-ONE-W*
NanoDrop One^{C} spectrophotometer *(Pedestal and cuvette positions)*	ND-ONEC-W*
Accessories and Consumables	**Product Number**
NanoDrop One Productivity kit *(Contains: 0.2–2.0 µL pipette, screen wipe, USB device, PR-1 kit, and PV-1 solution)*	ND-PP1
NanoDrop One^{C} Productivity kit *(Contains: 0.2–2.0 µL pipette, screen wipe, USB device, PR-1 kit, PV-1, stir bars, and quartz cuvette)*	ND-PP1C
DYMO® LabelWriter® 450 printer with labels	PNTR-LW400
PR-1 Reconditioning Compound kit	CHEM-PR1-KIT
PV-1 Performance Verification solution	CHEM-PV-1

* Wi-Fi model not available in all countries. Please contact your NanoDrop distributor to confirm the correct part number in your region.

Contact your local NanoDrop representative.

Email nanodrop@thermofisher.com for ordering information and technical support.

NanoDrop One. One step closer.

www.thermoscientific.com/nanodrop

FL52746_E 09/15M

Thermo SCIENTIFIC

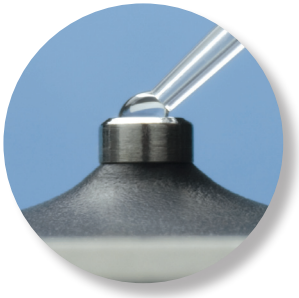
A Thermo Fisher Scientific Brand

Thermo Scientific NanoDrop Products

Instruments for microvolume analysis of biomolecules

Revolutionary technology. Elegant simplicity.

Pipetting 1 µL onto pedestal



Measuring 1 µL sample

NanoDrop™ 2000c Spectrophotometer

Full-spectrum microvolume and cuvette measurements in a single instrument

A full-spectrum UV-Vis spectrophotometer offering a complete solution by integrating both a patented sample-retention system* for microvolume samples and a cuvette option. Dual sampling modes accommodate very low and very high-concentration samples.

NanoDrop™ 2000 Spectrophotometer

Full-spectrum microvolume measurements

Provides the same accuracy, full-spectrum analysis, and benefits as the NanoDrop 2000c without the added flexibility and sensitivity of the cuvette option.

NanoDrop™ 8000 Spectrophotometer

Higher throughput, full-spectrum microvolume measurements

Delivers full-spectrum UV-Visible absorbance measurements for up to eight samples at one time. Use an eight-channel pipette to dispense samples from tubes or plates onto a linear array of pedestals, measure and wipe clean.

NanoDrop™ Lite Spectrophotometer

Basic microvolume measurements

A basic microvolume instrument which uses our patented sample-retention system to deliver the same exceptional accuracy and reproducibility as other NanoDrop instruments. It's ideal for labs looking for trusted NanoDrop technology, but not requiring the full performance of a NanoDrop 2000|2000c or NanoDrop 8000.

NanoDrop™ 3300 Fluorospectrometer

Full-spectrum microvolume fluorescence measurements

Brings the sensitivity and selectivity of fluorescence spectroscopy to microvolume samples. Using our patented sample-retention technology, it performs broad-spectrum fluorescence analysis in a versatile, high-performance instrument.

*Patents US6628382 and US6809826

Thermo SCIENTIFIC

NanoDrop 2000c|2000
Spectrophotometer

NanoDrop Lite
Spectrophotometer

NanoDrop 8000
Spectrophotometer

NanoDrop 3300
Fluorospectrometer

Choose the NanoDrop instrument that's right for you

	UV-Vis				Fluorescence
	NanoDrop 2000c	**NanoDrop 2000**	**NanoDrop 8000**	**NanoDrop Lite**	**NanoDrop 3300**
Full-Spectral Data	✓	✓	✓		✓
Pre-Programmed Methods for Nucleic Acids	✓	✓	✓	✓	✓
Measures Nucleic Acids 260/280 Ratio	✓	✓	✓	✓	
Measures Nucleic Acids 260/230 Ratio	✓	✓	✓		
Pre-Programmed Methods for Protein	✓	✓	✓	✓	✓
Measures Purified Protein A280	✓	✓	✓	✓	
Pre-Programmed Methods for Colorimetric Assays (i.e. BCA)	✓	✓	✓		
Measures Fluorescently-Labeled Samples	✓	✓	✓		✓
Custom Methods Editor	✓	✓	✓		✓
PC Based	✓	✓	✓		✓
Built-in Cuvette Mode	✓				
Qualification Procedures IQ/OQ	✓	✓	✓		
User Calibration Verification	✓	✓	✓	✓	

Realize the difference.
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Our trial program allows you to try any NanoDrop instrument in your lab with your own samples, completely free of charge. Once you have used a NanoDrop instrument, we are confident you will quickly realize the benefits of our revolutionary technology.

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FREE Trial Offer!

Thermo SCIENTIFIC
Part of Thermo Fisher Scientific

MILLIPORE

- *Leading-edge void-free membranes to match virtually any separation challenge*
- *Short flow path for higher flux and higher resolution separation capability*
- *Choice of flow channel configuration providing process optimization capability*
- *Predictable, fast, scale-up*
- *True linear scalability from laboratory size modules to industrial assemblies for processing thousands of liters*

DATA SHEET

Pellicon® 2 Filters and Holders

High-performance tangential flow filters for biopharmaceutical process development, scale-up/scale-down and concentration/ purification/cell harvesting applications

Typical Applications

Concentration, desalting or buffer exchange of:

- Protein solutions
- Polysaccharide solutions
- Virus suspensions

Harvest, washing or clarification of:

- Cell cultures and lysates
- Colloidal suspensions
- Viral cultures

Superior TFF Performance

For research, process development, scale-up and production, Pellicon 2 filters and holders offer the following benefits:

Consistent High Flux and High Product Recovery

Millipore's Biomax® polyethersulfone and Ultracel® PLC-composite regenerated cellulose membranes have void-free structures that guard against leakage of solutes through microdefects normally associated with voids beneath the thin skins of conventional UF membranes (Figures 1 and 2).

These void-free membranes are more permeable, resulting in high-flux with equivalent or superior product retention (Figure 3). These void-free membranes provide the advantages of fast, high yield processing and smaller systems.

The long established Durapore® hydrophilic PVDF microfiltration membrane is well known for its exceptional combination of high flux, low protein binding and high product recoveries.

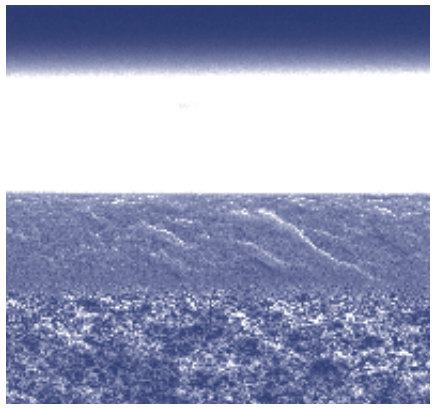


Figure 1. *Void-free Biomax 10 modified polyethersulfone membrane*

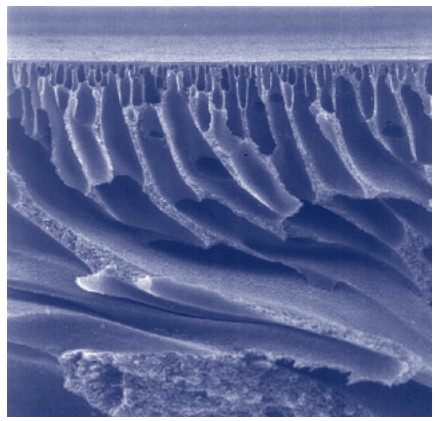


Figure 2. *Conventional 10 kD polyethersulfone membrane with sub-surface voids*

Easy, Reliable Linear Scale-Up from the Lab to the Production Plant

Pellicon 2 Mini filters scale-up easily and reliably from the laboratory to the production plant (Figures 4 and 5). By ensuring every flow channel has the same length, height and turbulence promoter as well as flow direction and materials of construction, we maintain the same ultrafilter/microfilter performance at all scales. Thus, rapid and reliable translation of processes from lab to manufacturing scale is easily achieved.

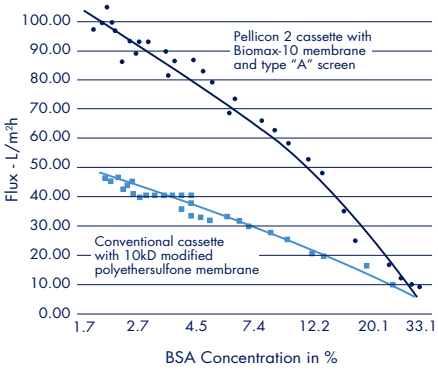
Linear Scale-Up

Mini filters (0.1 m^2/1.1 ft^2) and holders are designed for laboratory ultrafiltration/microfiltration of 100 mL to 10 L volumes, yet scale up linearly to Pellicon 2 Cassette (0.5 m^2/5.4 ft^2) and Maxi (2.5 m^2/26.9 ft^2) filters used in the pilot or manufacturing plant to process volumes from one liter to thousands of liters.

Thus, whether you operate 0.1 m^2 or 100 m^2 of installed area, every Pellicon 2 filter operates with the same pressure drop, flow velocity and concentration profile for true, rapid and simple linear scale-up.

Pellicon 2 Filters Proof of Performance

Improved Flux



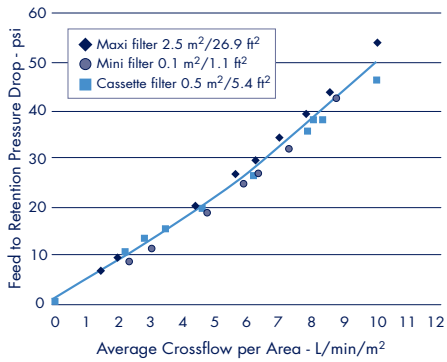
Feed pressure: 5.6 bar/80 psi
Retentate pressure: 2.1 bar/30 psi
Temperature: 10 – 13.5 °C
Initial volume 28 L
Final volume: 2 L

Conclusion

Pellicon 2 filters with Biomax membranes provide up to two-times the process flux of conventional cassettes resulting in faster processing and smaller systems.

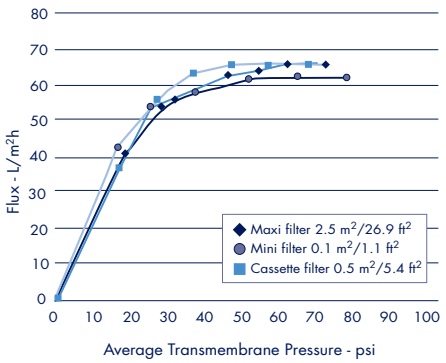
Figure 3. *Flux versus BSA concentration*

Linear Scalability



Temperature: 8 °C

Figure 4. *Feed to retentate pressure drop versus average crossflow on a 10% BSA solution*



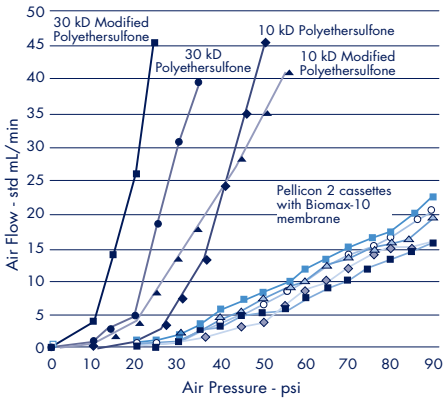
Temperature: 8 °C
Feed to retentate pressure drop: 2.8 bar/40 psi

Conclusion

(Figures 4 and 5) Pellicon 2 family of cassette filters scale linearly from 0.1 to 0.5 to 2.5 m^2 (1.1 to 5.4 to 26.9 ft^2) sizes for rapid, accurate and safe process scale-up and transfer.

Figure 5. *Flux versus average transmembrane pressure on a 10% BSA solution.*

Improved Reliability



Conclusion

The void-free structure of Biomax membranes is demonstrated by low, linear air diffusion values. This performance ensures better process reliability and safety and better product retention for higher yields.

Figure 6. *Integrity test comparison-air flow through wetted cassettes*

Greater Process Reliability and Reproducibility

The combination of defect-free membranes with Millipore's highly reliable manufacturing processes, offers greater consistency of process parameters.

The high quality of Millipore's ultrafiltration membranes is further ensured by our pioneering multiple-solute mixed-dextran retention profile test. Unlike the single solute protein retention test, Millipore's retention profile test measures and ensures reproducible retention performance of our UF membranes over the entire range of molecular weights retained by the membrane, not ¡ust at one or two molecular weights.

Low Product Loss

Pellicon 2 filters have a low minimum working volume – as low as 175 mL of retentate volume per square meter of membrane area. This low retentate volume permits high concentration factors to be reached with low starting volumes and maximizes the recovery of small sample volumes.

To prevent product loss, Pellicon 2 filters are 100% tested in manufacturing to ensure that every filter is integral.

In addition, Biomax and Ultracel membranes are exposed to a new high-pressure integrity test that provides greater sensitivity. The integrity test procedure and specifications are supplied so users can confirm integrity at high pressure when the filter is installed (Figure 6).

Biocompatibility

All wetted parts have been tested and meet the requirements of the USP Class VI biological test for plastics.

Superior Filter Quality

Pellicon cassettes are subjected to a complete array of quality control release tests.

A Certificate of Quality is included with every cassette.

Each casette is identified with a unique serial number.

Validatable

Since 1973, Pellicon filters and systems have been successfully used for development and scale-up of processes for manufacturing injectable protein and polysaccharide drugs, in the serum fractionation, biotechnology, vaccine and pharmaceutical industries.

Pellicon 2 filters and systems were developed based upon Millipore's experience serving these applications, and are supported by an extensive Validation Support Data Package proving performance claims and demonstrating the suitability of these filters for drug manufacturing in validated processes. This package is available upon request.

Millipore can further assist your validation efforts through:

- Design and fabrication of standard and custom turnkey TFF systems for drug manufacturing facilities
- Installation and operational qualification services for these systems
- Validation support services for tangential flow filter use in drug manufacturing processes.
- Training on TFF process scale-up, optimization and development.

A Choice of Feed Channel Screens

For optimal performance in a range of applications Pellicon 2 filters incorporate three types of feed-channel screens:

- *Type A screen (tight screen)* is optimized to operate Biomax membranes with maximum flux with low-viscosity solutions.
- *Type C screen (coarse screen)* is optimized to operate PLC series membranes with maximum flux. The Type C screen is also available with Biomax-50, 100, 300, 500 and Biomax 1000 membranes for concentration and diafiltration of viscous solutions.
- *Type V screen (open channel)* is optimized for very viscous solutions or solutions with higher levels of suspended solids.

Normalized Recirculation Rates

Parameter	Unit	Typical ΔP		
Screen Type		A	C	V
Recirculation Rate	L/min/m^2	4/6	5/35	
Differential Pressure	bar/psi	1.4/20	0.4/6	

Screen Selection Guidelines

Solution Type	Screen Type
Dilute protein solution or low viscosity solutions (MAbs, interferons)	A screen (tight screen)
Concentrated protein solutions or high viscosity solutions (IgG, biopolymers)	C screen (course screen)
High viscosity solutions (polysaccharides, certain microfiltration or clarification applications)	V screen (loose screen)

For More Detailed Information

Request literature number P17512 – User Guide for Pellicon Filters.

Specifications

Temperature Range

Mini, Cassette and Maxi:
4 to 50 °C

Maximum Forward Transmembrane Pressure

Device Size (m^2)	Biomax	Ultracel
0.1	6.8 bar (100 psi) Max	6.8 bar (100 psi) Max
0.5	6.8 bar (100 psi) at 30 °C	3.4 bar (50 psi) at 30 °C
2.5	6.8 bar (100 psi) at 30 °C	3.4 bar (50 psi) at 30 °C

Maximum Reverse Transmembrane Pressure

Device Size (m^2)	Biomax	Ultracel
0.1	0.33 bar (5 psi)	0.33 bar (5 psi)
0.5	0.33 bar (5 psi)	0.33 bar (5 psi)
2.5	0.33 bar (5 psi)	0.33 bar (5 psi)

Prefiltration Required

Mini, Cassette and Maxi:
100 µm

Dimensions

Device	Width	Length	Thickness
Mini	5.6 cm	21 cm	1.5 cm (V screen-2.16 cm)
Cassette	17.8 cm	21 cm	1.5 cm (V screen-2.16 cm)
Maxi	17.8 cm	21 cm	7.6 cm (V screen-9.0 cm)

Membrane Selection Guideline

Membrane Type	Materials	Benefits
Biomax	Modified polyethersulfone	Highest flux ultrafiltration membrane Excellent chemical resistance Void-free structure for higher yield and reliability
Ultracel PLC	Regenerated cellulose (ideal for protein solutions < 20 g/L) PLC membranes are composite membranes cast on a microporous substrate for defect-free membranes with superior adhesion. Brings higher resolution, improved yields and superior back-pressure resistance	Extremely low protein binding hydrophilic membrane Highest product recovery and improved performance with difficult to process streams (antifoams, lipids, protein transmission applications)
Durapore	Hydrophilic PVDF	Very hydrophilic microporous membrane for cell harvest or clarification applications

Pellicon 2 Membrane Selection Chart

Approximate Molecular Weight (range of solutes retained >99%, kD)		Membrane	NMWL (kD) or Microns	Membrane Material	pH Range
High Flux Biomax Membranes – Void-free for Higher Yield and Reliability					
12 – 25	(growth factors, hormones)	Biomax-5	5	modified polyethersulfone	1 – 14
25 – 50	(growth factors, hormones)	Biomax-8	8	modified polyethersulfone	1 – 14
50 – 100	(albumin, hemoglobin)	Biomax-10	10	modified polyethersulfone	1 – 14
100 – 140	(enzymes)	Biomax-30	30	modified polyethersulfone	1 – 14
140 – 300	(IgG's)	Biomax-50	50	modified polyethersulfone	1 – 14
300 – 500	(small viruses and antigens)	Biomax-100	100	modified polyethersulfone	1 – 14
>500	(IgM's, large viruses)	Biomax-300	300	modified polyethersulfone	1 – 14
>0.03 µm	(large viruses, colloids, particulates)	Biomax-500	500	modified polyethersulfone	1 – 14
>0.03 µm	(large viruses, cells, colloids, particulates)	Biomax-1000	1000	modified polyethersulfone	1 – 14
Ultracel PLC Series – for High Recoveries					
8 – 18	(proinsulin, hematopoetic factors)	PLCCC	5	regenerated cellulose	2 – 13
18 – 60	(hemoglobin, enzymes)	PLCGC	10	regenerated cellulose	2 – 13
60 – 200	(monoclonal IgG's)	PLCTK	30	regenerated cellulose	2 – 13
200 – 500	(small viruses, viral antigens)	PLCHK	100	regenerated cellulose	2 – 13
>500	(large viruses, IgM's)	PLCMK	300	regenerated cellulose	2 – 13
>0.03 µm	(large viruses, cells, colloids, particulates)	PLCXK	1000	regenerated cellulose	2 – 13
Durapore Membranes – for Microporous Applications					
Clarify cell lysates and protein solutions, clarify viral cultures		VVPP	0.1 µm	hydrophilic PVDF	2 – 11
Harvest & wash colloidal suspensions, bacterial cells; clarify protein solutions and viral cultures		GVPP	0.22 µm	hydrophilic PVDF	2 – 11
Harvest & wash colloidal suspensions, cell & viral cultures, clarify protein solutions & viral cultures		HVMP	0.45 µm	hydrophilic PVDF	2 – 11
Harvest cell cultures or colloidal suspensions		DVPP	0.65 µm	hydrophilic PVDF	2 – 11

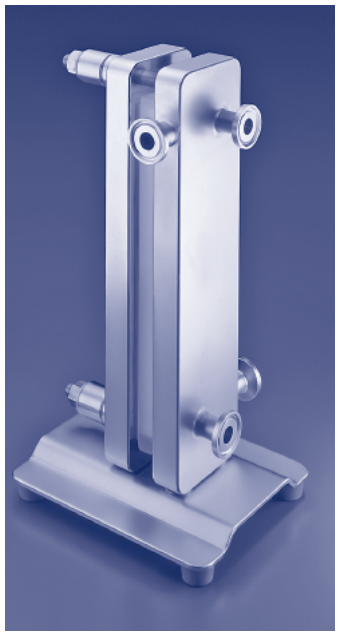
Ordering Information

Pellicon 2 Filters

	Filters with A Screens (Tight Screen)			Filters with Type C Screens (Coarse Screen)		
Membrane	**0.1 m²/1.1 ft²**	**0.5 m²/5.4 ft²**	**2.5 m²/26.9 ft²**	**0.1 m²/1.1 ft²**	**0.5 m²/5.4 ft²**	**2.5 m²/26.9 ft²**
Biomax Series – Modified Polyethersulfone						
Biomax 5	P2B0 05A 01	P2B0 05A 05	P2B0 05A 25	+	+	+
Biomax 8	P2B0 08A 01	P2B0 08A 05	P2B0 08A 25	+	+	+
Biomax 10	P2B0 10A 01	P2B0 10A 05	P2B0 10A 25	+	+	+
Biomax 30	P2B0 30A 01	P2B0 30A 05	P2B0 30A 25	+	+	+
Biomax 50	P2B0 50A 01	P2B0 50A 05	P2B0 50A 25	P2B0 50C 01	P2B0 50C 05	P2B0 50C 25
Biomax 100	P2B1 00A 01	P2B1 00A 05	P2B1 00A 25	P2B1 00C 01	P2B1 00C 05	P2B1 00C 25
Biomax 300	+	+	+	P2B3 00C 01	P2B3 00C 05	P2B3 00C 25
Biomax 500	+	+	+	P2B5 00C 01	P2B5 00C 05	P2B5 00C 25
Biomax 1000	+	+	+	P2B0 1MC 01	P2B0 1MC 05	P2B0 1MC 25
Ultracel PLC Series – Regenerated Cellulose, Composite Construction						
5 kD	NA	NA	NA	P2C0 05C 01	P2C0 05C 05	P2C0 05C 25
10 kD	NA	NA	NA	P2C0 10C 01	P2C0 10C 05	P2C0 10C 25
30 kD	NA	NA	NA	P2C0 30C 01	P2C0 30C 05	P2C0 30C 25
100 kD	NA	NA	NA	P2C1 00C 01	P2C1 00C 05	P2C1 00C 25
300 kD	NA	NA	NA	P2C3 00C 01	P2C3 00C 05	P2C3 00C 25
1000 kD	NA	NA	NA	P2C0 1MC 01	P2C0 1MC 05	P2C0 1MC 25
Durapore – Hydrophilic PVDF						
0.1 µm	+	+	+	P2VV PPC 01	P2VV PPC 05	P2VV PPC 25
0.22 µm	+	+	+	P2GV PPC 01	P2GV PPC 05	P2GV PPC 25
0.45 µm	+	+	+	P2HV MPC 01	P2HV MPC 05	P2HV MPC 25
0.65 µm	+	+	+	P2DV PPC 01	P2DV PPC 05	P2DV PPC 25

Each Pellicon filter is packed one per box and includes Operating Instructions. A Certificate of Quality is included in every box.
Silicone intercassette gaskets are required for use with Pellicon 2 filters. Two gaskets are packed in the box with every Pellicon 2 filter.
+ = On request (custom order)
NA = not available

Filters with V Screens (Loose Screen)		
0.1 m²/1.1 ft²	**0.5 m²/5.4 ft²**	**2.0 m²/21.5 ft²**
P2B0 05V 01	P2B0 05V 05	P2B0 05V 20
P2B0 08V 01	P2B0 08V 05	P2B0 08V 20
P2B0 10V 01	P2B0 10V 05	P2B0 10V 20
P2B0 30V 01	P2B0 30V 05	P2B0 30V 20
P2B0 50V 01	P2B0 50V 05	P2B0 50V 20
P2B 100V 01	P2B1 00V 05	P2B1 00V 20
P2B3 00V 01	P2B3 00V 05	P2B3 00V 20
P2B5 00V 01	P2B5 00V 05	P2B5 00V 20
P2B0 1MV 01	P2B0 1MV 05	P2B0 1MV 20
P2C0 05V 01	P2C005V 05	P2C0 05V 20
P2C0 10V 01	P2C0 10V 05	P2C0 10V 20
P2C0 30V 01	P2C0 30V 05	P2C0 30V 20
P2C1 00V 01	P2C1 00V 05	P2C1 00V 20
P2C3 00V 01	P2C3 00V 05	P2C3 00V 20
P2C0 1MV 01	P2C0 1MV 05	P2C01MV 20
P2VV PPV 01	P2VV PPV 05	P2VV PPV 20
P2GV PPV 01	P2GV PPV 05	P2GV PPV 20
P2HV MPV 01	P2HV MPV 05	P2HV MPV 20
P2DV PPV 01	P2DV PPV 05	P2DV PPV 20



Pellicon 2 Mini Holder

Pellicon 2 Mini holder operates one to three Mini filters in parallel for total areas of 0.1 to 0.3 m² (1.1 – 3.3 ft²). This sanitary holder is tightened with a small torque wrench to compress the filters between a manifold plate that conveys fluids in and out of the filters and an end plate that seals the filters together. The Mini holder is designed for process development and small volume pharmaceutical manufacturing.

Materials of Construction

Manifold and End Plates:
316 L stainless steel

Base, Tie Rods, Spacers and Washers:
304 stainless steel

Feet:
Thermoplastic rubber

Gaskets:
Silicone

Nuts:
Silicone bronze

Separator Plates

An optional separator plate allows processing simultaneously with up to three 0.1 m²/1.1 ft² cassettes to determine the best molecular weight cut-off in a single study on the same feed material.

Connections

All manifold connections are standard ½-inch sanitary clamp type.

Operating Parameters

Temperature Range:
4 to 50 °C. The Mini holder can be autoclaved without filters installed. The filters themselves cannot be autoclaved.

Maximum Pressure:
6.8 bar

Dimensions

Height: 260 mm; *Width:* 114 mm

Length: 140 mm; *Weight:* 5 kg

Holder Manifold Volume:

Feed plus retentate: 5.3 mL

Permeate: 6.4 mL

Stainless Steel Pellicon Holder

XX42P0080

The stainless steel Pellicon filter holder, designed for sanitary applications, can be used alone or to expand existing cassette ultrafiltration (CUF) systems or to replace existing holders.

It requires only to be connected to an existing sanitary pump and piping for tangential flow microporous filtration or ultrafiltration.

It can accomodate up to 5 m^2/55 ft^2 filter area as shipped with long tie rods or 0.5 to 2.5 m^2 (5.4 – 26.9 ft^2) with accessory short tie rods.

Materials of Construction

Wetted Surfaces:
316 L stainless steel

Non-wetted Surfaces:
Silicon bronze nuts

Dimensions

Length: 28 cm; *Width:* 19 cm

Height: 25 cm

Operating Parameters

Operating Temperature Range:
4 to 50 °C. The Pellicon holder can be autoclaved without pressure gauges and filters; holder with gauges cannot be steamed. Pellicon filters cannot be steamed or autoclaved.

Connections

Sanitary ¾" TC connections;
1½" TC connections for gauges.

Shipping Weight

24 kg

Process-scale Pellicon Holder

The Pellicon Process-scale Holder is a unique innovation for production scale Pellicon systems. This holder, vertically mounted, can hold up to 80 m^2/880 ft^2 of membrane area.

Benefits

- Extremely compact footprint
- Easy to change cassettes
- Easy to vent and fully drain
- Simple connections
- Up to 4 levels. Can be easily extended in levels for simple membrane area expansion
- Each level up to 20 m^2/220 ft^2
- Uses standard and Maxi Cassettes
- Can be adapted for series or parallel configurations
- Simplifies pipework connection
- Hydraulic closure systems are available for the stainless-steel Pellicon holder and the process-scale Pellicon holder. These systems are convenient, reliable and easy to use to enable rapid and repeatable loading operation and storage of Pellicon 2 cassettes.

Materials of Construction

Manifold segment, fitting blocks and end plate 316 L stainless steel; tie rods 304 and 304 L stainless steel.

Ordering Information

Pellicon 2 Filter Holders

Description	Catalogue No.
Pellicon 2 Mini filter holder	XX42 PMI NI
Pressure gauges One diaphragm-protected digital pressure gauge, 0 – 7 bar, ¾-inch fittings	XX42 PSG 01L
Pressure gauge adapters	XX42 PM0 01
Fitting kit Contains all tees, clamps, gaskets and a valve to connect tubing and pressure gauges to the Pellicon 2 Mini holder	XX42 PFK 01
Pellicon filter holder (for cassettes and Maxi filters)	XX42 P00 80
Pellicon 2 double thick gasket	PSSP 2XC 10
Pellicon Process-scale holder support and plate	XX42 SSP LT
Pellicon Process-scale holder	On request

To Place an Order or Receive Technical Assistance

For additional information call your nearest Millipore office:
In the U.S. and Canada,
call toll-free **1-800-MILLIPORE (1-800-645-5476)**
In the U.S., Canada and Puerto Rico, fax orders to **1-800-MILLIFX (1-800-645-5439)**
Outside of North America contact your local office. To find the office nearest you visit **www.millipore.com/offices.**
Internet: **www.millipore.com**
Technical Service:
www.millipore.com/techservice

MILLIPORE

A Typical Pellicon Production Processing System

Millipore supplies a range of standard and custom engineered systems. These systems can contain from 1 m^2/11 ft^2 to several hunded m^2 of membrane area, with Clean-in-Place (CIP) or Steam-in-Place (SIP) integrated as appropriate. Systems can also be supplied with integrated process vessels in manual or fully automatic versions.

All systems are designed, engineered and manufactured in ISO® 9001 registered facilities, and are supplied with extensive validation data support packages.

Please contact us to discuss your specific application and process requirements.

Pellicon XL Devices for Process Development

For process development of volumes from 50 mL to 1 liter, Millipore offers Pellicon XL devices. This small volume TFF filter is designed for true scalability by providing the same flow path, channel length, and channel height as the Pellicon 2 cassettes. Based on proven TFF membrane technology, Pellicon XL devices ensure reliable, consistent and predictable performance.

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Printed in U.S.A. 04-324

Data Sheet

Pellicon® 3 Cassettes with Biomax® Membrane

The device of choice for applications requiring high flux, low to moderate protein binding, and harsh chemical cleaning and/or sanitization.

Pellicon® 3 cassettes with Biomax® membrane are the optimum tangential flow filtration (TFF) devices for the filtration of solutions containing therapeutical proteins, albumin, hormones, vaccines and growth factors. These advanced, high-performance cassettes are ideal for today's higher titer therapeutic antibodies as well as the more demanding filtration processes that require higher operating pressures, temperatures and caustic cleaning regimes.

From small-scale to full-scale production, Pellicon® 3 cassettes are designed for use in research, process scale-up/scale-down, applications development and full-scale manufacturing. The Pellicon® 3 design and automated manufacturing process provides unbeatable performance consistency between cassette sizes. Pellicon® 3 devices also offer greater cassette size selection for improved scale-up and scale-down process development. The streamlined design allows operators to quickly and easily handle, install and remove Pellicon® 3 cassettes. The materials of construction are compatible with a broad range of chemical cleaning agents that many TFF systems require to ensure proper sanitization.

Benefits

- Optimum product recovery using proven macrovoid-free membrane technology
- Fast, reliable scale up/down from lab to production scale
- Rugged, reliable design ideally suited to filtration processes with higher operating pressures, temperatures and caustic cleaning regimes
- Automated manufacturing delivers unbeatable performance consistency and reliability
- Easy to install and clean
- Extreme temperature and chemical compatibility

EMD Millipore is a division of Merck KGaA, Darmstadt, Germany

Applications

- **Monoclonal antibodies**
- **Albumin**
- **Hormones**
- **Vaccines**
- **Growth Factors**
- **Recombinant protein**
- **Nanoparticules**

Optimum Product Recovery and High Yields

High flux and retention properties of the Biomax® membrane result in faster processing speeds with higher yields, which means shortened processing times and a bioprocessing system that can be smaller and more compact.

Biomax® membranes are composed of polyethersulfone and are resistant to harsh chemicals used in cleaning, biological decontamination and sanitization. The polyethersulfone Biomax® membrane has been modified to reduce non-specific protein binding compared to conventional polyethersulfone membranes. The technology offers a mechanically robust design able to withstand process upsets and extreme operating conditions.

Fast, Reliable Linear Scale-Up from the Lab to the Production Plant

Offered in four sizes, 88 cm^2, 0.11 m^2, 0.57 m^2 and 1.14 m^2, all Pellicon® 3 cassettes are constructed of identical materials and have the same flow channel length, height, turbulence promoter and flow direction. This ensures that every Pellicon® 3 cassette maintains the same performance profile at every scale, from 250 milliliters to thousands of liters.

Streamlined Installation and Rugged Design

Pellicon® 3 cassettes incorporate a hard polypropylene jacket and end cap design that protects the membrane surface from impacts and potential damage. The end cap includes integral seals which simplify the installation by eliminating the need for external gaskets between each device.

Reliable Product Performance Delivering Exceptional Consistency and Reproducibility

Our controlled, automated manufacturing process provides the highest level of cassette performance consistency. The high level of process control ensures consistent, repeat performance in terms of scale up to scale down, from run to run and campaign to campaign. All cassettes are manufactured in accordance with GMP.

Extreme Temperature and Chemical Capability

Pellicon® 3 cassettes are manufactured using the most modern polymers and plastics enabling continuous operation at 50 °C and 1.0N NaOH up to 200 hours. These materials of construction ensure low extractables in a wide range of solvents, acids and bases.

Quality Assurance

All Pellicon® 3 cassettes are manufactured using the same equipment, process and quality assurance. Each Pellicon® 3 cassette manufacturing lot is 100% integrity tested during manufacturing to ensure that every filter is integral, robust and within specification. Additionally, Pellicon® 3 cassettes are subjected to a complete array of quality control release tests.

Each cassette is identified with a unique serial number and shipped with an individual Certificate of Quality.

* Contact your local representative for additional information.

Single-Pass TFF

Pellicon® 3 cassettes run in single-pass TFF mode is a simple and efficient way to increase production capacity by reducing process volumes and tank requirements. Single-Pass TFF systems can concentrate process streams without the recirculation required in traditional TFF steps and require a smaller pump and less piping resulting in a more compact footprint and lower cost. For concentrated final formulations, Single-Pass TFF can increase recovery due to lower hold-up volume. Single-Pass TFF also enables continuous processing where in-line concentration is coupled to other process steps.

Single-Pass TFF has several applications such as:

- Product concentration/volume reduction
- In-line delution/de-salting
- Final formulation/concentration

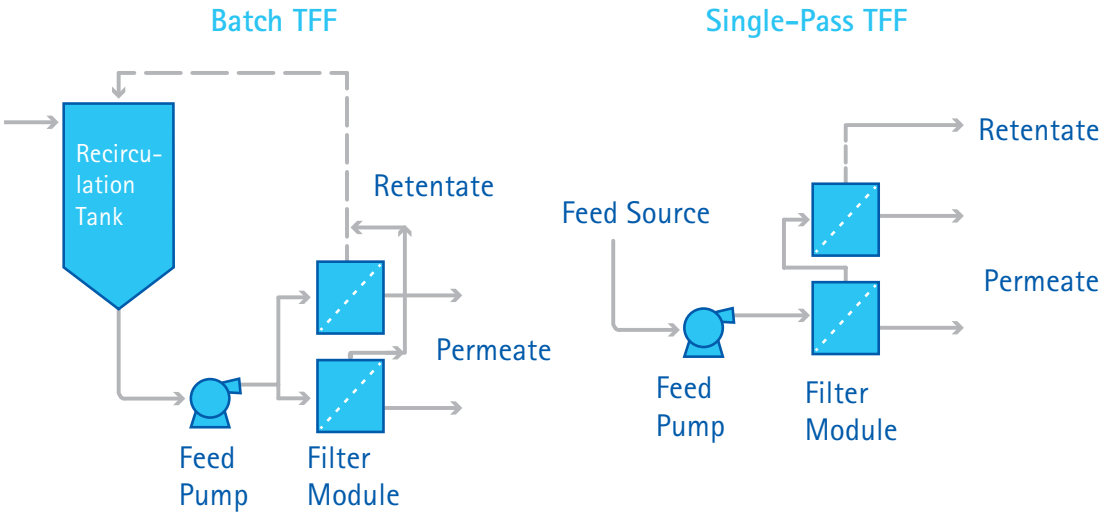

Figure 1.
Batch TFF vs. Single-Pass TFF

Specifications

Materials and Assembly

Materials of Construction:	Polypropylene Polyethylene Polyethersulfone Thermoplastic elastomer Stainless steel (0.57 m^2 and 1.14 m^2 cassettes only)
Preservative:	1.6% Phosphoric Acid, 1.1% Acetic Acid, 20% glycerin and water
Membrane:	Biomax® PES—Polyethersulfone
Assembly Design:	Automated assembly and testing of heat sealed packets bound together by an injection-molded polypropylene jacket

Hold Up Volume

Area	Feed Channel (mL)	Permeate Channel (mL)
88 cm^2	1.8	2.8
0.11 m^2	9	7
0.57 m^2	69	39
1.14 m^2	134	88

Pellicon® 3 Cassette (88 cm^2)

Maximum Operating Conditions

Recommended Feed Flow Rate:	4–8 L/m^2/min
Maximum Inlet Pressure:	<100 psi
Forward Transmembrane Pressure:	80 psi (5.5 bar) at 4-40 °C, 200 hours continuous (4 hours continuous, micro format only) 40 psi (2.7 bar) at 4-50 °C, 50 hours continuous
Reverse Transmembrane Pressure:	30 psi (2.1 bar) at 25 °C, 3 min intervals, 10 cycles (5 cycles, micro format only)
Maximum Caustic Exposure:	1.0 N NaOH at 50 °C up to 200 hours (Contact EMD Millipore for exposure parameters.)
Operating pH Range:	2 - 14

Pellicon® 3 Cassette (0.11 m^2)

Pellicon® 3 Cassette (0.57 m^2)

Regulatory Information

Component Material Toxicity:	Component materials were tested and meet the criteria of the USP <88> Biological Reactivity Tests for Class VI Plastics.
Good Manufacturing Practices:	These products are manufactured in an EMD Millipore facility which adheres to FDA Good Manufacturing Practices.
ISO® 9001 Quality Standard:	This product was manufactured in an EMD Millipore facility whose Quality Management System is approved by an accredited registering body to the appropriate ISO® 9001 Quality Systems Standard.
100% Integrity Tested in Manufacturing:	Each unit must pass the EMD Millipore integrity test based on air flow through the fully-wetted membranes of the filter.
Validated Production Process:	This product was fabricated using a validated manufacturing process. Principles of statistical process control and determinations of process capability have been applied to critical variables in the device fabrication process. In-process controls are used to assure stability of the process.

Pellicon® 3 Cassette (1.14 m^2)

Ordering Information

Pellicon® 3 Cassettes with Biomax® Membrane			
Description	Catalogue No.		
	10kD NMWL	30kD NMWL	50kD NMWL
88 cm^2	P3B 010 A00	P3B 030 A00	P3B 050 A00
0.11 m^2	P3B 010 A01	P3B 030 A01	P3B 050 A01
0.57 m^2	P3B 010 A05	P3B 030 A05	P3B 050 A05
1.14 m^2	P3B 010 A10	P3B 030 A10	P3B 050 A10

Accessories

Pellicon® 3 Cassette Holders			
Holder Type	Cassette Size	Area Range	Catalogue No.
Stainless Steel Mini-Holder	88 cm^2 and 0.11 m^2	88 cm^2 to 0.55 m^2	XX42PMINI
Acrylic Cassette Holder Low Retentate Volume	0.57 m^2 and 1.14 m^2	0.57 m^2 to 5.7 m^2	XX42PRV60
Stainless Steel Holder	0.57 m^2 and 1.14 m^2	0.57 m^2 to 5.7 m^2	XX42P0080
Stainless Steel Cassette Holder and Assembly	0.57 m^2 and 1.14 m^2	0.57 m^2 to 5.7 m^2	XX42P0K80
Manifold Support Plate	0.57 m^2	1.14 m^2	XXPEL3MAP
Process Scale Holder	0.57 m^2 and 1.14 m^2	1.14 m^2 and up	Contact Local Representative
Hydraulic Process Scale Holder	0.57 m^2 and 1.14 m^2	1.14 m^2 and up	Contact Local Representative

Cleaning	
Description	Catalogue No.
Sodium hydroxide solution 0.5 mol/L suitable for biopharmaceutical production EMPROVE® bio	137060
Sodium hydroxide solution 1 mol/L suitable for biopharmaceutical production EMPROVE® bio	137031
Sodium hydroxide solution 25% low iron suitable for biopharmaceutical production EMPROVE® bio	480659

Single-Pass TFF Accessories	
Description	Catalogue No.
Diverter plate and silicon gasket kit for 88 cm^2 cassette	XXSPTFF01
Diverter plate for 0.57 and 1.14 m^2 cassettes	XXSPTFF02
Retentate collection plate for 0.57 and 1.14 m^2 cassettes	XXSPTFF03

To Place an Order or Receive Technical Assistance

In the U.S. and Canada,
call toll-free 1-800-645-5476

For other countries across Europe and the world, please visit: **www.emdmillipore.com/offices**

For Technical Service, please visit:
www.emdmillipore.com/techservice

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MASTERFLEX® L/S® 07522-20

OPERATING MANUAL:

L/S® DIGITAL PUMP DRIVES

Model Nos.

07522-20
07522-30
07575-30
07575-40

A-1299-1110
Edition 01

Cole-Parmer®
1-800-MASTERFLEX (627-8373) (U.S. and Canada only) • 11 (847) 549-7600 (Outside U.S.)
(847) 549-7600 (Local) • www.masterflex.com • techinfo@coleparmer.com

A-1299-1110

REV	ECR/ECN	DATE	DESCRIPTION	By
1	18946	130715	First Edition	BK

SAFETY PRECAUTIONS

DANGER: High voltages exist and are accessible. Use extreme caution when servicing internal components.

WARNINGS: Tubing breakage may result in fluid being sprayed from pump. Use appropriate measures to protect operator and equipment.

Turn drive off before removing or installing tubing. Fingers or loose clothing could get caught in drive mechanism.

WARNINGS: Do not operate the pump drive in a manner not specified in the documentation. Misuse of the pump drive may result in a hazard and may compromise the safety protection built into the pump drive. If the pump drive is damaged, turn it off and not use it until service-trained personnel can check its safety.

Single-Phase Only. Not to be used with Split-Phase lines.

The Power switch on the Back Panel is not the main disconnect. Main disconnect is accomplished by disconnecting the detachable power supply cord at the appliance coupler or at the main plug. Ensure the power cord is easily accessible and removable, in the event of an emergency, which requires immediate disconnection.

The operator should check the detachable power supply cord condition. The equipment should not be operated if the power supply cord is cracked or broken. Any obvious damage to the enclosure (from a drop or fall) should be checked by service personnel for loose or damaged parts inside.

CAUTIONS: Power must be turned off before connecting the external remote control cable to prevent damage to the drive.

Do not contaminate the lubricant in the container, on the shaft or on the seal with foreign material.
Failure to observe this precaution may result in damage to the seal and premature failure of the seal.

No foreign matter should be allowed under the gasket on the back of the front plate or under the heads of the screws.
Failure to observe this precaution may result in leakage during washdown of the drive

Do not block the rear panel of the pump drive. The power switch must always be easy to access. The power cord must always be easy to disconnect.

Replace the power cord only with one of the same type and rating. The minimum power ratings are stated on the rear panel.

The power cord set supplied with your pump drive meets the requirements of the country where you purchased the pump drive. If you use the pump drive in another country, you must use a power cord set that meets the requirements of that country.

When using hazardous chemical and biological agents, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of the system.

SAFETY PRECAUTIONS (continued)

CAUTION: To avoid electrical shock, the power cord protective grounding conductor must be connected to ground. Not for operation in wet locations as defined by EN61010-1.

CAUTIONS: Keep fingers away from rotor while pump is in operation. Stop pump before loading or unloading tubing.

To reduce the possibility of tipping, use the stacking clip provided with the unit.

Explanation of Symbols

CAUTION: Risk of Danger. Consult Operator's manual for nature of hazard and corrective actions.

CAUTION: Risk of crushing. Keep fingers away from rotor while pump is in operation. Stop pump before loading or unloading tubing.

CAUTION: Hot Surface. Do not touch.

CAUTION: Risk of electric shock. Consult Operator's manual for nature of hazard and corrective actions.

WARNING: Product Use Limitation

This product is not designed for, nor intended for use in patient connected applications; including, but not limited to, medical and dental use, and accordingly has not been submitted for FDA approval.

This product is not designed for, nor intended for use in hazardous duty areas as defined by ATEX or the NEC (National Electrical Code); including, but not limited to use with flammable liquids. Consult the factory for products suitable for these types of applications

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Section 1 Introduction

The Digital drive controls the speed of MASTERFLEX® Pump Heads to provide flow rates from 0.001 to 3400 mL/min.

Mount up to 2 (600 rpm) or 4 (100 rpm) MASTERFLEX Pump Heads and all MASTERFLEX-compatible Pump Heads.

Application Solutions

Advantages of Peristaltic Pumps:

- Handle abrasive slurries and corrosive fluids with minimal wear. Ideal for titanium dioxide or diatomaceous earth filter aid applications.
- No seals in contact with the medium pumped.
- No valves to clog.
- Inner surfaces are smooth and easy to clean.
- Fluid contacts only the tubing or tube material.
- Suction lift and priming up to 8m water column at sea level.
- Low shearing for handling the most shear sensitive of fluids like latex or fire fighting foam.
- Capable of running dry and pumping fluids with high quantities of entrained air, such as black liquor soap.
- High volumeteric efficiency allows operation in metering or dosing applications where high accuracy is required.
- Handle extremely viscous fluids.
- Tubing and tube materials are available that are suitable for food and pharmaceutical use.

General Description

The MASTERFLEX L/S Digital Peristaltic Pump Drive offers flow rate capacities from 0.001 mL/min to 3400 mL/min using MASTERFLEX Standard, EASY-LOAD® or High-Performance Pump Heads. Even lower flow rates can be achieved with our multichannel and cartridge Pump Heads. Features include a small footprint, plus non-stainless steel drives that are stackable.

The MASTERFLEX digital pump provides a motor speed repeatability of 0.1 percent to maximize productivity in precision liquid dosing, batch dispensing and filling applications. A turndown ratio up to 6000-to-1, bidirectional flow and self-priming capabilities allow for smooth, seamless operation and an extremely broad flow range within one tubing size.

In addition to high accuracy, precision, repeatability and resolution of speed (or flow rate), the MASTERFLEX drive features a multi-language, intuitive, man/machine interface with a four-line graphical LCD display providing direct readout of pump speed (rpm), flow rate (user-selected units), number of dispenses, and menu options.

The easy-to-use keypad eliminates setpoint overshoot and provides easy navigation through menu options that include a number of on-screen programming features.

These drives use high precision, no-maintenance brushless motors for improved reliability. This, combined with its high turndown, superior accuracy, and intuitive interface make the MASTERFLEX drives ideally suited where ultra-precise, repeatable flow control is required. The pump accommodates a variety of product fill volumes and batch dispensing profiles, and fluid only contacts the tubing, providing for contamination-free pumping.

MASTERFLEX pumps are self-priming, can operate dry without damage, are suitable for most chemicals and contain no valves or seals. See *Pump Head* and *Tubing Guides* within this CD.

Section 2 Installation and Setup

Before Starting Drive

- The drive should be mounted on a flat horizontal surface, and no more than two (2) Pump Heads should be added for 600 rpm drives or four (4) Pump Heads for 100 rpm drives.
- The ambient air temperature should not exceed 104° F (40° C) and adequate air flow should be provided for.

⚠ ***CAUTION: Do not block the rear panel of the pump drive. The power switch must always be easy to access. The power cord must always be easy to disconnect.***

- Tubing should be clean and routed so that bend radii are at a minimum four (4) times the tube diameter and as short as possible.

⚠ ***WARNING: Turn drive off before removing or installing tubing. Fingers or loose clothing could get caught in drive mechanism.***

- Use a tube size of appropriate diameter for the flow rate and viscosity required.
- To maintain the best accuracy of flow rates, re-calibrate tubing regularly. See *Tubing Calibration Section* of this manual.
- For tubing selection and compatibility, see *Tubing Selection Guide* within this CD.
- For Pump Head information, see *Pump Head* information within this CD.
- When cleaning or performing maintenance, please remove power from the drive.

⚠ ***CAUTION: The power cord set supplied with your pump drive meets the requirements of the country where you purchased the pump drive. If you use the pump drive in another country, you must use a power cord set that meets the requirements of that country.***

⚠ ***DANGER: High voltages exist and are accessible. Use extreme caution when servicing internal components.***

Mounting the Pump Head

- Mount Pump Head and load tubing (See *Pump Head* information within this CD). Check to make sure that rollers are clean and free of defects.

⚠ ***CAUTION: When using hazardous chemical and biological agents, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of the system.***

Section 3 Operation

Turning On the Drive

WARNING: Do not operate the pump drive in a manner not specified in the documentation. Misuse of the pump drive may result in a hazard and may compromise the safety protection built into the pump drive. If the pump drive is damaged, turn it off and not use it until service-trained personnel can check its safety.

1. Plug the power cord into the IEC Connector, located on the rear of the drive. Plug the opposite end of the power cord into an electrical outlet.

2. Flip the power switch located on the rear of the drive.

3. Upon turning on the drive for the first time you will be prompted to select a language. The selected language will be set as the default but can be changed at any time by selecting "LANGUAGE" on the main menu.

4. After selecting your language, the Main Menu will now appear on the LCD screen. (**NOTE:** Each start-up after the initial will revert to the mode of operation screen previously in use.)

5. If the language is accidently changed and the user would like to reset it to the default language (English), press and hold the UP/DOWN (▲/▼) keys during power up.

6. To restore drive to default settings, press and hold the LEFT/RIGHT (◀/▶) keys during power up.

CAUTION: To avoid electrical shock, the power cord protective grounding conductor must be connected to ground. Not for operation in wet locations as defined by EN61010-1.

CAUTION: Power must be turned off before connecting the external remote control cable to prevent damage to the drive.

WARNING: Tubing breakage may result in fluid being sprayed from pump. Use appropriate measures to protect operator and equipment.

Control Panel

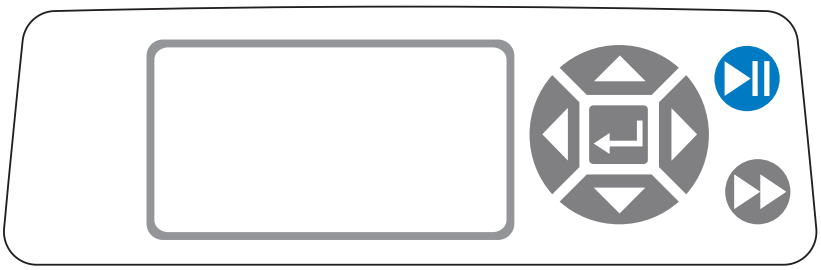


Figure 3-1. Control Panel

- To navigate all menus on the drive use the directional pad directly to the right of the LCD screen.
- The (ENTER) key located in the middle of the directional pad is used to enter or select a highlighted field or option. This key is often referred to as the ENTER key in this manual.
- The (START/STOP) **key** located at the top right of the control panel is used to start and pause the drive. This key is functional only when in one of the four operating modes: Continuous, Time Dispense, Copy Dispense, or Volume Dispense. This key is often referred to as the START/STOP **key** in this manual.
- The (PRIME) key located at the bottom right of the control panel is used to access the prime (fast forward) function. While pressed, this key operates the drive at the maximum allowed speed/flow rate and in the direction shown on the display. When released, the drive returns to its original speed or flow rate.

Priming the Pump

1. Mount Pump Head to drive.
2. Insert appropriate tubing into Pump Head.
3. Insert tube inlet into supply fluid.
4. Insert supply outlet into desired container.
5. Turn on pump using switch located on the back of the drive.
6. Press and hold the PRIME key on the drive console to prime the pump. Priming will stop when key is released.

CAUTION: Keep fingers away from rotor while pump is in operation. Stop pump before loading or unloading tubing.

Main Menu

CONTINUOUS MODE refer to *Continuous Mode* in this manual.

TIME DISPENSE MODE refer to *Time Dispense Mode* in this manual.

COPY DISPENSE MODE refer to *Copy Dispense Mode* section in this manual.

VOLUME DISPENSE MODE refer to *Volume Dispense Mode* section in this manual.

REMOTE CONTROL MODE refer to *Remote Control Mode* section in this manual.

CUMULATIVE VOLUME: The drive stores and displays the cumulative volume in units based on flow rate units (see SETUP MENU in this section). The Cumulative Volume can also be reset to zero.

NOTE: The Cumulative Volume is dependent on the Tubing Size selected. (See *SETUP MENU* in this section.)

SOUNDS: An audible "beep" can be enabled to indicate a keypad press, the end of a dispense and/or the end of a batch.

AUTOSTART: By default the drive will not restart when power is applied. To enable this feature select AUTOSTART and then ON. The drive will now restart when power is reapplied.

DISPLAY CONTRAST: This display can be adjusted using the UP/DOWN (▲/▼) arrows after selecting this menu item.

LANGUAGE: After selecting this menu, the user will be able to select one of seven different languages.

NOTE: If the language is accidentally changed and the user would like to reset it to the default language (English), press and hold the UP/DOWN (▲/▼) keys when power is reapplied.

DEFAULT SETTINGS: Selecting this menu item and pressing the ENTER key will restore default settings. To restore drive to default settings the user may also press and hold the LEFT/RIGHT (◀/▶) keys when power is reapplied.

Tubing Calibration

1. Mount Pump Head to drive.

2. Insert appropriate tubing into Pump Head.

3. Insert tube inlet into supply fluid.

4. Insert tube outlet into desired container. Container should be a graduated container or a container placed on a scale may be used for increased accuracy.

   If using a scale, an acceptable weight to volume conversion for water is 1 gram = 1 mL.

5. Turn on drive using power switch located on the rear of the drive.

6. Go to the Main Menu or Mode Setup Menu by selecting the SETUP icon and pressing the ENTER key. Use the UP and DOWN keys to highlight TUBING CAL in the Main or Setup Menu and press the ENTER key.

7. Set the drive for the desired flow direction, tube size, and flow rate. Note that these settings are retained and transferred to other mode screens when entering or leaving the TUBING CAL screen.

   - The flow direction is set using the directional keypad to highlight the directional arrow. Pressing ENTER will toggle arrow between CW and CCW.
   - The tube size is set using the directional keypad to highlight the tube size field. Press ENTER and use the UP/DOWN keys to select the tube size. Press ENTER to SAVE the selection and return to TUBING CAL screen.
   - The estimated flow rate is set using the directional keypad to highlight the flow rate field. Press ENTER and use the LEFT/RIGHT keys to select the digit to be changed. Use the UP/DOWN keys to adjust the flow rate value. Press ENTER to SAVE the setting and EXIT field using arrow keys. The drive will adjust this flow rate after calibration is complete.
   - Note that the calibration volume is fixed and cannot be changed.

8. Press and hold the prime key on the drive console to prime the pump. Priming will stop when key is released.

9. Place a measuring container at the pump outlet. Highlight the START field and press the ENTER key. The drive will run based on the default volume at the estimated flow rate selected.

Tubing Calibration (continued)

10. Upon completion of the calibration run period, the CAL VOLUME field will be highlighted. Press the ENTER key and adjust the CAL VOLUME to the measured quantity. Use the LEFT/RIGHT keys to select digit to be changed, use the UP/DOWN keys to adjust the value, and press ENTER to SAVE setting and EXIT the field.

    A lower case "c" should now be displayed when the calibrated tubing size is selected. The volume units will depend on the flow rate units. The flow rate unit mL/min will result in a volume unit of mL; oz/min will result in a volume unit of oz.

Tubing Calibration Notes

- If the drive is stopped during calibration, empty the container and re-start the procedure.
- Calibration time at maximum allowable flow rate (default max flow rate) is 5-10 seconds and at minimum allowable flow rate (approximately 4% of the maximum flow rate) is 4 minutes. Select the CUSTOM tube size for other tubing sizes or lower flow rates.
- Minimum and maximum flow rates will change after a tubing calibration due to a re-calculation of the vol/rev.
- Optimum results are best obtained after tubing has been broken in by running in pump for at least 10 minutes. Steps 8-10 can be repeated as necessary to optimize the accuracy of the tubing cal.

CAL RUN TIME FORMULA

60 / (flow rate [mL/min] / cal volume [mL]) = cal run time (seconds)

INVALID CAL RUN TIME EXAMPLE

- tube size 13 flow rate range is 0.006 mL/min – 36.0 mL/min
- at flow rate of 1 mL/min, cal run time calculation is as follows:
  60 / (1 mL/min / 6 mL) = 360 seconds
  360 seconds exceed the max run time of 4 minutes (240 seconds)

Setup Menu

All four operation mode screens contain a SETUP icon in the upper right hand that gives quick access to the SETUP menu. The exact options that can be accessed through the SETUP menu will depend on the operating mode currently in use:

1. **Selecting the SETUP Menu:** In any of the four operating modes, use the directional pad and enter key to select the SETUP icon from the mode operation screen.

2. **Navigating the SETUP Menu:** Use the directional pad and the ENTER key to select desired setting.

A breakdown of the setting features common to all modes follows. Other settings are related to the specific operating mode currently in use and can be accessed through the mode operation screen as well.

Flow Unit: Select desired flow unit to be displayed.

Tubing Size: Size and Maximum Flow Rate are displayed. Select desired tubing size.

Flow Rate: Set the flow rate in flow unit listed at the top of the screen. (**NOTE:** To change flow unit, see *Flow Unit* above.) When the entire rate field is highlighted, press ENTER. The digits can be navigated individually using the UP/DOWN arrows; switch between digits using the LEFT/RIGHT arrows. After selecting an optimal flow rate, press ENTER again to validate.

Tubing Calibration: See *Tubing Calibration*.

Pump Direction: Select the direction of the pump flow.

Sounds: Select a beep for keypad, end of dispenses, and batches.

Remote Control: See *Remote Control*.

Keypad Lockout: Allows for the keypad to be locked and unlocked.

Cumulative Volume: View and reset cumulative volume.

Main Menu: Return to the Main Menu.

Exit: Return to the Mode Operation screen.

Continuous Mode Screen

Display Legend: Below is a screenshot of the screen display for the drive in Continuous Mode. An explanation of the information on the screen follows.

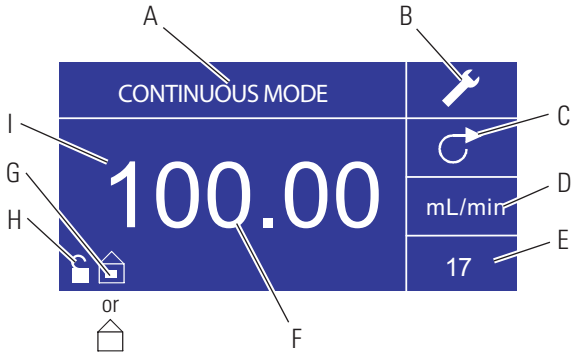


Figure 3-2. Continuous Mode Screen

A. **Mode Display:** Current operating mode in which the drive will operate. Pressing ENTER key when highlighted will cycle through the different operation modes.

B. **Setup** : Pressing the ENTER key on this icon goes to the Setup screen. The Setup screen contains most functions that can be accessed from the Continuous Mode operation screen, including: flow units, tubing size, flow rate, pump direction, remote control, and keypad lockout. The Setup screen also provides access to tubing calibration, sounds, cumulative volume and the Main Menu.

C. **Flow Direction:** Pressing the ENTER key on this icon toggles between clockwise and counterclockwise flow direction.

D. **Flow Units:** Pressing the ENTER key on this icon goes to the Flow Unit selection screen. **NOTE:** % and rpm are available in Continuous Mode only. When switching to Copy Dispense or Volume Dispense Modes % and rpm units will change to mL/min with values dependent on tubing size selected.

E. **Tubing Size:** Pressing the ENTER key on this icon goes to the tubing size selection screen.

F. **Current Flow Rate:** The center digits show the flow rate of the drive in the unit of measure selected and shown to the right (see position D, Figure 3-2).

G. **Local/Remote** or : Pressing the ENTER key on this icon goes to the Remote Control setup screen. This icon indicates whether your drive is in local or remote control mode. If the solid rectangle appears in the center of the figure the drive is set to be operated locally. If the solid rectangle does not appear in the center of the figure the drive is set to be operated by remote control.

H. **Key Pad Lock** : Pressing the ENTER key on this icon goes to the Keypad Lockout screen. Locking the keypad will prevent someone from changing the settings on the drive. When locked this icon changes to .

Continuous Mode Operation

Figure 3-3. Continuous Mode Operation

1. **Getting Started:** From the Main Menu, use the ENTER key to select Continuous Mode to enter the Continuous Mode Operation screen.

2. **Calibrating Tubing:** Before operating the pump, insert desired tubing into the Pump Head. For more information, see "*Tubing Calibration*".

3. **Preparing External Supplies:** Insert tube inlet into supply fluid. Next, insert tube outlet into desired container.

4. **Starting the Drive:** From this operation screen, simply pressing the START/STOP **key** will start the drive at the speed/flow rate and direction shown. In Continuous Mode the drive will operate at the displayed speed/flow rate and direction continuously.

5. **Stopping the Drive:** To pause or stop the drive, press the START/STOP **key** in the top right hand corner of the console.

6. **Changing Speed/Flow Rate:** To change the speed/flow rate of the drive, use the directional pad to highlight the numeric field in the center of the display and press the ENTER key. This puts you in a position to change the speed/flow rate of the drive at the farthest digit to the right (tenths, hundredths, thousandths, etc depending on flow unit). Pressing the UP arrow on the directional pad will increase the speed/flow rate by one value and pressing the DOWN arrow will decrease the speed/flow rate by one value. Pressing the ENTER key again will show all the possible digits that can be manipulated for the specific flow unit currently in use; use the LEFT/RIGHT arrows on the directional pad to move between digits and the UP/DOWN arrows to increase or decrease the value, respectively. Once desired speed/flow rate is selected, press ENTER key a final time to set the drive to operate at that speed/flow rate.

7. **Changing Flow Unit:** To change the flow unit of the drive pause the drive using the START/STOP **key.** Next, use the directional pad to select the Flow Units icon and press the ENTER key. Use the UP/DOWN arrow on the directional pad to select the desired flow unit and press the ENTER key to choose that unit. The drive will now operate in that flow unit. Press the START/STOP **key** to resume operating the drive.

Time Dispense Mode Screen

Display Legend: Below is a screenshot of the screen display for the drive in Time Dispense Mode. An explanation of the information on the screen follows.

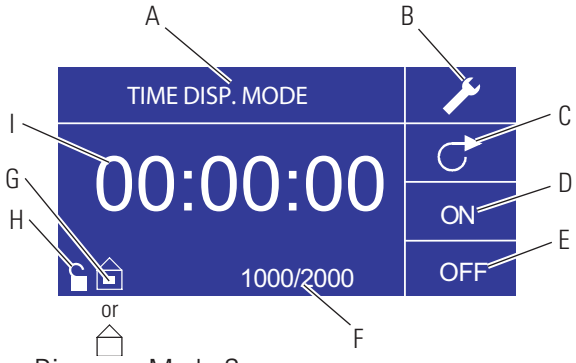


Figure 3-4. Time Dispense Mode Screen

A. **Mode Display:** Current operating mode.

B. **Setup** : The Setup screen can be used to select flow units, tubing size, flow rate, tubing calibration, sounds, cumulative volume, and Main Menu. The Setup screen contains some functions that can be accessed from the Time Dispense Mode operation screen, including: pump direction, on/off time, batch count, remote control, and keypad lockout.

C. **Flow Direction:** Pressing the ENTER key on this icon toggles between clockwise and counterclockwise flow direction.

D. **Pump ON Time:** When this field is highlighted the drive is ON. **NOTE:** The drive will not show 00:00 when switching from ON to OFF Time.

E. **Pump OFF Time:** When this field is highlighted the drive is OFF.

F. **Batch Count:** Displays the number of cycles dispensed in the batch.

G. **Local/Remote** or : Pressing the ENTER key on this icon goes to the Remote Control setup screen. This icon indicates whether your drive is in Local or Remote Control mode. If the solid rectangle appears in the center of the figure the drive is set to be operated locally. If the solid rectangle does not appear in the center of the figure the drive is set to be operated by remote control.

H. **Key Pad Lock** : Pressing the ENTER key on this icon goes to the Keypad Lockout screen. Locking the keypad will prevent someone from changing the settings on the drive. When locked this icon changes to .

I. **Time Display:** The center digits show the remaining time of the drive in the ON or OFF Time highlighted on the right of the display (position D or E, Figure 3-4).

Time Dispense Mode Operation

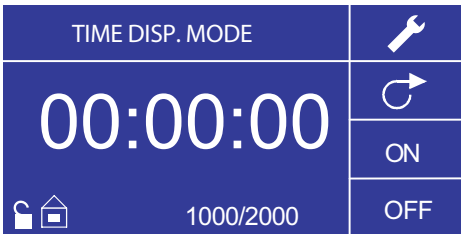


Figure 3-5. Time Dispense Mode Operation

1. **Getting Started:** From the Main Menu, use the enter key to select Time Dispense Mode to enter the Time Dispense Mode Operation screen.

2. **Calibrating Tubing**: Before operating the pump, insert desired tubing into the Pump Head. For more information, see "*Tubing Calibration*".

3. **Choosing Settings:** Select desired flow unit, tube size, flow rate, pump direction, etc. For more information see "*SETUP Menu*."

4. **Preparing Tubing:** Insert tube inlet into supply fluid. Next, insert tube outlet into desired container.

5. **Selecting Flow Rate:** Use the directional pad and ENTER key to select the Setup icon. Use the UP/DOWN arrows on the directional pad to select Flow Rate. In the Flow Rate selection screen, press the ENTER key and then use the UP/DOWN arrows on the directional pad to select a desired flow rate. For faster entry, use the LEFT/RIGHT arrows on the directional pad to move between digits and the UP/DOWN arrows to increase or decrease the value, respectively. Press ENTER one more time to validate the selected flow rate. Use the directional pad to select EXIT to return to the Time Dispense Mode Setup Screen.

6. **Setting ON Time:** To set the ON Time, use the directional pad and ENTER key to select the ON field (see position D, Figure 3-4). Doing so will highlight the timer in the center of the screen (see position I, Figure 3-4). Pressing ENTER again, allows the timer to be set using the UP/DOWN arrows. Switch between digits using the LEFT/RIGHT arrows. Having selected an optimal ON Time, press ENTER again to validate. The drive will now run for the time appearing in the center of the screen.

Time Dispense Mode Operation (continued)

7. Setting OFF Time: To set the OFF Time, use the directional pad and ENTER key to select the OFF field (see position E, Figure 3-4). Doing so will highlight the timer in the center of the screen (see position I, Figure 3-4). Pressing ENTER again, allows the timer to be set using the UP/DOWN arrows. Switch between digits using the LEFT/RIGHT arrows. Having selected an optimal OFF Time, press ENTER again to validate. The drive will stop running for the time appearing in the center of the screen. **NOTE:** If the OFF Time is set to 00:00:00, the drive requires a START/STOP **input** from the keypad or the remote I/O Connector to start the next dispense.

8. Selecting Batch Size: Before running the drive at the selected ON/OFF Times, select a batch size for the operation. To do so, use the directional pad and the ENTER key to select the BATCH icon (see position F, Figure 3-4). In the Batch Count screen, press the ENTER key and then use the UP/DOWN arrows on the directional pad to select a batch size. Switch between digits using the LEFT/RIGHT arrows. Press ENTER one more time to validate the selected batch size. When set to zero (0) the drive will run for an infinite number of cycles and the ∞ symbol is displayed. Use the directional pad to select EXIT to return to the Time Dispense Operation Screen.

9. Starting the Drive: The drive is now set to operate, press the START/STOP **key** in the upper right hand corner to start the drive. The drive can be paused at any time throughout the batch to adjust flow direction, tubing size, flow units, flow rate, etc.

10. Resetting Batch: To reset a batch, use the directional pad and the ENTER key to select the BATCH icon (see position F, Figure 3-4). In the Batch Count screen, use directional pad to select RESET and press the ENTER key to reset the batch count, select EXIT to return to the main Time Dispense Mode operation screen.

Copy Dispense Mode Screen

Display Legend: Below is a screenshot of the screen display for the drive in Copy Dispense Mode. An explanation of the information on the screen follows.

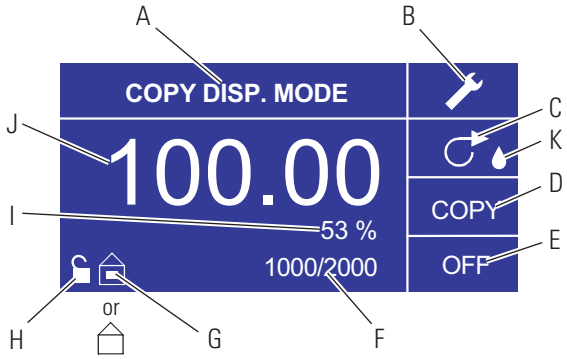


Figure 3-6. Copy Dispense Mode Screen

A. **Mode Display:** Current operating mode.

B. **Setup** : The Setup screen can be used to select flow units, tubing size, flow rate, tubing calibration, sounds, cumulative volume, and Main Menu. The Setup screen contains some functions that can be accessed from the Time Dispense Mode operation screen, including: pump direction, on/off time, batch count, remote control, and keypad lockout.

C. **Flow Direction:** Pressing the ENTER key on this icon toggles between clockwise and counterclockwise flow direction.

D. **Copy Amount Screen:** See *Copy Setting Screen*, Figure 3-8.

E. **Pump OFF Time:** Highlighted when the drive is OFF.

F. **Batch Count:** Displays the number of cycles dispensed in the batch.

G. **Local/Remote** **or** : Pressing the ENTER key on this icon goes to the Remote Control setup screen. This icon indicates whether your drive is in local or remote control mode. If the solid rectangle appears in the center of the figure the drive is set to be operated locally. If the solid rectangle does not appear in the center of the figure the drive is set to be operated by remote control.

H. **Keypad Lock** : Pressing the ENTER key on this icon goes to the Keypad Lockout screen. Locking the keypad will prevent someone from changing the settings on the drive. When locked this icon changes to .

I. **Percentage Completed:** This icon displays the portion of fluid dispensed as a percentage.

J. **Copy Volume:** Displays the Copy Volume while dispensing or the OFF Time.

K. **Anti-Drip:** A waterdrop icon present indicates that the Anti-Drip function is on. For further information see Anti-Drip Function page 3-27.

Copy Dispense Mode Operation

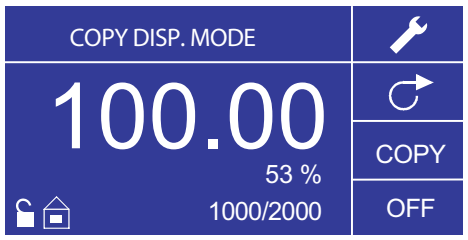


Figure 3-7. Copy Dispense Mode Operation

1. **Getting Started:** From the Main Menu, use the ENTER key to select Copy Dispense Mode to enter the Copy Dispense Mode operation screen.

2. **Calibrating Tubing:** Before operating the pump, insert desired tubing into the Pump Head. For more information, see "*Tubing Calibration*".

3. **Choosing Settings:** Select desired flow unit, tube size, flow rate, pump direction, etc. For more information see "Using the SETUP Menu."

4. **Preparing Tubing:** Insert tube inlet into supply fluid. Next, insert tube outlet into desired container.

5. **Setting Copy Amount:** See *Copy Setting Operation.*

6. **Setting OFF Time:** Use the directional pad and ENTER key to select OFF on the display to enter the Pump OFF Time. Use the directional pad and ENTER key to set the Pump OFF Time. The timer in the center of the screen will be highlighted, and using the UP/DOWN arrows will increase/decrease the farthest right digit of the time interval. Switch between digits using the LEFT/RIGHT arrows. After selecting an optimal OFF Time, press ENTER again to validate. The drive will now rest for the time appearing in the center of the screen. **NOTE:** If the OFF Time is set to 00:00:00, the drive requires a START/STOP **input** from the keypad or the remote I/O Connector to start the next dispense.

7. **Setting Batch Size:** Use the directional pad and ENTER key to select the Batch Count icon from the operation screen (see position F, Figure 3-6). From Batch Count screen use the UP/DOWN arrows to select batch size. Press ENTER to validate batch size. When set to zero (0) the drive will run for an infinite number of cycles and the ∞ symbol is displayed. Select EXIT to return to the Copy Dispense Mode screen.

   - Batch count may be reset from BATCH COUNT screen by selecting RESET.

Copy Dispense Mode Operation (continued)

8. Operating Drive: Press the START/STOP key to operate the drive at the settings selected and displayed on the screen. Press again to pause or stop the drive. Drive will automatically stop once batch is complete.

9. Reset Batch Count: Use the directional pad and the ENTER key to select the BATCH COUNT icon (see position F, Figure 3-6). In the BATCH COUNT screen, select RESET and press the ENTER key to reset the batch count. Select EXIT to return to the Copy Mode Operation screen.

10. Maximum Dispense Time: The specification for the maximum dispense in Copy Mode is over 80+ hours at 600 rpm. Actual maximum volume is dependant on tubing size and flow units selected.

COPY Setting Screen

Display Legend: Below is a screenshot of the screen display for the drive in Copy Setting Mode. An explanation of the information on the screen follows.

