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EFFECT OF NUTRIENT LIMITATION AND LIGHT IN THE PRODUCTION OF POLYHYDROXYBUTYRATES AND GLYCOGEN BY CYANOBACTERIA CULTIVATED IN WASTEWATER

Report and Annexes

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Resum

Els plàstics no degradables són un greu problema mediambiental. L'ús de recursos vius per produir plàstics biodegradables podria ser una possible solució. Els polihidroxibutirats (PHB) que són polímers amb propietats similars al polipropilè i el glicogen, que pot convertir-se en bioetanol, son produïts per els cianobacteris i altres microorganismes. Els cianobacteris tenen l'avantatge d'acumular aquestes substàncies utilitzant només CO_2 i energia solar. El tractament d'aigües residuals es podria utilitzar com a medi de cultiu per cianobacteris per tal de reduir els costos de producció dels bioplàstics.

Els principals objectius d'aquest treball han estat estudiar l'ús dels cianobacteris com a tractament terciari en les aigües residuals, l'efecte de les limitacions de nutrients i les hores de llum en l'acumulació de PHB i glicogen, i la millor manera d'operar un reactor per seleccionar cianobacteris davant algues verdes.

S'ha aconseguit eliminar el 64,8% del nitrogen i el 70,5% del fòsfor provinent del tractament secundari i el digestat. Demostrant la capacitat dels cianobacteris per eliminar contaminants. Una concentració màxima de glicogen de 838 mg/L s'ha obtingut en condicions de limitació de nitrogen i 12h de llum i una de PHB de 104,2 mg/L en condicions de limitació de fòsfor i 24h de llum. Per tan es pot concloure que la limitació de nitrogen te més efecte en l'acumulació de glicogen, i la de fòsfor en la de PHB. Tanmateix, segons el Key performance indicator (KPI) econòmic definit, els beneficis obtinguts no justifiquen les despeses en il·luminació. Finalment, es pot dir que les condicions de cultiu òptimes són limitació de fòsfor i il·luminació durant 12h diàries.

Resumen

Los plásticos no degradables son un grave problema medioambiental. El uso de recursos vivos para producir plásticos biodegradables podría ser una posible solución. Los polihidroxibutirats (PHB) que son polímeros con propiedades similares al polipropileno y el glucógeno, que puede convertirse en bioetanol, son producidos por las cianobacterias y otros microorganismos. Las cianobacterias tienen la ventaja de acumular estas sustancias utilizando sólo CO_2 y energía solar. El tratamiento de aguas residuales se podría utilizar como medio de cultivo para cianobacterias para reducir los costes de producción de los bioplásticos.

Los principales objetivos de este trabajo han sido estudiar el uso de las cianobacterias como tratamiento terciario en las aguas residuales, el efecto de las limitaciones de nutrientes y las horas de luz en la acumulación de PHB y glucógeno, y la mejor manera de operar un reactor para seleccionar cianobacterias ante algas verdes.

Se ha conseguido eliminar el 64,8% del nitrógeno y el 70,5% del fósforo proveniente del tratamiento secundario y del digestado. Demostrando la capacidad de las cianobacterias para eliminar contaminantes. Una concentración máxima de glucógeno de 838 mg / L se ha obtenido en condiciones de limitación de nitrógeno y 12h de luz y una de PHB de 104,2 mg / L en condiciones de limitación de fósforo y 24h de luz. Por lo tanto se puede concluir que la limitación de nitrógeno tiene más efecto en la acumulación de glucógeno, y la de fósforo en la de PHB. Sin embargo, según el Key performance indicator (KPI) económico definido, los beneficios obtenidos no justifican los gastos en iluminación. Finalmente, se puede decir que las condiciones de cultivo óptimas son limitación de fósforo e iluminación durante 12h diarias.

Abstract

Non-degradable plastics are a serious environmental problem. The use of living resources to produce biodegradable plastics could be a possible solution. Polyhydroxybutyrates (PHB), which are polymers with similar properties to polypropylene, and glycogen, which can be converted into bioethanol, are produced by cyanobacteria and other microorganisms. Cyanobacteria have the advantage of accumulating these substances using only CO_2 and solar energy. Wastewater treatment could be used as a culture medium for cyanobacteria to reduce the bioplastics production costs.

The main objectives of this work are to study the use of cyanobacteria as tertiary wastewater treatment, the effect of nutrient limitations and light hours on the accumulation of PHB and glycogen, and the best way to operate a reactor to select cyanobacteria against green algae.

It has been possible to eliminate 64,8% of the nitrogen and 70,5% of the phosphorus coming from the secondary treatment and the digestate. Proving the ability of cyanobacteria to remove contaminants. A maximum glycogen concentration of 838 mg / L was obtained under nitrogen limitation and 12 h light conditions and a PHB of 104,2 mg / L under phosphorus limitation and 24 h light conditions. Therefore, it can be deduced that nitrogen limitation has more effect on glycogen accumulation, and phosphor on PHB. However, according to the defined economical Key performance indicator (KPI) the product benefits do not justify the lighting expenses. Finally, it can be said that the optimal cultivation conditions are limitation of phosphor and 12h light per day.

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1. Introduction

Plastics have become one of the most widely used materials over the world, their applications are universally important: they are found in automobiles, home appliances, computer equipment, packaging, medical applications and in other multitude of objects and utensils of common use. It has been estimated that more than 100 million tonnes of plastics are produced every year worldwide and 60 kg per capita are consumed in Europe. 40% of the produced plastics are discarded to landfill and several hundred thousand tonnes into marine environments [1] accumulating in the global environment at a rate of 25×10^6 t per year provoking serious threats to the solid waste management program [2]. So, today demand for biodegradable plastics has become one of the most important targets in current research. In the early 1920s, Lemoigne a microbiologist at Pasteur Institute in Paris isolated a polymer from *Bacillus megaterium* by chloroform extraction and demonstrated that it was a polyester of 3-hydroxybutyric acid. Later, it was found that PHB is only one type in a huge family of polymers collectively known as polyhydroxyalkanoates (PHA). Among the 150 different types of polyhydroxyalkanoids identified so far, PHB is the more widespread homopolymer in different taxonomic group of prokaryotes including cyanobacteria. The PHB properties including thermoplastic process ability, absolute water resistance and biodegradability makes PHB a good option to substitute common plastics such as polypropylene. The main drawback of these bioplastics are they high production costs compared to the petroleum derived plastics. Cyanobacteria can be an alternative host system due to their minimal nutrients requirements, as they are photosynthetic microorganisms. Cyanobacteria have the capacity of accumulate PHB under photoautotrophic conditions, this means that they have the ability to convert carbon dioxide, a greenhouse gas, into environmentally friendly plastics just with sunlight energy [2]. Another way to reduce the costs associated with PHB production, could be using cyanobacteria born in wastewater. Substrates found in waste water could be used as a feed, considerably reducing the production costs. However, the problem of using wastewater to growth cyanobacteria is the competition with green algae [3].

Cyanobacteria can not only accumulate PHB but also other interesting compounds such as carbohydrates in the form of glycogen, which can be converted into bioethanol by means of bacterial fermentation or sacarification processes.

Cyanobacteria, besides solar energy and carbon dioxide, need nutrients, such as nitrogen or phosphorus for their growth. If a nutrient is limited or omitted from the cultivation medium, they change their metabolic strategies resulting in a higher accumulation of interesting metabolites such as carbohydrates or PHB.

1.1. Project Objectives

The objectives set for this project are:

- To conduct a bibliographic revision on wastewater treatment, cyanobacteria production of glycogen and PHB and types of photobioreactors used in cyanobacteria cultures.
- To check the possible use of a 30L photobioreactor to grow wastewater-born cyanobacteria, as well as in the tertiary wastewater treatment.
- To investigate the effect of nitrogen and phosphor limitation and light hours in the production of PHB and glycogen in a mixed cyanobacteria culture.
- To study which reactor operation best select cyanobacteria increasing thus the accumulation of PHB.

1.2. Project scope

The purpose of this project is to find cyanobacteria cultivation conditions that allow to reduce the bioplastics production costs while increasing the cyanobacteria PHB productivity, in order to facilitate the transition towards industrial application. One possible way to reduce the production costs is the valorisation of the biomass used to treat wastewater. With this strategy nutrients for cyanobacteria cultivation will be extracted from wastewater treatment processes, which is in many industries anyway necessary. Thus, the sub-products of this treatment will be revalorized. Furthermore, it should be taken into account that cyanobacteria are photosynthetic microorganisms that fix CO2, so the use of this process should be seen as a carbon capture and storage (CCS) technology.

In order to become closer to the industrial application of this systems, in this project three main experiments have been done. First, a study of the use of high rate algae ponds as a wastewater treatment process and the possibility of using a closed photobioreactor as a tertiary treatment and to cultivate cyanobacteria was done. After that different nitrogen and phosphorus limitations and light conditions were tested in order to find how nutrient limitation and light conditions affect PHB and glycogen accumulation on wastewater born cyanobacteria. Furthermore, different reactor operation have been studied in order to select cyanobacteria in a mixed culture.

2. Wastewater treatment

In this section a short review of the conventional methods used for waste water treatment will be done. Thereafter, the treatment used for this project, the high rate algae ponds will be explained.

2.1. Conventional wastewater treatment

Conventional wastewater treatment consists of a combination of physical, chemical, and biological processes and operations to remove solids, organic matter and, sometimes, nutrients from wastewater. The wastewater treatment process is divided into levels depending on the degree of treatment. The levels are pretreatment, primary, secondary, tertiary and advanced treatment. In Figure 1 there is an scheme of the conventional wastewater process[4].

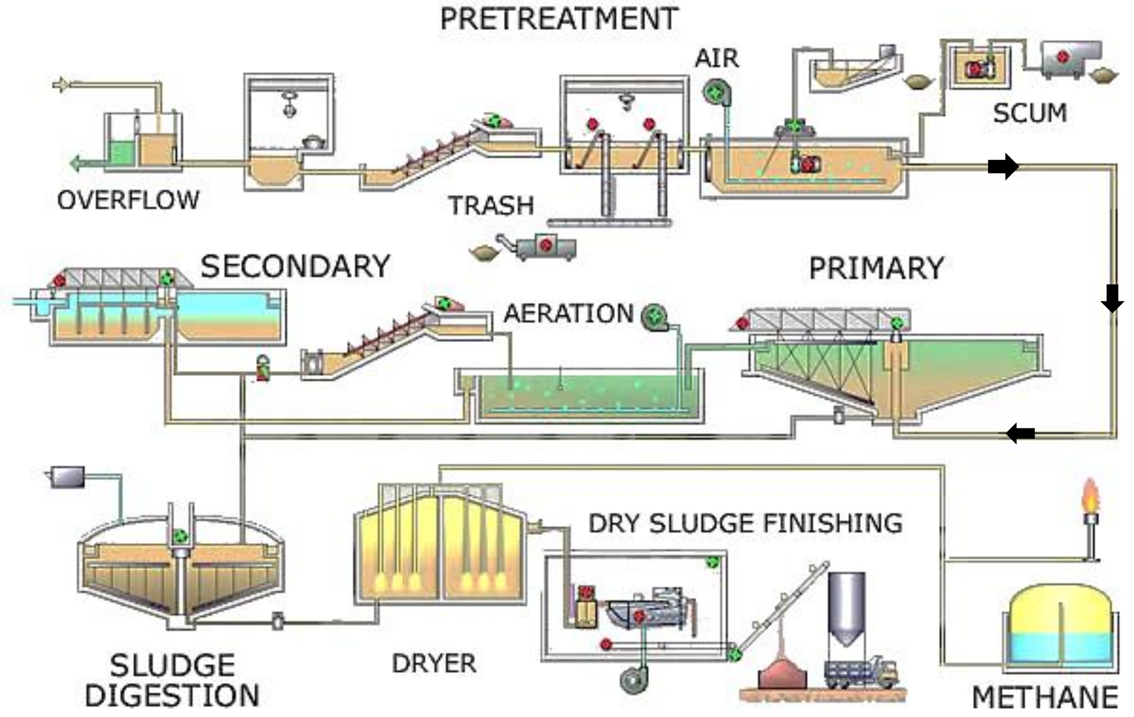


Figure 1. Conventional wastewater treatment process [5]

a) **Pretreatment**

The aim of the pretreatment is to remove coarse solids and other large material often found in wastewater. This material removal is needed for the correct operation and maintenance of the subsequent treatment units. Pretreatment operations typically include coarse screening, grit removal and, in some cases, large objects removal [4].

b) Primary treatment

The objective of the primary treatment is to remove the sedimentary organic and inorganic solids by sedimentation and skimming (elimination of materials that float). In this treatment step it is eliminated about 25-50% of the biochemical oxygen demand, 50-70% of the total suspended solids and 65% of the oil and grease. The primary treatments are done in sedimentation tanks or clarifiers (Figure 2), which may be round or rectangular with 3 to 5 m deep and with and hydraulic retention time of 2 or 3 hours. Settled solids are normally removed by the action of rakes which scrape the solids to the bottom of the tank where they are pumped to sludge processing units. Some of the sludge treatment used in big sewage plants is anaerobic digestion. In the digestion process anaerobic bacteria metabolize the organic material in sludge converting it into biogas and digestate. The biogas is composed by CH_4, CO_2, H_2, H_2S, being methane the major component, so it could be used as a combustible, and digestate which is a mixture of mineral components (N, P, K, Ca) (See 6.1.1.1 Anaerobic biodigester). Thereby, the volume for the ultimate sludge disposal is reduced and the sludge is stable at the same time that biogas is generated [4].

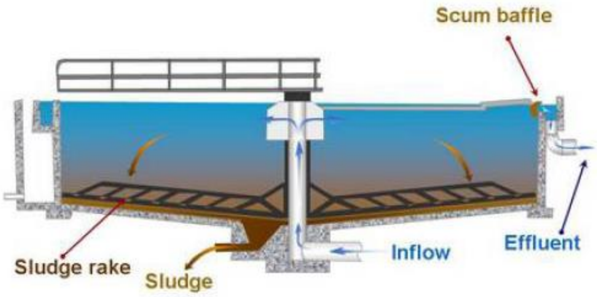


Figure 2. Sedimentation tank

c) Secondary treatment

The aim of the secondary treatment is the removal of organic residual and suspended solids of the effluent from primary treatment. In most cases secondary treatment uses biological treatment process. Aerobic biological treatment is performed in the presence of oxygen by aerobic microorganisms that metabolize the organic matter in wastewater thereby the microorganisms population grow and the organic matter is converted into inorganic end-products (CO_2, NH_3 and H_2O). The microorganisms used in this treatment step must be separated from the treated water. This is done by sedimentation tanks as in primary treatment. The solid removed, usually called secondary biological sludge, are normally combined with those from primary treatment and processed the same way [4].

There are several aerobic biological processes used for secondary treatment. The main difference is how the oxygen is supplied and the rate at which the organic matter is metabolized [4].

a. Activated sludge: Consist in an aeration tank containing a suspension of wastewater and microorganisms. The tank content is mixed by aeration devices which also supply the oxygen to microorganisms. Aeration devices are commonly submerged diffusers that release compressed air. The normal hydraulic retention time is between 3 to 8 hours. A portion of the biological sludge is recycled to maintain a high mixed-liquor and the remainder is removed to sludge treatment.
b. Trickling Filters: They consist of a basin or tower filled with support media such as stones, plastic or wooden salts. The microorganisms remain in the media and form a biological layer. After that, the organic matter in the water pass through the film and the biological layer metabolize it. Oxygen is supplied by the natural air flow. Periodically, part of the biofilm is removed from the media to maintain a correct thickness.
c. Rotating biological contactors (RBCs): Are fixed-film reactors in which organisms are fixed to slowly rotating discs that are partially submerged in flowing wastewater. Oxygen is supplied from the air when the film is out of the water and from the liquid, which have dissolved oxygen due to disc rotation, when submerged

Figure 3. a) Aeration tank, b) trickling filters, c) Rotating biological contactors

d) Tertiary treatment

It is used when specific wastewater constituents which cannot be eliminated by secondary treatment must be removed. This treatments are needed to remove nitrogen and phosphorus compounds, additional suspended solids, heavy metals and dissolved solids [4].

2.2. High rate algae ponds

The high rate algae ponds are used as a secondary wastewater treatment. They consist in a race-way pond that usually have a paddlewheel that provides mixing at a horizontal velocity of approximately 0,15-0,3 m/s. The configuration could be done in a unique loop or multiple loops around central division walls. CO_2 could be injected in counter current or just use the CO_2 produced by the heterotrophic bacteria [6]. The microalgae that grow in the pond provide oxygen to heterotrophic microorganisms that degrade the organic residues. The algae used in the wastewater treatment could be reused in order to feed animals or to generate biofuels. This kind of system are much simpler to use and cheaper than other methods such as activated sludge [7], although they need much more land taking into account the biological oxygen demand (BOD) that they can remove[6].

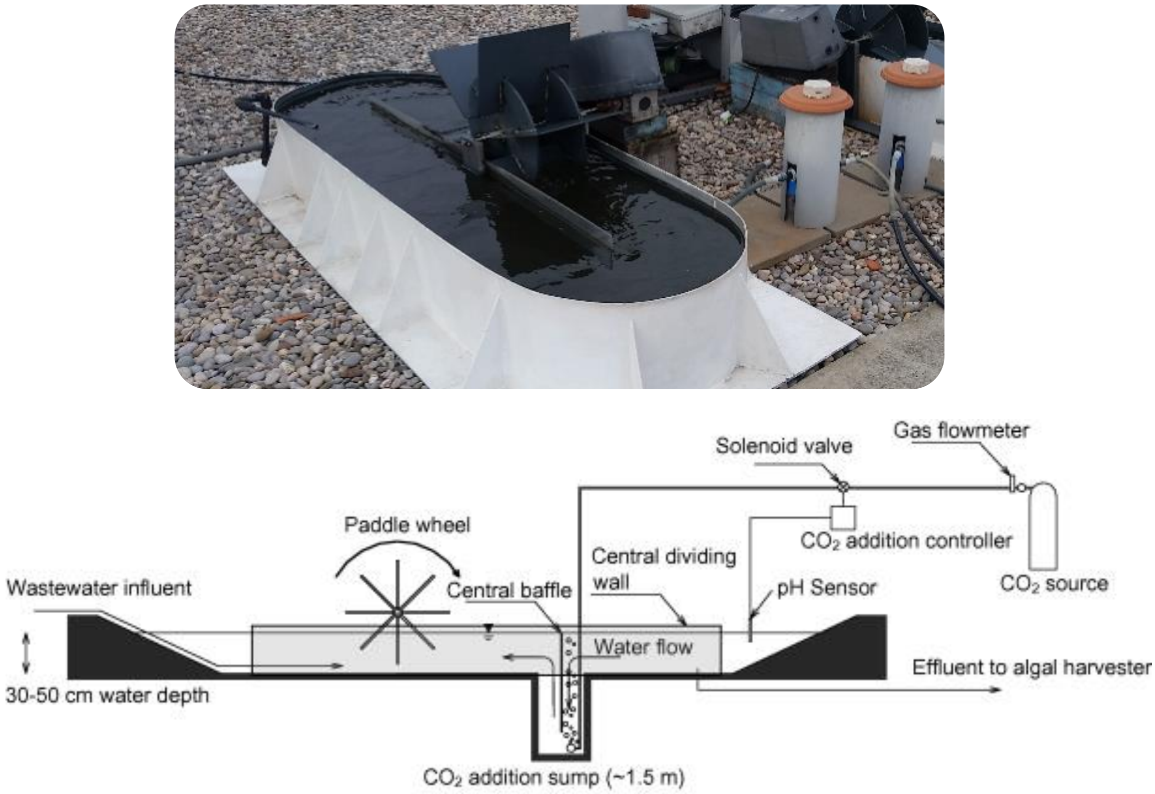


Figure 4. Picture of the high rate algae pond used in this project and scheme of a HRAP [6].

The efficiency of this process is related to the photosynthetic activity that causes variations in the dissolved oxygen and the pH. At midday the concentration of oxygen in water is high because of the high sun intensity. The concentration could exceed 20 mg/L and pH 11 due to the photosynthesis where CO_2 is consumed increasing the pH and oxygen is released rising the dissolved oxygen concentration [6]. Nevertheless, at night those parameters could take low values, even anoxic values. At night the aeration process is mainly due to the diffusion between atmosphere and the pond, but this is not enough for the aerobic microorganism, causing low process efficiency [7]. Therefore the good or bad functioning of the high rate algae pond is really dependent on the environmental parameters such as light energy, temperature, pH, dissolved oxygen and nutrients [7].

- **Light intensity**: light is one of the most important parameter affecting the whole system operation as it is the energetic source necessary by the microalgae system to do the photosynthesis. However, high light intensities could even cause photo inhibition, which damage the light receptors of algae decreasing the photosynthetic rate [6]. The high biomass content can cause that the light just arrives up to the first 0.15 m, for that reason it is necessary to use a paddlewheel to mix the culture and ensure that all the algae biomass receives light.

- **Temperature:** biologic reactions, algae species and physical chemistry parameters, such as pH, electric conductivity, gas solubility and alkalinity are temperature dependent. The optimum temperature in microalgae cultures is between 20 and 24 ºC. Temperature lower than 16 ºC causes a low growing rate and temperature higher than 35 ºC causes algae death for many species.
- **pH:** The pH affects the biochemical processes related to the algae growth including the CO_2 availability. The CO_2 dissolution is high pH dependant. The CO_2 in contact with water is transformed into carbonic acid and then in carbonate and bicarbonate. Depending on which is the pH of water the chemical compound will change. The optimum pH is around 8. In the following chemical reaction it can be seen the chemical equilibrium of CO_2 and the equilibrium constants at 25 ºC [8].

$$CO_2 + H_2O \xleftrightarrow[\text{Pkh}]{} H_2CO_3 \xleftrightarrow[\text{pKa1}]{} H^+ + HCO^-_3 \xleftrightarrow[\text{pKa2}]{} 2H^+ + CO_3^{2-} \qquad \text{(Eq. 1)}$$

pKH = 1,47

pKa1=3,6

pKa2=10,33

- **Dissolved oxygen**: during the day high levels of dissolved oxygen could be achieved, this levels could even cause bacterial oxidation and affect the algae productivity.
- **Nutrients**: the critical nutrient necessary for algae growth are nitrogen compounds. Wastewater provide the necessary nitrogen for algae growth.

3. Cyanobacteria

Cyanobacteria are microorganism of the domain bacteria. Nevertheless, as they are photosynthetic prokaryotes they are sometimes considered as algae (blue green algae) [9]. What makes the differences between bacteria and algae is that bacteria are prokaryote and algae are all eukaryote. The main distinction of cyanobacteria with respect to another bacterial type is that they are photosynthetic organisms. Cyanobacteria are the oldest photosynthetic organisms on earth. They were originated between 2,6 – 3,5 billion years ago. It is though that the photosynthetic organelle in eukaryotes was created due to endosymbiosis with cyanobacteria. So it could be said that they are the predecessor of the whole plant kingdom. They exist in many different forms, such as unicellular, filamentous, planktonic, colonial, etc. They can grow in different ecological habitats (marine, freshwater, terrestrial). Cyanobacteria capacity to fix carbon dioxide thanks to photosynthesis makes them really interesting microorganisms for the industrially and environmentally friendly production of different chemicals and biofuels, in front of heterotrophic bacteria which requires of more complexes media. Furthermore, they are really efficient in the conversion of sunlight into biomass compared with plants [10]. They have three modes of growth, photoautrophic, mixotrophic and heterotrophic. In presence of light, the photoautotroph mode is exploit and CO_2 is used as a carbon source and light as energy. In the heterotrophic mode they obtain the carbon from organic sources. And in the mixotrophic conditions they are able to use at the same time inorganic and organic carbon in presence of light. This represents an advantage over just photoautotroph or heterotroph organisms [11].

In this section a review on the microorganisms capable of accumulating PHB, paying attention on the cyanobacteria will be done. An explanation of the cyanobacteria metabolic pathways to accumulate PHB and glycogen will be also done in order to understand how different nutrient starvation could affect the glycogen and PHB production.

3.1. Microorganisms that accumulate PHA

There are more than 250 different polyhydroxyalkanoates (PHA) producers [12]. PHA is found in heterotrophic bacteria, archaea, cyanobacteria and plants. Heterotrophic bacteria have the advantage that are easily cultivated for massive production [13]. The more used kind of bacteria are gram-negative bacteria for its great production amounts in front of gram-positive bacteria [14]. Archaea are considered as extremophiles, the unique archaea reported to be capable of storing PHA are haloarchaeal species, so salt is needed to sustain their growth [14]. The first case of PHA accumulation by archaea was reported in 1970 in the Dead Sea by *Halobacterium marismortui.* PHA has also been found in plants as membrane constituents, their functions in plants seems to be voltage-gated calcium channels or DNA transport protection of the macromolecules [1]. In the case of cyanobacteria they have the advantage of being photosynthetic microorganisms, so their nutrients requirements are even lower than those from heterotrophic bacteria, so the production cost could be drastically reduced by using these microorganisms. Furthermore, they contribute to CO_2 fixation. The problem of cyanobacteria is that its PHA accumulation rate is low in comparison with heterotrophic bacteria. Although it is generally though that PHA is used as a carbon storage compound, its function in cyanobacteria is not really clear as they have glycogen as energy storage and it is unlikely that both macromolecules are used for the same purpose.

It is thought that PHB could be a specific energy storage for the amino acids, chlorophyll and carotenoids synthesis. PHB could also act as a sink of electrons as it is a reduced molecule [15]. Or also as a mechanisms to decrease the excess of reducing equivalents [16].

In Table 1 there is a summary of the more important PHB producers of each of the microorganisms domains, the carbon source used to feed them and the PHA concentration that has been reported.

Table 1. Most important microorganisms capable to store PHA

Microorganisms	**Carbon source**	**PHA content (%)**	**Reference**
Gram negative heterotrophic bacteria			
Azohydromonas australica	Malt waste	70,1	[17]
Azohydromonas lata	Sucrose	50-88	[18][19][20]
	Fructose, glucose	76,5-79,4	[21]
Burkholderia cepacia	Xylose	58,4	[22]
	Glycerol	31,3	[23]
	Fructose, glucose, sucrose	50,4-59,0	[21]
Cupriavidus necator	4-Hydroxyhexanoic acid	67,2	[24]
	CO_2	60	[25]
Gram positive heterotrophic bacteria			
Bacillus megaterium	Citric acid, glucose, glycerol, succinic acid	9-50	[26]
Corynebacterium glutamicum	Acetic acid, citric acid, glucose, glycerol, succinic acid	4-32	[26]
Archaea			
Haloferax mediterranei	Vinasse	50-73	[27]
	Hydrolyzed whey	72,8	[28]
	Glycerol and crude glycerol from biodiesel production	75-76	[29]
Various archaeal strains	Fructose, glucose, glycerol	0,8-22,9	[30]

Cyanobacteria			
Spirulina platensis	CO_2	6	[31]
Synechococcus MA19	CO_2	7-50	[32]
Synechocystis sp. PCC6803	Acetate	10	[33]
Synechocystis sp. PCC6803	Fructose/acetate	38	[34]
Nostoc muscorum	Acetate/glucose	45-60	[35]
Anabaena cylindrical 10C	Acetate + propionate	2	[36]
Oscillatoria limosa	CO_2/Acetate	6	[15]
Aphanocapsa sp	CO_2/glucose	2,16	[37]
Chrococcus sp	CO_2/glucose	3,11	[37]

A mixed culture of cyanobacteria and microalgae have been used in this project. The species from this culture are not always the same and the predominant species could change with time. The morphology of the most important cyanobacteria present in the mixed culture is reviewed next:

a) *Aphanocapsa* sp.: their cells are small (0,2-2 µm) and more or less spherical. They form microscopic colonies. Cell colorations vary from grey, olive, bright blue-green, and sometimes appear colourless [38], [39].

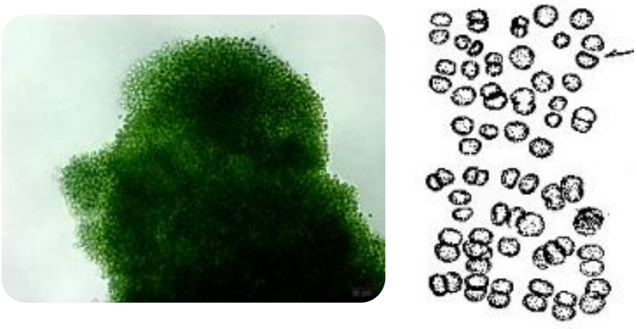


Figure 5. Microscope image and schematic drawing of Aphanocapsa sp. [38], [39].

b) Chrococcus sp.*:* They form colonies, usually with not many cells. The cells usually are hemispherical with homogeneous or granular content. Their colour is pale or bright blue-green, yellowish, pinkish or violet [38].

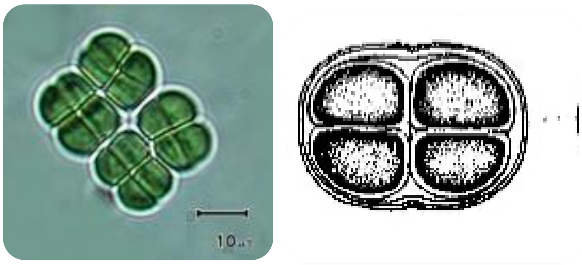


Figure 6. Microscope image and schematic drawing of Chroococcus sp. [38].

c) *Pseudanabaena* sp.: they are filamentous, usually solitary or agglomerated in very fine mats. They measure around 0,8-3 µm wide and they are not very long. They are composed by cylindrical cells [38].

Figure 7. Microscope image and schematic drawing of Pseudanabaena sp. [38].

d) *Synechocystis* sp*:* They are solitary, spherical cells that never form colonies. For short times they are seen as pairs [38].

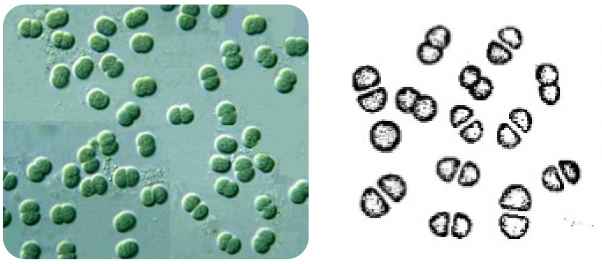


Figure 8. Microscope image and schematic drawing of Synechocystis sp. [38].

3.2. Cyanobacteria metabolic pathways

The photosynthetic microorganisms, such as microalgae or cyanobacteria capture the solar energy by the photosynthetic reactions. The photosynthetic reactions are divided in two main groups, the light reactions and the dark reactions. In the light reactions the photosynthetic pigments absorb the solar energy and this energy is used to split the water into protons, electrons and oxygen. The electrons and protons are used to synthetize energy carriers (NADPH and ATP). In the dark reactions carbon dioxide is fixed and reduced in the Calvin cycle using the NADPH and ATP generated in the light phase. The Calvin cycle assimilates CO_2 by the ribulose-1,5-biphosphate carboxylase oxygenase (commonly known as Rubisco). 6 molecules of carbon dioxide reacts with 6 molecules of the ribulose-1,5-biphosphate to form 12 molecules of three carbon compounds called 3-phosphoglycerate (3-PGA). One of them is used as substrate for the formation of pyruvate, while the others are used to accomplish the cycle [9]. Pyruvate is the precursor of many interesting compounds. Following the Calvin cycle the 3-phospohoglycerate molecules are converted into glyceraldehyde 3-phosphate (G3P) and from two molecules of G3P, Fructose 6-phosphate (F6P) is generated. Some of those formed molecules are used for the glycogenesis.