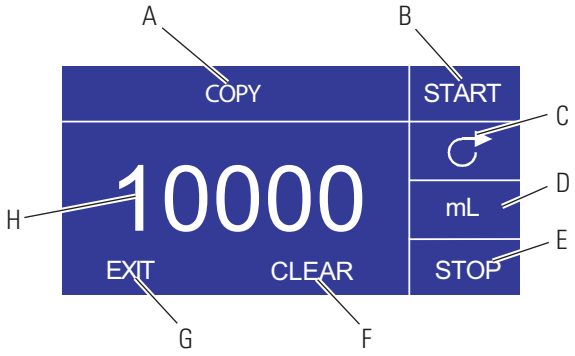


Figure 3-8. COPY Setting Screen

A. **Mode Display:** Current operating mode.

B. **START:** This icon will start drive allowing for copy volume to be set.

C. **Flow Direction:** Pressing the ENTER key on this icon toggles between clockwise and counterclockwise flow direction.

D. **Volume Unit:** This is dependent on the flow rate selected.

E. **STOP:** This stops the Copy and sets the volume to be dispensed. It is displayed in position H.

F. **CLEAR:** Selecting this will clear the number displayed on the screen and will allow for a new copy volume to be selected.

G. **EXIT:** Return to Copy Dispense Mode.

H. **Volume:** This is the amount that was dispensed during the copy.

COPY Setting Operation

Figure 3-9. COPY Setting Operation

1. **Getting Started:** From the COPY DISPENSE MODE Screen select COPY and ENTER.

2. **Clear Volume:** Using the directional Keypad select CLEAR and ENTER.

3. **Establish Copy Volume:** 3 methods are available to the user.

    a. Place the desired container at the tubing outlet. Press the START/STOP **key** to initiate the dispensing of fluid. When you have reached the desired volume press the START/STOP **key** again. Select EXIT and press ENTER. The drive will store the value of the copy in memory and use that value in the COPY DISPENSE MODE.

    b. Place the desired container at the tubing outlet. Select the START field on the screen and press the ENTER key to initiate the dispensing of fluid. The drive will now highlight the STOP field on the screen. When you have reached the desired volume press the ENTER key to stop. Select EXIT and press ENTER. The drive will store the value of the copy in memory and use that value in the COPY DISPENSE MODE.

    c. Place the desired container at the tubing outlet. Close the contacts on the START/STOP **input** to initiate the dispensing of fluid. When you have reached the desired volume, close and release the contacts on the START/STOP **input.** Select EXIT and press ENTER. The drive will store the value of the copy in memory and use that value in the COPY DISPENSE MODE.

NOTE: The value displayed as the volume in the COPY SETTING screen and the COPY DISPENSE Mode screen depend on the flow units selected. RPM, and % are invalid. If these units have been selected the drive will display a volume in mL, in the COPY DISPENSE MODE, that is dependent on the tubing size selected.

See *TUBING CALIBRATION* to improve the accuracy of this conversion.

Volume Dispense Mode Screen

Display Legend: Below is a screenshot of the screen display for the drive in Volume Dispense Mode. An explanation of the information on the screen follows.

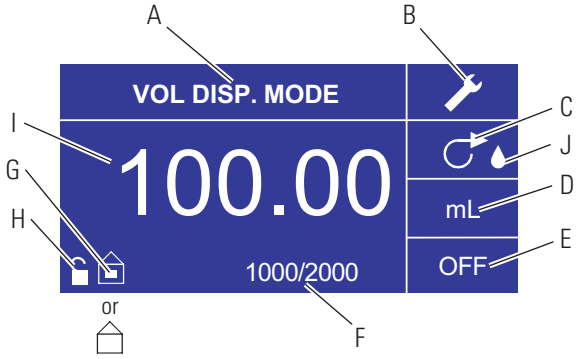


Figure 3-10. Volume Dispense Mode Screen

A. **Mode Display:** Current operating mode.

B. **Setup** : The Setup screen can be used to select flow units, tubing size, flow rate, tubing calibration, sounds, cumulative volume, and Main Menu. The Setup screen contains some functions that can be accessed from the Time Dispense Mode operation screen, including: pump direction, on/off time, batch count, remote control, and keypad lockout.

C. **Flow Direction:** Pressing the ENTER key on this icon toggles between clockwise and counterclockwise flow direction.

D. **Flow Units:** Select desired flow unit.

E. **Pump OFF Time:** Highlighted when the drive is OFF.

F. **Batch Count:** Displays the number of cycles dispensed in the batch.

G. **Local/Remote** **or** : Pressing the ENTER key on this icon goes to the Remote Control setup screen. This icon tells you whether your drive is in local or remote control mode. If the solid rectangle appears in the center of the figure the drive is set to be operated locally. If the solid rectangle does not appear in the center of the figure the drive is set to be operated by remote control.

H. **Keypad Lock** : Pressing the ENTER key on this icon goes to the Keypad Lockout screen. Locking the keypad will prevent someone from changing the settings on the drive. When locked this icon changes to .

I. **Volume:** Displays the Volume while dispensing or the OFF Time.

J. **Anti-Drip:** A waterdrop icon present indicates that the Anti-Drip function is on. For further information see Anti-Drip Function page 3-27.

Volume Dispense Mode Operation

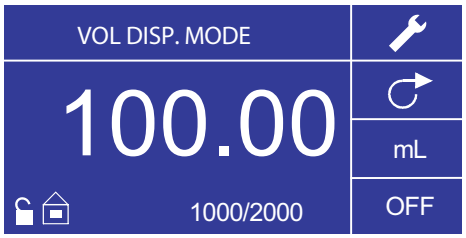


Figure 3-11. Volume Dispense Mode Operation

1. Getting Started: From the Main Menu, use the ENTER key to select Volume Dispense Mode to enter the Volume Dispense Mode operation screen.

2. Calibrating Tubing: Before operating the pump, insert desired tubing into the Pump Head. For more information, see "*Tubing Calibration*".

3. Choosing Settings: Select desired flow unit, tube size, flow rate, pump direction, etc. For more information see "*SETUP Menu*."

4. Preparing Tubing: Insert tube inlet into supply fluid. Next, insert tube outlet into desired container.

5. Setting Desired Volume: Using the directional pad highlight the numeric field in the center of the display and press the ENTER key. This puts you in a position to change the fluid volume of the drive at the farthest digit to the right (tenths, hundredths, thousandths, etc., depending on your volume unit). Pressing the UP arrow on the directional pad will increase the volume by one value and pressing the DOWN arrow will decrease the volume by one value. Pressing the ENTER key again will show all the possible digits that can be manipulated for the specific volume unit currently in use; use the LEFT/RIGHT arrows on the directional pad to move between digits and the UP/DOWN arrows to increase or decrease the value, respectively. Once desired volume is selected, press ENTER a final time to set the drive to operate at that volume. Press the START/STOP **key** to resume operating the drive.

6. Setting Pump OFF Time: Use the directional pad and ENTER key to select OFF on the display (see position E, Figure 3-10) to enter the OFF TIME. Use the directional pad and ENTER key to set the pump rest time. The timer in the center of the screen will be highlighted, and using the UP/DOWN arrows will increase/decrease the farthest right digit of the time interval. If ENTER is pressed a second time while the timer is highlighted, the digits can be navigated individually using the UP/DOWN arrows; switch between digits using the LEFT/RIGHT arrows. After selecting an optimal OFF time, press ENTER again to validate. The drive will now rest for the time appearing in the center of the screen. **NOTE:** If the OFF Time is set to 00:00:00, the drive requires a START/STOP **input** from the keypad or the remote I/O Connector to start the next dispense.

Volume Dispense Mode Operation (continued)

7. Setting Batch Size: Use the directional pad and ENTER key to select the Batch Count icon from the operation screen (see position F, Figure 3-10). From Batch Count screen use the UP/DOWN arrows to select batch size. Press ENTER to validate batch size. When set to zero (0) the drive will run for an infinite number of cycles and the ∞ symbol is displayed. Select EXIT to return to drive operation screen.

    - Batch count may be reset from the Batch Count screen by selecting RESET.

8. Operating the Drive: Press the START/STOP key to operate the drive continuously at the settings selected and displayed on the screen. Press again to pause or stop the drive. Drive will automatically stop once batch is complete.

9. Reset Batch Count: Use the directional pad and the ENTER key to select the BATCH COUNT icon (see position F, Figure 3-10). In the BATCH COUNT screen, select RESET and press the ENTER key to reset the batch count. Select EXIT to return to the COPY MODE OPERATION screen.

10. Maximum Dispense Time: The specification for the maximum dispense volume in Volume Mode is over 80+ hours at 600 rpm. Actual maximum volume is dependant on tubing size and flow units selected.

Remote Control Menu

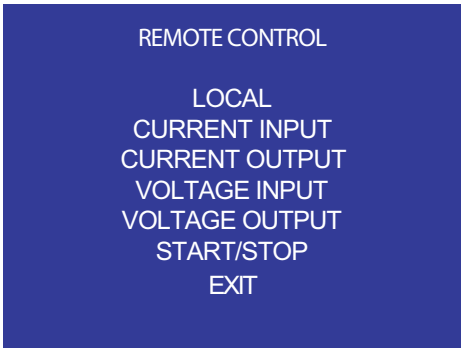


Figure 3-12. Remote Control Menu Screen

NAVIGATION: From the Main Menu or SETUP Menu select REMOTE CONTROL and ENTER.

LOCAL: When this is selected the drive is controlled by the front panel keypad, Start/Stop **Input,** Directional Input or Prime Input.

CURRENT INPUT: When this is selected, the drive is in remote control. This allows the user to input a current signal to control the flow. The user has an option to adjust the minimum, maximum and middle set points for current and flow. By default the minimum (MIN) current is set to 4.2 mA and the flow is set to 0. The maximum (MAX) is set to 20 mA and the flow is set to maximum. The middle (MID) is auto calculated for a current and flow that is centered between the MIN and the MAX. The MID can be adjusted if other profiles are needed. The scaling can be inverted if necessary. To confirm CURRENT INPUT MODE is selected, select EXIT after returning to the Remote Control Menu, then select CONTINUOUS PUMP MODE. To deselect Remote Current Input Mode select LOCAL and ENTER.

NOTE: When Current Input is selected the drive will not start until the REMOTE CONTROL MODE is exited and CONTINUOUS PUMP MODE is selected.

CURRENT OUTPUT: This allows the user to adjust the current output for a given flow. The user has an option to adjust the minimum, maximum and middle setpoints for current and flow. By default the minimum (MIN) flow is set to 0.00 and the current is set to 4.0 mA. The maximum (MAX) is set to maximum flow and the current is set to 20.0 mA. The middle (MID) is auto calculated for a current and flow that is centered between the MIN and the MAX. The MID can be adjusted if other profiles are needed. This allows for a three-point calibration of the current output. The flow is linear between these points. The scaling can be inverted if necessary. **NOTE:** Selecting Current Output will not put user into REMOTE CONTROL MODE. Only selecting VOLTAGE INPUT or CURRENT INPUT will put the user into Remote Control Mode, as indicated by the empty house icon (see position G, Figure 3-2). **NOTE:** The Current Output indicates the Running Command Speed. Use the Motor Running contacts (normally open/closed) to indicate if pump is running.

Remote Control Menu (continued)

VOLTAGE INPUT: When this is selected, the drive is in remote control. This allows the user to input a voltage signal to control the flow. The user has an option to adjust the minimum, maximum and middle setpoints for voltage and flow. By default the minimum (MIN) voltage is set to 00.1 V DC and the flow is set to 00.0. The maximum (MAX) is set to 10.0 V DC and the flow is set to maximum. The middle (MID) is auto-calculated for a voltage and flow that is centered between the MIN and the MAX. The MID can be adjusted if other profiles are needed. The scaling can be inverted, if necessary. To confirm VOLTAGE INPUT MODE is selected, select EXIT after returning to the Remote Control Menu, then select CONTINUOUS PUMP MODE. To deselect Remote Voltage Input Mode select Local and ENTER.

NOTE: When Voltage Input is selected the drive will not start until the REMOTE CONTROL MODE is exited and CONTINUOUS PUMP MODE is selected.

VOLTAGE OUTPUT: This allows the user to adjust the voltage output for a given flow. The user has an option to adjust the minimum, maximum and middle set points for voltage and flow. By default the minimum (MIN) flow is set to 00.00 and the voltage is set to 00.0V DC. The maximum (MAX) is set to maximum flow and the voltage is set to 10.0V DC. The middle (MID) is auto calculated for a voltage and flow that is centered between the MIN and the MAX. The MID can be adjusted if other profiles are needed. This allows for a three point calibration of the voltage output. The flow is linear between these points. The scaling can be inverted if necessary. **NOTE:** Selecting Voltage Output will not put the user into Remote Control Mode. Only selecting Voltage Input or Current Input will put the user into Remote Control Mode, as indicated by the empty house icon (see position G, Figure 3-2). **NOTE:** The Voltage Output indicates the Running Command Speed. Use the Motor Running contacts (normally open/closed) to indicate if pump is running.

START/STOP: The START/STOP **input** can be configured to be OFF (factory default), or ON for the drive to run.

With the OFF selected (factory default), use of the START/STOP **input** is optional. When the START/STOP **input** is open, the drive can still be started using the START/STOP **key,** PRIME key, or PRIME input. In remote modes the drive will also run if there is sufficient current or voltage at the input.

Closing the START/STOP **input** will cause the drive to run until the START/STOP **input** opens or the START/STOP **key** is pressed. In Time dispense, Copy dispense, and Volume dispense mode, only a momentary START/STOP closure is needed to start the drive. If the drive is already running in one of the dispense modes, a momentary START/STOP closure will stop the drive. In SET COPY MODE, the START/STOP **input** functions the same as in CONTINUOUS MODE; closing it will cause the drive to run until it opens.

Remote Control Menu (continued)

The function of the START/STOP **input** is considerably simplified when the ON is selected. The drive will not run under any condition unless the START/STOP **input** is closed.

Table 3-1. Continuous Mode Operation

MENU SETTINGS SETUP OPTIONS		START/STOP INPUT	INTERNAL MODE		mA or V MODE
AUTO START	START/STOP REQUIRED		Drive State When Powered OFF	Drive Response When Powered ON	Drive Running (sufficient level) When Powered OFF Drive Response when Powered ON (sufficient level present)
OFF	OFF	OPEN	Running	Not running	Not running
OFF	OFF	OPEN	Not running	Not running	Not running
OFF	OFF	CLOSED	Forced run due to S/S CLOSED	Not running	Not running
OFF	ON	OPEN	Forced not running due to S/S OPEN	Not running	Not running
OFF	ON	CLOSED	Forced run due to S/S CLOSED	Not running	Not running
ON	OFF	OPEN	Running	Running	Running
ON	OFF	OPEN	Not running	Not running	Running
ON	OFF	CLOSED	Forced run due to S/S CLOSED	Running	Running
ON	ON	OPEN	Forced not running due to S/S OPEN	Not running	Not running
ON	ON	CLOSED	Forced run due to S/S CLOSED	Running	Running

NOTE: In Continuous Mode when using the START/STOP **input** the drive is started with a closed contact and stopped when the contacts are opened.

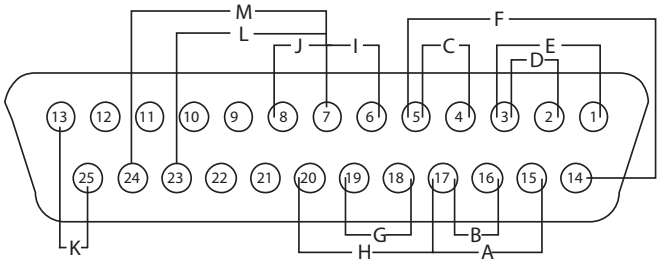
Table 3-2. Dispense Mode Operation

MENU SETTING SETUP OPTIONS		START/STOP INPUT	Drive State When Powered OFF	Drive Response When Powered ON
AUTO START	START/STOP REQUIRED			
OFF	OFF	OPEN	Running	Not running
OFF	OFF	OPEN	Not running	Not running
OFF	OFF	CLOSED*	Forced run due to S/S CLOSED	Not running
OFF	ON	OPEN	Forced not running due to S/S OPEN	Not running
OFF	ON	CLOSED	Forced run due to S/S CLOSED	Not running
ON	OFF	OPEN	Running	Running
ON	OFF	OPEN	Not running	Not running
ON	OFF	CLOSED*	Forced run due to S/S CLOSED	Running
ON	ON	OPEN	Forced not running due to S/S OPEN	Not running
ON	ON	CLOSED	Forced run due to S/S CLOSED	Running

*** NOTE:** In Dispense Modes and START/STOP MENU SETUP Option OFF the drive will start a dispense with a momentary contact closure and stop with a momentary contact closure during both the dispense period and interval period.

DB-25 Pin Configuration with Wiring Scheme

Contact Arrangements



A. START/STOP
B. CW/CCW
C. OUTPUT 0-20mA; 4-20mA
D. INPUT 0-20mA; 4-20mA
E. INPUT 0-10V
F. OUTPUT 0-10V
G. TACH OUTPUT
H. PRIME
I. MOTOR RUNNING N.O. (1A @ 24 V)
J. MOTOR RUNNING N.C. (1A @ 24 V)
K. 24V (150mA max.)
L. General Alarm
M. Local.Remote Indicator

Figure 3-13. DB-25 Pin Configuration with Wiring Scheme

Pin No. DB-25	Description
1	Speed Control Voltage Input (0-10 V)
2	Speed Control Current Input (0-20 mA)
3	Speed Control Input Ground Return
4	Speed Signal Current Output (0-20 mA)
5	Speed Signal Output Ground Reference
6	(Motor Running N.O. Default) 1A @24 V (open collector)
7	Motor Running Ground Return
8	(Motor Running N.C. Default) 1A @24 V (open collector)
14	Speed Signal Voltage Output (0-10 V)
15	Remote Start/Stop Input
16	Remote CW/CCW Input
17	Remote Start/Stop, CW/CCW, Prime Grnd Ref.
18	Tach Ground Reference
19	Tach Output (open collector)
20	Remote Prime Input
9	Reserved – Not Used
10	Reserved – Not Used
11	Reserved – Not Used
12	Reserved – Not Used
21	Reserved – Not Used
22	Reserved – Not Used
23	General Alarm (Open Collector)
24	Local.Remote Indicator (Open Collector)
25	Aux 24V+ (150 mA)
13	Aux 24V- (150 mA)

NOTE: Pins 5, 13, 17, and 18 are at earth ground, all are suitable for use with START/STOP, PRIME, Direction, Tach, LOCAL/REMOTE, General Alarm Signals and Current and Voltage Outputs.

CAUTION: Power must be turned off before connecting the external remote control cable to prevent damage to the drive.

NOTE: Open collector outputs in "low impedance" state are at earth ground and when in "high impedance" state are essentially floating. See Open Collector page following.

31-Pin Configuration with Wiring Scheme

Contact Arrangements

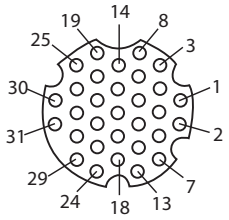


Figure 3-14. 31-Pin Configuration with Wiring Scheme

Pin No.	Description
1	Speed Control Voltage Input (0-10 V)
2	Speed Signal Voltage Output (0-10 V)
3	Speed Control Current Input (0-20 mA)
4	Remote Start/Stop Input
5	Speed Control Input Ground Return
6	Remote CW/CCW Input
7	Speed Signal Current Output (0-20 mA)
8	Remote Start/Stop, CW/CCW, Prime Grnd Ref.
9	Speed Signal Output Ground Reference
10	Tach Ground Reference
11	(Motor Running N.O. Default) 1A @24 V (open collector)
12	Tach Output (open collector)
13	Motor Running Ground Return
14	Remote Prime Input
15	(Motor Running N.C. Default) 1A @24 V (open collector)
16	Reserved – Not Used
17	Reserved – Not Used
18	Reserved – Not Used
19	Reserved – Not Used
20	General Alarm
21	Reserved – Not Used
22	Local.Remote Indicator
23	Reserved – Not Used
24	Aux 24V+ (150 mA)
25	Aux 24V- (150 mA)
26	Reserved – Not Used
27	Reserved – Not Used
28	Reserved – Not Used
29	Reserved – Not Used
30	Reserved – Not Used
31	Reserved – Not Used

NOTE: Pins 8, 9, 10, and 25 are at earth ground, all are suitable for use with START/STOP, PRIME, Direction, Tach, LOCAL/REMOTE, General Alarm Signals and Current and Voltage Outputs.

⚠ ***CAUTION: Power must be turned off before connecting the external remote control cable to prevent damage to the drive.***

NOTE: Open collector outputs in "low impedance" state are at earth ground and when in "high impedance" state are essentially floating. See Open Collector page following.

Remote Control Inputs and Outputs

INPUTS

Remote CW/CCW, Remote Start/Stop, Remote Prime, & Aux. In:

The remote control inputs work with current sinking outputs (open-collector NPN transistor outputs without passive pull-up resistors) or contact closures to DC common (earth ground). A continuous active low to the Remote Start/Stop **input** causes the drive to run, while a continuous active low to the Remote CW/CCW input causes the drive to run CCW. The motor is brought to a controlled stop before reversing direction. A continuous active low to the Remote Prime input causes the drive to run at full rated speed.

Table 3-3. Remote Control Inputs and Outputs

CURRENT CLOSED INPUT	1 mA TYP
VOLTAGE OPEN INPUT	3.2 V TYP
THRESHOLD CURRENT TO ACTIVATE	0.5 mA TYP

Remote Analog Input:

4-20 mA Input:	250 ohms typical input impedance ref. to signal ground. 4 mA, Stop; 20 mA, Full Speed (Default Settings) 10 Bit Resolution
Overload Capability:	10 V or 40 mA max.
0-10 V Input:	10 K ohms typical input impedance ref. to signal ground. 0 V, Stop; 10 V, Full Speed (Default Settings) 10 Bit Resolution

OUTPUTS

4-20 mA Output:	0 to 600 ohms max. load referenced to earth ground. 4 mA, Stop; 20 mA, Full Speed (Default Settings) 10 Bit Resolution
0-10 V Output:	1.0 K ohms min. load referenced to earth ground. 0 V, Stop; 10 V, Full Speed (Default Settings) 10 Bit Resolution
Tach Output:	Open Collector, 1.0A @ 28V DC
Frequency range:	100 to 6000 Hz or 100 to 1000 Hz, 50% Duty Cycle. (10 Hz = 1 pump rpm)
Logic Outputs:	Open Collector, 1.0 A @ 28V DC
Motor Running Outputs:	Normally Open and Normally Closed when drive is running.
General Alarm Output:	Open (High Impedance) when an alarm is displayed.
Local/Remote Indicator:	Open (High Impedance) when in remote control mode (Voltage Input, Current Input, or RS232).

Open Collector Outputs

Some remote outputs (Tachometer, Local/Remote, Motor Running and Alarm) are “open collector” type outputs and cannot be wired in the same manner as relay outputs. An open collector output is not isolated and must be configured differently than a relay output.When the open collector output is active, the output is effectively switched to earth ground and if improperly terminated could result in damage to the drive and/or external equipment.

Recommendation

When connecting to open collector outputs, the output should be connected to a current limiting resistor and then to a positive supply source which is less than 28V DC. Typically this would be connected to a 24V PLC input (see Figure 3-15).

NOTE: when using the 24V supply on the interface connector, current draw must be limited to 150 mA.

NOTE: DO NOT connect 120V supply lines to open collector outputs!

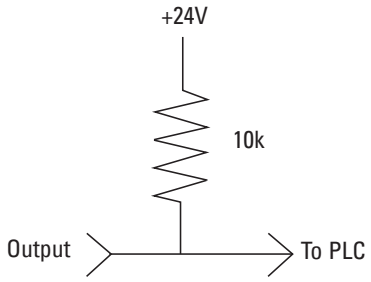


Figure 3-15. Terminating Open Collector Outputs to a PLC

Anti-Drip Function

The same drive offers an Anti-Drip feature. The tendency of fluid to drip after a dispense is dependant on several factors including tubing size, tubing orientation, and the viscosity of the fluid. To minimize this drip the drive will reverse direction after a dispense to draw the fluid back at the end of the tubing.

To access this feature select in either Copy Dispense Mode or Volume Dispense Mode ANTI-DRIP.

Figure 3-16. Anti-Drip Screen

If the ANTI-DRIP function is desired, select ON and a second screen will appear which will allow the user to input how many degrees of reverse rotation the drive will perform. Typical values range from 5 to 45 degrees. To exit without changing the current setup select EXIT.

Figure 3-17. Anti-Drip Degrees Screen

With the number highlighted press the ENTER key and use the UP and DOWN, and RIGHT and LEFT arrows to change the digits. Press the ENTER key and then select EXIT to save the setting. The drive will now reverse after every dispense.

Section 4 Maintenance

Replacement Parts and Accessories

WARNINGS: The Power switch on the Back Panel is not the main disconnect. Main disconnect is accomplished by disconnecting the detachable power supply cord at the appliance coupler or at the main plug. Ensure the power cord is easily accessible and removable, in the event of an emergency, which requires immediate disconnection.

The operator should check the detachable power supply cord condition. The equipment should not be operated if the power supply cord is cracked or broken. Any obvious damage to the enclosure (from a drop or fall) should be checked by service personnel for loose or damaged parts inside.

CAUTIONS: Replace the power cord only with one of the same type and rating. The minimum power ratings are stated on the rear panel.

The power cord set supplied with your pump drive meets the requirements of the country where you purchased the pump drive. If you use the pump drive in another country, you must use a power cord set that meets the requirements of that country.

Description	Part Number
Fuse-T3.15A, 5 x 20 mm	77500-25
Gear Service Kit (600)	07553-06
Gear Only (600 rpm)	07553-09
Gear Service Kit (100 rpm)	07553-08
Replacement seal kit (NEMA)*	07575-01
Replacement gear and shaft kit (NEMA)*	07575-02

*For washdown drives only

Fuse Replacement

1. Place the power switch in the off position.
2. Disconnect the AC power input line cord from the receptacle.
3. Remove and check the fuse and replace if defective.

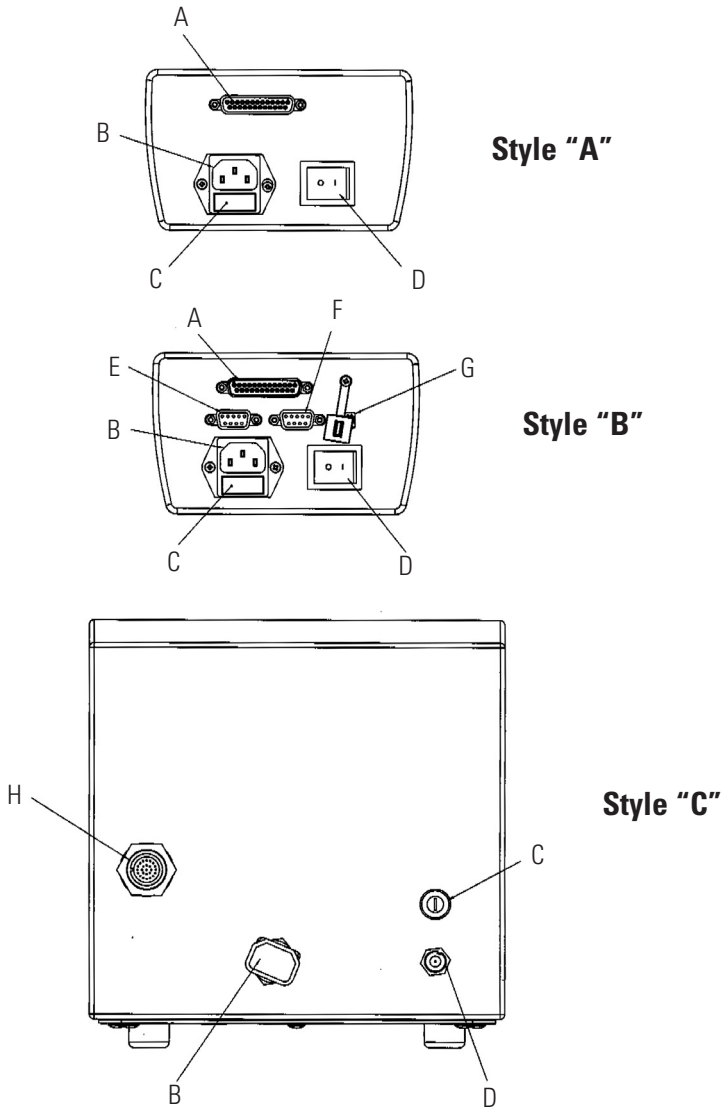
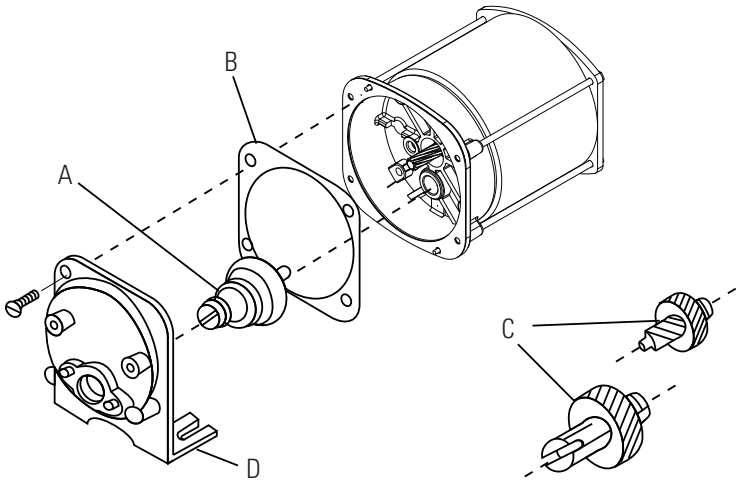


Figure 4-1. Fuse Replacement

Item	Description
A	I/O Receptacle DB-25 Pin (Style A and B)
B	IEC Power Entry Module / Line Cord
C	T3.15A (5 × 20 mm) Fuse – Do Not Substitute
D	Power Switch – All settings are retained in memory

Item	Description
E	RS-232C IN (Style B)
F	RS-232C OUT (Style B)
G	USB Port (Style B)
H	I/O Receptacle 31-Pin (Style C)

Gear Replacement



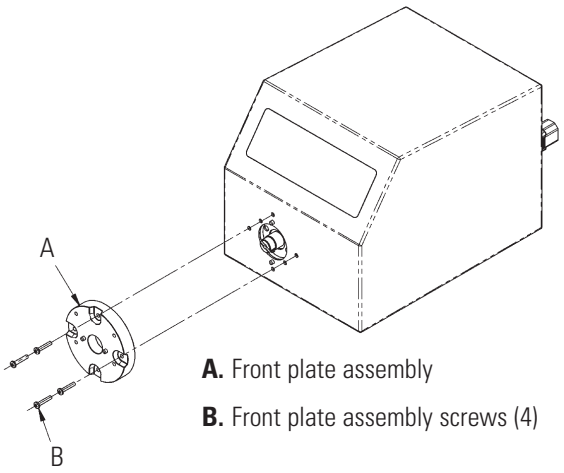
A. 6-600 rpm gear assembly (included in service kit 07553-06)
B. Gasket
C. 1-100 rpm gear set (included in service kit 07553-08)
D. Gear Case cover assembly

Figure 4-2. Motor

Shaft Seal Inspection (Stainless Steel and Powder Coated Enclosures Only)

1. Remove any pump(s) attached to the front of the drive. Clean any foreign material from the outside diameter of the drive shaft.

2. Remove the four (4) screws (see Figure 4-3, Item B) that hold the front plate assembly (see Figure 4-3, Item A) to the drive, and pull the front plate assembly off the drive. #8-32 screws may be installed in the pump-mounting holes to provide handles for pulling the plate assembly off. Retain Item B screws for Step 8. DO NOT substitute screws.



A. Front plate assembly
B. Front plate assembly screws (4)

Figure 4-3. Shaft Seal Inspection

Shaft Seal Inspection (continued)

3. Turn the front plate over so that the seal is visable. Wipe the elastomeric seal lips with a clean cloth to remove any grease and foreign material.

4. Inspect the elastomeric seal lips for tears or cuts or missing material. If any of the above mentioned conditions exist, replace the seal assembly using the 07575-01 replacement seal kit.

5. Wipe the exposed part of the drive shaft with a clean cloth. Wipe from the drive outward, to remove all grease and foreign matter.

6. Inspect the shaft surface, in the area touched by the seal. Look for a rough finish, or grooves parallel to the shaft length. If the shaft end is worn or damaged as described above, replace the gear and shaft with the 07575-02 kit. A polished groove, concentric to the outside of the shaft, is not a defect, as long as the groove is no more than 0.002 inches deep.

7. Prior to re-assembly, re-lubricate the shaft and the seal with the food-grade lubricant provided with the unit.

⚠ ***CAUTION: Do not contaminate the lubricant in the container, on the shaft or on the seal with foreign material.***

Failure to observe this precaution may result in damage to the seal and premature failure of the seal.

8. Slide the front plate assembly back over the shaft and onto the locating pins, in the orientation desired. (4 configurations, each 90 degrees of rotation apart, are possible.) Reinstall the four (4) screws, removed in step 2 (see Figure 4-3).

⚠ ***CAUTION: No foreign matter should be allowed under the gasket on the back of the front plate or under the heads of the screws.***

Failure to observe this precaution may result in leakage during washdown of the drive.

Cleaning

Keep the drive enclosure clean with mild detergents. Do not immerse or use excessive fluid when cleaning.

Section 5 Troubleshooting

Troubleshooting Chart

Symptom	Cause	Remedy
Motor does not rotate, Display does not light.	No Power.	1. Check fuse and replace, if necessary.
		2. Check that unit is plugged into a live line.
		3. Check connection of power cord.
		4. Check the line cord for continuity and replace if defective.
		5. Return for servicing.
Motor does not rotate. Display lights.	Defective Remote Control or Setting Error.	1. Place power switch in OFF position.
		2. Check that remote cable connector is fully inserted into the receptacle.
		3. Reapply power.
		4. If motor still does not rotate, select remote control in Main Menu or Setup Menu and verify settings.
		5. Return to Mode screen and verify icon shows Remote Control Mode.
		6. See *Remote Control Mode* in this manual for further details.
	START/STOP Mode "ON" without an input at I/O Connector.	1. See *Remote Control Mode* in this manual for further details.
		2. Select "OFF" in START/STOP Menu to run without an input at the I/O Connector cable.
Drive does not follow Serial or USB Commands	Hardware or Firmware issue.	1. Verify cable connection to drive.
		2. COM Port selection error. See *WINLIN software*. (Hyper terminal not included)

Error Definitions

Error #2 Motor Overspeed

Description:	The drive has exceeded commanded speed value.
Error Condition(s):	The motor has exceeded the commanded speed value by 20%.
Actions:	Drive will stop immediately. Verify load is correct and power cycle drive. If error persists consult factory.

Error #3: Instantaneous Over-Current

Description:	Motor is drawing too much current for a short duration of time.
Error Condition(s):	The motor current is above 4.0 A peak.
Actions:	Drive will stop immediately. Verify that pump head is not binding and that the load is not above recommended maximum load. If error persists consult factory.

Error #4: Bad Flash Checksum

Description:	Run-time checksum (checked at power-on) contains a bad checksum value.
Error Condition(s):	Checksum is checked at power-on for an invalid value.
Actions:	Power cycle the drive. If error persists consult factory.

Error #7: Bad EEPROM Checksum (Settings)

Description:	Bad EEPROM checksum on parameter values and settings, or its data is out of range.
Error Condition(s):	1) Checksum value in EEPROM does not match calculated value. 2) Data in EEPROM is out of range.
Actions:	Error will be cleared after 10 seconds and parameters will be reset to default values. If error persists consult factory.

Error #8: Bad EEPROM Checksum (Factory Cal)

Description:	Bad EEPROM checksum for Factory Cal
Error Condition(s):	1) Checksum value in EEPROM does not match calculated value. 2) Data in EEPROM is out of range.
Actions:	Error will be cleared after 10 seconds and parameters will be reset to default values. If error persists consult factory.

Error Definitions (continued)

Error #9: EEPROM Write Verification Error

Description:	Data written to EEPROM does not match.
Error Condition(s):	Data values do not match.
Actions:	Error will be cleared after 10 seconds and parameters will be reset. If error persists consult factory.

Error #10: Bus Over Voltage

Description:	The measured AC voltage reported by the drive is too high.
Error Condition(s):	The drive voltage is above 260V AC.
Actions:	The pump will stop immediately, check the supply line voltage. If error persists consult factory.

Error #11: Bus Under Voltage

Description:	The measured AC voltage reported by the drive is too low.
Error Condition(s):	The drive voltage is below 90V AC.
Actions:	The pump will stop immediately, check the supply line voltage.
NOTE:	This error when displayed during power down is considered normal and proper. If error persists consult factory.

Error #12: Motor Stall / Motor Under Speed

Description:	The motor was commanded to run, but has either slowed down significantly or has stopped.
Error Condition(s):	The motor speed is below 95% of the desired speed for too long a period of time.
Actions:	The motor will be commanded to stop. Verify the pump turns freely and is not binding. If error persists consult factory.

Error #14: Ambient Over Temperature

Description:	The motor control board is overheating.
Error Condition(s):	The temperature value from motor control board is above given threshold value.
Actions:	The pump will stop immediately. Verify that the ambient air temperature is less than 104° F (40° C). Verify the pump turns freely and that there is no restriction of air flow. If error persists consult factory.

Error Definitions (continued)

Error #15: Motor Feedback Fault

Description:	Communications to the motor control board is not correct, has disappeared, or some other communications fault.
Error Condition(s):	No data coming back over the serial port from the motor control board.
Actions:	The drive will attempt to stop the pump. Power cycle drive. If error persists consult factory.

Error #16: Invalid Interrupt or Address

Description:	Software jumps to an invalid address, invalid interrupt, or other abort/exception (i.e., Data Abort Exception). This may occur due to invalid pointer references, or ram memory corruption, etc.
Error Condition(s):	These are handled by an Abort Exception/Interrupt within the CPU and should branch out to their respective exception handler functions.
Actions:	Power cycle the drive to reset error. If error persists consult factory.

Error #18: Watchdog Error

Description:	Program has stopped running as the watch dog has not been updated, i.e., Software Locked up.
Error Condition(s):	Interrupt triggered when the Watchdog has not been updated.
Actions:	Power cycle drive to reset error. If error persists consult factory.

Section 6 Accessories

1.	Footswitch w/DB-25 male	07523-92
2.	Connector DB-25 male	07523-94
3.	Dispensing Wand DB-25 male	07523-97
4.	Footswitch (NEMA)*	07575-84
5.	Remote control cable (NEMA)*, 25ft (7.62 m)	07575-80

*For washdown drives only.

Section 7 Specifications

Output

Speed:	
600 rpm models	0.1 to 600 rpm
100 rpm models	0.02 to 100 rpm
Torque output, Maximum:	
600 rpm models	180 oz-in (13 kg•cm) 540 oz-in Starting
100 rpm models	360 oz-in (26 kg•cm) 1080 oz-in Starting
Speed regulation:	
All models	Line ±0.1% F.S. Load ±0.1% F.S. Drift ±0.1% F.S.
Display:	
All models	128 x 64 LCD w/ LED Backlight
Remote outputs:	
All models	Voltage speed output (0–10V DC @ 1 kΩ min)
All models	Current speed output (0–20 mA @ 0–600 Ω)
600 rpm models	Tach output (100 to 6000 Hz, 50% duty cycle, 10 Hz/rpm)
100 rpm models	Tach output (100 to 1000 Hz, 50% duty cycle, 10 Hz/rpm)
All models	Motor running output (N.O. & N.C. open collector, 1A @ 28V DC)

Input

Supply voltage limits:	
All models	90 to 260 Vrms @ 50/60 Hz (Universal Input) Single Phase Only
Current, max.:	
All models	1.8A @ 115 Vrms, or 1.1A @ 230 Vrms
Remote Inputs:	
All models	STOP/START, CW/CCW, PRIME (Contact closure)
All models	Voltage input (0–10V DC @ 10 kΩ), ±50V common mode range
All models	Current input (0–20 mA or 4–20mA @ 250 Ω), ±50V common mode range

Construction

Dimensions (L × W × H):	
Models w/plastic enclosure	10.5 in × 8 in × 8 in (267 × 203 × 203 mm)
Models w/stainless steel or powder coated steel enclosure	14.0 in x 9 in x 9.5 in (356 × 229 × 241 mm)
Weight:	
Models w/plastic enclosure	13 lb (5.9 kg)
Models w/stainless steel or powder coated steel enclosure	26 lb (11.8 kg)
Enclosure Rating:	
Models w/plastic enclosure	IP 33 per IEC 60529
Models w/stainless steel or powder coated steel enclosure	IP 66 per IEC 60529/NEMA 4X – indoor use

Environment

Temperature, Operating:	
All models	0° to 40°C (32° to 104°F)
Temperature, Storage:	
All models	–25° to 65°C (–13° to 149°F)
Humidity (non-condensing):	
Models w/plastic enclosure	10% to 90%
Models w/stainless steel or powder coated steel enclosure	10% to 100%
Altitude:	
All models	Less than 2000 m
Pollution Degree:	
Models w/plastic enclosure	Pollution Degree 2 (Indoor use — lab, office)
Models w/stainless steel or powder coated steel enclosure	Pollution Degree 3 (Indoor use — Sheltered locations)
Chemical Resistance:	
Models w/plastic enclosure	Exposed material is aluminum, ABS plastic and vinyl
Models w/stainless steel or powder coated steel enclosure	Exposed material is 316 enclosure stainless steel, vinyl and powder coated steel
Compliance:	Conforms to ANSI/UL Std 61010-1 Certified to CAN/CSA Std C22.2 No. 61010-1. This product has been tested to the requirements of CAN/CSA-C22.2 No. 61010-1, second edition, including Amendment 1, or a later version of the same standard incorporating the same level of testing requirements. (For CE Mark): EN61010-1 (EU Low Voltage Directive) and EN61326 (EU EMC Directive)

Section 8 Warranty, Product Return and Technical Assistance

Warranty

Use only MASTERFLEX precision tubing with MASTERFLEX pumps to ensure optimum performance. Use of other tubing may void applicable warranties.

This product is warranted against defects in material or workmanship, and at the option of the manufacturer or distributor, any defective product will be repaired or replaced at no charge, or the purchase price will be refunded to the purchaser, provided that: (a) the warranty claim is made in writing within the period of time specified on the warranty card, (b) proof of purchase by bill of sale or receipted invoice is submitted concurrently with the claim and shows that the product is within the applicable warranty period, and (c) the purchaser complies with procedures for returns set forth in the general terms and conditions contained in the manufacturer's or distributor's most recent catalog.

This warranty shall not apply to: (a) defects or damage resulting from: (i) misuse of the product, (ii) use of the product in other than its normal and customary manner, (iii) accident or neglect, (iv) improper testing, operation, maintenance, service, repair, installation, or storage, (v) unauthorized alteration or modification, or (b) post-expiration dated materials.

THIS WARRANTY IS THE EXCLUSIVE REMEDY OF THE PURCHASER, AND THE MANUFACTURER AND DISTRIBUTOR DISCLAIM ALL OTHER WARRANTIES, WHETHER EXPRESS, IMPLIED, OR STATUTORY, INCLUDING WITHOUT LIMITATION, WARRANTIES OF MERCHANTABILITY AND FITNESS FOR A PARTICULAR PURPOSE. NO EMPLOYEE, AGENT, OR REPRESENTATIVE OF THE MANUFACTURER OR DISTRIBUTOR IS AUTHORIZED TO BIND THE MANUFACTURER OR DISTRIBUTOR TO ANY OTHER WARRANTY. IN NO EVENT SHALL THE MANUFACTURER OR DISTRIBUTOR BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

The warranty period for this product is two (2) years from date of purchase.

Product Return

To limit charges and delays, contact the seller or Manufacturer for authorization and shipping instructions before returning the product, either within or outside of the warranty period. When returning the product, please state the reason for the return. For your protection, pack the product carefully and insure it against possible damage or loss. Any damages resulting from improper packaging are your responsibility.

Technical Assistance

If you have any questions about the use of this product, contact the Manufacturer or authorized seller.

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Application Note

A Hands-On Guide to Ultrafiltration/ Diafiltration Optimization using Pellicon® Cassettes

In ultrafiltration (UF) tangential flow filtration (TFF) systems, operating parameter selection will have far reaching impact as the process is scaled to full-scale manufacturing levels. While there are many factors that contribute to final system design, several key parameters should be optimized early in the process development phase. The goal is to develop a robust process with the following success criteria: superior product quality, consistent and high product yield, reproducible process flux and time, and a cleaning regime that allows extended membrane reuse.

The following basic experiments should be considered during development of processing methodology:

- Optimization
  - Impact of transmembrane pressure (TMP) and feed flow on process flux and retention
  - Impact of product concentration and buffer conditions on process flux and retention
  - Impact of diavolumes on buffer exchange and contaminant removal
- Paper design and full process simulation with chosen processing parameters

Typically, the first three experiments are performed sequentially to bracket process performance and obtain data for analysis. This information is then combined with actual manufacturing considerations (batch volume, process time, etc.) to design a process simulation.

The purpose of a process simulation is to duplicate the entire manufacturing process in a scale-down format, to confirm sizing, and to assess preliminary product quality and yield. The intent is to develop an optimized process, on the bench, that will efficiently scale-up to meet full-scale manufacturing expectations.

Sequence	Purpose
1. TMP Excursion at Initial Concentration ($C_{b\ initial}$)	• Determine TMP for UF/DF • Determine Feed Flow (Q_F) for UF/DF • Demonstrate Flux Stability • Confirm Retention of Product
↓	
2. Concentration / Volume Reduction ($C_{b\ initial} \rightarrow C_{b\ final}$)	• Determine Flux as Function of Concentration • Determine Placement of Diafiltration Step • Determine Flux as Function of Buffer Conditions
↓	
3. TMP Excursion at Final Concentration ($C_{b\ final}$)	• Determine TMP for UF/DF • Determine Feed Flow (Q_F) for UF/DF • Confirm Retention of Product
↓	
4. Diafiltration / Buffer Exchange	• Determine Diavolume Requirement • Confirm Retention of Product during DF
↓	
5. Product Recovery	• Crude Assessment of Step Yield • Product Quality Evaluation

Figure 1. Basic Optimization Experiments

Use this step-by-step guide to develop a robust UF/DF process with Pellicon® cassettes (cutoffs of 100 kD and lower) that will deliver superior product quality, reproducible results, and high yields.

EMD Millipore is a division of Merck KGaA, Darmstadt, Germany

The following are step-by-step protocols for basic optimization experiments.

Set-up and Installation Procedure

Refer to the *Maintenance Procedures for Pellicon® and Pellicon® 2 Cassette Filters (P17512)* or the *Pellicon® 3 Filters Installation and User Guide (AN1065EN00)* when performing actual set-up and installation of Pellicon® cassettes.

1. Assemble the TFF system as shown in Figure 2.
2. Install the Pellicon® cassette(s) (Pellicon® 2 Mini with 0.1 m^2 membrane area, Pellicon® 3 with 0.11 m^2 membrane area) in the appropriate Pellicon® holder.
3. Flush the system with water, clean with the appropriate cleaning agent (per appropriate maintenance guide), and flush again.

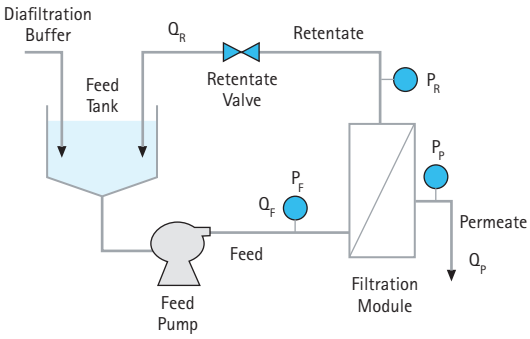

Figure 2. Schematic of a TFF System

Equilibration Procedure

1. Add 3 L/m^2 of the appropriate buffer to the feed tank. *Example:* 0.1 m^2 membrane area x 3 L/m^2 = 0.3 L buffer
2. Direct the retentate and permeate to a waste container.
3. Start the feed pump and achieve the following conditions by partially closing the retentate valve and adjusting the pump speed:
   - Feed flow of 5 L/min/m^2
   - Retentate pressure of 2 – 15 psi (0.14 – 1.03 bar) to achieve approximately 30% conversion
4. When half the buffer has been flushed, put the system in total recycle mode[1] and recirculate for 10 minutes; verify that the pH and conductivity in the system have been equilibrated to the level of the starting buffer.
5. Direct the retentate and permeate to a waste container.
6. When the feed tank level reaches the minimum level, open the retentate valve fully and stop the feed pump to prevent the introduction of air into the system.

Part 1. TMP Excursion at Initial Concentration

1. Add sufficient volume of product to the feed reservoir such that final volume or concentration target can be reached or slightly exceeded (approximately 1 – 1.5 L of final product at final concentration per m^2). *Example:* if $C_{initial}$ = 10 g/L and $C_{retentate}$ = 80 g/L, then the concentration factor is 8X. If the minimum achievable final volume for 0.1 m^2 is 0.1 L, calculate the required initial volume:

   $$V_{initial} = V_{minimum} \times VCF = 0.1\ L \times 8X = 0.8\ L$$

2. Open the retentate valve fully and configure system in total recycle mode.
3. Start the feed pump and achieve the following conditions by partially closing the retentate valve and adjusting the pump speed:
   - Recommended feed flow (Q_F) rate for the membrane device, typically 5 L/min/m^2 for Pellicon® 2 and 3 cassettes
   - Minimal TMP, typically 2 – 5 psi (0.14 – 0.34 bar) for more open membranes and 10 psi (0.69 bar) for tighter membranes.
4. Recirculate the product for 10 – 15 minutes and ensure that stable process flux is achieved[2].
5. Record temperatures, pressures, and flows; sample feed and permeate for product retention[3].
6. Increase TMP by 5 – 10 psid (0.34 – 0.69 bar) by manipulating the retentate valve while keeping the feed flow constant. For more open membranes increase by 2 – 5 psid (0.14 – 0.34 bar). Repeat steps 4 and 5.
7. Repeat step 6 until flux begins to level off[4]; typically 4 – 6 TMP values are evaluated in total.
8. Open the retentate valve fully and allow system to continue in a total recycle.
9. Increase or decrease the feed flow by 2 – 3 L/min/m^2 and repeat steps 4 through 8. If desired, a third feed flow rate can be investigated.
10. Plot the data as shown in Figure 3.

Tight membranes (1 kD, 5 kD, etc.)	Can use large TMP increases since optimum is typically > 30 psi
Open membranes (50 kD, 100 kD, etc.)	Can use small TMP increases since optimum is typically < 10 psi

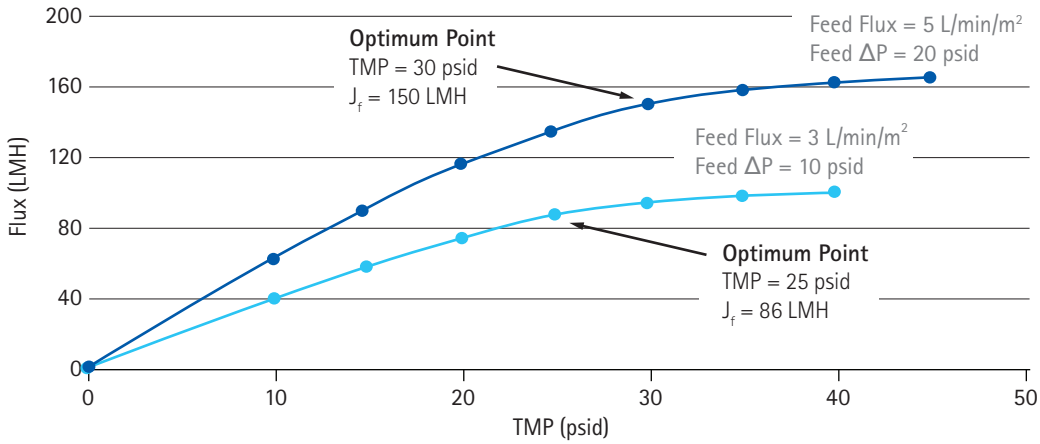

Figure 3.
TMP Excursion at Two Feed Flows

	Q_1 = 5 L/min/m²	Q_2 = 3 L/min/m²	Q_1/Q_2
A [m²]	Volume / Time / 150 LMH	Volume / Time / 86 LMH	0.57
Q_F [L/min]	(5 L/min/m²) x Volume / Time / 150 LMH	(3 L/min/m²) x Volume / Time / 86 LMH	0.95

Table 1. Membrane Area vs. Pump Feed Rate (Figure 3)

Calculations

The appropriate combination of feed flow rate and TMP will maximize flux while minimizing the impact of pumping and shear on the product. The appropriate combination of these two parameters will also minimize processing time and/or membrane area. To calculate the optimum feed flow, compare the required membrane area with the required pump rate at each of the two feed flow conditions, as shown in Table 1.

Membrane Area [m²] =
Process Volume [L] / (Flux [LMH] x Process Time [h])

In Figure 3:

$Area_{Q1} = 0.57 \times Area_{Q2}$

Pump feed rate [L/min] =
Feed flux [L/min/m²] x Area [m²]

In Figure 3:

$Pump\ feed\ rate_{Q1} = 0.95 \times Pump\ feed\ rate_{Q2}$

In this example it is advantageous to run at the higher feed flow, Q_1, since it only requires 57% of the membrane area used at the lower feed flow rate at almost the identical pump feed rate.

Note:

- Anticipated final volume of over-concentrated product must exceed minimum working volume of membrane system at selected feed flow rate (Q_F); avoid introduction of air and maintain uniform mixing at end of volume reduction.
- Move from least to greatest fouling conditions:
  - Do not test into pressure-independent regime (past the knee of the flux vs. TMP curve)[4]
  - Avoid exceeding 30 – 40% conversion ratios
- Check hysteresis if possible by returning the system to the initial conditions and taking a final flux measurement; compare initial flux performance to final flux performance at initial conditions.
- Ensure that choice of TMP and feed flow have corresponding retention values that are acceptable (> 0.998) at both initial and final product concentration and in each buffer[5].
- There is often very little performance difference versus feed flow rate at low product concentration. However, at the higher concentrations that will be investigated in Parts 2 and 3, the benefits of different feed flow rates should become more pronounced.

Part 2. Concentration

1. Use the product from Part 1 in the starting buffer. Based on desired final product concentration factor, add additional feed volume as needed to ensure sufficient volume at end of concentration[6].
2. Sample feed to confirm product concentration.
3. Put the system in total recycle.
4. Start the feed pump and achieve the optimum TMP and feed flow as determined in Part 1 by partially closing the retentate valve and adjusting the pump speed.
5. Direct the permeate to a separate container to concentrate product and reduce volume.
6. Record temperatures, pressures, and flows throughout the concentration; sample feed and permeate for product retention[7].
7. Concentrate slightly beyond desired final product concentration.
8. Repeat the TMP excursion outlined in Part 1 to determine optimum TMP at the final concentration in the starting buffer.
9. Diafilter with one diavolume to get product into final buffer and dilute with final buffer back to initial concentration.
10. Repeat the TMP excursion to determine the optimum TMP at the initial concentration in the final buffer.
11. Repeat Part 2 steps 2 – 7 once in final buffer using the optimum TMP as determined above.
12. Plot the data as shown in Figure 4, remembering to apply a temperature correction in the flux calculations[8].

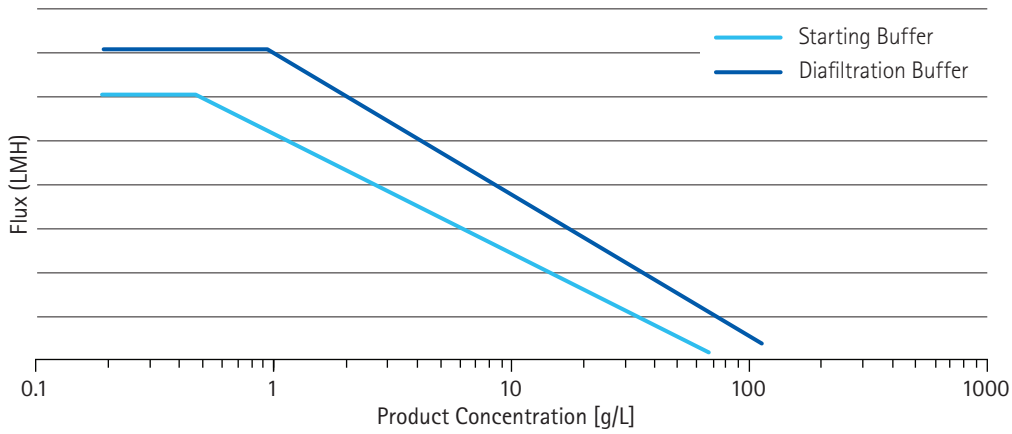

Figure 4. Flux vs. Concentration

Calculations

The tradeoff between flux and diafiltration buffer volume create an optimum bulk concentration at which to perform diafiltration; this can be calculated using the DF optimization parameter at each data point:

DF Optimization Parameter =
Concentration [g/L] x Flux [LMH]

Plotting the DF optimization parameter as a function of product concentration yields the optimum concentrations for diafiltration in both the starting and final buffers, as shown in Figure 5.

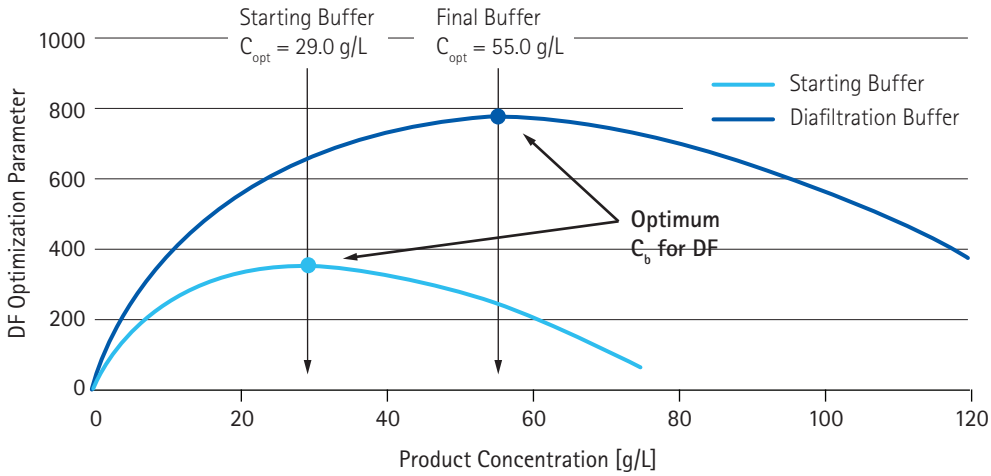

Figure 5. DF Optimization

There is an alternative approach that may be used to calculate the optimum concentration at which to perform diafiltration (C_{opt}). It assumes that the product is completely retained and that the passage of the permeating species is constant.

If the flux versus concentration data is plotted as shown in Figure 4, then the gel concentration, C_g, is the concentration at which the permeate flux reaches zero (example: ~80 g/L in the starting buffer, ~110 g/L in the final buffer). The optimum concentration at which to perform diafiltration is then calculated as[9]:

C_{opt} [g/L] = C_g [g/L] / e

In Figure 4:
Starting buffer C_{opt} = 80 / 2.71828 = 29.4 g/L
Final buffer C_{opt} = 110 / 2.71828 = 40.5 g/L

The C_g/e method can only be used when the flux vs. concentration data allows for accurate extrapolation to zero flux.

Note:

- Ensure enough feed material and appropriate system working volume in order to achieve the final concentration.
- Based on the results of the additional TMP excursions performed in Part 2, the TMPs used for concentration in both the starting and final buffers should be changed and the concentration should be repeated to obtain more accurate data.
  - If the optimum TMP for the dilute solution occurs in the pressure-independent region (past the knee of the curve) for the concentrated solution, then the TMP should be decreased to the lowest optimum value.
  - If the optimum TMP for the dilute solution occurs within the pressure-dependent region (before the knee of the curve) for the concentrated solution, then the TMP may be increased to the highest optimum value to further optimize the flux and reduce the processing time.
- Optimum concentration for diafiltration will be different for each buffer; choose an average or the most conservative.
  - Restrictions on buffer usage or minimum recirculation volume often dictate the concentration at which diafiltration occurs.
  - If the required final concentration is significantly less than the optimum concentration for diafiltration, over concentration followed by dilution is a possible option, although rarely chosen. It should only be considered in cases where diafiltration buffer is limited and the product is stable at the higher concentrations.

Part 3. TMP Excursion at Final Concentration

1. Use the product from Part 2 at the final concentration in the final buffer.
2. Repeat steps 2 – 10 of Part 1.

Calculations

Reference Part 1.

Note:

Reference Part 1 and Part 2 notes.

Part 4. Diafiltration

1. Use the product from Part 3 at the optimum concentration for diafiltration; dilute as needed using the final buffer.
2. Configure the system for constant volume diafiltration.
3. Start the feed pump and achieve the optimum TMP and feed flow as determined in Part 1 and Part 3.
4. Diafilter the product with the chosen number of diavolumes:
   - Choose the number of diavolumes based on the product purity specifications (if known, see calculation below) and add a safety factor of 2 diavolumes, or
   - Use 3 – 5 diavolumes as an initial estimate for upstream UF/DF steps, or
   - Use 7 – 12 diavolumes as an initial estimate for final formulation UF/DF steps
5. Record temperatures, pressures, and flows at every diavolume; sample feed and permeate for both product retention, and retention and concentration of the contaminant of interest.
6. Plot the data as shown in Figure 6.

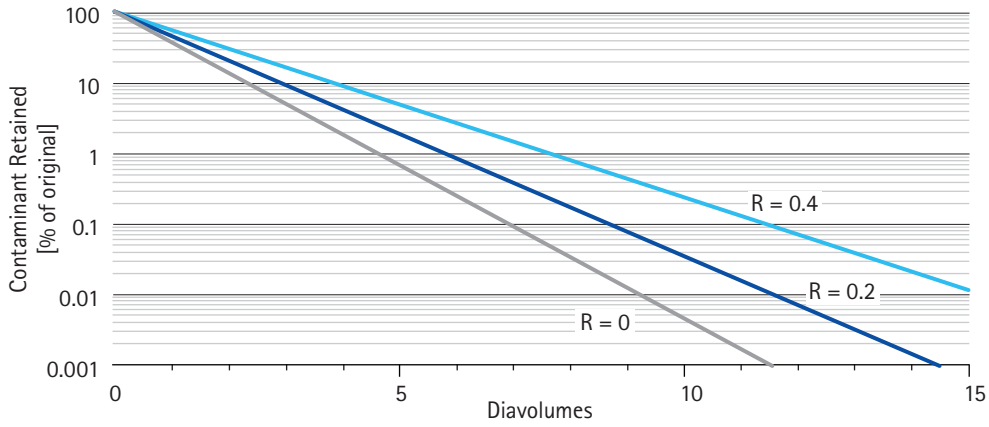

Figure 6. contaminant removal vs. Diavolumes

Calculations

The percentage of the original contaminant in the retentate at each diavolume can be calculated from the retention values using the following:

$$\text{Remaining Contaminant [\%]} = 100 \times e^{(\text{Retention} - 1) \times N}$$

where N is the number of diavolumes.

However, since contaminant concentration is being directly measured in each feed sample throughout diafiltration, plot these concentrations as a percentage of the original and use the above equation to plot several lines of theoretical retention, as shown in Figure 6. This plot will help demonstrate the contaminant removal at various retentions.

Select the whole number of diavolumes based on the acceptable contaminant levels for the product; always add 2 – 3 diavolumes as a 10-fold safety factor for critical diafiltration steps, such as final formulation. For upstream steps, add 1 – 2 diavolumes. If the goal of diafiltration is not to wash out a contaminant but rather to reach a target pH or conductivity, then the measurement of that quality can be plotted against the number of diavolumes instead.

Note:

- If it appears necessary to diafilter past ~ 14 diavolumes, any dead-legs or poor mixing areas in the system will increase the apparent retention of the contaminant and make further removal difficult.
- Ensure that choice of TMP and feed flow have corresponding product retention values that are acceptable (> 0.998) throughout diafiltration.

Part 5. Product Recovery

There are various methods for product recovery at large-scale[10]. However, at small-scale, sufficient product recovery can be achieved by manually tilting the system and breaking the piping at low-points to drain the product. Samples of the final retentate should then be analyzed for product concentration and quality.

1. After the product has been drained from the system, add one system volume of diafiltration buffer to the feed tank.
2. Recirculate at the selected feed flow rate with the retentate valve fully open for 10 minutes.
3. Recover the buffer in a separate container using the same methods that were used to recover the product. Samples of this buffer rinse should be analyzed for product concentration.
4. After the product is recovered, the system should be cleaned with the appropriate solutions[11].

Calculations

Ideally, the total product mass recovered in the retentate, permeate, and buffer flush as well as unrecoverable holdup volume should equal the total mass of product in the feed. If the total product mass recovered is less than the initial product mass, it is typically due to adsorption and/or solubility losses during processing[12]. However, it is important to perform a mass balance and calculate total yield to ensure optimum process parameters.

The theoretical yield can also be calculated based on the membrane retention and compared to the actual yield.

$$\text{Theoretical Yield [\%]} = 100 \times e^{(\text{Retention} - 1)(N + \ln X)}$$

where N = number of diavolumes and X = concentration factor.

$$\text{Actual Yield [\%]} = 100 \times (V_{retentate}\,[L] \times C_{retentate}\,[g/L]) / (V_{initial}\,[L] \times C_{initial}\,[g/L])$$

$$\text{Mass Balance [\%]} = 100 \times \{(V_{retentate}\,[L] \times C_{retentate}\,[g/L]) + (V_{permeate}\,[L] \times C_{permeate}\,[g/L]) + (V_{rinse}\,[L] \times C_{rinse}\,[g/L])\} / (V_{initial}\,[L] \times C_{initial}\,[g/L])$$

Note:

- All calculations are estimates; during these optimization steps, the product has undergone more processing than normal. Product degradation and yield may be slightly affected. For a true indication of processing on product quality, perform the entire optimized process using fresh feed and new membranes.
- Product can be very viscous when recovered and may affect assays; perform serial dilutions for more accurate assay results.
- Actual yield and mass balance percentages should be close to 100% and/or theoretical yield. If significant losses occur, process parameters (including membrane type) may have to be changed and then re-optimized.
- In a robust process, adsorption and solubility losses should be very low.

Paper Design and Process Simulation

The optimization parameters obtained from the previous experiments can be combined to design a full process simulation: concentration, diafiltration, (concentration,) and recovery. If time permits, a process simulation should be run immediately following the optimization work, and should employ the following:

- New set of cassettes; same membrane type, same cassette path length
- Fresh feedstock
- Fresh buffer(s)
- Optimized process parameters
- See detailed process simulation calculations below.

After performing the process simulation, the system should be cleaned with the appropriate solution according to EMD Millipore recommendations[11]. If possible, the process should be rerun using the cleaned membranes to determine the effectiveness of the cleaning cycle and the consistency of membrane performance from run-to-run. If the cleaning cycle does not prove effective, the cleaning parameters or cleaning solutions will need to be changed and the cleaning cycle will have to be tested again.

Calculations

The membrane area can be optimized to allow the entire process (both concentration and diafiltration) to be completed in the specified timeframe (3 – 4 hours is recommended). The average flux for each concentration and diafiltration step can be estimated from the optimization data and combined with the desired volumes to be processed. The approximate required membrane area can then be calculated for both manufacturing scale and scale-down runs.

Assume an example process scenario (this would have been determined by optimization data, DF parameter, etc.):

- 2.9X Concentration:
  10 g/L to 29 g/L; flux decreases from 150 LMH to 80 LMH
- 7X Diafiltration:
  29 g/L; flux increases from 80 LMH to 85 LMH
- 3.4X Concentration:
  29 g/L to 100 g/L; flux decreases from 85 LMH to 20 LMH
- Desired process time is 4 hours

Manufacturing scale volumes as determined by the customer:

- Feed volume = 5000 L
- Retentate volume at end of 2.9X concentration = 5000 L/2.9 = 1724 L
- Permeate volume removed during 2.9X concentration = 5000 L – 1724 L = 3276 L
- 7X Diafiltration buffer volume = 7 x 1724 L = 12,068 L
- Retentate volume at end of 3.4X Concentration = 1724 L/3.4 = 507 L
- Permeate volume removed during 3.4X concentration = 1724 L – 507 L = 1217 L

Average process flux for concentration step[13]:

$J_{avg} = J_{final} + 0.33 (J_{initial} - J_{final}) = J_{initial} \times 0.33 + J_{final} \times 0.67$

For 2.9X concentration:

$J_{avg} = 150 \text{ LMH} \times 0.33 + 80 \text{ LMH} \times 0.67 = 103 \text{ LMH}$

For 3.4X concentration:

$J_{avg} = 85 \text{ LMH} \times 0.33 + 20 \text{ LMH} \times 0.67 = 41 \text{ LMH}$

Average process flux for diafiltration step:

For diafiltration the average flux can be estimated as the initial and final process flux during the diafiltration step.

Required area:

$\text{Area} = [(\text{Permeate volume/Average flux})_{\text{Concentration}} + (\text{Permeate volume/Average flux})_{\text{Diafiltration}} + \ldots] \text{ / Time}$

In this example:

$\text{Area} = [(3{,}276 \text{ L}/103 \text{ LMH}) + (12{,}068 \text{ L}/83 \text{ LMH}) + (1{,}217 \text{ L}/41 \text{ LMH})] / 4 \text{ hours} = 51.6 \text{ m}^2$

Add 20% safety factor:

$\text{Area} = 62 \text{ m}^2$

To perform a scale-down process simulation, the same volume to area ratio is used and scaled based on either the feed volume that can be used for the simulation or the area of the desired filtration device. For example, if the process is to be performed on one Pellicon® 2 Mini cassette (with an area of 0.1 m^2), then the required feed volume will be:

$\text{Scale-down feed volume} = 0.1 \text{ m}^2 \times (5000 \text{ L}/62 \text{ m}^2) = 8 \text{ L}$

Instead, if there is a specific volume of feedstock to process (example: 25 L), then the required membrane area will be:

$\text{Scale-down membrane area} = 25 \text{ L} \times (62 \text{ m}^2/5000 \text{ L}) = 0.3 \text{ m}^2$

The process parameters, including Pellicon® device type, should be consistent between scales, allowing the process to be completed in a similar timeframe with similar fluxes, pressures and loadings. The concentration factors, number of diavolumes and feed quality should be kept consistent at all scales to ensure robust scalability. However, to demonstrate process robustness and repeatability, the process should be tested at pilot scale before proceeding to manufacturing.

Definitions

Apparent Sieving (S_{app})
The fraction of a particular protein that passes through the membrane to the permeate stream based on the measurable protein concentrations in the feed and permeate streams. A sieving coefficient can be calculated for each protein in a feedstock.

S_{app} [–] = (Concentration in permeate, C_P) / (Concentration in feed, C_b)

Concentration Factor (CF)
The amount that the product has been concentrated in the feed stream. This depends on both the volume concentration factor and the retention. As with the VCF, for a Fed-Batch concentration process, calculate CF based only on the volume of feedstock added to the TFF application.

CF [–] = Final product concentration / initial product concentration = VCF(R_{app})

Conversion Ratio (CR)
The fraction of the feed side flow that passes through the membrane to the permeate.

CR [–] = Q_P / Q_F

Diavolume (DV or N)
A measure of the extent of washing that has been performed during a diafiltration step. It is based on the volume of diafiltration buffer introduced into the unit operation compared to the retentate volume. If a constant-volume diafiltration is being performed, where the retentate volume is held constant and diafiltration buffer enters at the same rate that permeate leaves, a diavolume is calculated as:

DV or N [–] = Total buffer volume introduced to the operation during diafiltration/retentate volume

Intrinsic Sieving (S_i)
The fraction of a particular protein that passes through the membrane to the permeate stream. However, it is based on the protein concentration at the membrane surface. Although it cannot be directly measured, it provides a better understanding of the membrane's inherent separation characteristics.

S_i [–] = (Concentration in permeate, C_P) / (Concentration at membrane wall, C_w)

Mass Flux (J_m)
The mass flow of protein through the membrane normalized for the area of membrane (m^2) through which it is passing.

J_m [g m^{-2} h^{-1}] = Q_P x C_P / area

Permeate Flux (J_f)
The permeate flow rate normalized for the area of membrane (m^2) through which it is passing.

Pressure Drop (ΔP)
The difference in pressure along the feed channel of the membrane from inlet to outlet.

ΔP [bar] = $P_F - P_R$

Retention (R)
The fraction of a particular protein that is retained by the membrane. It can also be calculated as either apparent or intrinsic retention. Retention is often also called rejection.

R_{app} [–] = $1 - S_{app}$ or $R_i = 1 - S_i$

Transmembrane Pressure (TMP)
The average applied pressure from the feed to the permeate side of the membrane.

TMP [bar] = $[(P_F + P_R)/2] - P_P$

Volume Concentration Factor (VCF or X)
The amount that the feed stream has been reduced in volume from the initial volume. For instance, if 20 L of feedstock are processed by ultrafiltration until 18 L have passed through to the permeate and 2 L are left in the retentate, a ten-fold concentration has been performed so the Volume Concentration Factor is 10. In a Fed-Batch concentration process, where the bulk feedstock is held in an external tank and added to the TFF operation continuously as permeate is removed, VCF should be calculated based only on the volume that has been added to the TFF operation.

VCF or X [–] = Total starting feed volume added to the operation / current retentate volume

References/Footnotes

1. Total recycle means retentate and permeate lines return to feed vessel
2. If process flux is unstable, it may be necessary to allow additional time or investigate other membrane options
3. Retention samples are not required at every data point; sampling at lowest and highest TMP is typical
4. The point at which the flux levels off is defined as the point around which the slope of the flux vs. TMP curve decreases to ≤ 50% of the previous slope. This point is also referred to as the "knee" of the curve.
5. These other conditions are described in more detail in Parts 2 and 3.
6. *Example:* 10X concentration with a final volume of 300 mL requires (300 mL x 10) = 3 L of feed
7. Retention samples are not required at every data point; initial and final concentration are typical. Typical data recording interval is approximately every 10 – 15 minutes.
8. See Guide: *Maintenance Procedures for Pellicon® and Pellicon® 2 Cassette Filters (P17512)* or *Pellicon® 3 Filters Installation and User Guide (AN1065EN00)*
9. Ng P, Lundblad J, and Mitra G, *Optimization of Solute Separation by Diafiltration,* **Separation Science, 11**(5): 499-502, 1976.
10. See Technical Brief: *Protein Concentration and Diafiltration by Tangential Flow Filtration (TB032)*
11. See Guide: *Maintenance Procedures for Pellicon® and Pellicon® 2 Cassette Filters (P17512)* or *Pellicon® 3 Filters Installation and User Guide (AN1065EN00)*
12. See Technical Note: *Increase Product Yield in Your UF/DF Processes (AN1026EN00)*
13. Average flux can also be calculated for each step by dividing the total process volume by the total process time

To Place an Order or Receive Technical Assistance

In the U.S. and Canada, call toll-free 1-800-645-5476

For other countries across Europe and the world, please visit: **www.emdmillipore.com/offices**

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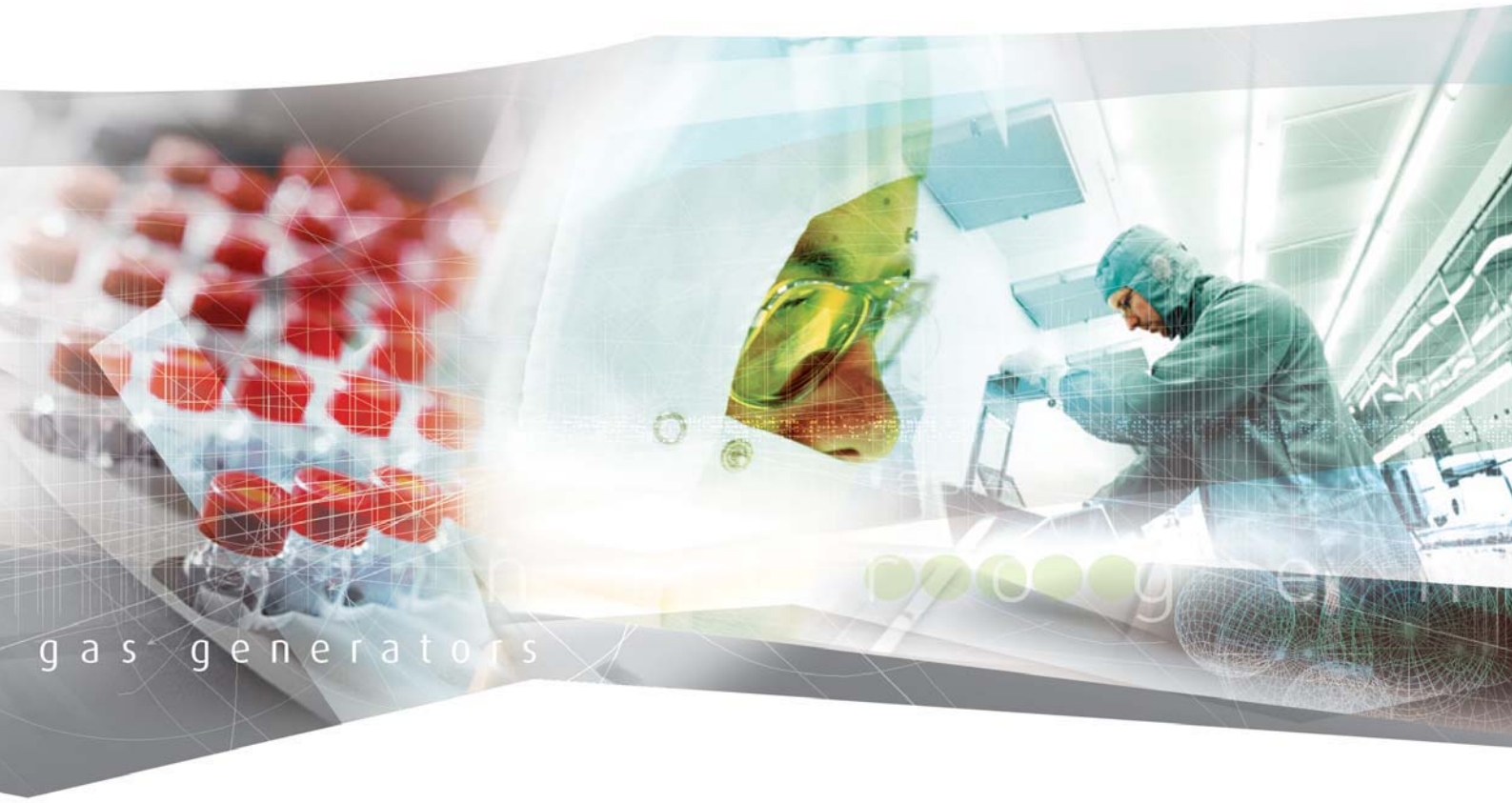
domnick hunter



Laboratory Gas Generators

LABGAS
Pure & Simple

Don't Buy Gas, Make It!

PAYBACK IN LESS THAN 1 YEAR

Generate laboratory gases for all of your analytical applications.

Hydrogen

Eliminate high pressure cylinders from the laboratory by generating a continuous source of UHP hydrogen gas.

- GC-FID, NPD, FPD, TCD, ELCD, HALL
- GC-carrier gas
- Total Hydrocarbon Analysers (THA)

Nitrogen

Eliminate high pressure cylinders by producing nitrogen gas from compressed air simply and cost effectively.

- LCMS (single and multiple units)
- ICP
- ELSD
- GC-FID, ECD, NPD, AED
- GC-carrier gas
- Solvent evaporation

Zero Air

Through the purification of a compressed air supply Zero Air Generators are ideal for use in FID applications.

- GC-FID, NPD, FPD
- THA

Clean Dry Air

Desiccant dryers to provide a constant flow of clean, dry compressed air.

- NMR, AA, GC, ATD, Rheometer, Sample Prep, Auto-Samplers and many other applications

CO_2 free air

Replace high pressure O_2 and N_2 cylinders with CO_2 moisture free compressed gas.

- TOC analyser
- FT-IR Purge
- Microscope Purge

Increased flexibility, Improved economy, Greater control

- **Performance**
  Ultra high purity gas generators will improve your analysis
- **Safety**
  Low pressure, minimal stored gas - no cylinder handling
- **Reliability**
  Never run out of gas
- **Compact design**
  Free up laboratory floor space
- **Cost effective**
  Quick paybacks, can be less than one year

An evolutionary development of domnick hunter gas generators provide real benefits in the laboratory:

• Compact design
- Can be used anywhere in the laboratory

• Aesthetic and ergonomic design
- Designed to integrate into any laboratory and complement scientific instrumentation

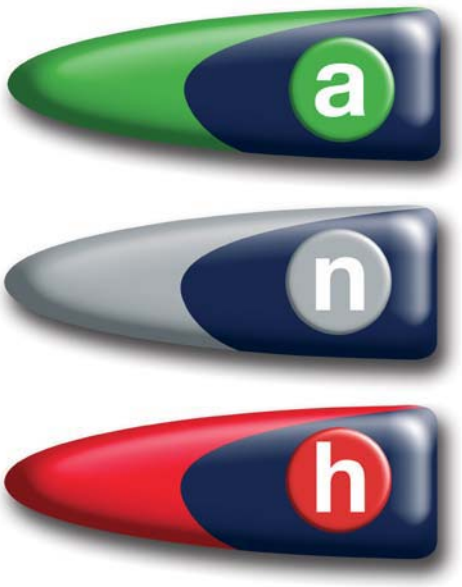
• Quiet operation
- No need to worry about disruption to your laboratory operations

• Minimal maintenance
- Maximum reliability with low cost of ownership
- domnick hunter gas generators require minimal attention and maintenance. Diagnostic capabilities include audible and visual indicators.

• Modular construction
- Common design
- Common spares
- Common operator interface
- Easy to operate and maintain

• Global product approvals
- We take care of the approvals process - leaving you free to concentrate on the key tasks in the laboratory

Diagnostic Indication
Common Operator Interface



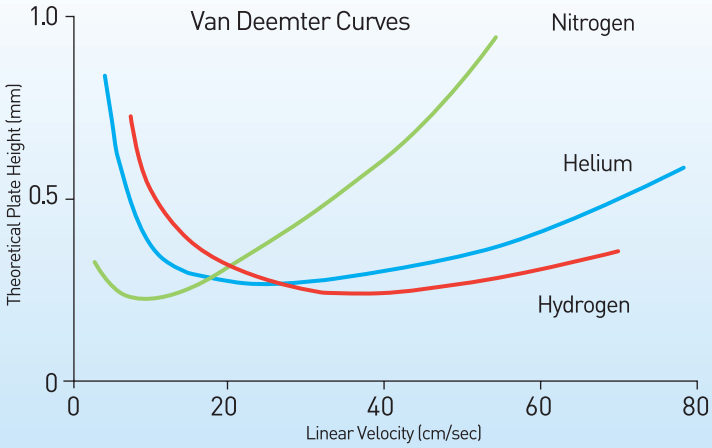
All domnick hunter gas generators carry a 2 year warranty which can be supported by IQ/OQ certification if requested.

Applications Guide

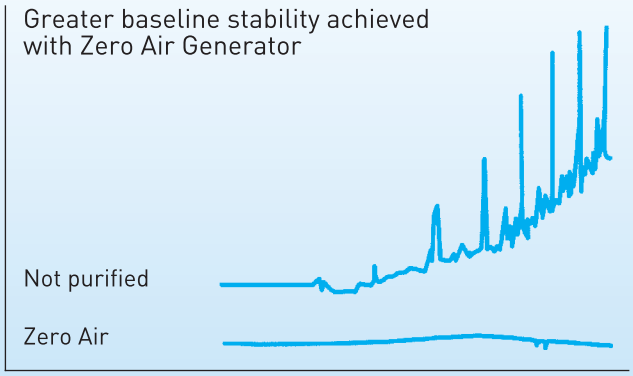
Instrument	Gas Requirement	Purity	Flow Rate	Generator Recommendation
Products for Gas Chromatography				
GC-FID	Hydrogen for fuel gas Zero air for flame gas Hydrogen for capillary carrier gas Nitrogen for packed carrier gas Nitrogen for make-up gas	UHP Hydrocarbon-free UHP UHP, zero grade UHP, zero grade	30-50 cc/min 300-500 cc/min up to 10 cc/min 20-50 cc/min 30-50 cc/min	**Hydrogen** **Zero Air** **Hydrogen** **Zero Nitrogen** **Zero Nitrogen**
GC-FPD	Hydrogen for fuel gas Zero Air for flame gas	UHP Hydrocarbon free	60-90 cc/min 90-120 cc/min	**Hydrogen** **Zero Air**
GC-NPD	Hydrogen for capillary gas Nitrogen for make up gas	UHP UHP, zero grade	up to 50 cc/min up to 30 cc/min	**Hydrogen** **Zero Nitrogen**
GC-ECD	Nitrogen for carrier gas Nitrogen for make up gas	UHP, zero grade UHP, zero grade	up to 60 cc/min up to 100 cc/min	**Zero Nitrogen** **Zero Nitrogen**
GC-TCD	Hydrogen as carrier gas	UHP	up to 50 cc/min	**Hydrogen**
GC-ATD	Dry air purge	Clean and dry air	less than 2L/min	**Clean Dry Air**
GC-AED	Nitrogen for carrier gas	UHP, zero grade	less than 1L/min	**Zero Nitrogen**
GC-ELCD,HALL	Hydrogen as reaction gas	UHP	70 to 200 cc/min	**Hydrogen**
Products for LCMS Instruments				
LCMS API/ LCMS APCI, Electrospray, LCMS/MS, TOF	Air for nebulizer gas Nitrogen for curtain and sheath shield gas	Clean and dry air, hydrocarbon free 99.5%	18L/min 5 to 35L/min	**Clean Dry Air** or **Zero Air** **Nitrogen**
Products for Spectroscopy				
FT-IR Spectrometer	Purge gas for sample compartment, optics, air bearing and microscope	Clean dry, CO_2 free	14 to 85L/min	**CO_2 free air**
NMR Spectrometer	Air for lifting spinning and ejecting	Clean and dry air	up to 300L/min	**Clean Dry Air**
ICP Spectrometer	Nitrogen or zero nitrogen for purge gas	99.99 + %	up to 9L/min	**Nitrogen** or **Zero Nitrogen**
AA Spectrometer	Air for oxidant gas	Clean and dry air	28 to 200L/min	**Clean Dry Air**
Products for Analyzers				
TOC	Dry air or Nitrogen for carrier gas or combustion gas	Clean dry, CO_2 free/ hydrocarbon free UHP	100-500 cc/min 50-700 cc/min	**CO_2 free air**/ **Zero Air** **Zero Nitrogen**
THA	Zero air for FID Hydrogen for fuel gas	Hydrocarbon free UHP	50 to 500 cc/min 5 to 50 cc/min	**Zero Air** **Hydrogen**
DSC	Air for air shield	Clean and dry	100cc/min	**Clean Dry Air**
TGA	Nitrogen or dry air as furnace gas	Clean and dry air or high purity N_2	100cc/min	**Clean Dry Air** or **Nitrogen**
TOD	Nitrogen carrier gas	UHP, Zero grade	300cc/min	**Zero Nitrogen**
CO_2 analyzer	Calibration air	Clean dry, CO_2 free hydrocarbon free	550 to 1000cc/min	**CO_2 free air**
Other laboratory applications				
Sample Prep Autosamplers	Nitrogen for solvent evaporation Air for pneumatic controls Nitrogen for sample injector	95% to 99% Clean and dry UHP, zero grade	up to 130L/min 28L/min 550cc/min	**Nitrogen** **Clean Dry Air** **Zero Nitrogen**
Circular Dichroism	Nitrogen	UHP, zero grade		**Zero Nitrogen**
ELSD Detector	Nitrogen	98%	2-8 L/min	**Nitrogen** or
Particle sizing by Laser Diffraction	Clean and dry air for nebulizing gas	Clean and dry air		**Clean Dry Air**

domnick hunter gas generators improve your analysis

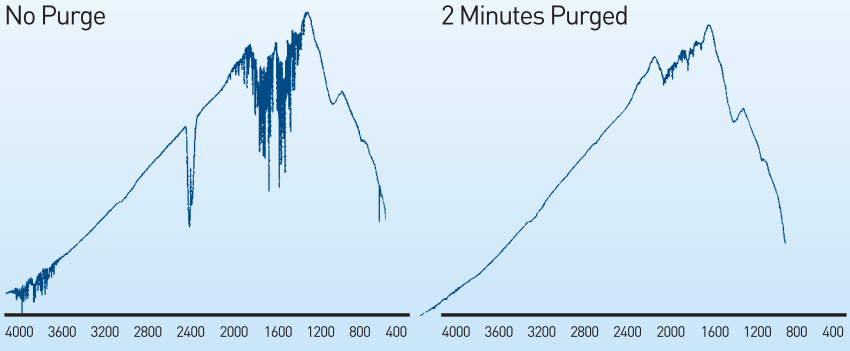
H_2 generator



Zero Air



CO_2 removal



Hydrogen Generators

Eliminate high pressure hydrogen cylinders from the laboratory by generating a continuous source of UHP hydrogen gas for applications such as:

- GC-FID, NPD, FPD, TCD, ELCD, HALL
- GC-carrier gas
- THA

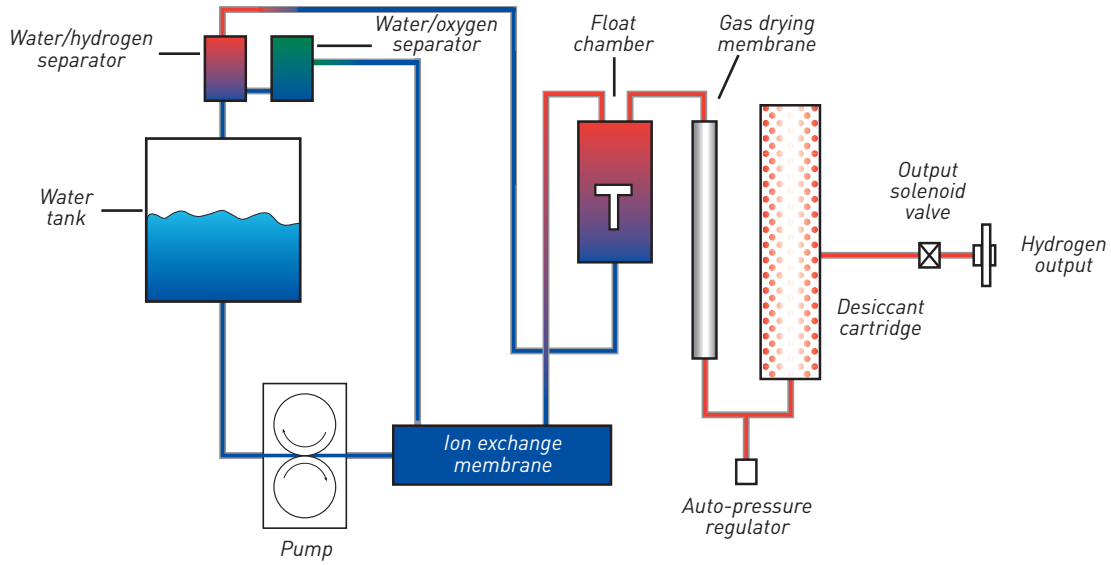
Benefits

- **Continuous supply of GC quality hydrogen gas on demand**
- **Ultra compact design**
- **Improved productivity and chromatography results -** Hydrogen is a faster carrier gas and more sensitive when compared to helium, reducing analysis times by 25 to 35% without significant loss of resolution
- **Extended column life -** Hydrogen, when used as a carrier gas, requires lower elution temperatures and thus improves the column service life
- **Safety -** Hydrogen production at low pressure and only when required, eliminates the risks of explosion. Eliminates problems relating to handling gas cylinders.
- **Improved laboratory safety -** Through automatic leak and low water detection, remote start/stop/alarm and elimination of long gas lines
- **Economy -** No gas cylinder rental, no price inflation
- **Increased efficiency in the laboratory -** 24 hour operation; no interruption of analysis due to cylinder changes
- **No caustic solutions required**
- **Recommended by major instrument manufacturers**

Technical Features

- Self test fault diagnosis with digital display and audible alarm: detection of internal and external H_2 leaks, H_2 overpressure, water level, water conductivity, display of H_2 product flow and total flow
- Simplified use and attention by easy access to maintenance components (desiccant cartridge and de-ionisation cartridge)
- Improved independent operation due to its 5 litre water tank
- Automatic water filling (optional feature)
- Generator protected against the harsh lab environment by means of domnick hunter patented filters (avoids rapid degradation of the generator water quality and thus increases service life)

How the generator works



domnick hunter hydrogen generators use a special ion exchange membrane to produce a flow of ultra-pure hydrogen. Use of the electrolytic dissociation process enables water to be broken down into hydrogen and oxygen.

The oxygen is released into the air, while the hydrogen is retained to form the product flow.

A long-life desiccant cartridge purifies the hydrogen even further so that it attains the desired grade of purity and ensures constant reproducible results.

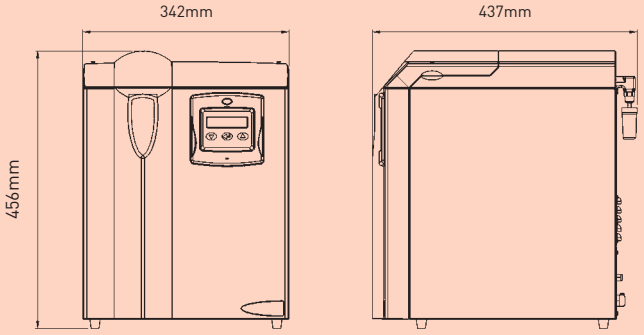
Having proved its worth in thousands of systems throughout the world, this technology eliminates the need to use liquid electrolytes, such as caustic solutions; since it only uses de-ionised water and electricity, continuous operation is assured.

Technical Specifications

Model	20H	40H	60H
H_2 Flow rate cc/min	160	250	500
H_2 Purity *	>99.99999%		
H_2 Pressure (electronically adjustable)	0-7 bar (0-100 psi)		
De-ionised Water Quality	>1 Megohm		
Water Capacity	5 Litres		
Supply Voltage	90 - 264 Volts (47 - 63 Hz)		
Electrical Consumption	100W	170W	240W
Weight (Empty)	24Kg (53 lbs)		
Drying Type	Desiccant		
Ambient Operating Temperature	+5 - 40°C (41 - 104°F)		
Outlet Connector	⅛" Compression (Swagelok)		

* When used with palladium purifier

Dimensions



Nitrogen Generators

(Including Zero N_2 and dry air)

Produce nitrogen gas from compressed air simply and cost effectively. Replace nitrogen gas cylinders for the following applications:

- LCMS (single and multiple units)
- ICP
- ELSD
- GC-carrier gas
- GC-FID, NPD, ECD, AED
- Solvent evaporation

Benefits

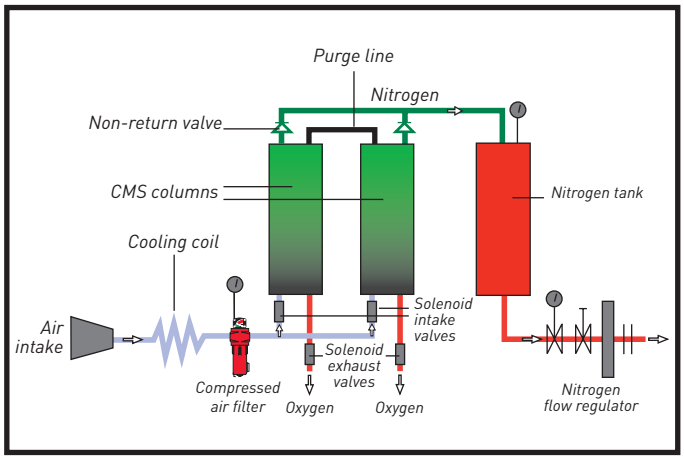
- **Designed to meet specific analytical instrument gas purity and flow requirements**
- **Improved analytical performance -** Production of a constant flow of gas improves the consistency of the analysis results and hence reproducibility
- **Increased laboratory efficiency with a constant, guaranteed supply**
- **Improved safety -** No handling high-pressure gas cylinders or liquid dewars. Nitrogen production at controlled low pressures
- **Simple installation -** Only one set up operation required for reliable service over time. Installation on or below a laboratory bench top, saving space in the laboratory
- **Economy -** Quick return on investment - No gas cylinder rental bottles, no price inflation
- **Recommended by major instrument manufacturers**
- **Combined N_2 and Dry Air Generators -** In addition to the stand alone nitrogen, zero N_2 and dry air gas generators, domnick hunter also manufactures models that provide nitrogen and dry air from a single unit (models G6 and G7).

Technical Features

Principle: passage of compressed air through a bed of carbon molecular sieve (CMS) using presure swing adsorption technology, the most reliable on the market

- N_2 flow: 550 cc/min to 30L/min (for larger capacities please consult domnick hunter)
- O_2 purity: 3% to 10 ppm
- On-line purity monitoring capability
- Digital hours counter
  Audible and visual maintenance indicator
- Economy mode option: Enables the compressor to switch off when nitrogen supply is not required
- Oil-free air compressor available: The quietest models available
- Available with or without built-in air compressor
- Quick and easy servicing: less than 10 minutes every 6 months
- A 25L/min version is also available

How the generator works



The technology used to produce a continuous flow of high purity N_2 is pressure swing adsorption (PSA).

This technology uses a combination of molecular sieves to selectively eliminate O_2 and other contaminants in the ambient air.

The CMS column(s) alternate between the purification and regeneration modes to ensure continuous N_2 production.

The gas generator is designed to take pre-filtered compressed air at 7 or 8.5 barg (102 or 123 psi g) (depending on model) from the existing laboratory supply or via the integrated oil-free compressor.

This flow of filtered compressed air then passes through the CMS column which is in the purification mode. At this point, the O_2, CO_2, humidity and hydrocarbons are eliminated from the compressed air, producing a flow of clean and dry high purity nitrogen.

For zero N_2 generators, a heated catalyst oxidises additional hydrocarbons from the N_2 gas flow providing zero grade N_2 with a remaining hydrocarbon content of <0.1ppm

Weights

Model	without compressor (Kg)	with compressor (Kg)
G1	52	56
G2 / G4	77	90
G3	71	83
G5	51	55
G6	54	58
G7	80	93
G8 / G9	50	54

Nitrogen Generator Selection Chart

Model		Gas Flow L/min (up to)	Outlet Pressure bar g	Purity ppm	Purity %	
G1	0	550 cc/min	5	10	-	Nitrogen N_2
	1	750 cc/min		10	-	
G2	0	1.5	5	10	-	
	1	3		10	-	
G3	0	2.5	5	100	-	
	1	4		-	0.1	
	2	5		-	0.5	
	3	7		-	1	
	4	8		-	2	
G4	0	5	5	100	-	
	1	6		-	0.1	
	2	10		-	0.5	
	3	12.5		-	1	
	4	14		-	2	
	5	12	7	-	0.5	
	6	18*		-	0.5	
	7	20*		-	1	
	8	25*		-	1	
G5	0	1	5	10	-	Zero N_2
G6	0	N_2 : 600 cc/min Air : 1.5	5	10 -55°C adp	-	N_2 & Dry Air
G7	0	N_2 : 3 Air : 3	5	10 -55°C adp	-	N_2 & Dry Air
G8	0	3	5	-55°C adp		Dry Air
G9	0	6	5	-55°C adp		Dry Air

0	Without compressor
1	With compressor

0	Without Economy Mode
1	With Economy Mode**

E	230 V / 50 HZ
W	110 V / 60 HZ

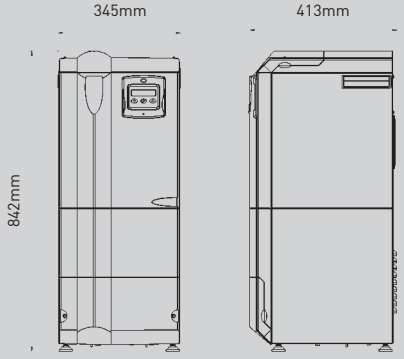
G4	3	0	1	E	Selected Product Code

Please Note:

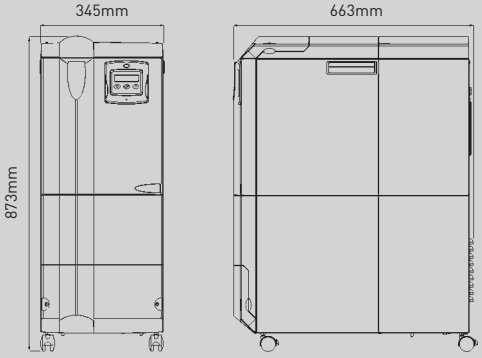
1 All 5 bar (72.5 psi g) delivery generators without compressor require 7 bar (102 psi) air inlet
2 All 7 bar delivery generators without compressor require 8.5 bar (123 psi) air inlet
3 *This unit is only available without compressor
4 **Economy mode only available on models G1-G4

Dimensions

Models G1 / G5 / G6 / G8 / G9



Models G2 - G4 / G7



Zero Air Generators

By simply connecting to a clean, dry compressed air supply, a Zero Air Generator will remove hydrocarbons, making it ideal for use in FID applications:

- GC-FID, NPD, FPD
- THA

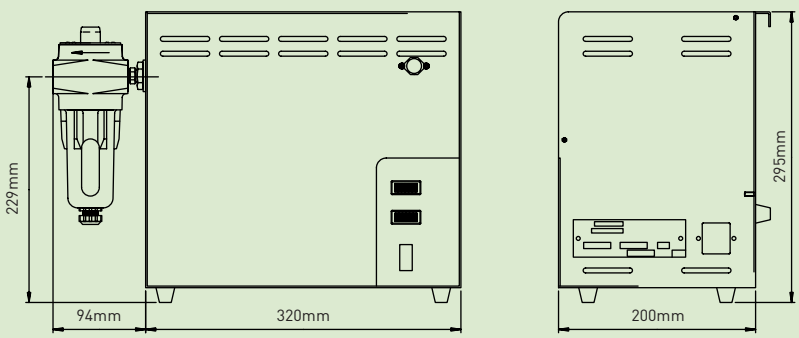
Benefits

- **Improved analytical performance -** The reduction of methane (CH_4) to less than 0.1 ppm reduces background noise and improves the base line stability. This enables a lower detection limit and hence increases the sensitivity of your analysis
- **Simple installation -** From any source of clean, dry compressed air, this generator will provide gas free from any trace of hydrocarbons.
- **Improved Safety -** Elimination of high pressure gas cylinders. Only needs compressed air and a standard electrical socket
- **Consistent gas purity improves instrument performance**
- **Economy -** Quick return on investment typically 1 year. No gas cylinder rental, no price inflation
- **Increased efficiency in the laboratory -** 24 hour operation; no interruption of analyses due to gas cylinder replacement. Reduced re-calibration of equipment
- **Recommended by major instrument manufacturers**

Technical Features

- Elimination of the main contaminants in your compressed air, (hydrocarbons including CH_4) via a heated platinised catalyst
- Models:
  UHP-10ZA Zero Air Purifier 1L/min
  UHP-35ZA Zero Air Purifier 3.5L/min
  Larger flowrates (up to 50L/min) on request
- G¼ air connections
- Purity < 0.1 ppm (CH_4)
- Quick and easy servicing : changing the filters once a year takes just a few minutes

Dimensions



Clean dry air

domnick hunter desiccant dryers are ideal for laboratory use, providing a constant flow of clean, dry compressed air for:

- NMR, AA, GC, ATD, Rheometer, Sample Prep, Auto Samplers and many other applications

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- Point of use installation provides dry and clean air where you need it
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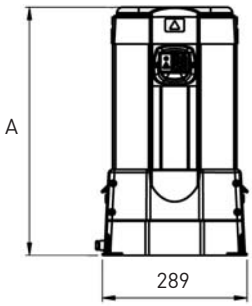
- Compact and lightweight
- Can be bench or wall mounted
- Quick and easy servicing. Maintenance alarm activates every 12000 hours of operation
- Larger flowrate models available



Model	Output Air Flow L/min (cfm)		Dimension A mm (ins)		Weight Kg (lbs)	
DAS 1	70	(2.5)	422	(16.6)	11	(24.2)
DAS 2	115	(4.1)	500	(19.7)	13	(28.7)
DAS 3	182	(6.4)	616	(24.2)	16	(35.3)
DAS 4	227	(8.0)	692	(27.2)	18	(39.7)
DAS 5	295	(10.4)	847	(33.3)	20	(44.1)
DAS 6	340	(12.0)	906	(35.7)	23	(50.7)
DAS 7	453	(16.0)	1098	(43.2)	28	(61.7)

Max. Working pressure: 12 bar g (175 psi g)
Dewpoint: -40°C (-40°F)
Purity: Non-methane HC's <0.01 ppm
particles <0.1 micron
oil <0.01mg/m^3

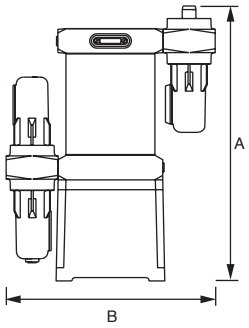
CO_2 free air

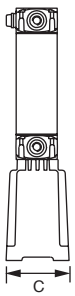
Replaces high pressure oxygen or nitrogen gas cylinders with CO_2 and moisture free compressed gas for:

- TOC Analyser
- FT-IR Purge
- Microscope purge

Benefits

- Reduced signal to noise ratio improves instrument performance
- Protects sensitive optics and air bearings from moisture
- Constant, guaranteed purity supply increases laboratory efficiency
- Tested and approved by most TOC and FT-IR instrument manufacturers
- Compact design frees up floor space





Model	Output Air Flow L/min	Dimensions mm (ins)						Weight Kg (lbs)	
		A		B		C			
C02RP015	1.5	380	(15)	310	(12)	90	(4)	8	(18)
C02RP140	14	470	(18)	310	(12)	90	(4)	10	(22)
C02RP280	28	710	(28)	310	(12)	90	(4)	12	(27)
C02RP850	85	1020	(40)	420	(17)	150	(6)	36	(80)

Max. Working pressure: 10.5 bar g (152 psi g)
CO_2 Content: <1ppm
Pressure Dewpoint @ 7bar g (100 psi g): -70°C (-100°F)
Purity : Non methane HC's <0.003 ppm
particles <0.1 micron

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PyroPure Multiple-Effect Stills

The Mueller PyroPure P6000 Series is Built to Last

Mueller PyroPure multiple-effect stills (MES) are the simplest, most reliable means of producing pyrogen-free water-for-injection (WFI) that meets all U.S. Pharmacopoeia requirements. The MES is designed with efficiency in mind. Because the system recovers the latent heat of vaporization occurring within its own process to heat feedwater and uses feedwater as its primary source of cooling, the MES is an energy and money-saving model of ingenuity. Due to the absence of moving parts, the PyroPure MES requires less maintenance and is much quieter than mechanical compression stills. Multiple-effect stills also lack the seal and associated oil supply required by mechanical compression stills; therefore, there is no danger of contamination due to seal breakdown associated with mechanical compression. The PyroPure MES is manufactured according to FDA current Good Manufacturing Practices (cGMPs) and ASME-BPE requirements.

Each PyroPure MES is designed to minimize operating costs associated with production of WFI by minimizing the required utility steam and coolant consumption. This is accomplished by utilizing sources of energy within the various process streams to preheat the feedwater and thus use the feedwater as a cooling source. Using the feedwater as a coolant source also reduces the utility steam consumed to elevate the temperature of the feedwater. The feedwater ultimately enters the tubes of the first effect evaporator where utility steam is applied to the shell to evaporate the feedwater. The resulting steam produced is then directed to the separation column where a tangential inlet produces centrifugal force that separates the entrained water droplets away from the pure steam. This pure steam is then used as the heating source for the subsequent effect.

Simple Design, Reliable Operation

- External evaporators access for inspection and preventative maintenance on critical o-rings and gaskets.
- The separation columns contain no internal components that require inspection or periodic maintenance.
- All maintenance, including replacement of critical components, can be performed with only 24" of space on all sides (including the top) of the equipment.
- ASME-BPE certified fittings are used throughout.
- WFI condensers have removable tube bundles for easy cleaning and inspection of product contact surfaces.
- Minimal instrumentation is required upon operation of the equipment. Only two control loops are needed which minimizes the calibration required as well as the potential for downtime.
- All elastomers in contact with feedwater and product are provided with USP Class VI certifications.
- All components are fully drainable including the optional feedwater pump.

As the pure steam is condensed in the shell side of the subsequent evaporator, the resulting WFI flows through feedwater preheating devices and to the WFI condenser for subcooling to the required product temperature. Only pure steam discharged from the last effect of the still is condensed in the product condenser. The final product as well as the feedwater supplied to the still is measured for conductivity to ensure compliance with specifications.

Control of the multiple-effect still is accomplished by two control loops. The first control loop monitors the first effect temperature and manipulates the plant steam control valve as needed to maintain the specified temperature. The second control loop monitors the product temperature and manipulates a coolant control valve to maintain the specified product temperature. Level switches in the separation columns provide control for the feedwater supply and provide alarm capabilities to ensure that all effects are operating correctly. The control and operational simplicity results in a design that requires no rotating equipment, flow measurement devices or pressure transmitters.

Models are available with 3 to 6 effects to provide the best solution for your application. Additional effects will result in further reduced utility consumption while a minimum of effects will provide the lowest capital cost solution and occupy the smallest footprint. All product contact surfaces are polished to 20 Ra maximum and electropolished. Surfaces in contact with feedwater are polished to 25 Ra maximum. All surfaces in contact with feedwater and product are manufactured from 316/316L stainless steel.

System Components

Condenser. PyroPure condensers have a double-tube-sheet design that provides users with the efficiency of heat exchange and at the same time ensures that pure vapor and distillate will never come into contact with feedwater and coolant. To facilitate maintenance, all PyroPure condensers are mounted at an angle to allow full drainage of the pure distillate through the distillate outlet port installed at the lowest point of the vessel. The condenser is designed to allow the removal of the U-tube bundle, making it easy for the user to inspect the critical pure distillate contact surfaces.

Controls. The standard control system is an Allen Bradley PLC with an Allen Bradley operator interface mounted in a NEMA rated panel. Ethernet is provided on the standard control system to facilitate communications with adjacent equipment or data archiving systems. Mueller can also provide other Allen Bradley control components, as well as control systems from Siemens and Mitsubishi. Control and electrical panels are supplied with a UL 508a label.

Steam Separator. Mixture of water and vapor leaves the evaporator at high velocity and enters the separator through a tangential port, a natural vortex is formed. The centrifugal force of the vortex separates water droplets and contaminants out of the spiraling vapor. Pure vapor rises up through the steam separator and out of the port at the top of the separator. The steam separator has no baffles or demister, there are no auxiliary surfaces for condensation to collect and stagnate. Concerns over the potential for bacterial growth are eliminated.

Preheaters. Each still is equipped with a preheater for each effect to provide for maximum energy recovery and efficiency. As the water flows under pressure from each effect to the next the pressure of the water is reduced which will result in “flashing” of the water into steam. The preheater recovers this energy into the feedwater to reduce the overall plant steam consumption.

Evaporator. The natural circulation design of the PyroPure evaporator ensures maximum surface wetting, eliminating the hot, dry areas that lead to the stress-cracking associated with other designs. The tube bundle creates a large heat transfer surface which vaporizes feedwater on contact. The PyroPure multiple-effect still has fully drainable external evaporators, eliminating the need for the excess headroom required for evaporator removal with other designs. The evaporator on the first effect of the multiple-effect still is double tube-sheet to prevent cross-contamination. All other effects have single-tubesheet evaporators.

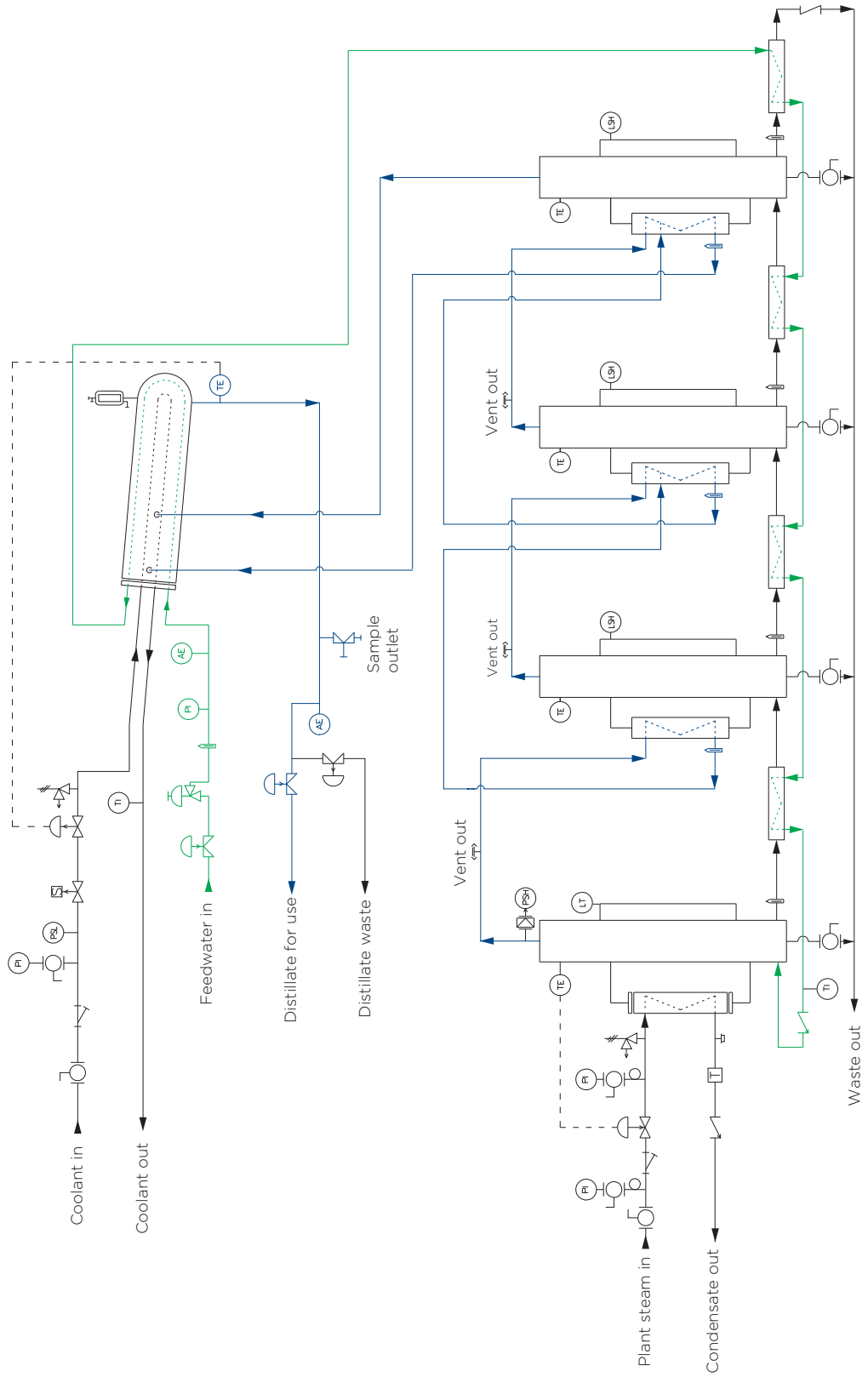
Options

Feedwater Pump System. The feedwater pump system enhances feedwater pressure and is required if feedwater supply pressure is not equivalent to the plant steam pressure. When purchased, the feedwater pump system will be installed on the MES framework.

Pure Steam Option. Multiple-effect stills can be configured to produce pure steam from the first effect. Simultaneous WFI and pure steam production is also available.

The Mueller PyroPure P6000 Series is Built to Last

Schematic of Operation



Specifications

Plant Steam (psig): 110 • Distillate (°F): 190 • Feedwater (°F): 75 • Coolant Inlet (°F): 60

	Capacity[1]		Supply Steam[2]		Coolant Supply[3]		Approximate Dimensions		Distillate Outlet Ht		Est. Weight	
Model	gph	lph	lb/hr	kg/hr	gph	lph	HxWxD (in)	HxWxD (cm)	in	cm	lb	kg
MES 6009-3	90	341	345	156	214	810	113x62x40	287x157x102	90	229	2,500	1,135
MES 6015-3	165	625	625	283	424	1,604	114x62x40	290x157x102	89	226	3,050	1,385
MES 6015-4	150	568	467	212	267	1,011	114x75x40	290x191x102	89	226	3,200	1,453
MES 6015-5	140	530	382	173	185	702	114x88x40	290x224x102	89	226	3,350	1,521
MES 6032-4	305	1,154	958	435	569	2,155	112x80x45	284x203x114	85	216	3,600	1,635
MES 6032-5	275	1,041	753	342	382	1,447	112x94x45	284x239x114	85	216	4,100	1,861
MES 6032-6	240	908	593	269	256	969	112x108x45	284x274x114	85	216	4,600	2,088
MES 6040-5	400	1,514	1,060	481	544	2,059	112x94x45	284x239x114	85	216	4,400	1,998
MES 6040-6	390	1,476	922	418	408	1,544	112x108x45	284x274x114	85	216	4,800	2,542
MES 6064-4	630	2,385	1,926	874	1,169	4,425	128x100x52	325x254x132	102	259	4,900	2,225
MES 6064-5	600	2,271	1,554	705	811	3,070	128x119x52	325x302x132	102	259	5,600	2,543
MES 6064-6	500	1,893	1,198	543	542	2,052	128x138x52	325x351x132	102	259	6,300	2,860
MES 6076-5	690	2,612	1,811	822	945	3,578	131x119x52	333x302x132	102	259	6,800	3,087
MES 6076-6	660	2,498	1,557	706	706	2,671	131x138x52	333x351x132	102	259	7,500	3,405
MES 6110-5	1,100	4,163	2,393	1,085	1,896	7,176	136x147x53	345x373x135	107	272	12,100	5,494
MES 6110-6	1,070	4,050	2,055	932	1,558	5,897	136x168x53	345x423x135	107	272	15,000	6,810
MES 6140-6	1,300	4,921	2,467	1,119	1,877	7,105	151x184x58	384x467x147	117	297	16,400	7,446
MES 6175-6	1,810	6,852	3,423	1,553	2,642	10,001	180x184x58	457x467x147	145	368	16,800	7,620
MES 6200-6	3,200	12,112	6,126	2,779	4,797	18,157	155x209x64	394x531x163	113	287	34,500	15,649
MES 6300-6	3,700	14,004	7,060	3,202	5,547	20,994	188x192x64	478x488x163	87	221	56,500	25,628

[1] Distillate 170°F (77°C) to 190°F (88°C) (customer determined). Gravity flow.

[2] Plant steam 110 to 125 psig (7.6 to 8.6 bar) dry and saturated (capacity based on 110 psig).

[3] Coolant water at 32°F to 100°F (0°C to 38°C) at 40 psig (2.8 bar) (flow rates based upon a distillate outlet temperature of 190°F [88°C] and cooling water inlet temperature of 60°F [16°C] and cooling water outlet temperature of 160°F [71°C]).

Additional requirements:

- *Feedwater: Feedwater supply 10 percent over distillate capacity. If feedwater pressure is less than plant steam pressure, a feedwater booster pump may be required. (Max. of 1 ppm silica or total hardness. No chlorine, chlorides, or amines.)*
- *Electrical Service (Standard): Without pump: 115 VAC, single phase, 60/50 Hz; with pump 460 VAC, 3 phase, 60Hz.*

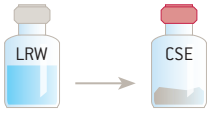
Pharma&Biotech

Traditional Kinetic Limulus Amebocyte Lysate (LAL) Assay Procedure Quick Guide

This is a step by step guide depicting how to perform a traditional kinetic LAL assay. Prior to initiating the assay procedure, allow reagent vials to equilibrate to room temperature. The incubating microplate reader should also be turned on and a plate template created in the WinKQCL™ Software.

Step 1

Reconstitute Control Standard Endotoxin (CSE) with LAL Reagent Water (LRW) to yield a solution containing 50 EU/ml or 100 EU/ml depending on assay method being used.



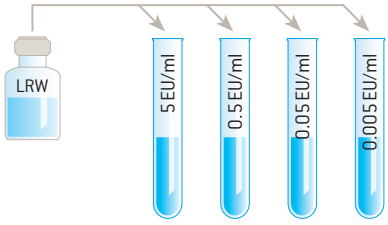
Step 2

Vortex for 15 minutes.

Step 3

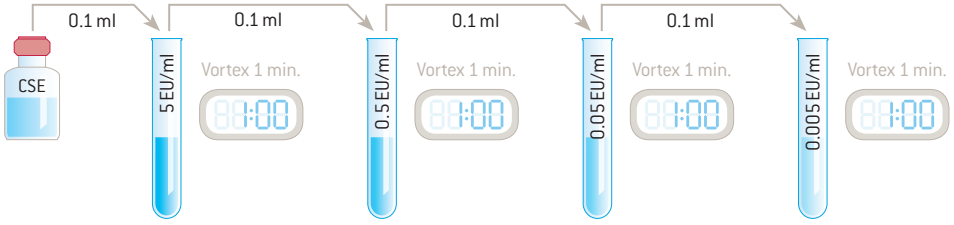
Label the tubes with the appropriate endotoxin concentration and add 0.9 ml of LRW to each.

(Example based on a test with an operating standard curve of 0.005–50 EU/ml.)



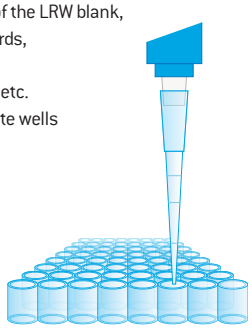
Step 4

Prepare a series of endotoxin standards.



Step 5

Dispense 100 µl of the LRW blank, endotoxin standards, product samples, positive controls, etc. into the appropriate wells of the microplate.



Step 6

Pre-incubate the plate for ≥10 minutes at 37°C ± 1°C in the microplate reader.

Step 7

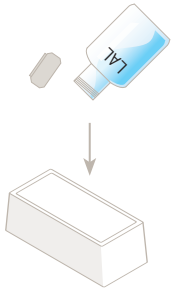
Immediately prior to use, reconstitute LAL and gently swirl.

kinetic chromogenic

kinetic turbidimetric

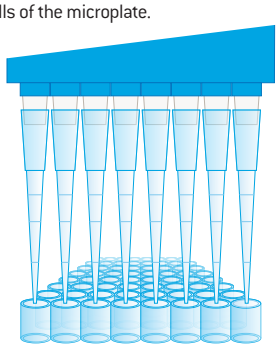
Step 8

Pour LAL into a reagent reservoir and mix gently.



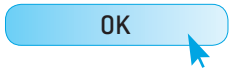
Step 9

Use an eight channel pipettor to dispense 100 µl of LAL into the appropriate wells of the microplate.



Step 10

Initiate the test by clicking the **OK** button in the WinKQCL™ Software.



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Materials, Equipment & Documents Needed

Reagents

- Limulus Amebocyte Lysate (LAL) Reagent (Kinetic-QCL™ or PYROGENT™-5000 Reagent)
- Control Standard Endotoxin (CSE)
- LAL Reconstitution Buffer (Required for the PYROGENT™-5000 Kinetic Turbidimetric LAL Assay)
- LAL Reagent Water (LRW) (# W50-640, W50-100, W50-500)

Kits are available in a wide range of sizes and configurations.
Please contact your local sales representative for additional information.

Accessories

- Glass dilution tubes (# N207)
- Individually wrapped serological pipettes (optional)
- Tips
- 96-well plates (# 25-340)
- Reagent reservoirs (# 00190035)

Use pyrogen-free accessories that have been qualified for endotoxin testing.

Equipment and Software

- Eight channel pipettor
- Incubating absorbance microplate reader
- WinKQCL™ Software
- Pipettors
- Timer
- Vortex mixer

Supporting Documents

- Certificate of Analysis (CoA), www.lonza.com/coa
- Limulus Amebocyte Lysate (LAL) Kinetic-QCL™ Package Insert or
  Limulus Amebocyte Lysate (LAL) PYROGENT™-5000 Package Insert

Points to Consider

- Use matched LAL and CSE reagents
- Plastic tubes are not recommended for making endotoxin dilutions
- Follow all suggested endotoxin vortexing times
- Use pyrogen-free accessories that have been qualified for endotoxin testing
- Equilibrate reagents to room temperature before use
- Do not vortex the LAL
- Avoid bubbles when plating reagents into the 96-well plate
- Avoid contaminating samples, standards, LRW and accessories
- Equipment should be installed, validated and maintained appropriately

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 TS-RT-DS013 10/13

TECHNICAL BRIEF

Steaming-in-place and integrity testing of a sterilizing-grade filter assembly

Millipore Steam Sterilization & Integrity Testing Procedures

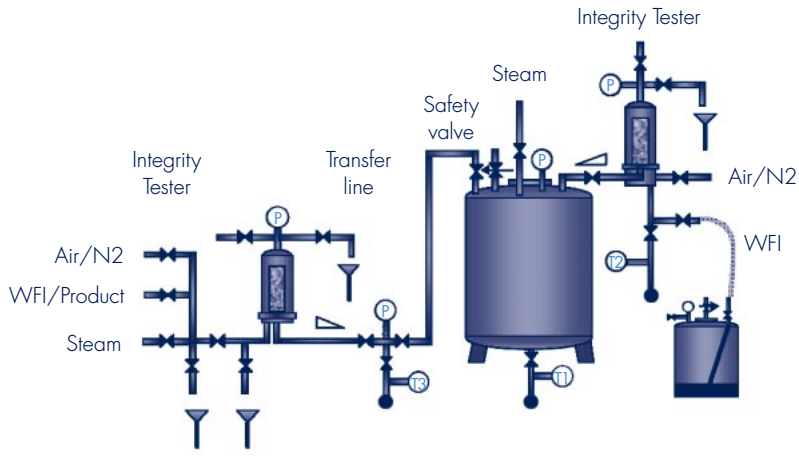
The intent of this technical brief is to provide a Standard Operating Procedure (SOP) for the steam sterilization and integrity testing of a filter assembly typically used in aseptic processing (see figure 1). Because steam sterilization is the most common source of damaging a filter, it is recommended to test the integrity of sterilizing-grade filters after sterilization, before the filtration process[1,2,3] Therefore, the SOP includes both the Steaming-In-Place (SIP) and the post-SIP, pre-use integrity testing of the vent and product filters.

The operations described in this SOP should be performed in the following sequence:

1. SIP of the hydrophobic filter assembly, like Millipore's Aervent® CTGR 5 inch or 10 inch or LAGR 4 inch devices, used for the venting of a sterile holding tank (vent filter).
2. Post-SIP, pre-use integrity testing of the vent filter.
3. SIP of the hydrophilic sterilizing-grade filter assembly, like Millipore's Durapore® CVGL 10 inch, LAGL 4 inch or MCGL devices, used for the sterile filtration of the product (product filter).
4. Post-SIP, pre-use integrity testing of the product filter.

The recommended post-SIP, pre-use filter integrity test for hydrophobic vent

Sterile Filter Assembly: Product Filter, Sterile Tank and Vent Filter



MILLIPORE

filters is the HydroCorr™ test. Unlike classical testing procedures like diffusion and bubble point for these filters, which require alcohol/water mixtures as a wetting agent, the HydroCorr™ test requires only clean water. As this test is performed at the upstream side of the vent filter and no downstream manipulation is required, it is perfectly suited to assess the integrity of the filter and to demonstrate that the SIP procedure has been adequately conducted, without damaging the vent filter.

The recommended post-SIP, pre-use filter integrity test for hydrophilic product filters is the Enhanced Bubble Point test.

Integrity testing involves wetting of the filter with the standard wetting medium and is a possible source of breaking sterility of the sterilized system. From the product filter to the vent filter on the sterile tank is a sealed, closed and sterile system. Water used to wet the filter before integrity testing cannot be drained downstream of the closed filter system, and will remain in the sterile product tank. Using the product to be filtered as the wetting agent facilitates pre-use integrity testing and avoids the downstream evacuation of the testing medium since the product can be directly routed to the holding tank.

Product bubble point test is perfectly suited to assess the integrity of the filter as well as to demonstrate that the SIP procedure has been adequately conducted, without damaging the product filter.

Filter Characteristics

Millipore's Aervent filters are sterilizing-grade vent filters that are constructed with a PTFE membrane. These filters have been qualified to withstand at least 40 SIP cycles at 135 °C for 30 minutes in the forward direction, as well as 40 cycles in the reverse direction (see the Validation Guide of the respective filters).

The maximum differential pressure allowed during SIP in the forward direction is 350 mbar. For reverse SIP, the recommended maximum ditterential pressure is 100 mbar throughout the entire SIP cycle.

Millipore's Durapore® filters are sterilizing grade filters that are constructed with a PVDF membrane. These filters have been qualified to withstand 5 to 30 SIP cycles at 135 °C for 30 minutes in the forward direction (see the Validation Guide of the respective filters). The maximum differential pressure allowed during SIP in the forward direction is 350 mbar.

Integrity Testing Parameters

The minimum bubble point specification for hydrophilic sterilizing-grade (0.22 µm) Durapore filters is 3450 mbar, after wetting with pure water for 5 min. at 200 mbar differential pressure.

The minimum product bubble point is determined by the bubble point ratio laboratory scale study. The bubble point ratio (BPR) approach is a proven method used for determining minimum bubble point values tor non-specified wetting fluids (see Millipore's Application Note No. AN1505EN00).

The recommended pressure for filling the housing prior to the HydroCorr test is 1 barg. Table 2 shows the HydroCorr integrity test criteria for Aervent type of filters.

Specifications

SIP Parameters

Minimum required steam supply pressure:	>1.2 barg
Minimum required compressed air supply:	> 1.5 barg (300 mbar above steam pressure)
Minimum temperature in the coldest points:	> 121.1 °C
Maximum reverse differential pressure across the filter during SIP:	< 100 mbar
Maximum forward differential pressure across the filter during cooling:	< 350 mbar
Minimum sterilization time:	30 min. at 121.1 °C
Cooling time:	30 min. (approximately)

Filter type	Wetting volume (L)
Millidisk® 20	5
Millidisk 40	10
Optiseal® 4 inch	10
Durapore 5 inch	18
Durapore 10 inch	35

Table 1: *Recommended volume for wetting Durapore filters prior to Bubble Point testing.*

Type of Aervent Filter	Catalogue number	Specification
4 inch Optiseal	LAGR04TP6	< 0.20 mL/min. @ 2620 mbar
5 inch Cartridge	CTGR75S01	< 0.38 mL/min. @ 2620 mbar
10 inch Cartridge	CTGR_1TP1	< 0.75 mL/min. @ 2620 mbar
20 inch Cartridge	CTGR_2TP1	< 1.50 mL/min. @ 2620 mbar
30 inch Cartridge	CTGR_3TP1	< 2.25 mL/min. @ 2620 mbar

Table 2: *HydroCorr integrity test criteria for Aervent filters.*

Steam-In-Place Procedure for a Sterile Tank Equipped With a Vent Filter

The manual operations that are described in this SOP should be performed respecting the given sequence. For automatic SIP procedures refer to Millipore's Technical Brief No. TB011EN00.

Prior to commencing the procedure, the following is assumed:

- Tank and adjacent piping is clean (e.g. by means of CIP) and empty;
- Filter housing is installed and the correct vent filter is put in place;
- All connections are checked for proper fitting;
- System has been checked for leak-tightness by means of a pressure hold test;
- All valves are closed and silicone tubing are attached to bleed valves and directed to a condensate drain;
- Use caution to avoid contact with steam or hot stainless steel surfaces;
- Wear protective glasses at all times and heat resistant protective gloves when necessary.

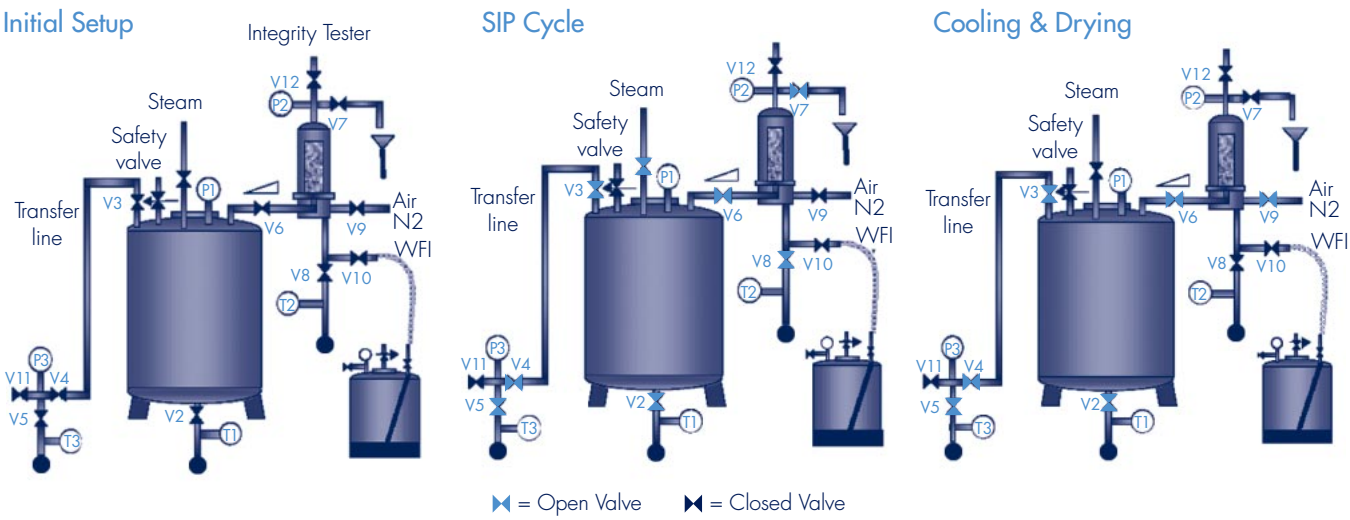
Standard Operating Procedure

1. Check that the steam supply and compressed gas pressures are set up at the required values.
2. Respectively open V2, V3, V4, V5, and V1 to introduce steam to the system and to purge air from the system.
3. Partially close bleed valves V2 and V5 to build tank pressure to at least 0.5 bar and wait for the temperature gauges T1 and T3 to indicate more than 100 °C.
4. Then, slowly open V6 to introduce steam to the vent filter. Crack open bleed valves V7 and V8 to establish a steady flow of steam and allow for condensate drainage and air removal from the filter housing. Note: It is of utmost importance to control the difference between pressure gauges P1 and P2 and keep the delta-P over the filter to a maximum of 100 mbar. For reverse direction SIP, use an Optiseal filter or a code 7* filter. Do not use a code 0** filter.
5. Ensure all air and condensate are effectively removed by keeping V2, V5, V7 and V8 cracked open so that a 15 cm wisp of steam and a continuous drip of water can be seen exiting.
6. When the temperature throughout the system reaches over 121.1 °C, as measured by the temperature gauges T1, T2 and T3, the timer is started. Sterilization time should be at least 30 minutes or longer if validation has decreed so. *During the sterilization phase both pressure and temperature should be recorded regularly.*
7. When the required sterilization time has been achieved, close the steam supply valve V1 and slowly open V9 to introduce sterile compressed gas into the system.

   CAUTION: Make sure that the system remains under positive pressure (as indicated by pressure gauges P1, P2 and P3) and control that the delta-P over the filter does not exceed 350 mbar.
8. Allow for steam purge from all bleed valves and close valves V7 and V8 to increase the flow of sterile gas through the system. Maintain the gas flow to cool down the system until the temperature gauges T1, T2 and T3 indicate approximately 40 °C.
9. Respectively close valves V5, V2 and V4, and keep V6 and V9 open to maintain a positive pressure into the sterile system while it is not in use.

* *2–226 O-ring locking outlet with spear assembly* ** *2–222 O-ring outlet*

Recommended SIP Process for a Sterile Tank Equipped with a Vent Filter



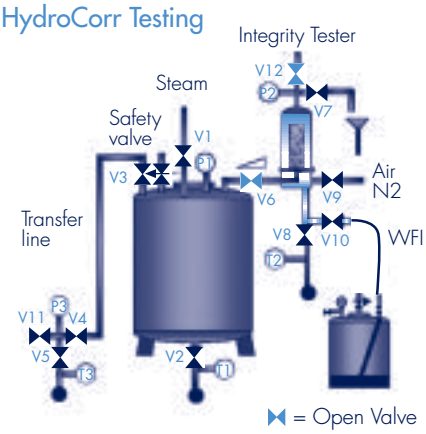
Post SIP, Pre-Use Vent Filter Integrity Test Procedure

1. Close the compressed gas valve V9, keep V6 open, and open V7 to vent the system. Wait for the pressure as measured by P1 and P2 to drop to atmospheric pressure.
2. Fill the pressure vessel with clean pure water and attach the inlet of the vessel to a compressed gas supply at 1 bar. Attach the outlet tubing of the pressure vessel to V10.
3. Slowly open V10 to have water entering the filter housing. Ensure that the filling pressure does not exceed 1 bar and that air cannot enter the housing (e.g. empty pressure vessel). Continue filling until water is seen exiting the hose attached to V7.

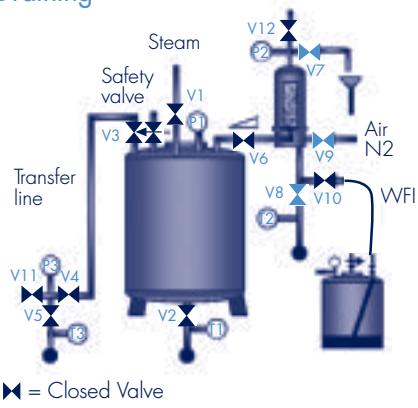
   Note: Should the filter housing be installed on top of a rather tall tank, it may prove useful to increase the pressure to adjust for gravity influence while the filling operation commences.
4. Close V10 and bleed air from the pressure vessel by slowly opening the pressure relief valve on top of the vessel until atmospheric conditions are reached.
5. Close V7, open V12 and attach an automatic filter integrity tester to V12.
6. Double check that V6 is open and that V7, V8, V9 and V10 are all fully closed and run the HydroCorr test.
7. When the test is finished and a positive result (i.e. pass) is obtained, close V6 and V12 and detach the filter integrity tester.
8. Open V7 and V8 to drain water from the housing. The draining can be facilitated by carefully opening V9 and applying pressure until the system is empty.
9. Fully open V9 to allow for drying of the filter over a period of 30 min.
10. Close V7 and V8, open V6 and keep V9 open to build and maintain a positive pressure into the system while it is not in use.

Recommended Post SIP Pre-use Filter Integrity Test for Hydrophobic Vent Filters

HydroCorr Testing



Draining



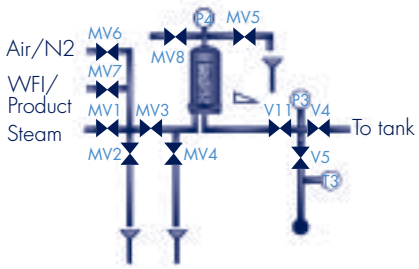
Steam-In-Place Procedure for Product Filter

The manual operations that are described in this SOP should be performed respecting the given sequence. For automatic SIP procedures refer to Millipore's Technical Brief No. TB011EN00.

Prior to commencing the procedure, the following is assumed:

- Filter housing is installed and the correct product filter is put in place;
- Product filter is dry;
- All valves are closed and silicone tubing are attached to bleed valves and directed to a condensate drain;
- Use caution to avoid contact with steam or hot stainless steel surfaces;
- Wear protective glasses at all times and heat resistant protective gloves when necessary.

Recommended SIP for the Product Filter



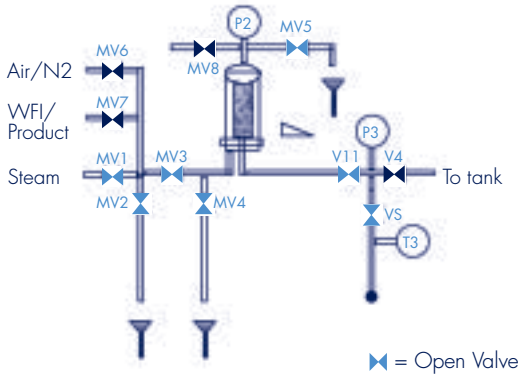
Standard Operating Procedure

1. Check that the steam supply and compressed gas pressures are set up at the required values.
2. Open MV1 and MV2 and purge the steam line until complete absence of condensate.
3. Fully open MV4 and MV5 to allow for subsequent air and condensate evacuation.
4. Slowly open MV3 to progressively introduce steam and heat up the filter.
5. Partially close bleed valves MV2, MV4 and MV5 so that a wisp of steam and a continuous drip of water can be seen exiting.
6. Respectively open V11 and crack open bleed valve V5 to establish a steady flow of steam and allow for condensate drainage and air removal from the filter housing.

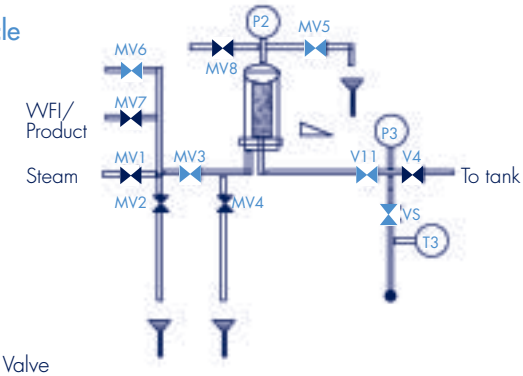
   Note: It is of utmost importance to control the difference between pressure gauges P3 and P4 and keep the delta-P over the filter to a maximum of 350 mbar.
7. Ensure all air and condensate are effectively removed by keeping MV2, MV4, MV5, and V5 cracked open so that a 15 cm wisp of steam and a continuous drip of water can be seen exiting.

Recommended SIP and Cooling Process for Hydrophilic Product Filter

SIP Cycle

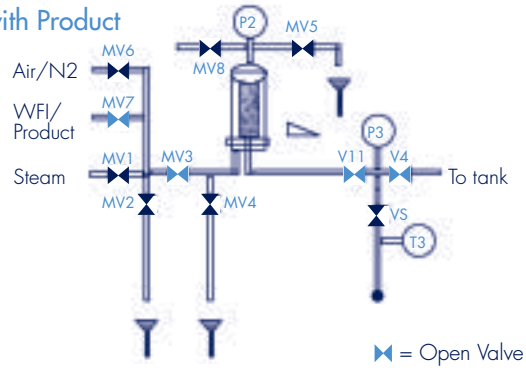


Cooling Cycle

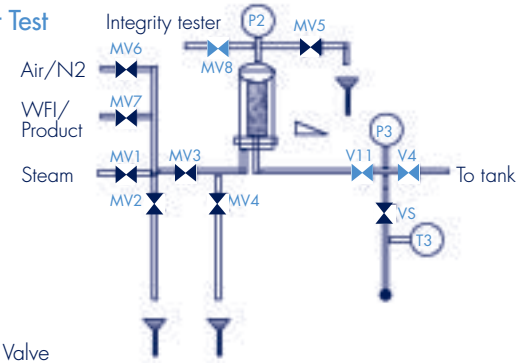


Recommended Post SIP Pre-use Filter Wetting and Integrity Test for Hydrophilic Product Filter

Wetting with Product



Bubble Point Test



8. When the temperature downstream of the product filter, as measured by the temperature gauge T3, reaches over 121.1 °C, the timer is started. Sterilization time should be at least 30 minutes or longer, as established during validation. During the sterilization phase both pressure and temperature should be recorded regularly.
9. At completion of the sterilization cycle, close the steam supply valve MV1 and slowly open MV6 to introduce compressed gas into the system.

   CAUTION: Make sure that the system remains under positive pressure (as indicated by pressure gauges P3 and P4) and control that the delta-P over the filter does not exceed 350 mbar.
10. Allow for steam purge from all bleed valves and close valves MV2 and MV4 to increase the flow of gas through the system. Maintain the gas flow to cool down the system until the temperature gauge T3 indicates approximately 30 °C.
11. Respectively close valves V5, V11 and MV5, and keep MV6 and MV3 open to maintain a positive pressure into the sterile filter system while it is not in use.

Post SIP, Pre-Use Filter Integrity Test Procedure

1. Ensure that V3, V6 and V7 on the vent filter are open and that the sterile tank downstream of the product filter is vented at the atmospheric pressure.
2. Maintain MV3 open, close the compressed gas supply valve MV6, and open MV5 to vent the system. Wait for the pressure as measured by P4 to drop to atmospheric pressure.
3. If possible, set the inlet product pressure at 2.8 bar. Gradually open MV7 to fill the filter housing with product and vent the filter housing from MV5, until all upstream air has been released.
4. When product is seen exiting the hose attached to MV5, close the vent valve MV5 and continue to maintain the 2.8 bar pressure for at least one minute to dissolve any residual gas within the filter and ensure membrane wetting.

5. Fully open the downstream valve V11 and gradually open V4 to set the differential pressure (P4–P3) at approximately 200 mbar.
6. Continue to flow product through the filter to the sterile tank at appropriate pressure differential for at least five minutes.
7. Then close MV7 and MV3 to isolate the filter and fully open V4.
8. Open MV8 and attach an automatic filter integrity tester.
9. Double-check that MV8, V11, V4, V3, V6 and V7 are open and that MV3 and MV4 are all fully closed and run the enhanced bubble point test.
10. When the test is finished and a positive result (i.e. pass) is obtained close MV8 and detach the filter integrity tester.
11. Open MV7, MV3 and MV5 to re-start the filtration of the product.
12. When product is seen through MV5, close MV5 and continue the filtration of the product.

References

1. Revision of annex 1 to *EC Guide to GMP for Sterile Medicinal Products*; 1997. "The integrity of the sterilised filter should be verified before use and should be confirmed immediately after use by an appropriate method such as a bubble point, diffusive flow or pressure hold test."
2. *FDA Guideline on Sterile Drug Products Produced by Aseptic Processing*; 1987. "Normally, integrity testing of the filter is performed after the filter unit is assembled and sterilized prior to use. More importantly, however, such testing should be conducted after the filter is used in order to detect any filter leaks or perforations that may have occurred during filtration. Forward flow, bubble point and pressure hold tests are acceptable integrity tests."
3. PDA Technical Report No. 26, *Sterilizing Filtration of Liquids*; 1998. "Integrity testing of sterilizing grade production filters pre and post use is a fundamental element of sterility."

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Thermo Scientific TSU700 Series -86ºC Freezers

Ultimate protection, optimum capacities, energy savings

* Internal performance data, data on file
** Comparison to high-performance mode

Built specifically to protect the most critical of samples, Thermo Scientific™ TSU Series -86ºC freezers achieve outstanding thermal performance, safety and security through state-of-the-art engineering.

Performance:
Outstanding BTU (British Thermal Unit) reserve, leading to fast door-opening recovery times so sample integrity is not compromised.*

Capacity:
Store up to 700 2-inch boxes in a 12.85 sq. ft. (1.19 m^2) footprint. That's 70,000 2mL vials or 118,300 1mL tubes!

Energy-Efficiency:
Save up to 15% in energy usage with our energy-savings mode.** Or choose our high-performance mode for applications requiring ultra-tight temperature uniformity. Both modes are accessible through our easy-to-use touch-screen interface.