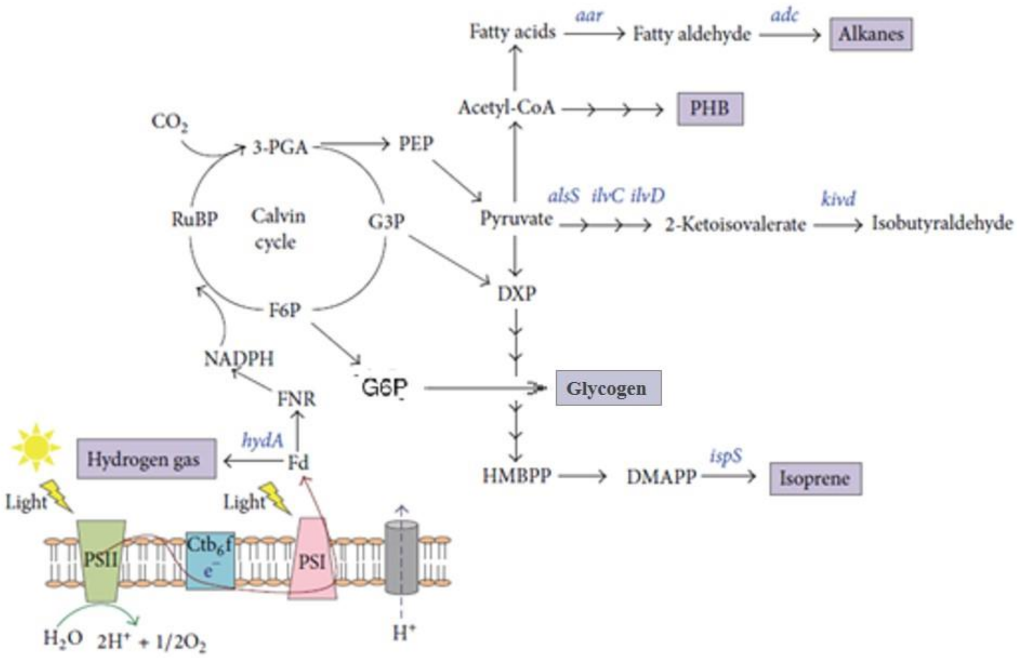


Figure 9. Scheme of the metabolism of a cyanobacteria. [[10]*modified]*

Many metabolic pathways are derived from pyruvate, such as Polyhydroxybutyrates (PHB) formation. The PHB is generated from acetyl CoA which comes from the oxidation of pyruvate as it can be seen in the following reaction:

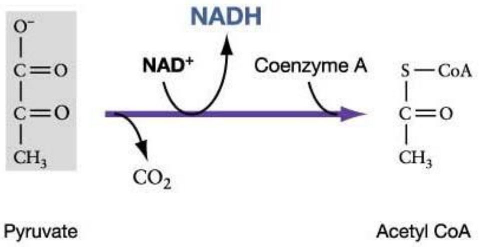


Figure 10.Pyruvate reaction to form Acetyl CoA [40].

Three enzymatic reactions should occur in order to generate the PHB. First, the 3-Ketothiolase converts two acetyl-CoA molecules in to one Acetoacetyl-CoA molecule. After that reaction acetoacetyl-CoA-reductase converts acetoacetyl-CoA in to D-3-hydroxybutyryl-CoA and finally the PHB synthase links the D-3-hydroxybutyryl-CoA to the existing PHB molecules via an ester bond making the polymer grow [11][2].

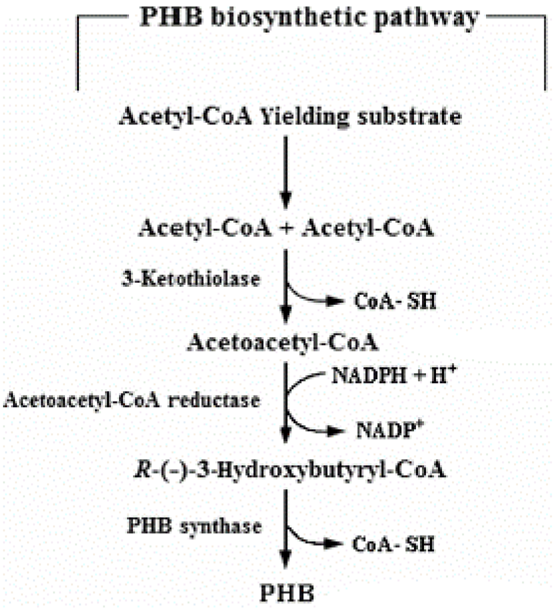


Figure 11. Metabolic pathway for the PHB synthesis [modified[11]*]*

In the case of glycogen it can be synthetized from F6P. The F6P is transformed into glucose 3-phosphate (G3P) which is at the same time converted into glucose-1-phosphate (G1P). G1P is converted to ADP-glucose by the action of glucose-1-phosphate adenyltransferase. The ADP-glucose is polymerized in order to form glycogen by the action of glycogen synthase. The AGPase activity regulates the glycogen synthesis in cyanobacteria. AGPase is allosterically activated by an increase in 3-phosphoglycerate (3PGA), and allosterically inhibited by an increase in inorganic phosphorus (Pi) content. In the following image it can be seen a scheme of the glycogen biosynthesis and the regulatory enzymes.

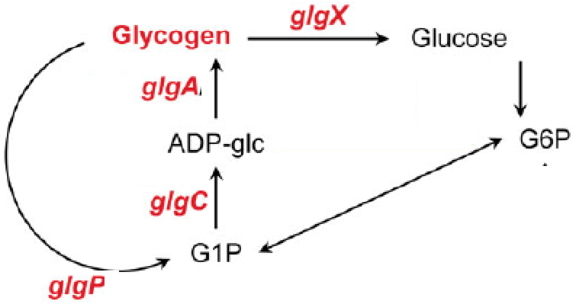


Figure 12. Scheme of the glycogen synthesis and the regulatory enzymes. Abbreviations: glgP, glycogen phosphorylase; glgC, glucose-1-phosphate adenylyltransferase; glgA, glycogensynthase; glgX, isoamylase. [[41] *modified]*

Cyanobacteria, besides solar energy and carbon dioxide, need nutrients, such as nitrogen, phosphorus, potassium, etc. for their growth. If a nutrient is limited or omitted from the cultivation medium, they change their metabolic strategies resulting to the alteration of their biomass composition. The alteration of the biomass composition is related to the accumulation of interesting metabolites such as carbohydrates or PHB.

Nitrogen is an essential nutrient. It is necessary for the generation of vital compounds such as DNA or proteins. It affects the synthesis of photosynthetic pigments such as chlorophyll. Under nitrogen starvation, the photosynthetically fixed carbon is turned from the protein synthesis metabolic pathway to the lipid or carbohydrate synthesis pathways, resulting into an accumulation either of carbohydrates or lipids. In the case of microalgae it is though that the nitrogen limitation causes lipid accumulation although it is possible that this is species-dependant. In the case of cyanobacteria the majority of the articles reported an accumulation of carbohydrates, in some cases up to a 70% of carbohydrates where found to be accumulated [9]. Nitrogen starvation also affects the PHA production. In this case the NADPH molecules availability diminishes, that inhibits the amino acid biosynthetic routes and turns the residual NADPH into PHB [11].

Phosphorus is also an essential nutrient that participates in the synthesis of vital organic molecules such as RNA, DNA, phospholipids and ATP. The carbohydrates synthesis is not an inorganic phosphorous consuming process and the ADP-glucose pyrophosphorylase which is the enzyme that controls carbohydrates synthesis is activated by the 3-phosphoglycerate and is inhibited by inorganic phosphorous. For that reason the level of accumulated carbohydrates is determined by the ratio of 3-phosphoglycerate to inorganic phosphorus. It is known that carbohydrates start to accumulate when the intracellular phosphorus drops below a threshold level[9]. Phosphorous also affects the PHA accumulation. It is thought that the PHA increase in phosphorus inadequacy could be due to a surplus of reducing power, as a result of the fact that NADPH is continually produced by photosynthetic electron flow whereas ATP generation declines with phosphorous deficiency [11].

Not only nutrients deprivations affect the accumulation of PHA and glycogen. Light quality and quantity also affects in the culture growth and the biomass composition. The microalgal and cyanobacteria growth rate increases with irradiance up to a level where photoinhibition occurs due to damage in the photosynthetic receptors. The majority of microalgae become saturated with a light flux of 200-400 µmolphotons $m^{-2}s^{-1}$. However, light penetration inside the reactor decreases as the cell density increases by the self-shading effect. In general it is though that more light increases the carbohydrate and PHA content [15]. It has also been seen that a rapid shift in light can cause a glycogen accumulation [16].

4. Cyanobacteria cultivation

4.1. Cultivation conditions

There are many parameters that affects the cyanobacteria growth, such as light, carbon source, nutrients, pH, and temperature.

a) **Light**

Cyanobacteria need light in order to do the photosynthesis. They need light within a wavelength between 400nm and 700nm. For satisfying biomass production, the light intensity has to be in a certain range. If the intensity is too low, light becomes a limiting factor, and if light is too high can lead to photoinhibition. That means that cyanobacteria will be not able to repair the photosystem II, which leads to a loss in the photosynthetic activity. Cells should be in constant and sufficient agitation in order that all the cells get light, this means turbulent flow inside the reactor. This cycle prevents the microorganisms from light starvation and also allows that the dark reactions to be completed and thus restore the capacity of the photosynthetic apparatus [42].

b) Carbon source

All the photoautotrophic organisms needs an inorganic carbon source. In almost all the species the inorganic carbon is introduced in the form of hydrogen carbonate. As explained in 3.2 cyanobacteria fix the carbon via the Calvin Cycle driven by the energy gained in the photosynthesis. CO_2 could be used as the sole carbon source for microalgal production, thus avoiding emitting a greenhouse gas into the atmosphere. If CO_2 is used is important to control the dosage in order to avoid unfavourable pH. The best way to dosage CO_2 is by using small bubbles aerators and injecting it just to maintain a correct pH [42].

c) Nutrients

Cyanobacteria needs other nutrients a part from carbon to grow, such as phosphorous or nitrogen. In the case of phosphorous they need less than 0,03 mg/L. Nitrogen content can be up to 10% of the dry matter weight. In many cases cyanobacteria prefer to assimilate nitrogen in form of ammonium. They do not assimilate other forms of nitrogen if ammonium is present. Other kind of nutrients such as iron and trace metal micronutrients (Zn, Cu, Ni) are also necessary for the cyanobacteria growth [42] .

d) pH

Cyanobacteria are not able to grow in pH lower than 4. The pH influences in the dissolution of inorganic carbon, therefore it also affects the cyanobacteria growth. Their optimum pH is between 7,5 and 10 depending on the strain and the environment. [42].

e) Temperature

The temperature at which cyanobacteria can grow depends also on the strain. It has been reported growth from 4ºC up to 75ºC. However, the optimal cultivation temperature is between 20 to 30 ºC [42].

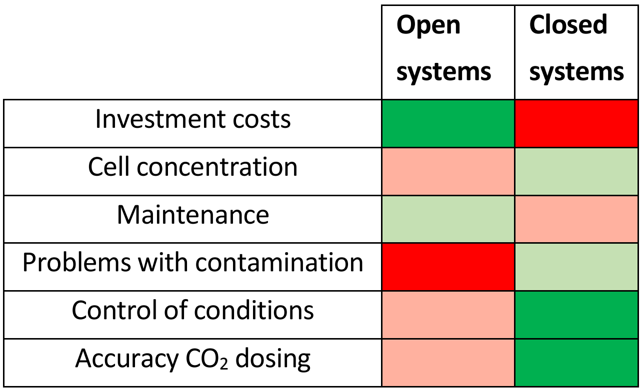
4.2. Photobioreactors

Cyanobacteria are cultivated in photobioreactors. Photobioreactors (PBR) are reactors in which phototrophs (microbial, algal or plant cells) are grown or used to carry out a photobiological reaction there are two main ways of cultivate photoautotrophic microorganisms at large scale production: open and closed systems. There are many ways to operate the same photobioreactor type, such as continuous operation, semi-continuous, batch, sequential bioreactors, etc. In this part a review on the most used photobioreactors and different operation manners that can be applied into a vertical column reactor, which is the type of reactor used for this project, will be done.

4.2.1. Types of photobioreactor

There are two big families of photobioreactors; Open, such as high rate algae ponds and closed. Table 2 illustrates the advantages and disadvantages of this two photobioreactor families have.

Table 2. Advantages and disadvantages of the open systems or closed systems. In green are the aspects in which the system is very good in red the aspect in which the system is very bad and in light colours the aspects in which the system is more or less good (light green) and in light red the aspect in which the system is rarely favourable [42].



	Open systems	**Closed systems**
Investment costs	🟩	🟥
Cell concentration	🟧	🟢
Maintenance	🟢	🟧
Problems with contamination	🟥	🟢
Control of conditions	🟧	🟩
Accuracy CO_2 dosing	🟧	🟩

In Table 2 shows that the open systems has a low investment costs but they have lot of contamination problems, control of conditions, CO_2 dosing is difficult to control and the cell concentration is low. However, they are easily to maintain. On the contrary, the closed systems are easier to control both the CO_2 dosing and the other parameters (pH, temperature). Furthermore, the cell concentration is high and the contaminations problems are avoided. However, they require from a biggest investment than open systems and their maintenance is also more expensive.

4.2.1.1. Open systems

Open ponds cultivation are the most used system for the commercial cultivation of microalgae. The open cultivation systems can be divided into three main systems: inclined systems, circular ponds and raceway ponds. The last one is the more used open system[43].

a) **Inclined systems**: in this type of reactors turbulence required for the algae growth is created by gravity. The culture flows from the top to the bottom of a sloping surface. That allows to have very thin culture layers (< 1cm) and a high turbulent flow. However, this systems presents several problems, such as sedimentation in the points where turbulence is low, strong losses by evaporation, high desorption of CO_2 and considerably energetic requirements.

b) **Circular ponds**: They are open vessels with a rotating arm that provides mixing. They are not commercially used due to the high energetic costs for mixing.

c) **Raceway ponds**: They consist on a raceway track where algae and nutrients are pumped around by a paddle. Its depth is around 25-30 cm, they cannot operate at a water level lower than 15cm. Due to the paddlewheel the algae is maintained in suspension. The ponds operate continuously with CO_2 and nutrients being constantly introduced by one side and algae containing water is removed in the other side. This constructions are inexpensive but its costs strongly depends in the ground characteristics. Their major drawbacks are the possible contamination, the evaporation in hot climates and the lack of control.

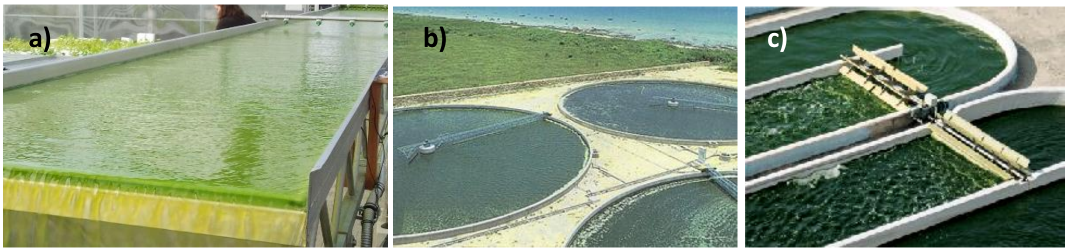


Figure 13. a) Inclined system image [78]*; b) circular pond image* [79]*; c) raceway pond image* [80]

4.2.1.2. Closed systems

There exist several types of closed systems. The most common ones are tubular reactors, laminar or flat panel reactors and vertical cylinders photobioreactors.

a) **Tubular**: they are the most used PBR for outdoor cultivation. They consist in narrow diameter tubes that allow the light to penetrate up to the tube centre. This maximizes the surface area available for photosynthesis. Usually as algae need to spend certain amount of time in darkness the tubular reactor is connected to a tank and the culture is constantly recirculated. The tube diameter usually ranges from 10-30 cm. They can be in form of horizontal / serpentine, vertical near horizontal, conical or inclined. Aeration and mixing of the cultures in tubular photobioreactors are usually done by air-pump or airlift systems. The benefits of the tubular reactors are a large illumination surface area, fairly good biomass productivities and they are relatively cheap. Their drawbacks are the pH, and dissolved CO_2 and O_2 gradients [44], [45].

b) **Flat panel**: They consist in glass flat chambers that can be titled at the proper angle to maximize the solar radiation capture, thus the main benefit of this type of photobioreactor is the large illumination surface area [43]. The dissolved oxygen that can be accumulated is low compared to the tubular photobioreactors. But great photosynthetic efficiencies were found in this kind of PBR. The benefits of using this PBR are the large illumination, their good biomass productivity, the relatively low price and maintenance. However, they require from many support materials, it is difficult to control the temperature and algal growth could exist in the walls [45].

c) **Vertical cylinders**: They are simple transparent cylinder reactor usually surrounded by a water jacket allowing temperature maintenance. The main benefit of this reactor type are good mixing, good temperature control, low photoinhibition and photo-oxidation. The problem of those reactors are the low illumination surface area, they require of sophisticated materials to be constructed and algal cultures suffer from shear stress [45]. This type of photobioreactor is the one used to realize this project.

Figure 14. a) Tubular photobioreactor [81];*b) flat panel photobioreactor* [82]; *c) vertical cylinder photobioreactor* [83].

4.2.2. Operation of the photobioreactors

In this project it has been used the vertical cylinder photobioreactor. Nevertheless there are several ways to operate this type of reactor. The operations used in this project are continuous, semi-continuous, batch and sequencing batch reactor. In this part a short explanation on this reactor operations will be done.

First of all, two generic concepts must be reviewed in order to understand the differences between the reactors operations: the hydraulic retention time and the solids retention time:

- **Reactor hydraulic retention time (HRT)**: It is the average time a compound remains inside the reactor.

$$HRT = \frac{Total\ Volume\ [m^3]}{Influent\ flow\ rate\ \ [\frac{m^3}{days}]} \qquad \text{(Eq. 2)}$$

- **Reactor solids retention time (SRT)**: The SRT represents the time spent by microorganisms in the system, or the time available for micoorganisms to reproduce. If SRT is longer than the regeneration time of an organism, it will proliferate. Otherwise it will be washed out of the system [46]. It is calculated by (Eq. 3.

$$SRT = \frac{Total\ Volume\ [m^3] \cdot VSS_{reactor}\ [\frac{mg}{L}]}{Influent\ flow\ rate\ \ \left[\frac{m^3}{days}\right] \cdot VSS_{effluent}[\frac{mg}{L}]} \qquad \text{(Eq. 3)}$$

Once this concepts are defined the different operations used in this project can be reviewed.

a) **Continuous reactor**: In this kind of operation the influent is all the time introduced into the reactor and the reactor content emerge continuously.
b) **Semi-continuous:** this operation way consist on a partial extraction of the reactor content and replacement for new nutrients solutions every defined amount of time in order to accomplish the defined hydraulic retention time.
c) **Batch**: this reactors are inoculated at the initial time and all the reactor content is extracted after a defined amount of time.
d) **Sequencing batch reactor (SBR)**: this operation manner is very used in the wastewater treatment. This reactors have the capability of having different hydraulic and solids retention times. Its operation consist in 5 phases.
    1. Filling: where the nutrients are introduced into the reactor.
    2. Reaction
    3. Settle: In this phase agitation is stopped and the biomass of the culture settles.
    4. Draw: Part of the liquid is extracted.

5. Idle: is the time between the decantation and the filling

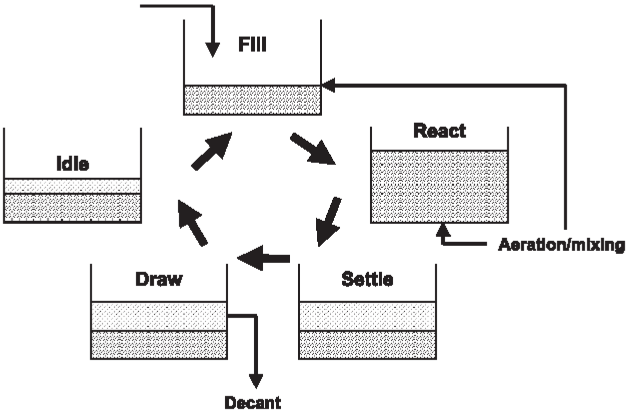


Figure 15. Scheme of a sequencing batch reactor [84].

5. Useful substances accumulated by Cyanobacteria

Cyanobacteria are able to accumulate different substances that are industrially and environmentally interesting, such as polyhydroxyalkanoates and glycogen. In this section it will be discussed the chemical structure and properties of polyhydroxyalkanoates and glycogen.

5.1. Polyhydroxyalkanoates

Plastics have become a really popular material due to the fact that they are easily modelled into almost any shape, they have high chemical resistance and stability, light weight, low price, and are more or less elastic. What makes them undesirable is the difficulty in their disposal [1][11]. Their big molecular size seems to be the main responsible for biodegradation resistance and their persistence in soils for a long time. It has been estimated that more than 100 million tonnes of plastics are produced every year worldwide. 40% of the produced plastics are discarded to landfill and several hundred thousand tonnes into marine environments. One option in dealing with non-biodegradable plastics has been incineration. However, harmful chemicals like hydrogen chloride and hydrogen cyanide are released to the atmosphere in this process. Recycling is also a solution but the big variety of plastics makes the recycled final product application difficult [1].

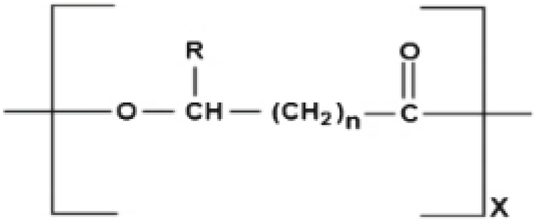
It seems that the best solution would be the replacement of non-biodegradable by biodegradable plastics. There are three types of biodegradable plastics: photodegradable, semi-biodegradable, and completely biodegradable. Photodegradable plastics have incorporated groups that are sensitive to light, so they can be disintegrated by ultra-violet radiation. However, landfills lack of sunlight what makes them remain non-degraded. Semi-biodegradable plastics are the ones that have polyethylene linked with starch. The bacteria in the soil will attack the starch and small polymer fragments will remain, this small fragments can be degraded by other bacteria. Completely biodegradable plastics are plastics generated by bacteria or cyanobacteria such as polyhydroxyalkanoates (PHA)[1].

The PHA are a versatile class of polymers with more than 100 different monomers that can be obtained from renewable raw materials such as corn, wheat, beets, sugar, potatoes, vegetable oils, or microorganisms [47]. Furthermore, they seem to have lots of different application such as in pharmaceuticals where they can be used in retarded drugs release, in agriculture where it is used to regularly discharge pesticides, plant growth regulators and herbicides, in medicine where could be exploited for the development of absorbable structures, bone plates, tendon repair devices, spinal fusion cages, ocular cell implants, skin substitutes, heart valves etc., in disposals such as food trays, razors, feminine hygienic products, bags, etc [11].

PHA is a natural carbon storage for prokaryotes that includes both heterotrophic bacteria and autotrophic bacteria (cyanobacteria). If it is wanted to produce PHA in eukaryotes the corresponding genes must be transferred. Using genetically modified plants is not the best option as they are not well accepted due to their environmental problems and they would be also a competitor for agricultural land which is not desirable considering the world food problems. The major benefits of using cyanobacteria are that they contribute to the atmospheric CO_2 consumption, its low nutrient requirements, they are aquatic microorganism so they do not need a cultivable land, so they could play an important role in the carbon capture and reusing technologies. The main drawback is, from an economic point of view, their natural low PHA content, almost always below 10% [42] in front of the heterotrophic bacteria which can accumulate around 80% of dry cell [48].

5.1.1. Chemical structure and properties

Polyhydroxyalkanoates (PHA) are predominantly linear, formed by hydroxyalkanoic acid as monomer units. The carboxyl group of one monomeric unit forms an ester bond with the hydroxyl group of the adjacent monomer. In Figure 16 it is shown an image of a monomer of a PHA. The X can attain a value of 35000 units. Each PHA monomer has an R chain group which can differ from a hydrogen atom to a methyl to tridecyl. The alteration in this side chains are contributing factor towards the diversification of PHA thermoplastics family [11]. In Table 3 there is summary of the molecular names that different kinds of PHA can have depending on its monomer structure.



$$\left[-O-\overset{R}{\overset{|}{CH}}-(CH_2)_n-\overset{O}{\overset{\|}{C}}- \right]_X$$

Figure 16. Monomer of a PHA[11]

Table 3. Molecular names of PHA depending on its monomer structure [49].

N value	R group	Molecule name
1	hydrogen	Poly(3-hydroxypropionate) [P(3HP)]
	methyl	Poly(3-hydroxybutyrate) [P(3HB)]
	ethyl	Poly(3-hydroxyvalerate) [P(3HV)]
	propyl	Poly(3-hydroxycaproate) [P(3HC)]
	butyl	Poly(3-hydroxyheptanoate) [P(3HH)]
	pentyl	Poly(3-hydroxyoctanoate) [P(3HO)]
	hexyl	Poly(3-hydroxynonanoate) [P(3HN)]

	heptyl	Poly(3-hydroxydecanoate) [P(3HD)]
	octyl	Poly(3-hydroxyundecanoate) [P(3HUD)]
	nonyl	Poly(3-hydroxydodecanoate) [P(3HDD)]
2	hydrogen	Poly(4-hydroxybutyrate) [P(4HB)]
3	hydrogen	Poly(5-hydroxyvalerate) [P(5HV)]

The PHAs can be divided into three big families: the short-chain length (SCL-PHA), the medium-chain length (MCL-PHA) and the long chain length (LCL-PHA). Among the SCL-PHA the most common are the Polyhydroxybutyrates. PHB properties such as glass transition temperature, melting temperature, young modulus and tensile strength seems to be similar to the ones from polypropylene. Other additional characteristics of PHB are non-toxicity, biodegradability, biocompatibility, hydrophobicity, piezoelectricity, and an optical purity [11].

The most important thermal properties for polymers are melting temperature (Tm) and glass transition temperature (Tg). Both of them are measured in order to know in which temperatures a polymer can be processed and used. The melting temperature is the temperature in which the polymer changes from solid to liquid state. The glass transition temperature is defined as the temperature at which the mechanical properties of a polymer drastically change because of the internal movement of the polymer chains. In the case of mechanical properties the most important for a polymer is Young's modulus, elongation-to-break and tensile strength. The Young's modulus studies the polymer stiffness or toughness measuring the relationship between stress (force per area) and strain (proportional deformation). A low value of young's modulus means a high flexibility. Young's modulus can be computed by the following equation:

$$E = \frac{Fa \cdot L_0}{A \cdot \Delta L} \qquad \text{(Eq. 4)}$$

Where E is the Young's modulus, F is the force exerted, A is the actual cross-sectional area through which the force is applied, ΔL is the amount by which the length of the object changes, L_0 is the original length of the object.

Elongation-to-break measure the magnitude that a substance will stretch before breaking. It is measured as a % of the initial polymer length. Finally, tensile strength measures the force required to break a material. In the following table there is a comparison between the most important thermal and mechanical properties of a biopolymer generated by cyanobacteria (PHB) and a petroleum based polymer (polypropylene).

Table 4. Properties comparison between PHB and polypropylene

Property	**Cyanobacteria (*Nostoc Muscorum*) PHB**[11]	**Polypropylene**
T_m(ºC)	175	171[50]
T_g (ºC)	0.9	-10[51]
Young modulus (GPa)	3.8	1.8[52]
Elongation-to-break (%)	4.9	150[53]
Tensile Strength (MPa)	30.2	33[53]

In Table 4 it can be seen how PHB properties can commercially compete with polypropylene, some of them are even better than those of polypropylene. The weaker property of PHB is elongation-to-break that could be improved synthetizing the co-polymer with hydroxyvalerate (HV) that achieve almost a 90% of elongation to break.

Although all the aforementioned properties can well compete with polypropylene properties, actually where PHB stands out is in its biodegradability. PHB are biodegradable in different environments such as compost, landfill, and aquatic systems. The ability to degrade PHB is widely distributed among bacteria and fungi. Even in extreme aquatic conditions, such as seasonal changes of oxygen concentration (anoxic to oxic), low pressure or no sunlight, the PHB is degraded. PHB depolymerases are excreted by a huge number of bacteria and fungi. The degradation occurs at the surface of PHB by enzymatic hydrolysis, so the degradation rate depends on the surface area available for colonization. In the degradation process the bacteria excrete PHA depolymerases which hydrolyze the polymer into water-soluble monomers which can be assimilated by microorganisms as nutrients, so the degradation components can be considered as non-toxic in nature[47].

5.2. Glycogen

The consumption of biofuels has risen in the last decade due to the interest of economical potencies on reducing its dependence on petroleum. However, the benefits of using biofuels has been questioned many times. From an environmental perspective damages in the biodiversity, are done by the intensification of monocultures and the use of fertilizers and pesticides. But the most concerning problem of classical biofuels is the food crisis. The increasing demand of biofuels makes that a great part of the crops produced are destined to the biofuels production causing a rice in the crops prices making them unaffordable for the most poor countries [54]. That is why microalgae and cyanobacteria biomass has been drawing attention for the production of biofuels. It has several advantages over the terrestrial biomass production. One of the most important benefits of using microalgae and cyanobacteria are that they are among the most efficient microorganism regarding solar conversion into interesting products. Moreover, they could be cultivated in non-arable land and their cultivation requirements are low, also another benefit is that microalgae and cyanobacteria could be used to treat wastewater[9].

Although carbohydrates have a lower energy content in comparison to lipids. They are preferable in the biofuels production[9]. In this project the culture used is mainly dominated by cyanobacteria which main storage carbohydrate is glycogen [55]. That is why in this section it will be explained the chemical structure of glycogen and which processes are used in order to convert carbohydrates in to biofuels.

5.2.1. Chemical structure of glycogen

Glycogen is a multi-branched polysaccharide of glucose monomers that is used as an energy storage molecule. Glycogen is branched with chains every 8 to 12 glucoses or so. The glucoses monomers are linked by (1-4) α-glycosidic bonds. The glycosidic bond is a covalent bond between the hemiacetal group of the glucose and the hydroxide group of the next glucose monosaccharide. The α indicates that both the hemicetal group and the hydroxide are in the same plane position. The branches are linked with a (1-6) α-glycosidic bond between the first glucose of the new branch. In Figure 17 there is an image of the molecular structure of glycogen. It could be clearly seen how the glycosidic bond changes when the chain is ramified [56].

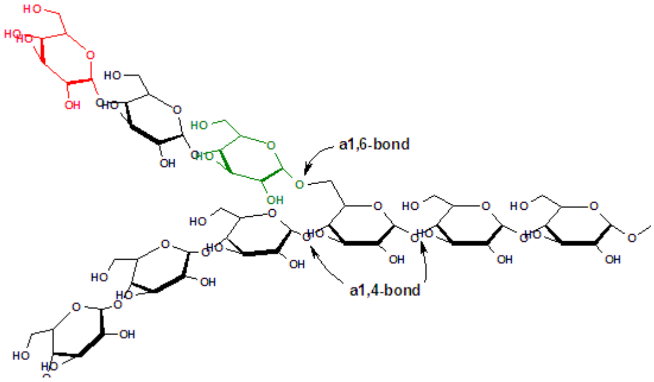


Figure 17. Molecular structure of glycogen: (1-6) α-glycosidic and (1-4) α-glycosidic bonds[56].

5.2.2. Chemical process to convert biomass into biofuels

Once the biomass of the photobioreactor is rich in carbohydrates different conversion technologies could be used in order to convert those carbohydrates into biofuels. In this section fermentation as a way to transform carbohydrates and sugars into bioethanol will be explained.

In Figure 18 there is a diagram of the process that the biomass follows in order to be converted into bioethanol:

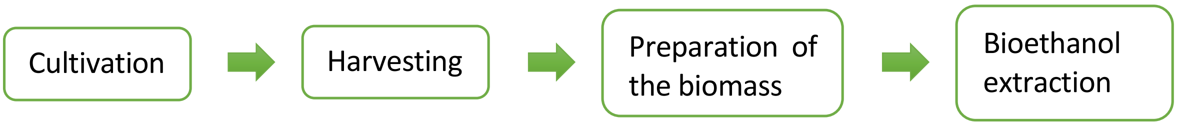


Figure 18. Bioethanol production process

The cultivation step should be done in a way that permits to accumulate the biggest quantity of carbohydrates in order to form more bioethanol [57].