Features and Benefits:

- Touch-screen interface featuring freezer health monitoring system, event log, advanced settings and USB port
- On-board data storage – store up to 15 years worth of temperature and event data on our on-board computer
- Polystyrene insulated inner doors help maintain cabinet temperature during openings and feature embedded rare earth magnets, eliminating the need for exposed latches or magnets
- Optional proximity card access control for enhanced security
- 4x7 heated outer door gaskets provide four touchpoints of security and seven zones of protection, maximizing cabinet temperature and minimizing frost build-up
- Rugged steel construction with a corrosion-resistant coating
- Pressure equalization port (PEP) allows for quick re-entries after door opening

Thermo SCIENTIFIC

Thermo Scientific TSU700 Series -86ºC Freezers (Temperature Range: -50ºC to -86ºC)

Model No.	Cabinet Capacity	Sample Capacity	Voltage (Hz)	Amps/Breaker (Plug)	Maximum Shelf Weight	Interior Dimensions H x D x W	Exterior Dimensions H x D x W	Shipping Weight
TSU700D	33.5 cu. ft. (949 liters)	700 boxes (2-inch)	208-230V/60Hz	12/15 (NEMA 6-15)	285 lbs. (128.4 kg)	51.2 x 28.3 x 40 in. (130 x 71.9 x 101.6 cm)	78 x 37.6 x 49.2* (198.1 x 95.5 x 125 cm)	951 lbs. (432 kg)
TSU700V			230V/50Hz	12/15 (European)				

Racking Systems for Boxes

Model No.	Description	Dimensions H x W x D	Boxes/ Rack	Racks/ Shelf	Racks/ Freezer	Boxes/ Freezer
920090	Sliding drawer for 2-inch boxes	11.9 x 5.5 x 26.9 in. (30.2 x 14 x 68.3 cm)	25	7	28	700
1950520	Adjustable side access for 2-inch boxes	11.6 x 5.4 x 26.75 in. (29.5 x 13.7 x 67.9 cm)	25	7	28	700
920091	Sliding drawer for 3-inch boxes	11.9 x 5.5 x 26.9 in. (30.2 x 14 x 68.3 cm)	15	7	28	420
1950521	Adjustable side access for 3-inch boxes	11.6 x 5.4 x 26.75 in. (29.5 x 13.7 x 67.9 cm)	15	7	28	420

Racking Systems for Microplates

Model No.	Description	Dimensions H x W x D	Plates/ Rack	Racks/ Shelf	Racks/ Freezer	Plates/ Freezer
1950642	Sliding drawer for standard or deepwell	11.9 x 5.5 x 26.9 in. (30.2 x 14 x 68.3 cm)	35	7	28	980
1950523	Side access for standard plates	11.9 x 5.5 x 25.7 in. (30.2 x 14 x 65.3 cm)	105	7	28	2940
1950592	Side access with locking rod for standard or deepwell plates	11.9 x 5.5 x 25.7 in. (30.2 x 14 x 65.3 cm)	147	7	28	4116

Options (Field-Installed Requires Qualified Professional)

Model No.	Description
LN4567	Factory-installed LN2 back-up
FLN4567	Field-installed LN2 back-up
CO4567	Factory-installed CO2 back-up
FCO4567	Field-installed CO2 back-up
CR4567	Factory-installed inkless chart recorder
FCR567FT	Field-installed inkless chart recorder
CRP4567	Factory-installed ink chart recorder
FCRP567FT	Field-installed ink chart recorder
RAC34567	Factory-installed access key option
FFAC34567	Field-installed access key option
WC34567	Factory-installed water-cooled condenser
SS34567	Factory-installed stainless steel interior

Accessories

Model No.	Description
ACU34567	Access key pack U.S. (ISO15693 protocol)
ACE34567	Access key pack EU (ISO14443 protocol)
RSK700SD4	Racking shelf kit (7 racks, 175 boxes)
SK700	Shelf kit (one shelf and clips)
17020	Chart paper ink (pack of 50)
AF34567	Replacement air filter
400159	Replacement back-up battery
6903	Alarm delay module
4425	Cryo gloves (medium)
4426	Cryo gloves (large)
TF-ULT700	Seismic restraint kit

Speciality Plugs (Factory-Installed)

Model No.	Description
AR230V16A	Argentina
AU230V16A	Australia
BR230V16A	Brazil
CH230V16A	China
DK230V16A	Denmark
UK230V16A	Great Britain
IN230V16A	India
IS230V16A	Israel
IT230V16A	Italy
SW230V16A	Switzerland

* Door opening clearance is 34.5" (86 cm).

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Pall Corporation

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Resolute® Chromatography Columns

Description

Resolute columns are ideally suited for process purification in the biopharmaceutical industry. A patented nozzle valve provides all column functions required for packing, unpacking and running of the column in a closed system. Based on established and well proven designs, the Resolute column provides improved column performance and true linear scalability combined with reproducible column packing methods for today's high performance media.

Pall continues to work closely with the biopharmaceutical manufacturing industry to advance the state of the art for both chromatographic performance and productivity. The packing process is controlled by a slurry packing system which,

combined with a Resolute column, offers a complete solution for process chromatography from development to manufacturing scale.

Features and Benefits

- Standard column diameters from 280 mm to 1200 mm – all with selectable bed heights from 100 mm up to 600 mm
- Bed Height: fixed or adjustable (200 mm adjustment)
- Nozzle Valve: choice of manually operated Resolute DM, or pneumatically actuated Resolute DP columns
- Choice of bed supports in polyethylene or stainless steel
- Piston seal and precision bore tube eliminates need for additional mechanical compression or pneumatic activation of adjuster seals
- Compatible with a wide range of Chromatographic Media

Sanitary Design

- Fully flushed flow path and adjuster seal for clean-in-place (CIP)
- Minimum dead space fixed cell seal arrangement
- Reduced risk of corrosion
  - non metallic nozzle flow path for high salt and low pH conditions
  - forged stainless steel tube eliminates weld seams on tube wall and flanges
- Visible valve flow path aids detection of entrapped air
- Sanitary clamp terminations
- Leachate free acrylic tube — no phthalates
- Peroxide cured seals — no sulphur containing leachate
- Process wetted materials meet regulatory requirements

Column Options

Standard Columns

- Actuated nozzle valve; fits 400 to 1200 mm diameter columns
- Bed supports:

- Polyethylene 10/20/60 µm
- Stainless steel 10/20/50 µm

- Lockable castors and adjustable feet supplied as standard*
- Up to 1000 mm diameter only.

Engineered Columns

- Operating pressure up to 7 bar
- Columns from 400 mm to 2000 mm diameter
- Certification to ASME Div VIII sec 1 or PD5500.CE where applicable
- End-cell Rotation Frames (ERF)
- Alternative materials: Hastalloy C22, Stainless Steel 1.4435, 1.4439
- Hydraulic Actuators — see technical specifications for operating modes

View additional chromatography columns.

Specifications

Material Specifications

Process Wetted Components

Column Tube	Acrylic (cast PMMA) or Stainless Steel 316L (1.4404)
Distribution Cell	Polypropylene
Nozzle Body	PVDF/Acrylic(1)
Process Terminations and Slurry Nozzle Tip	PEEK
Slurry Inlet Port	180 and 280 mm columns: (2) PEEK 400 to 1200 mm columns: (2) Stainless Steel 316L
Bed Support	Polyethylene sinter or Stainless Steel mesh
Seals	EPDM (Peroxide cured) FEP encapsulated silicone
Wiper Blade	PTFE

External Components

Stand, Adjustable Feet	Stainless Steel 316L
Castor	Stainless Steel 304L
Castor Tyre	Polyurethane

(1) PVDF/PVDF version available for increased chemical resistance.
(2) Components not in mobile phase flow path.

Column Specifications

Resolute Fixed and Adjustable Columns 180 – 1200 mm (7 – 47 in.) diameter

Operating Pressure	180 – 280 mm (7 – 11 in.): 5 bar (73) psi 400 – 1200 mm (15.7 – 47 in.): 3 bar (44) psi
Operating Temperature	2 – 30 ºC (35 – 86 °F)
Bed Support, Type and Rating	Stainless Steel Mesh: 10, 20, 50 µm Polyethylene Sinter: 10, 20, 60 µm
Product Flow Path	Stainless Steel surface finish < 0.6 µm Ra, Electropolished
Exterior Components	Stainless Steel surface finish < 0.9 µm Ra, Electropolished
Pressure Retaining Plates	Stainless Steel surface finish < 1.5 µm Ra, 240 (UK) Grit Sateen
Column Frame	Stainless Steel surface finish Bright polished
Media Transfer Nozzle	DM:

	Manual actuation DP: Manual control with pneumatic actuation

Hydraulic Actuators (Optional)

Type	Mode of Operation	Application
HEP Hydraulic Endcell Positioning	Hydraulic linear actuator when column is empty or primed and has an open flow path with < 1 bar column pressure	Set up prior to Pack in Place and Maintenance
DAP Dynamic Axial Packing	Hydraulic linear actuator when column is empty or filled with slurry and has an open flow path with < 3.0 bar column pressure	Flow Packing at optimal linear velocity
/M Option Hoist-free Maintenance	Actuators and integral support and safety devices move and secure Top Adjuster and Column Tube (/M option available for either HEP or DAP model)	Access to top and bottom bed supports and seals for changeout

Ordering Information

This is a list of typical part numbers for this product range. For part numbers and configurations that are that are not listed, please contact your Pall representative.

Column Capacity and Ordering Information

Description	Diameter	CSA	Adjustable Height	Adjustable Capacity	Operating Pressure
Resolute 280	280 mm (11.in.)	620 cm^2	100 – 300 mm	6.2 – 18.5 L	5 bar (73 psi)
Resolute 400	400 mm (15.7 in.)	1260 cm^2	100 – 300 mm	12.6 – 37.7 L	3 bar (44 psi)
Resolute 600	600 mm (23.6 in.)	2830 cm^2	100 – 300 mm	28.3 – 84.8 L	3 bar (44 psi)
Resolute 800	800 mm (2.6 ft)	5030 cm^2	100 – 300 mm	50.3 – 150 L	3 bar (44 psi)
Resolute 1000	1000 mm (3.2 ft)	7850 cm^2	100 – 300 mm	79 – 235 L	3 bar (44 psi)
Resolute 1200	1200 mm (3.9 ft)	11310 cm^2	100 – 300 mm	113 – 339 L	3 bar (44 psi)
Resolute 1400	1400 mm (4.6 ft)	15390 cm^2	100 – 300 mm	154 – 461 L	3 bar (44 psi)
Resolute 1600	1600 mm (5.2 ft)	20110 cm^2	100 – 300 mm	201 – 603 L	3 bar (44 psi)
Resolute 1800	1800 mm (5.9 ft)	25450 cm^2	100 – 300 mm	255 – 763 L	3 bar (44 psi)
Resolute 2000	2000 mm (6.6 ft)	31420 cm^2	100 – 300 mm	314 – 942 L	3 bar (44 psi)

Contact Information

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Fax: ++43 1 49192 400
Email: Pall-Austria-Office@europe.pall.com

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- *Leading-edge void-free membranes to match virtually any separation challenge*
- *Short flow path for higher flux and higher resolution separation capability*
- *Choice of flow channel configuration providing process optimization capability*
- *Predictable, fast, scale-up*
- *True linear scalability from laboratory size modules to industrial assemblies for processing thousands of liters*

DATA SHEET

Pellicon® 2 Filters and Holders

High-performance tangential flow filters for biopharmaceutical process development, scale-up/scale-down and concentration/ purification/cell harvesting applications

Typical Applications

Concentration, desalting or buffer exchange of:

- Protein solutions
- Polysaccharide solutions
- Virus suspensions

Harvest, washing or clarification of:

- Cell cultures and lysates
- Colloidal suspensions
- Viral cultures

Superior TFF Performance

For research, process development, scale-up and production, Pellicon 2 filters and holders offer the following benefits:

Consistent High Flux and High Product Recovery

Millipore's Biomax® polyethersulfone and Ultracel® PLC-composite regenerated cellulose membranes have void-free structures that guard against leakage of solutes through microdefects normally associated with voids beneath the thin skins of conventional UF membranes (Figures 1 and 2).

These void-free membranes are more permeable, resulting in high-flux with equivalent or superior product retention (Figure 3). These void-free membranes provide the advantages of fast, high yield processing and smaller systems.

The long established Durapore® hydrophilic PVDF microfiltration membrane is well known for its exceptional combination of high flux, low protein binding and high product recoveries.

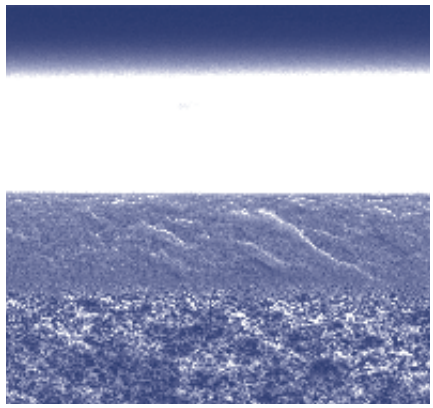


Figure 1. *Void-free Biomax 10 modified polyethersulfone membrane*

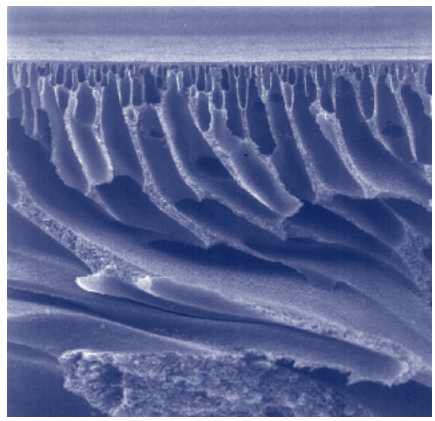


Figure 2. *Conventional 10 kD polyethersulfone membrane with sub-surface voids*

Easy, Reliable Linear Scale-Up from the Lab to the Production Plant

Pellicon 2 Mini filters scale-up easily and reliably from the laboratory to the production plant (Figures 4 and 5). By ensuring every flow channel has the same length, height and turbulence promoter as well as flow direction and materials of construction, we maintain the same ultrafilter/microfilter performance at all scales. Thus, rapid and reliable translation of processes from lab to manufacturing scale is easily achieved.

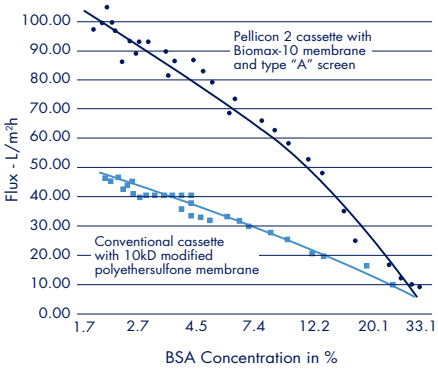
Linear Scale-Up

Mini filters (0.1 m^2/1.1 ft^2) and holders are designed for laboratory ultrafiltration/microfiltration of 100 mL to 10 L volumes, yet scale up linearly to Pellicon 2 Cassette (0.5 m^2/5.4 ft^2) and Maxi (2.5 m^2/26.9 ft^2) filters used in the pilot or manufacturing plant to process volumes from one liter to thousands of liters.

Thus, whether you operate 0.1 m^2 or 100 m^2 of installed area, every Pellicon 2 filter operates with the same pressure drop, flow velocity and concentration profile for true, rapid and simple linear scale-up.

Pellicon 2 Filters Proof of Performance

Improved Flux



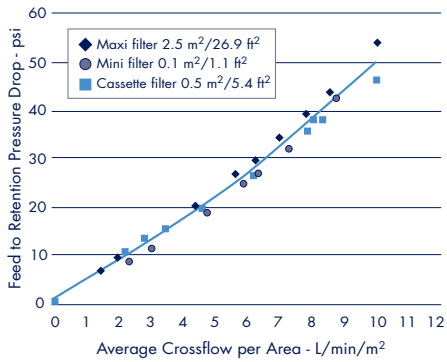
Feed pressure: 5.6 bar/80 psi
Retentate pressure: 2.1 bar/30 psi
Temperature: 10 – 13.5 °C
Initial volume 28 L
Final volume: 2 L

Conclusion

Pellicon 2 filters with Biomax membranes provide up to two-times the process flux of conventional cassettes resulting in faster processing and smaller systems.

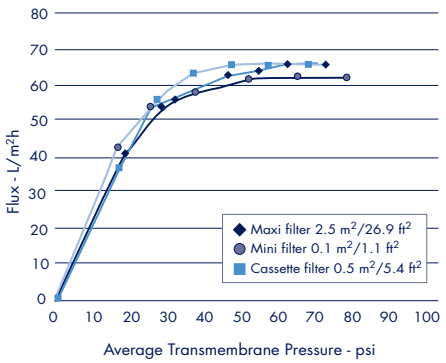
Figure 3. *Flux versus BSA concentration*

Linear Scalability



Temperature: 8 °C

Figure 4. *Feed to retentate pressure drop versus average crossflow on a 10% BSA solution*



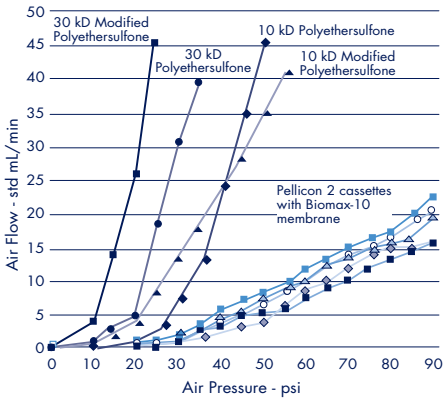
Temperature: 8 °C
Feed to retentate pressure drop: 2.8 bar/40 psi

Conclusion

(Figures 4 and 5) Pellicon 2 family of cassette filters scale linearly from 0.1 to 0.5 to 2.5 m^2 (1.1 to 5.4 to 26.9 ft^2) sizes for rapid, accurate and safe process scale-up and transfer.

Figure 5. *Flux versus average transmembrane pressure on a 10% BSA solution.*

Improved Reliability



Conclusion

The void-free structure of Biomax membranes is demonstrated by low, linear air diffusion values. This performance ensures better process reliability and safety and better product retention for higher yields.

Figure 6. *Integrity test comparison-air flow through wetted cassettes*

Greater Process Reliability and Reproducibility

The combination of defect-free membranes with Millipore's highly reliable manufacturing processes, offers greater consistency of process parameters.

The high quality of Millipore's ultrafiltration membranes is further ensured by our pioneering multiple-solute mixed-dextran retention profile test. Unlike the single solute protein retention test, Millipore's retention profile test measures and ensures reproducible retention performance of our UF membranes over the entire range of molecular weights retained by the membrane, not ¡ust at one or two molecular weights.

Low Product Loss

Pellicon 2 filters have a low minimum working volume – as low as 175 mL of retentate volume per square meter of membrane area. This low retentate volume permits high concentration factors to be reached with low starting volumes and maximizes the recovery of small sample volumes.

To prevent product loss, Pellicon 2 filters are 100% tested in manufacturing to ensure that every filter is integral.

In addition, Biomax and Ultracel membranes are exposed to a new high-pressure integrity test that provides greater sensitivity. The integrity test procedure and specifications are supplied so users can confirm integrity at high pressure when the filter is installed (Figure 6).

Biocompatibility

All wetted parts have been tested and meet the requirements of the USP Class VI biological test for plastics.

Superior Filter Quality

Pellicon cassettes are subjected to a complete array of quality control release tests.

A Certificate of Quality is included with every cassette.

Each casette is identified with a unique serial number.

Validatable

Since 1973, Pellicon filters and systems have been successfully used for development and scale-up of processes for manufacturing injectable protein and polysaccharide drugs, in the serum fractionation, biotechnology, vaccine and pharmaceutical industries.

Pellicon 2 filters and systems were developed based upon Millipore's experience serving these applications, and are supported by an extensive Validation Support Data Package proving performance claims and demonstrating the suitability of these filters for drug manufacturing in validated processes. This package is available upon request.

Millipore can further assist your validation efforts through:

- Design and fabrication of standard and custom turnkey TFF systems for drug manufacturing facilities
- Installation and operational qualification services for these systems
- Validation support services for tangential flow filter use in drug manufacturing processes.
- Training on TFF process scale-up, optimization and development.

A Choice of Feed Channel Screens

For optimal performance in a range of applications Pellicon 2 filters incorporate three types of feed-channel screens:

- *Type A screen (tight screen)* is optimized to operate Biomax membranes with maximum flux with low-viscosity solutions.
- *Type C screen (coarse screen)* is optimized to operate PLC series membranes with maximum flux. The Type C screen is also available with Biomax-50, 100, 300, 500 and Biomax 1000 membranes for concentration and diafiltration of viscous solutions.
- *Type V screen (open channel)* is optimized for very viscous solutions or solutions with higher levels of suspended solids.

Normalized Recirculation Rates

Parameter	Unit	Typical ΔP		
Screen Type		A	C	V
Recirculation Rate	L/min/m^2	4/6	5/35	
Differential Pressure	bar/psi	1.4/20	0.4/6	

Screen Selection Guidelines

Solution Type	Screen Type
Dilute protein solution or low viscosity solutions (MAbs, interferons)	A screen (tight screen)
Concentrated protein solutions or high viscosity solutions (IgG, biopolymers)	C screen (course screen)
High viscosity solutions (polysaccharides, certain microfiltration or clarification applications)	V screen (loose screen)

For More Detailed Information

Request literature number P17512 – User Guide for Pellicon Filters.

Specifications

Temperature Range

Mini, Cassette and Maxi:
4 to 50 °C

Maximum Forward Transmembrane Pressure

Device Size (m^2)	Biomax	Ultracel
0.1	6.8 bar (100 psi) Max	6.8 bar (100 psi) Max
0.5	6.8 bar (100 psi) at 30 °C	3.4 bar (50 psi) at 30 °C
2.5	6.8 bar (100 psi) at 30 °C	3.4 bar (50 psi) at 30 °C

Maximum Reverse Transmembrane Pressure

Device Size (m^2)	Biomax	Ultracel
0.1	0.33 bar (5 psi)	0.33 bar (5 psi)
0.5	0.33 bar (5 psi)	0.33 bar (5 psi)
2.5	0.33 bar (5 psi)	0.33 bar (5 psi)

Prefiltration Required

Mini, Cassette and Maxi:
100 µm

Dimensions

Device	Width	Length	Thickness
Mini	5.6 cm	21 cm	1.5 cm (V screen-2.16 cm)
Cassette	17.8 cm	21 cm	1.5 cm (V screen-2.16 cm)
Maxi	17.8 cm	21 cm	7.6 cm (V screen-9.0 cm)

Membrane Selection Guideline

Membrane Type	Materials	Benefits
Biomax	Modified polyethersulfone	Highest flux ultrafiltration membrane Excellent chemical resistance Void-free structure for higher yield and reliability
Ultracel PLC	Regenerated cellulose (ideal for protein solutions < 20 g/L) PLC membranes are composite membranes cast on a microporous substrate for defect-free membranes with superior adhesion. Brings higher resolution, improved yields and superior back-pressure resistance	Extremely low protein binding hydrophilic membrane Highest product recovery and improved performance with difficult to process streams (antifoams, lipids, protein transmission applications)
Durapore	Hydrophilic PVDF	Very hydrophilic microporous membrane for cell harvest or clarification applications

Pellicon 2 Membrane Selection Chart

Approximate Molecular Weight (range of solutes retained >99%, kD)		Membrane	NMWL (kD) or Microns	Membrane Material	pH Range
High Flux Biomax Membranes – Void-free for Higher Yield and Reliability					
12 – 25	(growth factors, hormones)	Biomax-5	5	modified polyethersulfone	1 – 14
25 – 50	(growth factors, hormones)	Biomax-8	8	modified polyethersulfone	1 – 14
50 – 100	(albumin, hemoglobin)	Biomax-10	10	modified polyethersulfone	1 – 14
100 – 140	(enzymes)	Biomax-30	30	modified polyethersulfone	1 – 14
140 – 300	(IgG's)	Biomax-50	50	modified polyethersulfone	1 – 14
300 – 500	(small viruses and antigens)	Biomax-100	100	modified polyethersulfone	1 – 14
>500	(IgM's, large viruses)	Biomax-300	300	modified polyethersulfone	1 – 14
>0.03 µm	(large viruses, colloids, particulates)	Biomax-500	500	modified polyethersulfone	1 – 14
>0.03 µm	(large viruses, cells, colloids, particulates)	Biomax-1000	1000	modified polyethersulfone	1 – 14
Ultracel PLC Series – for High Recoveries					
8 – 18	(proinsulin, hematopoetic factors)	PLCCC	5	regenerated cellulose	2 – 13
18 – 60	(hemoglobin, enzymes)	PLCGC	10	regenerated cellulose	2 – 13
60 – 200	(monoclonal IgG's)	PLCTK	30	regenerated cellulose	2 – 13
200 – 500	(small viruses, viral antigens)	PLCHK	100	regenerated cellulose	2 – 13
>500	(large viruses, IgM's)	PLCMK	300	regenerated cellulose	2 – 13
>0.03 µm	(large viruses, cells, colloids, particulates)	PLCXK	1000	regenerated cellulose	2 – 13
Durapore Membranes – for Microporous Applications					
Clarify cell lysates and protein solutions, clarify viral cultures		VVPP	0.1 µm	hydrophilic PVDF	2 – 11
Harvest & wash colloidal suspensions, bacterial cells; clarify protein solutions and viral cultures		GVPP	0.22 µm	hydrophilic PVDF	2 – 11
Harvest & wash colloidal suspensions, cell & viral cultures, clarify protein solutions & viral cultures		HVMP	0.45 µm	hydrophilic PVDF	2 – 11
Harvest cell cultures or colloidal suspensions		DVPP	0.65 µm	hydrophilic PVDF	2 – 11

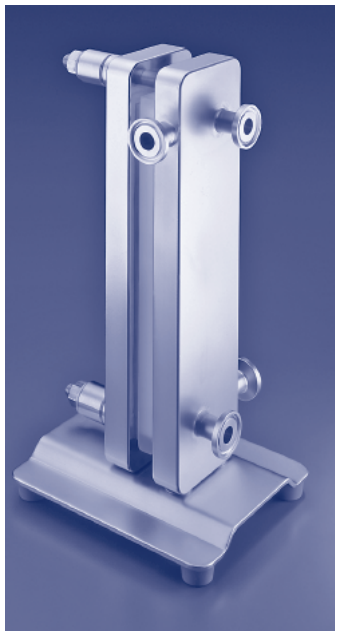
Ordering Information

Pellicon 2 Filters

	Filters with A Screens (Tight Screen)			Filters with Type C Screens (Coarse Screen)		
Membrane	**0.1 m²/1.1 ft²**	**0.5 m²/5.4 ft²**	**2.5 m²/26.9 ft²**	**0.1 m²/1.1 ft²**	**0.5 m²/5.4 ft²**	**2.5 m²/26.9 ft²**
Biomax Series – Modified Polyethersulfone						
Biomax 5	P2B0 05A 01	P2B0 05A 05	P2B0 05A 25	+	+	+
Biomax 8	P2B0 08A 01	P2B0 08A 05	P2B0 08A 25	+	+	+
Biomax 10	P2B0 10A 01	P2B0 10A 05	P2B0 10A 25	+	+	+
Biomax 30	P2B0 30A 01	P2B0 30A 05	P2B0 30A 25	+	+	+
Biomax 50	P2B0 50A 01	P2B0 50A 05	P2B0 50A 25	P2B0 50C 01	P2B0 50C 05	P2B0 50C 25
Biomax 100	P2B1 00A 01	P2B1 00A 05	P2B1 00A 25	P2B1 00C 01	P2B1 00C 05	P2B1 00C 25
Biomax 300	+	+	+	P2B3 00C 01	P2B3 00C 05	P2B3 00C 25
Biomax 500	+	+	+	P2B5 00C 01	P2B5 00C 05	P2B5 00C 25
Biomax 1000	+	+	+	P2B0 1MC 01	P2B0 1MC 05	P2B0 1MC 25
Ultracel PLC Series – Regenerated Cellulose, Composite Construction						
5 kD	NA	NA	NA	P2C0 05C 01	P2C0 05C 05	P2C0 05C 25
10 kD	NA	NA	NA	P2C0 10C 01	P2C0 10C 05	P2C0 10C 25
30 kD	NA	NA	NA	P2C0 30C 01	P2C0 30C 05	P2C0 30C 25
100 kD	NA	NA	NA	P2C1 00C 01	P2C1 00C 05	P2C1 00C 25
300 kD	NA	NA	NA	P2C3 00C 01	P2C3 00C 05	P2C3 00C 25
1000 kD	NA	NA	NA	P2C0 1MC 01	P2C0 1MC 05	P2C0 1MC 25
Durapore – Hydrophilic PVDF						
0.1 µm	+	+	+	P2VV PPC 01	P2VV PPC 05	P2VV PPC 25
0.22 µm	+	+	+	P2GV PPC 01	P2GV PPC 05	P2GV PPC 25
0.45 µm	+	+	+	P2HV MPC 01	P2HV MPC 05	P2HV MPC 25
0.65 µm	+	+	+	P2DV PPC 01	P2DV PPC 05	P2DV PPC 25

Each Pellicon filter is packed one per box and includes Operating Instructions. A Certificate of Quality is included in every box.
Silicone intercassette gaskets are required for use with Pellicon 2 filters. Two gaskets are packed in the box with every Pellicon 2 filter.
+ = On request (custom order)
NA = not available

Filters with V Screens (Loose Screen)		
0.1 m²/1.1 ft²	**0.5 m²/5.4 ft²**	**2.0 m²/21.5 ft²**
P2B0 05V 01	P2B0 05V 05	P2B0 05V 20
P2B0 08V 01	P2B0 08V 05	P2B0 08V 20
P2B0 10V 01	P2B0 10V 05	P2B0 10V 20
P2B0 30V 01	P2B0 30V 05	P2B0 30V 20
P2B0 50V 01	P2B0 50V 05	P2B0 50V 20
P2B 100V 01	P2B1 00V 05	P2B1 00V 20
P2B3 00V 01	P2B3 00V 05	P2B3 00V 20
P2B5 00V 01	P2B5 00V 05	P2B5 00V 20
P2B0 1MV 01	P2B0 1MV 05	P2B0 1MV 20
P2C0 05V 01	P2C005V 05	P2C0 05V 20
P2C0 10V 01	P2C0 10V 05	P2C0 10V 20
P2C0 30V 01	P2C0 30V 05	P2C0 30V 20
P2C1 00V 01	P2C1 00V 05	P2C1 00V 20
P2C3 00V 01	P2C3 00V 05	P2C3 00V 20
P2C0 1MV 01	P2C0 1MV 05	P2C01MV 20
P2VV PPV 01	P2VV PPV 05	P2VV PPV 20
P2GV PPV 01	P2GV PPV 05	P2GV PPV 20
P2HV MPV 01	P2HV MPV 05	P2HV MPV 20
P2DV PPV 01	P2DV PPV 05	P2DV PPV 20



Pellicon 2 Mini Holder

Pellicon 2 Mini holder operates one to three Mini filters in parallel for total areas of 0.1 to 0.3 m² (1.1 – 3.3 ft²). This sanitary holder is tightened with a small torque wrench to compress the filters between a manifold plate that conveys fluids in and out of the filters and an end plate that seals the filters together. The Mini holder is designed for process development and small volume pharmaceutical manufacturing.

Materials of Construction

Manifold and End Plates:
316 L stainless steel

Base, Tie Rods, Spacers and Washers:
304 stainless steel

Feet:
Thermoplastic rubber

Gaskets:
Silicone

Nuts:
Silicone bronze

Separator Plates

An optional separator plate allows processing simultaneously with up to three 0.1 m²/1.1 ft² cassettes to determine the best molecular weight cut-off in a single study on the same feed material.

Connections

All manifold connections are standard ½-inch sanitary clamp type.

Operating Parameters

Temperature Range:
4 to 50 °C. The Mini holder can be autoclaved without filters installed. The filters themselves cannot be autoclaved.

Maximum Pressure:
6.8 bar

Dimensions

Height: 260 mm; *Width:* 114 mm

Length: 140 mm; *Weight:* 5 kg

Holder Manifold Volume:

Feed plus retentate: 5.3 mL

Permeate: 6.4 mL

Stainless Steel Pellicon Holder

XX42P0080

The stainless steel Pellicon filter holder, designed for sanitary applications, can be used alone or to expand existing cassette ultrafiltration (CUF) systems or to replace existing holders.

It requires only to be connected to an existing sanitary pump and piping for tangential flow microporous filtration or ultrafiltration.

It can accomodate up to 5 m^2/55 ft^2 filter area as shipped with long tie rods or 0.5 to 2.5 m^2 (5.4 – 26.9 ft^2) with accessory short tie rods.

Materials of Construction

Wetted Surfaces:
316 L stainless steel

Non-wetted Surfaces:
Silicon bronze nuts

Dimensions

Length: 28 cm; *Width:* 19 cm

Height: 25 cm

Operating Parameters

Operating Temperature Range:
4 to 50 °C. The Pellicon holder can be autoclaved without pressure gauges and filters; holder with gauges cannot be steamed. Pellicon filters cannot be steamed or autoclaved.

Connections

Sanitary ¾" TC connections;
1½" TC connections for gauges.

Shipping Weight

24 kg

Process-scale Pellicon Holder

The Pellicon Process-scale Holder is a unique innovation for production scale Pellicon systems. This holder, vertically mounted, can hold up to 80 m^2/880 ft^2 of membrane area.

Benefits

- Extremely compact footprint
- Easy to change cassettes
- Easy to vent and fully drain
- Simple connections
- Up to 4 levels. Can be easily extended in levels for simple membrane area expansion
- Each level up to 20 m^2/220 ft^2
- Uses standard and Maxi Cassettes
- Can be adapted for series or parallel configurations
- Simplifies pipework connection
- Hydraulic closure systems are available for the stainless-steel Pellicon holder and the process-scale Pellicon holder. These systems are convenient, reliable and easy to use to enable rapid and repeatable loading operation and storage of Pellicon 2 cassettes.

Materials of Construction

Manifold segment, fitting blocks and end plate 316 L stainless steel; tie rods 304 and 304 L stainless steel.

Ordering Information

Pellicon 2 Filter Holders

Description	Catalogue No.
Pellicon 2 Mini filter holder	XX42 PMI NI
Pressure gauges One diaphragm-protected digital pressure gauge, 0 – 7 bar, ¾-inch fittings	XX42 PSG 01L
Pressure gauge adapters	XX42 PM0 01
Fitting kit Contains all tees, clamps, gaskets and a valve to connect tubing and pressure gauges to the Pellicon 2 Mini holder	XX42 PFK 01
Pellicon filter holder (for cassettes and Maxi filters)	XX42 P00 80
Pellicon 2 double thick gasket	PSSP 2XC 10
Pellicon Process-scale holder support and plate	XX42 SSP LT
Pellicon Process-scale holder	On request

To Place an Order or Receive Technical Assistance

For additional information call your nearest Millipore office:
In the U.S. and Canada,
call toll-free **1-800-MILLIPORE (1-800-645-5476)**
In the U.S., Canada and Puerto Rico, fax orders to **1-800-MILLIFX (1-800-645-5439)**
Outside of North America contact your local office. To find the office nearest you visit **www.millipore.com/offices.**
Internet: **www.millipore.com**
Technical Service:
www.millipore.com/techservice

MILLIPORE

A Typical Pellicon Production Processing System

Millipore supplies a range of standard and custom engineered systems. These systems can contain from 1 m^2/11 ft^2 to several hunded m^2 of membrane area, with Clean-in-Place (CIP) or Steam-in-Place (SIP) integrated as appropriate. Systems can also be supplied with integrated process vessels in manual or fully automatic versions.

All systems are designed, engineered and manufactured in ISO® 9001 registered facilities, and are supplied with extensive validation data support packages.

Please contact us to discuss your specific application and process requirements.

Pellicon XL Devices for Process Development

For process development of volumes from 50 mL to 1 liter, Millipore offers Pellicon XL devices. This small volume TFF filter is designed for true scalability by providing the same flow path, channel length, and channel height as the Pellicon 2 cassettes. Based on proven TFF membrane technology, Pellicon XL devices ensure reliable, consistent and predictable performance.

Lit. No. DS1210EN00 Rev. A 10/04
Printed in U.S.A. 04-324

Criterion XT™ Precast Gel Instruction Guide

Catalog Number
345-9898

BIO-RAD

For Technical Service Call Your Local Bio-Rad Office
or in the US, Call **1-800-4BIORAD** (1-800-424-6723)

Table of Contents

Section 1
General Information

1.1 Introduction

Criterion is the next generation of dedicated precast gel systems. The innovative, easy-to-use design produces superior resolution while allowing you to run more samples per gel. Compared to any other precast gel system, Criterion produces more results while providing significant cost and time savings. Some of the unique features and benefits provided are:

- 12 month shelf life for Bis-Tris gels
- 8 month shelf life for Tris-acetate gels
- Room temperature storage for Bis-Tris gels
- Easy sample preparation without extra anti-oxidant addition steps
- Patented integral buffer chamber that eliminates buffer leaks
- Up to 26 sample capacity per gel
- Flexibility to run one or two gels
- Multichannel pipet compatible gels
- Outlined and numbered wells that simplify sample loading
- J-foot that improves gel drying and blotting results

US Patents #5,073,246, #5,656,145, #6,093,301 and other patents issued and pending.

1.2 Criterion XT Precast Gels

Criterion XT precast gels are formulated at pH near neutrality to optimize gel matrix stability, significantly delaying acrylamide hydrolysis, which occurs in traditional Laemmli systems. Specially optimized buffers result in tight, consistently resolved bands throughout the life of the gel.

This versatile system allows the separation of small to large proteins using just two gel buffer systems: Criterion XT Bis-Tris precast gels for small to mid-sized proteins and Criterion XT Tris-acetate precast gels for large proteins.

The Criterion XT Bis-Tris gels are based on a Bis-Tris·HCl buffer system (pH 6.4) that uses discontinuous chloride and MES or MOPS ion fronts to form moving boundaries to stack and then separate denatured proteins by size. The chemistry of the XT Bis-Tris gels allows maximum stability and consistent results for a minimum of one year. Running the same XT Bis-Tris gels with the XT MES denaturing running buffer or the XT MOPS denaturing running buffer will produce different migration patterns. A combination of these two running buffers and our three XT Bis-Tris gels can produce up to six different migration patterns in the small and mid-size range.

The Criterion XT Tris-acetate gels are based on a Tris-acetate buffer system (pH 7.0). It uses discontinuous acetate and Tricine ion fronts to form moving boundaries to stack and then separate large denatured proteins by molecular weight. The Criterion XT Tris-acetate gels can also be used to separate proteins by their charge-to-mass ratio (under native-PAGE conditions). This is possible because the XT Tris-acetate gels are made without SDS, allowing the sample buffer and running buffer to dictate the separation mechanism. The nonreducing and nondenaturing environment of native PAGE allows the detection of biological activity and can improve antibody detection. Native PAGE can also be used to resolve multi-protein bands where molecular mass separation by SDS-PAGE would reveal only one and for the separation of intact protein

complexes. Separation by native PAGE with XT Tris-acetate gels uses discontinuous acetate and glycine ion fronts to form moving boundaries to stack and separate proteins by both size and charge.

Protein samples for the Criterion XT precast gel system are prepared in a reducing denaturing sample buffer. The sample buffer contains XT reducing agent, a pH neutralized and stabilized solution of TCEP as the reducing agent; heat and SDS are used to denature the proteins. In addition, the use of TCEP in combination with Bio-Rad's optimized running buffers maintains proteins in a fully reduced state during the electrophoresis run, eliminating the need for an anti-oxidant in the upper buffer chamber. Criterion XT Tris-acetate precast gels can also be used for native PAGE. Proteins are prepared in a nonreducing, nondenaturing sample buffer, which maintains the proteins' native structure and charge density.

1.3 Criterion System Specifications

Gel material	Polyacrylamide
Gel dimensions (W x L)	13.3 x 8.7 cm
Gel thickness	1.0 mm
Resolving gel height	6.5 cm
Cassette dimensions (W x L)	15.0 x 10.6 cm
Cassette material	Styrene copolymer
Comb material	Polycarbonate
Storage tray material	PET
Upper running buffer volume	60 ml
Lower running buffer volume	800 ml
Storage conditions	Bis-Tris gels: Store flat at ambient temperature; DO NOT FREEZE
	Tris-acetate gels: Store flat at 4°C; DO NOT FREEZE
Gel shelf life	12 months for Bis-Tris gels; 8 months for Tris-acetate gels

1.4 Criterion XT Comb Configurations

Comb	Load Volume	Comments
12+2 well	45 µl with two 15 µl reference wells	Multichannel pipet compatible
18-well	30 µl	
26-well	15 µl	Multichannel pipet compatible
Prep+2 well	800 µl with two 15 µl reference wells	
IPG	11 cm ReadyStrip™ IPG strip	
IPG+1 well	11 cm ReadyStrip IPG strip with one 15 µl reference well	

Section 2
Setup and Basic Operation

2.1 Setting Up and Running Criterion XT Gels

1. Each Criterion XT gel is packaged individually in a plastic storage tray. Remove the cover by gently pulling the corner tab up and diagonally across the package. Remove the gel from the package.

2. Remove the comb and gently rinse the wells with ddH_2O or running buffer.

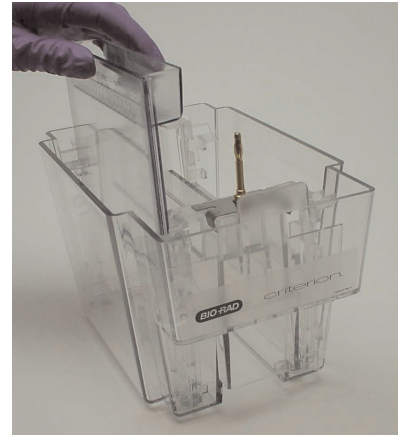
3. Remove the tape from the bottom of the cassette by pulling the tab across the gel.

4. Insert the Criterion XT gel into one of the slots in the Criterion cell tank. Ensure that each integral buffer chamber faces the center of the cell.

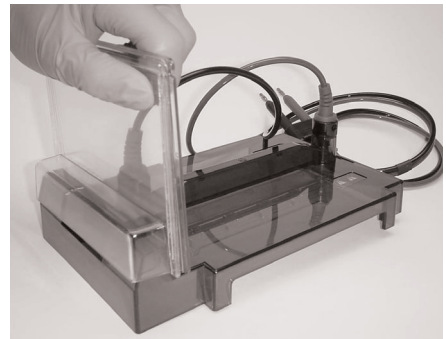
5. Fill each integral buffer chamber with 60 ml running buffer.

6. Load samples using a Hamilton syringe or a pipet with gel loading tips. A sample loading guide can be placed on the outer edge of the cassette to aid in aligning pipet tips with the wells. This is especially useful with multichannel pipets.

7. Fill each half of the lower buffer tank with 400 ml of running buffer to the marked fill line.

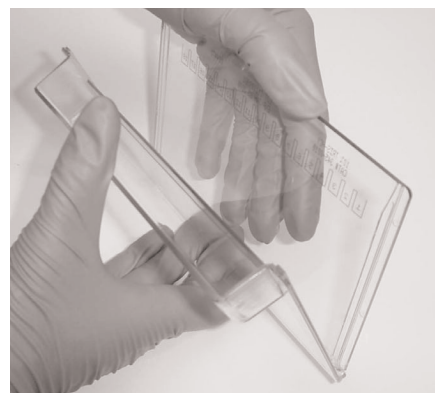


8. Place the lid on the tank, aligning the color-coded banana plugs and jacks. See section 3.6 for power conditions.



2.2 Opening Criterion XT Cassettes and Removing the Gels

1. After electrophoresis is complete, turn off the power supply and disconnect the electrical leads.
2. Remove the lid from the tank and remove the Criterion XT gel(s) from the cell. Pour off and discard the upper running buffer.
3. Invert the cassette and place the integral buffer chamber over the cassette-opening tool built into the Criterion cell lid.
4. Firmly press down on the cassette to crack the cassette welds on both sides of the cassette. The cassette will split open approximately 1/3 of the way.
5. Alternatively, the gel cassette can be opened by sliding the tapered back of the comb into the slits on either side of the cassette.
6. Pull the two halves of the cassette apart to completely expose the gel.
7. Remove the gel by either floating the gel into a fixing or staining solution or by carefully lifting the gel from the cassette.



Section 3
SDS-PAGE and Native PAGE

3.1 Criterion XT Gel Selection Guide

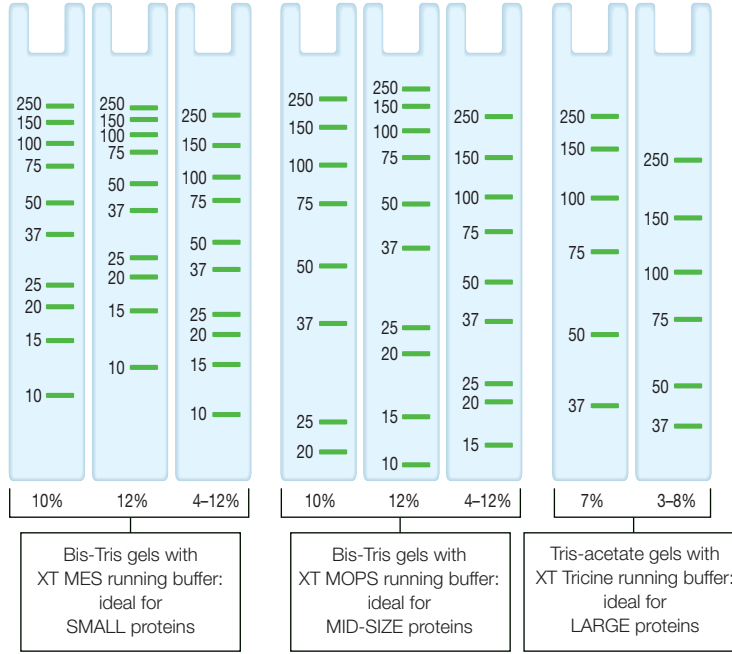
Criterion XT gels are available in a wide selection of single acrylamide percentages and gradients for the separation of proteins by SDS-PAGE or native PAGE.

Optimal Separation

Bis-Tris Gels	**With XT MES Running Buffer**	**With XT MOPS Running Buffer**
10%	2.5–200 kD	14–220 kD
12%	1–30 kD	6–66 kD
4–12%	2.5–200 kD	10–300 kD
Tris-Acetate Gels*	**With XT Tricine Running Buffer**	**With Tris/Glycine Running Buffer**
7%	36–200 kD	N/A
3–8%	40–400 kD	N/A

*Because Criterion XT Tris-acetate gels are made without SDS, they can be used to separate proteins by both SDS-PAGE and native PAGE.

Criterion XT Protein Migration Chart



3.2 Bis-Tris Gel Composition

Gel buffer	Bis-Tris·HCl, pH 6.4
Cross linker	5% C
Stacking gel	4% T, 5% C
Storage buffer	Bis-Tris·HCl, pH 6.4
Shelf life	12 months; individual expiration date is printed on each cassette; store flat at ambient temperature

3.3 Tris-Acetate Gel Composition

Gel buffer	Tris-acetate, pH 7.0
Cross linker	3.8% C
Stacking gel	4% T, 3.8% C
Storage buffer	Tris-acetate, pH 7.0
Shelf life	8 months; individual expiration date is printed on each cassette, store flat at 4°C

3.4 Criterion XT Buffers and Reagents

Bis-Tris running buffer for SDS-PAGE	20x XT MOPS (dilute to 1x) For separation of mid-size proteins Catalog #161-0788	or	XT MES (dilute to 1x) For separation of small proteins Catalog #161-0789
Tris-acetate running buffer for SDS-PAGE	20x XT Tricine (dilute to 1x) For separation of large proteins Catalog #161-0790		
Tris-acetate running buffer for Native-PAGE	10x Tris-Glycine (dilute to 1x) Catalog #161-0732		
XT sample buffer	Catalog #161-0791		
XT reducing agent	Catalog #161-0792		

3.5 Sample Preparation

Determine the appropriate protein concentration of your sample based on the detection method and load volume used. (See section 4.1 for approximate stain sensitivities.) XT sample buffer is a 4x concentrate and can be used with both dilute and concentrated samples. Refer to the sample preparation guide below:

Sample Preparation Guide

SDS-PAGE	**Native-PAGE**
25 µl XT sample buffer 5 µl XT reducing agent x µl sample	50 µl Native sample buffer x µl sample
Make up to 100 µl with ddH_20	Make up to 100 µl with ddH_2O
Heat sample at 95°C for 5 min.	Do not heat sample

3.6 Running Conditions

Gel type	**Bis-Tris (for SDS-PAGE)**	**Bis-Tris (for SDS-PAGE)**	**Tris-Acetate (for SDS-PAGE)**	**Tris-Acetate (for Native-PAGE)**
Running buffer	**XT MOPS**	**XT MES**	**XT Tricine**	**Tris/Glycine**
Power conditions	200 V constant	200 V constant	150 V constant	200 V constant
Run time	60 min	45 min	65 min	75 min
Starting current	165–175 mA/gel	185–200 mA/gel	170–180 mA/gel	70–80 mA/gel
Final current	60–70 mA/gel	90–110 mA/gel	85–95 mA/gel	25–35 mA/gel

Section 4
2-D Electrophoresis

4.1 Equilibration

Use existing equilibration protocols as described in the ReadyPrep 2-D Starter Kit (catalog #163-2105 or bulletin 411009) or existing protocols and buffers used for Tris-HCl gels.

4.2 Agarose Overlay

Make a solution of 0.6% low melt agarose and 0.002% Bromophenol blue. To make 10 ml of the agarose overlay, mix 9.5 ml of the above agarose with 0.5 ml of 20x XT Running Buffer. Use the XT Running Buffer that will be used to run the second dimension gel.

Section 5
Staining and Detection

5.1 SDS-PAGE and Native PAGE Detection

Total Protein Gel Stain Method	Sensitivity	Optimal Protein Load	Advantages	Disadvantages
Coomassie Blue R-250	36–47 ng	~0.5 µg/band	Laboratory standard	Requires MeOH
Bio-Safe™ Coomassie stain	8–28 ng	~0.5 µg/band	Nonhazardous, uses no MeOH	More steps than Coomassie R-250
Zinc stain	6–12 ng	~0.2 µg/band	High-contrast, fast, reversible stain	Negative stain, must be photographed; SDS-PAGE only
Silver Stain Plus™ kit	0.6–1.2 ng	~0.01 µg/band	Simple, robust, mass spectrometry compatible	Will not stain glycoproteins
Silver stain	0.6–1.2 ng	~0.01 µg/band	Stains complex proteins: i.e., glycoproteins and lipoproteins	Not mass spectrometry compatible
SYPRO Orange protein stain	4–8 ng	~0.2 µg/band	Will not stain nucleic acids; mass spectrometry compatible	Optimization required for maximum sensitivity
SYPRO Ruby protein gel stain	1–10 ng	~0.2 µg/band	Broad dynamic range, simple robust protocol	Requires imaging instrument for maximum sensitivity

Total Protein Blot Stain Method	Sensitivity	Optimal Protein Load	Advantages	Disadvantages
SYPRO Ruby protein blot stain	2–8 ng	~0.2 µg/band	Compatible with mass spectrometry, Edman-based sequencing, and standard immunological procedures	Multiple-step protocol; Requires imaging instrument for maximum sensitivity
Colloidal gold stain	1 ng	~0.1 µg/band	Sensitive, one step	Not compatible with nylon membranes
Enhanced colloidal gold detection kit	10–100 pg	~0.1 µg/band	Increases sensitivity of colloidal gold stain	Multiple steps
Amido Black	100–1,000 ng	~5 µg/band	Standard membrane stain, economical	Low sensitivity

Immunoblot Detection Method	Sensitivity	Optimal Protein Load	Advantages	Disadvantages
4CN colorimetric (HRP)*	500 pg	~0.25 µg/band	Fast detection	Results may fade
DAB colorimetric (HRP)	500 pg	~0.25 µg/band	Fast detection	Contains toxic chemicals
Opti-4CN colorimetric (HRP)	100 pg	~0.05 µg/band	Color does not fade	More expensive than 4CN
Amplified Opti-4CN™ colorimetric (HRP)	10 pg	~0.005 µg/band	High sensitivity, low background	Amplification requires additional steps
BCIP/NBT colorimetric (AP)	100 pg	~0.05 µg/band	Sensitive; multiple antigens	May detect endogenous enzyme activity
Amplified AP*	10 pg	~0.005 µg/band	High sensitivity	Amplification requires additional steps
Immun-Star™ chemiluminescent (AP)	10 pg	~0.005 µg/band	Long-lasting signal, short and multiple exposures possible	Requires visualization on film or instrumentation

*(HRP) horseradish peroxidase; (AP) alkaline phosphatase

Section 6
Blotting

Criterion XT gels are blotted using the same buffers and protocols used to blot Tris-HCl and other polyacrylamide gels. Please refer to the Criterion blotter instruction manual (bulletin 4006190) for detailed instructions on how to blot gels. Tris/Glycine (Towbin) transfer buffer is recommended for western transfer of the Criterion XT pre-cast gels.

Section 7
Troubleshooting

Improper storage of Criterion XT gels can produce numerous artifacts. Criterion XT Bis-Tris gels should be stored flat at ambient temperature. Criterion XT Tris-acetate gels should be stored flat at 4°C. Avoid freezing. If you suspect your gels have been stored improperly, DO NOT USE THEM.

Problem	Possible Cause	Solution
Samples do not migrate into gel	Tape at the bottom of the cassette not removed	Remove tape
	Insufficient buffer in integral buffer chamber	Fill integral buffer chamber with 60 ml running buffer
	Insufficient lower electrode buffer	Fill both halves of the lower buffer tank with 400 ml running buffer when running two gels
	Electrical disconnection	Check electrodes and connections
Bands “smile” across gel, band pattern curves upward at both sides of the gel	Excess heating of gel	Check buffer composition
		Completely fill both halves of the lower buffer tank with 400 ml running buffer when running two gels
		Do not exceed recommended running conditions
Skewed or distorted bands, lateral band spreading	Excess salt in samples	Remove salts from sample by dialysis or desalting column prior to sample preparation
	Insufficient sample buffer or wrong formulation	Check buffer composition and dilution instructions

Problem	Possible Cause	Solution
Vertical streaking	Samples overloaded	Dilute sample
		Selectively remove predominant protein in the sample
	Sample precipitation	Centrifuge samples to remove particulates prior to sample loading
Gels run too fast, provide poor resolution, and gel temperature is too high	Running buffer is too concentrated	Check buffer composition
Artifact bands at ~60–70 kD	Possible skin keratin contamination	Wear gloves while cleaning all dishware and while handling and loading gel
		Filter all solutions through nitrocellulose
		Use 10% iodoacetamide to eliminate keratin bands

Section 8 Ordering Information

8.1 Criterion XT Gels

Criterion XT Bis-Tris Gels	12+2 Well	18-Well	26-Well	Prep Well	IPG+1 Well	IPG Well
10% Bis-Tris	345-0111	345-0112	345-0113	345-0114	345-0115	345-0116
12% Bis-Tris	345-0117	345-0118	345-0119	345-0120	345-0121	345-0122
4–12% Bis-Tris	345-0123	345-0124	345-0125	345-0126	345-0127	345-0128

Criterion XT Tris-Acetate Gels	12+2 Well	18-Well	26-Well	Prep Well	IPG+ 1 Well	IPG Well
3–8% Tris-Acetate	345-0129	345-0130	345-0131	345-0132	345-0133	345-0134
7% Tris-Acetate	345-0135	345-0136	345-0137	345-0138	345-0139	345-0140

8.2 Criterion XT Buffers and Kits

Catalog #	Description
161-0788	XT MOPS Running Buffer, 20x, 500 ml
161-0789	XT MES Running Buffer, 20x, 500 ml
161-0790	XT Tricine Running Buffer, 20x, 500 ml
161-0791	XT Sample buffer, 4x, 10 ml
161-0792	XT Reducing Agent, 20x, 1 ml
161-0793	XT MOPS Buffer Kit, includes 500 ml 20x XT MOPS running buffer, 10 ml 4x XT sample buffer, 1 ml 20x XT reducing agent
161-0796	XT MES Buffer Kit, includes 500 ml 20x XT MOPS running buffer, 10 ml 4x XT sample buffer, 1 ml 20 x XT reducing agent
161-0797	XT Tricine Buffer Kit, includes 500 ml 20x XT MOPS running buffer, 10 ml 4x XT sample buffer, 1 ml 20x XT reducing agent

8.3 Other Related Products

161-0738	Native Sample Buffer, 30 ml
161-0734	10x Tris/Glycine, 1 L
161-0404	Bromophenol Blue, 10 g
161-0311	Certified Low-Melt Agarose, 25 g
163-2107	ReadyPrep 2-D Starter Kit Equilibration Buffer I, with DTT
163-2108	ReadyPrep 2-D Starter Kit Equilibration Buffer II, with DTT

8.4 Criterion Gels

Criterion Tris-HCl Gels	12+2 Well 45 µl Samples	18-Well 30 µl Samples	26-Well 15 µl Samples	Prep+2 Well 800 µl Samples	IPG Well 11 cm IPG Strip	IPG+1 Well 11 cm IPG Strip
5% Tris-HCl	345-0001	345-0002	345-0003	345-0004	-	-
7.5% Tris-HCl	345-0005	345-0006	345-0007	345-0008	-	-
10% Tris-HCl	345-0009	345-0010	345-0011	345-0012	345-0013	345-0101
12.5% Tris-HCl	345-0014	345-0015	345-0016	345-0017	345-0018	345-0102
15% Tris-HCl	345-0019	345-0020	345-0021	345-0022	-	-
18% Tris-HCl	345-0023	345-0024	345-0025	345-0026	-	-
4–15% Tris-HCl	345-0027	345-0028	345-0029	345-0030	345-0031	345-0103
4–20% Tris-HCl	345-0032	345-0033	345-0034	345-0035	345-0036	345-0104
8–16% Tris-HCl	345-0037	345-0038	345-0039	345-0040	345-0041	345-0105
10.5–14% Tris-HCl	345-9949	345-9950	345-9951	345-9952	345-9953	345-0106
10-20% Tris-HCl	345-0042	345-0043	345-0044	345-0045	345-0046	345-0107

Criterion TBE Gels	12+2 Well 45 µl Samples	18-Well 30 µl Samples	26-Well 15 µl Samples	Prep+2 Well 800 µl Samples
5% TBE	345-0047	345-0048	345-0049	345-0050
10% TBE	345-0051	345-0052	345-0053	345-0054
15% TBE	345-0055	345-0056	345-0057	345-0058
4–20% TBE	345-0059	345-0060	345-0061	345-0062

Criterion Peptide Gels	12+2 Well 45 µl Samples	18-Well 30 µl Samples	26-Well 15 µl Samples	Prep+2 Well 800 µl Samples
16.5% Peptide	345-0063	345-0064	345-0065	345-0066
10–20% Peptide	345-0067	345-0068	345-0069	345-0070

Criterion IEF Gels	12+2 Well 45 µl Samples	18-Well 30 µl Samples	26-Well 15 µl Samples	Prep+2 Well 800 µl Samples
IEF pH 3–10	345-0071	345-0072	345-0073	345-0074
IEF pH 5–8	345-0075	345-0076	345-0077	345-0078

Criterion Zymogram Gels	12+2 Well 45 µl Samples	18-Well 30 µl Samples	26-Well 15 µl Samples	Prep+2 Well 800 µl Samples
10% Zymogram, gelatin	345-0079	345-0080	345-0081	-
12.5% Zymogram, casein	345-0082	345-0083	345-0084	-

Criterion TBE-Urea Gels	12+2 Well 45 µl Samples	18-Well 30 µl Samples	26-Well 15 µl Samples	Prep+2 Well 800 µl Samples
5% TBE-Urea	345-0085	345-0086	345-0087	-
10% TBE-Urea	345-0088	345-0089	345-0090	-
15% TBE-Urea	345-0091	345-0092	345-0093	-

8.5 Criterion Gel Accessories

345-9920	Criterion Staining/Blotting Trays, 12
345-9901	Criterion Empty Cassettes, 1.0 mm with 12+2 comb, 10
345-9902	Criterion Empty Cassettes, 1.0 mm with 18-well comb, 10
345-9903	Criterion Empty Cassettes, 1.0 mm with 26-well comb, 10
345-9904	Criterion Empty Cassettes, 1.0 mm with prep+2 comb, 10
345-9905	Criterion Empty Cassettes, 1.0 mm with IPG comb, 10
165-6006	Criterion Sample Loading Guide, 12+2 well, 1
165-6007	Criterion Sample Loading Guide, 18-well, 1
165-6008	Criterion Sample Loading Guide, 26-well, 1

8.6 Protein Standards

161-0362	Precision Plus Protein™ Unstained Standards (10–250 kD), 500 µl, 100 applications
161-0373	Precision Plus Protein All Blue Standards (10–250 kD), 500 µl, 100 applications
161-0317	SDS-PAGE Standards, broad range, 200 µl
161-0303	SDS-PAGE Standards, high range, 200 µl
161-0304	SDS-PAGE Standards, low range, 200 µl
161-0324	Kaleidoscope Prestained Standards, broad range, 500 µl
161-0325	Kaleidoscope Polypeptide Standards, 500 µl
161-0326	Polypeptide SDS-PAGE Standards (1.4–26.6 kD), 200 µl, 400 applications

8.7 Detection Reagents

Total Protein Gel Stains

161-0436	Coomassie Blue R-250 Stain Solution, 1 L
161-0438	Coomassie Blue R-250 Destain Solution, 1 L
161-0400	Coomassie Brilliant Blue R-250, 10 g
161-0786	Bio-Safe Coomassie Stain, 1 L
161-0440	Zinc Stain and Destain Kit
161-0449	Silver Stain Plus Kit
161-0443	Bio-Rad Silver Stain Kit
170-3120	SYPRO Orange Protein Stain, 500 µl
170-3125	SYPRO Ruby Protein Gel Stain, 1 L
161-0434	IEF Gel Staining Solution, 1 L

Immunoblot Detection

170-6431	HRP Conjugate Substrate Kit, 4CN
170-6535	HRP Color Development Reagent, DAB
170-8238	Amplified Opti-4CN Kit
170-8235	Opti-4CN Substrate Kit
170-6432	BCIP/NBT AP Conjugate Substrate Kit
170-6412	Amplified Alkaline Phosphatase Kit
170-5012	Immun-Star™ Substrate Pack
170-5040	Immun-Star HRP Substrate, 500 ml

Total Protein Blot Stains

170-3127	SYPRO Ruby Protein Blot Stain, 200 ml
170-6527	Colloidal Gold Total Protein Stain, 500 ml
170-6517	Enhanced Colloidal Gold Detection Kit
161-0402	Amido Black 10B, 25 g

8.8 Blotting Membranes

162-0175	Immun-Blot PVDF Membrane, 10 x 15 cm, 10 sheets
162-0232	0.2 µm Nitrocellulose/Filter Paper Sandwich, 8.5 x 13.5 cm, 20 pack
162-0233	0.2 µm Nitrocellulose/Filter Paper Sandwich, 8.5 x 13.5 cm, 50 pack
162-0234	0.45 µm Nitrocellulose/Filter Paper Sandwich, 8.5 x 13.5 cm, 20 pack
162-0235	0.45 µm Nitrocellulose/Filter Paper Sandwich, 8.5 x 13.5 cm, 50 pack
162-0236	Sequi-Blot™ PVDF/Filter Paper Sandwich, 8.5 x 13.5 cm, 20 pack
162-0237	Sequi-Blot PVDF/Filter Paper Sandwich, 8.5 x 13.5 cm, 50 pack

8.9 Equipment

165-6001	Criterion Cell, includes tank, lid with power cables, three sample loading guides
170-4070	Criterion Blotter With Plate Electrodes
170-4071	Criterion Blotter With Wire Electrodes

Catalog Number
345-9898

Bio-Rad Laboratories, Inc.

Life Science Group

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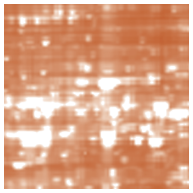


ELECTROPHORESIS

Criterion™ XT Precast Gels

Get unsurpassed results with the Criterion system:

- Separate up to 26 samples on one gel
- Resolve more proteins on every gel
- Never lose track of critical gel information
- Eliminate gel trimming with the patented* J-foot design
- Load gels quickly and easily with sample loading guides and multichannel pipet-compatible combs
- Open cassettes safely and easily without a separate tool

All the Benefits of the Criterion™ Format With an Extended Shelf Life

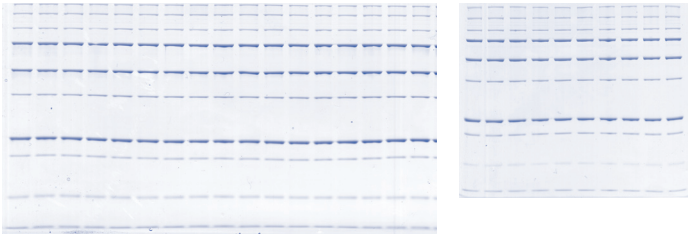
Introduction

The Criterion system is a convenient, high-quality precast gel system. Its superior features and numerous benefits are ideally suited for use with Criterion XT precast gels, which offer a shelf life of 12 months for Bis-Tris gels and 8 months for Tris-acetate gels.

Criterion XT precast gels are formulated at a near-neutral pH, significantly delaying acrylamide hydrolysis compared to traditional Laemmli systems. Their chemical composition allows maximum stability and consistent results for up to one year.

Criterion XT gels are designed to work with optimized sample and running buffers without the need for antioxidant addition. Like traditional Laemmli systems, Criterion XT gels use discontinuous buffer ion fronts that form moving boundaries to stack and then separate proteins (see diagram to right).

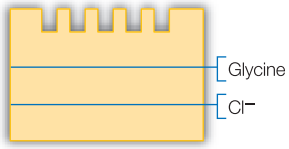
This high-performance, versatile system allows separation of small to large proteins using just two types of gels: Criterion XT Bis-Tris gels for small to midsized proteins and Criterion XT Tris-acetate gels for large proteins.



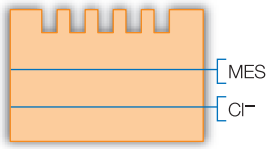
Criterion XT gels separate more samples with comparable resolution. Precision Plus Protein™ standards separated on a Criterion XT 12% Bis-Tris 18-well gel with XT MOPS buffer (left) and a competitor's 12% Bis-Tris 10-well mini gel (right). Both gels were stained with Bio-Safe™ Coomassie stain and imaged on a Molecular Imager® GS-800™ calibrated densitometer.

* U.S. patent 6,093,301.

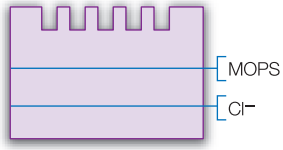
Leading and Trailing Ions



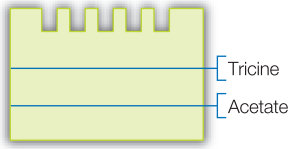
Tris-HCl gel with Tris/glycine buffer (Laemmli system)



Bis-Tris gel with XT MES running buffer for small proteins (Criterion XT system)



Bis-Tris gel with XT MOPS running buffer for midsized proteins (Criterion XT system)



Tris-acetate gel with XT Tricine running buffer for large proteins (Criterion XT system)

BIO-RAD

Choice of Separation Mechanism

Bis-Tris Gels

Criterion XT Bis-Tris gels are formulated using a Bis-Tris-HCl buffer system (pH 6.4) for separation of proteins by molecular weight. By selecting from two running buffers, you can expand the separation capability of a single Bis-Tris gel type. To further refine your resolution range, refer to the migration chart on the right for the acrylamide concentration appropriate for your proteins of interest.

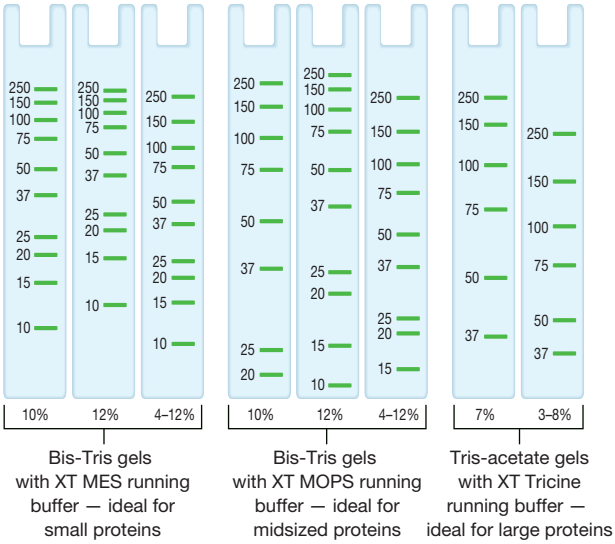
Tris-Acetate Gels

Criterion XT Tris-acetate gels are formulated using a Tris-acetate buffer system (pH 7.0) that separates large denatured proteins by molecular weight when run with XT Tricine running buffer. These gels are made without SDS, so they can also be used with nondenaturing sample and running buffers (native PAGE conditions) to separate proteins by mass-to-charge ratio. The nonreducing, nondenaturing conditions of native PAGE preserve biological activity and can improve antibody detection. Native PAGE can also resolve multiple protein bands where molecular mass separation by SDS-PAGE would reveal only one.

For native PAGE on Criterion XT gels, nonreducing, nondenaturing native sample buffer and Tris/glycine running buffer can be used to maintain protein secondary structure and native charge density.

Gel and Buffer Selection Guide

The banding patterns below indicate the optimal separation ranges (in kD) for each acrylamide percentage in combination with the buffer system specified.



Recommended Standards

Because of the pH differences between traditional Laemmli and Criterion XT buffer systems, some prestained protein standards may migrate differently. The standards listed below are recommended with Criterion XT gels.

For molecular weight determination:

- Precision Plus Protein unstained standards
- Unstained SDS-PAGE standards

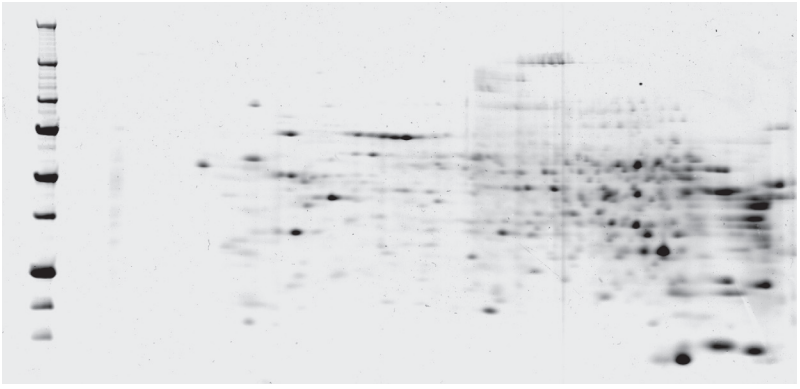
For molecular weight estimation:

- Precision Plus Protein all blue standards
- Kaleidoscope™ prestained standards
- Prestained SDS-PAGE standards

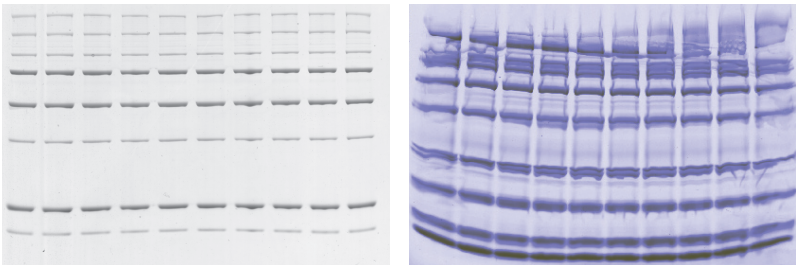
The Criterion precast gel system.

Greater Resolving Area for 1-D and 2-D Electrophoresis

Compared to traditional mini formats, the Criterion system provides 60% more resolving area in the first dimension and 24% more resolving area in the second dimension.* Use existing IPG strip equilibration protocols for second-dimension analysis on Criterion XT gels.



Criterion XT gels yield excellent 2-D electrophoresis results. Mouse liver extract separated on an 11 cm ReadyStrip™ IPG strip, pH 3–10, followed by second-dimension separation on a 4–12% Criterion XT Bis-Tris gel with Precision Plus Protein standards. The gel was run with XT MES running buffer, stained with SYPRO Ruby protein gel stain, and imaged on the Molecular Imager FX™ system.



Improvement of shelf life and gel quality with neutral pH formulation. Left, a typical neutral-pH gel at 20 weeks; right, a typical Tris-HCl (Laemmli system) gel at 14 weeks.

Converting to Criterion XT Gels

Existing Gel	Criterion XT Gel
10–20%, 16.5% peptide/Tricine	10%, 12%, 4–12% Bis-Tris with XT MES buffer
10% Tris-HCl	10% Bis-Tris with XT MOPS buffer
12% Tris-HCl	12% Bis-Tris with XT MOPS buffer
15%, 4–15%, 4–20% Tris-HCl	4–12% Bis-Tris with XT MOPS buffer
5% Tris-HCl	3–8% Tris-acetate with XT Tricine buffer
7.5% Tris-HCl	7% Tris-acetate with XT Tricine buffer

Blotting Criterion XT Gels

Criterion XT gels provide excellent transfer efficiency when blotting with standard Tris/glycine and Towbin buffer in the Criterion blotter. For instructions and tips on blotting Criterion gels, refer to the Criterion Blotter Instruction Manual.

* Criterion XT gels and buffers are formulated at near-neutral pH, resulting in better protein stability (ideal for downstream applications such as mass spectrometry).

Criterion Gel Cassette
The Criterion gel cassette (15.0 x 10.5 cm) with integral buffer chamber runs exclusively on Criterion cells. The Criterion gel is 13.3 cm wide x 8.5 cm long (1.0 mm thick).

Criterion Cell
The Criterion cell runs 1 or 2 Criterion gels. Lower buffer chamber holds 720 ml; upper buffer chamber/gel cassette chamber holds 60 ml.

Criterion™ Dodeca™ Cell
The Criterion Dodeca cell runs up to 12 gels and uses 6 L of buffer.

Criterion Blotter
The Criterion blotter is highly efficient, transferring most proteins in 30–60 min using 1.3 L of buffer. The unique gel blot assembly tray facilitates setup.