The harvesting step allows to remove the aqueous solution from the biomass. Although in this project centrifugation has been used there are several harvesting methods[57]:

5.2.2.1. Centrifugation

Most microalgae and cyanobacteria can be recovered from dilute suspension through centrifugal forces. Centrifugation allows to have good harvesting with short time. However centrifugation energetic requirements are really high, so this process has a negative effect on the net energy and CO_2 balances in the biofuels production by microalgal and cyanobacteria means.

5.2.2.2. Filtration

This is the best accepted harvesting method due to its high efficiency, simplicity and continuous operation. Furthermore the medium used for the cultivation could be recovered.

5.2.2.3. Ultrasounds

Ultrasounds with a high frequency (MHz) and a low amplitude facilitates microalgal or cyanobacteria cells to aggregate and precipitate. Ultrasound may be unsuccessfully used in industry due to its low efficiency and high energetic costs.

5.2.2.4. Immobilization

This method use an entrapment matrix in which microalgal or cyanobacteria cells are embedded and continuously growing from the beginning of the cultivation. Thus, the beds where microalgae and cyanobacteria are cultivated could be removed easily without a large energy input.

The preparation of the biomass consist on the disruption of cell walls through various mechanical, chemical or enzymatic methods in order to enhance the carbohydrates availability and break down the carbohydrates molecules into sugars. After the biomass is prepared the yeast is supplied into the biomass for the fermentation process. Yeast is able to convert sugars into ethanol producing

CO_2.This CO_2 could be recycled as a carbon source for the cyanobacteria and microalgae culture. In Figure 19 there is a scheme of the fermentation reaction where glucose is converted into pyruvate generating 2 ATP and then this pyruvate is converted into acetaldehyde releasing two molecules of CO_2. Finally, the acetaldehyde is converted into ethanol [57].

After fermentation the bioethanol is purified so it can be finally be used as a combustible and the remaining biomass can be used as a feed in an anaerobic digester to produce biomethane [57].

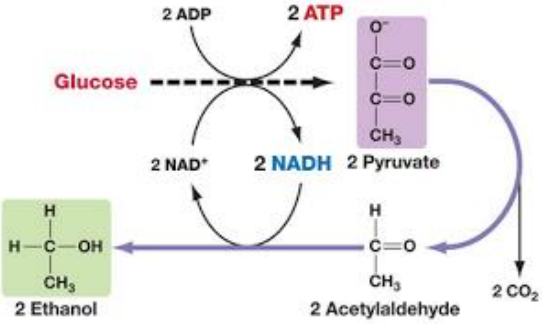


Figure 19. Fermentation reactions to form ethanol from glucose[58].

6. Material and methods

6.1. Material

In this part an explanation of the reactors used to develop this project will be done. Furthermore it will be explained the necessary reagents and equipment needed to realize the experiments explained in part 6.23.

6.1.1. Reactors

In this project three different reactors have been used. The anaerobic digester which produce biogas through the algae digestion. The 30L photobioreactor which is used as a tertiary wastewater treatment and as a culture medium to grow the cyanobacteria. The 3L vertical cylinder reactors were used to check which nutrients and light condition enhance the bioplastics and glycogen accumulation and also to see the reactors operation that allows a greater selection of cyanobacteria.

6.1.1.1. Anaerobic biodigester

Nowadays, almost all our biodegradable waste (food, garden waste, sewage, etc.) is send to landfill where it breaks down and realize methane which is a greenhouse gas. The anaerobic digestion allows to make this process in a closed reactor thus it is possible to recover the methane that could be used as a combustible [59].

The anaerobic digestion is a biological process where the organic matter is degraded to biogas and digestate through the action of bacteria in absence of oxygen. The biogas is composed by CH_4, CO_2, H_2, H_2S. The major component is CH_4 (50%-70%) that is why it could be used as combustible. The digestate is a mixture of mineral components (N, P, K, Ca) that can be used as nutrients in the photobioreactor [60].

Figure 20 summarizes the process that organic matter follows in the anaerobic digester.

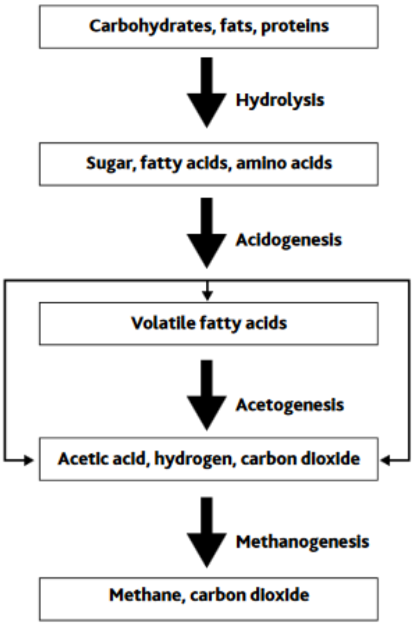


Figure 20. Scheme of the steps of anaerobic digestion [61]

The anaerobic digestion is based in 4 main steps[61][62].

1. **Hydrolysis**: Extracellular enzymes produced by hydrolytic microorganism, such as cellulose, amylase, protease and lipase, decompose complex organic polymers into soluble monomers. The proteins are broken down into amino acids, lipids into fatty acids, and carbohydrates into sugars. An example of hydrolysis reaction is the following:

$$C_6H_{10}O_5 + 2H_2O \rightarrow C_6H_{12}O_6 + H_2 \qquad \text{(Eq. 5)}$$

2. **Acidogenesis**: The small molecules resulting from hydrolysis are converted by fermentative bacteria into a mixture of volatile fatty acids (acetic, propionic, butyric) and other minor products such as hydrogen and CO_2. Usually this is the fastest step. The following equations shows the typical acidogenic reactions. In those equations the glucose is converted into ethanol (Eq. 6), propionate (Eq. 7), and acetic acid (Eq. 8):

$$C_6H_{12}O_6 \leftrightarrow 2CH_3CH_2OH + 2CO_2 \qquad \text{(Eq. 6)}$$

$$C_6H_{12}O_6 + 2H_2 \leftrightarrow 2CH_3CH_2COOH + 2H_2O \qquad \text{(Eq. 7)}$$

$$C_6H_{12}O_6 \rightarrow 3CH_3COOH \qquad \text{(Eq. 8)}$$

3. **Acetogenesis**: The acetogenic bacteria converts the volatile fatty acids into acetate, CO_2 and H_2. The following equations represent the basic acetogenic reactions. The first one represents the conversion of propionate to acetate. Glucose and ethanol are also converted to acetate in the acetogenesis.

$$CH_3CH_2COO^- + 3H_2O \leftrightarrow CH_3COO^- + H^+ + HCO_3^- + 3H_2 \qquad \text{(Eq. 9)}$$

$$C_6H_{12}O_6 + 2H_2O \leftrightarrow 2CH_3COOH + 2CO_2 + 4H_2 \qquad \text{(Eq. 10)}$$

$$CH_3CH_2OH + H_2O \leftrightarrow CH_3COO^- + 2H_2 + H^+ \qquad \text{(Eq. 11)}$$

4. **Methanogenesis**: The products form step 3 are converted into methane by methanogens. In the following equations it is showed the chemical reactions occurring in this step.

$$CO_2 + 4H_2 \rightarrow CH_4 + 2H_2O \qquad \text{(Eq. 12)}$$

$$2C_2H_5OH + CO_2 \rightarrow CH_4 + 2CH_3COOH \qquad \text{(Eq. 13)}$$

$$CH_3COOH \rightarrow CH_4 + CO_2 \qquad \text{(Eq. 14)}$$

The biodigester used in this project is a complete-mix digester. In Figure 21 there is image of the digester used:

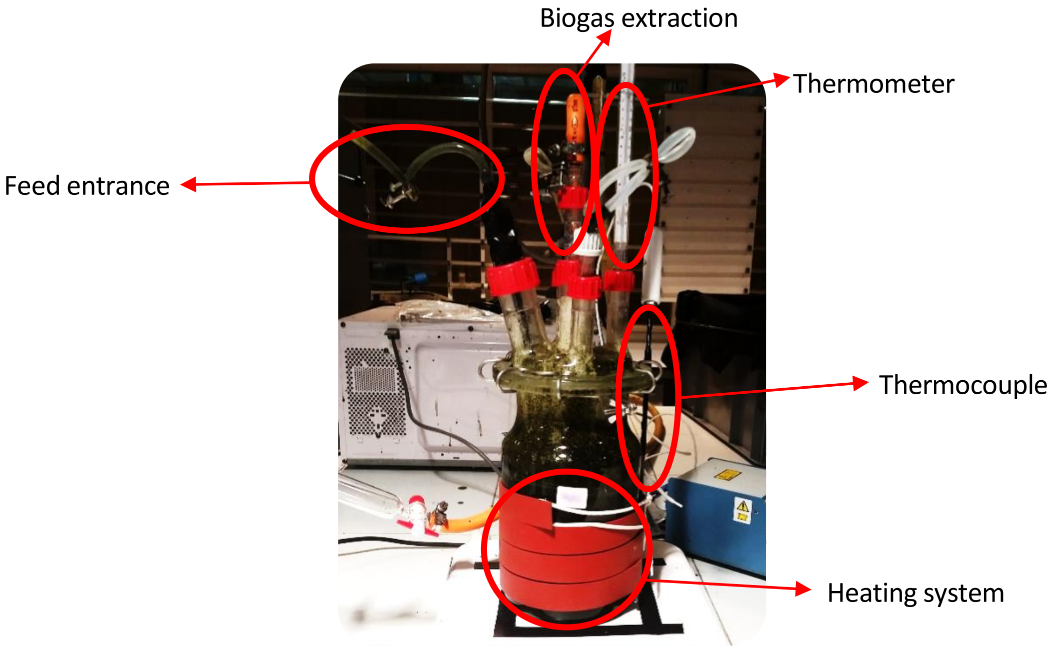


Figure 21. Anaerobic digester

The anaerobic digester used is equipped with a heater automatically controlled by a thermocouple which maintain 35ºC which is the adequate temperature for the digestion [61]. A thermometer which measures the inside reactor temperature, a biogas extractor from where the biogas is recovered and a hose by which the reactor is purged and fed.

6.1.1.2. 30L photo bioreactor

The 30L photobioreactor is a vertical cylinder reactor. In Figure 22 and Figure 23 there are images of the photobioreactor used. In annex 2 there can be found the reactor plans. This reactor is controlled automatically by a computer software. The pH increase is controlled by CO_2 injection which at the same time is used by cyanobacteria as inorganic carbon source. The CO_2 injected amount is controlled by the computer software which when receives the signal of a pH higher than 7,5 from the pH meter stars injecting CO_2 which is provided by a CO_2 gas bottle.

This reactor has a hydraulic retention time of 10 days which means that all the biomass inside the reactor will be completely renewed every 10 days. That retention time is accomplished by feeding the reactor with 3L every day.

$$HRT = \frac{Total\,Volume}{influent\ flow\ rate} = \frac{30\ L}{3\ \frac{L}{day}} = 10\ days \qquad \text{(Eq. 15)}$$

The PBR is fed by the feeding tank. This tank has a rotor available in order to make the feed dissolution homogenous. The feed dissolution is formed by the digestate extracted from the anaerobic digester and water from the high rate algae ponds. The quantity of digestate is introduced in a proportion of 5:300 mL of digestate per mL of influent. The harvesting tank is used to recover the PBR content that is extracted from the reactor every day. The system also has a light available. This light is surrounded by a screen in order to focus all the light to the reactor direction and avoid light loses. That light is controlled by a timer which turn on it just 12h a day.

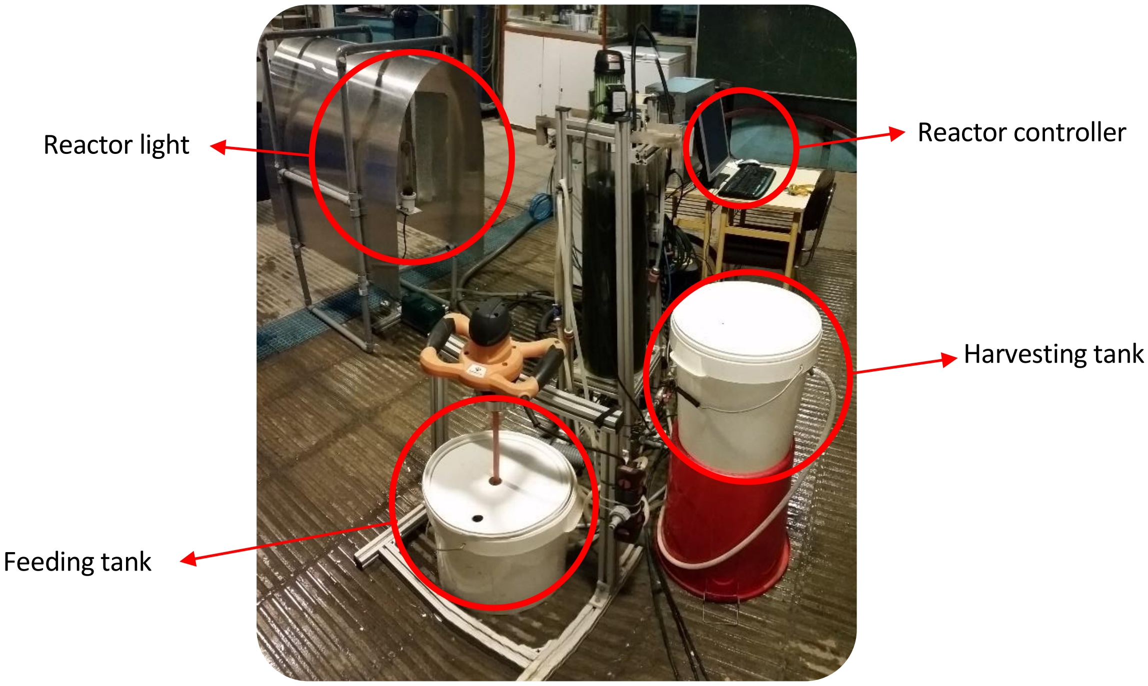


Figure 22. Image of the 30L PBR

Figure 23. 30L PBR tank

6.1.1.3. 3L vertical cylinder photobioreactors

They has been used to study which nutrient conditions and light are the best ones in order to produce high quantities of PHA and glycogen in a mixed cyanobacteria culture and which is the best operation to select cyanobacteria. They have a maximum capacity of 3L but they have been used at lower volumes than the maximum to avoid overflow when acid is added to control the pH. The pH is monitored by an electronic controller which when pH higher than 8,7 is detected activates a pump that introduces chlorhydric acid to the reactor. In some of the experiments it is also necessary to have a basic solution to avoid pH lower than 6,8. This reactors are jacketed photobioreactors in order to avoid a high raise in the temperature due to the heat emitted by the lamps. They also have an agitator that maintains the organic matter in continuous agitation, thus the light that arrives to each cell will be the same. In Figure 24 there is an image of the used reactor.

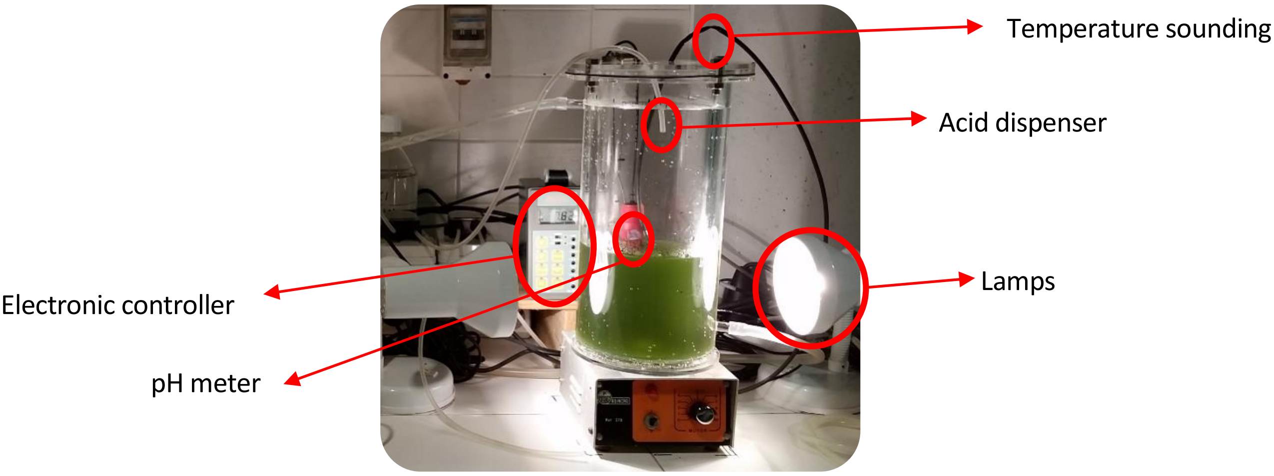


Figure 24. 3L vertical cylinder photobioreactor

In order to change the operation manner of those reactors different timers and pumps were connected. It was only necessary to automatize the continuous and SBR reactors, as batch and semi-continuous could be operated manually. In the continuous reactor two pumps were connected: one to continually fill the reactor and one that empties the reactor at the same time. In the case of SBR three timers were needed. First of all when the reactor is in agitation it is extracted manually the volume to accomplish the solid retention time. Then, timer 1 is activated and agitation is stopped for 1h. When half of this hour has passed it is considered that the reactor is completely settled and timer 2 is activated emptying the not settled content to accomplish the hydraulic retention time. When the emptying is completed timer 3 is activated and fills the reactor with new nutrients.

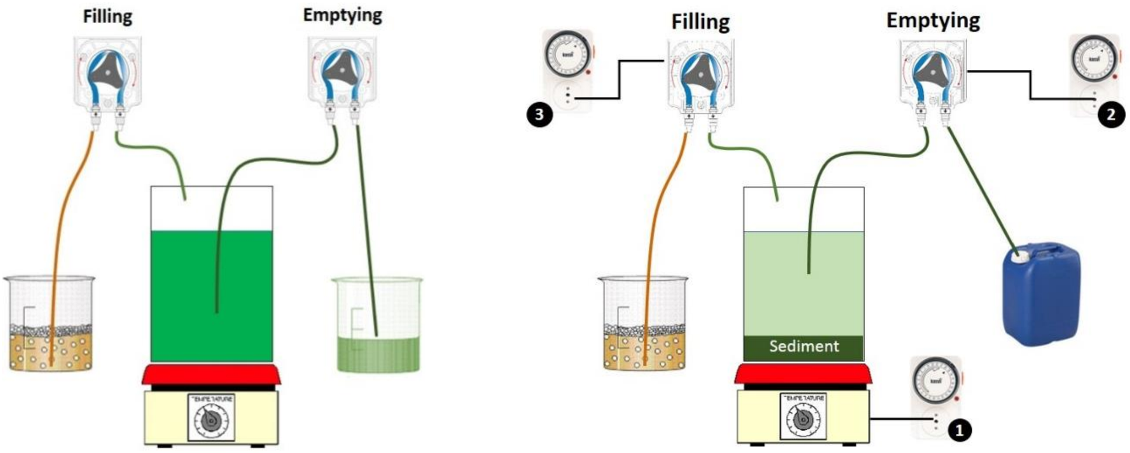


Figure 25. Continuous and SBR reactors

6.1.2. Reagents

In this section it will be explained which reagents are needed to make all the analysis and experiments.

- **BG11 culture medium**: It is the medium used in the batch reactors. It has been modified depending on the needed nutrient limitation. For example if nitrogen limitation wants to be done the $NaNO_3$ is eliminated of the solution and for phosphor limitation the K_2HPO_4 is eliminated. In Table 5 there is a summary of the solutions needed to prepare one litre of this culture [63].

Table 5. Preparation of the BG11 culture medium

Solution	Concentration [g/L]	Quantity used in the medium [mL]
$NaNO_3$[1]	15	100
K_2HPO_4[2]	4	10
$MgSO_2\cdot 7H_2O$	7,5	10
$CaCl_2\cdot 2H_2O$	3,6	10

---

[1] This solution is eliminated to prepare the nitrogen limited medium

[2] This solution is eliminated to prepare the phosphorous limited medium

Citric acid		0,6	10
Ammonium ferric citrate green		0,6	10
$EDTANa_2$		0,1	10
Na_2CO_3		2	10
Trace metal solution	H_2BO_3	2,86	1
	$MnCl_2\cdot 4H_2O$	1,81	
	$ZnSO_4\cdot 7H_2O$	0,22	
	$Na_2M_0O_4\cdot 2H_2O$	0,39	
	$CuSO_4\cdot 5H_2O$	0,08	
	$Co(NO_2)_2\cdot 6H_2O$	0,05	

- **COD**
    - Potassium dichromate solution 0,04167M:
    - Sulphuric acid with Ag_2SO_4 as additive at a rate of 5.5 g Ag_2SO_4/kg H_2SO_4.
- **Alkalinity**
    - Chlorhydric acid 0,02N
- **Chlorophyll**
    - Alkalized acetone
- **Ammonia**
    - Sodium nitroprussiade $NaFe(CN)_5NO\cdot 2H_2O$ 0,5%
    - Phenol solution 10% in ethanol.
    - Alkali solution: sodium citrate $C_6H_5Na_3O_7\cdot 3H_2O$ and NaOH
    - Oxidant solution formed by alkali solution and bleach (80:20)
    - Patron solution of ammonia
- **PHAs**
    - $CHCl_3 + C_{17}H_{36}$: 50 mg heptadecane + 20mL chloroform in 50mL volumetric flask.
    - PHB patron: PHB pellet of 30mg in a 10mL volumetric flask + 9mL chloroform
- **Glycogen**
    - Chlorhydric acid 0,1N
    - Concentrate sulphuric acid (95-98%)
    - Phenol 5% in water

6.1.3. Equipment

The equipment used for the analysis are listed below. More information about the use of those equipment could be found in the analytical methods section. The glass material has not been specified as it is the usual in a chemical laboratory.

a) Oxygen electrode
b) Neophelometer
c) Conductivity meter
d) TOC equipment (Analytikjena multi N/C 2100S)
e) Digester
f) Anion chromatograph (Diones ICS-1000)
g) Polytron
h) COD reader (MD 600)
i) pH meter
j) Centrifuge
k) Oven
l) Spectrophotometer (Spectronic Genesys 8)
m) Gas chromatograph (Agilent 7820A)
n) Muffle

Figure 26. Laboratory equipment

6.2. Aalytical methods

In order to control the changes in the mixed cyanobacteria culture and understand what is happening inside the reactors, different parameters have been continuously controlled. In this section a review on the different methodologies used to control the cultures will be reported. Otherwise specified, those methodologies are taken from standard methods for the examination of water and wastewater [64].

6.2.1. Turbidity

Water turbidity is caused by suspended matter. The measurement of turbidity express the optical property that causes light to be scattered and absorbed rather than transmitted with no change in the direction or flux level through the sample. Turbidity could be directly related with the total suspended matter. Nevertheless, depending on the sample interferences could be found. For example, samples with particles consisting on light absorbing materials could cause interferences when they are present in high concentrations. So, turbidity is an easy and quick way of measuring the biomass inside the reactor. In order to measure turbidity an electronic nephelometer is used.

The nephelometer measurements are based in a comparison of the light scattered by a standard reference and the light scattered by a sample under defined conditions. This nephelometer consist on a light source illuminating the sample and photoelectric detectors with readout devices capable to detect intensity of light scattered at 90ºC to the path of incident light. The units of measurement are the nephelometric turbidity units (NTU).

The measurements of turbidity must be done right after extracting the samples to avoid flocculation or changes with temperature. The procedure that should be followed in order to measure turbidity is the following:

1. Agitate gently the sample and wait until bubbles disappear
2. Pour the sample into the cell. Dry the cell with a tissue paper.
3. Introduce the cell into the dispositive and measure.

6.2.2. Dissolved Oxygen (OD)

The dissolved oxygen is a measurement of the oxygen present in the sample. It is an easy and quick measurement of the photosynthetic activity in the culture. To measure oxygen a membrane electrode has been used. The oxygen sensitive membranes are composed of two solid metal electrodes in contact with a supported electrolyte separated of the sample by a selective membrane.

It is preferable the OD to be measured in situ. If this is not possible samples should not be agitated and should be not in contact with air as the concentration of gases could be modified. In order to do the measurements first of all the electrode should be cleaned with deionized water and carefully dried with a tissue paper. Then, the electrode can be introduced into the sample and the device will show the oxygen concentration. The membrane electrode device can be seen in the following scheme:

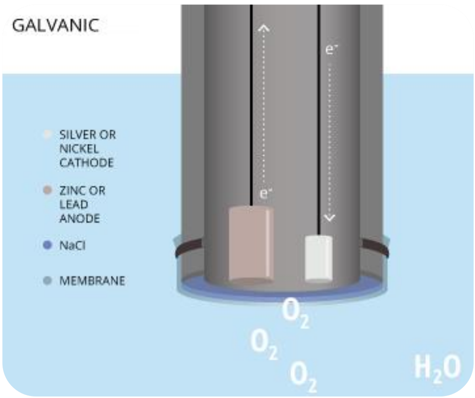


Figure 27. Galvanic membrane electrode [65].

6.2.3. Conductivity

The conductivity is a measurement of the capacity of an aqueous solution to conduct electric current. This measurement depend on the presence of ions. This conductivity is used to control the medium salinity, since the algae cultivated are fresh water species and high salinities could cause its dead. Conductivity can be related with salinity by an empirical relation between the salinity of a standard solution and the conductivity of this solution.

The conductivity has been measured by a conductivity meter. This conductivity meter is calibrated with different standard KCl dissolutions which permits the user to establish the internal cell constant in order to match the measured conductivity with the standard conductivity:

$$k_{measured} = C \cdot G = k_{solution} \qquad \text{(Eq. 16)}$$

Being k the conductivity C the cell constant in cm and G the solution conductance.

Once the conductivity meter is calibrated the sample can be measured. To do that clean the electrode with DI water and introduce it into the sample. Wait until the measurement is stable.

6.2.4. Total and volatile suspended solids (TSS, VSS)

The total suspended solids is the portion of solids retained by a filter of 2μm nominal pore size. The suspended solids is the weight loss after ignition of the solids retained in the filter. Volatile solids indicates the quantity of organic matter present in the sample, although its precision is not really high as ignition is not confined to organic matter. It includes losses due to decomposition or volatilization of some mineral salts.

The procedure to analyse the TSS and the VSS is the following:

1. Prepare the filters by numbering them and filtering approximately 25mL of DI water. Then put them inside a muffle at 550ºC for 20 minutes. Let them cold and weight them. The filters can be stored in a 100ºC oven.
2. Filter the sufficient quantity to have an enough solids deposition in the filter using a kitasatos.
3. Let the filters dry in an oven at 100ºC for 24h and weight them.
4. Combust the samples in a muffle furnace for 20 min and weight the filters again.

The total suspended solids and the volatile suspended solids can be computed by the following formulas:

$$TTS\ \left[\frac{mg}{L}\right] = \frac{(grams\ after\ filtration - grams\ before\ filtration) * 1000}{Filtred\ volume\ [L]} \quad \text{(Eq. 17)}$$

$$TTV\ \left[\frac{mg}{L}\right] = \frac{(grams\ after\ filtration - grams\ after\ muffla) * 1000}{Filtred\ volume\ [L]} \quad \text{(Eq. 18)}$$

6.2.5. Chemical oxygen demand (COD)

It is the amount of oxidant that reacts with a sample under controlled conditions. The oxidant consumed is expressed in terms of oxygen equivalents. Both organic and inorganic compounds are oxidized. The COD is used as a measurement of pollutants in wastewater. It can present interferences, the most important ones are those related with the chloride ion. Chloride reacts with silver ion to precipitate and thus inhibit the catalytic activity. For that reason COD has been not measured for the 3L vertical cylindrical PBR experimental reactors as chloride acid is used to control the pH in this reactors. The information given by COD can be extracted from other analysis such as TOC.

The COD methodology that has been used in the analysis is the colorimetric method. In this method most types of inorganic matter are oxidized by a boiling mixture of chromic and sulphuric acids. This result in the change of chromium from the hexavalent (VI) state to the trivalent (III) state. This species are coloured and absorb in the visible region. The $Cr_2O_7^{2-}$ absorb in the 400 nm region and the Cr^{3+} absorb in the 600nm region where the dichromate has nearly no absorption, so measuring in this longitudes can be determined the amount of chromate ion formed and thus the COD value.

$$COD = \frac{mg\ O_2 in\ final\ volume \cdot 1000}{mL\ Sample} \quad \text{(Eq. 19)}$$

The analytical method followed is the next:

1. Add 2,5 mL of sample. The sample must be filtrated if the soluble COD wants to be measured, otherwise total COD will be measured. Three triplicates will be done for each sample and three blank controls will be prepared adding 2,5 mL of DI water to the sample tubes.
2. Then add 1,5mL of potassium dichromate solution. And add 3,5 mL of sulphuric acid + Ag^+
3. Incubate the samples in a digester 2h at 150ºC.
4. Once the 2h are finished let the sample tubes cold to room temperature.
5. Clean the tubes with alcohol and introduce the blank to the COD reader.
6. Use the 800 program in the COD reader MD 600 and press zero in order to find the zero value.
7. Once the zero in registered press test in order to see which is the value of the blank. If it is lower than 8 press zero again, otherwise try with another blank control sample and repeat steps 7 and 8.
8. Once the zero in found introduce the samples and press test. 5 measurements would be done for each sample to minimize the error.

Figure 28. Tubes being incubated in the digester

6.2.6. Total, organic and inorganic carbon (TC, IC, TOC, TN)

This methodology allows to measure the quantity of organic carbon, inorganic carbon and total nitrogen in a sample. The total carbon will be just the sum of both inorganic and organic carbon. The methodology used is the high-temperature combustion method. In this method the sample is homogenized and injected to a reaction chamber packed with an oxidative catalyst where water is vaporized and all the organic and inorganic matter is oxidized to CO_2 and transported by a gas carrier and analysed by an infrared analyser. Then inorganic carbon is measured by injecting the sample into a reaction chamber where it is acidified and all inorganic carbon is converted into CO_2 and transferred to the detector. Then TOC value is computed by difference.

$$TOC = TC - IC \qquad \text{(Eq. 20)}$$

The methodology followed in order to measure TOC, IC and TN is the next:

1. Homogenize the sample with a polytron. The homogenization time will be determined by the quantity of solids in the sample. If soluble carbon should be measured this step could be avoided.
2. While the sample is in agitation fill the 500 µL syringe.
3. Open the air bottle.
4. Open the software and charge the method to TOC DIF + TN
5. Once the software give the message that it is prepared to receive the sample introduce the sample to the IC analyser.
6. Once the IC measurements have finished the program indicates that the sample can be introduced to the reactor. Introduce the sample to the reactor with the syringe and TC will be measured. When all the measurements are finished the TOC will be directly computed by difference.

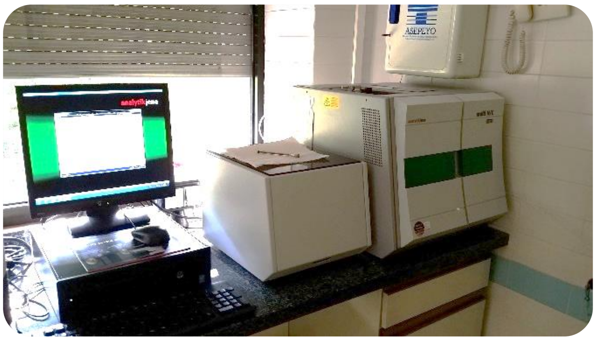


Figure 29. TOC analyser

Figure 30. Injection of the simple with a syringe

6.2.7. Alkalinity

Alkalinity is the capacity of a sample to neutralize an acid. It is taken as an indication of the concentration of carbonate, bicarbonate and hydroxide. The method used to measure alkalinity is the titration method. The hydroxyl ions present in the sample as a result of dissociation or hydrolysis of the solutes react with the acid. The procedure that must be followed is the next one:

1. Calibrate the pH meter with the standard dissolutions of pH 7 and 4,01.
2. Fill the burette with chlorhydric acid 0,02N
3. Add the acid to the sample until a pH equal to 4,5 is achieved.

With the quantity of acid titrated the alkalinity can be computed by (Eq. 21):

$$Alkalinity = mg\frac{CaCO_3}{L} = \frac{A \cdot N \cdot 50\,000}{mL\ Sample} \quad \text{(Eq. 21)}$$

Where A is the mL of standard acid titrated and N the normality of the acid.

6.2.8. Chlorophyll

Chlorophyll is a measure of the biomass of photosynthetic organism present in the culture. First of all it is needed to extract the pigments from the cells. This pigments are extracted with alkalinized acetone. The procedure that should be followed is the next one:

1. Concentrate the sample by centrifugation at 2400 rpm 10min.
2. Add 3mL of alkalinized acetone and macerate at 500 rpm for 5min
3. Add 5mL of acetone and store the sample 24h in dark.
4. Clarify the sample by centrifuging 20 min at 2000 rpm.

Once the pigments are extracted the chlorophyll concentration could be measured by a spectrophotometer. In order to determine the chlorophyll a the optical densities (OD) of 664, 647 and 630nm respectively are used. The OD of 750 nm is a correction factor for the sample turbidity. Finally, the amount of chlorophyll a could be determined by (Eq. 22):

$$Chlorophyll\ a\ \left[\frac{mg}{L}\right] = \frac{\left(11.85 \cdot (OD664) - 1.54 \cdot (OD647) - 0.08 \cdot (OD630)\right) \cdot extracted\ volume\ [mL]}{Sample\ Volume\ [mL]} \quad \text{(Eq. 22)}$$

6.2.9. Anions

Anions where determined by ion chromatography. In order to apply this methodology previous filtration is needed to remove particles larger than 0,45 µm. The measured anions were fluoride, chloride, nitrite, nitrate, bromide, orthophosphates and sulphate. The sample merges with the eluent stream and is pumped into the column where anions are separated on the basis of their affinity for the active sites of the column material.

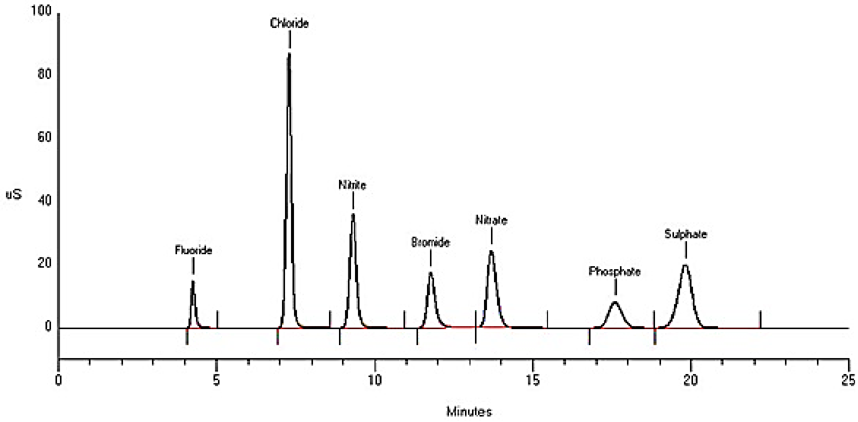


Figure 31. Typical separation of anions [66]

6.2.10. **Ammonia** [67]

Ammonia present in the sample is measured by the phenolhypochlorite method. The reagents necessary to measure ammonia through this method are summarized in the materials section. The procedure consist in adding 2ml of phenol solution 2mL nitroprussiate solution and 5mL of oxidation reagent to a 50mL previously filtered sample, mixing thoroughly after each addition. Then, wait 1 and a half hour and measure its absorbance at 640 nm. A blank should be done in order to establish the absorbance of the mixture of phenol, nitroprussiate and oxidation solution as the zero value. The concentration of ammonia can be calculated by substituting the absorbance in a previously computed calibration curve:

$$C = 1{,}195 \cdot A - 0{,}008 \qquad \textit{(Eq. 23)}$$

Where C is the concentration and A the absorbance found with the spectometer.

Figure 32. Aspect of the samples after ammonia test

6.2.11. **PHB quantification**

In order to quantify the PHB quantity present in our sample the following methodology has been followed. First of all a calibration curve is needed. To do that, it is placed 1mL of patron solution and 1mL of chloroform and heptadecane ($CHCl_3 + C_{17}H_{36}$) solution. Then the tube is mixed with a vortex for a minute. After that, 1mL of the solution is transferred to an enumerated tube. Then 1mL of $CHCl_3 + C_{17}H_{36}$ is added to the first dissolution and it is placed in the vortex for a minute. Then 1mL of that sample is transferred to a second enumerated tube. This procedure is repeated until 6 concentrations levels are obtained. Finally 1mL of methanol 20% solution is added to all the enumerated tubes. This samples are analysed with a gas chromatograph and thus the calibration curve is obtained.

Once the calibration curve is done the samples can be quantified. To do that 2mg of dried sample are weighted. Then, 1mL of $CHCl_3 + C_{17}H_{36}$ and 1mL of methanol 20% solution are added. The sample are placed in the digester for 5h at 100ºC. Once the 5h have passed the tubes are placed in ice for 15 or 30 min. Then 0,5mL of water are added and the part that is dissolved with chloroform is transferred to vials with molecular sieves in order to measure them with a gas chromatograph.

6.2.12. Glycogen quantification

Simple sugars, oligosaccharides, polysaccharides and their derivatives give an orange yellow colour when treated with phenol and concentrate sulphuric acid. Dubois has developed a method that allows measuring submicro amounts of sugars [68]. This method requires of a calibration curve. This curve is done by preparing 7 different glucose concentration solutions. Then 1mL of glucose solution is added to a test tube. After that 0,5 mL of phenol and 2,5 mL of sulphuric acid concentrate are added. Then the tubes are closed and placed in a vortex to completely mix. Then, the tubes are leave it cold for 10 min and after that they are placed in a 35ºC bath for 15min. After that the samples are measured with a spectrophotometer at 492nm.

Once the calibration curve is done the samples can be measured. First, it is necessary to extract the glycogen from the cells and to break it into glucose molecules. To do that approximately 1,5mg of sample are weighted. Then 2mL of chlorhydric acid 0,1N are added to the sample. After that the tubes are introduced in the digester for 2h at 100ºC. Once the glycogen is extracted and broken into small molecules the same procedure as in the calibration curve is followed (addition of phenol, and concentrated sulphuric acid, vortex, 10 min rest, 15min 35ºC bath and measured with spectrophotometer at 492 nm). The glycogen concentration in mg/L can be computed by (Eq. 24):

$$C = 73{,}51 \cdot A - 2{,}098 \qquad \text{(Eq. 24)}$$

Where C is the concentration and A the absorbance found with the spectrometer.

6.3. Experimental methodology

In this part the experimental procedure developed to achieve the objectives will be explained. The experimental methodology is divided in two parts the first one which talks about the control of the 30L PBR and the second one, which is the main part of the project, which explains the experiments realized with the 3L vertical cylinder reactors.

6.3.1. 30 L photobioreactor

To understand how this reactor is integrated in the wastewater treatment a brief summary of all the complete process will be done. In the next image there is a scheme of the process. First of all the water arriving from the sewage system is stored in a big tank (1a). Then this water passes by a primary decanter in order to remove the heavy solids by decantation (1b). After that the water from the primary decanter pass to the high rate algae ponds which works as a secondary wastewater treatment (2). There the algae and other microorganisms removes the suspended organic matter. Then, it is necessary to eliminate the microorganisms present in the ponds. It is done by a secondary decanter which separates the microorganisms from water. The microorganisms are used in the digester where they are degraded into biogas and a solution of minerals called digestate (3a). The water extracted from the secondary decanter is mixed with the digestate and used as feed for the 30L PBR (3b). This PBR is used as a tertiary treatment where nutrients such as nitrogen or phosphorus are removed at the same time that cyanobacteria are grown.

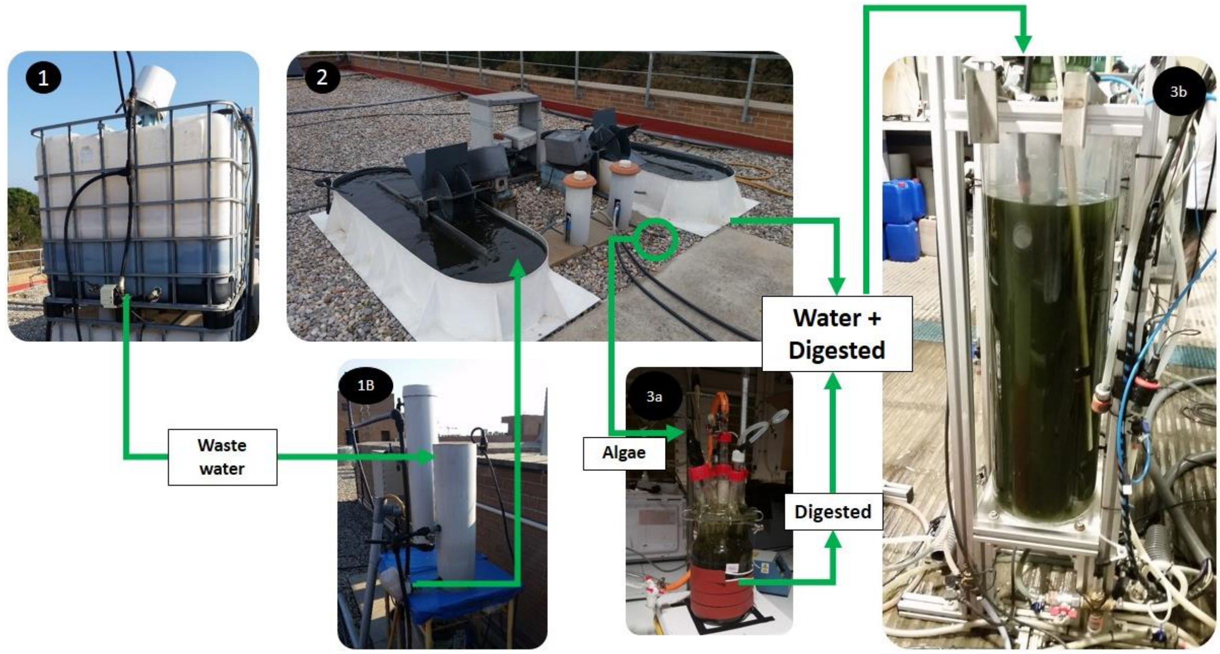


Figure 33. Process scheme

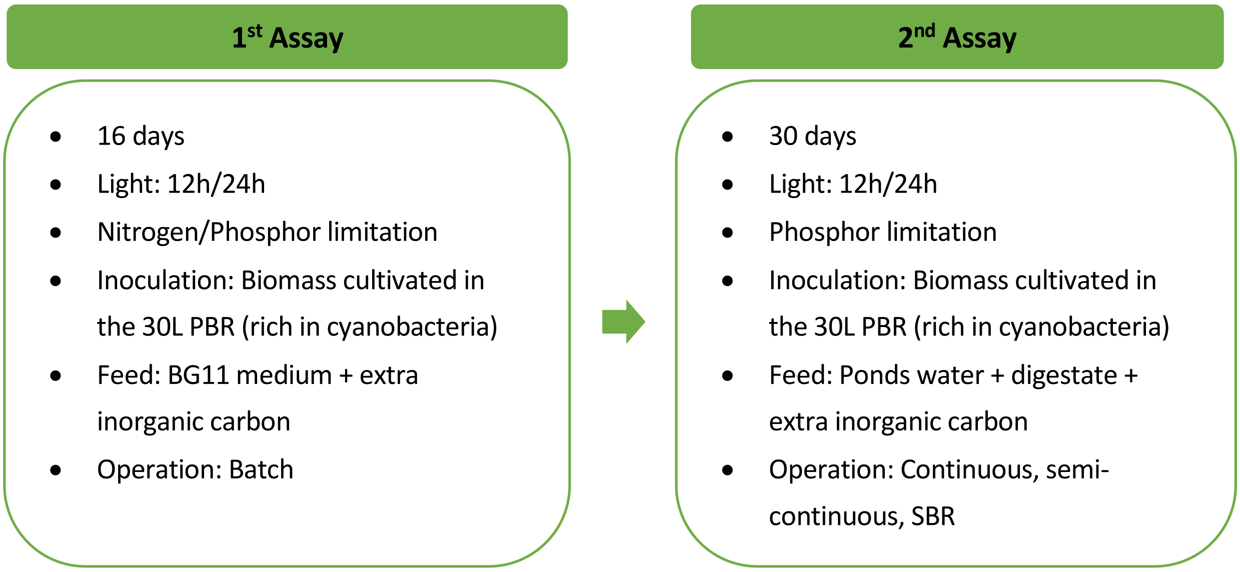
This reactor is completely monitored in order to know how it is working. To do that different analysis are planed during the week. In the following table there is a calendar with the analysis performed every day.

Table 6. Planning of the analysis

Monday	Tuesday	Wednesday	Thursday	Friday
• Turbidity • Oxygen	• Turbidity • Oxygen • Ammonia • Anions • SST/SSV • Alkalinity	• Turbidity • Oxygen • COD • TOC/TC/IC • Chlorophyll	• Turbidity • Oxygen • Ammonia • Anions • SST/SSV • Alkalinity	• Turbidity • Oxygen

6.3.2. 3L vertical cylinder reactors

This is the main experimental part of the project. As it was explained before the 3L vertical cylinder reactors have been used to accomplish two main objectives: investigate the effect of nitrogen and phosphor limitation and light hours in the production of PHB and glycogen in a mixed cyanobacteria culture and observe which reactor operation best selects cyanobacteria.



1st Assay

- 16 days
- Light: 12h/24h
- Nitrogen/Phosphor limitation
- Inoculation: Biomass cultivated in the 30L PBR (rich in cyanobacteria)
- Feed: BG11 medium + extra inorganic carbon
- Operation: Batch

2nd Assay

- 30 days
- Light: 12h/24h
- Phosphor limitation
- Inoculation: Biomass cultivated in the 30L PBR (rich in cyanobacteria)
- Feed: Ponds water + digestate + extra inorganic carbon
- Operation: Continuous, semi-continuous, SBR

Figure 34.Planning of the realized experiments

The first assay consist in two reactors one limited from nitrogen and the other limited from phosphor. This experiments last for two weeks. The experiments was done twice one with illumination for 24h a day and the other with illumination 12h a day. The culture for this assay was rich in cyanobacteria. The experiment was elongated up to 16 day due to the fact that in previous assays it has been seen that with 8 days saturation was not reached, on the contrary the tendency seems to continue to grow.

This experiments were completely monitored to understand what is happening with the system. The experimental calendar was the following one:

Table 7. Analysis Planning

Monday	Tuesday	Wednesday	Thursday	Friday
• Turbidity • Oxygen • Conductivity • TOC/TC/IC total and soluble • Anions • SST/SSV • Chlorophyll • PHA + Carbohydrates	• Turbidity • Oxygen • Conductivity • TOC/TC/IC soluble • PHA + Carbohydrates	• Turbidity • Oxygen • Conductivity • TOC/TC/IC total and soluble • Anions • SST/SSV • Chlorophyll • PHA + Carbohydrates	• Turbidity • Oxygen • Conductivity • TOC/TC/IC soluble • PHA + Carbohydrates	• Turbidity • Oxygen • Conductivity • TOC/TC/IC total and soluble • Anions • SST/SSV • Chlorophyll • PHA + Carbohydrates

Finally, a second experiment has been done in order to select the major quantity of cyanobacteria to maximize the PHA productivity in our culture. In the previous results it has been seen that phosphor limitation allows to achieve higher cyanobacteria content. This reactor where fed with water from the ponds and digestate with a relation water/digestate 100:6 mL in order to connect the whole process. The ponds water is rich in nitrogen and its phosphor content is low allowing to have a phosphorous content lower than 2mg/L and a N to P ratio of approximately 10-12. Nevertheless, water from ponds was diluted by half to avoid nutrients accumulation. To study the best way to select cyanobacteria 4 types of operation has been tried:

- Continuous reactor: hydraulic and solids retention time of 10 days.
- Semi-Continuous reactor: hydraulic and solids retention time of 10 days.
- SBR1: hydraulic retention time of 2 days and a solid retention time of 10 days.
- SBR2: hydraulic retention time of 2 days and a solid retention time of 5 days.

The SBR are expected to have the capacity of eliminating *scenedesmus sp.* (green algae) present in the inoculated biomass, as this kind of reactor operation selects the microorganisms that precipitate more easily. Cyanobacteria are known to be quickly settled while *scenedesmus* remains in suspension.

In Table 8 there are the week analysis done for this reactors:

Table 8. Analysis planning

Monday	Tuesday	Wednesday	Thursday	Friday
• Turbidity • Oxygen • Anions • SST/SSV • Chlorophyll	• Turbidity • Oxygen • TOC/TC/IC/TN soluble+total • PHA + Carbohydrates	• Turbidity • Oxygen • Anions • SST/SSV • Chlorophyll	• Turbidity • Oxygen • TOC/TC/IC/TN soluble+total • PHA + Carbohydrates	• Turbidity • Oxygen • Anions • SST/SSV

7. Results and discussion

7.1. 30 L Photobioreactor

Cyanobacteria have the capacity of transform contaminants present in wastewater, such as nitrogen or phosphor, and use them for its own benefit. The aim of this experiment is to observe cyanobacteria growth and its capacity to remove contaminants from ponds. For that reason, as the reactor works approximately always in the same way, a mean was computed with the values collected during 135 days.

In Table 9 there are the mean values of the contaminants presents in the effluent from the ponds (secondary treatment), the solution used to feed the PBR and the nutrients inside the PBR, as well as the 95% confidence interval associated with these values.

Table 9. Nutrients present in the pond effluent, reactor feed and PBR

Contaminants	Pond		Reactor feed		PBR	
	Mean	95% CI	Mean	95% CI	Mean	95% CI
$N\text{-}NH_4^+$	**0,192**	0,137	**7,556**	1,639	**0,285**	0,187
$N\text{-}NO_2^-$	**1,167**	0,513	**0,947**	0,343	**0,658**	0,324
$N\text{-}NO_3^-$	**17,215**	4,630	**17,151**	3,516	**8,113**	3,315
$P\text{-}PO_4^{3-}$	**1,528**	0,361	**1,506**	0,351	**0,444**	0,322
IC	**26,084**	4,795	**25,707**	2,700	**20,929**	4,795
TIN	**14,791**	7,277	**26,858**	4,436	**9,456**	4,098
N/P	**9,681**	--	**17,838**	--	**21,294**	--

In order to analyse in which cases there are significant differences between the nutrients present in each of the process stages a one-way Anova statistical analysis has been done using the software *GraphPad Prisma6*. One-way anova is a statistical method used to compare the mean of three or more samples. It tests the null hypothesis, which says that no variation exists between variables. This hypothesis is presumed to be true until statistical evidences nullifies it for the alternative hypothesis. The one-way anova allows to compute the p-value which indicates the evidence against the null hypothesis. A high value of p indicates a strong evidence of the null hypothesis, which means no statistical differences, a value of $p \leq 0,05$ means low significance (*), $p \leq 0,01$ medium significance (**), $p \leq 0,001$ high significance (***), $p < 0,001$ indicates really high significances (****).

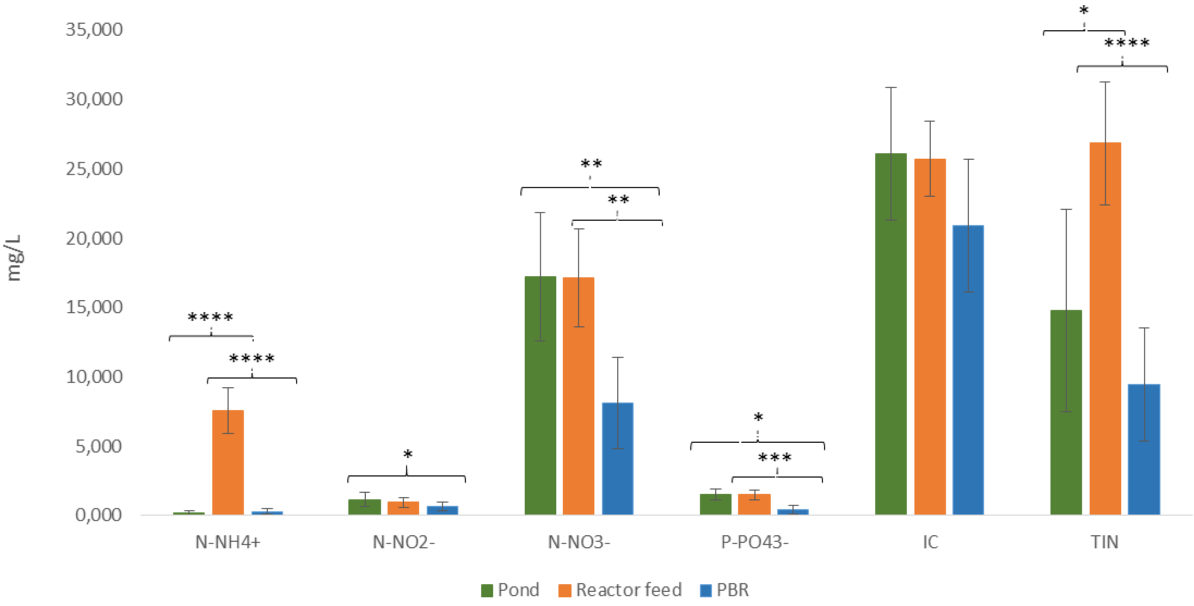


*Figure 35. Nutrients present in the pond effluent, reactor feed and PBR. Indication of the statistical difference between bars. * p≤0.05 low significance, ** p≤0.01 medium significance, *** p≤0.001 high significance, **** p<0.001 indicates really high significances.*

Figure 35 shows the concentration of the water contaminants present in the different process stages. In the case of ammonia, we can see that there are really significant differences between the reactor feed and the other two. So, the microorganisms present in the PBR are capable of eliminate the high ammonia concentrations present in the digestate. In the case of $N\text{-}NO_2^-$ there are small significant differences between the ponds and the PBR, therefore not a high removal % of nitrites may be expected. Talking about nitrates, we can see that there are significant differences between the pond effluent, the reactor feed and the PBR. This means that PBR allows the removal of nitrates. Nitrogen can easily change from ammonia to nitrite or nitrate and vice versa. For this reason it has been also studied the total inorganic nitrogen (TIN), which is the sum of the total amoniacal nitrogen (TAN), the $N\text{-}NO_2^-$ and the $N\text{-}NO_3^-$. It can be observed that there are really significant differences between the concentration of TIN in the feed and the concentration in the PBR. This demonstrates the microorganisms present in the PBR have a high capacity to remove inorganic nitrogen. Significant differences are also found with $P\text{-}PO_4^{3-}$, so phosphor elimination is also possible with that system. For inorganic carbon there are no significant differences, but it should be remembered that this reactor is connected to a CO_2 gas bottle that regulates the pH by CO_2 injection, so an extra source of inorganic carbon is present that is not included in the calculation of the reactor feed.

Finally the degree of elimination of nutrients can be computed by (Eq. 25).

$$Contaminatn\ elimination\ [\%] = \frac{C_{feed}[\frac{mg}{L}] - C_{PBR}[\frac{mg}{L}]}{C_{feed}[\frac{mg}{L}]} \cdot 100 \qquad \text{(Eq. 25)}$$

In Table 10 there are the obtained efficiencies. High elimination efficiencies are achieved by using this system as a tertiary treatment, especially in the case of ammoniacal nitrogen and phosphor.

Table 10. Water treatment efficiencies

Water treatment efficiencies	
N-NH_4^+	96,2%
N-NO_2^-	30,5%
N-NO_3^-	52,7%
P-PO_4^{3-}	70,5%
TIN	64,8%

These process will be applicable in the urban wastewater treatment, if the obtained values meet the European directive 91/271/EEC [69]. In Table 11 there is a comparison between the obtained values and the limit values of Directive 91/271/EEC. The TSS are much higher than the limit ones. So, to apply this process as a substitute to the conventional wastewater treatment an extra process stage, such as decantation, is necessary to eliminate the suspended biomass. Table 11 shows that the soluble values are all below the limit of discharge. Therefore, if biomass is eliminated the resulting effluent will obey the law. Furthermore, the eliminated biomass is an interesting by-product for the production of biofuels and bioplastics.

Table 11. Comparison of the results with directive 91/271/EEC [69]. In red there are the values over the limit from the directive. In green the values below the limit values of the directive

	Directive 91/171/CEE	**Total**	**Soluble**
DQO [mg O2/L]	**125**	--	61,36
TSS [mg/L]	**35**	450	--
Total Phopsphor [mg/L]	**2**	--	0,444
Total Nitrogen [mg/L]	**15**	31,4	9,45

Not only contaminants have been removed but also cyanobacteria have been grown in the PBR. It has been found that the amount of volatile suspended solids is 0,369 ± 0,118 g/L and that there is a chlorophyll amount of 1,328 ± 0,333 mg/L which indicate the presence of photosynthetic microorganisms (microalgae and cyanobacteria). Figure 36 shows that in November the microorganisms present in the photobioreactor are mainly green algae of *Scenedesmus* sp. (a). Some cyanobacterias are observed but in small quantities (b). In January the presence of cyanobacteria becomes higher. The specie that dominates is *Aphanocapsa* sp. These cyanobacteria have been found either in completely isolated flocs (b) or in flocs with a high presence of green algae (c). Some other cyanobacteria species have been found such as *Pseudanabaena* sp. although they are present in a much lower quantity.

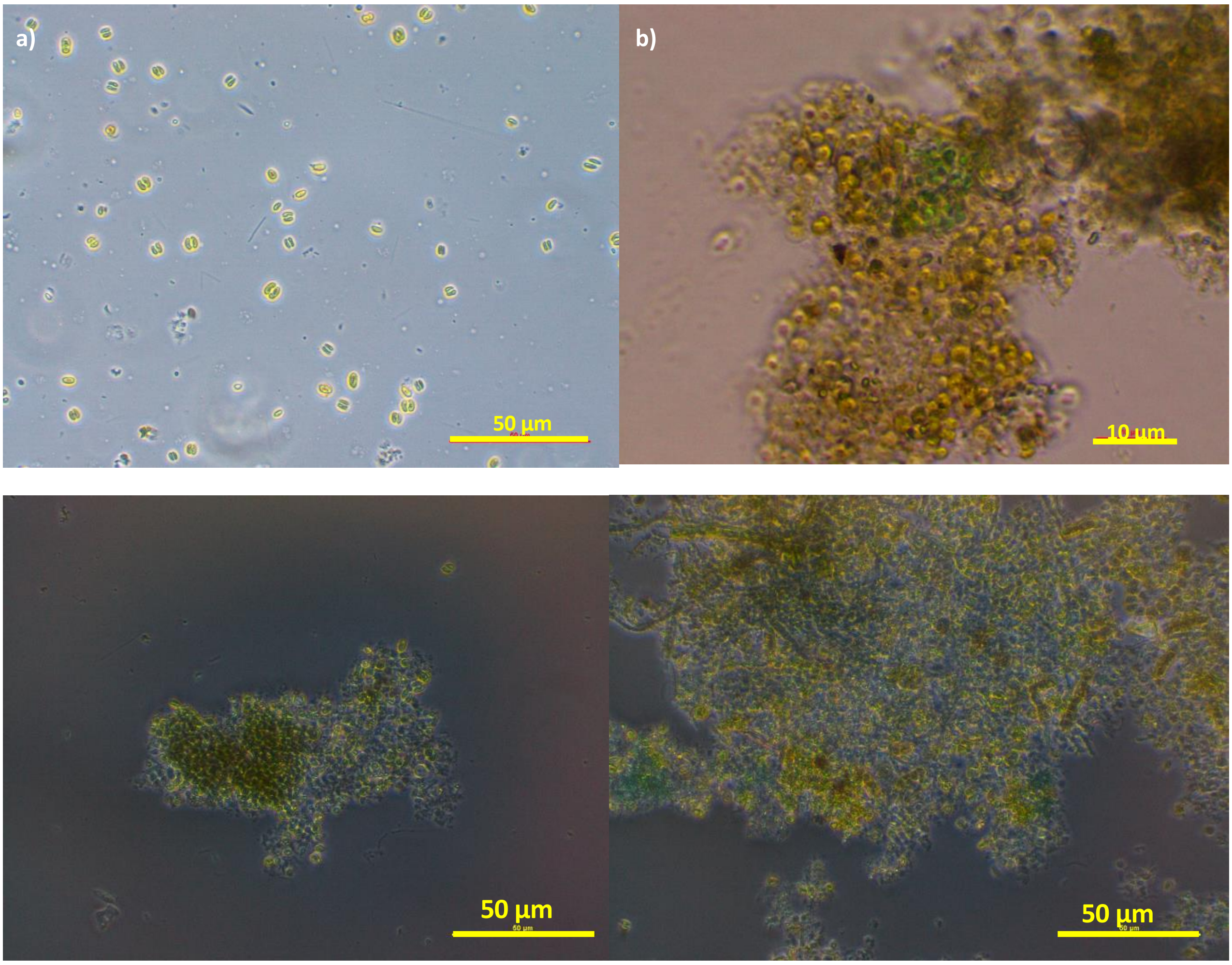


Figure 36. Microscopy images from the 30L PBR culture A) and B) pictures are from November and c) and d) from January. A) Isolated cells from scenedesmus sp. Observed in phase contrast microscopy (x400). B) Floc mainly dominated by green algae. Observed in bright light microscopy (x1000). B) Floc completely dominated by cyanobacteria from Aphanocapara sp. observed in phase contrast microscopy (400x). c) Algal floc with a high presence of cyanobacteria from Aphanocapsa sp with some Pseudanabaena sp. Observed in phase contrast microscopy (400x).

Our group, in a previous study done by *DM. Arias et al.* [3] observed the effect of nitrogen, phosphor and inorganic carbon charges on the cyanobacteria dominance in a mixed culture of cyanobacteria and microalgae. In Table 12 the results obtained in this project are compared with the ones obtained in [3].

Table 12. Comparison between the main quality parameters of [3] during the three experimental periods and *the ones obtained in this project*

	Period 1	Period 2	Period 3	This project
pH	8,1	8,4	8,6	8,01
DO [mg/L]	8,8	6,8	6,4	8,68
TSS [g/L]	0,52	1,05	0,49	0,45
VSS [g/L]	0,41	0,84	0,39	0,37
Biomass production [g VSS/d·L]	0,041	0,084	0,039	0,037
Chlorophyll a [mg/L]	3,9	4,5	3,7	1,4
Alkalinity [mg $CaCO_3$/L]	92,5	112,3	224,4	185,9
Total ammoniacal nitrogen (TAN) [mg/L]	0,3	0,3	0,16	0,29
N-NO_2^- [mg/L]	0,41	0,09	0,06	0,66
N-NO_3^- [mg/L]	31,18	12,49	3,95	8,11
Total inorganic nitrogen (TIN) [mg/L]	31,94	12,88	4,12	9,46
P-PO_4^{3-} [mg/L]	1,36	0,9	0,2	0,44
TAN removal	>95%	<96%	>95%	96%
N-NO_3^- removal	--	--	91%	52%
P-PO_4^{3-} removal	44%	47%	>95%	70%
Lv-P [mg P/L·d]	0,28	0,23	0,16	0,15
Influent N:P ratio	13	10	18	18

The volumetric phosphor load and the influent N:P ratio obtained in this project approximate the ones of period 3. However, in this case the P-PO_4^{3-} and the TIN present in the PBR are higher. This is caused by a low removal % of either nitrates or phosphates compared to period 3. A really low concentration of chlorophyll "a" is present in this project compared to any of the periods. That low chlorophyll content may explain the low removal % as there are less photosynthetic microorganisms than in the other cases. Although the phosphor content in this project is higher than in period 3 the biomass production is similar, but the quantity of cyanobacteria present is more like the one from period 1. Nevertheless, it seems that the culture is changing towards cyanobacteria domination. So, if feed conditions are maintained period 3 will be likely achieved. It should be also taken into consideration that the initial culture is mainly dominated by *scenedesmus* sp. this microalgae is a high competitor of cyanobacteria even when low phosphor charges are applied. For that reason it is interesting to study other ways to select cyanobacteria, as done in 7.2.2.