Specifications

Gel dimensions	13.3 x 8.7 cm (W x L), 1.0 mm thick	Gel shelf life (from date of manufacture)	12 months for Bis-Tris gels 8 months for Tris-acetate gels 12 weeks for Tris-HCl, Tris-Tricine, zymogram, TBE, TBE-urea gels 26 weeks for IEF gels
Cassette dimensions	10.6 x 15.0 cm (W x L)		
Cassette material	Styrene copolymer		
Comb material	Polycarbonate		
Storage tray material	PET	Buffer volume	Upper, 60 ml; lower, 400 ml
Gel storage conditions	Store flat; do not freeze Ambient temperature for Bis-Tris gels 4°C for all other gel types		

Ordering Information

Description	 12+2 Well*, ** 45 µl	 18-Well 30 µl	 26-Well* 15 µl	 Prep+2 Well 800 µl	 IPG+1 Well** 11 cm IPG Strip
Criterion XT Bis-Tris Gels					
10% Resolving Gel	345-0111	345-0112	345-0113	—	345-0115
12% Resolving Gel	345-0117	345-0118	345-0119	345-0120***	345-0121
4–12% Resolving Gel	345-0123	345-0124	345-0125	345-0126***	345-0127
Criterion XT Tris-Acetate Gels					
7% Resolving Gel	345-0135	345-0136***	345-0137***	—	—
3–8% Resolving Gel	345-0129	345-0130	345-0131	—	345-0133***

Catalog #	Description
Criterion XT Buffers and Reagents	
161-0788	**XT MOPS Running Buffer**, 20x, 500 ml
161-0789	**XT MES Running Buffer**, 20x, 500 ml
161-0790	**XT Tricine Running Buffer**, 20x, 500 ml
161-0791	**XT Sample Buffer**, 4x, 10 ml
161-0792	**XT Reducing Agent**, 20x, 1 ml
161-0793	**XT MOPS Buffer Kit**, includes 500 ml of 20x XT MOPS running buffer, 10 ml of 4x XT sample buffer, 1 ml of 20x XT reducing agent
161-0796	**XT MES Buffer Kit**, includes 500 ml of 20x XT MES running buffer, 10 ml of 4x XT sample buffer, 1 ml of 20x XT reducing agent
161-0797	**XT Tricine Buffer Kit**, includes 500 ml of 20x XT Tricine running buffer, 10 ml of 4x XT sample buffer, 1 ml of 20x XT reducing agent
161-0738	**1x Native Sample Buffer**, 3 ml
161-0734	**10x Tris/Glycine**, 1 L
Criterion Cell and Systems	
165-6001	**Criterion Cell**, includes electrophoresis buffer tank, lid with power cables, 3 sample loading guides (12+2 well, 18-well, 26-well), instructions
165-6024	**Criterion Cell/Plate Blotter System**, includes Criterion cell and Criterion blotter with plate electrodes
165-6025	**Criterion Cell/Wire Blotter System**, includes Criterion cell and Criterion blotter with wire electrodes
Criterion Dodeca Cell	
165-4130	**Criterion Dodeca Cell**, includes electrophoresis buffer tank with built-in cooling coil, lid with power cables, instructions

Catalog #	Description
Criterion Accessories	
345-9921	**Criterion Staining/Blotting Trays**, with lids, 2
345-9920	**Criterion Staining/Blotting Trays**, with lids, 12
Criterion Membrane/Filter Paper Sandwiches	
162-0232	**Nitrocellulose/Filter Paper Sandwiches**, 0.2 µm, 8.5 x 13.5 cm, 20 pack
162-0234	**Nitrocellulose/Filter Paper Sandwiches**, 0.45 µm, 8.5 x 13.5 cm, 20 pack
162-0238	**Immun-Blot PVDF/Filter Paper Sandwiches**, 8.5 x 13.5 cm, 20 pack
162-0236	**Sequi-Blot PVDF/Filter Paper Sandwiches**, 8.5 x 13.5 cm, 20 pack
Bio-Safe Coomassie Stain	
161-0786	**Bio-Safe Coomassie Stain**, 1 L
161-0787	**Bio-Safe Coomassie Stain**, 5 L
Protein Standards	
161-0324	**Kaleidoscope Prestained Standards**, broad range, 500 µl
161-0309	**Prestained SDS-PAGE Standards**, high range, 500 µl
161-0305	**Prestained SDS-PAGE Standards**, low range, 500 µl
161-0318	**Prestained SDS-PAGE Standards**, broad range, 500 µl
161-0363	**Precision Plus Protein Unstained Standards**, 1 ml, 100 applications
161-0373	**Precision Plus Protein All Blue Standards**, 500 µl, 50 applications
161-0303	**SDS-PAGE Standards**, high range, 200 µl
161-0304	**SDS-PAGE Standards**, low range, 200 µl
161-0317	**SDS-PAGE Standards**, broad range, 200 µl

* Multichannel pipet compatible. ** Includes reference well(s). *** Please allow up to 2 weeks for delivery.

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Web site *www.bio-rad.com* ***USA*** *800 4BIORAD* ***Australia*** *61 02 9914 2800* ***Austria*** *01 877 89 01* ***Belgium*** *09 385 55 11* ***Brazil*** *55 21 3237 9400* ***Canada*** *905 712 2771* ***China*** *86 21 6426 0808* ***Czech Republic*** *420 241 430 532* ***Denmark*** *44 52 10 00* ***Finland*** *09 804 22 00* ***France*** *01 47 95 69 65* ***Germany*** *089 318 84 0* ***Greece*** *30 210 777 4396* ***Hong Kong*** *852 2789 3300* ***Hungary*** *36 1 455 8800* ***India*** *91 124 4029300* ***Israel*** *03 963 6050* ***Italy*** *39 02 216091* ***Japan*** *03 5811 6270* ***Korea*** *82 2 3473 4460* ***Mexico*** *52 555 488 7670* ***The Netherlands*** *0318 540666* ***New Zealand*** *0508 805 500* ***Norway*** *23 38 41 30* ***Poland*** *48 22 331 99 99* ***Portugal*** *351 21 472 7700* ***Russia*** *7 495 721 14 04* ***Singapore*** *65 6415 3188* ***South Africa*** *27 861 246 723* ***Spain*** *34 91 590 5200* ***Sweden*** *08 555 12700* ***Switzerland*** *061 717 95 55* ***Taiwan*** *886 2 2578 7189* ***United Kingdom*** *020 8328 2000*

criterion XT™

Timeless Beauty.

With up to a 12-month shelf life, new Criterion™ XT gels give you beautiful results any time.

BIO-RAD

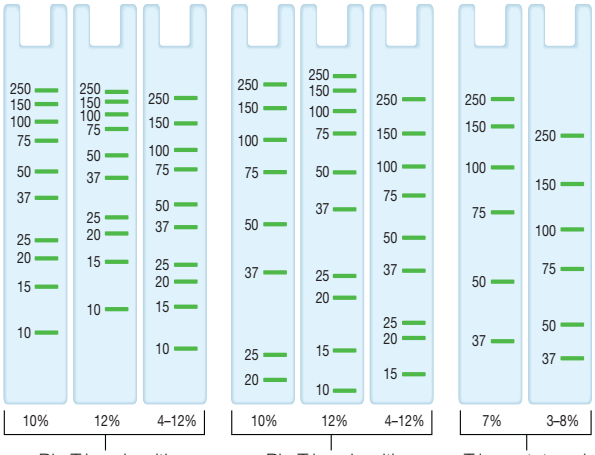
Criterion XT Precast Gels

With eXTended shelf life and room temperature storage!

Criterion XT gels are the highest-quality extended shelf-life gels available for protein electrophoresis with all the benefits of the Criterion system, including an integrated upper buffer chamber for ease-of-use, and gels that run up to 26 samples in an hour.

- Formulated at near-neutral pH to ensure:
  - – Long shelf life (12 months for Bis-Tris, 8 months for Tris-acetate)
  - – Better protein stability — Ideal for downstream applications such as protein sequencing and mass spectrometry
- Optimized sample and running buffers for sharp bands and minimal preparation time
- Choose from 3 buffer systems for flexibility in protein separations (see migration charts at right)

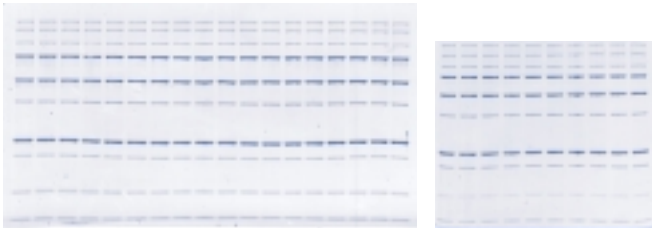
Visit **criterion.bio-rad.com** to schedule a demo.



Bis-Tris gels with XT MES running buffer: ideal for small proteins

Bis-Tris gels with XT MOPS running buffer: ideal for mid-size proteins

Tris-acetate gels with XT Tricine running buffer: ideal for large proteins



Criterion XT Gels Separate More Samples: Demonstrated by Bio-Rad Precision Plus Protein™ standards separated on a Criterion XT 12% Bis-Tris gel (left) and the leading competitor's 12% Bis-Tris gel (right).

SDS-PAGE Gel to Criterion XT Gel Conversion Table

If you use ...	Then go with ...
10–20%, 16.5% peptide/ Tricine gels	10%, 12%, 4–12% Bis-Tris gels with XT MES buffer
10% Tris-HCl gels	10% Bis-Tris gels with XT MOPS buffer
12% Tris-HCl gels	12% Bis-Tris gels with XT MOPS buffer
15%, 4–15%, 4–20% Tris-HCl gels	4–12% Bis-Tris gels with XT MOPS buffer
5% Tris-HCl gels	3–8% Tris-acetate gels with XT Tricine buffer
7.5% Tris-HCl gels	7% Tris-acetate gels with XT Tricine buffer

Ordering Information

Description	12+2 Well Comb* 45 µl Samples	18-Well Comb 30 µl Samples	26-Well Comb* 15 µl Samples	Prep+2 Well Comb 800 µl Samples	IPG+1 Well Comb 11 cm IPG Strip	IPG-Well Comb 11 cm IPG Strip
Criterion XT Bis-Tris Gels						
10% Resolving Gel	345-0111	345-0112	345-0113	345-0114	345-0115	345-0116
12% Resolving Gel	345-0117	345-0118	345-0119	345-0120	345-0121	345-0122
4–12% Resolving Gel	345-0123	345-0124	345-0125	345-0126	345-0127	345-0128
Criterion XT Tris-Acetate Gels						
3–8% Resolving Gel	345-0129	345-0130	345-0131	345-0132	345-0133	345-0134
7% Resolving Gel	345-0135	345-0136	345-0137	345-0138	345-0139	345-0140

* Multichannel pipet compatible.

Criterion Cell

165-6001 Criterion Cell, includes tank, lid with power cables, sample loading guides and instructions

Criterion XT Buffers and Reagents

161-0788 XT MOPS Running Buffer, 20x, 500 ml
161-0789 XT MES Running Buffer, 20x, 500 ml
161-0790 XT Tricine Running Buffer, 20x, 500 ml
161-0791 XT Sample Buffer, 4x, 10 ml
161-0792 XT Reducing Agent, 1 ml

161-0793 XT MOPS Buffer Kit, includes 500 ml 20x XT MOPS running buffer, 10 ml 4x sample buffer, 1 ml XT reducing agent
161-0796 XT MES Buffer Kit, includes 500 ml 20x XT MES running buffer, 10 ml 4x sample buffer, 1 ml XT reducing agent
161-0797 XT Tricine Buffer Kit, includes 500 ml 20x XT Tricine running buffer, 10 ml 4x sample buffer, 1 ml XT reducing agent

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Bulletin 2912 US/EG Rev A *02-747 1202 Sig 1102-1*

SSI5 SSI5-2 SSI5R SSI5R-2
Orbital Shaking Incubator
Operational Manual

Models:
SSI5 SSI5-2
SSI5R SSI5R-2
Previously designated as
SI6 SI6-2
SI6R SI6R-2
4861534
1/2016

Table of Contents

Table of Contents

These units are TUV CUE listed as orbital shaking incubators for professional, industrial, or educational use where the preparation or testing of materials is done at approximately atmospheric pressure and no flammable, volatile, or combustible materials are being heated.

These units have been tested to the following requirements:

CAN/CSA C22.2 No. 61010-1:2012
CAN/CSA C22.2 No. 61010-2-010 + R:2009
UL 61010-1:2004 + R:2005-07 + R:2008-10
UL 61010A-2-010:2002
UL 61010-1:2012
EN 61010-1:2010
EN 61010-2-010:2003

Using the Unit Safely

Introduction

Thank you for choosing a SHEL LAB shaking orbital incubator. SHEL LAB sets the standard for quality and reliability. Your unit is backed by over 30 years of design and manufacturing excellence in the scientific, research, and medical equipment industries.

Your unit is a general-purpose incubator designed for professional, industrial or educational use where

- the preparation or testing of materials is done at approximately atmospheric pressure, and
- no flammable, volatile or combustible materials are being heated.

These units are not intended for use at hazardous or household locations.

Before you use the unit, read this entire manual carefully to understand how to install, operate, and maintain the unit in a safe manner. Your satisfaction with the unit will be maximized as you read about its safety and operational features. Keep this manual on-hand so it can be used by all operators of the unit. Be sure all operators of the unit are given appropriate training before you put the unit in service.

Use the unit only in the way described in this manual. Failure to follow the guidelines and instructions in this manual may be dangerous and illegal.

General Safety Considerations

Your incubator and its recommended accessories have been designed and tested to meet strict safety requirements.

For continued safe operation of your incubator, always follow basic safety precautions including:

- Read this entire manual before using the incubator.
- Be sure you follow any city, county, or other ordinances in your area regarding the use of this unit.
- Use only approved accessories. Do not modify system components. Any alterations or modifications to your incubator may be dangerous and will void your warranty.
- Always plug the unit's power cord into a grounded electrical outlet that conforms to national and local electrical codes. If the unit is not grounded, parts such as knobs and controls may conduct electricity and cause serious injury.
- Do not connect the unit to a power source of any other voltage or frequency beyond the range stated on the power rating overlay at the rear of the unit.
- Do not modify the power cord provided with the unit. If the plug does not fit an outlet, have a proper outlet installed by a qualified electrician.
- Avoid damaging the power cord. Do not bend it excessively, step on it, place heavy objects on it. A damaged cord can easily become a shock or fire hazard. Never use a power cord after it has become damaged.

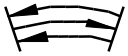
Precautions for Your Unit

Observe the following additional safety guidelines for your unit.

- **Operating Conditions** For optimum performance, use your incubator at room temperatures between 18 and 25°C, at no greater than 80% relative humidity (at 25°C). If you intend to operate the unit in conditions outside of these limits, contact customer service.
- **Installing the Unit** Installation of the unit can be performed by the end user
- **Lifting and Handling** The incubator is heavy and should be moved by a lifting device, such as pallet jack. If you must lift the device by hand, always observe the following guidelines:
    - Do not move the incubator while it is plugged into the power source.
    - Remove all moving parts, such as shelves and trays, before you move the unit. Make sure the door is securely shut.
    - Use at least four people to lift the incubator.
    - Lift the unit from its bottom surface only.
    - Do not use doors, handles or knobs to lift or stabilize the unit.
    - Keep the unit from tipping.
- **Servicing Your Unit** Only qualified personnel should service your unit. Faulty service may be dangerous and will invalidate the unit's warranty. Do not operate the unit if any parts are damaged or missing.
- **Maintenance** Unplug the unit from its power source before attempting any maintenance.

Meanings of Symbols

In this manual and on labels attached to the product, graphic symbols have the following meanings:

Symbol	Indcation
	You should consult this manual for a description or discussion of a control or item
	Temperature
	Over Temperature Safety
°C	Degrees Centigrade
	AC Power
	Manual Adjustable Components
	Oscillator
	Timer
	Light
	Indicates "**Unit should be recycled"** (Not disposed of in land-fill)

About this Manual

Throughout this manual, the words WARNING and CAUTION have the following meanings:

WARNING

A potentially hazardous situation that, if not avoided, could result in serious injury or death.

CAUTION

A potentially hazardous situation that, if not avoided, may result in minor or moderate injury or damage to the equipment.

Features of Your Unit

Product description

Your shaking orbital incubator provides:

- **Controlled environment** For continuous growth of biological organisms.
- **Vibration-free operation** A unique adjustable counterbalance system provides vibration-free operation regardless of load.
- **Large chamber** A large six cubic foot chamber facilitates throughput. Shelves in the top provide space for static incubation during shaking sessions.
- **Refrigeration** The SSI5R and SSI5R-2 (SI6R SI6R-2) are refrigerated, which supports insect cell culture and entomology studies.
- **Load Flexibility** Our unique counterbalance weighting system is adjustable to accommodate off-center loads, varying capacities and stroke lengths, which in turn allows smoother running.
- **Oxygen transfer** An adjustable orbit provides maximum oxygen transfer and offers three circular/stroke sizes, from vigorous to gentle, to accommodate different types of cells.
- **Sample protection** All major functions—temperature, RPM, and time—have audio and visual alarms that immediately alert you to deviations from set parameters.
- **Over-temperature protection** Provided by a safety thermostat that is independent of the main temperature controller. Guards your samples from inadvertent overheating.

Key Features

- A brushless DC motor offers quiet and maintenance-free orbital shaking motion.
- A PID microprocessor controller provides precise uniformity.
- The rotation platform is included with each unit and is self-centering for easy installation.
- Large LED displays are easy to read.
- Digital keypad operation allows calibration of the main temperature controller to a reference thermometer.
- A fluorescent light allows you to see all that's going on.
- An interior electrical outlet and a one-inch hermetically sealed, double-paned glass viewing window.
- Unit exteriors are formed of cold-rolled steel finished with corrosion resistant powder coat paint.
- Chamber interiors and shelves are made of polished stainless steel, which provides excellent durability and an easy-to-clean surface.
- An interlock switch stops the shaking mechanism if the door is opened.
- The SSI5R and SSI5R-2 (SI6R SI6R-2) refrigerate using a 1/6-horsepower motorized compressor that does not use CFCs or HCFC's.

Receiving Your Unit

Unpacking and Inspecting Your Unit

Before leaving our factory, all units are packaged in high quality shipping materials designed to provide protection from transportation related damage.

Once a unit leaves our factory, however, safe delivery becomes the responsibility of the carrier who is liable for loss or damage to your unit. Damage sustained during transit is not covered under your unit warranty.

When you receive your unit, inspect it for concealed loss or damage to its interior and exterior. Should you find any damage to the unit, follow the carrier's procedure for claiming damage or loss.

Inspection Guidelines

- Carefully inspect the package for damage. If the package is damaged, report the damage to the carrier service that delivered the unit.
- If the carton is not damaged, open the crate and remove its contents. Verify that all of the following equipment is included with the unit:
    - 1 shelf 5100531
    - 1 sample tray 9750758
    - 4 shelf clips 1250512
    - 6 counter weights (some of which are located in the metal pocket at the back of the unit)
    - 4 leveling feet
- Carefully check all packaging before discarding.

Save the unit's shipping crate until you are sure all is well. If you need to return your unit for any reason, see "Getting Your Unit Serviced" on page 20.

Recording Data Plate Information

Once you have determined the unit is free from damage, locate the data plate at the back of the unit.
The data plate indicates your unit's model number and serial number. Record this information on the space provided on page 20, "Getting Your Unit Serviced" for easy future reference.

Installing the Unit

Installation Overview

To install your unit, you need to:

1. Select a suitable operating location for the unit.
2. Level the unit.
3. Sterilize the unit.
4. Install the sample tray.
5. Plug the unit into a power source.

Selecting a Location for the Unit

The operating location of your unit has a significant impact on your unit's performance and how often it must be cleaned and disinfected. Use the following guidelines to select the best location for your unit.

CAUTION

DO NOT MOUNT YOU UNIT TO A FLAMMABLE SURFACE.

- **Operating Conditions** For optimum performance, use your incubator at room temperatures between 18 to 25°C (65 to 77 °F) and at no greater than 80% relative humidity (at 25°C).

  If you intend to operate the unit in conditions outside of these limits, contact your customer service representative.

- **Exposure** Avoid exposing the unit to the following:
  - Direct sun
  - High air movement, such as air vents, heating and cooling ducts, doors and other heavy traffic areas.
  - Extreme heat from steam radiators, stoves, ovens, autoclaves, or other sources of heat.
- **Level Surface** The unit must be located on a solid, flat and level surface.
- **Space requirements** Allow a minimum of 20 cm (8 in.) between the rear and sides of the unit, and any walls or partitions that can obstruct free airflow. Allow enough room so that the door can swing open at least 90 degrees. **Do not block access to the power cord, circuit breaker or fuses.**
- **Cleanliness** Good laboratory quality control practice is the most efficient and reliable way to keep your unit free from contamination.

  If it is important that the interior of your unit remain sterile, always pay attention to the following guidelines:
  - Keep the air in the laboratory as clean as possible.
  - Keep the floor around the unit clean.
  - If the unit must be placed at the floor level, use a platform, such as a caster platform. This facilitates movement of the unit during cleaning and allows for easier access to the back of the unit.
  - Minimize the number of times access is made to the chamber during normal operation. Each time the door is opened, room air is drawn in and can lead to contamination of the unit.

After deciding on the location for your unit, follow the installation instructions below.

Leveling the Unit

The unit must sit level from side to side and from front to back. While the unit does not have to be absolutely level, each of the four feet should be in firm contact with the surface on which the incubator is to be run.

Install the four leveling feet in the four holes in the bottom of the unit. When the feet are installed, you can raise or lower a corner of the unit by turning its foot clockwise or counterclockwise, respectively.

To level the incubator

1. Insert a leveling foot into each of the four holes at the bottom of the unit.
2. Adjust the foot at each corner until the unit stands level and solid without rocking.

If you move the incubator to a different location, be sure to re-level the incubator at the new location.

Sterilizing Your Unit

The interior of your incubator was cleaned at the factory but is not sterile. For information on

sterilizing your unit, see “Disinfecting Your Unit“ on page 14.

Installing Sample Tray

Your unit comes with a sample tray as standard equipment.

To install the sample tray

1. Enclose all corners of the shaking mechanism within the lips of the sample tray. This can be done easily by positioning the front two corners and then setting the rest of the tray down.
2. Shake the tray by its handles to confirm that it is firmly in place.

Plugging the Unit into a Power Source

We recommend that you plug your incubator into a circuit separate from other equipment. This prevents damage or destruction of the incubator caused by overloading or failure of other equipment on the same circuit.

The electrical supply circuit to the incubator must conform to all national and local electrical codes. The voltage supplied to your unit should not vary more than 10%.

WARNING

For your own safety, do not plug the unit into a power source until you have read and understood the safety and operational instructions in this manual.

To connect the unit to a power source

1. Be sure the plug and the cord are in good condition. If the power cord is worn, cut or damaged in any way, do not use it. Contact customer service for a replacement power cord. For information on contacting customer service, see page 20.
2. Plug the service cord firmly into a grounded electrical outlet. If the plug does not fit the outlet, have a proper outlet installed by a qualified electrician.

Operating the Unit

Control Panel Overview

Before turning the incubator on for the first time, take a moment to familiarize yourself with its controls and features. Following is an overview of the control panel.

1. Main temperature control
   - Displays current chamber temperature.
   - Controls temperature set point and calibration.
2. Shaker Speed (RPM) Control
   - Displays shaker platform speed.
   - Controls the rotational speed (RPM) of the shaker mechanism.
3. Oscillation timer
   - Permits timed shaking at a preset RPM.
4. Over Temperature Protection
   - Provides backup protection for the main temperature control.
   - Keeps the chamber temperature from inadvertently rising above the set point.
5. Alarms
   - Error status lights and an audible alarm immediately alerts you to deviations of temperature, RPM, or time.
6. RPM Switch
   - Activates and deactivates the shaker platform.
7. Light Switch
   - Controls the fluorescent light inside the chamber.
8. Timer Switch
   - Activates and deactivates the timer.
9. Power Switch
   - Controls all power to the unit. The switch is lit by a green light when the power is on.

Getting the Unit Ready for Use

WARNING

This equipment is NOT intended for the processing of Flammable materials.

Use the following guidelines to prepare the unit for regular use. The guidelines illustrate how to use all the features of your incubator. Your laboratory protocol will determine your actual use of these features.

1. Turn the unit on.
   See "Turning the Unit On" below on this page.
2. Set the chamber to the desired temperature and wait for the chamber temperature to stabilize.
   See "Setting the Chamber Temperature" on page 11.
3. Calibrate the main temperature control.

At any time, use the following features when appropriate.

- Turn the shaking mechanism on and adjust the speed of the shaking mechanism.
  See on page 11.
- Set the Over Temperature Protection (OTP) to guard your samples from inadvertent over-heating.
  See on page 12.
- To account for the weight of different sample loads, you will need to adjust the number of counterweights being used.
  See on page 12.
- To adjust the movement of the shaking mechanism from vigorous to gentle, you will need to adjust the shaking stroke and counterweight position.
  See on page 13.

Turning the Unit On

The unit is equipped with an On/OFF switch that controls power to the entire unit. The switch is lit by a green light when the power is on.

To turn the unit on

1. Be sure the unit is plugged in.
2. Push the *Power* switch to the On (I) position.

3. When you turn the unit on for the first time, use a screwdriver or coin to turn the *Safety Temp* knob fully clockwise to its maximum position. This deactivates the Over-Temperature Protection (OTP) feature. For more information on the OTP, see on page 12.

Setting the Chamber Temperature

You raise or lower the temperature in the chamber using the main temperature controller, which consists of a digital display and UP and DOWN arrow pads marked *Set Temp*.

To set the chamber temperature

- To set temperature, press and release either up or down key and display will blink. Then, press and hold either up or down key to scroll up or down for set point.

  When you press either the Up or Down arrow key, the display starts to blink from bright to dim and shows the temperature set point, which is the temperature to which the unit will stabilize.

  The incubator accepts the new set point after you release the arrow pads for 5 seconds. At that time, the display stops blinking and indicates the present chamber temperature.

After setting the chamber temperature, wait at least 1 hour for the chamber temperature to stabilize to ambient conditions. To achieve maximum temperature stability, wait 24 hours before you begin using the unit.

Calibrating the Main Temperature Control

Calibrating your unit ensures that the temperature inside the incubator matches the temperature reading of a certified reference thermometer.

We recommend that you calibrate your unit once it has been installed in its working environment and the chamber temperature has been stable at the set point for several hours.

You should calibrate your unit at or as close to the temperature set point as possible. To maximize your results, calibrate the unit each time you operate the unit at a new temperature.

Use only a Certified (NIST) temperature-measuring device to calibrate your unit.

To verify that your unit needs calibration

1. Be sure the temperature within the chamber has stabilized at the set point for several hours.
2. Insert a certified reference thermometer through the access porthole. To attain the best calibration, place the thermometer as close to the location of the samples. Be sure the thermometer is not touching any shelving.
3. Allow the reference thermometer to stabilize until it displays a constant value for one hour.
4. Compare the temperatures displayed by the incubator and reference thermometer.

If they match, you do not need to calibrate your unit for that temperature. If they do not match, you need to calibrate your unit.

To calibrate your unit

1. Simultaneously press and hold the *Set Temp* Up and Down arrow keys.

After approximately 5 seconds, the temperature reading will blink off and on. Release the Up and Down arrow key.

2. While the display is blinking, press the Up or Down arrow keys to select the temperature that matches your reference thermometer. When you hold an arrow key, the display scrolls through the temperature settings.

The incubator accepts the new temperature reading after you release the arrow pads for 5 seconds. At that time, the display stops blinking.

3. For best results, re-verify the calibration after the unit has remained on for 24 hours and its temperature has varied by no more than + 1 °C.

Setting the Shaker Speed

Your unit is equipped with a shaker mechanism that provides maximum oxygenation of your samples.

You control the shaking mechanism using the shaker control—which consists of a digital display that shows RPM (rotations per minute) in increments of 1 and UP/DOWN arrow pads marked *Set RPM*—and the *RPM* switch.

To turn the shaking mechanism on

1. Be sure the door is completely closed.
2. Push the *RPM* switch to the On (I) position.

   The shaker mechanism will increase the speed up to the current set point, which is the speed at which the unit will rotate per minute. Note that the shaker motor runs continuously as long the RPM switch is On (I).

To adjust the shaker speed (RPM)

1. Press either the Up or Down arrow key once. The display starts to blink from bright to dim and shows the RPM set point.
2. Press the Up or Down arrow keys to select the desired RPM.

   The incubator accepts the new set point after you release the arrow pads for 5 seconds. At that time, the display stops blinking and indicates the present RPM.

   Even if you turn the RPM switch off (O), the controller remembers the last RPM value used.

   You can adjust the movement of the shaking mechanism. See page 13.

Using the Timer

Using the incubator's timer, you can run the shaker platform at a preset RPM for a preset time. The timer can be set at intervals of one (1) minute up to a maximum of a 999 (16 hours 39 minutes).

Upon completion of the timing cycle, the TIMER alarm LIGHT will turn ON and an alarm will sound.

You can interrupt the timer if you need to access the shaking platform before the timer completes.

To start a timed shaking process

1. Turn the TIMER switch to the ON (I) position.
2. Press the set timer up or down arrow key once. The display starts to blink off and on and shows the current set time. Press the up or down arrow to select the desired time.

Approximately five seconds after you release the Up or Down arrow keys, the display stops flashing and the timing interval begins.

To interrupt a timed shaking process

- Turn the TIMER switch and the RPM switch to the OFF (O) position.

CAUTION

Wait for the mechanism to come to a complete stop before entering the chamber.

To restart an interrupted timed shaking process

- Turn the RPM and TIMER switch to the ON (I) position.

Setting the Safety Temperature Alarm

You can prevent the chamber temperature from inadvertent over-heating by using the unit's Over-Temperature Protection (OTP), which consists of:

- a thermostat independent of the main temperature control.
- a knob, marked *Safety Temp*, to set the safety temperature threshold. The numbered scale around the knob is for reference only and does not correspond to any temperature points.
- an alarm, marked *Temp*, that sounds if the temperature exceeds the user-defined temperature threshold.

To set the safety temperature thermostat

1. For best results, calibrate your unit before you set the safety thermostat.

See "Calibrating the Main Temperature Control" on page 11.

2. Be sure the temperature within the chamber has stabilized at the set point for several hours.
3. Using a screw driver or a small coin, turn the *Safety Temp* knob counterclockwise until the *Temp* alarm light turns on and off, which designates that your OTP has been activated. The light will cycle on and off as the element is trying to energize on and off.
4. Turn the *Safety Temp* knob slightly clockwise until the *Temp* alarm light turns off.

   NOTE: Temp Alarm will only sound off when temp overshoot 1°C from setpoint.

The OTP is now set at approximately 1°C above the main temperature set point. If, for any reason, the chamber temperature rises to the safety thermostat setting, the *Temp* alarm will go off and the heating element will not raise the chamber temperature any further.

Adding or Removing Counterweights

To allow the smoothest operation of the shaker, you should adjust the number of counterweights used based on the weight of the load.

To add or remove counterweights

1. Unplug the unit from its power source. When the shaker mechanism comes to a complete halt, remove the sample tray.
2. Rotate the counterweight platform until the counterweight appears. Remove the wing nuts and add or remove counterweights according to the total weight of your samples, as shown below.

Total Sample Weight	Number of Counterweights
Up to 2.3 kg (5 lbs.)	2
Up to 4.5 kg (10 lbs.)	3
Up to 6.8 kg (15 lbs.)	4
Up to 9 kg (20 lbs.)	5
Up to 11.3 kg (25 lbs.)	6

3. Replace the wingnuts and sample tray.

Adjusting the Shaker Movement

You can adjust the shaker movement to gentle, moderate, or vigorous shaking. Which shaking movement you use depends on the oxygenation needs and cell strength of your samples.

When you change the stroke of the shaker mechanism, you also need to adjust the counterbalance position.

WARNING

Always disconnect the unit from its power supply before attempting this procedure. Serious injury can result if the drive plate operates accidentally.

To adjust the shaker movement

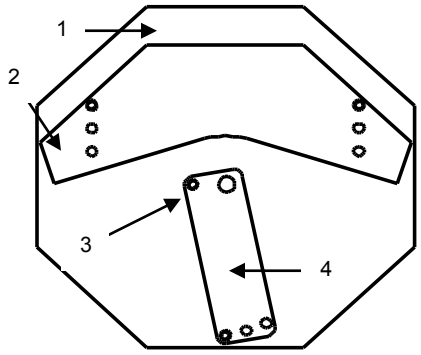
1. Unplug the unit from its power source. Remove the sample tray.
2. Rotate the counterweight platform until the stoke adjuster is in full view. Remove the wing nut and adjust the arm to any of the available options.
3. Re-add the wing nut.
4. Rotate the counterweight platform until the counterweights are in full view. Remove the wingnuts and adjust the counterweights according to the diagrams shown below.
5. Replace the counterweight wingnuts.

The following diagrams show the various positions of the shaker mechanism and counterweights. The dimensions shown are the total stroke of the oscillator.

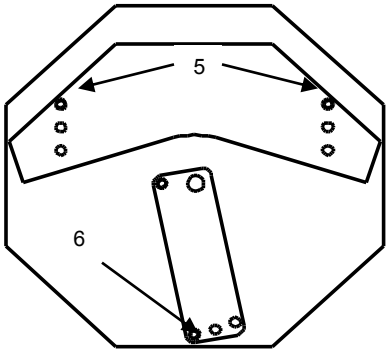
For example, ½ designates a pattern that is + ¼ inch from center. The indicated parts on the mechanism are:

1. Drive plate
2. Counterweight
3. Pivot nut: Do not adjust.
4. Stroke adjuster
5. Counterweight studs: secure using wingnuts.
6. Stroke adjuster bolt: secure using wingnut.

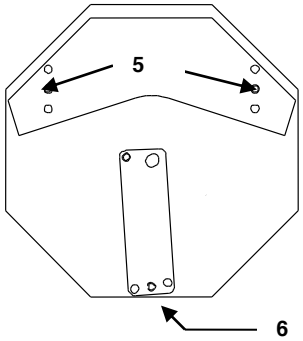
Oscillation Plate Overview ½" (1.3 cm) stroke setup -- light shaking ¾" (1.9 cm) stroke setup -- moderate shaking 1" (2.5 cm) stroke setup -- vigorous shaking



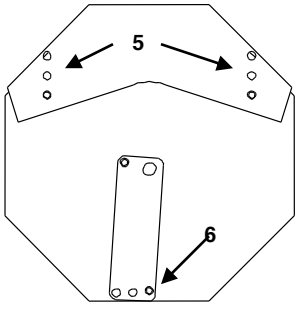
Oscillation Plate Overview



½" (1.3 cm) Stroke Setup – Light Shaking
(left-hand counterweight stud)



¾" (1.9 cm) Stroke Setup – Moderate Shaking
(middle counterweight stud)



1" 2/5 cm) Stroke Setup – Vigorous Shaking
(right-hand counterweight stud)

Interior Accessory Outlet

This unit features an accessory outlet to provide power for equipment such as magnetic stirrers, rockers, etc. The weight of this equipment should not exceed 22 pounds (10 kg) per shelf. This equipment may provide additional heat that could affect the temperature range of this incubator. We recommend testing the incubator and any accessory equipment to ensure that the desired operating conditions can be met.

Caution: This incubator operates at conditions that might damage accessory equipment. Verify that your accessory equipment is capable of operating under the same conditions as the incubator.

The outlet is located inside the incubator in the upper right rear of the chamber. The voltage available at the accessory outlet is the SAME as the voltage supplied to the incubator. For example, a 120-vac incubator will have 120 vac at the accessory outlet, and a 240-vac incubator will have 240 vac at the accessory outlet. DO NOT exceed 500 va rating of the accessory outlet.

Maintaining the Unit

The only regular maintenance required for your unit is to keep it clean and free from contamination. Use the guidelines and instructions in this section to maximize the life of your incubator and help prevent contamination of your samples.

WARNING

Do NOT Use Flammable Cleaning Detergents.

Do NOT store Flammable materials In, On or Near this equipment.

Disinfecting Your Unit

Although your operating conditions and related protocol should determine the actual decontamination procedures you use, always keep the following guidelines in mind when decontaminating your unit:

- **Use cleaning materials known to be compatible with your unit. If any questions arise about compatibility issues, contact Customer Service, see page-21.**
- Clean and disinfect the incubator interior on a regular basis. If the inside of your incubator smells strangely or contains rust, mold, or dirt, you need to clean your incubator more frequently
- Dust the outside walls of the incubator at least every two months.
- For incubators placed on the floor, move the incubator every two months to clean and disinfect the floor below.
- Clean all gaskets and hinges every month.
- **Do not use chlorine-based bleaches or cleaners** with abrasives as they will corrode and pit the interior of you incubator and any other stainless steel surfaces. Use only non-abrasive cleaners.
- Do not use spray cleaners that might leak through openings and cracks and get on electrical parts. These cleaners may also contain solvents that will harm the coatings.
- Do not use hard tools such as metal wire brushes or steel wool. Use only soft tools such as plastic brushes.
- Do not depend on the use of antibiotics to maintain completely sterile conditions, as this is an inadequate technique for sterilization. Instead, use the aseptic techniques described in this section to maintain sterile conditions in the incubator.
- You can use an autoclave to decontaminate stainless steel parts by following the manufacturer's instructions.

  Otherwise, wash all parts and surfaces with soap and water to remove any organic material. Disinfect the parts with a 70% alcohol solution. Rinse with distilled water and wipe dry with a soft cloth.

A Typical Decontamination Procedure

Following is decontamination procedure that will suit most situations.

WARNING

Regardless of which decontamination procedure you follow, always turn the unit off and disconnect the service cord from its power supply.

Before you reattach the unit to its power supply, be sure all cleaners are evaporated and dry.

To decontaminate the unit

1. Unplug the unit from its power source.
2. Remove all interior parts, including shelves and shelf clips.
3. Remove all gaskets and hinges. Clean and disinfect all mounting grooves for the door gaskets.
4. Clean and disinfect all rubber or plastic tubing, as well as the fan and fan housing.
5. Clean and disinfect all access ports, shaft holes, electrical pass-throughs and any other passages into the chamber.
6. Wash and disinfect all interior surfaces.
7. Let the chamber dry out fully before replacing the removed parts or reattaching the unit to a power supply.

Control Maintenance

The main temperature controller, over-temperature protection thermostat and main temperature probe do not require any maintenance. If the unit appears to be having trouble maintaining a temperature, see "Troubleshooting" on page 15.

Troubleshooting

Solving Problems

Should the proper function of your unit come into question, use this section to help you determine what the problem is and how to fix it.

Check if your question is similar to those listed below. Then follow the guidelines found in that section:

- The temperature control inside the unit does not appear to be working correctly. What's wrong?
- The refrigeration of my SSI5R (SI6R) does not appear to be working correctly. What's wrong?

WARNING

Replacing fuses with wrong type and value can result in serious damage to the equipment and property. ONLY Replace fuses with the same type and amperage indicated on the replacement fuse labels.

There are no user serviceable components inside the unit. Potentially lethal voltages exist. Do not attempt to service your unit beyond the procedures described here. See "Getting Your Unit Serviced on page-21"

Temperature

The temperature inside the unit is difficult to control. What's wrong?

What is the problem?	Possible Causes	To solve the problem...
The temperature indicated by the Main Temperature Control is higher than my reference thermometer.	• Controller is calibrated too high.	1. Calibrate the Main Temperature Controller. See page 11. 2. Call customer service. See page 21.
Display reads "HI" or "400+".	• Probe is unplugged *- Or -* • Wire to the probe is broken. *- Or -* • Probe is plugged in backwards.	1. Be sure the temperature probe is properly plugged in. *If this doesn't solve the problem...* 2. Call customer service. See page 21
Chamber temperature spikes over the set point.	• Unit is not calibrated properly.	1. Calibrate the Main Temperature Controller. See page 11.
The temperature indicated by the Main Temperature Control is lower than my reference thermometer.	• The temperature inside the unit has not yet stabilized after the door has been opened. *- Or -* • The temperature inside the unit has not yet stabilized after the unit has been turned off or a power failure. *- Or -* • Controller is calibrated too low. *- Or -*	1. Wait for the temperature indicated by the Main Temperature Controller to stabilize. If you have just turned the unit on, wait 24 hours for the incubator to stabilize at a warmer temperature. A fluctuation of no more than + 0.1 °C is normal. *If this is not the problem...* 2. Recalibrate the Main Temperature Controller. See page 11.

What is the problem?	Possible Causes	To solve the problem...
	• Over Temperature Protection (OTP) is set too low. *- Or -* • Heating element failure.	*If this doesn't solve the problem...* 3. Be sure your reference thermometer is certified. *If this is not the problem...* 4. Turn the OTP fully clockwise. *If this doesn't solve the problem...* Call customer service. See page 21
The Main Temperature Control displays "LO".	• Probe has shorted out.	Call customer service. See page 21
The unit will not heat up to set point.	• The amperage and voltage of the unit's power source do not match the unit's requirements. *- Or -* • Over Temperature Protection (OTP) is set too low.	1. Make sure the power source matches the data plate. (ie. voltage, hz, etc.) *If this does not solve the problem...* 2. Turn the OTP clockwise until the heating light or safety light turns on.
The unit will not heat at all.	• The OTP is not set high enough. *- Or -* • Temperature Controller failure. *- Or -* • Element failure.	1. For diagnostics purposes, turn the OTP fully clockwise. See OTP section. Call customer service. See page 21
The indicated temperature inside the chamber is fluctuating more than + 0.1 °C.	• The unit has not had time to stabilize to ambient conditions. *- Or -* • Temperature sensor not positioned properly. *- Or -* • The temperature sensor is faulty. *- Or -* • Electrical noise	1. If you have just turned the unit on, wait 24 hours for the incubator to stabilize at a warmer temperature. *If this is not the problem...* 2. If you have just opened the unit's door, wait for the temperature to stabilize. *If this is not the problem...* 3. Stabilize ambient conditions. *If this is not the problem...* 4. Call customer service. See page 21
Cannot adjust set points or calibration.	• This is a controller failure.	1. Turn entire unit off and then on to reset the unit. This may temporarily solve the problem, but controller may be faulty. *If this does not solve the problem...* 2. Call customer service. See page 21

What is the problem?	Possible Causes	To solve the problem...
Refrigeration (SSI5R models only) (SI6R)		
The unit will not cool.	• The evaperator has too much ice built up on it. *- Or -* • The unit is not calibrated correctly. *- Or -* • There is not enough space between the unit and adjacent walls or partitions. *- Or -* • The door seal does not work properly.	1. For diagnostics purposes, turn the OTP fully clockwise. See OTP section. *If this does not solve the problem...* 2. Recalibrate the Main Temperature Controller. See " " on page . *If this does not solve the problem...* 3. Be sure there is 5 cm (2 in.) of space between the rear and sides of the unit, and any walls or partitions that can obstruct free airflow.
Ice built up in the chamber.	• The door gasket leaks. • The door is opened too often. • There's an open container letting moisture collect inside the chamber.	1. Check door seal. 2. Try to limit door opening/closing. 3. Seal the container.
Power		
The unit will not turn on.	• Power cord not firmly plugged into the outlet. *- Or -* • The unit or wall fuse/circuit breaker has blown. *- Or -* • The outlet is defective. *- Or -* • The unit is plugged into a circuit that already has too many electrical loads.	1. Be sure the voltage and frequency specifications of the outlet are within the range stated on the power rating overlay at the rear of the unit. *If this does not solve the problem...* 2. Check the power cord at the electrical outlet for proper fit. 3. Make sure the unit is plugged in firmly. *If this does not solve the problem...* 4. Replace fuse/circuit breaker in the unit or wall as necessary. *If this does not solve the problem...* 5. Make sure the outlet is in proper working condition. 6. Replace if defective. *If this does not solve the problem...* 7. Check to see what other loads are on the same circuit as the unit. We recommend that you plug your incubator into a circuit separate from other equipment. 8. Call customer service. See page 21
The unit fuse/circuit breaker blows often.	• Wrong fuse installed. • Wire is shorting out.	1. Check fuse for right amperage. 2. Call customer service. See page 21

What is the problem?	Possible Causes	To solve the problem...
The wall fuse/circuit break blows often.	• Too many things may be plugged in.	1. Check to see what other loads are on the same circuit as the unit. We recommend that you plug your incubator into a circuit separate from other equipment.
The front panel displays fail to turn on but the rest of the unit receives power.	• Controller failure	1. Call customer service. See page 21
The Main Temperature Controller is locked up.	• Controller failure	1. Turn entire unit off and then on to reset the unit. This may correct the problem, but the controller may still be faulty. 2. Call customer service.
Mechanical		
The door is not sealing.	• The door gasket does not function properly. *- Or -* • The door latch bolts are not tightened enough. *- Or -* • The hinges are not adjusted properly. *- Or -* • The door has been twisted. *- Or -* • The unit has been damaged and the body is not square.	1. Check the gasket visually. Make sure it's secure and smooth and free from rolls or tears, which would interfere with the magnetic seal. *If this does not solve the problem...* 2. Tighten the door latch bolts with a screwdriver. *If this does not solve the problem...* 3. To tighten hinges, use wrench to adjust and to check if the bolts are tight. *If this does not solve the problem...* 4. Call customer service. See page 21
The shaker motor squeals continuously with a constant pitch. Changes in intensity only when rpm varies. Stops when the oscillate switch is turned off. Sound appears to be coming directly from the motor, not the mechanism.	• Motor cable plugs not seated properly. *- Or -* • May have motor bearing failure..	1. May need to replace motor. Perform a visual inspection on motor to decide. *If this does not solve the problem...* 2. Call customer service. See page 21
Contamination		
The chamber is contaminated.	• Your unit is not cleaned and decontaminated often enough. *- Or -* • If your unit becomes contaminated even after you follow an appropriate maintenance regimen, the source of the contamination is probably not the incubator.	1. See "Maintaining the Unit" on page 14 for recommendations and instructions on decontaminating your unit. *If this does not solve the problem...* 2. There are many sources of contamination, such as water baths, hoods, autoclaves, reagents, disposables, incubators and personnel. If your unit becomes contaminated even after you follow an appropriate maintenance regimen, the source of the contamination is probably not the incubator. 3. Call customer service.

Getting Your Unit Serviced

Getting Assistance

Your incubator will provide years of trouble-free operation. Should you require assistance, however, SHEL LAB's customer service and support is available to assist you.

If your unit is still covered under warranty, repair or replacement will be made at no cost to you according to the warranty given at the back of this manual. If the warranty for your unit has expired, you can still return the unit for repair. If the unit proves to be beyond repair, we will promptly inform you of its condition and, if you want, return the unit to you.

Obtaining Nameplate Information

Before you contact customer service, obtain the following information about your unit from the data plate at the back of the unit. Use the spaces below to record the information.

- _____
Model Number

- _____
Serial Number

Returning Your Unit

If you need to return your unit for any reason, first contact your customer representative for return authorization number (RA#). Be sure to print the RA# clearly on the package in which you ship your unit.

No return is accepted without:

- prior authorization by SHEL LABS
- a clearly visible RA# on the package.

SHEL LAB Contact Information

Please allow at least 24 hours from the time that you contact our service manager for service to be scheduled.

Contact Information

Sheldon Manufacturing Inc.
P.O. Box 627
Cornelius, Oregon 97113
Phone: (503) 640-3000
Toll free: 1-800-322-4897
Fax: (503) 640-1366

Email:**tech@Shellab.com**
Internet:http://www.Shellab.com/~Shellab

Replacement Parts and Accessories

Replacement Parts

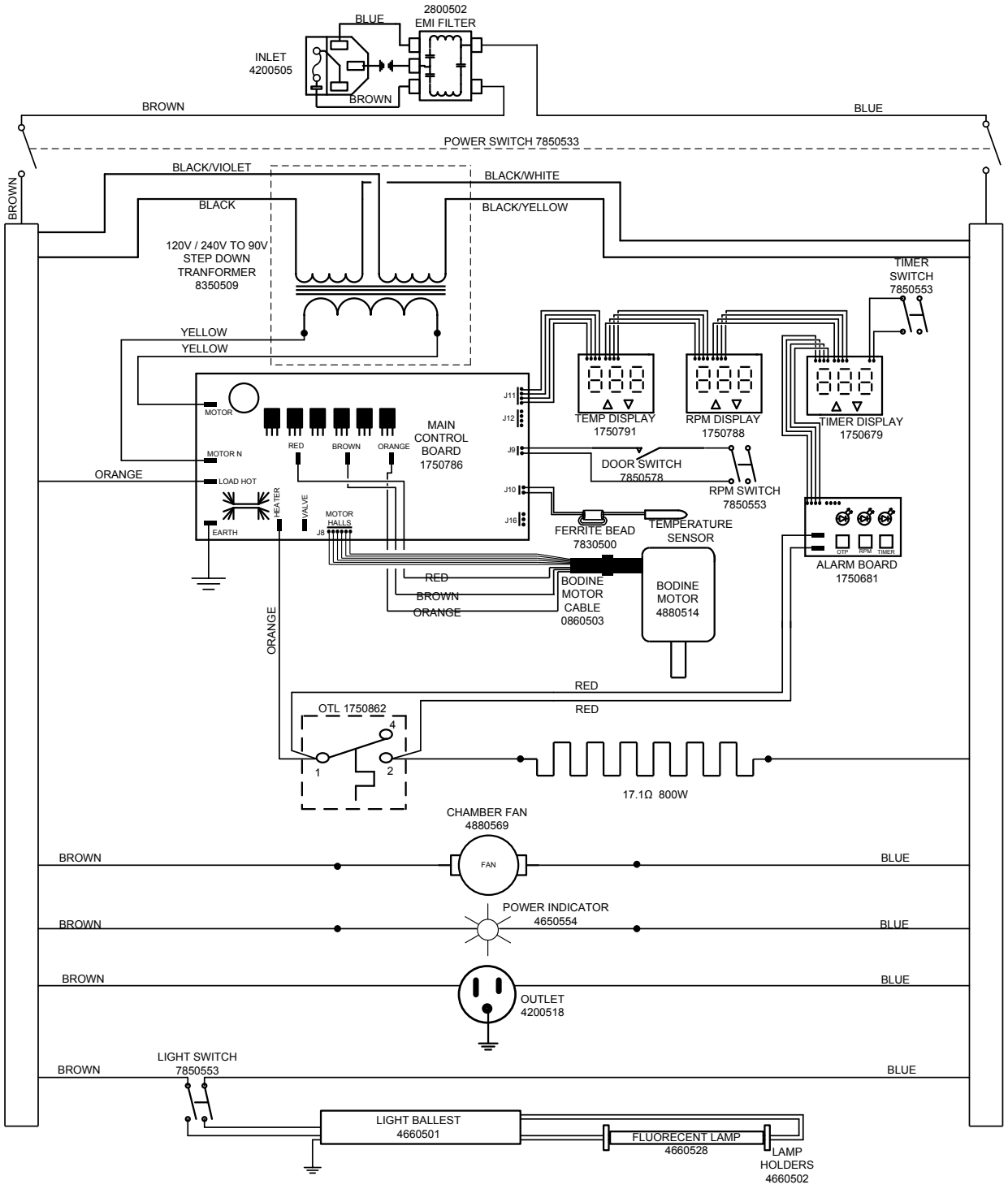
Part	115V	220V
Adjustable feet	2700500	2700500
Alarm Display Vertical	1750681	1750681
Counterweight, Single	5460662	5460662
Door Gasket	3450562	3450562
Drive Belt, Oscillator	0500512	0500512
Element Coil	9570703	9570738
Flask Clamps, 1 Liter	9530532	9530531
Flask Clamps, 125ml	9530530	9530530
Flask Clamps, 250ml	9530531	9530531
Flask Clamps, 500ml	9530526	9530526
Fluorescent Lamp	4650528	4650528
Fuse 120V	3300513	N/A
Fuse 230V	N/A	3300515
Fuse Holder	3300501	3300501
Knob, Safety Thermostat	4450506	4450506
Light Ballast	4660501	4660506
Light Cover	9510502	9510502
Light Cover Gasket	3450538	3450538
Light Holder	4660502	4660502

Part	115V	220V
Motor, Circulation	4880527	4880528
Outlet, Interior	4200518	6100531
Platform (Sample Tray)	9750758	9750758
Power Cord	1800510	1800500
Refrigeration Unit, SSI5R (SI6R)	7010521	7010543
Safety Thermostat	1750862	1750862
Shelf	5100531	5100531
Shelf Clips	1250512	1250512
Switch, Door	7850578	7850578
Switch, RPM, Timer Light	7850553	7850553
Switch, Power	4650554	4650554
Temp. Display Board	1750677	1750677
Timer Display Board	1750679	1750679
Transformer, Speed Control	8350508	8350508

Specifications

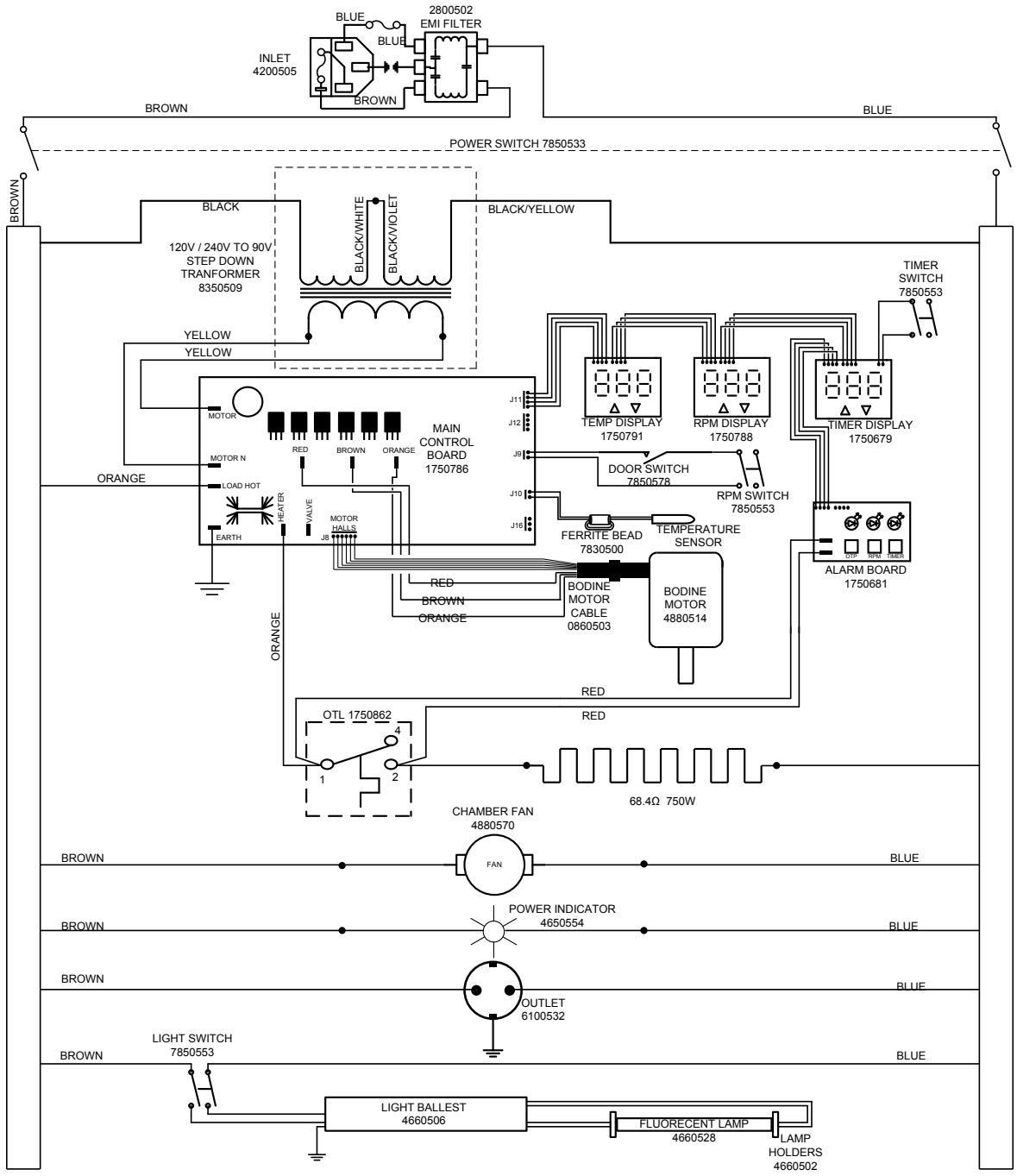
	SSI5 (SI6)	SSI5R (SI6R)
Temperature		
Unit Range	Ambient +8°C to 60 °C	10°C to 60°C
Uniformity	±0.8 °C at 37 °C	±0.8 °C at 37 °C
Accuracy	±0.1 °C	±0.1 °C
Alarms	Visual Safety Lamps	Visual Safety Lamps
Capacity		
Volume	156 m^3 (5.5 cu. ft.)	156 m^3 (5.5 cu. ft.)
Shelves Supplied	2 stainless steel	2 stainless steel
Shelf Dimensions	47 × 47 cm (18.5 × 18.5 in.)	47 × 47 cm (18.5 × 18.5 in.)
Total Shelf Capacity	10 kg (22 lbs)	10 kg (22 lbs)
Dimensions		
Interior (Width × Depth × Height)	48.3 × 48.9 × 59.7 cm (19 × 19.25 × 23.5 in.)	48.3 × 48.9 × 59.7 cm (19 × 19.25 × 23.5 in.)
Exterior (Width × Depth × Height)	72.4 × 73.6 × 106.7 cm (28.5 × 29 × 42 in.)	72.4 × 73.6 × 106.7 cm (28.5 × 29 × 42 in.)
Shaking Mechanism		
Motor	Brushless DC	Brushless DC
Speed, Sample	30 to 400rpm, ±4 rpm (1 rpm increments)	30 to 400rpm, ±4 rpm (1 rpm increments)
Controller	Microprocessor/Digital LED	Microprocessor/Digital LED
Stroke Length	1.3, 1.9, 2.54 cm (0.5, 0.75, 1.0 in.)	1.3, 1.9, 2.54 cm (0.5, 0.75, 1.0 in.)
Orbit Diameter	12 mm, 19 mm or 25 mm	12 mm, 19 mm or 25 mm
Shaking Capacity (stroke-limited)	10 kg (22 lbs.)	10 kg (22 lbs.)
Door Switch	Yes	Yes
Platform Dimensions	44 × 44 x 1.9 cm (17.25 × 17.25 x .75 in.)	44 × 44 x 1.9 cm (17.25 × 17.25 x .75 in.)
Refrigeration		
Compressor Type	NA	1/6 HP
Refrigerant	NA	R-134A (6.5 oz.)
Electrical		
Watts / Amps 110 – 120V~	850 watts / 8.5A	1100 watts / 11.5A
Watts / Amps 208 – 240V~	850 watts / 5.5A	1100 watts / 6.5A
Cycle / Phase	50/60 Hz / Single Phase	50/60 Hz / Single Phase
Certifications	CE (220V only)	CE (220V only)
Weight		
Net Weight	90 kg (198 lbs.)	114 kg (250 lbs.)
Shipping Weight	118 kg (260 lbs.)	136 kg (300 lbs.)

WIRE DIAGRAMS
SSI5 (SI6) 110V
(9851457)



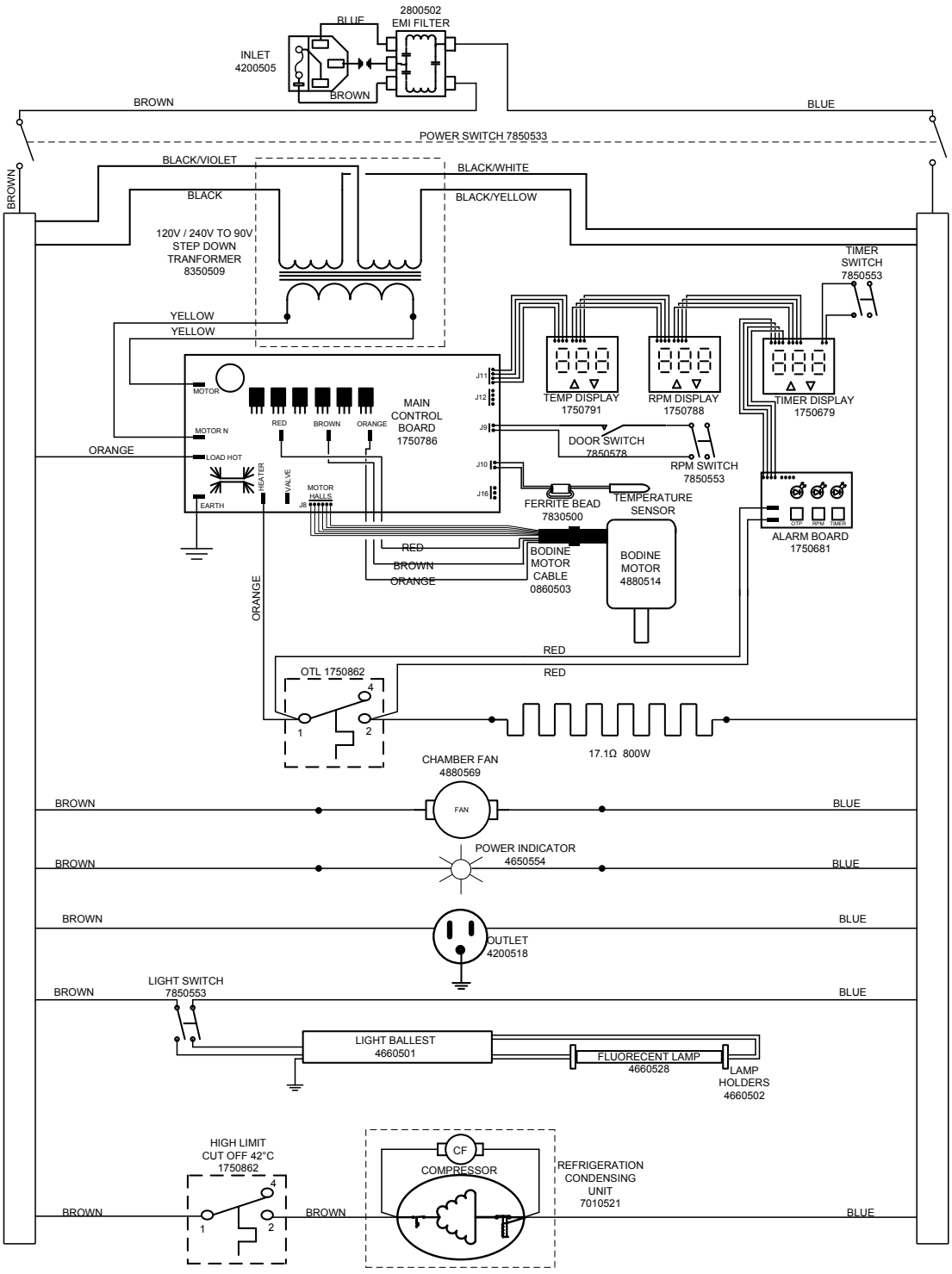
SSI5-2 (SI6-2) 220V

(9851458)



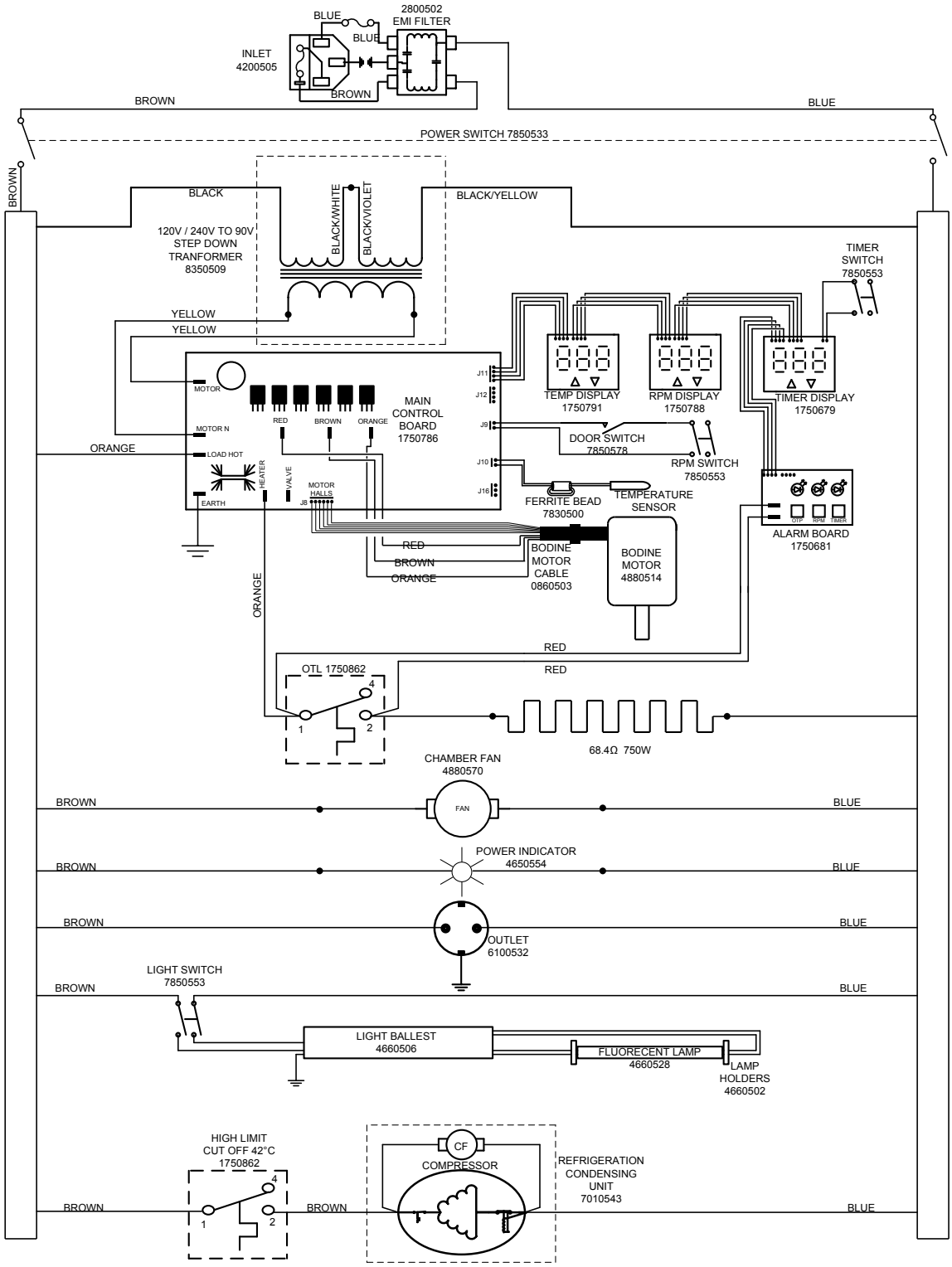
SSI5R (SI6R) 110V

(9851459)



SSI5R-2 (SI6R-2) 220V

(9851460)



TECAN.

Infinite® 200 PRO multimode microplate readers

Immediate access to all wavelengths in an affordable, scaleable detection family – with the patented NanoQuant Plate™ and Gas Control Module (GCM™)

DLReady™ LanthaScreen® Certified invitrogen TRANSCREENER® Red FI validated htrf This reader is HTRF® compatible quad4 monochromators™ 2µl NanoQuant Plate™ compatible

Detection solutions that grow with your ideas

Building on the success of the Infinite 200 series, Tecan has developed the Infinite 200 PRO, with enhancements that cater to the needs of today's scientists. The Infinite 200 PRO offers flexible, scaleable detection solutions for a wide range of assays, using monochromator- and filter-based technologies.

Access to a full range of leading detection methods

Infinite 200 PRO can provide a full range of leading detection methods in one easy-to-use modular instrument. Users can select from modules listed in the table below to create a perfect reader for their needs.

Infinite M200 PRO – Monochromator

The Quad4 Monochromators™ of the Infinite M200 PRO provides exceptional sensitivity, and allows the user to select any wavelength from UV to NIR, and to perform absorbance, excitation and emission scans. Users can access all wavelengths, and change from top to bottom reading, for easy measurement of multiplexed assays at the touch of a mouse click – no manual hardware changes are required.

- Fluorescence intensity top reading including TRF, with automated z-adjustment and background correction
- Enhanced fluorescence intensity bottom reading with OR (Optimal Read) function, including TRF
- Spectrally enhanced photomultiplier tube
- Absorbance
- Photon counting luminescence, including dual color luminescence
- Cuvette port for absorbance
- Temperature control
- Injectors
- NanoQuant Plate
- Gas control module (GCM)

Infinite F200 PRO – Filter

The Infinite F200 PRO uses a patented intelligent filter slide system with an integrated flash counter to monitor the number of flashes the filter is exposed to. And its fluorescence polarization module as well as its AlphaScreen/AlphaLISA module are perfectly suited for binding studies in homogenous mix and read assays. A dichroic filter allows TR-FRET applications, and the filter modules offer a cost-efficient solution for routine applications at fixed wavelengths.

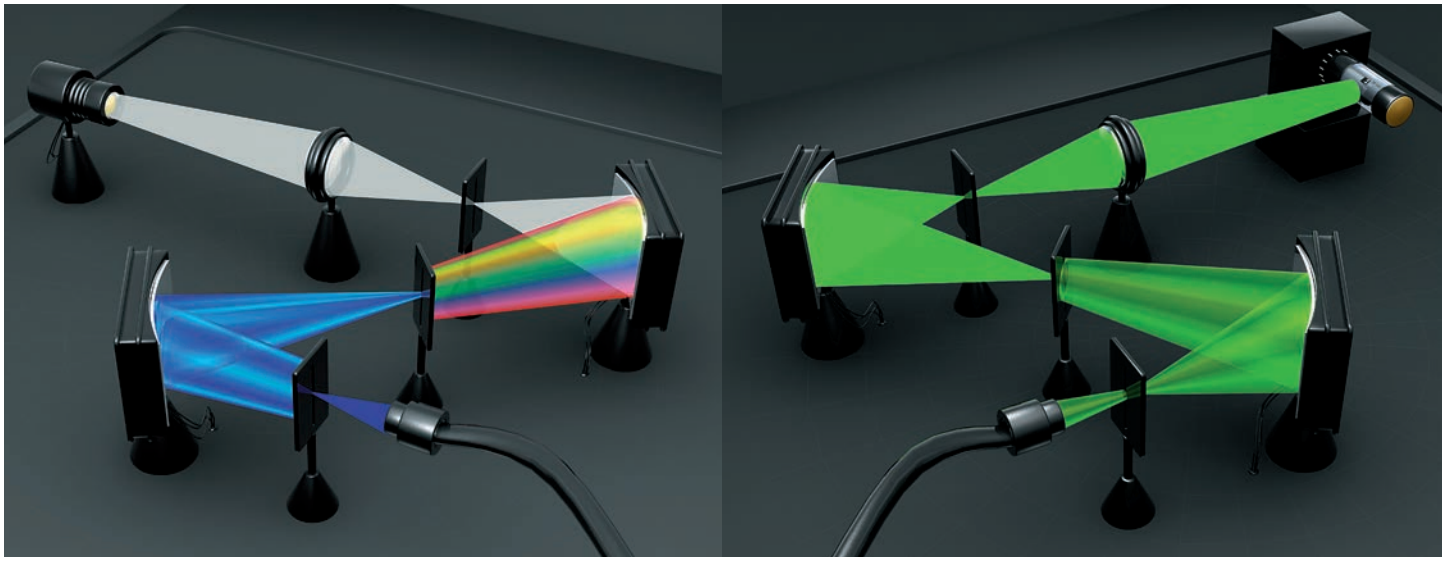
- Fluorescence intensity top reading including TRF
- Enhanced fluorescence intensity bottom reading with OR (Optimal Read) function, including TRF
- TR-FRET/HTRF®
- AlphaScreen® and AlphaLISA®
- Spectrally enhanced photomultiplier tube
- Absorbance
- Photon counting luminescence, including dual color luminescence
- Fluorescence polarization
- Temperature control
- Injectors
- NanoQuant Plate
- Gas control module (GCM)

Select your application, customize your detection device and perform your measurements quickly and easily

Broadly applicable modular detection solutions to widen application capabilities

Detection is at the heart of biopharmaceutical and diagnostic assay measurements. In today's rapidly changing application environment the Infinite 200 PRO's modular, cost-effective design permits fast wavelength selection.

The Infinite 200 PRO has been developed to deliver accuracy and performance in a format that allows you to build a versatile detection system to match your changing application needs. With the Quad4 Monochromators-based Infinite M200 PRO and filter-based Infinite F200 PRO detection options, the reader offers up to eight detection modes for sample measurements in 6- to 384-well plates, PCR plates or cuvettes. Three sets of advanced optics and three high performance detectors – optimized for the requirements of fluorescence, luminescence and absorbance reading – allow uncompromised performance in all detection modes.



The Quad4 Monochromators technology makes use of a double monochromator on both the excitation and emission side. The picture above outlines the double monochromator system architecture on the excitation (left picture) and the emission (right picture).

The Infinite 200 PRO offers unlimited flexibility for a wide range of biological assays and measurements including:

- DNA/RNA quantification
- Protein quantification
- Ion channel studies
- Ion flux studies
- Calcium ion detection
- Reporter gene and gene expression assays
- Cell viability and toxicity assays
- Cell-based assays
- Binding studies
- Enzyme assays
- ELISA
- Immunoassays
- Fluorescence and luminescence applications
- TR-FRET/HTRF applications
- AlphaScreen and AlphaLISA assays

Tecan's filter slide with patented system for monitoring the number of flashes.

Various modules are available with the Infinite M200 PRO and Infinite F200 PRO extended wavelength range and enhanced sensitivity

A spectrally enhanced photomultiplier tube extends emission wavelength range from 330 – 600 nm to 280 – 850 nm, allowing the use of red-shifted dyes and minimizing interference caused by autofluorescence. A UV Si photodiode absorbance detector provides excellent sensitivity for the wavelength range of 230 – 1,000 nm, even at low concentrations.

Superior performance in absorbance for low sample volumes

The instrument's improved wavelength accuracy for 260/280 nm absorbance measurements allows high sensitivity determination of DNA or RNA concentration. Up to 16 samples with volumes as low as 2 µl can be measured simultaneously with Tecan's patented (EP2045015) NanoQuant Plate. This highly precise measurement tool uses a separate quartz optic for each sample, and requires no additional plate calibration.