As a conclusion, it can be said that this kind of system can be used as a tertiary wastewater treatment as they are capable of eliminate 65% of the total nitrogen and a 70% of the phosphor. However, decantation of the biomass is necessary for this process to be used as substitute of conventional wastewater treatment. At the same time, it have been seen that cyanobacteria use these contaminants as nutrients and proliferate. The major problem is that there are many microorganisms that compete with cyanobacteria. It have been seen that low levels of phosphor enhance the cyanobacteria culture doninance. However, *scenedesmus* sp are high competitors even in low phosphor levels. For that reason, it is interesting to study other techniques that may enhance the selection of cyanobacteria in front of other microorganisms.

7.2. 3L Vertical cylinder reactors

These section include the main project results. The first part shows the results of the experiments done in order to find the best nutrients and light conditions to accumulate PHB and carbohydrates. In the second part there are the results of applying different reactor operations to observe which one causes a higher cyanobacteria selection.

7.2.1. Optimal nutrients and light conditions for PHB and carbohydrates accumulation

To understand which conditions favour the PHB and carbohydrates accumulation it is important to observe how the culture changes with time. For that reason different analysis have been done as explained in 6.3.2. In order to make the results interpretation easier a colour code has been established for all the graphics and tables:

Table 13. Colour code legend

12h light	P-limitation	🟧	🟠
	N-limitation	🟩	🟢
24h light	P-limitation	🟧	▲
	N-limitation	🟩	▲

Dissolved oxygen (DO) is an indicator that photosynthetic microorganisms are present in the culture. Dissolved oxygen measurements were done daily during the 15 experimental days. In the case of 12 h light culture it was measured after the dark phase. Table 14 show the mean dissolved oxygen values obtained after 15 experimental days.

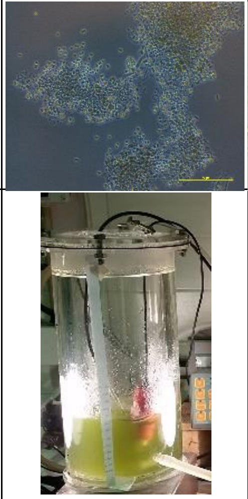
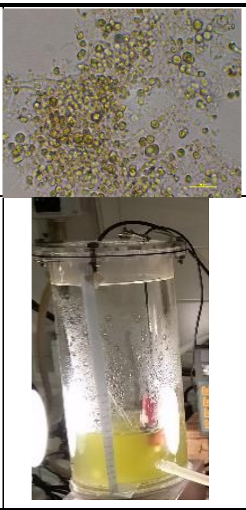
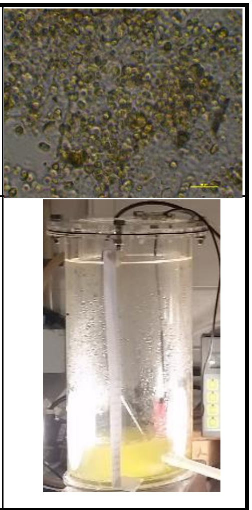
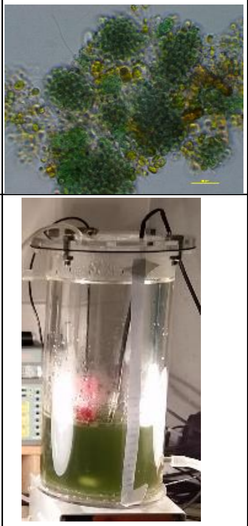
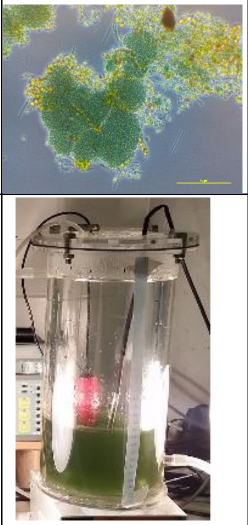
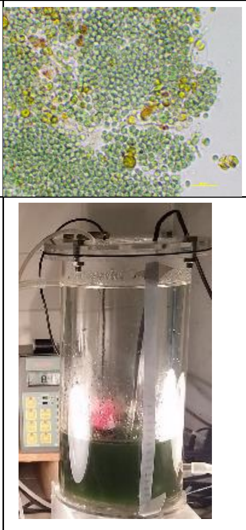
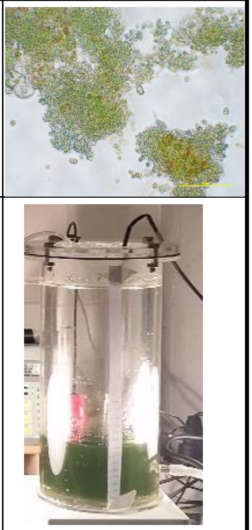
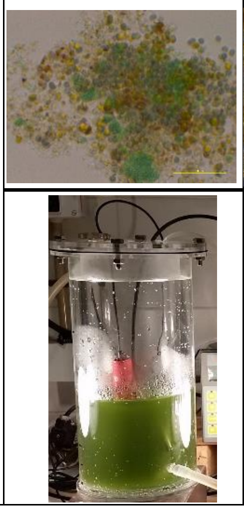
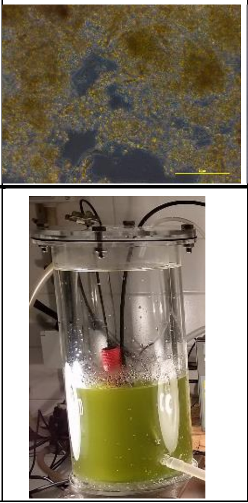
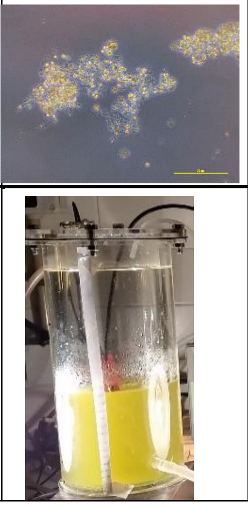
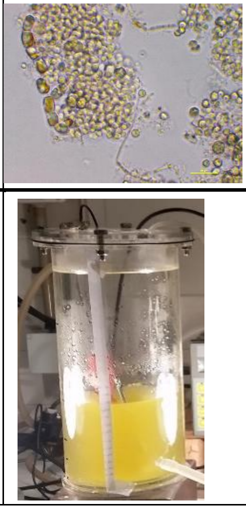
Table 14. Dissolved oxygen results

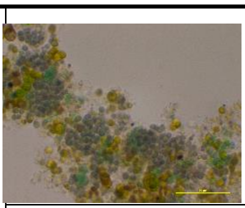
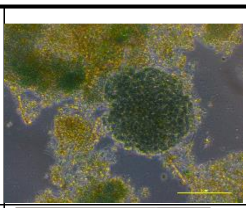
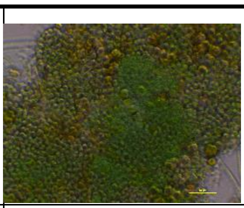
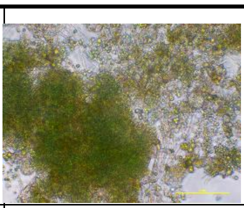
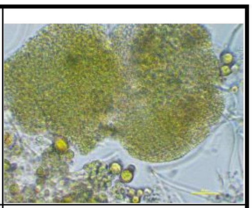
12h light		24h light	
N-limited	**P-limited**	**N-limited**	**P-limited**
6,18 ± 0,28 mg/L	6,56 ± 0,27 mg/L	7,07 ± 0,71 mg/L	7,60 ± 1,09 mg/L

From Table 14 it is observed that 24h light culture have a higher DO concentrations. However, these results are not conclusive as oxygen was not continually monitored, on the contrary in the 12h light culture, oxygen was measured after the dark phase were the oxygen concentration decreases due to the fact that photosynthetic activity is almost stopped while cellular respiration remains constant increasing thus the DO consumption/DO production ratio. Some measurements has been done at the light phase getting values up to 8 mg/L.

To observe how the culture population is changing microscopic observations and reactor pictures have been done. Table 15 shows that in all the cases the initial culture is formed by a mixed algae and cyanobacteria population. Cyanobacteria can be distinguished from green algae for its blue greener colour. In this case all the cyanobacteria found in the four cultures were mainly composed by *Aphanocapsa* sp.. Both 12h and 24h light cultures show that when the culture is limited from nitrogen the cyanobacteria and green algae loss its pigments due to chlorosis. That is why the culture turns to yellow. In the case of 24h light cultures it can be seen that this changes occur quicker than in the case of the 12h light culture. In day 4 the cellules from the N-limited 24 h light culture have already lost its pigments while in the case of N-limited 12h light that does not occur until day 12. That can be explained by a metabolism change during the dark hours. Cyanobacteria perform photosynthesis and respiration at the same time [70]. However, when no light is present the photosynthesis is stopped. Thus, the NADPH and ATP needed to maintain the cell alive, is synthesized almost only by the catabolism and cell respiration. Not enough energy and reducing power is present for the cell to reproduce. So the growth is reduced during the darkness hours and thus culture depigmentation is slower. In the case of the phosphorous limited cultures the cyanobacteria population increases until a point in which nitrogen quantity is really low.

Table 15. Microscopy and reactor pictures

		Day 1	Day 4	Day 9	Day 12	Day 15
12h light	**N-limited**					
	P-limited					
24h light	**N-limited**					

P-limited					
					

It is not enough with microscopic observations to see how the culture is changing. Different analysis has been done to see the culture growth.

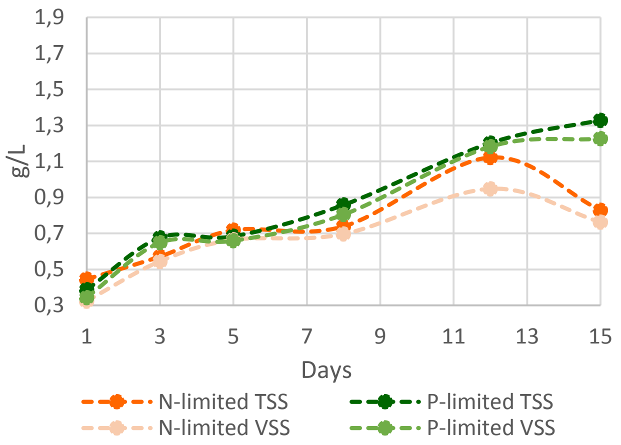


Figure 37. TSS and VSS for 24h light cultures

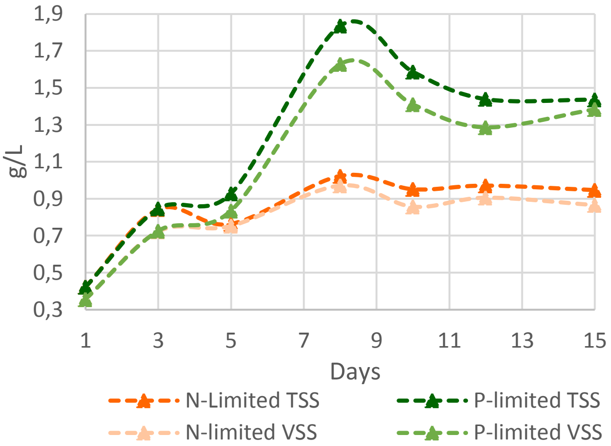


Figure 38. TSS and VSS for 24h light cultures

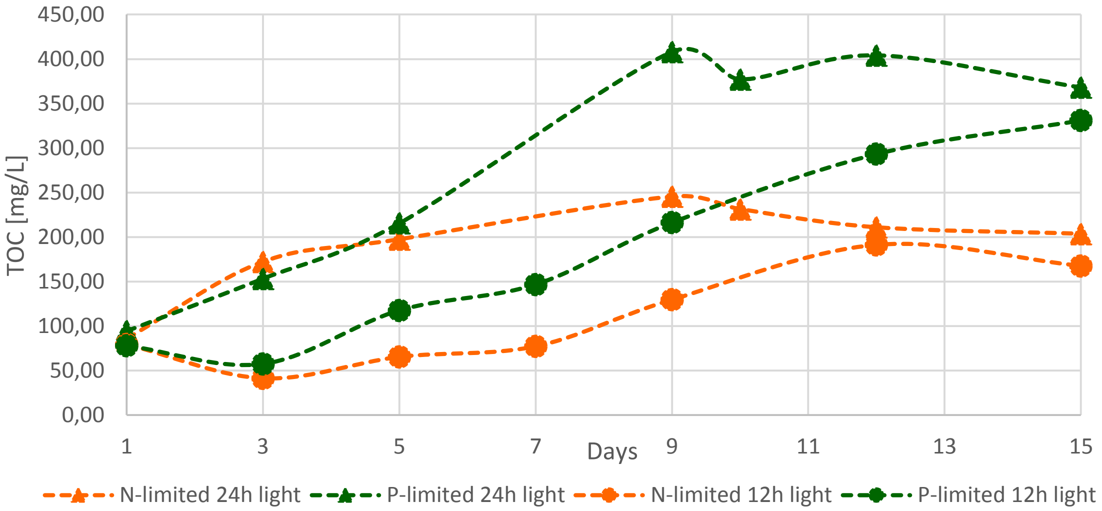


Figure 39. Total organic carbon (TOC)

In Figure 37 and 38 there are the results of the total and volatile suspended solids and in Figure 39 the ones for the total organic carbon. As there is no soluble organic matter, an increase in TOC indicates that microalgae and cyanobacteria are growing. Those results must be compared with the volatile suspended solids as they are the ones corresponding to organic matter. It can be seen that either TOC or VSS analysis in the 24h light cultures the growth is stabilized from day 9. On the contrary in the 12h light cultures the N-limited achieves steady state at day 12 and P-limited culture maintain its growth all the experimental time. Those differences between 12h and 24h light correspond to the metabolic changes during the darkness hours as previously explained.

An increase in either TOC or VSS could mean an increase on the quantity of cells in the culture or just that the cells are accumulating components. For that reason, chlorophyll measurements can be used to make a prediction of which of this two options is happening. If chlorophyll remains stable but VSS or TOC increase will mean that some components are been accumulated inside the cells. If both chlorophyll, VSS and TOC increases will mean an increment of the photosynthetic microorganisms population which may also cause an increase on the PHB or carbohydrates concentration. If chlorophyll decreases but VSS remains constant will mean a loss in the cellular pigments. This process is known as nitrogen chlorosis and has been demonstrated that has not to be accompanied by the loss of cell viability, it is just an acclimation process that enable the cells to survive prolonged periods without nitrogen. Chlorophyll “a” contains nitrogen that can be used as an intracellular nitrogen source. Furthermore, the protein anabolism is stopped due to a lack of the nitrogen necessary for the amino acid synthesis. For that reason cells stays in a dormant-like state until an addition of nitrogen is done [71], [72]. Finally, if both chlorophyll and VSS decreases it will mean that there is cellular death.

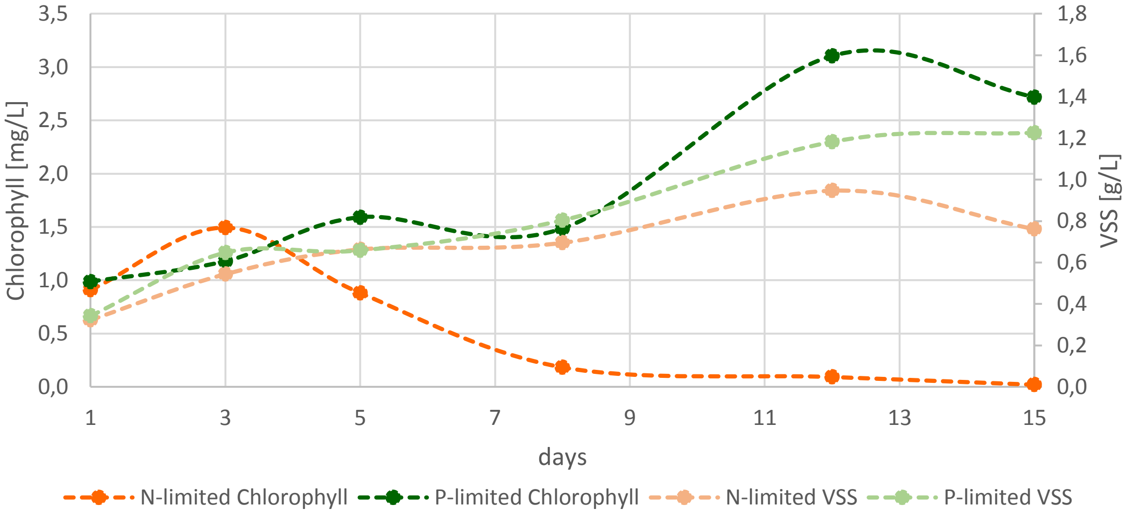


Figure 40. Chlorophyll and VSS 12h light cultures

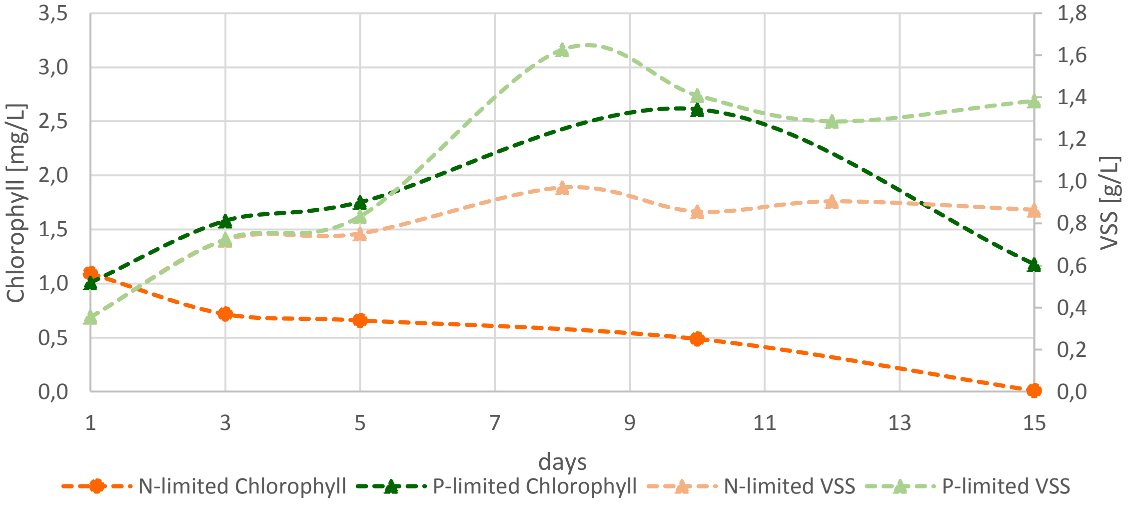


Figure 41. Chlorophyll and VSS 24h light culture

In Figures 40 and 41 it can be seen the chlorophyll "a" concentration compared to the volatile suspended solids. In the P-limited cultures both chlorophyll and VSS follows an increasing tendency. That means that the photosynthetic microorganisms present in the culture are growing. The 24h light culture reaches saturation earlier than the 12h light culture. On the contrary, the chlorophyll in the N-limited cultures drastically decreases up to almost 0 mg/L levels while VSS remain more or less stable. That confirms that in both cases chlorosis is taking place due to the lack of nitrogen.

A really important nutrient either for the PHB and carbohydrates accumulation is inorganic carbon. It is necessary for the PHB and glycogen synthesis that inorganic carbon is never limited. For that reason it is very important to control and add inorganic carbon whenever it is necessary.

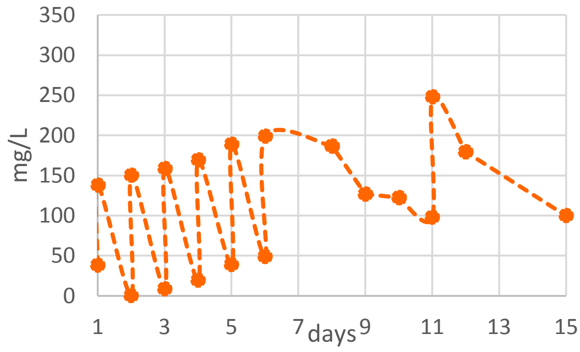


Figure 42. Inorganic carbon concentration N-limited culture 12h light

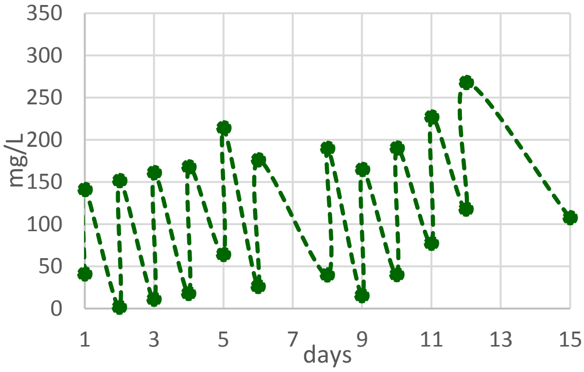


Figure 43. Inorganic carbon concentration P-limited culture 12h light

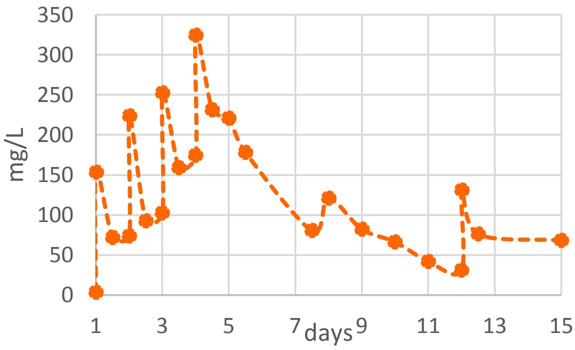


Figure 44. Inorganic carbon concentration N-limited culture 24h light

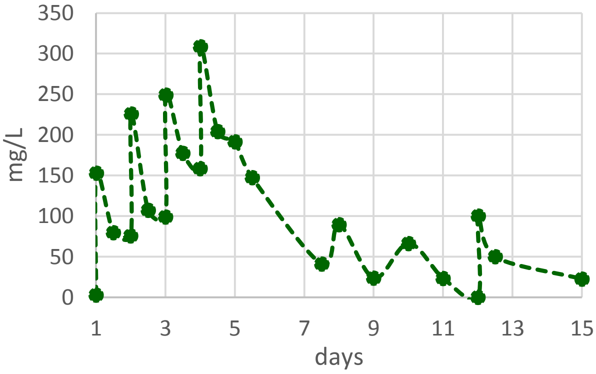


Figure 45. Inorganic carbon concentration N-limited culture 24h light

The peacks observed in the graphs from figures 42, 43, 44 and 45 correspond to an addition of bicarbonate. This additions has been done when a low inorganic carbon level was detected. In the case of 12 h light cultures high inorganic carbon consumption was observed until day 5. It can also be observed that there is some flat parts between the picks. Those parts correspond to the darkness hours when photosynthesys is almost stopped so no carbon is fixed. In 24h light cultures the consumption rate is higher, although in the N-limited culture the consumption rate also decreases at day 7. The higher consumption of inorganic carbon by the 24h light cultures is due to the fact that they are all the time fixing CO_2 in the Calvin cycle due to the continue presence of light.

Finally, after seeing how the culture parameters have evolved during the experiment it is time to see which conditions allows accumulating more PHB and carbohydrates.

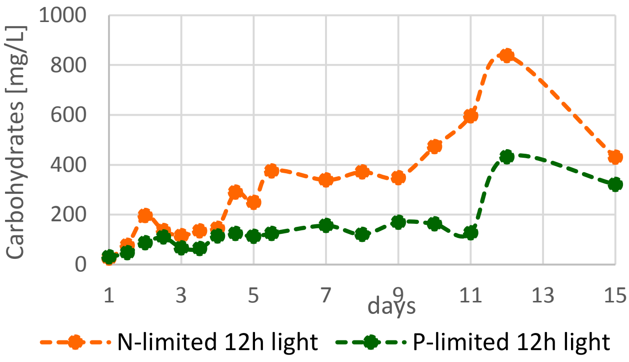


Figure 46. Carbohydrates concentrations 12h light experiments

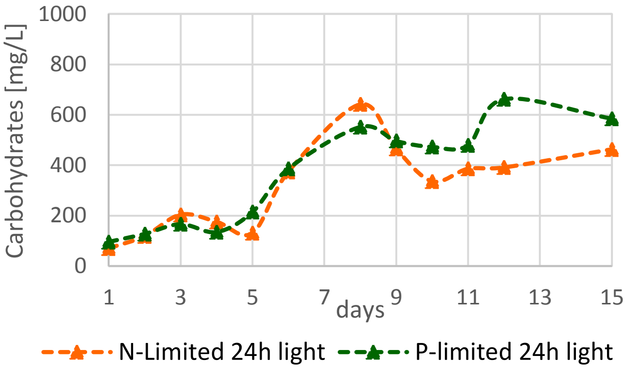


Figure 47. Carbohydrates concentrations 24h light experiments

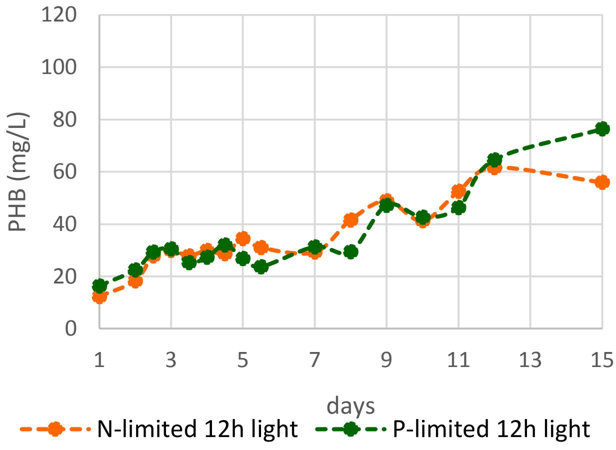


Figure 48. PHBs concentrations 12h light experiments

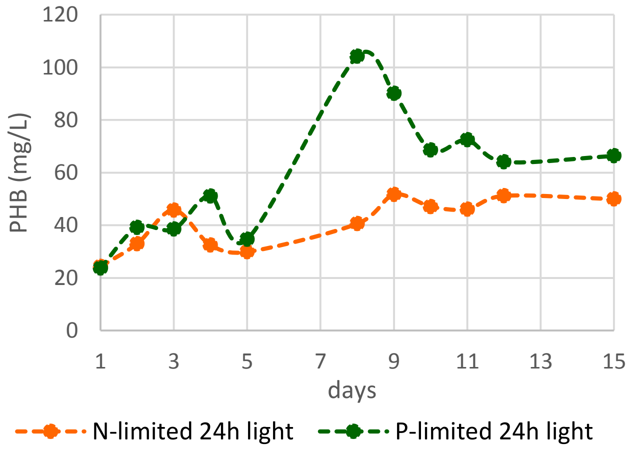


Figure 49. PHBs concentrations 24h light experiments

Table 16. Maximum carbohydrates and PHBs concentration levels

	12h light		24h light	
	N-limited	P-limited	N-limited	P-limited
Max Carbohydrates concentration [mg/L]	**838,056**	432,131	641,310	662,382
Max PHBs Concentration [mg/L]	61,614	76,361	51,635	**104,234**

In the case of carbohydrates a maximum is achived at day 12 with 12h ligh cultures and at day 8 in the case of 24h light cultures. In the case of PHBs in the 12 h cultures a maximum is not reached, so probably a highest concentration of PHBs can be achived if the experiment is maintained for longer. In the 24h light culture a maximum has been reached at day 8, after that day the PHB levels become stable. In the case of carbohydrates the culture conditions that get a highest accumulation levels are 12h light and nitrogen limitation. For PHBs 24h light and phosphorous limitation are the conditions that get highest accumulation levels. It can be said that the nitorgen limitation seems to enhance the carbohydrates acumulation more than phosphor limitation. However, phosphor limitation seems to enhance the PHBs accumulation. The role of PHBs in cyanobacteria is not really clear as these microorganisms lacks of a complete tricarboxylic acid cycle, so depolymerization of PHB can play only a minor role for energy production [16]. As explained in 3.2 different metabolic explanations has been found for why either nitrogen or phosphorous limitation conditions can enhance PHBs accumulation. It seems that the effect of nitrogen and phosphor is specie dependent. In this case it seems that the decreas in ATP due to the phosphor limitation and the surplus of reducing power generated has a highest effect on the PHB accumulation that the inhibition of the aminoacid synthesis due to nitrogen stravation. However, the inhibition of aminoacid synthesis seems to be more determinant for the carbohydrate accumulation.

Finally, in order to choose the best working conditions some key performance indicators (KPI) has been defined. A key performance indicators are a set of quantifiable measures that indicates the performance over time of a process. The defined KPIs are the following ones:

- **Environmental index:** In this case inorganic carbon has been added in the form of carbonate because there was not a CO_2 injection system available. However, it must be remembered that CO_2 can be used instead of carbonate as an inorganic carbon source and instead of chlorhydric acid in order to control the pH. For that reason it is important to know how much inorganic carbon (CO_2) is consumed. With that index it can be approximately seen the capacity of each system to convert CO_2 into other interesting cell constituents and thus it can be observed how good the system conditions are as a carbon capture and storage (CCS) technology. The environmental index can be computed as follows:

$$\text{Environmental index [\%]} = \frac{\text{Inorganic carbon added } \left[\frac{\text{mg}}{\text{L}}\right]}{\text{Final conc PHB } \left[\frac{\text{mg}}{\text{L}}\right] + \text{Final Conc Carbohydrates } \left[\frac{\text{mg}}{\text{L}}\right]} \cdot 100 \qquad \textit{Eq. 26}$$

- **Economic index**: it is impossible to have a culture of 24h using natural light, so it will be necessary to spent some energy to illuminate the culture at least 12 hours per day. In order to see If the money wasted in illumination is economically reasonable this economic index was used. The economic index considers the benefits of selling the PHB and biodiesel produced without taking into account the extraction and biorefinery costs as they will be the same for all the culture conditions. The economic index can be calculed with the following equation:

$$\begin{aligned}\text{Economic index}\left[\frac{\text{€}}{\text{L}}\right] &= \text{conc PHB}\left[\frac{\text{kg}}{\text{L}}\right]\cdot\text{Price PHB}\left[\frac{\text{€}}{\text{kg}}\right]+\text{Conc carbohydrates}\left[\frac{\text{kg}}{\text{L}}\right]\\ &\cdot\text{Price carbohydrate}\left[\frac{\text{€}}{\text{L}}\right]-60\text{W}\cdot 2\cdot\text{illumination hours }[\text{h}]\\ &\cdot\text{Price energy}\left[\frac{\text{€}}{\text{kWh}}\right]\end{aligned} \qquad \textit{(Eq. 27)}$$

The concentration of either PHB or carbohydrates refers to the final concentration minus the initial concentration. This is done in order to put all the concentration with the same base reference. As if you start from a higher base it is really probable that you get a higher final concentration.

The considered prices are the following ones:

Table 17. Considered prices for the economic index

Bioethanol price	0,41 €/kg	http://bit.ly/2p7cRF0
PHA price	5,00 €/kg	http://bit.ly/2q24Le2
Energetic costs	0,10 €/kWh	http://bit.ly/1CK8QD2

Finally, the values get for the indicators are summarized in Table 18:

Table 18. KPI results

	12h light		24h light	
	N-limited	P-limited	N-limited	P-limited
Environmental index [%]	144%	214%	195%	223%
Economic index [€/L]	$3{,}839\cdot10^{-4}$	$4{,}194\cdot10^{-4}$	-2,225	-2,224

The culture conditions that allows a highest carbon fixation is the 24h light limited from phosphor. It also seem that the P-limited cultures are more efficient in the carbon fixation and conversion into profitable components. Looking the economic index it can be clearly seen that expending extra money in illumination does not compensate the benefits generated by selling the components. The culture conditions that generate more benefits is the 12h light limited from phosphor.

Finally, after analysing all the obtained results and taking into account the KPIs. It can be said that in this case the optimal cultivation conditions found are 12h light with phosphor limitation.

Although the 24h light P-limited culture has a highest carbon conversion efficiency, the value from the culture with 12h light is really similar. Furthermore, using just 12h illumination makes this type of culture suitable for external cultivation, making sun the sole energetic source necessary. Even though the N-limited culture is capable of accumulating more carbohydrates the economic benefit that generates the PHB selling is higher than that from carbohydrates, so at the end it is preferable that PHBs concentration to be as high as possible.

7.2.2. Selection of cyanobacteria

In the used system there is a huge competence between different microorganisms born in wastewater, such as cyanobacteria, green algae or protozoa. To produce more PHB it is interesting to have a culture dominated by cyanobacteria, as they are the unique ones capable of accumulating PHB in such conditions. For that reason 4 reactor operation have been set in order to best select cyanobacteria. The reactor operation tried are presented in Table 19:

Table 19. Colour code and characteristics of the different analysis done

Continuous	HRT: 10 days SRT: 10 days	🟦
Semi-Continuous	HRT: 10 days SRT: 10 days	🟩
Sequencing batch reactor 1 (SBR1)	HRT: 2 days SRT: 10 days	🟧
Sequencing batch reactor (SBR 2)	HRT: 2 days SRT: 5 days	🟥

In the previous experiment it was observed that 12h light culture with phosphorous limitation are the preferable cultivation conditions. The effluent of the high rate algae ponds is rich in nitrogen and scarce from phosphorous. So small phosphor charges could be achived and at the same time the production of PHB and carbohydrates can be joined with the wastewater treatment.

In this experiment daily microscopic observations have been done in order to observe which of the systems have more cyanobacteria. In Table 20 there are some relevant microscopic images done during the experiment. Two types of microscopes have been used in this experiment. The optical microcop and the fluorescence microscop. The fluorescence is used to detect cyanobacteria as they have phycocyanin pigment which emites fluorescence when iluminated at 650 nm. In Table 20 there are the pictures of the microscopic observations. The black and white images correspond to fluorescence microscope.

Table 20. Microscopic observations

Day	1	2	3	6	7	8	10	15
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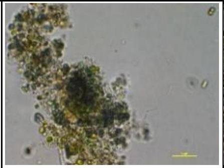
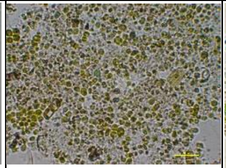
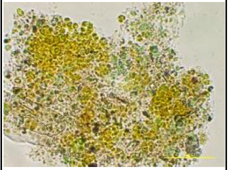
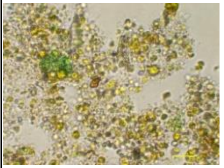
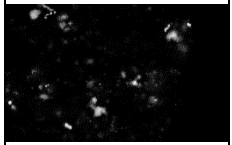
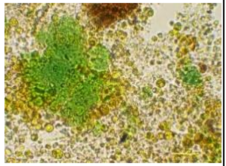
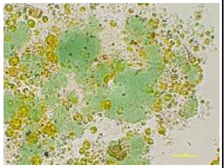
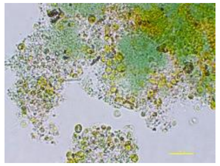
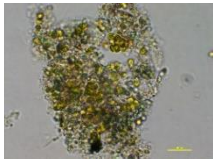
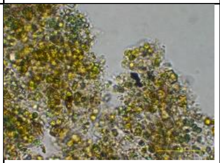
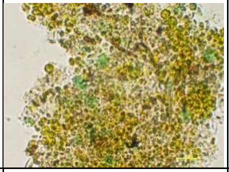
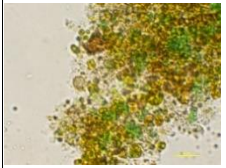
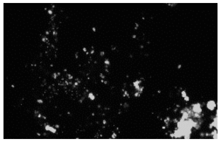
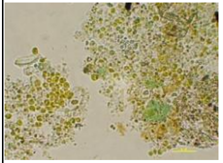
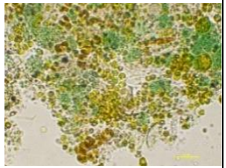
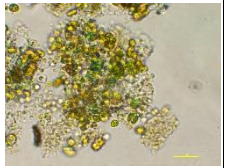
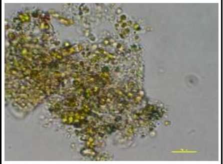
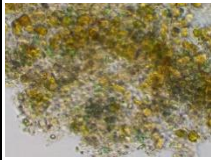
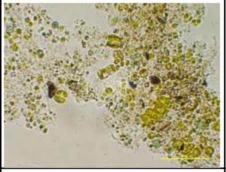
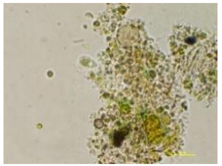
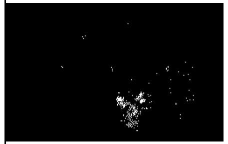
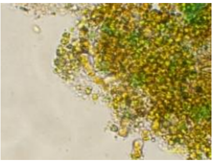
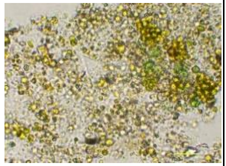
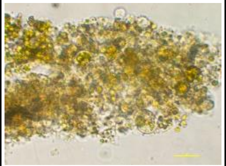
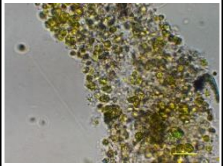
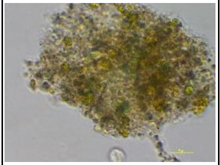
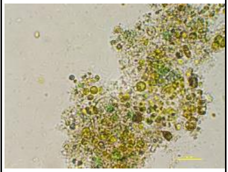
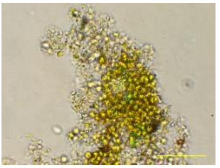
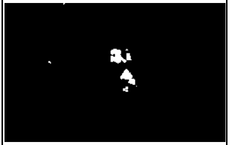
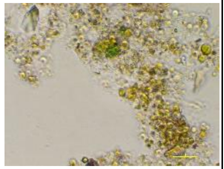
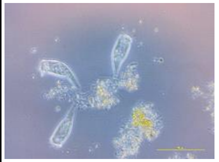
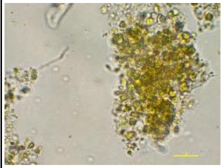
Continuous								
Semi-continuous								
SBR1								
SBR2								

16	17	20	22	24	29	30	31
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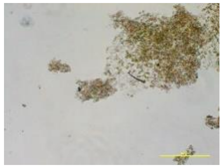
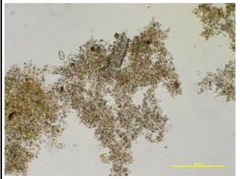
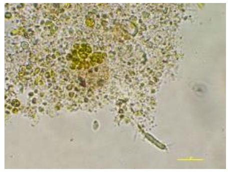
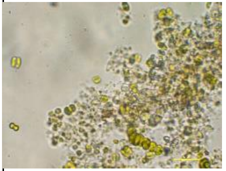
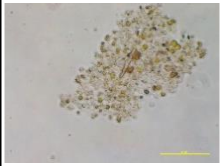
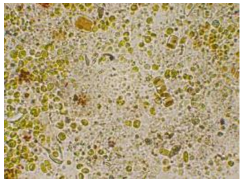
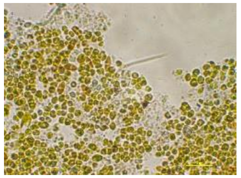
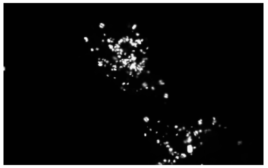
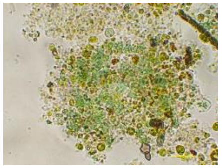
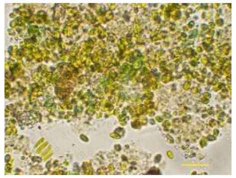
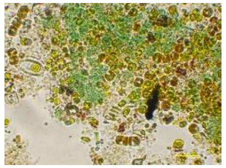
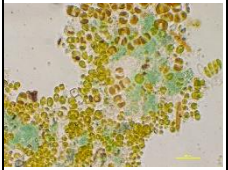
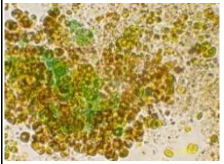
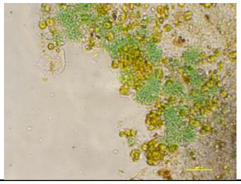
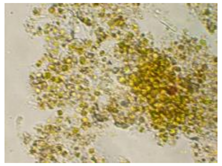
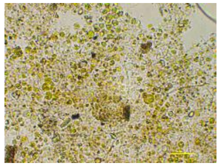
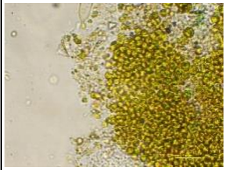
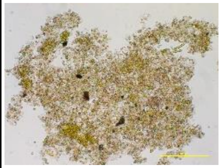
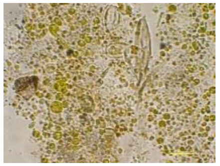
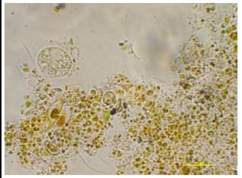
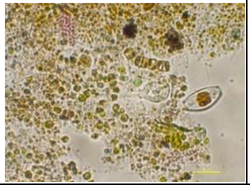
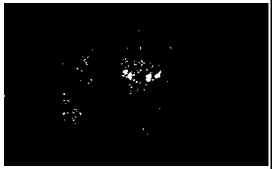
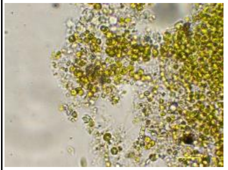
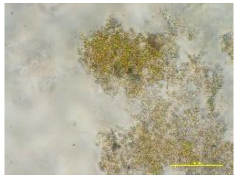
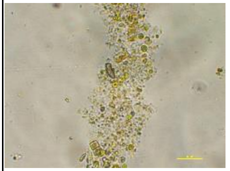
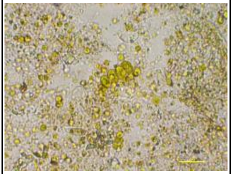
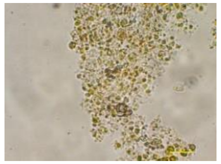
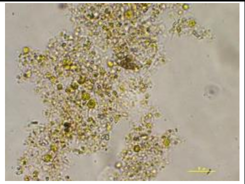
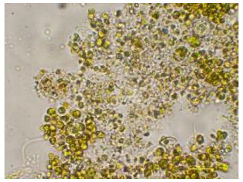
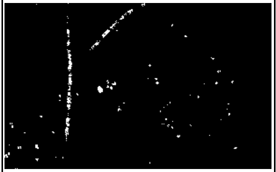
							
							
							
							

The microscopic observations show that firstly the cyanobacteria percentage was low. The main cyanobacteria species found are *Aphanocapsa* sp. and *Croococcus* sp. In the next experimental days the cyanobacteria population increases in all the systems, being the continuous reactor the one with a higher percentage. From day 4 to 7 the population of cyanobacteria in the SBRs systems decreases and the culture starts to die. Protozoa appear in these systems. In the case of continuous and semi-continuous culture the cyanobacteria presence remains at around a 30% of the culture population. This percentage is maintained till the end of the experiment. However, the green algae *Scenedesmus* sp. population increases in these reactors.

From day 7 to 16 the cyanobacteria population in the continuous reactor starts to increase being the system with a highest population of cyanobacteria. In the case of semi-continuous reactor the percentage of cyanobacteria remain constant although the *Scenedesmus* population increases. In the case of SBR reactors there is an increase of the presence of flagellated microorganisms (bacteria and protozoa). The cyanobacteria percentage is really low. SBR 1 is the SBR with a higher percentage of cyanobacteria.

At day 16 due to a problem with the acid dosage in the continuous reactor the pH decreases to 2. In this moment the majority of the microorganisms in this culture die. At this point this system cannot be anymore compared with the others. However, 2 days after normal pH is recuperated the culture recovers its green colour and the green algae becomes the main microorganism, demonstrating their survival capacity. In the case of semi-continuous reactor *Scenedesmus sp.* population follow their grow taking finally the place of cyanobacteria which decreases in quantity. In SBRs the cyanobacteria population follows decreasing becoming less than a 5% of the present microorganisms. There are almost no *Scenedesmus* present in those kind of reactor, however, the presence of other green algae species is really high.

As a sum up it can be said that semi-continuous and continuous reactors are the ones with a higher quantity of cyanobacteria. Semi-continuous system is able to maintain a high presence of cyanobacteria during all the experiment. However, at the end of the experiment the *scenedesmus* sp. present in this system starts competing with the cyanobacteria for the culture dominance. Protozoa have been observed in all the reactors, although its presence is higher in SBRs. Cyanobacteria presence in SBRs is really low.

In the next graphics there are the chlorophyll and volatile suspended solids results.

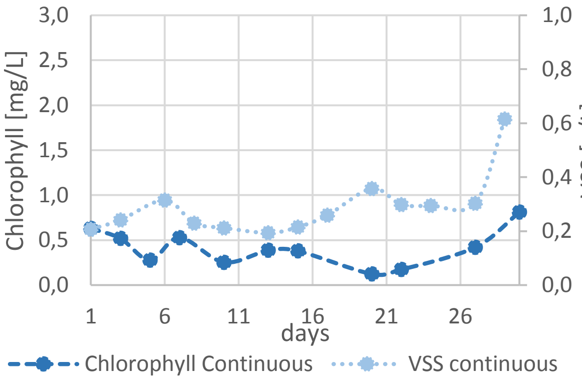


Figure 53. VSS and Chlorophyll results for Continuous system

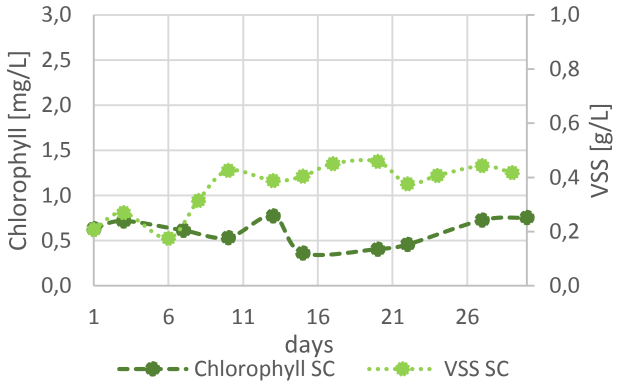


Figure 52. VSS and Chlorophyll results for Semi-continuous system

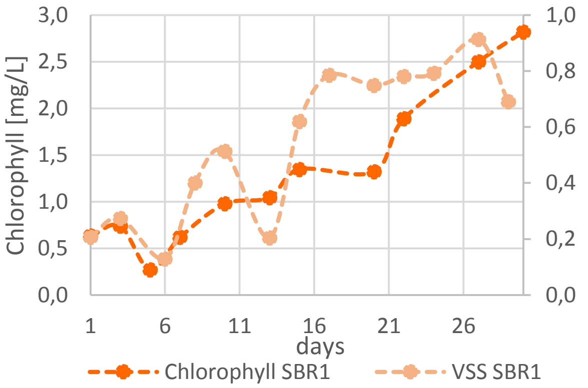


Figure 51. VSS and Chlorophyll results for SBR1 system

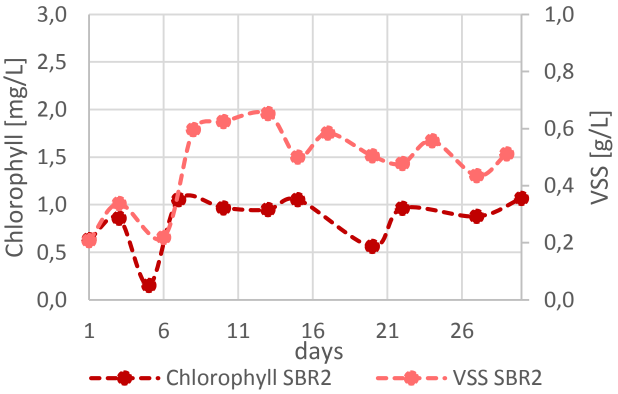


Figure 50. VSS and Chlorophyll results for SBR2 system

Figure 52, Figure 53, Figure 50 and Figure 51 show that chlorophyll and VSS have approximately the same tendency which means that an increase on the VSS is caused by an increase in the photosynthetic microorganisms. The VSS and Chlorophyll remain more or less constant in all the cultures except for the SBR1. In SBR1 there is an increase in the VSS and the chlorophyll. No increase in the cyanobacteria population has been observed in the microscopic analysis, so that rise can only be explained by an increase in the green algae population.

Some of the microorganisms present in SBRs are eliminated with the supernatant (see Operation of the photobioreactors). It is found that this quantity is around a 14% in SBR1 and a 15% in SBR2. This is a quite small quantity which can be neglected when computing the solid retention time.

In this experiment it is required to coordinate the wastewater treatment with the production of PHBs and carbohydrates. So, the culture medium is not an artificial medium as in the previous experiments otherwise it formed for water from the ponds and digestate from the anaerobic digesters. For that reason it is important to study the nutrients present in the culture

.

Table 21. Summary of the nutrients present in the culture

	Continuous	Semi-Continuous	SBR1	SBR2
Lv- PO_4^{3-} [mg/d·L]	0,098	0,098	0,492	0,492
P-PO_4^{3-} [mg/L]	1,001 ± 0,755	1,234 ± 0,627	1,165 ± 0,737	1,067 ± 0,559
TAM [mg/L]	5,419 ± 3,450	4,096 ± 3,004	3,710 ± 2,475	2,821 ± 1,919
N-NO_2^- [mg/L]	3,625 ± 1,190	3,893 ± 1,807	4,536 ± 2,742	7,258 ± 4,560
N-NO_3^- [mg/L]	7,748 ± 1,566	11,679 ± 3,593	9,234 ± 3,173	10,343 ± 3,809
$TN_{soluble}$ [mg/L]	25,112 ± 2,55	33,173 ± 3,411	40,973 ± 4,507	46,790 ± 3,818
N/P	25	27	35	44
$TOC_{soluble}$ [mg/L]	50,676 ± 6,274	46,984 ± 5,002	54,067 ± 11,839	49,546 ± 155,985
IC added [mg/L]	150	300	50	50

In Table 21 there is a summary of the most important nutrients present in the culture. In the case of phosphor it can be seen that the mean values are pretty high in all the cases. No limitation of phosphor is obtained by using water from the ponds. This predicts a low accumulation level of PHB and Carbohydrates. As seen in [3] volumetric loads of phosphor >0,28 mg P/d·L lead to low cyanobacteria concentrations and green algae dominated cultures. In the case of SBRs the phosphor volumetric load is much higher than 0,28 mg P/d·L. This can explain the low presence of cyanobacteria in this systems. A solution to reduce the load of phosphor in those reactors could be increase the HRT. If a higher hydraulic retention time is implemented the daily introduction of phosphor in the reactor will be reduced as less volume is replaced every day. There is also a high quantity of dissolved organic carbon in all the systems. This high presence of solved organic carbon may allow protozoa and heterotrophic bacteria living in this medium.

In SBRs although their amount of photosynthetic microorganisms is higher than in continue and semi-continue reactors the inorganic carbon consumption is much lower. That indicates that cyanobacteria and green algae have not a good access to inorganic carbon. It is probable that the selection of cyanobacteria in SBRs would work better if a lower SRT is applied. Lower SRT will lead to lower organic charges, eliminating thus the presence of heterotroph microorganisms and allowing a better access to nutrients by cyanobacteria.

Finally, the PHB and carbohydrates accumulated by these cultures have been studied. In Figure 54 and Figure 55 there are the obtained results:

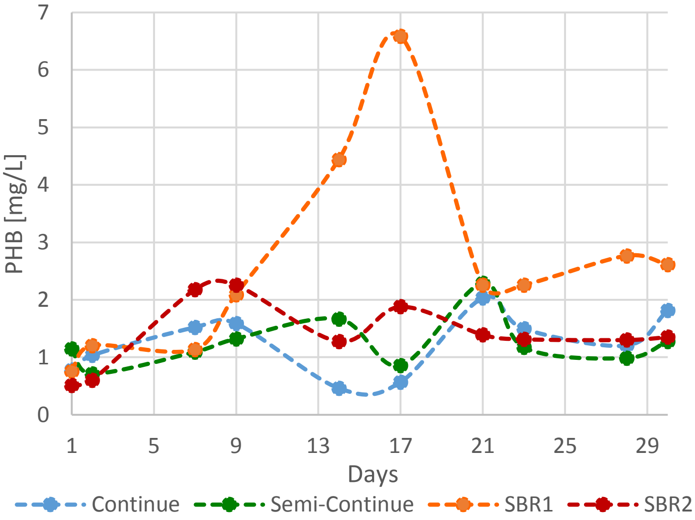


Figure 55. PHBs concentration during the experiment

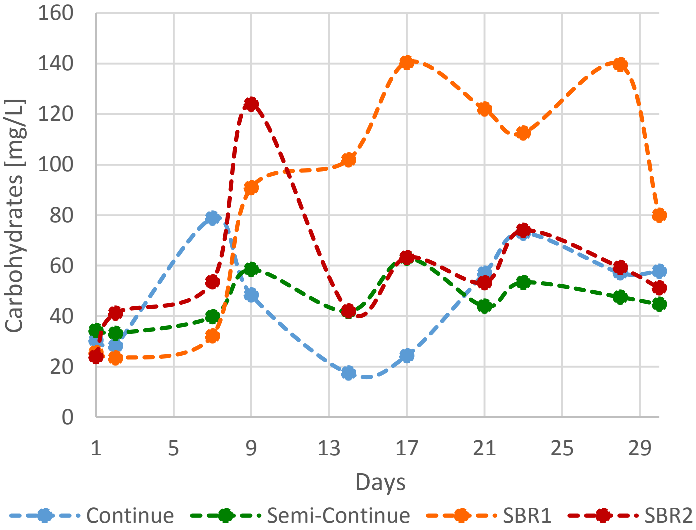


Figure 54. Carbohydrates concentration during the experiment

Table 22. Maximum concentration values

	Max PHA [mg/L]	Max Carbohydrates [mg/L]
Continue	2,03	78,77
Semi-continue	2,29	62,97
SBR1	6,59	140,40
SBR2	2,26	123,91
P-limited 12h light	76,361	432,131

Table 22 shows that the culture that accumulates more PHB and carbohydrates is SBR1 which have a solid retention time of 10 days. As previously observed this reactor is the only one that have a growing tendency in both VSS and chlorophyll. This tendency can explain the increase in the carbohydrates content, as if microorganismspopulation increases there will be more microorganisms accumulating carbohydrates. In the case of PHB there is an unexpected peak at day 17. Microscopic observations show that SBR1 is the SBR with a higher cyanobacteria content, which can explain that this reactor had a higher PHB concentration than SBR2. However, Semi-continuous reactor has a higher quantity of cyanobacteria, so this accumulation peak can only be explained by the presence of heterotrophic bacteria.

Although SBR1 is the one with higher accumulation amounts, the maximum achieved concentrations are much lower than in previous experiment as seen in Table 22 . This is because of the fact that cyanobacteria start to accumulate when the culture is completely depleted from phosphor [16]. If water from ponds wants to be used as culture medium a previous phosphorous elimination process should be done. New technologies that use nanocomposite magnetic particles functionalized with ZnFeZr-adsorbent are being investigated to efficiently and inexpensively eliminate and recover phosphates which can be afterwards used as fertilizers [73].

Finally, the maximum PHBs accumulation levels obtained in all this project have been compared with the ones obtained under photoautotrophic growth conditions in other articles. Table 23 shows that the dry cell percentage found in this project when using an artificial culture medium is around the ones obtained in other articles. It should be notice that the other articles use cultures formed by a unique cyanobacteria strain. So there are no other species competing by the nutrients. However, in this project the used biomass is formed by wastewater-born species, therefore there is a huge competition between cyanobacteria and other microorganisms, such as green algae. For that reason it can be concluded that the obtained results with the BG11 medium are pretty good in comparison with other articles. Nevertheless, the % obtained when using water from the ponds is much lower than in other articles. For that reason, more research is needed on how to eliminate cyanobacteria competitors. An improvement of the wastewater treatment process is also necessary in order to completely eliminate the phosphor present in the ponds effluent, to reach high accumulation levels.

Table 23. Comparison of the dry cell weigh % accumulation of PHBs

	Dry cell weight %	medium	**Reference**
Mixed culture (SBR1)	0,50%	Ponds Effluent	This project
Mixed culture (P-limited 12h light)	5,62%	K_2HPO_4 free BG11 medium	This project
Spirulina platensis	6%	Artificial medium	[31]
Spirulina maxima	1,2%	Mineral medium devoid of phosphorous	[16]
Nostocmus-corumNCCU- 442	6,44%	$NaNO_3$ free BG-11 medium	[37]
Synechococcus sp. PCC7942	1%	$NaNO_3$ free BG-11 medium	[74]
Aulosira fertilissima	6,4%	BG-11 Medium	[75]
Synechocystis sp. PCC 6803	11,2%	K_2HPO_4 free BG11 medium	[76]

8. Environmental analysis

The production of PHA by wastewater born cyanobacteria is in general an environmentally friendly process as the whole process is capable of consume CO_2 while they treat water without using any chemical, accumulate PHB and carbohydrates which are also economically and environmentally interesting products.

Although the general process is environmentally friendly, it should be remembered that there always exists a risk for the manipulator since wastewater which can contain pathogen microorganisms are treated. Furthermore, the chemical analysis done to control the culture use some dangerous substances, both for the environment and human health. For that reason some rules should be followed to avoid accidents and cause the minimum damage to the environment. This part summarizes the security rules and the precautions that should be followed in order to avoid possible laboratory accidents:

- Use the personal protective equipment (PPE), nylon gloves, laboratory glasses and lab coat.
- Correct labeling of the prepared solutions, products and ongoing experiments.
- Do not eat or drink in the laboratory.
- It should be known where are placed the emergency systems: extinguishers, showers, eye washes.
- Maintain clean and ordered the working space. In this laboratory as microorganisms from waste water are used it is necessary to clean the laboratory tables with bleach twice a week, in order to avoid contamination of the samples and human infection by pathogen microorganisms that could be present in wastewater.
- Clean your hands after working in the laboratory. This rule is very important to protect you against possible infections due to pathogen microorganisms from wastewater.
- Before using a chemical substance the safety data sheet should be read. All the substances should be stored and manipulated as indicated in the safety data sheets (Annex 3)
- The substances that emit toxic vapors should be manipulated under fume hoods. In this project this should be done for the following analysis:
    - Carbohydrates analysis as concentrate sulphuric acid is used.
    - Ammonia analysis due to phenol.
    - COD due to the potassium dichromate and sulphuric acid.
    - PHB quantification analysis due to chloroform.
- The generated residues should be deposited in the appropriate drum, for their subsequent treatment.

9. Conclusions

From the bibliographic review carried out and the experimental results obtained in this project the following conclusions can be deduced:

- The conventional wastewater treatment consist of 4 stages: pre-treatment, primary treatment, secondary treatment and tertiary treatment. High rate algae ponds (HRAP) can be used as an alternative secondary treatment. They are simpler and cheaper than activated sludge.

- Cyanobacteria are prokaryotic microorganisms capable of accumulating polyhydroxybutyrates and carbohydrates under photoautotrophic conditions. It has been found that nitrogen limitation changes the metabolism towards lipid and carbohydrates accumulation. Furthermore, the reduction of the amino acid synthesis causes the remaining NADPH to be converted into PHB. In the case of phosphor limitation it has been found that low levels of inorganic phosphor enhance glycogen accumulation. Moreover, the phosphor limitation causes a surplus of NADPH that is thought to turn on into PHB accumulation.

- There are two different types of photobioreactors: open and closed. Open ones are cheaper and easier to maintain. However, they suffer from contamination. On the contrary, the closed photobioreactors can be easily controlled and they reduce the contamination problems, but they are more expensive. Closed reactors can be operated in different ways: Continuous, semi-continuous, batch and sequencing batch reactor (SBR).

- The 30L photobioreactor is able to eliminate contaminants present in wastewater as it eliminates a 65% of the total inorganic nitrogen and a 70% of the phosphor. However, to substitute conventional wastewater treatment by this process it is necessary to eliminate the photobioreactor biomass by decantation. Cyanobacteria are able to grow in this kind of systems by using wastewater contaminants as a feed. The main problem is the competence with other microorganisms. To reduce it, low phosphor charge must be introduced.

- Nitrogen limitation enhance glycogen accumulation and phosphor limitation enhance PHB accumulation. 24h light and phosphor limitation is the system which reach higher PHB concentrations (104,23mg/L). Nevertheless, the economic KPI indicates that the generated benefits do not compensate the 24h illumination costs. Therefore, the optimal cultivation conditions are phosphor limitation and 12h light.

- The reactor operation that produce a higher increase of cyanobacteria presence are semi-continuous reactor. Sequencing batch reactors seems to have a high potential in the cyanobacteria cultivation. However, lower solid retention times should be applied to reduce the organic charge and thus decrease the presence of heterotroph microorganisms. Furthermore, an extra process stage is necessary to limit the phosphor from the ponds effluent, thus high PHB and glycogen concentrations will be possible.

- Cyanobacteria culture is a potential feasible method in the waste water treatment that provide PHB and glycogen as subproducts with possible worthwhile cost-benefits in the application to biodegradable plastics and bioenergy production if an adequate development is achieved.

10. Economic analysis

10.1 Materials and equipment costs

The materials and equipment costs refers to all the equipment, reagents and material used to realize all the experiments and analysis. It was divided into fungibles, reagents, laboratory equipment and reactors. For each of these categories an approximate shelf life has been established in order to compute the inversion amortization. The amortization can be computed as:

$$Amortization\ [\frac{€}{year}] = \frac{Adquisition\ costs\ [€]}{Shelf\ life\ [years]} \qquad (Eq.\ 28)$$

All the prices for the material and equipment costs were taken from Fisher Scientific catalogue except in the case of the reactors.

10.1.1. Fungibles

The *UPC Manual de gestió econòmica* [77] defines as a fungible anything that is consumed with it use or which have a shelf life lower than a year. In Table 24 there is the fungible material used in this project:

Table 24. Fungibles costs

Fungibles				
Material	**Characteristics**	**Quantity**	**unit price (€/kg)**	**Total price**
Electronic pipete Tip	5mL 500/bag	1	82,4	82,4
	1000 µl 1000/bag	1	20,27	20,27
Ependorf tub	1000 units/bag	1	36,8	36,8
Glass pasteur pipetes	1000 units/bag	1	42,81	42,81
Storage plastic bottles	500 mL 10 units/bag	3	46,8	140,4
Microscope slides	50 units	2	5,25	10,5
Microscope glass coverslips	100 units	1	8,7	8,7
Beakers	100 mL	10	4,194	41,94
	150 mL	3	4,69	14,07
	500 mL	1	6,95	6,95

Plastic beakers	500mL	3	3,55	10,65
Filter flask	glass 2L	2	91,36	182,72
Burete	25mL	1	46,9	46,9
Cylinders	25mL	1	7,175	7,175
	50mL	1	11,385	11,385
	100mL	1	7,9	7,9
Volumetric flask	50 mL	5	6,97	34,85
	100mL	1	7,03	7,03
	500mL	1	15,2	15,2
	1L	1	22,875	22,875
Erlenmeyer	100mL	10	4,903	49,03
Spatula	9x130 mm	1	2,44	2,44
	Microespatula spoon shaped 5x9 mm	1	4,26	4,26
	en forma de cuchara 5x130	1	1,83	1,83
Pliers		1	16,63	16,63
Water wash bottle	500 mL	2	4,95	9,9
Acetone flasks	250mL	1	9,0375	9,0375
Cubetes	Optical glass	2	56,95	113,9
Xeringe	0,5mL glass	1	153	153
Pipette	25mL aforate	1	9,05	9,05
	50 mL aforate	1	11,05	11,05
TOTAL				1131,66

10.1.2. Reagents

The reagents costs are the costs of buying the substances necessary to do the analysis and prepare the culture medium. No amortization was considered in this case, as the reagents will be completely expended during the project.