Gas Control Module

The patented Gas Control Module (GCM; EP2428792) for the Infinite 200 PRO offers a comprehensive solution for a variety of cell-based applications in this versatile multimode reader. Two independent gas inlets allow the automated control of CO_2 and O_2 concentration inside the reader chamber and help to maintain stable culture conditions during prolonged experiments and allow assays to be performed under anaerobic or physiological conditions. Maintaining the optimal CO_2 concentration within the measurement chamber helps stabilize pH and medium conditions, while the independent control of O_2 concentration (oxygen reduction is achieved using N_2) provides hypoxic or simulated in vitro growth conditions. Combining this with precise temperature control and efficient shaking, the Infinite 200 PRO makes cell-based assays more biologically relevant. In addition, the elimination of data gaps (e.g. overnight or on weekends) minimizes the number of repeated assays and leads to more consistent and reliable data than can be achieved manually.

The GCM's acoustic and visible warning system offers excellent measurement reliability. This system is able to detect if the gas pressure or flow changes dramatically during an experiment or quickly recognize if the target concentration deviates or will not be reached.

Altitude influences the atmospheric partial pressure of CO_2, affecting the measured value. The GCM's unique altitude correction function compensates for this, ensuring precise, stable measurement and regulation of gas concentration inside the reader chamber.

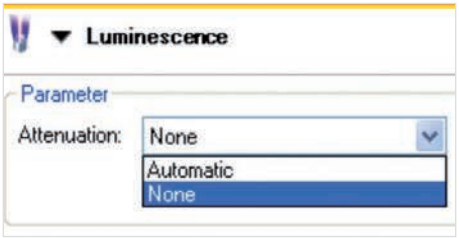
Tecan's impressive GCM allows the optimization of the gas mixture within the reader, providing the perfect solution for experiments with mammalian cells, hypoxia assays, cell viability studies, invasion assays, ischemia or reperfusion studies and many more.

Comprehensive format flexibility

The Infinite M200 PRO offers outstanding format flexibility, and can perform both fixed wavelength and scanning spectrophotometric measurements, using standard 1 × 1 cm cuvettes or low volume microcuvettes in an upright position. In addition, it is compatible with all standard microplate formats, from 6- to 384-wells, including low volume plates and Tecan's unique NanoQuant Plate.

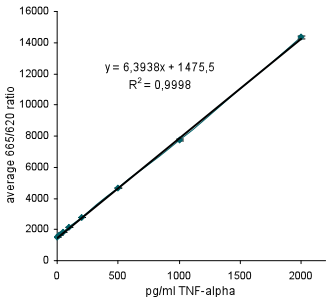
Ready to go luminescence

The luminescence module is capable of reading dual-color luminescence assays, with a photon counting detector that can record even the lowest light levels from an assay, and an integrated set of luminescence filters enable BRET1 and BRET2 applications. The dynamic range for luminescence measurements has also been improved, helping the analysis of sets of samples with wide variation, without the need to adjust sample concentrations.

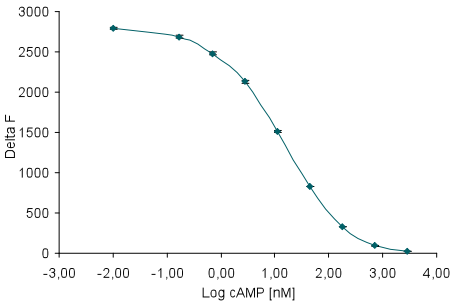


Access to advanced assay systems

A dichroic mirror allows TR-FRET (HTRF) assays on the Infinite F200 PRO, and enhances detection limits for TRF Top Europium and FI Top Fluorescein measurements. This sophisticated system makes the Infinite F200 PRO an attractive and cost effective option for these demanding applications.



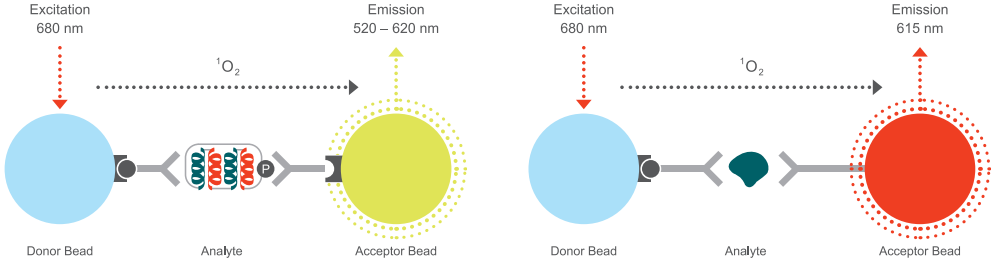
Human TNF-alpha kit: The measurement of a dilution series of the TNF-alpha standard shows a linear course (R^2 = 0.9998) from 2000 to 20 pg/ml TNF-alpha.

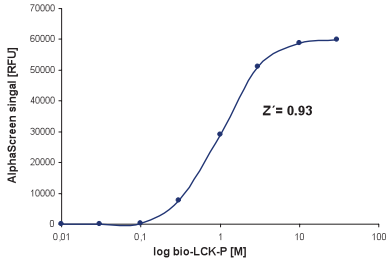


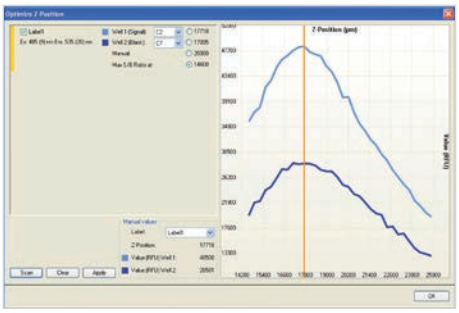
cAMP HiRange kit: The Delta F values obtained with the cAMP dilution series are inversely proportional to the cAMP concentration, resulting in the sigmoidal shape of the curve that is typical for competitive assays.

AlphaScreen and AlphaLISA for high sensitivity detection

AlphaScreen and AlphaLISA are homogeneous assay formats used for the measurement of biological interactions, both based on PerkinElmer's innovative bead technology.
With the Infinite F200 PRO, Tecan offers an affordable alternative to cost-intensive laser-based AlphaScreen and AlphaLISA detection systems. Based on its highly acclaimed fluorescence top optics, the AlphaScreen and AlphaLISA option for Infinite F200 PRO delivers highly sensitive and robust assay results with measurement times perfectly suited for low- to medium-throughput applications.

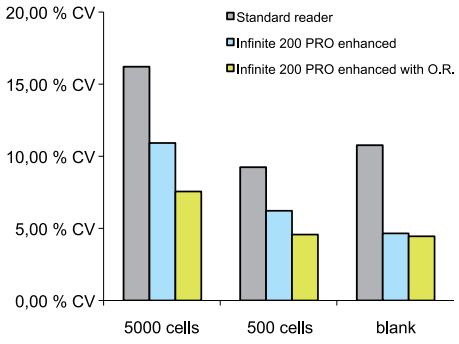






Automated, adjustable z-focus

Implementing assay miniaturization on the Infinite M200 PRO is helped by the automated, adjustable z-focus for FI Top measurements. Equally high sensitivity can be achieved for all plate formats, allowing the same high performance in low volume plates. This new feature, complete with background correction, is particularly suited to cell-based applications using autofluorescent growth media, providing automatic optimization of the signal-to-background ratio.



Cell-based applications

The Infinite 200 PRO benefits from enhanced FI Bottom reading. Its special Optimal Read (OR) function has been designed specifically to optimize and improve cell-based measurements. Very low CVs, high intra- and inter-well reproducibility can be achieved when measuring adherent cells in microplates, offering increased sensitivity.
The Infinite 200 PRO provides linear and orbital shaking – with adjustable amplitude in conjunction with frequency and duration – making it perfect for enzyme, bacterial and cell-based assays. The Infinite 200 PRO also allows temperature control for cellular and biochemical assays that require specific reaction temperatures, with top heating to avoid condensation in lidded plates, ensuring the best performance for covered MTP applications.

Optimized injector module

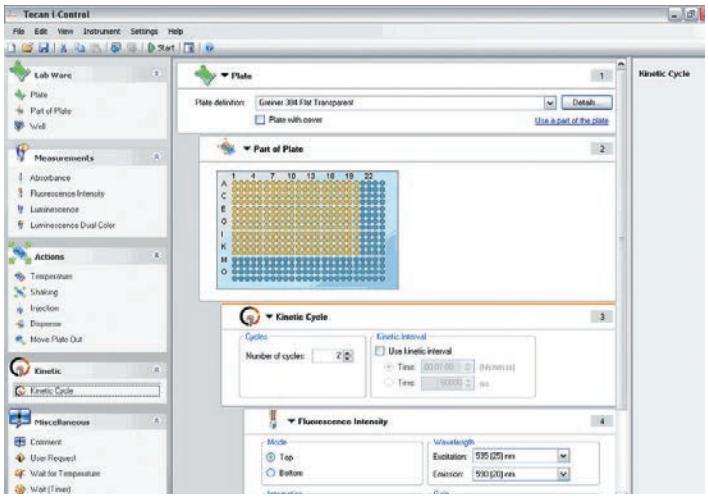
The injector module allows dispensing of up to two reagents per well, helping to replace a manual pipetting step or to trigger fast kinetic reactions in fluorescence, luminescence and absorbance modes. Its metal-free needles are ideal for ion studies, by preventing interference of metal ions in reactions. The injectors have variable volume and speed settings, and can be used together with the ratio mode to allow fast switching of wavelengths in a wide range of applications. The injector module has also been optimized for less wastage of substrates and buffers, with lower dead volumes for priming and the ability to tilt vessels, and its bulk reagent dispense function eliminates tedious pipetting steps for 6- to 384-well plates. Maintenance of the injectors is supported by easily accessible prime/wash buttons.

MultiCheck™ – QC package for Infinite 200 PRO series

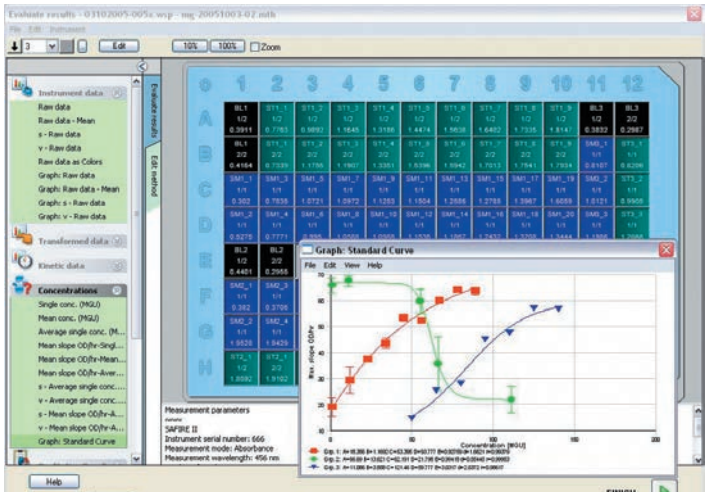
The Infinite 200 PRO has been designed to support users who need to meet GLP (Good Laboratory Practice) standards. A MultiCheck QC plate, which includes installation and operational (IQ OQ) checks and documentation, helps to ensure that all Infinite 200 PRO devices meet the standards needed for quality control laboratories, and satisfies the need to assure production standards in pharma and biotech settings.

Built-in performance features

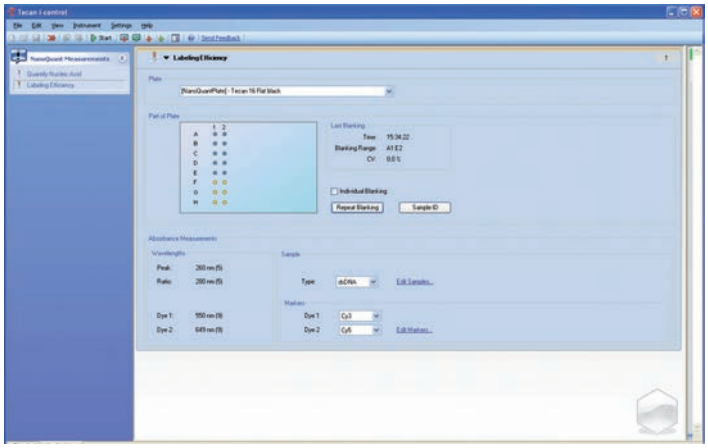
The Plate In/Out button is another useful feature that has been introduced, in response to popular demand.



Workflow oriented i-control software supports complex assay protocols.



Magellan software allows easy presentation and evaluation of data from multiple experimental groups on a microplate.



i-control application for nucleic acid quantification and measuring labeling efficiency.

Software designed for your workflow

Infinite 200 PRO users have complete access to intuitive software solutions that match their detection needs. The Infinite 200 PRO comes complete with i-control™ software interface that allows the user to define the workflow for each application.

Each workflow can be easily created by dragging and dropping the processing steps into the assay protocol sequence. The application workflow is then visible to the user, and can be saved for future use. Data sets are easily managed and exported to Windows® compatible formats like Excel®.

The i-control software includes an application-oriented tab for rapid DNA/RNA quantification in the NanoQuant Plate, and identifies dye incorporation by measuring nucleic acid labeling efficiency. For more advanced data processing, Tecan's proven Magellan™ software provides features that perfectly match the flexibility of the Infinite 200 PRO. Magellan Tracker is designed to meet 21 CFR Part 11 requirements for electronic records and signatures, in compliance with FDA regulations.

Highlights of Magellan software in combination with the Infinite 200 PRO include:

- Application-oriented workflow definition via drag-and-drop functionality
- Wizard-guided application definition for intuitive operation, available in different languages
- Easy conversion of data into results by Excel-style definition of transformations
- Advanced spectra calculation package – the perfect partner for your Infinite M200 PRO reader
- Convenient handling of dilution series and ICx calculations
- Kinetic data analysis with calculation of slopes, onsets and enzyme kinetics
- Pre-defined example files for a range of applications to help you get started immediately
- Comprehensive plate library for fast selection of your favorite microplate

Infinite M200 PRO and F200 PRO – Typical performance values*

Light source	UV Xenon flashlamp	
Wavelength selection		
Infinite M200 PRO	Quad4 Monochromators system (2 excitation and 2 emission monochromators)	
Bandwidth	Ex: < 5 nm for λ ≤ 315 nm and < 9 nm for λ > 315 nm; Em: < 20 nm	
	Absorbance	**Fluorescence**
Wavelength accuracy	< ± 0,5 nm for λ > 315 nm; < ± 0,3 nm for λ ≤ 315 nm	< ± 2 nm for λ > 315 nm; < ± 1 nm for λ ≤ 315 nm
Wavelength reproducibility	< ± 0,5 nm for λ > 315 nm; < ± 0,3 nm for λ ≤ 315 nm	< ± 1 nm for λ > 315 nm; < ± 0,5 nm for λ ≤ 315 nm
Infinite F200 PRO	Up to 4 filter pairs per slide	
Wavelength range	**Standard**	**Optional**
Fluorescence intensity	Ex 230 – 600 nm, Em 330 – 600 nm	Ex 230 – 850 nm, Em 280 – 850 nm
Fluorescence polarization	Ex 300 – 600 nm; Em 330 – 600 nm	Em 330 – 850 nm
Absorbance	230 – 1000 nm	
Detectors	Fluorescence – PMT, optional UV and red-sensitive	
	Absorbance – UV silicon photodiode	
	Luminescence – photon counting system with low dark current PMT	
Plate formats	6- to 384-well plates, cuvettes, NanoQuant Plate	
Temperature control	Ambient +5 °C up to 42 °C	
Shaking	Linear, orbital	
Fluorescence sensitivity [1] values	**Infinite F200 PRO**	**Infinite M200 PRO**
Fluorescence top reading [1]	85 amol / well (0,85 pM, 384-well plate)	170 amol / well (1,7 pM; 384-well plate)
Fluorescence bottom reading [1]	0,7 fmol / well (3,5 pM; 96-well plate)	1,2 fmol / well (6 pM; 96-well plate)
TRF [2]	2,8 amol / well (28 fM; 384-well plate)	90 amol / well (0,9 pM; 384-well plate)
FP [1]	< 4 mP standard deviation @ 1 nM Fluorescein	N / A
Luminescence sensitivity values	**Standard**	
Glow luminescence [3]	225 amol ATP / well (9 pM; low volume 384-well plate)	
Flash luminescence [4]	12 amol ATP / well (218 fM; 384-well plate)	
Absorbance		
Ratio accuracy 260 / 280 nm	± 0,07	
Precision @ 260 nm	< 0,2 %	
Accuracy @ 260 nm	< 0,5 %	
Measurement range	0 – 4 OD	
AlphaScreen		
Detection Limit	≤ 50 ng/ml Omnibeads [5]	
Uniformity	≤ 5 % CV [5]	
Z'value	≥ 0.8 [6]	
Typical reading time	< 11 min (384-well plate)	
Injectors		
Pump speed	100 – 300 µl/s	
Injection volume	selectable in 1 µl increments; max. volume: 800 µl per stroke	
Dead volume	100 µl including pump back	
Fastest Read Times		
96 well plate	20 sec	
384 well plate	30 sec	
Wavelength Ex / Em-scan, 96 well plate		
450 – 550 nm, 5 nm step	150 sec	

[1] Detection limit for Fluorescein, [2] Detection limit for Europium, [3] Detection limit for ATP (144-041 ATP detection kit SL (BioThema), [4] Detection for ATP (ENLITE® Kit)
[5] (PE # 6760626); (384-well small volume plates), [6] (P-Tyr-100 Assay Kit, PE # 6760620); (384-well small volume plates)

* Specifications are subject to change. Performance values represent the average observed factory tested values.
For product specifications refer to operators manual.

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Germany +49 79 51 94 170 **Italy** +39 02 92 44 790 **Japan** +81 44 556 73 11 **Netherlands** +31 18 34 48 17 4 **Singapore** +65 644 41 886 **Spain** +34 935 95 25 31
Sweden +46 8 750 39 40 **Switzerland** +41 44 922 89 22 **UK** +44 118 9300 300 **USA** +1 919 361 5200 **Other countries** +43 62 46 89 33

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Shaking Incubators

Floor Models

Versatile and Robust

Shaking incubators, also know as environmental shakers, are often used for cell culturing, cell aeration, and solubility studies. In addition to stable temperature conditions, they use an orbital agitation at variable speeds to affect the growth of cell cultures.

This is why the SHEL LAB floor model shaking incubators have adjustable stroke lengths to accommodate various cells and applications. This gives flexibility in adjusting the speed and orbit to meet each application. The SSI5R-HS achieves speeds up to 850 RPMs.

All models come equipped with a universal shaking platform, independent alarms, and microprocessor controls for temperature and speed adjustment.

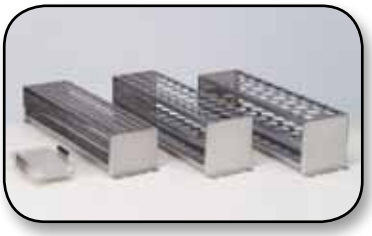
SSI5 Shaking Incubator Flask Clamp & Test Tube Rack Capacity

Description	Part Number	Size	Max #
Flask Clamp	9530528	25 ml	50
Flask Clamp	9530529	50 ml	50
Flask Clamp	9530530	125 ml	25
Flask Clamp	9530531	250 ml	25
Flask Clamp	9530526	500 ml	10
Flask Clamp	9530532	1000 ml	6
Flask Clamp	9530551	2000 ml	4
Flask Clamp	9530554	4 liter	4
Flask Clamp	9530555	6 liter	2
Fernbach Style	9530553	2.8 liter	2
Test Tube Shaking Rack	9751177 (100 Max)	10-13 ml	3
Test Tube Shaking Rack	9751178 (80 Max)	14-16 ml	3
Test Tube Shaking Rack	9751179 (60 Max)	18-20 ml	3
Test Tube Shaking Rack	9751180 (36 Max)	22-25 ml	3
Test Tube Shaking Rack	9751181 (29 Max)	50 ml	3

*Not to exceed units maximum load capacity of 22 lbs (10 kg)

Full Selection of Accessories

The easily removable rotation platform is included with each SHEL LAB unit.



Flask holders and accessories can be arranged in several combinations on the platform according to what best suits your application.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Orbital-Shaking Speed Alarm

2 Year Limited Warranty!

Shaking Incubators	110-120V 220-230V Details	Model Number SSI5 SSI5-2 Floor	 SSI5R SSI5R-2 Floor Refrigerated	 SSI5R-HS SSI5R-HS-2 High RPM
Exterior Dimensions (wxdxh)	Inches	28.5 x 29.5 x 40.5	28.5 x 29.5 x 40.5	28.5 x 29.5 x 40.5
	cm	72.4 x 75.0 x 102.9	72.4 x 75.0 x 102.9	72.4 x 75.0 x 102.9
Chamber Dimensions (wxdxh)	Inches	19.0 x 20.5 x 22.5	19.0 x 20.5 x 22.5	19.0 x 20.5 x 22.5
	cm	48.2 x 52.0 x 57.1	48.2 x 52.0 x 57.1	48.2 x 52.0 x 57.1
Incubator Chamber Capacity	cu ft	5	5	5
	L	144	144	144
Temperature Range	Celsius	8°C + Ambient to 60°C	10°C to 60°C	10°C to 60°C
Temperature Uniformity	Celsius	+/-0.8°C at 37°C	+/-0.8°C at 37°C	+/-0.8°C at 37°C
Platform Capacity	lbs (kg)	22 (10)	22 (10)	22 (10)
Orbital-Shaking Range	RPM	30-400	30-400	30-850
Timer Functionality	Minutes	1-999	1-999	1-999
Number of Shelves	Included	1	1	1

*Specifications are based on nominal values at an ambient temperature of 25°C and a line voltage of 120V or 240V respectively. The temperature data is determined in accordance with DIN 12880. We reserve the right to alter technical specifications at any time.
For a complete list of product specifications, call (800)322-4897 or visit the SHEL LAB website, www.shellab.com

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SHEL LAB®

www.shellab.com
1-800-322-4897

Thermo Scientific™ Refrigerators

Meet most laboratory requirements

Refrigerators feature an easy-to-clean interior with epoxy-coated, steel-wire shelves that resist most acids, solvents, and chemicals. Flammable material and explosion-proof models meet safety requirements of OSHA and the National Fire Protection Association.

Undercounter Refrigerator features adjustable hydraulic thermostat control and key lock.

Upright Refrigerator features adjustable hydraulic thermostat control, door lock, and storage basket.

Flammable Material Models are designed to completely insulate the storage compartment from chance of electrical sparking. Top-mounted thermostats.

Explosion-Proof Models are for use in hazardous locations where explosive conditions could potentially exist external to the cold storage unit. Top-mounted thermostats and door with lock. Suitable for use in Class I, Division I Group C and D hazardous environments.

Capacity	Temperature range	Dimensions (W x H x D)		No. of shelves	Power			Shpg wt lb (kg)	Catalog number	Price
		Interior	Overall		VAC	Hz	Watts			
Undercounter refrigerator										
5.6 cu ft (159 L)	2 to 7°C (36 to 45°F)	19" x 27½" x 13" (48 x 70 x 33 cm)	23½" x 33⅓" x 23¾" (60 x 85 x 60 cm)	3	120	60	160	120 (55)	GH-44202-00	
					240	50	160		GH-44202-05	
Upright refrigerator										
20 cu ft (566 L)	2 to 10°C (36 to 50°F)	17" x 56" x 23½" (43 x 142 x 60 cm)	32" x 70" x 28½" (81 x 178 x 72 cm)	5	120	60	480	280 (127)	GH-44202-30	
Flammable material refrigerators										
5.6 cu ft (159 L)	2 to 7°C (36 to 45°F)	19" x 27½" x 13" (48 x 70 x 33 cm)	23½" x 33⅓" x 23¾" (60 x 85 x 60 cm)	3	120	60	160	120 (55)	GH-44201-20	
20.6 cu ft (583 L)	2 to 10°C (36 to 50°F)	27" x 57" x 23" (69 x 145 x 58 cm)	32¼" x 76" x 30½" (82 x 193 x 77 cm)	4	120	60	480	230 (104)	GH-44201-30	
Explosion-proof refrigerators										
20 cu ft (566 L)	2 to 10°C (36 to 50°F)	27" x 57" x 23" (69 x 145 x 58 cm)	32" x 76" x 30½" (81 x 193 x 77 cm)	4	120	60	480	230 (104)	GH-44200-25	

Thermo Scientific™ General-Purpose and Safety Refrigerator/Freezers

Combination storage for biological materials or chemicals

01290-50

Undercounter Refrigerator/Freezer features adjustable thermostat control and manual defrost.

Upright Refrigerator/Freezer features automatic defrost and separate doors. UL listed.

Flammable Materials Models contain no internal electrical devices that can trigger the explosion of hazardous materials. They are supplied with a 6-ft (1.8-m) cord with three-prong plug.

Explosion-Proof Models protect against explosions both inside and outside of the unit. All motors, switches, and thermostats prohibit arcing that can ignite flammable air-vapor mixtures. They meet Class 1, Group C and D requirements for hazardous environments. Explosion-proof units must be hard-wired to voltage source.

All models feature CFC-free insulation and coolant.

Capacity		Temperature range		Dimensions (W x H x D)			No. of shelves	Power			Shpg wt		Catalog number	Price
				Interior		Overall								
Refrigerator	Freezer	Refrigerator	Freezer	Refrigerator	Freezer			VAC	Hz	Watts	lb	kg		
Undercounter compact refrigerator/freezer														
4.9 cu ft (144 L)	0.7 cu ft (21 L)	2 to 7°C (36 to 45°F)	–6 to 1°C (21 to 33°F)	18" x 21" x 15½" (46 x 53 x 39 cm)	15½" x 6" x 11" 39 x 15 x 28 cm)	23¾" x 33½" x 25½" (60 x 85 x 65 cm)	3	120	60	160	120	55	GH-01290-50	
								240	50	160	120	55	GH-44200-45	
Upright refrigerator/freezer														
6.8 cu ft (200 L)	2 cu ft (59 L)	–6 to 7°C (21 to 45°F)	–20 to –8°C (–4 to 18°F)	18½" x 35" x 11" (47 x 89 x 28 cm)	18½" x 14" x 9" (47 x 36 x 23 cm)	24" x 61" x 23" (61 x 155 x 59 cm)	6	120	60	120	160	73	GH-44200-40	
Flammable materials lab refrigerator/freezer with two doors														
8.6 cu ft (244 L)	2.4 cu ft (68 L)	–1 to 8°C (30 to 46°F)	–20 to –12°C (–4 to –10°F)	19½" x 39" x 13½" (50 x 99 x 34 cm)	18" x 12" x 17" (46 x 32 x 43 cm)	23½" x 60¾" x 30" (60 x 154 x 76 cm)	3 cabinet, 4 door	120	60	230	150	68	GH-44201-00	
								240	50	230	150	68	GH-44201-05	
Explosion-proof lab refrigerator/freezer with two doors														
8.6 cu ft (244 L)	2.4 cu ft (68 L)	–1 to 8°C (30 to 46°F)	–20 to –12°C –4 to –10°F)	19½" x 39" x 13½" (50 x 99 x 34 cm)	18" x 12" x 17" (46 x 32 x 43 cm)	23½" x 60¾" x 31" (60 x 154 x 79 cm)	3 cabinet, 4 door	120	60	360	150	68	GH-44200-00	
								240	50	360	150	68	GH-44200-05	

LRQ 4005852/B

Founded in 1875, Shimadzu Corporation, a leader in the development of advanced technologies, has a distinguished history of innovation built on the foundation of contributing to society through science and technology. We maintain a global network of sales, service, technical support and applications centers on six continents, and have established long-term relationships with a host of highly trained distributors located in over 100 countries. For information about Shimadzu, and to contact your local office, please visit our Web site at **www.shimadzu.com**

SHIMADZU

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Wharfside, Trafford Wharf Road, Manchester M17 1GP, UK
Phone: +44 161 888 4400 Fax: +44 161 888 4402
URL http://www.shimadzu.com/an/

SHIMADZU
Excellence in Science

MO305

Taking MALDI-TOF MS Beyond the Standard

AXIMA Confidence™

Axima Confidence™ - Sensitivity and Flexibility

The Axima Confidence™ is designed with the general analytical and life science laboratory in mind. Incorporating a variable repetition rate 50Hz N_2 laser, the system provides rapid, high quality MALDI mass spectra and an array of software tools for data processing and reporting.

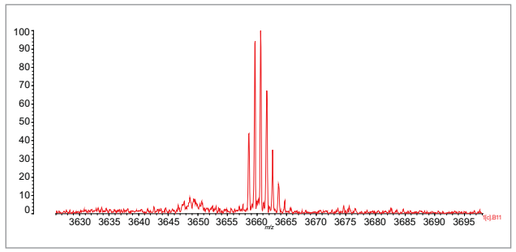
Linear mode allows the interrogation of high molecular weight samples, whilst reflectron mode, incorporating the patented curved-field reflectron (CFR), provides the high resolution and mass accuracy necessary for successful proteomics and life science experiments.

Positive and negative ion modes are included as standard allowing greater flexibility and extending the compound categories that may be analysed. The system also incorporates a patented beam blanker to optionally remove unwanted low mass ions and prevent detector saturation.

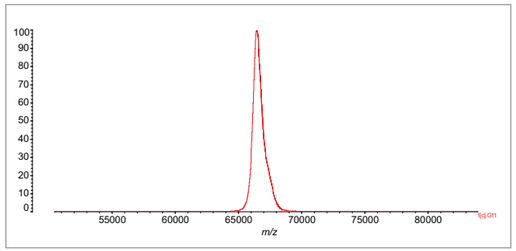
Excellent sensitivity is achieved using near normal (on-axis) laser irradiation and advanced ion optics for enhanced ion transmission. Pulsed extraction of ions from the MALDI source, in combination with the unique reflectron design, improves resolution and enhanced calibration algorithms with easy to use software facilitate the generation of more accurate data.

MS/MS may be easily performed using a seamless approach – ions of interest can be isolated using a precursor ion selection device, incorporated as standard, and data-rich fragment ion spectra quickly and simply acquired. The newly improved curved field reflectron design augments the low mass fragment region providing useful additional information.

Unparalleled flexibility is achieved by a variety of sample target formats including standard microtitre plate format 96 or 384 well targets. Fleximass™ microscope slide (plain or 48 well targets) and a wide variety of adaptors for unconventional sample layouts are also available. The standard sample target formats are fully compatible with common laboratory robots including the CHIP™.



ACTH fragment 7-38 showing >15,000 resolution FWHM



Bovine serum albumin in linear mode

Axima Confidence™ - Software solutions

Intuitive software incorporating data dependent workflows for achieving maximum results with minimum user input, making the system ideal for novice and expert users alike.

The Axima Confidence™ is controlled by the Launchpad™ suite of software, common to all Axima mass spectrometers, permitting manual or fully automated operation facilitating the seamless analysis of as few or as many samples as required. Intelligent optimization of acquisition conditions may be employed allowing auto-tuning for specific samples.

Ideally suited for life science and analytical environments alike, the system offers software packages specifically created for:

- Proteomics experiments
  - LC MALDI
    - Polymer analysis
      - Tissue imaging/biomarker discovery
        - Oligonucleotide/primer analysis

Application-centric data processing software packages are available to provide solutions to many commonly asked questions.

Intellimarque™ for proteomics experiments

Designed with the flexibility to adapt to user workflows: from a handful of samples to high throughput fully automated data generation, data-dependent peptide mass fingerprinting and MS/MS for protein identification are integrated into easy-to-use intuitive software.

- Peptide mass fingerprints are acquired and subjected to an optional integrated Mascot® database search.
- User definable limits for acceptance of PMF-based protein identification.
- Data-dependent MS/MS: using the results of the PMF search, MS/MS may be performed on ions that matched to the top ranked protein hit (confirmation MS/MS), in addition to those that were not (investigation MS/MS).
- Batch searching of these MS/MS spectra is then performed automatically to provide further and higher confidence protein identification.
- Data may be reprocessed and resubmitted for database searching at a later time to provide additional information.

PolymerAnalysis™

Polymers and copolymers can be characterised using our unique polymer software, PolymerAnalysis™, providing useful structural information and statistics in a text report format.

OliogoAnalysis™

Offers fully automated QC analysis of large numbers of oligonucleotides or peptides, complete with a report indicating the presence or absence of the target compound, an estimate of the purity and occurrence of known contaminants, adducts or truncated/extended analogues.

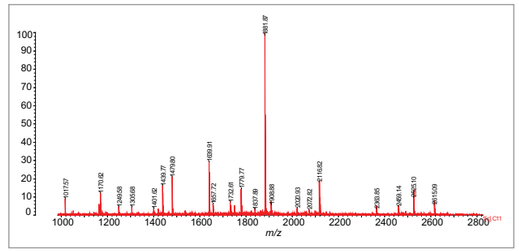
Biomarker discovery/Tissue imaging

This exciting area encompassing clinical sample screening, tissue imaging and profiling is comprehensively addressed using automated acquisition methods and refined data processing. Protein/peptide biomarkers, drugs and their metabolites can be rapidly screened directly from tissue sections and their location mapped and visualized using integrated software tools. Data can also easily be exported to alternative processing packages, including BioMap and NonLinear Dynamics PG600.

System support

All Axima systems can be fully supported throughout their lifetime using sophisticated web based service diagnostics and real time remote monitoring. Highly trained specialist local service support engineers are available to install and maintain Axima mass spectrometers. A wide range of service contracts are available, catering for all budgets and requirements, including IQ/OQ environments.

Full training courses are offered by MALDI experts at our regional corporate training centers or at the customer site and may be tailored for specific requirements and applications.



Glycogen phosphorylase B peptide mass fingerprint

SHIMADZU
Excellence in Science

AXIMA Confidence™

Specification

Technical Data

Sample Handling

- Fully automated sample introduction mechanism
- XY stage (10 µm step, 10 µm repeatability) for microtiter plate footprint MALDI target
- 2 mm thick plain, 96 and 384 sample targets
- Accepts thick (10 mm) targets with optional adaptor for a variety of biochip designs and alternative formats
- Turbomolecular pump (nominal 250 l/s) for fast SAC pumping with rotary backing
- Computer software driven target stage for accurate positioning of sample under the laser focus
- Raster software for scanning samples for 'sweet spots'

Sample Viewing System

- Monochrome CCD camera (25x magnification) controlled by software embedded in LAUNCHPAD™

Ionization Source

- Matrix assisted laser desorption ionization
- Pulsed Extraction (mass calibrated variable delay) or Continuous Extraction, under software control
- Variable ion extraction energy (linear +25 kV/-20 kV, reflectron +20 kV/-20 kV) under software control
- Positive and negative ion operation, as standard, through software selection

Laser

- 337 nm nitrogen laser, fixed focus
- 3 ns pulse width
- Nominal energy - 100 µJ per laser shot
- Maximum pulse rate - 50 Hz (50 laser shots per second)
- Near normal (on-axis) incidence of the laser beam to the sample
- Laser power and laser aim under software control

Analyzer

- Linear flight tube of 1.2 m drift length
- Reflectron effective drift length - 2.0 m
- Vacuum maintained by two turbomolecular pumps (nominal 250 l/s) with rotary backing
- Unique curved field reflectron system for seamless generation of MS/MS ions in a single spectrum
- Beam blanking to deflect unwanted strong signals e.g. matrix ions
- Precursor ion gate - pulsed electrostatic deflector

Detector

- Linear mode - electron multiplier (multiple dynode)
- Reflectron mode - fast micro-channel plate
- 2 GHz, 8 bit transient recorder, 16 bit accumulator
- Second transient recorder for simultaneous neutral detection - 125 MHz, 8 bit, 64 kB RAM

Control and Data System*

- Intel core i3 3.3GHz PC with 19" monitor
- 3 GB RAM
- 500 GB hard disc, DVD-RW
- Network adaptor and frame grabber
- Microsoft® Windows® 7 operating system

*Minimum specification subject to continuous improvement

Software

- LAUNCHPAD™ - operates under Microsoft® Windows® 7
- Software for automatic optimization of data generation
- Calculator for determination of theoretical masses of chemicals
- Calculator for determination and manipulation of peptide sequences
- Scanning software for the identification of 'sweet spots'
- Sample layout editor
- Sample scanning editor
- KOLA™ to access internet and intranet (Mascot® from Matrix Science Ltd.) database search engines for protein identification

Installation Data

Dimensions

- Size (w h d) - 0.7 m x 1.92 m x 0.85 m, minimum distance to wall at back is 100 mm
- Weight - 345 kg excluding data system

Installation Requirements

- Electrical - 200 VAC, 50/60 Hz, 1000 VA single phase OR
  230 VAC, 50/60 Hz, 1000 VA single phase
- A 'clean', stable and continuous mains supply is required for reliable operation
- PC - selectable 100-120 VAC, 50/60 Hz, 2.0 A single phase OR
  220-240 VAC, 50/60 Hz, 1.0 A single phase
- Monitor - auto-sensing 100-240 VAC, 50/60 Hz, 1.4-0.6 A
- Temperature - ambient 18° to 26° Celsius
- Relative humidity - less than 70% non condensing
- Vibration free, firm, level floor, at least 345 kg supported at four points

Performance Data

• **Mass range**	linear	-	1 to 500 kDa
	reflectron	-	1 to 80 kDa
• **Mass resolution**	linear	-	>5000 FWHM - ACTH 18-39((M+H)+ 2465 Da)
	reflectron	-	>15,000 FWHM - ACTH 7-38 ((M+H)+ 3660 Da)
	MS/MS	-	isotopic resolution of fragments - Angiotensin II
• **Accuracy**	linear	-	<30 ppm with internal calibration
			<200 ppm with external calibration**
	reflectron	-	<10 ppm with internal calibration
			<100 ppm with external calibration**
	MS/MS	-	0.02% of parent
• **Ion gate resolution**			>200 FWHM @ 1000 Da
• **Sensitivity**	linear	-	250 fmol (loaded) - bovine serum albumin
			250 amol - Glu-1-Fibrinopeptide B (loaded)
	reflectron	-	500 amol - Glu-1-Fibrinopeptide B (loaded)
	MS/MS	-	25 fmol (loaded) - Glu-1-Fibrinopeptide B

**Nearest neighbour external calibration on 384 well sample target, within 30 minutes.
All specifications are run on a standard 2 mm, 384 well, stainless steel sample plate unless otherwise stated.

The AXIMA range of instruments is designed and manufactured under the Kratos Analytical Ltd Quality Management System and is CE compliant.
Installation and initial training will be provided by a team of experienced engineers and application specialists world-wide. The instrument is covered by a 12 month warranty.
Please contact your local representative for details on full service contracts.

SHIMADZU

www.shimadzu.com/an/

Pure Steam Generator

Overview:

- Built to the exacting standards of the pharmaceutical industry and cGMP requirements.
- Wide range of sizes in both vertical and horizontal configurations available to fit your unique requirements.
- All components manufactured by MECO in the U.S. using the most advanced methods for design and fabrication.
- Supported by MECO 24-hour customer service and MASTERsupport Online Service Center
- Backed by warranty.
- Vertical design / Minimal floorspace needed for vertical generators.

Vertical Product Highlights:

- **Superior Design**
  - Vertical natural circulation evaporators
    - Scale effects reduced
    - Uniform wetting and heat transfer
  - Meets/exceeds cGMP/USP standards
    - Sanitary construction and connections
    - Double tubesheet heat exchangers
    - Sloped piping and low point drains
    - Minimum deadlegs
- **Superior Control**
  - PLC based control system
  - PID loop control of feedwater level
  - PID loop control of pure steam pressure
    - Allow 100% capacity turndown
  - Horizontal design
  - Perfect alternative when overhead space is limited

Horizontal Product Highlights:

- **Superior Design**
  - Horizontal submerged tube arrangement
  - Removable tube bundle
  - Uniform wetting and heat transfer
  - Meets/exceeds cGMP/USP standards
  - Sanitary construction and connections
  - Double tubesheet heat exchangers
  - Sloped piping and low point drains
  - Minimum deadlegs
- **Superior Control**
  - PLC based control system
  - PID loop control of feedwater level
  - PID loop control of pure steam pressure
  - Allow 100% capacity turndown
- **Design Details**
  - 25 Ra standard process contact surfaces
  - Galvanized rigid steel conduit
  - Stainless steel airlines
  - Blowdown cooler
  - Mirror finish stainless steel cladding
- **Design Details**
  - 25 Ra standard process contact surfaces
  - Galvanized rigid steel conduit
  - Stainless steel airlines
  - Blowdown cooler
  - Mirror finish stainless steel cladding

VERTICAL -Plant steam/pure steam pressure (psig)						
Model	100 / 60 lbs/hr	100 / 40 lbs/hr	110 / 60 lbs/hr	110 / 40 lbs/hr	120 / 40 lbs/hr	120 / 60 lbs/hr
CS280V	190	280	240	280	280	280
CS550V	300	480	370	550	440	560
CS800V	470	730	610	800	650	940
CS1700V	1150	1700	1430	1700	1700	1700
CS3000V	1930	3000	2430	3000	2880	3000
CS4000V	2740	4050	3420	4050	4060	4050
CS6600V	4300	6620	5370	6620	6340	6620
CS8100V	6440	8100	8000	8100	9440	8100
HORIZONTAL -Plant steam/pure steam pressure (psig)						
Model	100 / 60 lbs/hr	100 / 40 lbs/hr	110 / 60 lbs/hr	110 / 40 lbs/hr	120 / 60 lbs/hr	120 / 40 lbs/hr
CS5900H	2780	5220	3520	5950	4270	6660
CS10000H	4720	8900	6000	10130	7270	11310
CS13000H	6400	12040	8110	13680	9810	15310

Utility Steam Consumption, lbs/hr: 110% of pure steam output; Feedwater, lbs/hr: 110% of pure steam output; Feedwater Pressure, psig: 12-15 psig above pure steam pressure; Feedwater Quality: Hardness: None/Conductivity: <10μs/cm/Silica: <1.0 mg/lt; Feedwater Temperature: 70°F

MECO

U.S. /Canada/ UK / UAE Corporate Headquarters • 12505 Reed Road, Suite 100 • Sugar Land, TX 77478 • 800-450-6326 • www.mecobiopharm.com

Incubators

B.O.D., CO2, Humidified, Shaking

SHEL LAB®

Sheldon Manufacturing, Inc. is an ISO 9001:2008 certified manufacturer of high quality and innovative constant temperature equipment to the global market. Major product lines include incubators, humidity test chambers, ovens, water and bead baths, and anaerobic chambers for the life science, pharmaceutical, biomedical, environmental and industrial markets. Founded in 1970, Sheldon utilizes over 40 years of manufacturing expertise to aggressively pursue new product opportunities that add value to our customers' portfolio. Sheldon markets a complete line of products under the SHEL LAB and Lab Armor brands, which compliment our OEM manufacturing capabilities.

Contact us: US toll free - 1 (888) 227-1410 or (503) 640-3000

Visit our websites at www.shellab.com and www.labarmor.com.

MADE IN USA
FOR THE WORLD

Table of Contents

Microbiological Incubators

SHEL LAB General Purpose Incubators are the ideal solution for industrial protocols, biological research and environmental studies that demand accurate and repeatable results. Their best in class temperature uniformity is usually found only in more expensive, application specific incubators. SHEL LAB General Purpose Incubator's wide temperature and size range make them a perfect solution for any lab.

Applications include:

- Biochemical Studies
- Hematological Studies
- Bacterial Culturing and Research
- Microbiological Determinations
- Pharmaceutical Stability Assays
- Food Processing Quality Control
- Large Scale Roller Apparatus Applications

Microbiological Incubators
Laboratory Series

Heated doors and a unique air jacket design achieve precise temperature uniformity. An independent secondary temperature controller offers the added safety and security of over temperature production.

The laboratory incubator series models include a sealed, inner glass door which provides a view into the chamber without compromising samples or the chamber environment. Stainless steel panels and doors reduce contamination, provide durability and allow for easy cleaning.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Temperature Uniformity +/-0.35°C at 37°C
- Temperature Range Ambient +8°C to 70°C

Adjustable, No Tip Shelf Design

Interior Electrical Outlet

Throughwall Access Port

SMI12

2 Year Limited Warranty!

SMI6

SMI2

SMI11

CE TÜV SÜD NRTL C US

General Purpose Laboratory Incubators		Model Number				
	Details	SMI2 Bench Top	SMI7 Under Counter	SMI11 Double Doors	SMI6 Floor Model	SMI12 Dual/Stacked
Exterior Dimensions (wxdxh)	Inches	22.0 x 22.0 x 27.3	30.0 x 31.8 x 32.3	42.5 x 27.0 x 38.0	25.5 x 27.3 x 38.0	25.5 x 27.3 x 75.8
	cm	55.9 x 55.9 x 66.1	76.2 x 80.7 x 82.0	108 x 68.6 x 96.6	64.8 x 69.3 x 96.6	64.8 x 69.3 x 192.5
Chamber Dimensions (wxdxh)	Inches	15.0 x 15.0 x 15.0	23.7 x 24.0 x 19.7	36.2 x 20.0 x 25.7	19.2 x 20.0 x 25.7	19.2 x 20.0 x 25.7
	cm	38.1 x 38.1 x 38.1	60.3 x 60.9 x 50.2	92.0 x 50.8 x 65.4	48.9 x 50.8 x 65.4	48.9 x 50.8 x 65.4
Incubator Chamber Capacity	cu ft	2	6.5	10.8	5.7	5.7 (each)
	L	56	184	306	162	162 (each)
Number of Shelves	Included	2	2	6	3	3 (each)
	Maximum	6	9	12	12	12 (each)

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Microbiological Incubators
Air Jacket Economy Line

Precise Temperature Control - Superior Uniformity

- Temperature Uniformity +/- 0.5°C at 37°C
- Temperature Range Ambient + 5.0°C to 70°C

SHEL LAB general purpose incubators deliver a degree of temperature uniformity usually found only in more expensive, application specific models. Efficient gravity convection heating is supplied by economical low watt density heating elements. The heating element has a unique shield to protect against spills inside the chamber.

The rigidly constructed chamber is insulated with 2" of industrial fiberglass, minimizing heat loss and maximizing temperature uniformity.

The hydraulic controller is dependable and regulates the chamber temperature. Incubator door, with its positive door latch, is tightly sealed with a 1/2" silicone gasket. Glossy white interior enables easy contamination detection.

Model SMI1EM has a microprocessor based controller, for greater temperature accuracy.

1 Year Limited Warranty!

General Purpose Economy Incubators	Details	Model Number SMI1E	SMI1EM	SMI2E	SMI4E	SMI6E
Exterior Dimensions (wxdxh)	Inches	16.8 x 17.8 x 22.3	16.8 x 17.8 x 22.3	21.5 x 18.0 x 25.5	21.0 x 24.5 x 31.5	24.5 x 25.0 x 36.5
	cm	42.7 x 45.2 x 56.6	42.7 x 45.2 x 56.6	54.6 x 45.7 x 64.8	53.4 x 61.2 x 80.0	62.3 x 63.5 x 92.8
Chamber Dimensions (wxdxh)	Inches	12.0 x 12.0 x 14.0	12.0 x 12.0 x 14.0	17.0 x 12.0 x 17.0	16.0 x 19.0 x 22.0	20.0 x 19.0 x 28.0
	cm	30.4 x 30.4 x 35.5	30.4 x 30.4 x 35.5	43.1 x 30.4 x 43.1	40.6 x 48.2 x 55.8	50.8 x 48.2 x 71.1
Incubator Chamber Capacity	cu ft	1	1	2	4	6
	L	28	28	56	113	170
Number of Shelves	Included	2	2	2	2	2
	Maximum	6	6	6	6	6

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Microbiological Incubators
Large Capacity

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Temperature Uniformity +/-0.8°C at 37°C
- Temperature Range Ambient +8°C to 70°C

Optimized Floor Space

Large capacity incubators provide 30.8 & 38.6 cu.ft. chamber capacities while minimizing the amount of floor space used. Both models incorporate our microprocessor controller to achieve precise temperature uniformity. An independent, secondary temperature controller offers the added security of over temperature protection. The chamber floors are reinforced to support roller apparatus or shakers. Both models are supplied with six shelves capable of supporting the weight of benchtop instruments.

Features

- Four (4) Interior Electrical Outlets
- Prewired Panel for Chart Recorder
- Tempered Glass Viewing Window

2 Year Limited Warranty!

Large Capacity/Reach-In General Purpose Incubators	Details	Model Number SMI31	Model Number SMI39
Exterior Dimensions (wxdxh)	Inches	38.5 x 34.0 x 75.3	41.5 x 34.8 x 87.5
	cm	97.8 x 86.4 x 191.2	105.4 x 88.3 x 222.3
Chamber Dimensions (wxdxh)	Inches	32.2 x 26.0 x 63.5	35.0 x 26.0 x 73.2
	cm	81.9 x 66.0 x 161.2	88.9 x 66.0 x 186.0
Incubator Chamber Capacity	cu ft	30.8	38.6
	L	872	1092
Interior Outlet	Number	110V-4/ 220V-4	110V-4 / 220V-4
Access Port	Number	1	1
Number of Shelves	Included	6 (16 max)	6 (20 max)

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Refrigerated Incubators

SHEL LAB Refrigerated Incubators (often called B.O.D. Incubators or Low Temperature Incubators) are commonly used for applications such as B.O.D. Determinations, Plant and Insect Studies, Fermentation Studies, and Bacterial Culturing.

Units are equipped with a hermetically-sealed compressor, a circuit breaker to protect from electrical overload, and an easy-to-clean, fully insulated chamber. Gentle, continuous forced-air circulation ensures temperature uniformity and reproducible test conditions.

BOD Incubator Applications

- APHA Method at 20°C
- Plant Cell Growth
- Fermentation Studies
- Bacterial Culturing
- Mycology Studies

BOD Application

Biochemical Oxygen Demand (B.O.D.) incubators enable end users to determine levels of organic matter and nitrogen in wastewater samples. This wastewater must be effectively measured for contaminates, treated and then released back into the environment without posing a threat to the water supply system. Increased enforcement by government regulatory agencies charged with monitoring air and water quality has forced a greater number of organizations to actively test and treat their wastewater. B.O.D. incubators facilitate the storage of wastewater samples, and the SHEL LAB Low Temperature Incubators accommodate from 62 to 345 BOD bottles.

Refrigerated Incubators
Under Counter

The SRI3 space-saving low temperature incubator is ideal for small volume workloads and meets APHA specifications for Biochemical Oxygen Demand (B.O.D.) analysis.

Units are equipped with a hermetically-sealed compressor, a circuit breaker to protect from electrical overload, and an easy-to-clean fully insulated chamber that is corrosion-resistant. Gentle, continuous forced-air circulation ensures temperature uniformity and reproducible test conditions. Each unit also includes an independent over temperature safety controller, two shelves (adjustable in two inch increments), and a one amp interior outlet to allow the use of shakers, stirrers, roller bottles or other apparatus. This unit has a steel exterior with welded seams and corners and a double-coated, baked enamel finish. It is supplied with adjustable leveling feet and a condensation drip tray.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Temperature Uniformity +/- 0.5°C at 20°C
- Temperature Range 0°C to 45°C at 20°C Ambient

2 Year Limited Warranty!

Refrigerated Incubators	Details	Model Number SRI3
Exterior Dimensions (wxdxh)	Inches	24.0 x 21.0 x 33.8
	cm	61.0 x 53.4 x 85.8
Chamber Dimensions (wxdxh)	Inches	16.0 x 12.0 x 21.5
	cm	40.6 x 30.4 x 54.6
Incubator Chamber Capacity	cu ft	2.4
	L	68
Interior Outlet	Number	110V -1 / 220V-2
Bottle Capacity	Number	62
Number of Shelves	Included	2

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V).
Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Refrigerated Incubators
Large Capacity

The SHEL LAB Low Temperature Incubators have a temperature range of 20°C degrees below ambient to 45°C. The Refrigerated Incubators also include an independent over temperature safety controller, adjustable shelves in two inch increments and a one amp interior outlet to allow the use of shakers, stirrers, roller bottles or other apparatus.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Temperature Uniformity +/- 0.5°C at 20°C
- Temperature Range 0°C to 45°C at 20°C Ambient

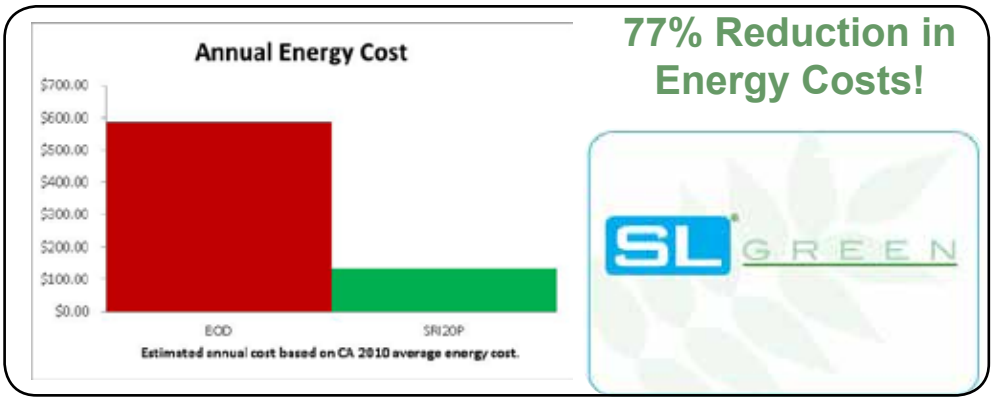
2 Year Limited Warranty!

Refrigerated Incubators	Details	Model Number SRI20
Exterior Dimensions (wxdxh)	Inches	34.5 x 34.5 x 77.5
	cm	87.7 x 87.7 x 196.9
Chamber Dimensions (wxdxh)	Inches	27.0 x 23.0 x 56.5
	cm	68.5 x 58.4 x 143.5
Incubator Chamber Capacity	cu ft	20.3
	L	574
Interior Outlet	Number	110V-1 / 220V -2
Bottle Capacity	Number	345
Number of Shelves	Included	4

**All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology. We reserve the right to change specifications at any time.

Refrigerated Incubators
Peltier Cooled Series

Innovative peltier cooling technology, eliminates the need for a refrigeration compressor in the peltier cooled series. These units use 78% less power than alternative models and reduce room air conditioning loads by 75%. They also include 75 pound capacity shelves, which eliminates sagging. These incubators meet APHA specifications for Biochemical Oxygen Demand (B.O.D.) and include a mechanical convection system to ensure even air distribution, digital temperature set controller, over temperature limit control, and a digital temperature display.



2 Year Limited Warranty!

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Temperature Uniformity +/- 0.5°C at 20°C
- Temperature Range 15°C to 40°C at 20°C Ambient

BOD Incubators		Model Number	
		SRI6P	SRI20P
	Details	Under Counter	Large Capacity
Exterior Dimensions (wxdxh)	Inches	30.0 x 31.5 x 33.5	30.0 x 31.5 x 69.5
	cm	76.2 x 80.1 x 85.1	76.2 x 80.1 x 176.6
Chamber Dimensions (wxdxh)	Inches	25.5 x 24.0 x 18.5	25.5 x 24.0 x 54.5
	cm	64.7 x 60.9 x 46.9	64.7 x 60.9 x 138.4
Incubator Chamber Capacity	cu ft	6.5	19.3
	L	185	546
Interior Outlet	Number	120V -1 / 230V-2	120V-1 / 230V -2
Bottle Capacity	Number	120	300
Number of Shelves	Included	2	5

**All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Refrigerated Incubators
Diurnal Plant Chamber

SHEL LAB Diurnal growth chambers are designed for studies requiring day and nighttime simulation. This unit features dual-program selector dials, which allows control of two temperature conditions and an ON/OFF illumination cycle relative to the program selected. Each system operates independently allowing for simulation of a diurnal cycle, such as an eight hour day cycle of 30°C with light followed by a sixteen hour night cycle of 18°C without light. Forced air circulation ensures the most reproducible test conditions. The chamber air is gently and continuously circulated at a rate that ensures temperature uniformity of all test samples.

The unit is equipped with a hermetically-sealed compressor and an independent over temperature safety controller. In addition, a one amp interior outlet allows use of shakers, stirrers, roller bottles or other apparatus. This chamber is ideal for plant growth studies.

- LED Display of Set point and Chamber Temperature
- High and Low Limit Temperature Protection
- Day/Night Light and Temperature Control
- Fan Assisted/Forced-Air Circulation
- Hermetically Sealed Compressor
- Interior Electrical Outlet

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Temperature Range 0°C to 45°C at 20°C Ambient
- Temperature Uniformity +/- 0.5°C at 20°C

2 Year Limited Warranty!

Diurnal Plant Chamber	Details	Model Number SRI21D
Exterior Dimensions (wxdxh)	Inches	34.5 x 34.5 x 77.5
	cm	87.7 x 87.7 x 196.9
Chamber Dimensions (wxdxh)	Inches	27.0 x 23.0 x 56.5
	cm	68.5 x 58.4 x 143.5
Incubator Chamber Capacity	cu ft	20.3
	L	574
Interior Outlet	Number	110V - 1 / 220V - 2
Number of Shelves	Included	4
Bottle Capacity	Number	317

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Refrigerated Incubators
Drosophila Chamber

The SHEL LAB Drosophila specific low temperature incubator that takes advantage of the range of temperatures acceptable in Drosophila culture allowing the condensing coil adequate cycling time, thus avoiding ice build-up. This incubator addresses all of the major performance issues associated with other fly-specific incubators on the market.

This incubator functions within the range of temperature preferred by fruit flies. The elements only activate if the chamber temperature goes below the programmed lowest acceptable level. The compressor will shut off and rest while the chamber temperature slowly rises in response to a door opening or heat from fan or optional light. This results in a longer lasting unit with less maintenance, reduced heat output and less noise from the compressor.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Temperature Range 0°C to 29°C

- Microprocessor controlled interior light mimics diurnal cycles that foster breeding
- Conformal coated refrigeration coils
- Robust, programmable heating and cooling control

Drosophila Chamber	Details	Model Number SRI21F Drosophila	SRI21FV Viewing Door
Exterior Dimensions (wxdxh)	Inches	34.5 x 34.5 x 77.5	34.5 x 34.5 x 77.5
	cm	87.7 x 87.7 x 196.9	87.7 x 87.7 x 196.9
Chamber Dimensions (wxdxh)	Inches	27.0 x 23.0 x 56.5	27.0 x 23.0 x 56.5
	cm	68.5 x 58.4 x 143.5	68.5 x 58.4 x 143.5
Incubator Chamber Capacity	cu ft	20.3	20.3
	L	574	574
Interior Outlet	Number	110V - 1 / 220V - 2	110V - 1 / 220V - 2
Number of Shelves	Included	8	8

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

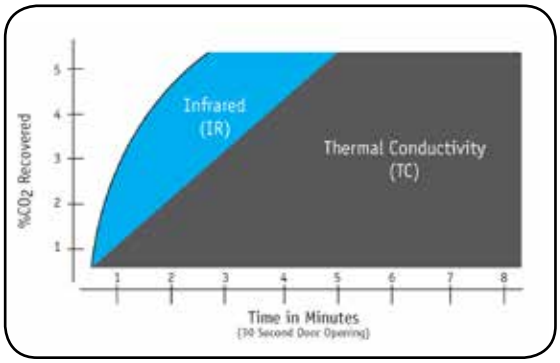
CO_2 Incubators

CO2 Incubator Applications

- Cell and Tissue Culture
- Immunology
- Genetic Engineering
- Protein Synthesis
- Virology
- Neurology
- Pharmacology
- In vitro Fertilization
- Human Vaccines
- Veterinary Vaccines
- Carcinogenicity Testing
- Monoclonal Antibodies

Infrared (IR) CO2 Sensors

For the fastest CO_2 recovery and most stable performance, this series features IR sensors.



Contamination Control

Extensive use of copper in the CO2 sample port, heated CO2 feed line, housing of the patented HEPA filtration system, and humidity pan with a copper SL decontamination token, adds reassurance that foreign microbes will not affect test results. Cleanup is a breeze with the all stainless steel chamber, and autoclavable door gasket. Optional copper shelves are available for most units for even more contamination control.

copper shelving option

CO2 Incubators
Water Jacket Economy

The SHEL LAB economy incubator was designed and manufactured to accommodate tight budgets, while maintaining the fundamental needs of quality and precision. These units have PID microprocessor controllers, a heated outer door and a tempered-glass inner door. They provide exceptional temperature uniformity, while minimizing cold spots that lead to condensation and ultimately, contamination. Although they do not have a humidity display, the extremely stable temperature environment maintains constant humidity through evaporation at up to 95%.

The audible/visual alarms for temperature and CO2 respond to out-of-tolerance conditions. They offer an independent overtemperature safety control to protect samples from overheating, and an optional CO2 tank switch/alarm to prevent prematurely exhausting the gas supply.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature & CO2 Alarm
- Inner Glass Viewing Door
- Temperature Uniformity +/- 0.2°C at 37°C
- Temperature Range Ambient + 8°C to 60°C
- CO2 Range 0 - 20%
- CO2 Recovery Rate < 5 Minutes
- Relative Humidity Up to 80%

Tissue & Cell Culture Applications

These incubators control three essential variables related to replicating the mammalian environment;

- Stable CO2 Level
- Controlled Temperature
- Relative Humidity (RH)

copper shelving option

5 Year Limited Warranty!

We stand behind our product quality, ShellLab CO_2 Incubators come with the most extensive warranties in the industry.

- 5 years parts (labor included in the US)
- 7 years IR sensor
- Lifetime water jacket chamber

Water Jacket CO2 Incubators	Details	Model Number SCO6WE Floor Model	SCO12WE Dual/Stacked Chambers
Exterior Dimensions (wxdxh)	Inches	26.0 x 26.3 x 40.3	26.0 x 25.8 x 51.0
	cm	66.0 x 66.7 x 102.3	66.1 x 65.5 x 203.2
Chamber Dimensions (wxdxh)	Inches	20.2 x 19.7 x 25.2	20.5 x 20.0 x 25.5 each
	cm	51.4 x 50.1 x 64.1	52.0 x 50.8 x 64.7 each
Incubator Chamber Capacity	cu ft	6	12 (6 each)
	L	165	342
Number of Shelves	Included	3 (16 Max)	3 each chamber (16 Max)

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

CO2 Incubators
Water Jacket Series

This series of CO2 Incubators offer dependable Infrared (IR) CO2 Sensor control and are ideal for sensitive tissue and cell culture applications. They provide the benefits of contamination control and uncompromising temperature uniformity for even the most demanding incubations.

Precision is easily maintained with push-button calibration of both temperature and CO2, and audio/visual alarms that signal high/low temperature and CO2 conditions. Modular controls and backup systems ensure confidence for incubating valuable samples, providing the dependable assurance you expect from a SHEL LAB incubator.

Patented Copper Coated HEPA Filter

A "Bacteriostatic" copper cage to trap particulate matter and reduce potential for chamber contamination. This filter removes 99.97% of all airborne microbes and isolated microbes 0.3 microns or larger.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature & CO2 Alarm
- Inner Glass Viewing Door
- Temperature Uniformity +/- 0.2°C at 37°C
- Temperature Range Ambient + 8°C to 60°C
- CO2 Range 0 - 20%
- Infrared (IR) CO2 Sensor Accuracy +/- 0.1%
- CO2 Recovery Rate < 5 Minutes
- Relative Humidity Up to 95%

5 Year Limited Warranty!

We stand behind our product quality, ShelLab CO_2 Incubators come with the most extensive warranties in the industry.

- 5 years parts (labor included in the US)
- 7 years IR sensor
- Lifetime water jacket chamber

Water Jacket CO2 Incubators	Details	Model Number SCO2W Bench Top	SCO5W Floor Model	SCO10W Dual/Stacked Chambers
Exterior Dimensions (wxdxh)	Inches	21.0 x 22.5 x 27.0	26.0 x 25.5 x 40.3	26.0 x 25.5 x 80.6
	cm	53.4 x 57.2 x 68.6	66.1 x 64.8 x 102.3	66.1 x 64.8 x 204.6
Chamber Dimensions (wxdxh)	Inches	15.7 x 15.7 x 10.2	19.2 x 19.7 x 23.0	19.2 x 19.7 x 23.0 each
	cm	40.0 x 40.0 x 26.0	48.9 x 50.1 x 58.4	48.9 x 50.1 x 58.4 each
Incubator Chamber Capacity	cu ft	1.5	5	10 (5 each)
	L	42	143	286
Number of Shelves	Included	3	3	3 each

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

CO2 Incubators
High Heat Decontamination

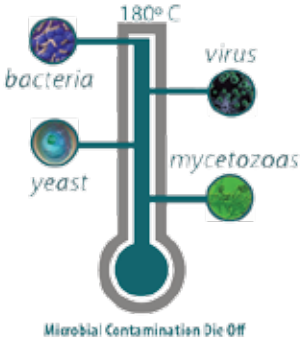
Protect your samples with high heat decontamination!

The model SCO6AD features a dry heat decontamination cycle, that maintains 180°C for 120 minutes. This industry best time and temperature relationship satisfies all global standards for decontamination.

This is decontamination at its fastest easiest and most effective - it is not necessary to remove the IR CO_2 sensor prior to activating the decontamination process and we feature the shortest cycle time on the market. This is a more convenient approach and eliminates potential damage to the sensitive IR sensor.

Other features of the SCO6AD include a USB interface for software communication, precise temperature control microprocessor and an independent over temperature safety controller.

The SHEL LAB SCO6AD is designed to stop microbial contamination caused by mycetozoa, yeast, viruses, and bacteria and the range of other microorganisms that thrive in incubator environments.



5 Year Limited Warranty!

We stand behind our product quality, Shel Lab CO_2 Incubators come with the most extensive warranties in the industry.

- 5 years parts (labor included in the US)
- 7 years IR sensor

Incubators, Infrared (IR) CO_2 Sensor	Details	Model Number SCO6AD
Exterior Dimensions (wxdxh)	chesDe	28.8 x 30.3 x 39.3
	cm	73.1 x 76.8 x 99.7
Chamber Dimensions (wxdxh)	Inches	20.2 x 20.0 x 25.5
	cm	51.4 x 50.8 x 64.7
Incubator Chamber Capacity	cu ft	5.9
	L	167
CO_2 Range	%	0-20%
CO_2 Sensor Accuracy	at 5%	+/-0.1%
CO_2 Recovery Rate	to 5%	<5 Minutes to 95% of Setpoint
Relative Humidity	at 37°C	Up to 95%
Temperature Range	Celsius	Ambient +5°C to 60°C
Temperature Uniformity	Celsius	+/- 0.25°C at 37°C
Over Temperature Protection	Yes/No	Yes
Temperature Alarm	Yes/No	Yes
CO_2 Alarm	Yes/No	Yes
Number of Shelves	Included	3

Al specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

CO2 Incubators
Air Jacket Series

The air jacket series of CO2 Incubators offer dependable Infrared (IR) CO2 Sensor control and are ideal for sensitive tissue and cell culture applications. They provide the benefits of contamination control and uncompromising temperature uniformity for even the most demanding incubations.

Precision is easily maintained with push-button calibration of both temperature and CO2, and audio/visual alarms that signal high/low temperature and CO2 conditions. Modular controls and backup systems ensure confidence for incubating valuable samples, providing the dependable assurance you expect from a SHEL LAB incubator.

Patented Copper Coated HEPA Filter

A "Bacteriostatic" copper cage to trap particulate matter and reduce potential for chamber contamination. This filter removes 99.97% of all airborne microbes and isolated microbes 0.3 microns or larger.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature & CO2 Alarm
- Inner Glass Viewing Door
- Temperature Uniformity +/- 0.25°C at 37°C
- Temperature Range Ambient + 8°C to 60°C
- CO2 Range 0 - 20%
- Infrared (IR) CO2 Sensor Accuracy +/- 0.1%
- CO2 Recovery Rate < 5 Minutes
- Relative Humidity Up to 95%

5 Year Limited Warranty!

We stand behind our product quality, ShelLab CO_2 Incubators come with the most extensive warranties in the industry.

- 5 years parts (labor included in the US)
- 7 years IR sensor

Copper Shelf Options	Part Number
Copper Shelf Kit (3 Shelves, 6 Slides)	89409-632
Copper Shelf Slides	89409-594
Copper Shelf	89409-592

Air Jacket CO2 Incubators	Details	Model Number SC05A Standard	SC010A Dual/Stacked Chambers
Exterior Dimensions (wxdxh)	Inches	27.3 x 28.0 x 37.8	27.3 x 28.0 x 75.6
	cm	69.3 x 71.2 x 95.9	69.3 x 71.2 x 191.8
Chamber Dimensions (wxdxh)	Inches	20.5 x 19.7 x 21.5	20.5 x 19.7 x 21.5 each
	cm	52.0 x 50.1 x 54.6	52.0 x 50.1 x 54.6 each
Incubator Chamber Capacity	cu ft	5	10 (5 each)
	L	142	284
Number of Shelves	Included	3	3 each

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

CO2 Incubators
Air Jacket Large Capacity

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Temperature Uniformity +/- 0.5°C at 37°C
- Temperature Range Ambient + 8°C to 60°C
- CO_2 Range 0 - 20%
- Infrared (IR) CO2 Sensor Accuracy +/- 0.1%
- CO_2 Recovery Rate < 5 Minutes

These units are well suited for roller bottle apparatus and high-volume tissue culture applications and are ideal for cell harvesting.

This large capacity incubator maximizes laboratory space in a convenient floor model design. Its chamber floor is specifically designed for easy movement of roller bottle apparatus by use of a flip-out ramp. Supplied with four one amp interior electrical outlets and gentle mechanical air convection that ensures excellent temperature uniformity, and eliminates cold spots. An infrared system accurately controls CO2 levels, provides fast CO2 recovery after door openings, and is not affected by temperature or humidity. This unit is supplied with six stainless steel shelves, which are adjustable on 1/2 inch increments.

SHEL LAB reach-in CO2 incubators are available in 31 cu.ft., 40 cu.ft., and 58 cu.ft. sizes.

2 Year Limited Warranty!

Air Jacket CO2 Incubators	Details	Model Number SC031	SC040	SC058
Exterior Dimensions (wxdxh)	Inches	39.5 x 33.8 x 75.3	42.5 x 34.5 x 87.0	51.0 x 44.8 x 80.0
	cm	100.4 x 85.8 x 191.2	108.0 x 87.7 x 221	129.6 x 113.7 x 203.2
Chamber Dimensions (wxdxh)	Inches	32.7 x 26.0 x 63.0	35.0 x 26.0 x 75.5	43.0 x 34.5 x 67.5
	cm	83.1 x 66.0 x 160.0	88.9 x 66.0 x 191.7	109.2 x 87.6 x 171.4
Incubator Chamber Capacity	cu ft	31	40	58
	L	879	1125	1641
Number of Shelves	Included	6	6	6
Interior Outlet	Included	4	4	4

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Humidified CO2 Incubators

SHEL LAB Model SCO26H, is the newest addition to the large capacity CO2 incubator line. This 26.1 cubic foot incubator features active humidity control up to 95%.

This unit has exceptional CO2 and temperature uniformity, along with a user controllable humidity system that is more accurate and responsive to door openings than a traditional water pan humidity system.

The SCO26H humidity system provides less evaporation of culture media and eliminates a potential source for contamination. Contamination is minimized by the heated glass door and an antimicrobial copper drain.

The triple-paned glass door allows for easy viewing of samples without having to open the incubator door, so samples can thrive in the stable environment within the chamber.

A gentle horizontal airflow heating system is used for quick temperature recovery after door openings. This airflow system obtains superior temperature control with minimal drying or disturbance of sample conditions. The CO2 is accurately controlled with an IR sensor, providing overall CO2 stability.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Temperature Uniformity +/- 0.5°C at 37°C
- Temperature Range Ambient + 5°C to 50°C
- CO2 Range 0 - 20%
- Infrared (IR) CO2 Sensor Accuracy +/- 0.1%
- CO2 Recovery Rate < 5 Minutes
- Relative Humidity Up to 95%
- Temperature & CO2 Alarms

2 Year Limited Warranty!

Humidified CO2 Incubators	Model Number	
	Details	SCO26H
Exterior Dimensions (wxdxh)	Inches	39.3 x 37.0 x 78.5
	cm	99.7 x 94.0 x 199.4
Chamber Dimensions (wxdxh)	Inches	30.7 x 26.0 x 56.5
	cm	78.1 x 66.0 x 143.5
Incubator Chamber Capacity	cu ft	26.1
	L	740
Number of Shelves	Included	6

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Humidity Chambers

SHEL LAB Humidity Test Cabinets provide a controlled environment for a wide range of industrial and biotechnology testing applications. This line is designed to duplicate a natural condition, which allows testing the limitations of a sample when exposed to various temperature and moisture fluctuations.

Microprocessor controls maintain temperature and humidity in approximate ranges of 35-70°C and 40-95%RH, respectively. An extra large water jacket minimizes condensation inside the chamber and supports optimum temperature uniformity.

Humidity is controlled by utilizing a low-pressure water vapor generator injecting saturated water vapor into the recirculating air duct. This process

2 Year Limited Warranty!

Humidity Cabinets	Details	Model Number	
		Medium (SHC10)	Large (SHC28)
Exterior Dimensions (wxdxh)	Inches	44.3 x 32.8 x 57.0	42.5 x 37.0 x 85.0
	cm	112.4 x 83.2 x 144.8	108.0 x 94.0 x 215.9
Chamber Dimensions (wxdxh)	Inches	30.0 x 21.0 x 30.0	30.2 x 26.0 x 62.0
	cm	76.2 x 53.3 x 76.2	76.8 x 66.0 x 157.4
Incubator Chamber Capacity	cu ft	10	28
	L	309	799
Temp. Range	Celsius	40°C to 70°C	Ambient +10°C to 70°C
Temp. Uniformity	Celcsus	+/-0.5°C at 37°C	+/-0.5°C at 37°C
Relative Humidity	Percent	Ambient + 10% to 95%	Ambient + 10% -to 95%
Number of Shelves	Included	3	6

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Humidity Chambers
Refrigerated

SHEL LAB Humidity Test Cabinets provide a controlled environment for a wide range of industrial and biotechnology testing applications. This line is designed to duplicate a natural condition, which allows testing the limitations of a sample when exposed to various temperature and moisture fluctuations. These humidity test chambers incorporate a refrigeration system that dramatically increases the operational range of the cabinet

Microprocessor controls maintain temperature and humidity in approximate ranges of 10-70°C and 40-95%RH, respectively. An extra large water jacket minimizes condensation inside the chamber and supports optimum uniformity conditions.