Table 25. Reagents costs

Reagents				
Reagents	**Characteristics**	**Quantity**	**unit price (€/kg)**	**Total price (€)**
$NaNO_3$	1L 1M	1	28,04	28,04
K_2HPO_4	250g 99+%	1	24,2	24,2
$MgSO_2\cdot 7H_2O$	500 g	1	44,1	44,1
$CaCl_2\cdot 2H_2O$	500 g	1	17,1	17,1
Citric acid	250 g 99%	1	15,2	15,2
Ammonium ferric citrate green	16,5-18,5% Fe 250g	1	27,9	27,9
$EDTANa_2$	1L 0,1M	1	45,16	45,16
Na_2CO_3	500 g 99,6%	1	26,6	26,6
H_2BO_3	500 g 99,6%	1	24,6	24,6
$MnCl_2\cdot 4H_2O$	100 g 99+%	1	15,7	15,7
$ZnSO_4\cdot 7H_2O$	500g 99%	1	27,2	27,2
$Na_2M_0O_4\cdot 2H_2O$	100 g 98%	1	27	27
$CuSO_4\cdot 5H_2O$	500g	1	23	23
$Co(NO_2)_2\cdot 6H_2O$	25g 98%	1	26,8	26,8
Potassium dichromate	50g 99,5%	1	12,7	12,7
Sulphuric acid	1L 95-98%	1	72,21	72,21
Ag_2SO_4	1L 10g/L	1	82,5	82,5
Chlorhydric acid	1L 37%	1	29,9	29,9
Acetone	2,5L extra pure	1	59,45	59,45
Magnesium carbonate	500g extra pure	1	27,5	27,5
Sodium nitroprusside	100g	1	64,7	64,7
Sodium citrate	500g	1	15,9	15,9
Sodium hydroxide	1Kg	1	21	21
Bleach	5L	1	1,19	1,19
ammonia	1L 25%	1	27,25	27,25
Heptadecane	100mL 99%	1	86,1	86,1
Chloroform	500mL 99+%	1	37,15	37,15
Phenol	500g 99+%	1	32,5	32,5
TOTAL				**942,65**

10.1.3. Laboratory equipment

The laboratory equipment are all the instruments and machines necessary to do the analysis. These equipment is considered to have a long shelf life of approximately 10 years.

Table 26. Laboratory equipment costs

Laboratory Equipment				
Material	**Characteristics**	**Quantity**	**unit price (€/unit)**	**Total price (€)**
Agitator	Model AGE	4	238	952
	RT Basic Magnetic Stirrers	2	136	272
Digester	ECO25	1	1065	1065
Anion chromatograph	Dionex ICS 1500 IC	1	14999	14999
gas chromatograph	G4350A AGILENT 7820A GC SYSTEM	1	12841,5	12841,5
TOC analyser	multi N/C® 2100/2100 S	1	12000	12000
Polytron	KINEMATICA 2500E Homogenizer	1	1337	1337
Oven	Memmert™ 400 x 560 720mm	1	2152,22	2152,22
Muffle	Thermolyne 2,1L	1	1350	1350
pH electrode		5	157	785
pH measurer		1	444	444
pH controller		4	1320	5280
conductivity meter	Hanna Instruments	1	373	373
Nephelometer		1	885	885
Dissolved oxygen electrode		1	860	860
Centrifuge	Sorvall™ ST 8 4 x 145mL (with TX-150 rotor), 6 x 50mL (with HIGHConic III rotor)	1	2081	2081
DQO reader	Range: 0 to 150mg/L	1	790	790
Microscope	Motic™ BA310 Trinocular Microscope	1	1922	1922
Fluorescence Microscope	OXION OX.3067	1	6406,95	6406,95
Computer	Lenovo Ideapad IP700-15ISK, I7-6700HQ, 8 GB RAM, GTX 950M	5	744	3720
Spectrophotometer		1	12195	12195

Analytic Balance	Max 210g	1	1235	1235
Peristaltic bombe	Fisherbrand™ FH10	18	740,52	13329,36
TOTAL				**97275,01**

10.1.4. Reactors

This part includes all the reactors andponds costs. The reactors are expected to have a long shelf life of approximately 10 years.

Table 27. Reactors costs

Reactors				
Material	**Characteristics**	**Quantity**	**unit price (€/kg)**	**Total price (€)**
3L vertical column PBR	Methacrylate	4	175	700
30L Photobioreactor	Methacrylate	1	116	116
3L Vertical column PBR lamp	Flexo Sobremesa Blanco Junior + 2 Halogen bulbs	8	42,54	340,32
30L Photobioreactor Lamp		1	146	146
High rate algae ponds		1	455	455
TOTAL				**1757,32**

10.2. Human resources

The human resources consider that in order to realize this project 1 project manager, 1 technical engineer and 1 laboratory technician are needed. The costs of hire those employees have been provided by GEMMA group administration. Finally, in order to compute the budget required for human resources during the seven month that the project last the following computation should be done:

$$Personal\ costs\ [€] = Personal\ costs\ [\frac{€}{h}] \cdot Dedication[h \cdot person] \qquad \text{(Eq. 29)}$$

Table 28. Personal costs

Human resources			
Position	**Personal costs (€/h)**	**Human resources (Hours·person)**	**Personal costs (€)**
Technical engineer	10	800	8000
Project manager	48	10	480
Laboratory technician	26	320	8320
TOTAL			**16800**

10.3. Water resources

In this part the water costs resulting from the use of water for the analysis and reactor refrigeration and material cleaning has been considered. Two different types of water has been used, tap and deionized water. The water quantity used comes from considering 12L of tap water used every day for the reactor refrigeration and material cleaning and 3L of deionized water daily used for the analysis and material cleaning.

Table 29. Water costs

Water costs			
Water	**Water quantity used (L)**	**Water cost (€/L)**	**Total Costs (€)**
Tap water	1680	0,003	4,8
Deionized water	420	0,4	168
TOTAL			**172,8**

10.4. Energetic costs

The energetic costs has been computed by considering the hours that each equipment has been in use during the seven month of the project, the power that each equipment consume and the electrical energy price. In this case considering that all the equipment can be connected at the same time the power that should be contracted is 8,12 kW. The price of the kWh found for this contracted power is 0,124 €/kWh (http://bit.ly/2qeRDmv). So, the formula used to compute the energetic costs is the following one:

$$Energetic\ costs\ [€] = 0{,}139\ \left[\frac{€}{kWh}\right] \cdot\ Equipment\ power\ [kW] \cdot hours\ in\ use\ [h] \qquad \text{(Eq. 30)}$$

Table 30. Energetic costs

Energetic costs					
Material	**Characteritics**	**Quantity**	**in use hours (h)**	**Power (W)**	**Costs (€)**
Microscope	Motic™ BA310	1	70	50 W	0,43
Fluorescence Microscope	OXION OX.3067	1	28	100 W	0,34
Agitator	Model AGE	4	5300	40 W	26,29
	130 x 30 x 65 mm	2	280	5 W	0,17
Digester		1	280	400 W	13,89

Anion chromatograph	Dionex ICS 1500 IC	1	700	1100 W	95,48
gas chromatograph	G4350A AGILENT 7820A GC SYSTEM	1	800	1500 W	148,8
TOC analyser	multi N/C® 2100/2100 S	1	420	1000 W	52,08
Polytron	KINEMATICA 2500E Homogenizer	1	14	500 W	0,87
Oven	Memmert UN160 400 x 560 720mm	1	5300	22 W	14,46
Muffle	Thermolyne 2,1L	1	42	1520 W	7,92
pH controller	Hanna Instruments	4	5300	100 W	65,72
Centrifuge	Sorvall™ ST 8 4 x 145mL	1	42	310 W	1,61
Computer	Lenovo Ideapad IP700-15ISK	5	800	500 W	49,60
Spectrophotometer		1	190	30 W	0,71
Analytic Balance	Max 210g	1	14	4 W	0,07
Peristaltic bombe	Fisherbrand™ FH10	18	98	250 W	3,04
Halogen bulb		8	3600	60 W	26,78
Fluorescent bulb		8	1120	36 W	4,99
Reactor bulb		1	2600	600 W	193,44
TOTAL					**706,64**

10.5. Total costs

Finally, with the shelf life of each of the categories the amortization of those equipment for 7 month has been computed. In Table 31 there is a summary of the total costs for each of the categories after taking into account the amortizations:

Table 31. Summary of the total cost for each category

Category	**Costs [€]**
Fungibles	1131,66
Reagents	942,65
Laboratory equipment	5674,38
reactors	102,51
Human resources	16800
Water costs	172,82
Energetic costs	706,64
TOTAL	25530,66

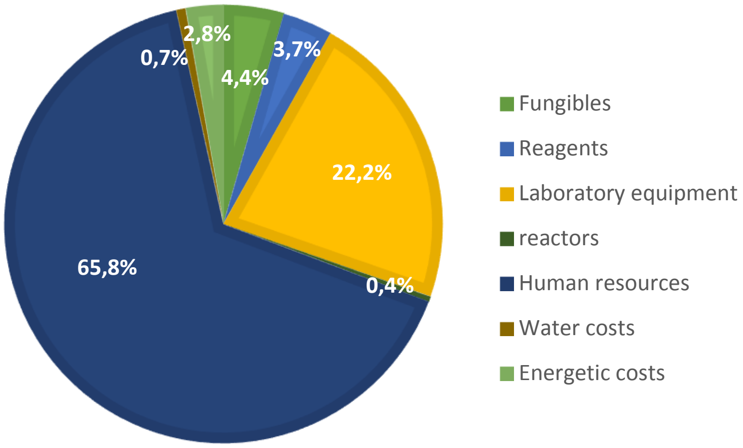


Figure 57. Distribution of the costs

In Table 30 it can be seen that the total quantity budget is 25530,66€. Figure 57 shows that a 65,8% of the budget is destinated to human resources and a 22,2% to laboratory equipment. The other costs are much less important.

11. References

[1] C. S. K. Reddy, R. Ghai, Rashmi, and V. C. Kalia, "Polyhydroxyalkanoates: An overview," *Bioresour. Technol.*, vol. 87, no. 2, pp. 137–146, 2003.

[2] S. Balaji, K. Gopi, and B. Muthuvelan, "A review on production of polyhydroxybutyrates from cyanobacteria for the production of bio plastics," *Algal Res.*, vol. 2, no. 3, pp. 278–285, 2013.

[3] D. M. Arias, E. Uggetti, M. J. García-Galán, and J. García, "Cultivation and selection of cyanobacteria in a closed photobioreactor used for secondary effluent and digestate treatment," *Sci. Total Environ.*, vol. 587–588, pp. 157–167, 2017.

[4] FAO corporate document reposity, "Wastewater treatment." [Online]. Available: http://www.fao.org/docrep/T0551E/t0551e05.htm.

[5] Environmental information system, "Flow diagram for a typical large-scale sewage treatment plant." [Online]. Available: http://www.envis.org/technology/wastewater-treatment/232-flow-diagram-for-a-typical-large-scale-sewage-treatment-plant.

[6] J. B. K. Park, R. J. Craggs, and A. N. Shilton, "Wastewater treatment high rate algal ponds for biofuel production," *Bioresour. Technol.*, vol. 102, no. 1, pp. 35–42, 2011.

[7] V. A. Cerón Hernández, C. A. Madera Parra, and M. Peña Varón, "Uso de lagunas algales de alta tasa para tratamiento de aguas residuales.," *Ing. y Desarro.*, vol. 33, no. 1, pp. 98–125, 2015.

[8] S. S. K. Lower, "Carbonate equilibria in natural waters," *Simon Fraser Univ.*, vol. Chem 1, no. Environmental Chemistry, pp. 1–26, 1999.

[9] G. Markou, I. Angelidaki, and D. Georgakakis, "Microalgal carbohydrates: An overview of the factors influencing carbohydrates production, and of main bioconversion technologies for production of biofuels," *Appl. Microbiol. Biotechnol.*, vol. 96, no. 3, pp. 631–645, 2012.

[10] N. S. Lau, M. Matsui, and A. A. A. Abdullah, "Cyanobacteria: Photoautotrophic Microbial Factories for the Sustainable Synthesis of Industrial Products," *Biomed Res. Int.*, vol. 2015, 2015.

[11] A. K. Singh, L. Sharma, N. Mallick, and J. Mala, "Progress and challenges in producing polyhydroxyalkanoate biopolymers from cyanobacteria," *J. Appl. Phycol.*, pp. 1–20, 2016.

[12] J.-Y. Chee, S.-S. Yoga, N. Lau, S. Ling, R. M. M. Abed, and K. Sudesh, "Bacterially Produced Polyhydroxyalkanoate (PHA): Converting Renewable Resources into Bioplastics," *Curr. Res. Technol. Educ. Top. Appl. Microbiol. Microb. Biotechnol.*, no. April 2016, pp. 1395–1404, 2010.

[13] N. Xiao and N. Jiao, "Formation of polyhydroxyalkanoate in aerobic anoxygenic phototrophic bacteria and its relationship to carbon source and light availability," *Appl. Environ. Microbiol.*, vol. 77, no. 21, pp. 7445–7450, 2011.

[14] G. Y. A. Tan, C. L. Chen, L. Li, L. Ge, L. Wang, I. M. N. Razaad, Y. Li, L. Zhao, Y. Mo, and J. Y. Wang, "Start a research on biopolymer polyhydroxyalkanoate (PHA): A review," *Polymers (Basel).*, vol. 6, no. 3, pp. 706–754, 2014.

[15] L. J. Stal, “Poly(hydroxyalkanoate) in cyanobacteria: an overview,” *FEMS Microbiol. Lett.*, vol. 103, no. 2–4, pp. 169–180, 1992.

[16] R. De Philippis, C. Sili, and M. Vincenzini, “Glycogen and poly-β-hydroxybutyrate synthesis in Spirulina maxima,” *J. Gen. Microbiol.*, vol. 138, no. 8, pp. 1623–1628, 1992.

[17] P. H. Yu, H. Chua, A. L. Huang, W. Lo, and G.-Q. Chen, “Conversion of food industrial wastes into bioplastics,” *Biochem. Biotechnol*, 1998.

[18] F. Wang and S. . Lee, “Poly(3-hydroxybutyrate) production with high productivity and high polymer content by a fed-batch culture of Alcaligenes latus under nitrogen limitation.,” *Environ. Microbiol*, 1997.

[19] E. Grothe, M. Moo-Young, and Y. Chisti, “Fermentation optimization for the production of poly(beta-hydroxybutyric acid) microbial thermoplastic.,” *Enzym. Microb. Technol*, 1999.

[20] T. Yamane, M. Fukunaga, and Y. Lee, “Increased PHB productivity by high-cell-density fed-batch culture of Alcaligenes latus, a growth-associated PHB producer,” *Biotechnol. Bioeng.*, 1996.

[21] J. Gomez, M. Rodrigues, R. Alli, B. Torres, C. B. Netto, M. Oliveira, and L. Da Silva, “Evaluation of soil Gram-negative bacteria yielding polyhydroxyalkanoic acids from carbohydrates and propionic acid,” *Microbilogy Biotechnol.*, 1996.

[22] W. Pan, J. A. Perrotta, A. J. Stipanovic, C. T. Nomura, and J. P. Nakas, “Production of polyhydroxyalkanoates by Burkholderia cepacia ATCC 17759 using a detoxified sugar maple hemicellulosic hydrolysate.,” *Microbiol. Biotechnol.*, 2012.

[23] C. Zhu, C. T. Nomura, J. A. Perrotta, A. J. Stipanovic, and J. P. Nakas, “Production and characterization of poly-ß-hydroxybutyrate from biodiesel-glycerol by Burkholderia cepacia ATCC 17759.,” *Biotechnology*, 2010.

[24] H. E. Valentin, E. Y. Lee, C. Y. Choi, and A. Steinbüchel, “Identification of 4-hydroxyhexanoic acid as a new constituent of biosynthetic polyhydroxyalkanoic acids from bacteria,” *Microbilogy Biotechnol.*, 1994.

[25] A. Ishizaki and K. Tanaka, “Production of poly-β-hydroxybutyric acid from carbon dioxide by Alcaligenes eutrophus ATCC 17697,” *Ferment. Bioeng.*, 1991.

[26] S. Shahid, R. Mosrati, J. Ledauphin, C. Amiel, P. Fontaine, J.-L. Gaillard, and D. Corroler, “Impact of carbon source and variable nitrogen conditions on bacterial biosynthesis of polyhydroxyalkanoates: Evidence of an atypical metabolism in Bacillus megaterium DSM 509.,” *Biosci. Bioeng.*, 2013.

[27] A. Bhattacharyya, A. Pramanik, S. K. Maji, S. Haldar, U. K. Mukhopadhyay, and J. Mukherjee, “Utilization of vinasse for production of poly-3-(hydroxybutyrate-co-hydroxyvalerate) by Haloferax mediterranei.,” *AMB Express*, 2012.

[28] M. Koller, A. Atlić, Y. Gonzalez-Garcia, C. Kutschera, and G. Braunegg, “Polyhydroxyalkanoate (PHA) biosynthesis from whey lactose.,” *Macromol*, 2008.

[29] C. Hermann-Krauss, M. Koller, A. Muhr, H. Fasl, F. Stelzer, and G. Braunegg, “Archaeal production of polyhydroxyalkanoate (PHA) co-and terpolyesters from biodiesel industry-derived by-products.,” *Archaea*, 2013.

[30] J. Han, J. Hou, H. Liu, S. Cai, B. Feng, J. Zhou, and H. Xiang, “Wide distribution among halophilic archaea of a novel polyhydroxyalkanoate synthase subtype with homology to bacterial type III synthases.,” *Environ. Microbiol.*, 2010.

[31] J. Campbell, S. E. J. Stevens, and D. L. Balkwill, “Accumulation of poly-ß-hydroxybutyrate in Spirulina platensis.,” *Bacteriology*, vol. 149, pp. 361–363, 1982.

[32] M. Miyake, K. Kataoka, M. Shirai, and Y. Asada, “Control of poly-beta-hydroxybutyrate synthase mediated by acetyl phosphate in cyanobacteria.,” *Bacteriology*, vol. 179, pp. 5009– 5013, 1997.

[33] S. Hein, H. Tran, and A. Steinbuchel, “Synechocystis sp. PCC6803 possesses a two-component polyhydroxyalkanoic acid synthase similar to that of anoxygenic purple sulfur bacteria.,” *Microbiology*, vol. 170, pp. 162–170, 1998.

[34] B. Panda and N. Mallick, “Enhanced poly-??-hydroxybutyrate accumulation in a unicellular cyanobacterium, Synechocystis sp. PCC 6803,” *Lett. Appl. Microbiol.*, vol. 44, no. 2, pp. 194–198, 2007.

[35] L. Sharma, A. Kumar Singh, B. Panda, and N. Mallick, “Process optimization for poly-ß-hydroxybutyrate production in a nitrogen fixing cyanobacterium, Nostoc muscorum using response surface methodology,” *Bioresour. Technol.*, vol. 98, no. 5, pp. 987–993, 2007.

[36] L. Lama, B. Nicolaus, and V. Calandrelli, “Effect of growth conditions on endo- and exopolymer biosynthesis in Anabaena cylindrica 10 C,” *Phytochemistry*, vol. 42, pp. 655–659, 1996.

[37] S. Ansari and T. Fatma, “Cyanobacterial polyhydroxybutyrate (PHB): Screening, optimization and characterization,” *PLoS One*, vol. 11, no. 6, pp. 1–20, 2016.

[38] J. Komárek and T. Hauer, “The on-line database of cyanobacterial genera.” [Online]. Available: http://www.cyanodb.cz/main.

[39] Center for freshwater biology, “Aphanocapsa.” [Online]. Available: http://cfb.unh.edu/CyanoKey/Aphanocapsa.html.

[40] AP biology Wiki, “Pyruvate Oxidation & The Citric Acid Cycle.” [Online]. Available: http://wiki.pingry.org/u/ap-biology/index.php/Pyruvate_Oxidation_%26_The_Citric_Acid_Cycle.

[41] S. Aikawa, S.-H. Ho, J.-S. Chang, and T. Hasunuma, “Improving polyglucan production in cyanobacteria and microalgae via cultivation design and metabolic engineering,” *Biotechnol. J.*, vol. 10, no. 6, pp. 886–898, 2015.

[42] B. Drosg, I. Fritz, F. Gattermayr, and L. Silvestrini, “Photo-autotrophic Production of Poly(hydroxyalkanoates) in Cyanobacteria,” *Chem. Biochem. Eng. Q.*, vol. 29, no. 2, pp. 145–156, 2015.

[43] A. Richmond, “Handbook of microalgal culture: biotechnology and applied phycology/edited by Amos Richmond.,” *Orton.Catie.Ac.Cr*, p. 472, 2004.

[44] Power plant CCS Report, “Types of PBR.” [Online]. Available: http://www.powerplantccs.com/ccs/cap/fut/alg/types_of_pbr.html.

[45] Oilgae, “Types of Photobioreactors.” [Online]. Available: http://www.oilgae.com/algae/cult/pbr/typ/typ.html.

[46] Smith, Elger, and Mleziva, “Implementation of solids retention time (SRT) control in wastewater treatment,” *YSI Solid Retent. Time White Pap.*, pp. 1–6, 2014.

[47] R. Jain, S. Kosta, and A. Tiwari, “Polyhydroxyalkanoates : A way to sustainable development of bioplastics,” *Chronicles Young Sci.*, vol. 1, no. 3, pp. 10–15, 2010.

[48] A. S. M. Chua, H. Takabatake, H. Satoh, and T. Mino, “Production of polyhydroxyalkanoates (PHA) by activated sludge treating municipal wastewater: Effect of pH, sludge retention time (SRT), and acetate concentration in influent,” *Water Res.*, vol. 37, no. 15, pp. 3602–3611, 2003.

[49] H. A. Wu, D. S. Sheu, and C. Y. Lee, “Rapid differentiation between short-chain-length and medium-chain-length polyhydroxyalkanoate-accumulating bacteria with spectrofluorometry,” *J. Microbiol. Methods*, vol. 53, no. 1, pp. 131–135, 2003.

[50] K. Wolfgang, *Kunststoffchemie für Ingenieure von der Synthese bis zur Anwendung*, 3rd ed. 2011.

[51] Polymer processing, “polypropylene.” [Online]. Available: http://www.polymerprocessing.com/polymers/PP.html.

[52] T. Maier, Clive; Calafut, *Polypropylene: the definitive user’s guide and databook*. 1998.

[53] British Plastics Federation, “polypropylene (PP).” [Online]. Available: http://www.bpf.co.uk/plastipedia/polymers/pp.aspx.

[54] international centre for trade and sustainable development, “Biocombustibles y crisis alimentaria: nuevas evidencias,” *Puentes*, 2008. [Online]. Available: http://www.ictsd.org/bridges-news/puentes/news/biocombustibles-y-crisis-alimentaria-nuevas-evidencias.

[55] S. Díaz-Troya, L. López-Maury, A. M. Sánchez-Riego, M. Roldán, and F. J. Florencio, “Redox regulation of glycogen biosynthesis in the cyanobacterium synechocystis sp. PCC 6803: Analysis of the AGP and glycogen synthases,” *Mol. Plant*, vol. 7, no. 1, pp. 87–100, 2014.

[56] J. Berg, J. L. Tymoczko, and L. Stryer, *Biochemistry*, 7th ed. 2011.

[57] J. Kumar, A. Singh, “Microalgae and Cyanobacteria Biofuels : A Sustainable Alternate to Crop-based Fuels,” no. December 2015, 2016.

[58] Biologist notes for A level, “Anaerobic respiration - Ethanol and Lactate pathways,” 2015. [Online]. Available: http://biology4alevel.blogspot.com.es/2015/08/91-anaerobic-respiration-

ethanol.html.

[59] Friends of Earth, “Briefing Anaerobic digestion,” no. September, pp. 1–7, 2007.

[60] Instituto para la diversificación y ahorro de la energía (IDAE), *Biomasa: Digestores anaerobios*. 2007.

[61] L. Chen and H. Neibling, “Anaerobic Digestion Basics,” pp. 1–6, 2014.

[62] Waste to energy research and technology council, “Anaerobic Digestion Process,” 2009. [Online]. Available: http://www.wtert.eu/default.asp?Menue=13&ShowDok=12.

[63] R. Y. Stanier, R. Kunisawa, M. Mandel, and G. Cohen-Bazire, “Purification and properties of unicellular blue-green algae (Order Chroococcales).,” *Bacteriol. Rev.*, vol. 35, no. 2, pp. 171–205, 1971.

[64] Amerian public health Association, “Standard Methods for the Examination of Water and Wastewater,” *Stand. Methods*, p. 541, 2012.

[65] Fundamentals of environmental Measurements, “Measuring Dissolved Oxygen.” [Online]. Available: http://www.fondriest.com/environmental-measurements/equipment/measuring-water-quality/dissolved-oxygen-sensors-and-methods/.

[66] LPD LAB SERVICES, “Anion - Ion Chromatography - IC Analysis.” [Online]. Available: http://www.lpdlabservices.co.uk/analytical_techniques/chemical_analysis/anion_chromatography.php.

[67] L. Solórzano, “determination of ammonia in natural waters by the phenolhypochlorite method,” pp. 799–801, 1968.

[68] M. Dubois, K. A. Gilles, and J. K. Hamilton, “Colorimetric method for determination of sugars and related substances,” *Anal. Chem.*, vol. 28, no. 3, pp. 350–356, 1956.

[69] The council of the european communities, “direvtive 91/271/EEC concerning urban wastewater treatment,” *Off. Jounal Eur. communities*, no. L, p. 10, 1991.

[70] W. F. J. Vermaas, “Photosynthesis and respiration in cyanobacteria,” *Encycl. Life Sci.*, pp. 1–7, 2001.

[71] J. Sauer, U. Schreiber, R. Schmid, U. Völker, and K. Forchhammer, “Nitrogen starvation-induced chlorosis in Synechococcus PCC 7942. Low-level photosynthesis as a mechanism of long-term survival.,” *Plant Physiol.*, vol. 126, no. May, pp. 233–243, 2001.

[72] R. Schwarz and K. Forchhammer, “Acclimation of unicellular cyanobacteria to macronutrient deficiency: Emergence of a complex network of cellular responses,” *Microbiology*, vol. 151, no. 8, pp. 2503–2514, 2005.

[73] A. Drenkova-Tuhtan, M. Schneider, M. Franzreb, C. Meyer, C. Gellermann, G. Sextl, K. Mandel, and H. Steinmetz, “Pilot-scale removal and recovery of dissolved phosphate from secondary wastewater effluents with reusable ZnFeZr adsorbent @ Fe3O4/SiO2 particles with magnetic

harvesting," *Water Res.*, vol. 109, pp. 77–87, 2017.

[74] T. Suzuki, M. Miyake, Y. Tokiwa, H. Saegusa, T. Saito, and Y. Asada, "A recombinant cyanobacterium that accumulates Plyhydroxybutyrate," vol. 18, no. 9, pp. 1047–1050, 1996.

[75] S. Samantaray and N. Mallick, "Production and characterization of poly-β-hydroxybutyrate (PHB) polymer from Aulosira fertilissima," *J. Appl. Phycol.*, vol. 24, no. 4, pp. 803–814, 2012.

[76] B. Panda and N. Mallick, "Enhanced poly-ß-hydroxybutyrate accumulation in a unicellular cyanobacterium, Synechocystis sp. PCC 6803," *Lett. Appl. Microbiol.*, vol. 44, no. 2, pp. 194–198, 2007.

[77] Universitat politècnica de Catalunya, "Manual de gestió econòmica," pp. 1–13.

[78] Life sciences und facility Management, "CCMA - Combining Competences in MicroAlgae." [Online]. Available: https://www.zhaw.ch/de/lsfm/forschung/formen-der-zusammenarbeit/ccma-combining-competences-in-microalgae/.

[79] Chlorella benefits, "The Different Kinds of Chlorella's Production," 2011. [Online]. Available: https://chlorelle.wordpress.com/.

[80] Algae Detective, "All about algae-Open ponds." [Online]. Available: http://www.algaedetective.com/open-ponds.html.

[81] Climate-KIC, "Micro algae biorefinery." [Online]. Available: http://algaerefinery.eu/.

[82] A. A. Vertès, N. Qureshi, H. P. Blaschek, and H. Yukawa, *Biomass to biofuels: Strategies for global industries*. John Wiley & Sons, 2010.

[83] Donewell, "Customised Solutions for Researchers." [Online]. Available: http://donewell.com.my/Customised_Solutions_for_Researchers.php.

[84] Environmental protection agency, "Sequencing Batch Reactor Systems." [Online]. Available: http://web.deu.edu.tr/atiksu/ana07/epa02.html.