A low-pressure water vapor generator, injecting saturated water vapor into the recirculating air duct, controls chamber humidification. This process is preferable to steam generation because steam introduces additional heat to the chamber atmosphere, which then compromises temperature control.

2 Year Limited Warranty!

Humidity Cabinets Refrigerated	Details	Model Number Medium (SHC10R)	Large (SHC28R)
Exterior Dimensions (wxdxh)	Inches	44.3 x 32.8 x 57.0	42.5 x 37.0 x 85.0
	cm	112.4 x 83.2 x 144.8	108.0 x 94.0 x 215.9
Chamber Dimensions (wxdxh)	Inches	30.0 x 21.0 x 30.0	30.2 x 26.0 x 62.0
	cm	76.2 x 53.3 x 76.2	76.8 x 66.0 x 157.4
Incubator Chamber Capacity	cu ft	10	28
	L	309	799
Temp. Range	Celsius	40°C to 70°C	10°C to 70°C
Temp. Uniformity	Celsius	+/- 0.5°C at 37°C	+/- 0.5°C at 37°C
Relative Humidity	Percent	Ambient + 10% to 95%	Ambient + 10% to 95%
Number of Shelves	Included	3	6

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Shaking Incubators

Shaking incubators, also know as environmental shakers, are often used for cell culturing, cell aeration, and solubility studies. In addition to stable temperature conditions, they use an orbital agitation at variable speeds to affect the growth of cell cultures.

This is why the SHEL LAB SI6 and SI6R Shaking Incubators have adjustable stroke lengths to accommodate various cells and applications. This gives flexibility in adjusting the speed and orbit to meet each application.

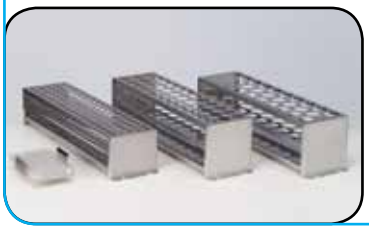
Most models come equipped with a universal shaking platform, independent alarms, and microprocessor controls for temperature and speed adjustment.

Applications include:

- Cell Cultures
- Cell Aeration
- Microbiology
- Increasing Solubility Rates
- Metabolism Studies
- Bacterial Cultures
- Bacteriology

Full Selection of Accessories

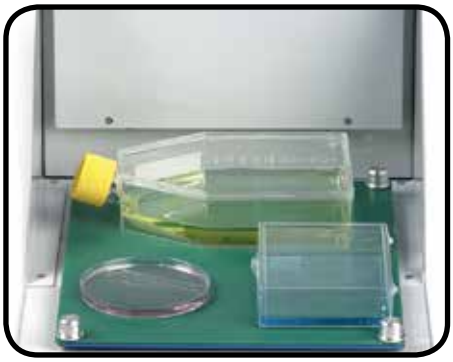
The easily removable rotation platform is included with each SHEL LAB unit.



Flask holders and accessories can be arranged in several combinations on the platform according to what best suits your application.

Shaking Incubators
Mini Shaker

The SHEL LAB Mini Shaker is the most compact shaking incubator in its class. The standard platform (included) features a non-slip, rubber coated surface, ideal for tissue culture flasks, petri dishes and staining trays. A convenient universal magnetic platform is also available for use with Erlenmeyer flasks and test tube racks. The unique, magnetic attachment method is the easiest way to instantly change between flask clamps of different sizes.



Flask holders and accessories can be arranged in several combinations on the platform according to what best suits your application.

A constant monitoring system verifies and maintains accuracy through the duration of the program. Sophisticated over-temperature and over-speed controls ensure long life, safety and sample integrity.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Orbital-Shaking Speed Alarm
- Digital Timer

Shaking Incubator	Details	Model Number SSI2
Exterior Dimensions (wxdxh)	Inches	11.3 x 15.8 x 11.5
	cm	28.6 x 40.1 x 29.3
Chamber Dimensions (wxdxh)	Inches	11.0 x 13.2 x 8.0
	cm	27.9 x 33.6 x 20.3
Incubator Chamber Capacity	cu ft	0.5
	L	13
Temperature Range	Celsius	5°C + Ambient to 60°C Increments of 0.1°C
Temperature Uniformity	Celsius	+/- 0.25%
Orbital-Shaking Range	RPM	30-300
Timer Functionality	Minutes	1-999

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Shaking Incubators
Bench Model

The SSI3 has a transparent hood that lifts up via a hydraulic system, so it can function in tight places. All of our shaking incubators feature stainless steel interiors which provide excellent durability and an easy-to-clean surface. Each unit has an easy-to-read LED display. The rotation platform is included with each unit and is self-centering for easy installation. The SSI3 includes a convenient, user adjustable counterbalance that provides optimal load flexibility

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Orbital-Shaking Speed Alarm
- Digital Timer
- Adjustable Orbit

2 Year Limited Warranty!

Shaking Incubator	Details	Model Number SSI3
Exterior Dimensions (wxdxh)	Inches	22.0 x 25.5 x 28.0
	cm	55.9 x 64.8 x 71.2
Chamber Dimensions (wxdxh)	Inches	19.0 x 18.0 x 16.5
	cm	48.2 x 45.7 x 41.9
Incubator Chamber Capacity	cu ft	3.3
	L	92
Temperature Range	Celsius	8°C + Ambient to 60°C
Temperature Uniformity	Celsius	+/-0.5°C at 37°C
Orbital-Shaking Range	RPM	30-400
Timer Functionality	Minutes	1-999

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V).
Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Shaking Incubators
Floor Model

Versatile and Robust

Shaking incubators, also know as environmental shakers, are often used for cell culturing, cell aeration, and solubility studies. In addition to stable temperature conditions, they use an orbital agitation at variable speeds to affect the growth of cell cultures.

This is why the SHEL LAB Shaking Incubators have adjustable stroke lengths to accommodate various cells and applications. This gives flexibility in adjusting the speed and orbit to meet each application. The SSI5R-HS achieves speeds up to 850 RPMs.

All models come equipped with a universal shaking platform, independent alarms, and microprocessor controls for temperature and speed adjustment.

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Orbital-Shaking Speed Alarm

2 Year Limited Warranty!

CE TÜV SÜD NRTL C US

Shaking Incubators	Details	Model Number SSI5 Floor	SSI5R Floor Refrigerated	SSI5R-HS High RPM
Exterior Dimensions (wxdxh)	Inches	28.5 x 29.5 x 40.5	28.5 x 29.5 x 40.5	28.5 x 29.5 x 40.5
	cm	72.4 x 75.0 x 102.9	72.4 x 75.0 x 102.9	72.4 x 75.0 x 102.9
Chamber Dimensions (wxdxh)	Inches	19.0 x 20.5 x 22.5	19.0 x 20.5 x 22.5	19.0 x 20.5 x 22.5
	cm	48.2 x 52.0 x 57.1	48.2 x 52.0 x 57.1	48.2 x 52.0 x 57.1
Incubator Chamber Capacity	cu ft	5	5	5
	L	144	144	144
Temperature Range	Celsius	8°C + Ambient to 60°C	10°C to 60°C	10°C to 60°C
Temperature Uniformity	Celsius	+/-0.8°C at 37°C	+/-0.8°C at 37°C	+/-0.8°C at 37°C
Platform Capacity	lbs (kg)	22 (10)	22 (10)	22 (10)
Orbital-Shaking Range	RPM	30-400	30-400	30-850
Timer Functionality	Minutes	1-999	1-999	1-999
Number of Shelves	Included	1	1	1

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.

Shaking Incubators
Large Capacity

The SHEL LAB SSI10 delivers all the features you appreciate in the SHEL LAB Shaking Incubator line, with even greater load capacity. The unit performs from 30-400 RPM's with a smooth, quiet oscillation. The door of the SSI10 is designed with hydraulic pistons making it easy to lift during loading and unloading.

These state of the art orbital shaking incubators feature a universal shaker platform, which accommodates a wide range of flask clamps, test tube racks and micro titer plate clamps. To support the loads over many years of use, four load-bearing positions are incorporated for optimal weight distribution.

For maximum load flexibility, the unique counter-balanced weighting system is adjustable to accommodate off-center loads and varying capacities.

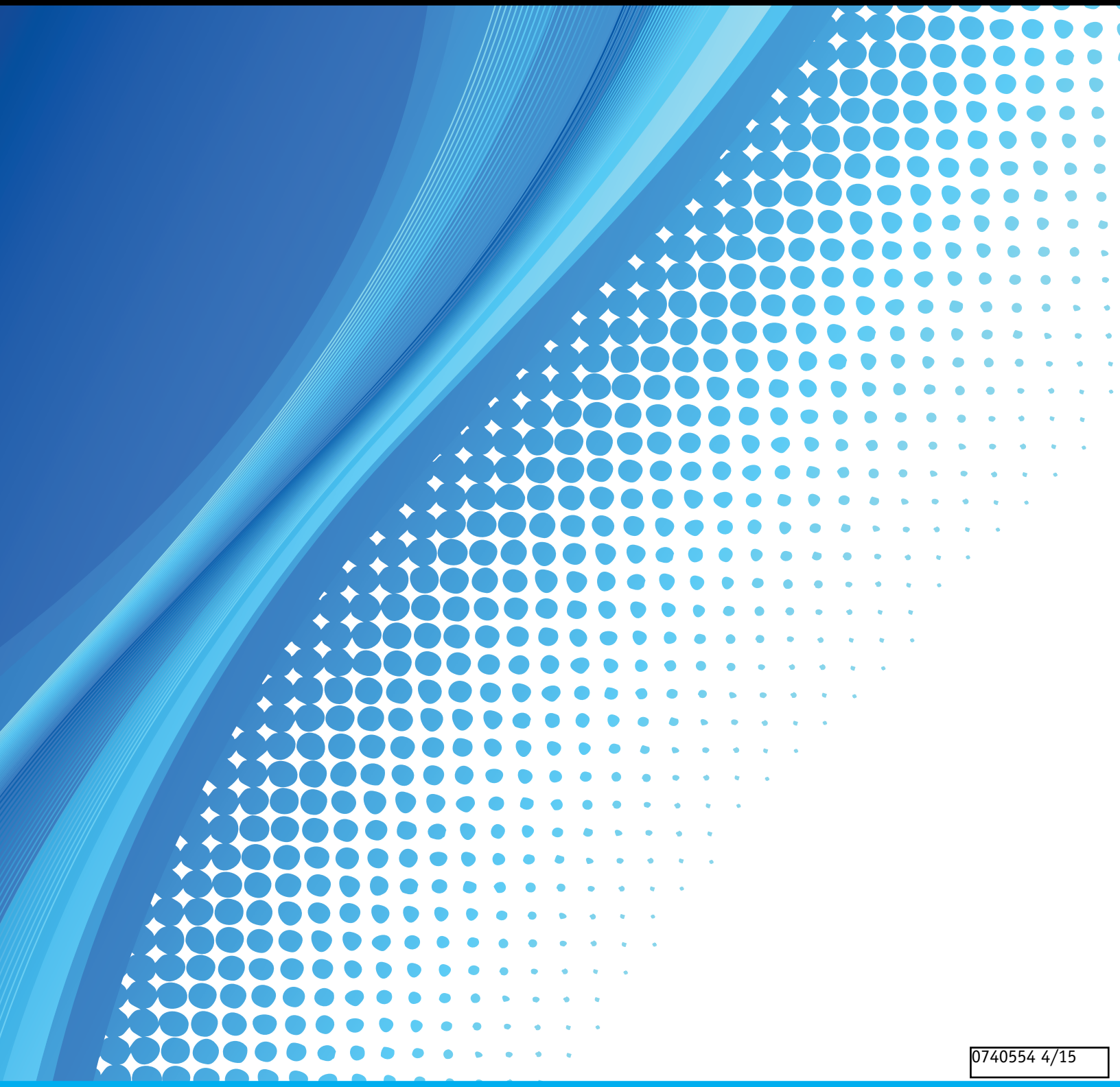
2 Year Limited Warranty!

Precise Temperature Control - Superior Uniformity

- Independent Over Temperature Thermostat
- Over Temperature Alarm
- Orbital-Shaking Speed Alarm

Shaking Incubators	Details	Model Number SSI10 Standard	SSI10R Refrigerated
Exterior Dimensions (wxdxh)	Inches	55.0 x 33.5 x 33.5	55.0 x 33.5 x 33.5
	cm	139.7 x 85.1 x 85.1	139.7 x 85.1 x 85.1
Chamber Dimensions (wxdxh)	Inches	35.5 x 25.5 x 19.7	35.5 x 25.5 x 19.7
	cm	90.1 x 64.7 x 50.1	90.1 x 64.7 x 50.1
Incubator Chamber Capacity	cu ft	10.3	10.3
	L	293	293
Platform Dimensions	Inches	32 x 22 x 3	32 x 22 x 3
	cm	81 x 56 x 7	81 x 56 x 7
Temperature Range	Celsius	8°C + ambient to 60°C	10°C - 60°C
Temperature Uniformity	Celsius	+/-0.5°C at 37°C	+/-0.5°C at 37°C
Platform Weight Capacity	lbs (kg)	45 (20)	45 (20)
Orbital-Shaking Range	RPM	30-400	30-400
Timer Functionality	Minutes	1-999	1-999

All specifications are determined by using average values on standard equipment at an ambient temperature of 25°C (77°F) and line voltages within +/-10% of unit type (115V/230V). Temperature specifications follow DIN 12880 methodology.
We reserve the right to change specifications at any time.



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SHEL LAB®

SAFETY DATA SHEET

B-PER® Bacterial Protein Extraction Reagent

SECTION 1: Identification of the substance/mixture and of the company/ undertaking

1.1 Product identifier

Product name : B-PER® Bacterial Protein Extraction Reagent

Product code : 0078243 0078248 78248B 0078248S 0090084 0090084F 1861468 1861469 1862487 1900286

SDS # : 2673

Product description : Not available.

Product type : Liquid.

Other means of identification : Not available.

1.2 Relevant identified uses of the substance or mixture and uses advised against

Not applicable.

1.3 Details of the supplier of the safety data sheet

National contact

Thermo Fisher Scientific Europe
Perbio Science
Industriezone III
Industrielaan 27
9320 Erembodegem Belgium

Manufacturer

Thermo Fisher Scientific
Pierce Biotechnology
P.O. Box 117
Rockford, IL 61105
United States
815.968.0747 or
800.874.3723
7 AM - 5 PM Central Time (GMT -06:00)

e-mail address of person responsible for this SDS : QA.Rockford@thermofisher.com

1.4 Emergency telephone number

National advisory body/Poison Center

Telephone number : CHEMTREC: 703-527-3887
CHEMTREC UK: +(44) 870 8200418
National Poisons Information Service (UK Only): 0870 600 6266

SECTION 2: Hazards identification

2.1 Classification of the substance or mixture

Product definition : Mixture

Classification according to Regulation (EC) No. 1272/2008 [CLP/GHS]

Not classified.

The product is not classified as hazardous according to Regulation (EC) 1272/2008 as amended.

Classification according to Directive 1999/45/EC [DPD]

The product is not classified as dangerous according to Directive 1999/45/EC and its amendments.

Classification : Not classified.

See Section 16 for the full text of the R phrases or H statements declared above.
See Section 11 for more detailed information on health effects and symptoms.

2.2 Label elements

Signal word : No signal word.

Hazard statements : No known significant effects or critical hazards.

SECTION 2: Hazards identification

Precautionary statements

Prevention : Not applicable.

Response : Not applicable.

Storage : Not applicable.

Disposal : Not applicable.

Supplemental label elements : Not applicable.

Annex XVII - Restrictions on the manufacture, placing on the market and use of certain dangerous substances, mixtures and articles : Not applicable.

2.3 Other hazards

Other hazards which do not result in classification : None known.

SECTION 3: Composition/information on ingredients

Substance/mixture : Mixture

There are no ingredients present which, within the current knowledge of the supplier and in the concentrations applicable, are classified as hazardous to health or the environment, are PBTs or vPvBs or have been assigned a workplace exposure limit and hence require reporting in this section.

SECTION 4: First aid measures

4.1 Description of first aid measures

Eye contact : Immediately flush eyes with plenty of water, occasionally lifting the upper and lower eyelids. Check for and remove any contact lenses. Get medical attention if irritation occurs.

Inhalation : Remove victim to fresh air and keep at rest in a position comfortable for breathing. Get medical attention if symptoms occur.

Skin contact : Flush contaminated skin with plenty of water. Remove contaminated clothing and shoes. Get medical attention if symptoms occur.

Ingestion : Wash out mouth with water. Remove victim to fresh air and keep at rest in a position comfortable for breathing. If material has been swallowed and the exposed person is conscious, give small quantities of water to drink. Do not induce vomiting unless directed to do so by medical personnel. Get medical attention if symptoms occur.

Protection of first-aiders : No action shall be taken involving any personal risk or without suitable training.

4.2 Most important symptoms and effects, both acute and delayed

Potential acute health effects

Eye contact : No known significant effects or critical hazards.

Inhalation : No known significant effects or critical hazards.

Skin contact : No known significant effects or critical hazards.

Ingestion : No known significant effects or critical hazards.

Over-exposure signs/symptoms

Eye contact : No specific data.

Inhalation : No specific data.

Skin contact : No specific data.

Ingestion : No specific data.

4.3 Indication of any immediate medical attention and special treatment needed

SECTION 4: First aid measures

Notes to physician : Treat symptomatically. Contact poison treatment specialist immediately if large quantities have been ingested or inhaled.

Specific treatments : No specific treatment.

SECTION 5: Firefighting measures

5.1 Extinguishing media

Suitable extinguishing media : Use an extinguishing agent suitable for the surrounding fire.

Unsuitable extinguishing media : None known.

5.2 Special hazards arising from the substance or mixture

Hazards from the substance or mixture : In a fire or if heated, a pressure increase will occur and the container may burst.

Hazardous thermal decomposition products : No specific data.

5.3 Advice for firefighters

Special protective actions for fire-fighters : Promptly isolate the scene by removing all persons from the vicinity of the incident if there is a fire. No action shall be taken involving any personal risk or without suitable training.

Special protective equipment for fire-fighters : Fire-fighters should wear appropriate protective equipment and self-contained breathing apparatus (SCBA) with a full face-piece operated in positive pressure mode. Clothing for fire-fighters (including helmets, protective boots and gloves) conforming to European standard EN 469 will provide a basic level of protection for chemical incidents.

SECTION 6: Accidental release measures

6.1 Personal precautions, protective equipment and emergency procedures

For non-emergency personnel : No action shall be taken involving any personal risk or without suitable training. Evacuate surrounding areas. Keep unnecessary and unprotected personnel from entering. Do not touch or walk through spilled material. Put on appropriate personal protective equipment.

For emergency responders : If specialised clothing is required to deal with the spillage, take note of any information in Section 8 on suitable and unsuitable materials. See also the information in "For non-emergency personnel".

6.2 Environmental precautions : Avoid dispersal of spilled material and runoff and contact with soil, waterways, drains and sewers. Inform the relevant authorities if the product has caused environmental pollution (sewers, waterways, soil or air).

6.3 Methods and materials for containment and cleaning up

Small spill : Stop leak if without risk. Move containers from spill area. Dilute with water and mop up if water-soluble. Alternatively, or if water-insoluble, absorb with an inert dry material and place in an appropriate waste disposal container. Dispose of via a licensed waste disposal contractor.

Large spill : Stop leak if without risk. Move containers from spill area. Prevent entry into sewers, water courses, basements or confined areas. Wash spillages into an effluent treatment plant or proceed as follows. Contain and collect spillage with non-combustible, absorbent material e.g. sand, earth, vermiculite or diatomaceous earth and place in container for disposal according to local regulations. Dispose of via a licensed waste disposal contractor.

6.4 Reference to other sections : See Section 1 for emergency contact information.
See Section 8 for information on appropriate personal protective equipment.
See Section 13 for additional waste treatment information.

SECTION 7: Handling and storage

The information in this section contains generic advice and guidance. The list of Identified Uses in Section 1 should be consulted for any available use-specific information provided in the Exposure Scenario(s).

7.1 Precautions for safe handling

Protective measures : Put on appropriate personal protective equipment (see Section 8).

Advice on general occupational hygiene : Eating, drinking and smoking should be prohibited in areas where this material is handled, stored and processed. Workers should wash hands and face before eating, drinking and smoking. Remove contaminated clothing and protective equipment before entering eating areas. See also Section 8 for additional information on hygiene measures.

7.2 Conditions for safe storage, including any incompatibilities

Store between the following temperatures: 20 to 25°C (68 to 77°F). Store in accordance with local regulations. Store in original container protected from direct sunlight in a dry, cool and well-ventilated area, away from incompatible materials (see Section 10) and food and drink. Keep container tightly closed and sealed until ready for use. Containers that have been opened must be carefully resealed and kept upright to prevent leakage. Do not store in unlabeled containers. Use appropriate containment to avoid environmental contamination.

7.3 Specific end use(s)

Recommendations : Not available.

Industrial sector specific solutions : Not available.

SECTION 8: Exposure controls/personal protection

The information in this section contains generic advice and guidance. Information is provided based on typical anticipated uses of the product. Additional measures might be required for bulk handling or other uses that could significantly increase worker or exposure or environmental releases.

8.1 Control parameters

Occupational exposure limits

No exposure limit value known.

Recommended monitoring procedures : If this product contains ingredients with exposure limits, personal, workplace atmosphere or biological monitoring may be required to determine the effectiveness of the ventilation or other control measures and/or the necessity to use respiratory protective equipment. Reference should be made to monitoring standards, such as the following: European Standard EN 689 (Workplace atmospheres - Guidance for the assessment of exposure by inhalation to chemical agents for comparison with limit values and measurement strategy) European Standard EN 14042 (Workplace atmospheres - Guide for the application and use of procedures for the assessment of exposure to chemical and biological agents) European Standard EN 482 (Workplace atmospheres - General requirements for the performance of procedures for the measurement of chemical agents) Reference to national guidance documents for methods for the determination of hazardous substances will also be required.

DNELs/DMELs

No DNELs/DMELs available.

PNECs

No PNECs available.

8.2 Exposure controls

Appropriate engineering controls : Good general ventilation should be sufficient to control worker exposure to airborne contaminants.

Individual protection measures

SECTION 8: Exposure controls/personal protection

Hygiene measures : Wash hands, forearms and face thoroughly after handling chemical products, before eating, smoking and using the lavatory and at the end of the working period. Appropriate techniques should be used to remove potentially contaminated clothing. Wash contaminated clothing before reusing. Ensure that eyewash stations and safety showers are close to the workstation location.

Eye/face protection : Safety eyewear complying with an approved standard should be used when a risk assessment indicates this is necessary to avoid exposure to liquid splashes, mists, gases or dusts. If contact is possible, the following protection should be worn, unless the assessment indicates a higher degree of protection: safety glasses with side-shields.

Skin protection

Hand protection : Chemical-resistant, impervious gloves complying with an approved standard should be worn at all times when handling chemical products if a risk assessment indicates this is necessary.

Body protection : Personal protective equipment for the body should be selected based on the task being performed and the risks involved and should be approved by a specialist before handling this product.

Other skin protection : Appropriate footwear and any additional skin protection measures should be selected based on the task being performed and the risks involved and should be approved by a specialist before handling this product.

Respiratory protection : Use a properly fitted, air-purifying or air-fed respirator complying with an approved standard if a risk assessment indicates this is necessary. Respirator selection must be based on known or anticipated exposure levels, the hazards of the product and the safe working limits of the selected respirator.

Environmental exposure controls : Emissions from ventilation or work process equipment should be checked to ensure they comply with the requirements of environmental protection legislation. In some cases, fume scrubbers, filters or engineering modifications to the process equipment will be necessary to reduce emissions to acceptable levels.

SECTION 9: Physical and chemical properties

9.1 Information on basic physical and chemical properties

Appearance

Physical state : Liquid. [Clear sparkling liquid.]

Color : Colorless.

Odor : Not available.

Odor threshold : Not available.

pH : 7.4 to 7.6

Melting point/freezing point : Not available.

Initial boiling point and boiling range : Not available.

Flash point : [Product does not sustain combustion.]

Evaporation rate : Not available.

Flammability (solid, gas) : Not available.

Burning time : Not applicable.

Burning rate : Not applicable.

Upper/lower flammability or explosive limits : Not available.

Vapor pressure : Not available.

Vapor density : Not available.

Relative density : Not available.

Solubility(ies) : Easily soluble in the following materials: cold water and hot water.

Solubility in water : Not available.

Partition coefficient: n-octanol/water : Not available.

Auto-ignition temperature : Not available.

SECTION 9: Physical and chemical properties

Decomposition temperature : Not available.

Viscosity : Not available.

Oxidizing properties : Not available.

9.2 Other information

No additional information.

SECTION 10: Stability and reactivity

10.1 Reactivity : No specific test data related to reactivity available for this product or its ingredients.

10.2 Chemical stability : The product is stable.

10.3 Possibility of hazardous reactions : Under normal conditions of storage and use, hazardous reactions will not occur.

10.4 Conditions to avoid : No specific data.

10.5 Incompatible materials : No specific data.

10.6 Hazardous decomposition products : Under normal conditions of storage and use, hazardous decomposition products should not be produced.

SECTION 11: Toxicological information

11.1 Information on toxicological effects

Acute toxicity

Conclusion/Summary : To the best of our knowledge, the toxicological properties of this product have not been thoroughly investigated.

Acute toxicity estimates

Not available.

Irritation/Corrosion

Conclusion/Summary : Not available.

Sensitization

Conclusion/Summary : Not available.

Mutagenicity

Conclusion/Summary : Not available.

Carcinogenicity

Conclusion/Summary : Not available.

Reproductive toxicity

Conclusion/Summary : Not available.

Teratogenicity

Conclusion/Summary : Not available.

Specific target organ toxicity (single exposure)

Not available.

Specific target organ toxicity (repeated exposure)

Not available.

Aspiration hazard

Not available.

SECTION 11: Toxicological information

Information on the likely routes of exposure : Routes of entry anticipated: Oral, Dermal, Inhalation.

Potential acute health effects

Eye contact : No known significant effects or critical hazards.

Inhalation : No known significant effects or critical hazards.

Skin contact : No known significant effects or critical hazards.

Ingestion : No known significant effects or critical hazards.

Symptoms related to the physical, chemical and toxicological characteristics

Eye contact : No specific data.

Inhalation : No specific data.

Skin contact : No specific data.

Ingestion : No specific data.

Delayed and immediate effects and also chronic effects from short and long term exposure

Short term exposure

Potential immediate effects : Not available.

Potential delayed effects : Not available.

Long term exposure

Potential immediate effects : Not available.

Potential delayed effects : Not available.

Potential chronic health effects

Not available.

Conclusion/Summary : Not available.

General : No known significant effects or critical hazards.

Carcinogenicity : No known significant effects or critical hazards.

Mutagenicity : No known significant effects or critical hazards.

Teratogenicity : No known significant effects or critical hazards.

Developmental effects : No known significant effects or critical hazards.

Fertility effects : No known significant effects or critical hazards.

Other information : Not available.

SECTION 12: Ecological information

12.1 Toxicity

Conclusion/Summary : Not available.

12.2 Persistence and degradability

Conclusion/Summary : Not available.

12.3 Bioaccumulative potential

Not available.

12.4 Mobility in soil

Soil/water partition coefficient (K_{OC}) : Not available.

Mobility : Not available.

SECTION 12: Ecological information

12.5 Results of PBT and vPvB assessment

PBT : Not applicable.

vPvB : Not applicable.

12.6 Other adverse effects : No known significant effects or critical hazards.

SECTION 13: Disposal considerations

The information in this section contains generic advice and guidance. The list of Identified Uses in Section 1 should be consulted for any available use-specific information provided in the Exposure Scenario(s).

13.1 Waste treatment methods

Product

Methods of disposal : The generation of waste should be avoided or minimized wherever possible. Disposal of this product, solutions and any by-products should at all times comply with the requirements of environmental protection and waste disposal legislation and any regional local authority requirements. Dispose of surplus and non-recyclable products via a licensed waste disposal contractor. Waste should not be disposed of untreated to the sewer unless fully compliant with the requirements of all authorities with jurisdiction.

Hazardous waste : Within the present knowledge of the supplier, this product is not regarded as hazardous waste, as defined by EU Directive 91/689/EEC.

Packaging

Methods of disposal : The generation of waste should be avoided or minimized wherever possible. Waste packaging should be recycled. Incineration or landfill should only be considered when recycling is not feasible.

Special precautions : This material and its container must be disposed of in a safe way. Empty containers or liners may retain some product residues. Avoid dispersal of spilled material and runoff and contact with soil, waterways, drains and sewers.

SECTION 14: Transport information

	ADR/RID	ADN	IMDG	IATA
14.1 UN number	Not regulated.	Not available.	Not regulated.	Not regulated.
14.2 UN proper shipping name	-	Not available.	-	-
14.3 Transport hazard class(es)	-	Not available.	-	-
14.4 Packing group	-	-	-	-
14.5 Environmental hazards	No.	No.	No.	No.
Additional information	-	-	-	-

14.6 Special precautions for user : **Transport within user's premises:** always transport in closed containers that are upright and secure. Ensure that persons transporting the product know what to do in the event of an accident or spillage.

SECTION 14: Transport information

14.7 Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code : Not available.

SECTION 15: Regulatory information

15.1 Safety, health and environmental regulations/legislation specific for the substance or mixture

EU Regulation (EC) No. 1907/2006 (REACH)

Annex XIV - List of substances subject to authorization

Annex XIV

None of the components are listed.

Substances of very high concern

None of the components are listed.

Annex XVII - Restrictions on the manufacture, placing on the market and use of certain dangerous substances, mixtures and articles : Not applicable.

Other EU regulations

Europe inventory : Not determined.

Seveso II Directive

This product is not controlled under the Seveso II Directive.

German hazard class for water : 1 Appendix No. 4

15.2 Chemical Safety Assessment : Not applicable.

SECTION 16: Other information

Indicates information that has changed from previously issued version.

Abbreviations and acronyms : ATE = Acute Toxicity Estimate
CLP = Classification, Labelling and Packaging Regulation [Regulation (EC) No. 1272/2008]
DMEL = Derived Minimal Effect Level
DNEL = Derived No Effect Level
EUH statement = CLP-specific Hazard statement
PBT = Persistent, Bioaccumulative and Toxic
PNEC = Predicted No Effect Concentration
RRN = REACH Registration Number
vPvB = Very Persistent and Very Bioaccumulative

Procedure used to derive the classification according to Regulation (EC) No. 1272/2008 [CLP/GHS]

Classification	Justification
Not classified.	

Full text of abbreviated H statements : Not applicable.

Full text of classifications [CLP/GHS] : Not applicable.

Full text of abbreviated R phrases : Not applicable.

Full text of classifications [DSD/DPD] : Not applicable.

Date of printing : 8/13/2015.

SECTION 16: Other information

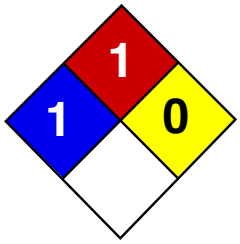
Date of issue/ Date of revision : 8/13/2015.

Date of previous issue : 10/28/2013.

Version : 1.01

Notice to reader

To the best of our knowledge, the information contained herein is accurate. However, neither the above-named supplier, nor any of its subsidiaries, assumes any liability whatsoever for the accuracy or completeness of the information contained herein.
Final determination of suitability of any material is the sole responsibility of the user. All materials may present unknown hazards and should be used with caution. Although certain hazards are described herein, we cannot guarantee that these are the only hazards that exist.



Health	1
Fire	1
Reactivity	0
Personal Protection	E

Material Safety Data Sheet
DNase I MSDS

Section 1: Chemical Product and Company Identification

Product Name: DNase I

Catalog Codes: SLD1084

CAS#: 9003-98-9

RTECS: RF0750000

TSCA: TSCA 8(b) inventory: DNase I

CI#: Not available.

Synonym: Nuclease, deoxyribo-; Deoxyribolnuclease I

Chemical Name: Deoxyribonuclease

Chemical Formula: Not available.

Contact Information:

Sciencelab.com, Inc.
14025 Smith Rd.
Houston, Texas 77396

US Sales: **1-800-901-7247**
International Sales: **1-281-441-4400**

Order Online: ScienceLab.com

CHEMTREC (24HR Emergency Telephone), call: 1-800-424-9300

International CHEMTREC, call: 1-703-527-3887

For non-emergency assistance, call: 1-281-441-4400

Section 2: Composition and Information on Ingredients

Composition:

Name	CAS #	% by Weight
DNase I	9003-98-9	100

Toxicological Data on Ingredients: Not applicable.

Section 3: Hazards Identification

Potential Acute Health Effects: Slightly hazardous in case of skin contact (irritant), of eye contact (irritant), of ingestion, of inhalation.

Potential Chronic Health Effects:
CARCINOGENIC EFFECTS: Not available. MUTAGENIC EFFECTS: Not available. TERATOGENIC EFFECTS: Not available. DEVELOPMENTAL TOXICITY: Not available. Repeated or prolonged exposure is not known to aggravate medical condition.

Section 4: First Aid Measures

Eye Contact:
Check for and remove any contact lenses. In case of contact, immediately flush eyes with plenty of water for at least 15 minutes. Get medical attention if irritation occurs.

Skin Contact: Wash with soap and water. Cover the irritated skin with an emollient. Get medical attention if irritation develops.

Serious Skin Contact: Not available.

Inhalation:
If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical attention.

Serious Inhalation: Not available.

Ingestion:
Do NOT induce vomiting unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. If large quantities of this material are swallowed, call a physician immediately. Loosen tight clothing such as a collar, tie, belt or waistband.

Serious Ingestion: Not available.

Section 5: Fire and Explosion Data

Flammability of the Product: May be combustible at high temperature.

Auto-Ignition Temperature: Not available.

Flash Points: Not available.

Flammable Limits: Not available.

Products of Combustion: Not available.

Fire Hazards in Presence of Various Substances:
Slightly flammable to flammable in presence of heat. Non-flammable in presence of shocks.

Explosion Hazards in Presence of Various Substances:
Slightly explosive in presence of open flames and sparks. Non-explosive in presence of shocks.

Fire Fighting Media and Instructions:
SMALL FIRE: Use DRY chemical powder. LARGE FIRE: Use water spray, fog or foam. Do not use water jet.

Special Remarks on Fire Hazards: As with most organic solids, fire is possible at elevated temperatures

Special Remarks on Explosion Hazards:
Fine dust dispersed in air in sufficient concentrations, and in the presences of an ignition source is a potential dust explosion hazard.

Section 6: Accidental Release Measures

Small Spill:
Use appropriate tools to put the spilled solid in a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and dispose of according to local and regional authority requirements.

Large Spill:
Use a shovel to put the material into a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and allow to evacuate through the sanitary system.

Section 7: Handling and Storage

Precautions:
Keep away from heat. Keep away from sources of ignition. Do not breathe dust. Keep away from incompatibles such as oxidizing agents.

Storage: Keep container tightly closed. Keep container in a cool, well-ventilated area. Do not store above 0°C (32°F).

Section 8: Exposure Controls/Personal Protection

Engineering Controls:
Use process enclosures, local exhaust ventilation, or other engineering controls to keep airborne levels below recommended exposure limits. If user operations generate dust, fume or mist, use ventilation to keep exposure to airborne contaminants below the exposure limit.

Personal Protection: Safety glasses. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Gloves.

Personal Protection in Case of a Large Spill:
Splash goggles. Full suit. Dust respirator. Boots. Gloves. A self contained breathing apparatus should be used to avoid inhalation of the product. Suggested protective clothing might not be sufficient; consult a specialist BEFORE handling this product.

Exposure Limits: Not available.

Section 9: Physical and Chemical Properties

Physical state and appearance: Solid. (Powdered solid.)

Odor: Not available.

Taste: Not available.

Molecular Weight: Not available.

Color: Beige. (Light.)

pH (1% soln/water): Not available.

Boiling Point: Not available.

Melting Point: Not available.

Critical Temperature: Not available.

Specific Gravity: Not available.

Vapor Pressure: Not applicable.

Vapor Density: Not available.

Volatility: Not available.

Odor Threshold: Not available.

Water/Oil Dist. Coeff.: Not available.

Ionicity (in Water): Not available.

Dispersion Properties: Not available.

Solubility: Not available.

Section 10: Stability and Reactivity Data

Stability: The product is stable.

Instability Temperature: Not available.

Conditions of Instability: Excess heat, dust generation, incompatible materials

Incompatibility with various substances: Reactive with oxidizing agents.

Corrosivity: Not available.

Special Remarks on Reactivity: Not available.

Special Remarks on Corrosivity: Not available.

Polymerization: Will not occur.

Section 11: Toxicological Information

Routes of Entry: Inhalation. Ingestion.

Toxicity to Animals:
LD50: Not available. LC50: Not available.

Chronic Effects on Humans: Not available.

Other Toxic Effects on Humans: Slightly hazardous in case of skin contact (irritant), of ingestion, of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: May affect genetic material (mutagenic)

Special Remarks on other Toxic Effects on Humans:
Acute Potential Health Effects: Skin: May cause skin irritation. Eyes: May cause eye irritation. Inhalation: May cause upper respiratory tract and mucous membrane irritation. Ingestion: No information. The toxicological properties of this substance have not been fully investigated.

Section 12: Ecological Information

Ecotoxicity: Not available.

BOD5 and COD: Not available.

Products of Biodegradation:
Possibly hazardous short term degradation products are not likely. However, long term degradation products may arise.

Toxicity of the Products of Biodegradation: Not available.

Special Remarks on the Products of Biodegradation: Not available.

Section 13: Disposal Considerations

Waste Disposal:
Waste must be disposed of in accordance with federal, state and local environmental control regulations.

Section 14: Transport Information

DOT Classification: Not a DOT controlled material (United States).

Identification: Not applicable.

Special Provisions for Transport: Not applicable.

Section 15: Other Regulatory Information

Federal and State Regulations: TSCA 8(b) inventory: DNase I

Other Regulations: EINECS: This product is on the European Inventory of Existing Commercial Chemical Substances.

Other Classifications:

WHMIS (Canada): Not controlled under WHMIS (Canada).

DSCL (EEC):
This product is not classified according to the EU regulations. S22- Do not breathe dust. S24/25- Avoid contact with skin and eyes.

HMIS (U.S.A.):

Health Hazard: 1

Fire Hazard: 1

Reactivity: 0

Personal Protection: E

National Fire Protection Association (U.S.A.):

Health: 1

Flammability: 1

Reactivity: 0

Specific hazard:

Protective Equipment:
Gloves. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Safety glasses.

Section 16: Other Information

References: Not available.

Other Special Considerations: Not available.

Created: 10/09/2005 05:24 PM

Last Updated: 05/21/2013 12:00 PM

The information above is believed to be accurate and represents the best information currently available to us. However, we make no warranty of merchantability or any other warranty, express or implied, with respect to such information, and we assume no liability resulting from its use. Users should make their own investigations to determine the suitability of the information for their particular purposes. In no event shall ScienceLab.com be liable for any claims, losses, or damages of any third party or for lost profits or any special, indirect, incidental, consequential or exemplary damages, howsoever arising, even if ScienceLab.com has been advised of the possibility of such damages.

SAFETY DATA SHEET

Version 4.8
Revision Date 09/01/2014
Print Date 04/01/2016

1. PRODUCT AND COMPANY IDENTIFICATION

1.1 Product identifiers

Product name : Imidazole

Product Number : I2399
Brand : Sigma-Aldrich

CAS-No. : 288-32-4

1.2 Relevant identified uses of the substance or mixture and uses advised against

Identified uses : Laboratory chemicals, Manufacture of substances

1.3 Details of the supplier of the safety data sheet

Company : Sigma-Aldrich
3050 Spruce Street
SAINT LOUIS MO 63103
USA

Telephone : +1 800-325-5832
Fax : +1 800-325-5052

1.4 Emergency telephone number

Emergency Phone # : (314) 776-6555

2. HAZARDS IDENTIFICATION

2.1 Classification of the substance or mixture

GHS Classification in accordance with 29 CFR 1910 (OSHA HCS)
Acute toxicity, Oral (Category 4), H302
Skin corrosion (Category 1B), H314
Serious eye damage (Category 1), H318
Reproductive toxicity (Category 1B), H360

For the full text of the H-Statements mentioned in this Section, see Section 16.

2.2 GHS Label elements, including precautionary statements

Pictogram

Signal word Danger

Hazard statement(s)
H302 Harmful if swallowed.
H314 Causes severe skin burns and eye damage.
H360 May damage fertility or the unborn child.

Precautionary statement(s)
P201 Obtain special instructions before use.
P202 Do not handle until all safety precautions have been read and understood.
P260 Do not breathe dust or mist.
P264 Wash skin thoroughly after handling.
P270 Do not eat, drink or smoke when using this product.
P280 Wear protective gloves/ protective clothing/ eye protection/ face

	protection.
P301 + P312	IF SWALLOWED: Call a POISON CENTER or doctor/ physician if you feel unwell.
P301 + P330 + P331	IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.
P303 + P361 + P353	IF ON SKIN (or hair): Remove/ Take off immediately all contaminated clothing. Rinse skin with water/ shower.
P304 + P340	IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing.
P305 + P351 + P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P310	Immediately call a POISON CENTER or doctor/ physician.
P321	Specific treatment (see supplemental first aid instructions on this label).
P363	Wash contaminated clothing before reuse.
P405	Store locked up.
P501	Dispose of contents/ container to an approved waste disposal plant.

2.3 Hazards not otherwise classified (HNOC) or not covered by GHS - none

3. COMPOSITION/INFORMATION ON INGREDIENTS

3.1 Substances

Synonyms : 1,3-Diaza-2,4-cyclopentadiene
Glyoxaline

Formula : $C_3H_4N_2$
Molecular weight : 68.08 g/mol
CAS-No. : 288-32-4
EC-No. : 206-019-2
Registration number : 01-2119485825-24-XXXX

Hazardous components

Component	Classification	Concentration
Imidazole		
	Acute Tox. 4; Skin Corr. 1B; Eye Dam. 1; Repr. 1B; H302, H314, H360	90 - 100 %

For the full text of the H-Statements mentioned in this Section, see Section 16.

4. FIRST AID MEASURES

4.1 Description of first aid measures

General advice
Consult a physician. Show this safety data sheet to the doctor in attendance.Move out of dangerous area.

If inhaled
If breathed in, move person into fresh air. If not breathing, give artificial respiration. Consult a physician.

In case of skin contact
Take off contaminated clothing and shoes immediately. Wash off with soap and plenty of water. Consult a physician.

In case of eye contact
Rinse thoroughly with plenty of water for at least 15 minutes and consult a physician.Continue rinsing eyes during transport to hospital.

If swallowed
Do NOT induce vomiting. Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.

4.2 Most important symptoms and effects, both acute and delayed
The most important known symptoms and effects are described in the labelling (see section 2.2) and/or in section 11

4.3 Indication of any immediate medical attention and special treatment needed
No data available

5. FIREFIGHTING MEASURES

5.1 Extinguishing media

Suitable extinguishing media
Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide.

5.2 Special hazards arising from the substance or mixture
Carbon oxides, Nitrogen oxides (NOx), Hydrogen cyanide (hydrocyanic acid)

5.3 Advice for firefighters
Wear self-contained breathing apparatus for firefighting if necessary.

5.4 Further information
No data available

6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures
Use personal protective equipment. Avoid dust formation. Avoid breathing vapours, mist or gas. Ensure adequate ventilation. Evacuate personnel to safe areas. Avoid breathing dust.
For personal protection see section 8.

6.2 Environmental precautions
Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

6.3 Methods and materials for containment and cleaning up
Pick up and arrange disposal without creating dust. Sweep up and shovel. Keep in suitable, closed containers for disposal.

6.4 Reference to other sections
For disposal see section 13.

7. HANDLING AND STORAGE

7.1 Precautions for safe handling
Avoid contact with skin and eyes. Avoid formation of dust and aerosols.
Provide appropriate exhaust ventilation at places where dust is formed.
For precautions see section 2.2.

7.2 Conditions for safe storage, including any incompatibilities
Keep container tightly closed in a dry and well-ventilated place.

7.3 Specific end use(s)
Apart from the uses mentioned in section 1.2 no other specific uses are stipulated

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Components with workplace control parameters
Contains no substances with occupational exposure limit values.

Derived No Effect Level (DNEL)

Application Area	Exposure routes	Health effect	Value
Workers	Inhalation	Long-term systemic effects	10.6 mg/m3
Workers	Skin contact	Long-term systemic effects	1.5mg/kg BW/d

Predicted No Effect Concentration (PNEC)

Compartment	Value
Soil	0.0425 mg/kg
Marine water	0.013 mg/l
Fresh water	0.13 mg/l
Marine sediment	0.0336 mg/kg

Fresh water sediment	0.336 mg/kg
Sewage treatment plant	10 mg/l
Aquatic intermittent release	1.3 mg/l

8.2 Exposure controls

Appropriate engineering controls
Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday.

Personal protective equipment

Eye/face protection
Face shield and safety glasses Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166(EU).

Skin protection
Handle with gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.

Full contact
Material: Nitrile rubber
Minimum layer thickness: 0.11 mm
Break through time: 480 min
Material tested:Dermatril® (KCL 740 / Aldrich Z677272, Size M)

Splash contact
Material: Nitrile rubber
Minimum layer thickness: 0.11 mm
Break through time: 480 min
Material tested:Dermatril® (KCL 740 / Aldrich Z677272, Size M)

data source: KCL GmbH, D-36124 Eichenzell, phone +49 (0)6659 87300, e-mail sales@kcl.de, test method: EN374
If used in solution, or mixed with other substances, and under conditions which differ from EN 374, contact the supplier of the CE approved gloves. This recommendation is advisory only and must be evaluated by an industrial hygienist and safety officer familiar with the specific situation of anticipated use by our customers. It should not be construed as offering an approval for any specific use scenario.

Body Protection
Complete suit protecting against chemicals, The type of protective equipment must be selected according to the concentration and amount of the dangerous substance at the specific workplace.

Respiratory protection
Where risk assessment shows air-purifying respirators are appropriate use a full-face particle respirator type N100 (US) or type P3 (EN 143) respirator cartridges as a backup to engineering controls. If the respirator is the sole means of protection, use a full-face supplied air respirator. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

Control of environmental exposure
Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

9. PHYSICAL AND CHEMICAL PROPERTIES

9.1 Information on basic physical and chemical properties

a)	Appearance	Form: crystalline Colour: white
b)	Odour	amine-like
c)	Odour Threshold	No data available
d)	pH	9 - 11 at 100 g/l at 23 °C (73 °F)
e)	Melting point/freezing point	Melting point/range: 88 - 91 °C (190 - 196 °F) - lit.
f)	Initial boiling point and	256 °C (493 °F) - lit.

	boiling range	
g)	Flash point	145 °C (293 °F) - closed cup
h)	Evaporation rate	No data available
i)	Flammability (solid, gas)	No data available
j)	Upper/lower flammability or explosive limits	No data available
k)	Vapour pressure	0.003 hPa (0.002 mmHg) at 20 °C (68 °F)
l)	Vapour density	No data available
m)	Relative density	1.030 g/cm3
n)	Water solubility	633 g/l at 20 °C (68 °F)
o)	Partition coefficient: n-octanol/water	log Pow: -0.02 at 25 °C (77 °F)
p)	Auto-ignition temperature	No data available
q)	Decomposition temperature	No data available
r)	Viscosity	No data available
s)	Explosive properties	No data available
t)	Oxidizing properties	No data available

9.2 Other safety information

Bulk density	550 kg/m3
Dissociation constant	7.15 at 25 °C (77 °F)

10. STABILITY AND REACTIVITY

10.1 Reactivity
No data available

10.2 Chemical stability
Stable under recommended storage conditions.

10.3 Possibility of hazardous reactions
No data available

10.4 Conditions to avoid
No data available

10.5 Incompatible materials
acids, Acid anhydrides, Strong oxidizing agents

10.6 Hazardous decomposition products
Other decomposition products - No data available
In the event of fire: see section 5

11. TOXICOLOGICAL INFORMATION

11.1 Information on toxicological effects

Acute toxicity
LD50 Oral - Rat - 970 mg/kg

Inhalation: No data available

Dermal: No data available

No data available

Skin corrosion/irritation
Skin - Rabbit
Result: Causes burns.

Serious eye damage/eye irritation
No data available

Respiratory or skin sensitisation
No data available

Germ cell mutagenicity
Did not show mutagenic effects in animal experiments. Tests on bacterial or mammalian cell cultures did not show mutagenic effects.

Carcinogenicity

IARC: No component of this product present at levels greater than or equal to 0.1% is identified as probable, possible or confirmed human carcinogen by IARC.

ACGIH: No component of this product present at levels greater than or equal to 0.1% is identified as a carcinogen or potential carcinogen by ACGIH.

NTP: No component of this product present at levels greater than or equal to 0.1% is identified as a known or anticipated carcinogen by NTP.

OSHA: No component of this product present at levels greater than or equal to 0.1% is identified as a carcinogen or potential carcinogen by OSHA.

Reproductive toxicity
May damage the unborn child.
Presumed human reproductive toxicant May damage the unborn child.

No data available

Specific target organ toxicity - single exposure
No data available

Specific target organ toxicity - repeated exposure
No data available

Aspiration hazard
No data available

Additional Information
RTECS: NI3325000

To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.

12. ECOLOGICAL INFORMATION

12.1 Toxicity

Toxicity to fish	static test LC50 - Leuciscus idus (Golden orfe) - 280 mg/l - 48 h
Toxicity to daphnia and other aquatic invertebrates	EC50 - Daphnia (water flea) - 341.5 mg/l - 48 h
Toxicity to algae	static test EC50 - Scenedesmus quadricauda (Green algae) - 133 mg/l - 72 h
Toxicity to bacteria	see user defined free text - other microorganisms - 45 mg/l - 0.5 h

12.2 Persistence and degradability

Biodegradability: aerobic - Exposure time 19 d
Result: 86 % - Readily biodegradable.

12.3 Bioaccumulative potential

No data available

12.4 Mobility in soil
No data available

12.5 Results of PBT and vPvB assessment
PBT/vPvB assessment not available as chemical safety assessment not required/not conducted

12.6 Other adverse effects

No data available

13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

Product
Offer surplus and non-recyclable solutions to a licensed disposal company. Contact a licensed professional waste disposal service to dispose of this material. Dissolve or mix the material with a combustible solvent and burn in a chemical incinerator equipped with an afterburner and scrubber.

Contaminated packaging
Dispose of as unused product.

14. TRANSPORT INFORMATION

DOT (US)
UN number: 3263 Class: 8 Packing group: II
Proper shipping name: Corrosive solid, basic, organic, n.o.s. (Imidazole)
Reportable Quantity (RQ):
Marine pollutant: No
Poison Inhalation Hazard: No

IMDG
UN number: 3263 Class: 8 Packing group: II EMS-No: F-A, S-B
Proper shipping name: CORROSIVE SOLID, BASIC, ORGANIC, N.O.S. (Imidazole)
Marine pollutant: No

IATA
UN number: 3263 Class: 8 Packing group: II
Proper shipping name: Corrosive solid, basic, organic, n.o.s. (Imidazole)

15. REGULATORY INFORMATION

SARA 302 Components
No chemicals in this material are subject to the reporting requirements of SARA Title III, Section 302.

SARA 313 Components
This material does not contain any chemical components with known CAS numbers that exceed the threshold (De Minimis) reporting levels established by SARA Title III, Section 313.

SARA 311/312 Hazards
Acute Health Hazard, Chronic Health Hazard

Massachusetts Right To Know Components
No components are subject to the Massachusetts Right to Know Act.

Pennsylvania Right To Know Components

	CAS-No.	Revision Date
Imidazole	288-32-4	

New Jersey Right To Know Components

	CAS-No.	Revision Date
Imidazole	288-32-4	

California Prop. 65 Components
This product does not contain any chemicals known to State of California to cause cancer, birth defects, or any other reproductive harm.

16. OTHER INFORMATION

Full text of H-Statements referred to under sections 2 and 3.

Acute Tox.	Acute toxicity
Eye Dam.	Serious eye damage
H302	Harmful if swallowed.
H314	Causes severe skin burns and eye damage.
H318	Causes serious eye damage.
H360	May damage fertility or the unborn child.
Repr.	Reproductive toxicity
Skin Corr.	Skin corrosion

HMIS Rating

Health hazard:	3
Chronic Health Hazard:	*
Flammability:	1
Physical Hazard	0

NFPA Rating

Health hazard:	3
Fire Hazard:	1
Reactivity Hazard:	0

Further information

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. The information in this document is based on the present state of our knowledge and is applicable to the product with regard to appropriate safety precautions. It does not represent any guarantee of the properties of the product. Sigma-Aldrich Corporation and its Affiliates shall not be held liable for any damage resulting from handling or from contact with the above product. See www.sigma-aldrich.com and/or the reverse side of invoice or packing slip for additional terms and conditions of sale.

Preparation Information

Sigma-Aldrich Corporation
Product Safety – Americas Region
1-800-521-8956

Version: 4.8 Revision Date: 09/01/2014 Print Date: 04/01/2016

SAFETY DATA SHEET

Version 5.3
Revision Date 02/26/2015
Print Date 04/01/2016

1. PRODUCT AND COMPANY IDENTIFICATION

1.1 Product identifiers

Product name : Isopropyl β-D-1-thiogalactopyranoside

Product Number : I6758
Brand : Sigma-Aldrich

CAS-No. : 367-93-1

1.2 Relevant identified uses of the substance or mixture and uses advised against

Identified uses : Laboratory chemicals, Manufacture of substances

1.3 Details of the supplier of the safety data sheet

Company : Sigma-Aldrich
3050 Spruce Street
SAINT LOUIS MO 63103
USA

Telephone : +1 800-325-5832
Fax : +1 800-325-5052

1.4 Emergency telephone number

Emergency Phone # : (314) 776-6555

2. HAZARDS IDENTIFICATION

2.1 Classification of the substance or mixture

GHS Classification in accordance with 29 CFR 1910 (OSHA HCS)
Carcinogenicity (Category 2), H351

For the full text of the H-Statements mentioned in this Section, see Section 16.

2.2 GHS Label elements, including precautionary statements

Pictogram

Signal word Warning

Hazard statement(s)
H351 Suspected of causing cancer.

Precautionary statement(s)
P201 Obtain special instructions before use.
P202 Do not handle until all safety precautions have been read and understood.
P281 Use personal protective equipment as required.
P308 + P313 IF exposed or concerned: Get medical advice/ attention.
P405 Store locked up.
P501 Dispose of contents/ container to an approved waste disposal plant.

2.3 Hazards not otherwise classified (HNOC) or not covered by GHS - none

3. COMPOSITION/INFORMATION ON INGREDIENTS

3.2 Mixtures

Synonyms	:	Isopropyl β-D-thiogalactoside IPTG
Formula	:	$C_9H_{18}O_5S$
Molecular weight	:	238.3 g/mol

Hazardous components

Component		Classification	Concentration
1,4-Dioxane			
CAS-No. EC-No. Index-No.	123-91-1 204-661-8 603-024-00-5	Flam. Liq. 2; Eye Irrit. 2A; Carc. 2; STOT SE 3; H225, H319, H335, H351	>= 0.1 - < 1 %

For the full text of the H-Statements mentioned in this Section, see Section 16.

4. FIRST AID MEASURES

4.1 Description of first aid measures

General advice
Consult a physician. Show this safety data sheet to the doctor in attendance.Move out of dangerous area.

If inhaled
If breathed in, move person into fresh air. If not breathing, give artificial respiration. Consult a physician.

In case of skin contact
Wash off with soap and plenty of water. Consult a physician.

In case of eye contact
Flush eyes with water as a precaution.

If swallowed
Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.

4.2 Most important symptoms and effects, both acute and delayed
The most important known symptoms and effects are described in the labelling (see section 2.2) and/or in section 11

4.3 Indication of any immediate medical attention and special treatment needed
No data available

5. FIREFIGHTING MEASURES

5.1 Extinguishing media

Suitable extinguishing media
Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide.

5.2 Special hazards arising from the substance or mixture
Carbon oxides, Sulphur oxides

5.3 Advice for firefighters
Wear self-contained breathing apparatus for firefighting if necessary.

5.4 Further information
No data available

6. ACCIDENTAL RELEASE MEASURES

6.1 Personal precautions, protective equipment and emergency procedures
Use personal protective equipment. Avoid dust formation. Avoid breathing vapours, mist or gas. Ensure adequate ventilation. Evacuate personnel to safe areas. Avoid breathing dust.
For personal protection see section 8.

6.2 Environmental precautions
Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

6.3 Methods and materials for containment and cleaning up

Pick up and arrange disposal without creating dust. Sweep up and shovel. Keep in suitable, closed containers for disposal.

6.4 Reference to other sections

For disposal see section 13.

7. HANDLING AND STORAGE

7.1 Precautions for safe handling

Avoid contact with skin and eyes. Avoid formation of dust and aerosols.
Provide appropriate exhaust ventilation at places where dust is formed.
For precautions see section 2.2.

7.2 Conditions for safe storage, including any incompatibilities

Keep container tightly closed in a dry and well-ventilated place.

Recommended storage temperature 2 - 8 °C

hygroscopic

7.3 Specific end use(s)

Apart from the uses mentioned in section 1.2 no other specific uses are stipulated

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

8.1 Control parameters

Components with workplace control parameters

Component	CAS-No.	Value	Control parameters	Basis
1,4-Dioxane	123-91-1	TWA	20.000000 ppm	USA. ACGIH Threshold Limit Values (TLV)
	Remarks	Liver damage Confirmed animal carcinogen with unknown relevance to humans Danger of cutaneous absorption		
		TWA	20 ppm	USA. ACGIH Threshold Limit Values (TLV)
		Liver damage Confirmed animal carcinogen with unknown relevance to humans Danger of cutaneous absorption		
		TWA	25 ppm 90 mg/m3	USA. OSHA - TABLE Z-1 Limits for Air Contaminants - 1910.1000
		Skin notation		
		TWA	100.000000 ppm 360.000000 mg/m3	USA. Occupational Exposure Limits (OSHA) - Table Z-1 Limits for Air Contaminants
		Skin designation The value in mg/m3 is approximate.		
		TWA	100 ppm 360 mg/m3	USA. Occupational Exposure Limits (OSHA) - Table Z-1 Limits for Air Contaminants
		Skin designation The value in mg/m3 is approximate.		
		C	1.000000 ppm 3.600000 mg/m3	USA. NIOSH Recommended Exposure Limits
		Potential Occupational Carcinogen See Appendix A 30 minute ceiling value		

8.2 Exposure controls

Appropriate engineering controls
Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday.

Personal protective equipment

Eye/face protection
Safety glasses with side-shields conforming to EN166 Use equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166(EU).

Skin protection
Handle with gloves. Gloves must be inspected prior to use. Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.

Full contact
Material: Nitrile rubber
Minimum layer thickness: 0.11 mm
Break through time: 480 min
Material tested:Dermatril® (KCL 740 / Aldrich Z677272, Size M)

Splash contact
Material: Nitrile rubber
Minimum layer thickness: 0.11 mm
Break through time: 480 min
Material tested:Dermatril® (KCL 740 / Aldrich Z677272, Size M)

data source: KCL GmbH, D-36124 Eichenzell, phone +49 (0)6659 87300, e-mail sales@kcl.de, test method: EN374
If used in solution, or mixed with other substances, and under conditions which differ from EN 374, contact the supplier of the CE approved gloves. This recommendation is advisory only and must be evaluated by an industrial hygienist and safety officer familiar with the specific situation of anticipated use by our customers. It should not be construed as offering an approval for any specific use scenario.

Body Protection
impervious clothing, The type of protective equipment must be selected according to the concentration and amount of the dangerous substance at the specific workplace.

Respiratory protection
Where risk assessment shows air-purifying respirators are appropriate use a full-face particle respirator type N100 (US) or type P3 (EN 143) respirator cartridges as a backup to engineering controls. If the respirator is the sole means of protection, use a full-face supplied air respirator. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU).

Control of environmental exposure
Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

9. PHYSICAL AND CHEMICAL PROPERTIES

9.1 Information on basic physical and chemical properties

a)	Appearance	Form: solid
b)	Odour	No data available
c)	Odour Threshold	No data available
d)	pH	No data available
e)	Melting point/freezing point	105 °C (221 °F)
f)	Initial boiling point and boiling range	No data available
g)	Flash point	No data available
h)	Evaporation rate	No data available

i)	Flammability (solid, gas)	No data available
j)	Upper/lower flammability or explosive limits	No data available
k)	Vapour pressure	No data available
l)	Vapour density	No data available
m)	Relative density	No data available
n)	Water solubility	No data available
o)	Partition coefficient: n-octanol/water	No data available
p)	Auto-ignition temperature	No data available
q)	Decomposition temperature	No data available
r)	Viscosity	No data available
s)	Explosive properties	No data available
t)	Oxidizing properties	No data available

9.2 Other safety information
No data available

10. STABILITY AND REACTIVITY

10.1 Reactivity
No data available

10.2 Chemical stability
Stable under recommended storage conditions.

10.3 Possibility of hazardous reactions
No data available

10.4 Conditions to avoid
Exposure to moisture

10.5 Incompatible materials
Strong oxidizing agents

10.6 Hazardous decomposition products
Other decomposition products - No data available
In the event of fire: see section 5

11. TOXICOLOGICAL INFORMATION

11.1 Information on toxicological effects

Acute toxicity
No data available

Inhalation: No data available

Dermal: No data available

No data available

Skin corrosion/irritation
No data available

Serious eye damage/eye irritation
No data available

Respiratory or skin sensitisation
No data available

Germ cell mutagenicity
No data available

Carcinogenicity

IARC: 2B - Group 2B: Possibly carcinogenic to humans (1,4-Dioxane)

NTP: Reasonably anticipated to be a human carcinogen (1,4-Dioxane)

OSHA: No component of this product present at levels greater than or equal to 0.1% is identified as a carcinogen or potential carcinogen by OSHA.

Reproductive toxicity
No data available

No data available

Specific target organ toxicity - single exposure
No data available

Specific target organ toxicity - repeated exposure
No data available

Aspiration hazard
No data available

Additional Information
RTECS: Not available

To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.

Liver - Irregularities - Based on Human Evidence
Liver - Irregularities - Based on Human Evidence (1,4-Dioxane)

12. ECOLOGICAL INFORMATION

12.1 Toxicity
No data available

12.2 Persistence and degradability
No data available

12.3 Bioaccumulative potential
No data available

12.4 Mobility in soil
No data available

12.5 Results of PBT and vPvB assessment
PBT/vPvB assessment not available as chemical safety assessment not required/not conducted

12.6 Other adverse effects

No data available

13. DISPOSAL CONSIDERATIONS

13.1 Waste treatment methods

Product
Offer surplus and non-recyclable solutions to a licensed disposal company. Contact a licensed professional waste disposal service to dispose of this material. Dissolve or mix the material with a combustible solvent and burn in a chemical incinerator equipped with an afterburner and scrubber.

Contaminated packaging
Dispose of as unused product.

14. TRANSPORT INFORMATION

DOT (US)
Not dangerous goods

IMDG
Not dangerous goods

IATA
Not dangerous goods

15. REGULATORY INFORMATION

SARA 302 Components
No chemicals in this material are subject to the reporting requirements of SARA Title III, Section 302.

SARA 313 Components
The following components are subject to reporting levels established by SARA Title III, Section 313:

	CAS-No.	Revision Date
1,4-Dioxane	123-91-1	2007-07-01

SARA 311/312 Hazards
Chronic Health Hazard

Massachusetts Right To Know Components

	CAS-No.	Revision Date
1,4-Dioxane	123-91-1	2007-07-01

Pennsylvania Right To Know Components

	CAS-No.	Revision Date
1,4-Dioxane	123-91-1	2007-07-01
Isopropyl-β-D-thiogalactopyranoside	367-93-1	

New Jersey Right To Know Components

	CAS-No.	Revision Date
1,4-Dioxane	123-91-1	2007-07-01
Isopropyl-β-D-thiogalactopyranoside	367-93-1	

California Prop. 65 Components

WARNING! This product contains a chemical known to the	CAS-No.	Revision Date
State of California to cause cancer.	123-91-1	2007-09-28
1,4-Dioxane		

16. OTHER INFORMATION

Full text of H-Statements referred to under sections 2 and 3.

Carc.	Carcinogenicity
Eye Irrit.	Eye irritation
Flam. Liq.	Flammable liquids
H225	Highly flammable liquid and vapour.
H319	Causes serious eye irritation.
H335	May cause respiratory irritation.
H351	Suspected of causing cancer.
STOT SE	Specific target organ toxicity - single exposure

HMIS Rating

Health hazard:	0
Chronic Health Hazard:	*
Flammability:	0
Physical Hazard	0

NFPA Rating

Health hazard:	0
Fire Hazard:	0
Reactivity Hazard:	0

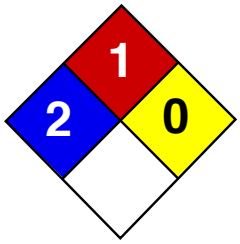
Further information

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. The information in this document is based on the present state of our knowledge and is applicable to the product with regard to appropriate safety precautions. It does not represent any guarantee of the properties of the product. Sigma-Aldrich Corporation and its Affiliates shall not be held liable for any damage resulting from handling or from contact with the above product. See www.sigma-aldrich.com and/or the reverse side of invoice or packing slip for additional terms and conditions of sale.

Preparation Information

Sigma-Aldrich Corporation
Product Safety – Americas Region
1-800-521-8956

Version: 5.3 Revision Date: 02/26/2015 Print Date: 04/01/2016



Material Safety Data Sheet
Kanamycin sulfate MSDS

Section 1: Chemical Product and Company Identification

Product Name: Kanamycin sulfate

Catalog Codes: SLK1033, SLK1111

CAS#: 25389-94-0

RTECS: NZ3225030

TSCA: TSCA 8(b) inventory: Kanamycin sulfate

CI#: Not available.

Synonym:

Chemical Name: Not available.

Chemical Formula: C18H36N4O11.H2SO4

Contact Information:

Sciencelab.com, Inc.
14025 Smith Rd.
Houston, Texas 77396

US Sales: **1-800-901-7247**
International Sales: **1-281-441-4400**

Order Online: ScienceLab.com

CHEMTREC (24HR Emergency Telephone), call:
1-800-424-9300

International CHEMTREC, call: 1-703-527-3887

For non-emergency assistance, call: 1-281-441-4400

Section 2: Composition and Information on Ingredients

Composition:

Name	CAS #	% by Weight
Kanamycin sulfate	25389-94-0	100

Toxicological Data on Ingredients: Kanamycin sulfate LD50: Not available. LC50: Not available.

Section 3: Hazards Identification

Potential Acute Health Effects: Hazardous in case of eye contact (irritant), of ingestion, of inhalation.

Potential Chronic Health Effects:
Hazardous in case of skin contact (irritant), of eye contact (irritant), of ingestion, of inhalation. CARCINOGENIC EFFECTS: Not available. MUTAGENIC EFFECTS: Not available. TERATOGENIC EFFECTS: Not available. DEVELOPMENTAL TOXICITY: Not available. The substance is toxic to the nervous system. Repeated or prolonged exposure to the substance can produce target organs damage.

Section 4: First Aid Measures

Eye Contact: Check for and remove any contact lenses. Do not use an eye ointment. Seek medical attention.

Skin Contact: No known effect on skin contact, rinse with water for a few minutes.

Serious Skin Contact: Not available.

Inhalation: Allow the victim to rest in a well ventilated area. Seek immediate medical attention.

Serious Inhalation: Not available.

Ingestion:
Do not induce vomiting. Loosen tight clothing such as a collar, tie, belt or waistband. If the victim is not breathing, perform mouth-to-mouth resuscitation. Seek immediate medical attention.

Serious Ingestion: Not available.

Section 5: Fire and Explosion Data

Flammability of the Product: May be combustible at high temperature.

Auto-Ignition Temperature: Not available.

Flash Points: Not available.

Flammable Limits: Not available.

Products of Combustion: These products are carbon oxides (CO, CO2), nitrogen oxides (NO, NO2...), sulfur oxides (SO2, SO3...).

Fire Hazards in Presence of Various Substances: Not available.

Explosion Hazards in Presence of Various Substances:
Risks of explosion of the product in presence of mechanical impact: Not available. Risks of explosion of the product in presence of static discharge: Not available.

Fire Fighting Media and Instructions:
SMALL FIRE: Use DRY chemical powder. LARGE FIRE: Use water spray, fog or foam. Do not use water jet.

Special Remarks on Fire Hazards: Not available.

Special Remarks on Explosion Hazards: Not available.

Section 6: Accidental Release Measures

Small Spill:
Use appropriate tools to put the spilled solid in a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and dispose of according to local and regional authority requirements.

Large Spill:
Use a shovel to put the material into a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and allow to evacuate through the sanitary system.

Section 7: Handling and Storage

Precautions:
Keep away from heat. Keep away from sources of ignition. Empty containers pose a fire risk, evaporate the residue under a fume hood. Ground all equipment containing material. Do not breathe dust. Avoid contact with eyes Wear suitable protective clothing In case of insufficient ventilation, wear suitable respiratory equipment If you feel unwell, seek medical attention and show the label when possible.

Storage:
Keep container dry. Keep in a cool place. Ground all equipment containing material. Keep container tightly closed. Keep in a cool, well-ventilated place. Combustible materials should be stored away from extreme heat and away from strong oxidizing agents.

Section 8: Exposure Controls/Personal Protection

Engineering Controls:
Use process enclosures, local exhaust ventilation, or other engineering controls to keep airborne levels below recommended exposure limits. If user operations generate dust, fume or mist, use ventilation to keep exposure to airborne contaminants below the exposure limit.

Personal Protection: Splash goggles. Lab coat.

Personal Protection in Case of a Large Spill:
Splash goggles. Full suit. Boots. Gloves. Suggested protective clothing might not be sufficient; consult a specialist BEFORE handling this product.

Exposure Limits: Not available.

Section 9: Physical and Chemical Properties

Physical state and appearance: Solid. (Crystalline solid.)

Odor: Not available.

Taste: Not available.

Molecular Weight: 582.58 g/mole

Color: White.

pH (1% soln/water): Not available.

Boiling Point: Not available.

Melting Point: Decomposes.

Critical Temperature: Not available.

Specific Gravity: Not available.

Vapor Pressure: Not applicable.

Vapor Density: Not available.

Volatility: Not available.

Odor Threshold: Not available.

Water/Oil Dist. Coeff.: Not available.

Ionicity (in Water): Not available.

Dispersion Properties: Not available.

Solubility: Not available.

Section 10: Stability and Reactivity Data

Stability: The product is stable.

Instability Temperature: Not available.

Conditions of Instability: Not available.

Incompatibility with various substances: Not available.

Corrosivity: Non-corrosive in presence of glass.

Special Remarks on Reactivity: Not available.

Special Remarks on Corrosivity: Not available.

Polymerization: No.

Section 11: Toxicological Information

Routes of Entry: Eye contact. Inhalation. Ingestion.

Toxicity to Animals:
LD50: Not available. LC50: Not available.

Chronic Effects on Humans: The substance is toxic to the nervous system.

Other Toxic Effects on Humans: Hazardous in case of ingestion, of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Special Remarks on other Toxic Effects on Humans: Not available.

Section 12: Ecological Information

Ecotoxicity: Not available.

BOD5 and COD: Not available.

Products of Biodegradation:
Possibly hazardous short term degradation products are not likely. However, long term degradation products may arise.

Toxicity of the Products of Biodegradation: The products of degradation are more toxic.

Special Remarks on the Products of Biodegradation: Not available.

Section 13: Disposal Considerations

Waste Disposal:

Section 14: Transport Information

DOT Classification: Not a DOT controlled material (United States).

Identification: Not applicable.

Special Provisions for Transport: Not applicable.

Section 15: Other Regulatory Information

Federal and State Regulations: TSCA 8(b) inventory: Kanamycin sulfate

Other Regulations: OSHA: Hazardous by definition of Hazard Communication Standard (29 CFR 1910.1200).

Other Classifications:

WHMIS (Canada): Not controlled under WHMIS (Canada).

DSCL (EEC): R36- Irritating to eyes.

HMIS (U.S.A.):

Health Hazard: 2

Fire Hazard: 1

Reactivity: 0

Personal Protection: j

National Fire Protection Association (U.S.A.):

Health: 2

Flammability: 1

Reactivity: 0

Specific hazard:

Protective Equipment:
Not applicable. Lab coat. Not applicable. Splash goggles.

Section 16: Other Information

References: Not available.

Other Special Considerations: Not available.

Created: 10/10/2005 08:20 PM

Last Updated: 05/21/2013 12:00 PM

MATERIAL SAFETY DATA SHEET

Section 1. Company Identification and Product Information			
Product Name or Identity:	**LB Broth, Lennox**		
Manufacturer's Name:	Acumedia Manufacturers, Inc.	**Emergency Phone No.:**	517/372-9200
	740 East Shiawassee	**Fax No.:**	517/372-0108
	Lansing, Michigan 48912	**e-mail:**	foodsafety@neogen.com
Date Prepared or Revised: August 2007			

Section 2. Composition / Information on Ingredients				
Hazardous Components Specific Chemical Identity:	**CAS-No.**	**%**	**EG-Number**	**Hazard Symbol**
Sodium Chloride, NaCl	7647-14-5	25%	231-598-3	Xi (Irritant)

Section 3. Health Hazard Identification			
Route(s) of Entry:	Inhalation? Yes	Skin? Yes	Ingestion? Yes
Health Hazards: *(Acute and Chronic)*	IRRITANT. Irritating to eyes, respiratory system, and skin.		
Carcinogenicity:	IARC Monographs? No	OSHA Regulated? No	
Signs and Symptoms of Exposure: Irritant if inhaled, coughing possible and breathing difficulties may be observed. Symptoms of ingestion can include nausea and vomiting. Can result in mild irritation if contact with skin for several hours. Contact with eye causes irritation, redness, and pain.			
Medical Conditions Generally Aggravated by Exposure: Chronic exposure can cause dermatitis. May be harmful if inhaled, causing respiratory tract irritation. May be harmful if absorbed through the skin.			

Section 4. First Aid Measures	
Emergency / First Aid Procedures:	Ingestion: If swallowed, wash out mouth with water, provided person is conscious. Never give anything by mouth to an unconscious person. Seek medical attention.
	Inhalation: If inhaled, supply fresh air or oxygen. Seek medical attention. If not breathing, apply artificial respiration. If breathing is difficult, give oxygen.
	Eye Contact: Rinse opened eye for at least 15 minutes under running water, lifting lower and upper eyelids occasionally. Seek medical attention.
	Skin Contact: Remove contaminated clothing. Immediately wash with plenty of soap and water for at least 15 minutes. Seek medical attention. Wash clothing before reuse.

Section 5. Fire and Explosion Hazard Data	
Flash Point (Method Used): N/A	**Flammable Limits**: LEL – N/A UEL – N/A
Extinguishing Media: Use alcohol foam, dry chemical, or carbon dioxide. Water may be ineffective.	
Special Fire Fighting Procedures: Firefighters should wear protective equipment and self-contained breathing apparatus. The product itself does not burn.	
Unusual Fire and Explosion Hazards: During heating or in case of fire, poisonous gases are produced. Fine dust dispersed in air in sufficient concentrations, and in the presence of an ignition source, is a potential dust explosion hazard.	

Section 6. Accidental Release Measures

Personal Precautions: Shut off all sources of ignition, ventilate spill area. Wear suitable protective clothing, gloves, and eye protection. Wear self-containing breathing apparatus, rubber boots, and heavy rubber gloves. Place contaminated material in a chemical waste container.

Environmental Precautions: Prevent dispersion of material. Do not allow to enter drains or water courses. Water runoff can cause environmental damage.

Clean-up Methods: Contact safety officer and ventilate area. Absorb spill with inert material, including dry-lime, sand, or soda ash, then place into a chemical waste container using non-sparking tools. Wash spill site.

Section 7. Handling and Storage

Handling: Protect against physical damage. Ensure good ventilation / exhaustion. Avoid contact with eyes, skin, and clothing. Avoid prolonged or repeated exposure. Do not use if skin is cut or scratched.

Storage: Keep container tightly closed. Keep away from incompatible material. Storage area should be cool, dry and well ventilated. Containers of this material may be hazardous when empty since they retain product residues.

Other Precautions: Remove contaminated clothing immediately. Ensure good ventilation. Prevent dust formation.

Section 8. Exposure Controls / Personal Protection

OES: N/A	**ACGIH TLV:** N/A

Engineering Measures: Respiratory protection is not required. Where protection from nuisance levels of dusts are desired, use type N95 (US) or type P1 (EN 143) dust masks. Use respirators and components tested and approved under appropriate government standards such as NIOSH (US) or CEN (EU). Proper ventilation, safety shower, and eye bath required.

Respiratory Protection (Specify Type)**:** With sufficient ventilation, breathing apparatus is not necessary. In the event of possible spill / exposure, use dust mask to EN 149 FFP2S.

Ventilation:	Local Exhaust: 50 – 100 CFM	**Special:** Safety shower and eye wash.
Protective Gloves: Compatible chemical-resistant gloves.		**Eye Protection:** Safety glasses or chemical goggles to EN 166, 167, and 168.

Other Protective Clothing or Equipment: Uniform, lab coat, or disposable lab wear.

Work / Hygienic Practices: Follow the usual precautionary measure for handling chemicals / powder. Keep away from food and beverages. Immediately remove all soiled and contaminated clothing. Avoid contact with eyes, skin, and clothing.

Section 9. Physical and Chemical Properties

Boiling Point: 1461°C (Sodium Chloride)	**Specific Gravity:** 2.16 g/cm^3 (Sodium Chloride)
Vapor Pressure: 1 mm at 865°C (Sodium Chloride)	**Melting Point:** 804 °C (Sodium Chloride)
Vapor Density (AIR = 1): N/A	**Solubility in Water:** Partly Soluble (Sodium Chloride)

Appearance and Odor: Solid, colorless or white, odorless (Sodium Chloride)

Section 10. Stability and Reactivity

Stability:	Unstable		
	Stable	X	Conditions to Avoid: Stable under recommended storage conditions.

Incompatibility *(Materials to Avoid)***:** Incompatible with strong oxidizing agents.

Hazardous Decomposition or Byproducts: Sodium oxide and Hydrogen chloride gas.

Hazardous Polymerization:	May Occur		
	Will Not Occur	X	Conditions to Avoid: Incompatible materials.

Section 11. Toxicological Information

LD_{50}: ORL-RAT, 3000 mg/kg (Sodium Chloride)

Section 12. Ecological Information

Ecotoxicity Tests: LC_{50} / 96h: 1,294.6 mg/L, *Lepomis macrochirus* (Bluegill) (Sodium Chloride)

Section 13. Disposal Considerations

Waste Disposal Method: Dispose in accordance with all applicable federal, state, and local environmental regulations. Keep waste separate. Contact a licensed professional waste disposal service to dispose of this material if questions arise. Do not allow product to reach ground water, water bodies, or sewage system.

Container Information: Do not remove labels from containers until they have been cleaned.

Section 14. Transport Information

Sodium Chloride:
Not Regulated

Section 15. Regulatory Information

EU Regulations

Hazard Symbol(s):

Sodium Chloride: Xi (Irritant)

Risk Phrases:

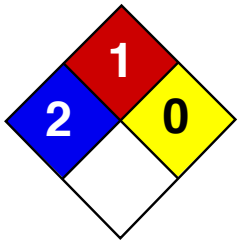
Sodium Chloride: R 36 / 38, Irritating to eyes and skin.

Safety Phrases:

Sodium Chloride: S 24 / 25 / 26, Avoid contact with skin and eyes. In case of contact with eyes, rinse immediately with plenty of water and seek medial advice.

Section 16. Other Information

This document is believed to be correct, but does not purport to be all inclusive and shall be used only as a guide. Acumedia Manufacturers, Inc. shall not be held liable for any damage resulting from handling or from contact with the above product. These suggestions should not be confused with state, municipal or insurance requirements, and constitute NO WARRANTY.



Material Safety Data Sheet
MES MSDS

Section 1: Chemical Product and Company Identification

Product Name: MES

Catalog Codes: SLM3343, SLM1198

CAS#: 4432-31-9

RTECS: KI7970000

TSCA: TSCA 8(b) inventory: MES

CI#: Not available.

Synonym: 2-(N-Morpholino)ethanesulfonic acid

Chemical Name: Not available.

Chemical Formula: C6H13NO4S.H2O

Contact Information:

Sciencelab.com, Inc.
14025 Smith Rd.
Houston, Texas 77396

US Sales: **1-800-901-7247**
International Sales: **1-281-441-4400**

Order Online: ScienceLab.com

CHEMTREC (24HR Emergency Telephone), call: 1-800-424-9300

International CHEMTREC, call: 1-703-527-3887

For non-emergency assistance, call: 1-281-441-4400

Section 2: Composition and Information on Ingredients

Composition:

Name	CAS #	% by Weight
MES	4432-31-9	100

Toxicological Data on Ingredients: MES LD50: Not available. LC50: Not available.

Section 3: Hazards Identification

Potential Acute Health Effects: Very hazardous in case of ingestion. Hazardous in case of skin contact (irritant), of eye contact (irritant).

Potential Chronic Health Effects:
Hazardous in case of skin contact (irritant), of eye contact (irritant), of ingestion, of inhalation. CARCINOGENIC EFFECTS: Not available. MUTAGENIC EFFECTS: Not available. TERATOGENIC EFFECTS: Not available. DEVELOPMENTAL TOXICITY: Not available. The substance is toxic to lungs, mucous membranes. Repeated or prolonged exposure to the substance can produce target organs damage.

Section 4: First Aid Measures

Eye Contact: Check for and remove any contact lenses. Do not use an eye ointment. Seek medical attention.

Skin Contact:

After contact with skin, wash immediately with plenty of water. Gently and thoroughly wash the contaminated skin with running water and non-abrasive soap. Be particularly careful to clean folds, crevices, creases and groin. Cover the irritated skin with an emollient. If irritation persists, seek medical attention. Wash contaminated clothing before reusing.

Serious Skin Contact:
Wash with a disinfectant soap and cover the contaminated skin with an anti-bacterial cream. Seek medical attention.

Inhalation: Allow the victim to rest in a well ventilated area. Seek immediate medical attention.

Serious Inhalation: Not available.

Ingestion:
Do not induce vomiting. Loosen tight clothing such as a collar, tie, belt or waistband. If the victim is not breathing, perform mouth-to-mouth resuscitation. Seek immediate medical attention.

Serious Ingestion: Not available.

Section 5: Fire and Explosion Data

Flammability of the Product: May be combustible at high temperature.

Auto-Ignition Temperature: Not available.

Flash Points: Not available.

Flammable Limits: Not available.

Products of Combustion: These products are carbon oxides (CO, CO2), nitrogen oxides (NO, NO2...), sulfur oxides (SO2, SO3...).

Fire Hazards in Presence of Various Substances: Not available.

Explosion Hazards in Presence of Various Substances:
Risks of explosion of the product in presence of mechanical impact: Not available. Risks of explosion of the product in presence of static discharge: Not available.

Fire Fighting Media and Instructions:
SMALL FIRE: Use DRY chemical powder. LARGE FIRE: Use water spray, fog or foam. Do not use water jet.

Special Remarks on Fire Hazards: Not available.

Special Remarks on Explosion Hazards: Not available.

Section 6: Accidental Release Measures

Small Spill:
Use appropriate tools to put the spilled solid in a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and dispose of according to local and regional authority requirements.

Large Spill:
Use a shovel to put the material into a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and allow to evacuate through the sanitary system.

Section 7: Handling and Storage

Precautions:
Keep away from heat. Keep away from sources of ignition. Empty containers pose a fire risk, evaporate the residue under a fume hood. Ground all equipment containing material. Do not breathe dust. Wear suitable protective clothing If you feel unwell, seek medical attention and show the label when possible. Avoid contact with skin and eyes

Storage:

Keep container dry. Keep in a cool place. Ground all equipment containing material. Keep container tightly closed. Keep in a cool, well-ventilated place. Combustible materials should be stored away from extreme heat and away from strong oxidizing agents.

Section 8: Exposure Controls/Personal Protection

Engineering Controls:
Use process enclosures, local exhaust ventilation, or other engineering controls to keep airborne levels below recommended exposure limits. If user operations generate dust, fume or mist, use ventilation to keep exposure to airborne contaminants below the exposure limit.

Personal Protection:
Splash goggles. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Gloves.

Personal Protection in Case of a Large Spill:
Splash goggles. Full suit. Dust respirator. Boots. Gloves. A self contained breathing apparatus should be used to avoid inhalation of the product. Suggested protective clothing might not be sufficient; consult a specialist BEFORE handling this product.

Exposure Limits: Not available.

Section 9: Physical and Chemical Properties

Physical state and appearance: Solid. (Powdered solid.)

Odor: Not available.

Taste: Not available.

Molecular Weight: 213.26 g/mole

Color: Colorless.

pH (1% soln/water): Not available.

Boiling Point: Not available.

Melting Point: Not available.

Critical Temperature: Not available.

Specific Gravity: Not available.

Vapor Pressure: Not applicable.

Vapor Density: Not available.

Volatility: Not available.

Odor Threshold: Not available.

Water/Oil Dist. Coeff.: Not available.

Ionicity (in Water): Not available.

Dispersion Properties: Not available.

Solubility: Not available.

Section 10: Stability and Reactivity Data

Stability: The product is stable.

Instability Temperature: Not available.

Conditions of Instability: Not available.

Incompatibility with various substances: Not available.

Corrosivity: Non-corrosive in presence of glass.

Special Remarks on Reactivity: Not available.

Special Remarks on Corrosivity: Not available.

Polymerization: No.

Section 11: Toxicological Information

Routes of Entry: Eye contact. Ingestion.

Toxicity to Animals:
LD50: Not available. LC50: Not available.

Chronic Effects on Humans: The substance is toxic to lungs, mucous membranes.

Other Toxic Effects on Humans:
Very hazardous in case of ingestion. Hazardous in case of skin contact (irritant).

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Special Remarks on other Toxic Effects on Humans: Not available.

Section 12: Ecological Information

Ecotoxicity: Not available.

BOD5 and COD: Not available.

Products of Biodegradation:
Possibly hazardous short term degradation products are not likely. However, long term degradation products may arise.

Toxicity of the Products of Biodegradation: The products of degradation are more toxic.

Special Remarks on the Products of Biodegradation: Not available.

Section 13: Disposal Considerations

Waste Disposal:

Section 14: Transport Information

DOT Classification: Not a DOT controlled material (United States).

Identification: Not applicable.

Special Provisions for Transport: Not applicable.

Section 15: Other Regulatory Information

Federal and State Regulations: TSCA 8(b) inventory: MES

Other Regulations: OSHA: Hazardous by definition of Hazard Communication Standard (29 CFR 1910.1200).

Other Classifications:

WHMIS (Canada): CLASS D-2A: Material causing other toxic effects (VERY TOXIC).

DSCL (EEC): R36/38- Irritating to eyes and skin.

HMIS (U.S.A.):

Health Hazard: 2

Fire Hazard: 1

Reactivity: 0

Personal Protection: E

National Fire Protection Association (U.S.A.):

Health: 2

Flammability: 1

Reactivity: 0

Specific hazard:

Protective Equipment:
Gloves. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Wear appropriate respirator when ventilation is inadequate. Splash goggles.

Section 16: Other Information

References: Not available.

Other Special Considerations: Not available.

Created: 10/09/2005 06:06 PM

Last Updated: 05/21/2013 12:00 PM

SAFETY DATA SHEET

HisPur Cobalt Superflow Agarose

Section 1. Identification

GHS product identifier : HisPur Cobalt Superflow Agarose

Other means of identification : Not available.

Product type : Liquid.

Product code : 0025228 0025228S 0025229 0025230 0025231 1862760

SDS # : 8960

Chemical formula : Not applicable.

CAS # : Not applicable.

Relevant identified uses of the substance or mixture and uses advised against

Not applicable.

Supplier's details : Thermo Fisher Scientific
Pierce Biotechnology
P.O. Box 117
Rockford, IL 61105
United States
815.968.0747 or
800.874.3723
7 AM - 5 PM Central Time (GMT -06:00)

Emergency telephone number (with hours of operation) : CHEMTREC: 800.424.9300
Outside US: 703.527.3887

Section 2. Hazards identification

OSHA/HCS status : This material is considered hazardous by the OSHA Hazard Communication Standard (29 CFR 1910.1200).

Classification of the substance or mixture : FLAMMABLE LIQUIDS - Category 3
SKIN CORROSION/IRRITATION - Category 2
SERIOUS EYE DAMAGE/ EYE IRRITATION - Category 2A
RESPIRATORY SENSITIZATION - Category 1
SKIN SENSITIZATION - Category 1
CARCINOGENICITY - Category 1B
TOXIC TO REPRODUCTION (Fertility) - Category 1A
TOXIC TO REPRODUCTION (Unborn child) - Category 1B

GHS label elements

Hazard pictograms :

Signal word : Danger

Hazard statements : Flammable liquid and vapor.
Causes serious eye irritation.
Causes skin irritation.
May cause allergy or asthma symptoms or breathing difficulties if inhaled.
May cause an allergic skin reaction.
May cause cancer.
May damage fertility or the unborn child.

Precautionary statements

Section 2. Hazards identification

Prevention : Obtain special instructions before use. Do not handle until all safety precautions have been read and understood. Use personal protective equipment as required. Wear protective gloves. Wear eye or face protection. In case of inadequate ventilation wear respiratory protection. Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking. Use explosion-proof electrical, ventilating, lighting and all material-handling equipment. Use only non-sparking tools. Take precautionary measures against static discharge. Keep container tightly closed. Avoid breathing vapor. Wash hands thoroughly after handling. Contaminated work clothing should not be allowed out of the workplace.

Response : IF exposed or concerned: Get medical attention. IF INHALED: If breathing is difficult, remove victim to fresh air and keep at rest in a position comfortable for breathing. If experiencing respiratory symptoms: Call a POISON CENTER or physician. IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water or shower. IF ON SKIN: Wash with plenty of soap and water. Take off contaminated clothing. If skin irritation or rash occurs: Get medical attention. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. If eye irritation persists: Get medical attention.

Storage : Store locked up. Store in a well-ventilated place. Keep cool.

Disposal : Dispose of contents and container in accordance with all local, regional, national and international regulations.

Hazards not otherwise classified : None known.

Section 3. Composition/information on ingredients

Substance/mixture : Mixture

Other means of identification : Not available.

CAS number/other identifiers

CAS number : Not applicable.

Ingredient name	%	CAS number
ethanol	10 - 20	64-17-5
Cobalt chloride (CoCl2), hexahydrate	0.1 - 1	7791-13-1

Any concentration shown as a range is to protect confidentiality or is due to batch variation.

There are no additional ingredients present which, within the current knowledge of the supplier and in the concentrations applicable, are classified as hazardous to health or the environment and hence require reporting in this section.

Occupational exposure limits, if available, are listed in Section 8.

Section 4. First aid measures

Description of necessary first aid measures

Eye contact : Immediately flush eyes with plenty of water, occasionally lifting the upper and lower eyelids. Check for and remove any contact lenses. Continue to rinse for at least 10 minutes. Get medical attention.

Inhalation : Remove victim to fresh air and keep at rest in a position comfortable for breathing. If it is suspected that fumes are still present, the rescuer should wear an appropriate mask or self-contained breathing apparatus. If not breathing, if breathing is irregular or if respiratory arrest occurs, provide artificial respiration or oxygen by trained personnel. It may be dangerous to the person providing aid to give mouth-to-mouth resuscitation. Get medical attention. If necessary, call a poison center or physician. If unconscious, place in recovery position and get medical attention immediately. Maintain an open airway. Loosen tight clothing such as a collar, tie, belt or waistband. In the event of any complaints or symptoms, avoid further exposure.

Section 4. First aid measures

Skin contact : Wash with plenty of soap and water. Remove contaminated clothing and shoes. Wash contaminated clothing thoroughly with water before removing it, or wear gloves. Continue to rinse for at least 10 minutes. Get medical attention. In the event of any complaints or symptoms, avoid further exposure. Wash clothing before reuse. Clean shoes thoroughly before reuse.

Ingestion : Wash out mouth with water. Remove dentures if any. Remove victim to fresh air and keep at rest in a position comfortable for breathing. If material has been swallowed and the exposed person is conscious, give small quantities of water to drink. Stop if the exposed person feels sick as vomiting may be dangerous. Do not induce vomiting unless directed to do so by medical personnel. If vomiting occurs, the head should be kept low so that vomit does not enter the lungs. Get medical attention. Never give anything by mouth to an unconscious person. If unconscious, place in recovery position and get medical attention immediately. Maintain an open airway. Loosen tight clothing such as a collar, tie, belt or waistband.

Most important symptoms/effects, acute and delayed

Potential acute health effects

Eye contact : Causes serious eye irritation.

Inhalation : May cause allergy or asthma symptoms or breathing difficulties if inhaled.

Skin contact : Causes skin irritation. May cause an allergic skin reaction.

Ingestion : Irritating to mouth, throat and stomach.

Over-exposure signs/symptoms

Eye contact : Adverse symptoms may include the following:
pain or irritation
watering
redness

Inhalation : Adverse symptoms may include the following:
wheezing and breathing difficulties
asthma
reduced fetal weight
increase in fetal deaths
skeletal malformations

Skin contact : Adverse symptoms may include the following:
irritation
redness
reduced fetal weight
increase in fetal deaths
skeletal malformations

Ingestion : Adverse symptoms may include the following:
reduced fetal weight
increase in fetal deaths
skeletal malformations

Indication of immediate medical attention and special treatment needed, if necessary

Notes to physician : Treat symptomatically. Contact poison treatment specialist immediately if large quantities have been ingested or inhaled.

Specific treatments : No specific treatment.

Protection of first-aiders : No action shall be taken involving any personal risk or without suitable training. If it is suspected that fumes are still present, the rescuer should wear an appropriate mask or self-contained breathing apparatus. It may be dangerous to the person providing aid to give mouth-to-mouth resuscitation. Wash contaminated clothing thoroughly with water before removing it, or wear gloves.

See toxicological information (Section 11)

Section 5. Fire-fighting measures

Extinguishing media

Suitable extinguishing media : Use dry chemical, CO_2, water spray (fog) or foam.

Unsuitable extinguishing media : Do not use water jet.

Specific hazards arising from the chemical : Flammable liquid and vapor. In a fire or if heated, a pressure increase will occur and the container may burst, with the risk of a subsequent explosion. Runoff to sewer may create fire or explosion hazard.

Hazardous thermal decomposition products : Decomposition products may include the following materials:
carbon dioxide
carbon monoxide

Special protective actions for fire-fighters : Promptly isolate the scene by removing all persons from the vicinity of the incident if there is a fire. No action shall be taken involving any personal risk or without suitable training. Move containers from fire area if this can be done without risk. Use water spray to keep fire-exposed containers cool.

Special protective equipment for fire-fighters : Fire-fighters should wear appropriate protective equipment and self-contained breathing apparatus (SCBA) with a full face-piece operated in positive pressure mode.

Section 6. Accidental release measures

Personal precautions, protective equipment and emergency procedures

For non-emergency personnel : No action shall be taken involving any personal risk or without suitable training. Evacuate surrounding areas. Keep unnecessary and unprotected personnel from entering. Do not touch or walk through spilled material. Shut off all ignition sources. No flares, smoking or flames in hazard area. Avoid breathing vapor or mist. Provide adequate ventilation. Wear appropriate respirator when ventilation is inadequate. Put on appropriate personal protective equipment.

For emergency responders : If specialised clothing is required to deal with the spillage, take note of any information in Section 8 on suitable and unsuitable materials. See also the information in "For non-emergency personnel".

Environmental precautions : Avoid dispersal of spilled material and runoff and contact with soil, waterways, drains and sewers. Inform the relevant authorities if the product has caused environmental pollution (sewers, waterways, soil or air).

Methods and materials for containment and cleaning up

Small spill : Stop leak if without risk. Move containers from spill area. Use spark-proof tools and explosion-proof equipment. Dilute with water and mop up if water-soluble. Alternatively, or if water-insoluble, absorb with an inert dry material and place in an appropriate waste disposal container. Dispose of via a licensed waste disposal contractor.

Large spill : Stop leak if without risk. Move containers from spill area. Use spark-proof tools and explosion-proof equipment. Approach release from upwind. Prevent entry into sewers, water courses, basements or confined areas. Wash spillages into an effluent treatment plant or proceed as follows. Contain and collect spillage with non-combustible, absorbent material e.g. sand, earth, vermiculite or diatomaceous earth and place in container for disposal according to local regulations (see Section 13). Dispose of via a licensed waste disposal contractor. Contaminated absorbent material may pose the same hazard as the spilled product. Note: see Section 1 for emergency contact information and Section 13 for waste disposal.

Section 7. Handling and storage

Precautions for safe handling

Protective measures : Put on appropriate personal protective equipment (see Section 8). Persons with a history of skin sensitization problems or asthma, allergies or chronic or recurrent respiratory disease should not be employed in any process in which this product is used. Avoid exposure - obtain special instructions before use. Avoid exposure during pregnancy. Do not handle until all safety precautions have been read and understood. Do not get in eyes or on skin or clothing. Do not ingest. Avoid breathing vapor or mist. Use only with adequate ventilation. Wear appropriate respirator when ventilation is inadequate. Do not enter storage areas and confined spaces unless adequately ventilated. Keep in the original container or an approved alternative made from a compatible material, kept tightly closed when not in use. Store and use away from heat, sparks, open flame or any other ignition source. Use explosion-proof electrical (ventilating, lighting and material handling) equipment. Use only non-sparking tools. Take precautionary measures against electrostatic discharges. Empty containers retain product residue and can be hazardous. Do not reuse container.

Advice on general occupational hygiene : Eating, drinking and smoking should be prohibited in areas where this material is handled, stored and processed. Workers should wash hands and face before eating, drinking and smoking. Remove contaminated clothing and protective equipment before entering eating areas. See also Section 8 for additional information on hygiene measures.

Conditions for safe storage, including any incompatibilities : Store in accordance with local regulations. Store in a segregated and approved area. Store in original container protected from direct sunlight in a dry, cool and well-ventilated area, away from incompatible materials (see Section 10) and food and drink. Store locked up. Eliminate all ignition sources. Separate from oxidizing materials. Keep container tightly closed and sealed until ready for use. Containers that have been opened must be carefully resealed and kept upright to prevent leakage. Do not store in unlabeled containers. Use appropriate containment to avoid environmental contamination.

Section 8. Exposure controls/personal protection

Control parameters

Occupational exposure limits

Ingredient name	Exposure limits
ethanol	**ACGIH TLV (United States, 2000).** TWA: 1880 mg/m³ 8 hours. **OSHA (United States, 0/1989).** CEIL: 7600 ppm TWA: 1000 ppm TWA: 1900 mg/m³ **MSHA (United States).** TWA: 1900 mg/m³ **NIOSH (United States, 0/1994).** TWA: 1000 ppm TWA: 1900 mg/m³ **ACGIH (United States, 0/1996).** TWA: 1880 mg/m³ **ACGIH (United States).** TWA: 1000 ppm **ACGIH TLV (United States, 6/2013).** STEL: 1000 ppm 15 minutes. **NIOSH REL (United States, 4/2013).** TWA: 1900 mg/m³ 10 hours. TWA: 1000 ppm 10 hours. **OSHA PEL (United States, 2/2013).** TWA: 1900 mg/m³ 8 hours. TWA: 1000 ppm 8 hours. **OSHA PEL 1989 (United States, 3/1989).** TWA: 1900 mg/m³ 8 hours. TWA: 1000 ppm 8 hours.
Cobalt chloride (CoCl2), hexahydrate	**ACGIH TLV (United States, 6/2013). Notes:**

Section 8. Exposure controls/personal protection

	as Co TWA: 0.02 mg/m³, (as Co) 8 hours. Form: Inorganic **ACGIH TLV (United States).** : 0.02 mg/m³ **OSHA PEL (United States).** : 0.1 mg/m³

Appropriate engineering controls : Use only with adequate ventilation. Use process enclosures, local exhaust ventilation or other engineering controls to keep worker exposure to airborne contaminants below any recommended or statutory limits. The engineering controls also need to keep gas, vapor or dust concentrations below any lower explosive limits. Use explosion-proof ventilation equipment.

Environmental exposure controls : Emissions from ventilation or work process equipment should be checked to ensure they comply with the requirements of environmental protection legislation. In some cases, fume scrubbers, filters or engineering modifications to the process equipment will be necessary to reduce emissions to acceptable levels.

Individual protection measures

Hygiene measures : Wash hands, forearms and face thoroughly after handling chemical products, before eating, smoking and using the lavatory and at the end of the working period. Appropriate techniques should be used to remove potentially contaminated clothing. Contaminated work clothing should not be allowed out of the workplace. Wash contaminated clothing before reusing. Ensure that eyewash stations and safety showers are close to the workstation location.

Eye/face protection : Safety eyewear complying with an approved standard should be used when a risk assessment indicates this is necessary to avoid exposure to liquid splashes, mists, gases or dusts. If contact is possible, the following protection should be worn, unless the assessment indicates a higher degree of protection: chemical splash goggles.

Skin protection

Hand protection : Chemical-resistant, impervious gloves complying with an approved standard should be worn at all times when handling chemical products if a risk assessment indicates this is necessary. Considering the parameters specified by the glove manufacturer, check during use that the gloves are still retaining their protective properties. It should be noted that the time to breakthrough for any glove material may be different for different glove manufacturers. In the case of mixtures, consisting of several substances, the protection time of the gloves cannot be accurately estimated.

Body protection : Personal protective equipment for the body should be selected based on the task being performed and the risks involved and should be approved by a specialist before handling this product. When there is a risk of ignition from static electricity, wear anti-static protective clothing. For the greatest protection from static discharges, clothing should include anti-static overalls, boots and gloves.

Other skin protection : Appropriate footwear and any additional skin protection measures should be selected based on the task being performed and the risks involved and should be approved by a specialist before handling this product.

Respiratory protection : Use a properly fitted, air-purifying or air-fed respirator complying with an approved standard if a risk assessment indicates this is necessary. Respirator selection must be based on known or anticipated exposure levels, the hazards of the product and the safe working limits of the selected respirator.

Section 9. Physical and chemical properties

Appearance

Physical state : Liquid. [Resin slurry.]

Color : Not available.

Odor : Alcohol-like.

Odor threshold : Not available.

pH : Not available.

Melting point : Not available.

Section 9. Physical and chemical properties

Boiling point : Not available.

Flash point : Closed cup: 23.333°C (74°F)

Burning time : Not applicable.

Burning rate : Not applicable.

Evaporation rate : Not available.

Flammability (solid, gas) : Highly flammable in the presence of the following materials or conditions: open flames, sparks and static discharge and heat.

Lower and upper explosive (flammable) limits : Not available.

Vapor pressure : Not available.

Vapor density : Not available.

Relative density : Not available.

Solubility : Not available.

Solubility in water : Not available.

Partition coefficient: n-octanol/water : Not available.

Auto-ignition temperature : Not available.

Decomposition temperature : Not available.

SADT : Not available.

Viscosity : Not available.

Section 10. Stability and reactivity

Reactivity : No specific test data related to reactivity available for this product or its ingredients.

Chemical stability : The product is stable.

Possibility of hazardous reactions : Under normal conditions of storage and use, hazardous reactions will not occur.

Conditions to avoid : Avoid all possible sources of ignition (spark or flame). Do not pressurize, cut, weld, braze, solder, drill, grind or expose containers to heat or sources of ignition.

Incompatible materials : Reactive or incompatible with the following materials:
oxidizing materials

Hazardous decomposition products : Under normal conditions of storage and use, hazardous decomposition products should not be produced.

Section 11. Toxicological information

Information on toxicological effects

Acute toxicity

Product/ingredient name	Result	Species	Dose	Exposure
ethanol	LC50 Inhalation Vapor	Rat	124700 mg/m³	4 hours
	LD50 Oral	Rat	7 g/kg	-
Cobalt chloride (CoCl2), hexahydrate	LD50 Oral	Rat	766 mg/kg	-

Conclusion/Summary : To the best of our knowledge, the toxicological properties of this product have not been thoroughly investigated.

Irritation/Corrosion

Section 11. Toxicological information

Product/ingredient name	Result	Species	Score	Exposure	Observation
ethanol	Eyes - Mild irritant	Rabbit	-	24 hours 500 milligrams	-
	Eyes - Moderate irritant	Rabbit	-	0.066666667 minutes 100 milligrams	-
	Eyes - Moderate irritant	Rabbit	-	100 microliters	-
	Eyes - Severe irritant	Rabbit	-	500 milligrams	-
	Skin - Mild irritant	Rabbit	-	400 milligrams	-
	Skin - Moderate irritant	Rabbit	-	24 hours 20 milligrams	-

Sensitization

Not available.

Mutagenicity

Product/ingredient name	Test	Experiment	Result
ethanol	DNA Damage	Subject: Bacteria	Positive
	DNA Damage	Subject: Bacteria	Positive
	Mutation in Microorganisms	Subject: Bacteria	Positive
	Mutation in Microorganisms	Subject: Bacteria	Positive
	Gene Conversion and Mitotic Recombination	Subject: Bacteria	Positive
	Sex chromosome loss and nondisjunction.	Subject: Insect	Positive
	Cytogenetic Analysis	Subject: Mammalian-Animal	Positive
	Cytogenetic Analysis	Subject: Mammalian-Animal	Positive
	Cytogenetic Analysis	Subject: Mammalian-Animal	Positive
	Cytogenetic Analysis	Subject: Mammalian-Animal	Positive
	DNA Adduct	Subject: Mammalian-Animal	Positive
	DNA Adduct	Subject: Mammalian-Animal	Positive
	DNA Damage	Subject: Mammalian-Animal	Positive
	Micronucleus Test	Subject: Mammalian-Animal	Positive
	-	Subject: Mammalian-Animal	Positive
	Other Mutation Test Systems	Subject: Mammalian-Animal	Positive
	Other Mutation Test Systems	Subject: Mammalian-Animal	Positive
	Sister Chromatid Exchange	Subject: Mammalian-Animal	Positive
	Specific Locus Test	Subject: Mammalian-Animal	Positive
	Sperm Morphology	Subject: Mammalian-Animal	Positive
	Cytogenetic Analysis	Subject: Mammalian-Human	Positive
	Cytogenetic Analysis	Subject: Mammalian-Human Cell: Germ	Positive
	Micronucleus Test	Subject: Mammalian-Human Cell: Somatic	Positive
	Micronucleus Test	Subject: Mammalian-Human	Positive
	DNA Inhibition	Subject: Mammalian-Human	Positive
	Cytogenetic Analysis	Subject: Mammalian-Human	Positive
	Cytogenetic Analysis	Subject: Mammalian-Human	Positive
	Cytogenetic Analysis	Subject: Mammalian-Human	Positive
	Sister Chromatid Exchange	Subject: Mammalian-Human	Positive

Carcinogenicity

Section 11. Toxicological information

Product/ingredient name	Result	Species	Dose	Exposure
ethanol	Equivocal - Oral - TD	Mouse	400 g/kg	57 weeks Intermittent
	Equivocal - Unreported - TDLo	Mouse	120 g/kg	18 weeks Intermittent
	Equivocal - Oral - TDLo	Mouse	320 mg/kg	50 weeks Intermittent

Classification

Product/ingredient name	OSHA	IARC	NTP
ethanol	+	1	-
Cobalt chloride (CoCl2), hexahydrate	+	2B	-

Reproductive toxicity

Product/ingredient name	Maternal toxicity	Fertility	Development toxin	Species	Dose	Exposure
ethanol	-	-	-	Mouse	Intraperitoneal: 2.9 g/kg	8 days
	-	-	-	Mouse	Intraperitoneal: 2900 mg/kg	8 days
	-	-	-	Dog - Male	Unreported: 100 mg/kg	1 days
	-	Positive	-	Rat	Intraperitoneal: 600 mg/kg	15 days
	Positive	-	-	Mammal - species unspecified	Oral: 206 g/kg	-
	-	Positive	-	Rat - Male	Unreported: 400 mg/kg	1 days
	-	-	Positive	Mouse	Intraperitoneal: 15 g/kg	8 days
	-	Positive	-	Rat - Female	Unreported: 2400 mg/kg	10 days
	-	Positive	-	Woman - Female	Unreported: 200 mg/kg	5 days
	-	Positive	-	Dog	Oral: 221 g/kg	-
	-	Positive	-	Mammal - species unspecified	Oral: 78 g/kg	-
	-	-	Positive	Mouse	Intraperitoneal: 22.8 g/kg	8 days
	-	-	Positive	Mouse	Intraperitoneal: 5.8 g/kg	5 days
	-	-	Positive	Rat	Intraperitoneal: 600 mg/kg	15 days
	-	Positive	Positive	Mouse	Intraperitoneal: 2900 mg/kg	8 days
	-	-	Positive	Mammal - species unspecified	Intravenous	-
	-	-	-	Mouse - Male	Oral: 1680 g/kg	70 days

Teratogenicity

Product/ingredient name	Result	Species	Dose	Exposure
ethanol	Positive - Oral	Woman - Female	41 g/kg	-

Specific target organ toxicity (single exposure)

Not available.

Specific target organ toxicity (repeated exposure)

Section 11. Toxicological information

Not available.

Aspiration hazard

Not available.

Information on the likely routes of exposure : Routes of entry anticipated: Oral, Dermal, Inhalation.

Potential acute health effects

Eye contact : Causes serious eye irritation.

Inhalation : May cause allergy or asthma symptoms or breathing difficulties if inhaled.

Skin contact : Causes skin irritation. May cause an allergic skin reaction.

Ingestion : Irritating to mouth, throat and stomach.

Symptoms related to the physical, chemical and toxicological characteristics

Eye contact : Adverse symptoms may include the following:
pain or irritation
watering
redness

Inhalation : Adverse symptoms may include the following:
wheezing and breathing difficulties
asthma
reduced fetal weight
increase in fetal deaths
skeletal malformations

Skin contact : Adverse symptoms may include the following:
irritation
redness
reduced fetal weight
increase in fetal deaths
skeletal malformations

Ingestion : Adverse symptoms may include the following:
reduced fetal weight
increase in fetal deaths
skeletal malformations

Delayed and immediate effects and also chronic effects from short and long term exposure

Short term exposure

Potential immediate effects : Not available.

Potential delayed effects : Not available.

Long term exposure

Potential immediate effects : Not available.

Potential delayed effects : Not available.

Potential chronic health effects

Not available.

General : Once sensitized, a severe allergic reaction may occur when subsequently exposed to very low levels.

Carcinogenicity : May cause cancer. Risk of cancer depends on duration and level of exposure.

Mutagenicity : No known significant effects or critical hazards.

Teratogenicity : May damage the unborn child.

Developmental effects : No known significant effects or critical hazards.

Fertility effects : May damage fertility.

Numerical measures of toxicity

Section 11. Toxicological information

Acute toxicity estimates

Not available.

Section 12. Ecological information

Toxicity

Product/ingredient name	Result	Species	Exposure
ethanol	Acute EC50 17.921 mg/l Marine water Acute EC50 2000 µg/l Fresh water Acute LC50 25500 µg/l Marine water Acute LC50 42000 µg/l Fresh water Chronic NOEC 4.995 mg/l Marine water Chronic NOEC 0.375 ul/L Fresh water	Algae - Ulva pertusa Daphnia - Daphnia magna Crustaceans - Artemia franciscana - Larvae Fish - Oncorhynchus mykiss Algae - Ulva pertusa Fish - Gambusia holbrooki - Larvae	96 hours 48 hours 48 hours 4 days 96 hours 12 weeks

Persistence and degradability

Product/ingredient name	Aquatic half-life	Photolysis	Biodegradability
ethanol	-	-	Readily

Bioaccumulative potential

Product/ingredient name	$LogP_{ow}$	BCF	Potential
ethanol	-0.35	0.66	low

Mobility in soil

Soil/water partition coefficient (K_{OC}) : Not available.

Other adverse effects : No known significant effects or critical hazards.

Section 13. Disposal considerations

Disposal methods : The generation of waste should be avoided or minimized wherever possible. Disposal of this product, solutions and any by-products should at all times comply with the requirements of environmental protection and waste disposal legislation and any regional local authority requirements. Dispose of surplus and non-recyclable products via a licensed waste disposal contractor. Waste should not be disposed of untreated to the sewer unless fully compliant with the requirements of all authorities with jurisdiction. Waste packaging should be recycled. Incineration or landfill should only be considered when recycling is not feasible. This material and its container must be disposed of in a safe way. Care should be taken when handling emptied containers that have not been cleaned or rinsed out. Empty containers or liners may retain some product residues. Vapor from product residues may create a highly flammable or explosive atmosphere inside the container. Do not cut, weld or grind used containers unless they have been cleaned thoroughly internally. Avoid dispersal of spilled material and runoff and contact with soil, waterways, drains and sewers.

Section 14. Transport information

Section 14. Transport information

	DOT Classification	IATA
UN number	UN1993	UN1993
UN proper shipping name	Flammable liquids, n.o.s. (ethanol)	Flammable liquids, n.o.s. (ethanol)
Transport hazard class(es)	3 	3 
Packing group	III	III
Environmental hazards	No.	No.
Additional information	-	-

Special precautions for user : **Transport within user's premises:** always transport in closed containers that are upright and secure. Ensure that persons transporting the product know what to do in the event of an accident or spillage.

Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code : Not available.

Section 15. Regulatory information

U.S. Federal regulations : **TSCA 8(a) CDR Exempt/Partial exemption**: Not determined

United States inventory (TSCA 8b): All components are listed or exempted.

Clean Air Act Section 112 (b) Hazardous Air Pollutants (HAPs) : Not listed

Clean Air Act Section 602 Class I Substances : Not listed

Clean Air Act Section 602 Class II Substances : Not listed

DEA List I Chemicals (Precursor Chemicals) : Not listed

DEA List II Chemicals (Essential Chemicals) : Not listed

SARA 302/304

Composition/information on ingredients

No products were found.

SARA 304 RQ : Not applicable.

SARA 311/312

Classification : Fire hazard
Immediate (acute) health hazard
Delayed (chronic) health hazard

Composition/information on ingredients

Section 15. Regulatory information

Name	%	Fire hazard	Sudden release of pressure	Reactive	Immediate (acute) health hazard	Delayed (chronic) health hazard
ethanol	10 - 20	Yes.	No.	No.	Yes.	Yes.
Cobalt chloride (CoCl2), hexahydrate	0.1 - 1	No.	No.	No.	Yes.	Yes.

SARA 313

	Product name	CAS number	%
Form R - Reporting requirements	Cobalt chloride (CoCl2), hexahydrate	7791-13-1	0.1 - 1
Supplier notification	Cobalt chloride (CoCl2), hexahydrate	7791-13-1	0.1 - 1

SARA 313 notifications must not be detached from the SDS and any copying and redistribution of the SDS shall include copying and redistribution of the notice attached to copies of the SDS subsequently redistributed.

State regulations

Massachusetts : The following components are listed: ETHYL ALCOHOL

New York : None of the components are listed.

New Jersey : The following components are listed: Agarose; ETHYL ALCOHOL; ALCOHOL; COBALT compounds

Pennsylvania : The following components are listed: Agarose; DENATURED ALCOHOL; COBALT COMPOUNDS

California Prop. 65

WARNING: This product contains a chemical known to the State of California to cause birth defects or other reproductive harm.

Ingredient name	Cancer	Reproductive	No significant risk level	Maximum acceptable dosage level
ethanol	No.	Yes.	No.	No.
methanol	No.	Yes.	No.	23000 µg/day (ingestion) 47000 µg/day (inhalation)

Canada inventory : All components are listed or exempted.

International regulations

International lists : **Australia inventory (AICS)**: All components are listed or exempted.
China inventory (IECSC): All components are listed or exempted.
Japan inventory: Not determined.
Korea inventory: All components are listed or exempted.
Malaysia Inventory (EHS Register): Not determined.
New Zealand Inventory of Chemicals (NZIoC): All components are listed or exempted.
Philippines inventory (PICCS): All components are listed or exempted.
Taiwan inventory (CSNN): Not determined.

Chemical Weapons Convention List Schedule I Chemicals : Not listed

Chemical Weapons Convention List Schedule II Chemicals : Not listed

Chemical Weapons Convention List Schedule III Chemicals : Not listed

Section 16. Other information

Hazardous Material Information System (U.S.A.)

Health	2
Chronic Health Hazard	
Flammability	3
Physical hazards	0

National Fire Protection Association (U.S.A.)

Health	2
Flammability	3
Instability/Reactivity	0
Special	

The customer is responsible for determining the PPE code for this material.

Caution: HMIS® ratings are based on a 0-4 rating scale, with 0 representing minimal hazards or risks, and 4 representing significant hazards or risks Although HMIS® ratings are not required on SDSs under 29 CFR 1910. 1200, the preparer may choose to provide them. HMIS® ratings are to be used with a fully implemented HMIS® program. HMIS® is a registered mark of the National Paint & Coatings Association (NPCA). HMIS® materials may be purchased exclusively from J. J. Keller (800) 327-6868.

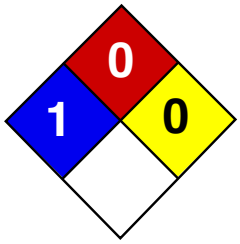
History

Date of printing	:	2/28/2014.
Date of issue/Date of revision	:	2/28/2014.
Date of previous issue	:	No previous validation.
Version	:	1
Prepared by	:	MSDS (Regulatory Specialist)
Key to abbreviations	:	ATE = Acute Toxicity Estimate BCF = Bioconcentration Factor GHS = Globally Harmonized System of Classification and Labelling of Chemicals IATA = International Air Transport Association IBC = Intermediate Bulk Container IMDG = International Maritime Dangerous Goods LogPow = logarithm of the octanol/water partition coefficient MARPOL 73/78 = International Convention for the Prevention of Pollution From Ships, 1973 as modified by the Protocol of 1978. ("Marpol" = marine pollution) UN = United Nations
References	:	Not available.

Indicates information that has changed from previously issued version.

Notice to reader

To the best of our knowledge, the information contained herein is accurate. However, neither the above-named supplier, nor any of its subsidiaries, assumes any liability whatsoever for the accuracy or completeness of the information contained herein.
Final determination of suitability of any material is the sole responsibility of the user. All materials may present unknown hazards and should be used with caution. Although certain hazards are described herein, we cannot guarantee that these are the only hazards that exist.



Health	1
Fire	0
Reactivity	0
Personal Protection	E

Material Safety Data Sheet
Sodium chloride MSDS

Section 1: Chemical Product and Company Identification

Product Name: Sodium chloride

Catalog Codes: SLS3262, SLS1045, SLS3889, SLS1669, SLS3091

CAS#: 7647-14-5

RTECS: VZ4725000

TSCA: TSCA 8(b) inventory: Sodium chloride

CI#: Not applicable.

Synonym: Salt; Sea Salt

Chemical Name: Sodium chloride

Chemical Formula: NaCl

Contact Information:

Sciencelab.com, Inc.
14025 Smith Rd.
Houston, Texas 77396

US Sales: **1-800-901-7247**
International Sales: **1-281-441-4400**

Order Online: ScienceLab.com

CHEMTREC (24HR Emergency Telephone), call: 1-800-424-9300

International CHEMTREC, call: 1-703-527-3887

For non-emergency assistance, call: 1-281-441-4400

Section 2: Composition and Information on Ingredients

Composition:

Name	CAS #	% by Weight
Sodium chloride	7647-14-5	100

Toxicological Data on Ingredients: Sodium chloride: ORAL (LD50): Acute: 3000 mg/kg [Rat.]. 4000 mg/kg [Mouse]. DERMAL (LD50): Acute: >10000 mg/kg [Rabbit]. DUST (LC50): Acute: >42000 mg/m 1 hours [Rat].

Section 3: Hazards Identification

Potential Acute Health Effects: Slightly hazardous in case of skin contact (irritant), of eye contact (irritant), of ingestion, of inhalation.

Potential Chronic Health Effects:
CARCINOGENIC EFFECTS: Not available. MUTAGENIC EFFECTS: Mutagenic for mammalian somatic cells. Mutagenic for bacteria and/or yeast. TERATOGENIC EFFECTS: Not available. DEVELOPMENTAL TOXICITY: Not available. Repeated or prolonged exposure is not known to aggravate medical condition.

Section 4: First Aid Measures

Eye Contact:

Check for and remove any contact lenses. In case of contact, immediately flush eyes with plenty of water for at least 15 minutes. Cold water may be used. Get medical attention.

Skin Contact:
Wash with soap and water. Cover the irritated skin with an emollient. Get medical attention if irritation develops. Cold water may be used.

Serious Skin Contact: Not available.

Inhalation:
If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical attention if symptoms appear.

Serious Inhalation: Not available.

Ingestion:
Do NOT induce vomiting unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. Loosen tight clothing such as a collar, tie, belt or waistband. Get medical attention if symptoms appear.

Serious Ingestion: Not available.

Section 5: Fire and Explosion Data

Flammability of the Product: Non-flammable.

Auto-Ignition Temperature: Not applicable.

Flash Points: Not applicable.

Flammable Limits: Not applicable.

Products of Combustion: Not available.

Fire Hazards in Presence of Various Substances: Not applicable.

Explosion Hazards in Presence of Various Substances:
Risks of explosion of the product in presence of mechanical impact: Not available. Risks of explosion of the product in presence of static discharge: Not available.

Fire Fighting Media and Instructions: Not applicable.

Special Remarks on Fire Hazards: When heated to decomposition it emits toxic fumes.

Special Remarks on Explosion Hazards:
Electrolysis of sodium chloride in presence of nitrogenous compounds to produce chlorine may lead to formation of explosive nitrogen trichloride. Potentially explosive reaction with dichloromaleic anhydride + urea.

Section 6: Accidental Release Measures

Small Spill:
Use appropriate tools to put the spilled solid in a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and dispose of according to local and regional authority requirements.

Large Spill:
Use a shovel to put the material into a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and allow to evacuate through the sanitary system.

Section 7: Handling and Storage

Precautions:
Keep locked up.. Do not ingest. Do not breathe dust. Avoid contact with eyes. Wear suitable protective clothing. If ingested, seek medical advice immediately and show the container or the label. Keep away from incompatibles such as oxidizing agents, acids.

Storage: Keep container tightly closed. Keep container in a cool, well-ventilated area. Hygroscopic

Section 8: Exposure Controls/Personal Protection

Engineering Controls:
Use process enclosures, local exhaust ventilation, or other engineering controls to keep airborne levels below recommended exposure limits. If user operations generate dust, fume or mist, use ventilation to keep exposure to airborne contaminants below the exposure limit.

Personal Protection:
Splash goggles. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Gloves.

Personal Protection in Case of a Large Spill:
Splash goggles. Full suit. Dust respirator. Boots. Gloves. A self contained breathing apparatus should be used to avoid inhalation of the product. Suggested protective clothing might not be sufficient; consult a specialist BEFORE handling this product.

Exposure Limits: Not available.

Section 9: Physical and Chemical Properties

Physical state and appearance: Solid. (Solid crystalline powder.)

Odor: Slight.

Taste: Saline.

Molecular Weight: 58.44 g/mole

Color: White.

pH (1% soln/water): 7 [Neutral.]

Boiling Point: 1413°C (2575.4°F)

Melting Point: 801°C (1473.8°F)

Critical Temperature: Not available.

Specific Gravity: 2.165 (Water = 1)

Vapor Pressure: Not applicable.

Vapor Density: Not available.

Volatility: Not available.

Odor Threshold: Not available.

Water/Oil Dist. Coeff.: Not available.

Ionicity (in Water): Not available.

Dispersion Properties: See solubility in water.

Solubility:
Easily soluble in cold water, hot water. Soluble in glycerol, and ammonia. Very slightly soluble in alcohol. Insoluble in Hydrochloric Acid.

Section 10: Stability and Reactivity Data

Stability: The product is stable.

Instability Temperature: Not available.

Conditions of Instability: Incompatible materials, high temperatures.

Incompatibility with various substances: Reactive with oxidizing agents, metals, acids.

Corrosivity: Not considered to be corrosive for metals and glass.

Special Remarks on Reactivity:
Hygroscopic. Reacts with most nonnoble metals such as iron or steel, building materials (such as cement) Sodium chloride is rapidly attacked by bromine trifluoride. Violent reaction with lithium.

Special Remarks on Corrosivity: Not available.

Polymerization: Will not occur.

Section 11: Toxicological Information

Routes of Entry: Inhalation. Ingestion.

Toxicity to Animals:
WARNING: THE LC50 VALUES HEREUNDER ARE ESTIMATED ON THE BASIS OF A 4-HOUR EXPOSURE. Acute oral toxicity (LD50): 3000 mg/kg [Rat.]. Acute dermal toxicity (LD50): >10000 mg/kg [Rabbit]. Acute toxicity of the dust (LC50): >42000 mg/m3 1 hours [Rat].

Chronic Effects on Humans: MUTAGENIC EFFECTS: Mutagenic for mammalian somatic cells. Mutagenic for bacteria and/or yeast.

Other Toxic Effects on Humans: Slightly hazardous in case of skin contact (irritant), of ingestion, of inhalation.

Special Remarks on Toxicity to Animals: Lowest Published Lethal Dose (LDL) [Man] - Route: Oral; Dose: 1000 mg/kg

Special Remarks on Chronic Effects on Humans:
Causes adverse reproductive effects in humans (fetotoxicity, abortion,) by intraplacental route. High intake of sodium chloride, whether from occupational exposure or in the diet, may increase risk of TOXEMIA OF PREGNANCY in susceptible women (Bishop, 1978). Hypertonic sodium chloride solutions have been used to induce abortion in late pregnancy by direct infusion into the uterus (Brown et al, 1972), but this route of administration is not relevant to occupational exposures. May cause adverse reproductive effects and birth defects in animals, particularly rats and mice (fetotoxicity, abortion, musculoskeletal abnormalities, and maternal effects (effects on ovaries, fallopian tubes) by oral, intraperitoneal, intraplacental, intrauterine, parenteral, and subcutaneous routes. While sodium chloride has been used as a negative control n some reproductive studies, it has also been used as an example that almost any chemical can cause birth defects in experimental animals if studied under the right conditions (Nishimura & Miyamoto, 1969). In experimental animals, sodium chloride has caused delayed effects on newborns, has been fetotoxic, and has caused birth defects and abortions in rats and mice (RTECS, 1997). May affect genetic material (mutagenic)

Special Remarks on other Toxic Effects on Humans:
Acute Potential Health Effects: Skin: May cause skin irritation. Eyes: Causes eye irritation. Ingestion: Ingestion of large quantities can irritate the stomach (as in overuse of salt tablets) with nausea and vomiting. May affect behavior (muscle spasicity/contraction, somnolence), sense organs, metabolism, and cardiovascular system. Continued exposure may produce dehydration, internal organ congestion, and coma. Inhalation: Material is irritating to mucous membranes and upper respiratory tract.

Section 12: Ecological Information

Ecotoxicity: Not available.

BOD5 and COD: Not available.

Products of Biodegradation:
Possibly hazardous short term degradation products are not likely. However, long term degradation products may arise.

Toxicity of the Products of Biodegradation: The product itself and its products of degradation are not toxic.

Special Remarks on the Products of Biodegradation: Not available.

Section 13: Disposal Considerations

Waste Disposal:
Waste must be disposed of in accordance with federal, state and local environmental control regulations.

Section 14: Transport Information

DOT Classification: Not a DOT controlled material (United States).

Identification: Not applicable.

Special Provisions for Transport: Not applicable.

Section 15: Other Regulatory Information

Federal and State Regulations: TSCA 8(b) inventory: Sodium chloride

Other Regulations: EINECS: This product is on the European Inventory of Existing Commercial Chemical Substances.

Other Classifications:

WHMIS (Canada): Not controlled under WHMIS (Canada).

DSCL (EEC):
R40- Possible risks of irreversible effects. S24/25- Avoid contact with skin and eyes.

HMIS (U.S.A.):

Health Hazard: 1

Fire Hazard: 0

Reactivity: 0

Personal Protection: E

National Fire Protection Association (U.S.A.):

Health: 1

Flammability: 0

Reactivity: 0

Specific hazard:

Protective Equipment:
Gloves. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Splash goggles.

Section 16: Other Information

References:
-Hawley, G.G.. The Condensed Chemical Dictionary, 11e ed., New York N.Y., Van Nostrand Reinold, 1987. -SAX, N.I. Dangerous Properties of Indutrial Materials. Toronto, Van Nostrand Reinold, 6e ed. 1984. -The Sigma-Aldrich Library of Chemical Safety Data, Edition II.

Other Special Considerations: Not available.

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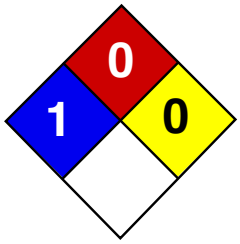

Material Safety Data Sheet
Sodium phosphate, dibasic MSDS

Section 1: Chemical Product and Company Identification

Product Name: Sodium phosphate, dibasic

Catalog Codes: SLS2365, SLS2986, SLS4408

CAS#: 7558-79-4

RTECS: WC4500000

TSCA: TSCA 8(b) inventory: Sodium phosphate, dibasic

CI#: Not available.

Synonym: Dibasic Sodium Phosphate; Disodium hydrogen phosphate; Disodium monohydrogen phosphate; Disodium orthophosphate; Disodium phosphoric acid; Phosphoric acid, disodium salt; Soda phosphate; Sodium hydrogen phosphate

Chemical Name: Sodium Monohydrogen Phosphate(2:1:1)

Chemical Formula: Na2HPO4

Contact Information:

Sciencelab.com, Inc.
14025 Smith Rd.
Houston, Texas 77396

US Sales: **1-800-901-7247**
International Sales: **1-281-441-4400**

Order Online: ScienceLab.com

CHEMTREC (24HR Emergency Telephone), call: 1-800-424-9300

International CHEMTREC, call: 1-703-527-3887

For non-emergency assistance, call: 1-281-441-4400

Section 2: Composition and Information on Ingredients

Composition:

Name	CAS #	% by Weight
Sodium phosphate, dibasic	7558-79-4	100

Toxicological Data on Ingredients: Not applicable.

Section 3: Hazards Identification

Potential Acute Health Effects: Slightly hazardous in case of skin contact (irritant), of eye contact (irritant), of ingestion, of inhalation.

Potential Chronic Health Effects:
CARCINOGENIC EFFECTS: Not available. MUTAGENIC EFFECTS: Not available. TERATOGENIC EFFECTS: Not available. DEVELOPMENTAL TOXICITY: Not available. Repeated or prolonged exposure is not known to aggravate medical condition.

Section 4: First Aid Measures

Eye Contact:

Check for and remove any contact lenses. In case of contact, immediately flush eyes with plenty of water for at least 15 minutes. Cold water may be used. Get medical attention if irritation occurs.

Skin Contact:
Wash with soap and water. Cover the irritated skin with an emollient. Get medical attention if irritation develops. Cold water may be used.

Serious Skin Contact: Not available.

Inhalation:
If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical attention.

Serious Inhalation: Not available.

Ingestion:
Do NOT induce vomiting unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. Loosen tight clothing such as a collar, tie, belt or waistband. Get medical attention if symptoms appear.

Serious Ingestion: Not available.

Section 5: Fire and Explosion Data

Flammability of the Product: Non-flammable.

Auto-Ignition Temperature: Not applicable.

Flash Points: Not applicable.

Flammable Limits: Not applicable.

Products of Combustion: Not available.

Fire Hazards in Presence of Various Substances: Not applicable.

Explosion Hazards in Presence of Various Substances:
Risks of explosion of the product in presence of mechanical impact: Not available. Risks of explosion of the product in presence of static discharge: Not available.

Fire Fighting Media and Instructions: Not applicable.

Special Remarks on Fire Hazards: Not available.

Special Remarks on Explosion Hazards: Not available.

Section 6: Accidental Release Measures

Small Spill:
Use appropriate tools to put the spilled solid in a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and dispose of according to local and regional authority requirements.

Large Spill: Use a shovel to put the material into a convenient waste disposal container.

Section 7: Handling and Storage

Precautions:
Do not ingest. Do not breathe dust. Wear suitable protective clothing. In case of insufficient ventilation, wear suitable respiratory equipment. If ingested, seek medical advice immediately and show the container or the label. Keep away from incompatibles such as acids, alkalis.

Storage: Keep container tightly closed. Keep container in a cool, well-ventilated area. Hygroscopic

Section 8: Exposure Controls/Personal Protection

Engineering Controls:
Use process enclosures, local exhaust ventilation, or other engineering controls to keep airborne levels below recommended exposure limits. If user operations generate dust, fume or mist, use ventilation to keep exposure to airborne contaminants below the exposure limit.

Personal Protection: Safety glasses. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Gloves.

Personal Protection in Case of a Large Spill:
Splash goggles. Full suit. Dust respirator. Boots. Gloves. A self contained breathing apparatus should be used to avoid inhalation of the product. Suggested protective clothing might not be sufficient; consult a specialist BEFORE handling this product.

Exposure Limits: Not available.

Section 9: Physical and Chemical Properties

Physical state and appearance: Solid. (Solid powder.)

Odor: Odorless.

Taste: Saline.

Molecular Weight: 141.96 g/mole

Color: White.

pH (1% soln/water): 9.1 [Basic.]

Boiling Point: Not available.

Melting Point:
Decomposition temperature: 240°C (464°F) Converted to Sodium Pyrophosphate @ about 240 deg. C

Critical Temperature: Not available.

Specific Gravity: Not available.

Vapor Pressure: Not applicable.

Vapor Density: 4.9 (Air = 1)

Volatility: Not available.

Odor Threshold: Not available.

Water/Oil Dist. Coeff.: Not available.

Ionicity (in Water): Not available.

Dispersion Properties: See solubility in water.

Solubility:
Easily soluble in hot water. Soluble in cold water. Insoluble in methanol, n-octanol.

Section 10: Stability and Reactivity Data

Stability: The product is stable.

Instability Temperature: Not available.

Conditions of Instability:

Exposure to moisture and to incompatible materials. When heated to decompostition, it emits toxic fumes of phosphoxides and sodium oxide.

Incompatibility with various substances: Reactive with acids, alkalis.

Corrosivity: Not available.

Special Remarks on Reactivity:
Hygroscopic; keep container tightly closed. Incompatible with magnesium, alkaloids, antipyrine, chloral hydrate, lead acetate, pyrogallol, resorcinol, strong mineral acids, strong organic acids.

Special Remarks on Corrosivity: Not available.

Polymerization: Will not occur.

Section 11: Toxicological Information

Routes of Entry: Inhalation. Ingestion.

Toxicity to Animals: Acute oral toxicity (LD50): 17000 mg/kg [Rat].

Chronic Effects on Humans: Not available.

Other Toxic Effects on Humans: Slightly hazardous in case of skin contact (irritant), of ingestion, of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans: Not available.

Special Remarks on other Toxic Effects on Humans:
Acute Potential Health Effects: Skin: Causes mild skin irritation. May cause dermatitis. Eyes: Causes mild eye irritation. Ingestion: May cause irritation of the digestive tract and may cause purging. It is slowly aborbed. Expected to be a low ingestion hazard for usual industrial handling. Ingestion of large doses may affect behavior/central nervous system (tetany). However, if a significant amount of phosphate is absorbed, hypophosphatemia will occur. Severe hypophosphatemia may result in hypocalcemia and tetany. Cardiovasular, respiratory, neurologic, and musculoskeletal effects may occur secondary to hypernatremia, hypophosphatemia, and hypocalcemia Inhalation: May cause respiratory tract and mucous membrane irritation. Low hazard for usual industrial handling. Chronic Potential Health Effects: Skin: High and repeated exposure may cause dermatitis.

Section 12: Ecological Information

Ecotoxicity: Not available.

BOD5 and COD: Not available.

Products of Biodegradation:
Possibly hazardous short term degradation products are not likely. However, long term degradation products may arise.

Toxicity of the Products of Biodegradation: The product itself and its products of degradation are not toxic.

Special Remarks on the Products of Biodegradation: Not available.

Section 13: Disposal Considerations

Waste Disposal:
Waste must be disposed of in accordance with federal, state and local environmental control regulations.

Section 14: Transport Information

DOT Classification: Not a DOT controlled material (United States).

Identification: : Not available. UNNA: 9147 PG: III

Special Provisions for Transport: Not applicable.

Section 15: Other Regulatory Information

Federal and State Regulations:
New York release reporting list: Sodium phosphate, dibasic Pennsylvania RTK: Sodium phosphate, dibasic Massachusetts RTK: Sodium phosphate, dibasic New Jersey: Sodium phosphate, dibasic California Director's List of Hazardous Sustances: Sodium phosphate, dibasic TSCA 8(b) inventory: Sodium phosphate, dibasic CERCLA: Hazardous substances.: Sodium phosphate, dibasic: 5000 lbs. (2268 kg)

Other Regulations: EINECS: This product is on the European Inventory of Existing Commercial Chemical Substances.

Other Classifications:

WHMIS (Canada): Not controlled under WHMIS (Canada).

DSCL (EEC):
This product is not classified according to the EU regulations. S24/25- Avoid contact with skin and eyes.

HMIS (U.S.A.):

Health Hazard: 1

Fire Hazard: 0

Reactivity: 0

Personal Protection: E

National Fire Protection Association (U.S.A.):

Health: 1

Flammability: 0

Reactivity: 0

Specific hazard:

Protective Equipment:
Gloves. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Safety glasses.

Section 16: Other Information

References: -Hawley, G.G.. The Condensed Chemical Dictionary, 11e ed., New York N.Y., Van Nostrand Reinold, 1987.

Other Special Considerations: Not available.

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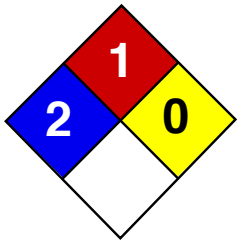

Material Safety Data Sheet
Urea MSDS

Section 1: Chemical Product and Company Identification

Product Name: Urea

Catalog Codes: SLU1063, SLU1132, SLU1093, SLU1162

CAS#: 57-13-6

RTECS: YR6250000

TSCA: TSCA 8(b) inventory: Urea

CI#: Not available.

Synonym: Carbamide

Chemical Name: carbonyldiamide

Chemical Formula: (NH2)2CO or CH4N2O

Contact Information:

Sciencelab.com, Inc.
14025 Smith Rd.
Houston, Texas 77396

US Sales: **1-800-901-7247**
International Sales: **1-281-441-4400**

Order Online: ScienceLab.com

CHEMTREC (24HR Emergency Telephone), call:
1-800-424-9300

International CHEMTREC, call: 1-703-527-3887

For non-emergency assistance, call: 1-281-441-4400

Section 2: Composition and Information on Ingredients

Composition:

Name	CAS #	% by Weight
Urea	57-13-6	100

Toxicological Data on Ingredients: Urea: ORAL (LD50): Acute: 8471 mg/kg [Rat]. 11000 mg/kg [Mouse].

Section 3: Hazards Identification

Potential Acute Health Effects: Hazardous in case of skin contact (irritant), of eye contact (irritant), of ingestion, of inhalation.

Potential Chronic Health Effects:
CARCINOGENIC EFFECTS: Not available. MUTAGENIC EFFECTS: Mutagenic for mammalian somatic cells. TERATOGENIC EFFECTS: Not available. DEVELOPMENTAL TOXICITY: Not available. The substance may be toxic to blood, cardiovascular system. Repeated or prolonged exposure to the substance can produce target organs damage.

Section 4: First Aid Measures

Eye Contact:
Check for and remove any contact lenses. In case of contact, immediately flush eyes with plenty of water for at least 15 minutes. Cold water may be used. Get medical attention.

Skin Contact:

In case of contact, immediately flush skin with plenty of water. Cover the irritated skin with an emollient. Remove contaminated clothing and shoes. Cold water may be used.Wash clothing before reuse. Thoroughly clean shoes before reuse. Get medical attention.

Serious Skin Contact:
Wash with a disinfectant soap and cover the contaminated skin with an anti-bacterial cream. Seek medical attention.

Inhalation:
If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical attention.

Serious Inhalation: Not available.

Ingestion:
Do NOT induce vomiting unless directed to do so by medical personnel. Never give anything by mouth to an unconscious person. Loosen tight clothing such as a collar, tie, belt or waistband. Get medical attention if symptoms appear.

Serious Ingestion: Not available.

Section 5: Fire and Explosion Data

Flammability of the Product: May be combustible at high temperature.

Auto-Ignition Temperature: Not available.

Flash Points: Not available.

Flammable Limits: Not available.

Products of Combustion: These products are carbon oxides (CO, CO2), nitrogen oxides (NO, NO2...).

Fire Hazards in Presence of Various Substances: Slightly flammable to flammable in presence of heat.

Explosion Hazards in Presence of Various Substances:
Risks of explosion of the product in presence of mechanical impact: Not available. Risks of explosion of the product in presence of static discharge: Not available.

Fire Fighting Media and Instructions:
SMALL FIRE: Use DRY chemical powder. LARGE FIRE: Use water spray, fog or foam. Do not use water jet.

Special Remarks on Fire Hazards: Not available.

Special Remarks on Explosion Hazards: Not available.

Section 6: Accidental Release Measures

Small Spill:
Use appropriate tools to put the spilled solid in a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and dispose of according to local and regional authority requirements.

Large Spill:
Use a shovel to put the material into a convenient waste disposal container. Finish cleaning by spreading water on the contaminated surface and allow to evacuate through the sanitary system.

Section 7: Handling and Storage

Precautions:
Keep locked up.. Keep away from heat. Keep away from sources of ignition. Empty containers pose a fire risk, evaporate the residue under a fume hood. Ground all equipment containing material. Do not ingest. Do not breathe dust. Wear suitable protective clothing. In case of insufficient ventilation, wear suitable respiratory equipment. If ingested, seek medical advice

immediately and show the container or the label. Avoid contact with skin and eyes. Keep away from incompatibles such as oxidizing agents.

Storage: Keep container tightly closed. Keep container in a cool, well-ventilated area. Do not store above 23°C (73.4°F).

Section 8: Exposure Controls/Personal Protection

Engineering Controls:
Use process enclosures, local exhaust ventilation, or other engineering controls to keep airborne levels below recommended exposure limits. If user operations generate dust, fume or mist, use ventilation to keep exposure to airborne contaminants below the exposure limit.

Personal Protection:
Splash goggles. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Gloves.

Personal Protection in Case of a Large Spill:
Splash goggles. Full suit. Dust respirator. Boots. Gloves. A self contained breathing apparatus should be used to avoid inhalation of the product. Suggested protective clothing might not be sufficient; consult a specialist BEFORE handling this product.

Exposure Limits: Not available.

Section 9: Physical and Chemical Properties

Physical state and appearance: Solid. (Crystals solid.)

Odor:
Almost odorless; May gradually develop slight odor of ammonia, especially in presence of moisture.

Taste: Cooling. Saline

Molecular Weight: 60.06 g/mole

Color: White.

pH (1% soln/water): Not available.

Boiling Point: Not available.

Melting Point: 132.7°C (270.9°F)

Critical Temperature: Not available.

Specific Gravity: 1.323 (Water = 1)

Vapor Pressure: Not applicable.

Vapor Density: 2.07 (Air = 1)

Volatility: Not available.

Odor Threshold: Not available.

Water/Oil Dist. Coeff.: The product is more soluble in water; log(oil/water) = -2.1

Ionicity (in Water): Not available.

Dispersion Properties: See solubility in water.

Solubility: Easily soluble in cold water, hot water.

Section 10: Stability and Reactivity Data

Stability: The product is stable.

Instability Temperature: Not available.

Conditions of Instability: Excess heat, excess dust generation, incompatible materials.

Incompatibility with various substances: Reactive with oxidizing agents.

Corrosivity: Not available.

Special Remarks on Reactivity:
Hygroscopic. Aborbs moisture from air. Reacts violently with Gallum Perchlorate. Reacts with chlorine to form chloramines. It also reacts with the following: sodium hypochlorite, sodium nitrate, calcium hypochlorite, $NaNO_2$, P_2Cl_5, nitrosyl perchlorate, strong oxidizing agents (permanganate, nitrate, dichromate, chloride)

Special Remarks on Corrosivity: Not available.

Polymerization: Will not occur.

Section 11: Toxicological Information

Routes of Entry: Inhalation. Ingestion.

Toxicity to Animals: Acute oral toxicity (LD50): 8471 mg/kg [Rat].

Chronic Effects on Humans:
MUTAGENIC EFFECTS: Mutagenic for mammalian somatic cells. May cause damage to the following organs: blood, cardiovascular system.

Other Toxic Effects on Humans: Hazardous in case of skin contact (irritant), of ingestion, of inhalation.

Special Remarks on Toxicity to Animals: Not available.

Special Remarks on Chronic Effects on Humans:
May cause adverse reproductive effects (fetotoxicity) and genetic material (mutagenicity) based on animal studies. Passes through the placental barrier in human and is present in breast milk.

Special Remarks on other Toxic Effects on Humans:
Acute Potential Health Effects: Skin: Causes skin irritation. Eyes: Causes eye irritation. Inhalation: Causes irritation of the respiratory tract, nose, and throat, coughing and sneezing. May also affect blood, metabolsim and urinary system. Ingestion: Causes digestive (gastrointestinal) tract irritation with nausea, vomiting, and diarrhea. May affect behavior (altered sleep time, change in motor activity), cardiovascular system (heart rate), and the brain. May also affect the blood and may cause tumorigenic effects. Chronic Potential Health Effects: Prolonged exposure may cause adverse reproductive effects. Laboratory experiments on animals have resulted in mutagenic effects. Prolonged exposure or exposure at high concentrations may cause eye damage.

Section 12: Ecological Information

Ecotoxicity: Not available.

BOD5 and COD: Not available.

Products of Biodegradation:
Possibly hazardous short term degradation products are not likely. However, long term degradation products may arise.

Toxicity of the Products of Biodegradation: The product itself and its products of degradation are not toxic.

Special Remarks on the Products of Biodegradation: Not available.

Section 13: Disposal Considerations

Waste Disposal:
Waste must be disposed of in accordance with federal, state and local environmental control regulations.

Section 14: Transport Information

DOT Classification: Not a DOT controlled material (United States).

Identification: Not applicable.

Special Provisions for Transport: Not applicable.

Section 15: Other Regulatory Information

Federal and State Regulations:
Minnesota: Urea TSCA 8(b) inventory: Urea

Other Regulations:
OSHA: Hazardous by definition of Hazard Communication Standard (29 CFR 1910.1200). EINECS: This product is on the European Inventory of Existing Commercial Chemical Substances.

Other Classifications:

WHMIS (Canada): Not controlled under WHMIS (Canada).

DSCL (EEC):
R36/38- Irritating to eyes and skin. R40- Possible risks of irreversible effects. S24/25- Avoid contact with skin and eyes.

HMIS (U.S.A.):

Health Hazard: 2

Fire Hazard: 1

Reactivity: 0

Personal Protection: E

National Fire Protection Association (U.S.A.):

Health: 2

Flammability: 1

Reactivity: 0

Specific hazard:

Protective Equipment:
Gloves. Lab coat. Dust respirator. Be sure to use an approved/certified respirator or equivalent. Splash goggles.

Section 16: Other Information

References: Not available.

Other Special Considerations: Not available.

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