Annex 1: Numeric results

A1.1. 30L Photobioreactor

Table 32. Turbidity, dissolved oxygen and pH results

Day	PBR		
	Turbidity	OD	pH
[-]	[UNT]	[mg/L]	[-]
1	52	7,88	7,77
3	48	10,60	7,79
4	60	14,40	8,10
7	76	11,35	7,69
8	197	8,80	7,81
9	152	11,60	7,92
10	185	7,10	7,20
11	192	8,02	7,54
14	167	9,10	7,46
15	127	8,10	7,80
16	199	8,34	7,96
17	223	8,70	7,80
18	308	9,80	7,72
21	212	8,48	7,74
23	271	6,60	7,73
24	364	7,20	7,49
25	357	7,58	7,65
28	332	7,66	7,46
29	360	7,76	7,69
30	558	8,38	7,69
31	414	9,45	7,80
32	511	7,68	7,44
35	360	7,80	7,47
36	341	8,86	7,30
37	338	6,80	7,46
39	291	7,83	7,40
42	423	6,40	7,03
43	296	12,33	7,49
44	355	9,38	7,44
45	375	9,32	7,55
46	-	10,80	7,64
49	362	10,69	7,56

50	433	3,71	8,25
51	981	13,65	7,79
52	795	8,14	7,40
53	459	9,88	7,50
91	362	--	7,62
92	763	--	7,62
93	714	7,32	7,64
94	401	7,61	8,62
95	472	8,95	8,12
98	369	7,42	7,84
99	319	8,81	8,22
100	473	9,68	8,21
101	407	--	8,24
102	411	8,19	9,99
105	340	10,35	9,57
106	434	9,94	9,21
107	367	9,57	8,12
108	--	9,21	7,90
109	379	8,18	8,12
112	367	7,07	7,95
113	--	--	8,19
114	358	8,15	8,05
115	504	8,03	--
116	469	7,91	8,00
119	347	8,48	8,63
120	382	8,40	9,66
121	419	8,41	9,54
122	350	8,57	8,57
123	294	8,31	9,80
126	314	8,10	8,10
127	--	8,13	9,02
128	313	8,28	9,26
129	331	8,05	8,89
130	678	7,70	6,86
133	--	8,53	8,53
134	556	7,93	7,93

Table 33. Total and volatile suspended solids results

Day	**Reactor feed**		**PBR**	
	TSS	**VSS**	**TSS**	**VSS**
[-]	[g/L]	[g/L]	[g/L]	[g/L]
1	0,259	0,196	0,068	0,072
8	0,285	0,235	0,327	0,308
15	0,412	0,328	0,317	0,283
23	0,436	0,337	0,311	0,261
29	0,459	0,379	0,817	0,640
36	0,595	0,495	0,411	0,337
45	0,396	0,341	0,348	0,309
51	0,508	0,416	1,105	0,857
93	0,515	0,413	0,643	0,541
107	0,375	0,416	0,367	0,300
115	0,311	0,273	0,782	0,609
120	0,361	0,283	0,359	0,280
122	0,291	0,256	0,448	0,361

Table 34. Chlorophyll results

Day	**Date**	**PBR**										
		λ 750	λ 664	λ 647	λ 630	A664	A647	A630	Va	Vm	Ca	Ca mean
[-]	[-]	[nm]	[nm]	[nm]	[nm]	[nm]	[nm]	[nm]	[mL]	[mL]	[mg/L]	[mg/L]
1	11/10/2016	0,024	0,121	0,070	0,059	0,097	0,046	0,062	5	6,9	0,778	0,682
		0,025	0,105	0,082	0,060	0,080	0,057	0,045	5	7,3	0,587	
9	19/10/2016	0,010	0,286	0,113	0,071	0,276	0,103	0,215	5	6,7	2,310	2,094
		0,011	0,242	0,096	0,061	0,231	0,085	0,181	5	6,9	1,878	
16	26/10/2016	0,001	0,126	0,042	0,031	0,125	0,041	0,095	5	6,6	1,069	0,973
		0,001	0,113	0,038	0,029	0,112	0,037	0,084	5	7,2	0,877	
23	02/11/2016	-0,010	0,094	0,028	0,020	0,104	0,038	0,074	5	6,9	0,846	0,581
		0,050	0,090	0,034	0,026	0,040	-0,016	0,064	5	7,8	0,316	
28	07/11/2016	-0,030	0,155	0,048	0,031	0,185	0,078	0,124	5	7,4	1,393	1,335
		-0,040	0,133	0,042	0,027	0,173	0,082	0,106	5	7,5	1,277	
29	08/11/2016	0,003	0,259	0,080	0,050	0,256	0,077	0,209	5	7,4	1,958	2,177
		0,005	0,276	0,087	0,053	0,271	0,082	0,223	5	6,4	2,396	
36	15/11/2016	-0,002	0,174	0,053	0,032	0,176	0,055	0,142	5	7,2	1,382	1,437
		-0,002	0,196	0,060	0,035	0,198	0,062	0,161	5	7,5	1,492	
45	24/11/2016	-0,092	0,009	-0,060	-0,061	0,101	0,032	0,070	5	7,3	0,782	0,869
		-0,097	0,023	-0,060	-0,070	0,120	0,037	0,093	5	7,1	0,956	
51	30/11/2016	-0,005	0,367	0,105	0,060	0,372	0,110	0,307	5	7,3	2,886	3,322
		-0,003	0,448	0,130	0,073	0,451	0,133	0,375	5	6,8	3,757	
93	11/01/2017	0,000	0,173	0,052	0,036	0,173	0,052	0,137	5	6,9	1,420	1,319

		0,001	0,162	0,047	0,033	0,161	0,046	0,129	5	7,5	1,218	
100	18/01/2017	-0,009	0,193	0,066	0,048	0,202	0,075	0,145	5	6,4	1,771	1,652
		-0,056	0,135	0,013	0,002	0,191	0,069	0,133	5	7	1,533	
107	25/01/2017	0,001	0,123	0,031	0,023	0,122	0,030	0,100	5	6,6	1,054	0,957
		0,003	0,108	0,025	0,016	0,105	0,022	0,092	5	7	0,859	

Table 35. Alkalinity results

Day	Reactor feed	PBR
[-]	[mg/L]	[mg/L]
1	216	65
3	184	80
8	178	151
10	182	151
15	196	154
17	192	159
23	214	150
25	224	147
29	227,8	176
31	244	158
36	248	116
43	226	234
44	84	219
45	120	199
50	800	407
52	90	370
92	202	220
94	202	180
99	310	164
101	302	160
106	298	174
108	290	181
113	290	181
115	292	185
118	274	192
120	252	205
125	280	194
127	264	182
132	276	206
136	282	179
142	258	148

Table 36. Ammonia results

Day	Date	Pond effluent			Reactor feed			PBR		
		abs	F D	Ammonia	abs	FD	Ammonia	abs	FD	Ammonia
[-]	[-]	[-]	[-]	[mg/L]	[-]	[-]	[mg/L]	[-]	[-]	[mg/L]
1	11/10/2016	0,115	1	0,129	0,044	50	2,229	0,343	1	0,402
8	18/10/2016	0,129	1	0,146	0,069	50	3,723	0,019	1	0,015
11	21/10/2016	0,089	1	0,098	0,073	50	3,962	0,104	1	0,116
15	25/10/2016	0,089	1	0,098	0,105	50	5,874	0,042	1	0,042
17	27/10/2016	0,126	1	0,143	0,082	50	4,500	0,031	1	0,029
23	02/11/2016	0,059	1	0,063	0,122	50	6,890	0,312	1	0,365
25	04/11/2016	0,138	1	0,157	0,162	50	9,280	0,056	1	0,059
29	08/11/2016	0,629	1	0,744	0,100	50	5,575	0,074	1	0,080
31	10/11/2016	0,134	1	0,152	0,137	50	7,786	0,157	1	0,180
36	15/11/2016				0,151	50	8,622	0,209	1	0,242
39	18/11/2016				0,092	50	5,097	0,030	5	0,139
43	22/11/2016				0,142	50	8,085	0,142	1	0,162
45	24/11/2016				0,122	50	6,890	0,090	1	0,100
52	01/12/2016				0,109	50	6,113	0,223	1	0,258
101	19/01/2017				0,236	51	13,975	0,064	2	0,137

Table 37. Anions results

Pond					
Nitrites [NO_2^-]	N- NO_2^-	Nitrates [NO_3^-]	N- NO_3^-	Orthophosphates [PO_4^{3-}]	P- PO_4^{3-}
[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
11,109	3,381	72,759	16,429	4,371	1,425
5,855	1,782	65,570	14,806	3,381	1,103
3,248	0,989	64,908	14,657	0,000	0,000
4,684	1,426	77,842	17,577	3,418	1,115
2,832	0,862	69,023	15,586	2,548	0,831
2,292	0,698	76,124	17,189	4,389	1,431
2,020	0,615	97,294	21,970	4,023	1,312
3,796	1,155	91,246	20,604	5,388	1,757
2,478	0,754	75,172	16,974	7,201	2,348
3,850	1,172	148,674	33,572	7,450	2,429

Day	Reactor feed					
	Nitrites [NO_2^-]	N- NO_2^-	Nitrates [NO_3^-]	N- NO_3^-	Orthophosphates [PO_4^{3-}]	P- PO_4^{3-}
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	5,486	1,670	67,551	15,253	2,140	0,698
3	5,486	1,670	30,250	6,831	6,951	2,267
8	4,233	1,288	70,804	15,988	2,970	0,969
15	3,053	0,929	64,567	14,580	3,826	1,248
17	3,687	1,122	65,384	14,764	2,118	0,691
24	2,147	0,653	66,197	14,948	3,248	1,059
31	2,502	0,761	102,329	23,107	4,928	1,607
36	3,602	1,096	112,683	25,445	6,736	2,197
43	5,070	1,543	122,565	27,676	6,145	2,004
101	0,000	0,000	62,840	14,190	5,605	1,828
134	5,186	1,578	70,340	15,883	6,121	1,996

Day	PBR					
	Nitrites [NO_2^-]	N- NO_2^-	Nitrates [NO_3^-]	N- NO_3^-	Orthophosphates [PO_4^{3-}]	P- PO_4^{3-}
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	2,162	0,658	12,930	2,920	0,000	0,000
3	0,000	0,000	22,202	5,013	2,064	0,673
8	1,473	0,448	11,407	2,576	0,000	0,000
17	1,548	0,471	10,241	2,312	0,000	0,000
24	5,006	1,524	34,360	7,759	2,218	0,723
31	1,682	0,512	72,400	16,348	3,383	1,103
36	2,711	0,825	65,919	14,885	2,982	0,972
43	2,534	0,771	57,849	13,063	4,332	1,413
101	0,000	0,000	62,624	14,141	0,000	0,000
134	5,316	1,618	37,041	8,364	0,000	0,000

A1.2. 3L Vertical cylinder reactors

A1.2.1. Optimal nutrients and light conditions for PHB and carbohydrates accumulation

Table 38. Dissolved oxygen results

	12h light		**24h light**	
	N-limited	**P-limited**	**N-limited**	**P-limited**
Day	**DO**	**DO**	**DO**	**DO**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	6,63	6,68	7,60	7,63
2	7,00	7,48	8,12	9,12
3	6,50	7,20	4,83	8,81
4	6,24	6,38	7,28	9,74
5	5,63	6,21	7,07	7,14
8	5,34	6,19	6,73	6,46
9	5,99	6,50	7,35	8,86
10	5,90	6,60	9,57	6,56
11	6,06	6,61	6,92	5,97
12	6,17	5,80	6,14	9,61
15	6,54	6,53	6,19	3,72
Mean	6,18	6,56	7,07	7,60
interval	0,28	0,27	0,71	1,09

Table 39. Total and volatile suspended solids results12h light experiments

12h light				
Day	**N-limited**		**P-Limited**	
	TSS	VSS	TSS	VSS
[-]	[g/L]	[g/L]	[g/L]	[g/L]
1	0,445	0,323	0,387	0,343
3	0,571	0,544	0,676	0,648
5	0,718	0,664	0,683	0,659
8	0,738	0,696	0,857	0,804
12	1,121	0,947	1,201	1,182
15	0,829	0,762	1,328	1,226

Table 40. Total and volatile suspended solids results 24h light experiments

24h light				
Day	N-limited		P-Limited	
	TSS	VSS	TSS	VSS
[-]	[g/L]	[g/L]	[g/L]	[g/L]
1	0,422	0,355	0,422	0,355
3	0,843	0,722	0,846	0,726
5	0,763	0,752	0,928	0,834
8	1,023	0,971	1,835	1,628
10	0,951	0,856	1,586	1,409
12	0,971	0,905	1,439	1,286
15	0,946	0,865	1,436	1,384

Table 41. TOC results

24h light		
Day	**N-limited**	**P-limited**
	TOC	**TOC**
[-]	[mg/L]	[mg/L]
1	86,31	94,54
3	172,37	153,41
5	197,55	215,62
9	245,50	408,14
10	231,37	377,25
12	211,00	404,28
15	203,75	368,13

12h light		
Day	**N-limited**	**P-limited**
	TOC	**TOC**
[-]	[mg/L]	[mg/L]
1	79,58	78,08
3	41,56	57,64
5	65,53	117,80
7	77,47	146,95
9	129,80	216,41
12	191,37	293,38
15	167,71	331,16

Table 42. Inorganic Carbon results 12h light experiment

12h light			
Day	**Date**	**N-limited**	**P-limited**
		IC	**IC**
[-]	[-]	[mg/L]	[mg/L]
1	30/01/2017	3,40	2,78
1,01	30/01/2017	153,40	152,78
1,5	30/01/2017	72,02	79,63
2	31/01/2017	73,88	75,65
2,01	31/01/2017	223,88	225,65
2,5	31/01/2017	92,53	107,21
3	01/02/2017	102,50	98,96
3,01	01/02/2017	252,50	248,96
3,5	01/02/2017	158,94	177,51
4	02/02/2017	174,50	158,60
4,01	02/02/2017	324,50	308,60
4,5	02/02/2017	231,55	204,02
5	03/02/2017	220,90	191,50
5,5	03/02/2017	178,32	147,26
7,5	05/02/2017	80,51	41,08
8	06/02/2017	120,90	89,63
9	07/02/2017	82,25	23,88
10	08/02/2017	66,38	66,37
11	09/02/2017	41,96	23,46
12	10/02/2017	31,19	0,00
12,01	10/02/2017	131,19	100,00
12,5	10/02/2017	76,30	50,00
15	13/02/2017	68,29	22,62

Table 43. Inorganic Carbon results 24h light experiments

24h light			
Day	**Date**	**N-limited**	**P-limited**
		IC	**IC**
[-]	[-]	[mg/L]	[mg/L]
1	16/01/2017	38,07	40,88
1,01	16/01/2017	138,07	140,88
2	17/01/2017	0,29	1,51
2,01	17/01/2017	150,29	151,51
3	18/01/2017	8,78	10,83
3,01	18/01/2017	158,78	160,83
4	19/01/2017	19,46	17,80

4,01	19/01/2017	169,46	167,80
5	20/01/2017	39,07	63,83
5,01	20/01/2017	189,07	213,83
6	21/01/2017	48,83	26,14
6,01	21/01/2017	198,83	176,14
8	23/01/2017	186,70	39,64
8,01	23/01/2017	186,70	189,64
9	24/01/2017	126,90	14,92
9,01	24/01/2017	126,90	164,92
10	25/01/2017	122,50	40,24
10,01	25/01/2017	122,50	190,24
11	26/01/2017	97,87	77,05
11,01	26/01/2017	247,87	227,05
12	27/01/2017	179,40	117,90
12,01	27/01/2017	179,40	267,90
15	30/01/2017	99,99	107,40

Table 44. Carbohydrates results

12h light		
Day	**N-limited**	**P-limited**
	mg/L	**mg/L**
1	26,018	30,483
1,5	76,576	47,308
2	196,084	86,551
2,5	135,725	109,795
3	115,377	66,862
3,5	135,236	64,134
4	144,668	114,689
4,5	289,991	124,345
5	249,408	112,236
5,5	375,600	124,978
7	338,846	156,156
8	371,633	120,999
9	348,670	169,944
10	472,091	163,119
11	595,917	127,137
12	838,056	432,131
15	430,112	321,478

24h light		
Day	**N-limited**	**P-limited**
	mg/L	**mg/L**
1	70,44	95,93
2	118,86	127,58
3	203,92	166,33
4	175,62	134,89
5	131,27	214,41
6	374,40	386,81
8	641,31	552,07
9	464,72	496,49
10	335,99	472,93
11	386,03	479,51
12	391,03	662,38
15	463,11	585,27

Table 45. PHB results

	12h light	
Day	**N-limited**	**P-limited**
	mg/L	**mg/L**
1	12,304	16,346
2	18,312	22,460
2,5	27,855	29,277
3	29,932	30,499
3,5	27,874	25,202
4	29,875	27,308
4,5	28,677	32,015
5	34,427	26,906
5,5	30,970	23,679
7	29,278	31,275
8	41,590	29,443
9	48,911	47,198
10	41,253	42,627
11	52,501	46,341
12	61,614	64,584
15	55,957	76,361

	24h light	
Day	**N-limited**	**P-limited**
	mg/L	**mg/L**
1	24,308	23,712
2	32,966	39,224
3	45,745	38,555
4	32,516	51,040
5	29,882	34,674
8	40,590	104,234
9	51,635	90,145
10	47,121	68,564
11	46,088	72,447
12	51,187	64,147
15	49,953	66,351

A1.2.2. Selection of Cyanobacteria

Table 46. Volatile and total suspended solids results

Day	**Continue**		**Semi-Continue**		**SBR1**		**SBR1**	
	TSS	**VSS**	**TSS**	**VSS**	**TSS**	**VSS**	**TSS**	**VSS**
[-]	[g/L]	[g/L]	[g/L]	[g/L]	[g/L]	[g/L]	[g/L]	[g/L]
1	0,267	0,207	0,267	0,207	0,267	0,207	0,267	0,207
3	0,341	0,241	0,373	0,269	0,383	0,273	0,488	0,337
6	0,403	0,315	0,279	0,175	0,175	0,129	0,297	0,219
8	0,389	0,229	0,360	0,315	0,445	0,400	0,843	0,596
10	0,256	0,212	0,451	0,425	0,660	0,513	0,708	0,624
13	0,133	0,193	0,312	0,388	0,693	0,204	0,626	0,652
15	0,220	0,215	0,452	0,404	0,891	0,620	0,644	0,500
17	0,301	0,259	0,521	0,451	1,021	0,785	0,776	0,584
20	0,428	0,357	0,548	0,459	0,959	0,749	0,636	0,504
22	0,589	0,299	0,421	0,376	0,960	0,781	0,551	0,477
24	0,333	0,295	0,551	0,407	1,049	0,792	0,712	0,557
27	0,336	0,303	0,551	0,443	1,213	0,913	0,500	0,435
29	0,675	0,615	0,476	0,417	0,996	0,691	0,645	0,511

Table 47. Chlorophyll results

	Continue	Semi-Continue	SBR1	SBR2
Day	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	0,633	0,633	0,633	0,633
3	0,520	0,718	0,741	0,859
5	0,279	-	0,269	0,151
7	0,529	0,613	0,619	1,051
10	0,255	0,533	0,977	0,965
13	0,388	0,775	1,045	0,947
15	0,381	0,361	1,347	1,053
20	0,125	0,406	1,322	0,560
22	0,174	0,456	1,889	0,962
27	0,422	0,727	2,501	0,876
30	0,813	0,754	2,819	1,066

Table 48. Anions results

Day	**Continuous**					
	Nitrites [NO$_2^-$]	**N- NO$_2^-$**	**Nitrates [NO$_3^-$]**	**N- NO$_3^-$**	**Orthophosphates [PO$_4^{3-}$]**	**P- PO$_4^{3-}$**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	14,679	4,468	22,185	5,010	0,000	0,000
3	9,934	3,023	49,071	11,081	0,000	0,000
6	16,932	5,153	54,863	12,388	0,000	0,000
8	17,900	5,448	56,081	12,663	0,000	0,000
10	2,169	0,660	30,112	6,799	11,482	3,744
17	6,868	2,090	23,947	5,407	0,000	0,000
20	15,060	4,583	22,403	5,059	0,000	0,000
21	15,869	4,830	24,754	5,590	--	--
24	22,479	6,841	34,977	7,898	8,074	2,633
27	15,469	4,708	30,408	6,866	5,974	1,948
32	0,000	0,000	32,010	7,228	5,254	1,713
Day	**Semi-continuous**					
	Nitrites [NO$_2^-$]	**N- NO$_2^-$**	**Nitrates [NO$_3^-$]**	**N- NO$_3^-$**	**Orthophosphates [PO$_4^{3-}$]**	**P- PO$_4^{3-}$**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	14,679	4,468	22,185	5,010	0	0,000
3	12,785	3,891	55,723	12,583	0	0,000
6	23,928	7,282	81,706	18,450	5,899	1,924
8	21,869	6,656	101,704	22,965	0	0,000
10	3,788	1,153	88,592	20,005	0	0,000
17	37,525	11,421	0,000	0,000	0,000	0,000

20	10,718	3,262	40,427	9,129	5,176	1,688
21	9,402	2,861	52,726	11,906	7,594	2,476
24	5,83	1,773	44,00	9,936	6,59	2,149
27	0,00	0,000	47,62	10,754	6,25	2,037
32	5,66	1,722	39,78	8,983	7,55	2,461

Day	**SBR 1**					
	Nitrites $[NO_2^-]$	**N- NO_2^-**	**Nitrates $[NO_3^-]$**	**N- NO_3^-**	**Orthophosphates $[PO_4^{3-}]$**	**P- PO_4^{3-}**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	14,679	4,468	22,185	5,010	0,000	0,000
3	9,260	2,818	68,759	15,526	0,000	0,000
6	50,759	15,448	63,565	14,353	0,000	0,000
8	0,000	0,000	0,000	0,000	0,000	0,000
10	0,000	0,000	0,000	0,000	0,000	0,000
17	21,399	6,513	33,469	7,558	11,101	3,620
20	15,764	4,798	53,820	12,153	0,000	0,000
21	12,496	3,803	45,969	10,380	6,068	1,979
24	18,607	5,663	42,468	9,590	6,627	2,161
27	6,084	1,852	71,389	16,120	7,357	2,399
32	0,000	0,000	27,320	6,169	4,905	1,600

Day	**SBR 2**					
	Nitrites $[NO_2^-]$	**N- NO_2^-**	**Nitrates $[NO_3^-]$**	**N- NO_3^-**	**Orthophosphates $[PO_4^{3-}]$**	**P- PO_4^{3-}**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	14,679	4,468	22,185	5,010	0,000	0,000
3	10,124	3,081	77,607	17,524	0,000	0,000
6	54,811	16,682	54,445	12,294	3,806	1,241
8	0,000	0,000	0,000	0,000	0,000	0,000
10	0,000	0,000	0,000	0,000	0,000	0,000
17	35,014	10,656	53,736	12,134	0,000	0,000
20	4,552	1,385	85,971	19,413	3,830	1,249
21	12,496	3,803	45,969	10,380	6,068	1,979
24	14,800	4,504	44,600	10,071	6,230	2,032
27	16,771	5,104	42,227	9,535	6,166	2,011
32	23,459	27,731	91,117	20,575	6,471	2,110

Table 49. Total Ammoniacal nitrogen results

Day	Continue	Semi-Continue	SBR1	SBR2
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	1,390	1,390	1,390	1,390
3	3,505	8,249	7,496	4,796
6	5,887	0,697	5,489	1,872
8	1,521	0,470	2,979	0,075
10	4,640	0,948	0,016	0,016
17	2,860	0,587	9,540	2,024
20	6,433	0,784	0,099	0,267
21	17,498	2,908	11,607	11,368
24	0,105	2,322	1,987	2,728
27	15,336	13,256	0,207	4,401
32	0,434	13,447	0,002	2,095

Table 50. TOC, IC, TC, TN total

Day	**Continuo**				**Semi-Continuo**				**SBR1**				**SBR2**			
	TOC	**IC**	**TC**	**TN**	**TOC**	**IC**	**TC**	**TN**	**TOC**	**IC**	**TC**	**TN**	**TOC**	**IC**	**TC**	**TN**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
2	125,40	21,98	147,40	47,89	155,50	24,12	179,60	54,19	161,80	30,08	191,90	56,09	184,80	33,36	218,10	61,83
7	280,90	39,60	320,50	54,13	368,80	7,39	376,20	72,11	275,70	84,20	359,90	78,11	366,90	71,28	438,20	84,98
9	275,60	25,07	300,60	4,98	378,40	35,17	413,60	67,51	373,20	33,27	406,50	81,42	443,20	34,95	478,20	85,18
14	284,10	17,00	301,10	48,61	391,70	19,79	411,50	70,66	450,10	37,27	487,40	87,61	465,30	30,88	496,20	91,01
16	274,70	6,03	280,70	46,40	363,00	16,75	379,80	60,68	403,40	95,47	498,80	94,16	390,90	105,00	495,90	87,73
22	318,00	14,83	332,80	59,70	393,80	24,60	418,40	66,70	434,10	52,86	487,00	113,10	42,66	66,64	489,20	86,30
29	356,70	13,97	370,70	54,87	418,40	11,82	430,20	58,13	407,60	36,38	444,00	104,10	441,60	45,61	487,20	83,40

Table 51. TOC, IC, TC, TN soluble

Day	Continuo				Semi-Continuo				SBR1				SBR2			
	TOC	IC	TC	TN	TOC	IC	TC	TN	TOC	IC	TC	TN	TOC	IC	TC	TN
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
2	20,00	26,21	46,21	35,35	21,38	21,23	42,61	35,22	23,34	23,48	46,81	31,41	24,40	26,43	50,83	35,69
7	71,52	39,68	111,20	32,63	48,15	5,84	53,99	45,63	119,00	73,91	192,90	57,16	65,21	59,24	124,40	57,49
8	57,72	30,93	88,65	25,62	47,71	169,42	67,13	43,22	83,72	50,52	134,24	54,40	62,62	42,58	106,67	56,28
9	43,92	22,18	66,10	18,61	47,26	33,00	80,27	40,81	48,44	27,13	75,57	51,63	60,03	25,91	88,94	55,06
13	46,12	20,72	66,84	20,39	43,88	75,36	69,25	36,62	44,50	27,52	72,02	47,54	50,03	24,55	76,08	51,81
14	48,32	19,26	67,58	22,17	40,50	17,72	58,22	32,42	40,56	27,90	68,47	43,44	40,03	23,18	63,21	48,55
15	50,33	62,32	62,64	22,55	41,49	66,70	58,18	31,49	48,47	103,16	101,64	41,27	45,78	105,79	101,56	48,95
16	52,33	5,37	57,70	22,92	42,47	15,67	58,14	30,56	56,37	78,42	134,80	39,09	51,53	88,40	139,90	49,34
17	51,36	73,73	75,09	26,22	48,11	65,79	63,89	30,93	54,16	53,49	107,66	37,72	54,18	76,46	130,60	46,92
20	50,39	42,09	92,48	29,51	53,74	15,90	69,64	31,30	51,95	28,56	80,51	36,34	56,83	64,51	121,30	44,50
22	61,42	14,25	75,67	28,13	62,08	19,20	81,28	29,43	42,32	41,95	84,27	33,41	45,04	55,23	100,30	46,51
23	51,37	59,03	60,40	25,16	57,02	63,37	70,39	27,92	45,84	31,88	77,71	33,07	45,07	49,65	94,73	40,65
27	41,31	3,81	45,12	22,18	51,95	7,53	59,49	26,41	49,35	21,81	71,15	32,72	45,10	44,06	89,16	34,78
29	63,37	13,16	76,54	20,14	52,05	10,76	62,81	22,46	48,93	28,21	77,14	34,45	47,79	36,93	84,72	38,55

Table 52. PHBs results

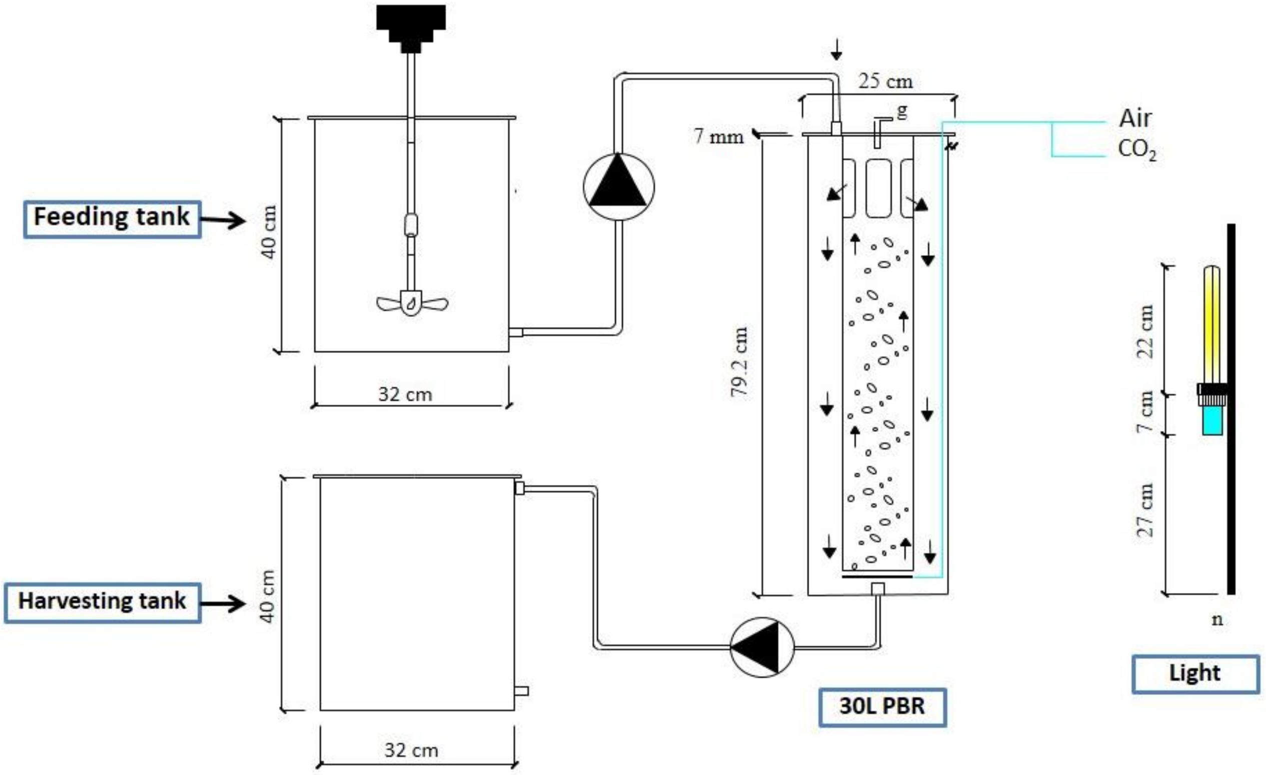
Day	**Continue**	**Semi-Continue**	**SBR1**	**SBR2**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	0,781	1,147	0,757	0,512
2	1,035	0,713	1,202	0,603
7	1,523	1,092	1,135	2,181
9	1,589	1,321	2,092	2,257
14	0,461	1,667	4,440	1,278
17	0,567	0,859	6,590	1,881
21	2,035	2,290	2,259	1,394
23	1,497	1,173	2,259	1,316
28	1,207	0,987	2,766	1,303
30	1,814	1,275	2,611	1,352

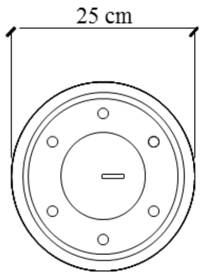
Table 53. Carbohydrates results

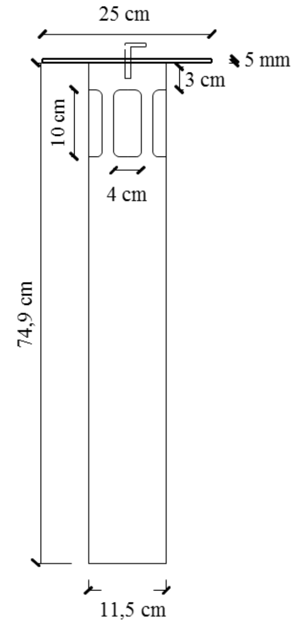
Day	**Continue**	**Semi-Continue**	**SBR1**	**SBR2**
[-]	[mg/L]	[mg/L]	[mg/L]	[mg/L]
1	30,468	34,377	25,577	23,915
2	28,326	33,199	23,397	41,128
7	78,773	39,746	32,192	53,651
9	48,452	58,475	90,792	123,906
14	17,461	41,838	101,891	42,124
17	24,356	62,973	140,404	63,237
21	57,005	43,921	121,940	53,254
23	72,872	53,398	112,521	73,957
28	57,126	47,554	139,559	59,280
30	57,826	44,667	79,965	51,169

Annex 2: Reactor plans

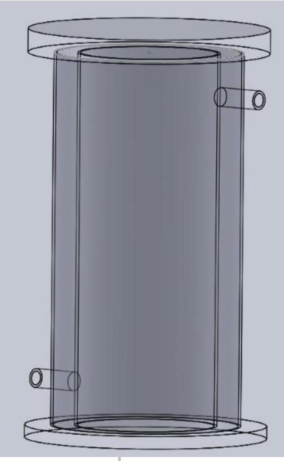
A2.1 30L Photobioreactor

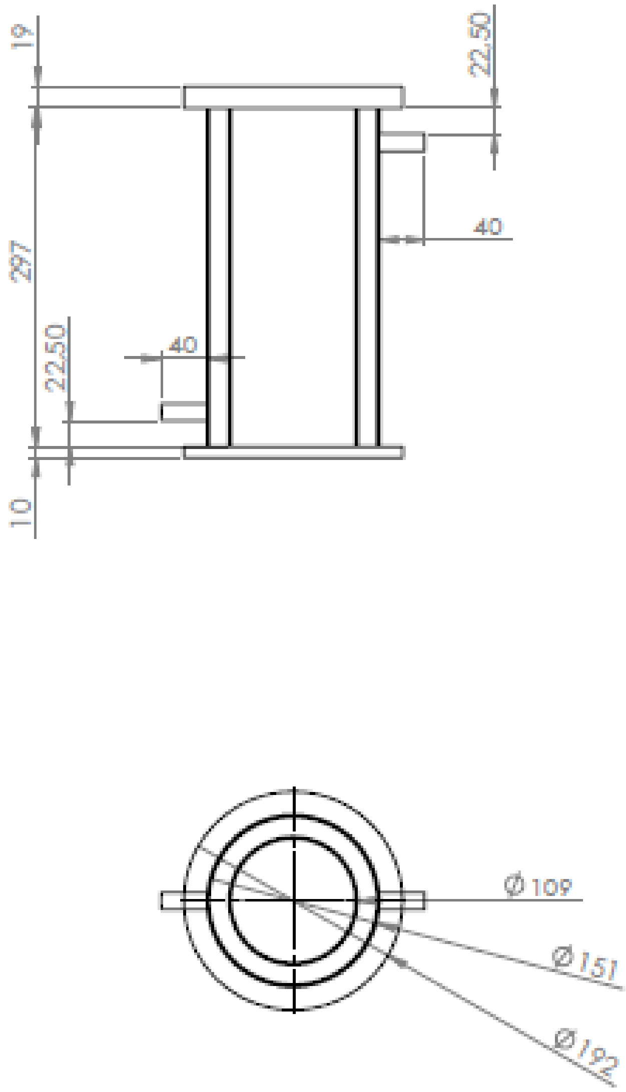






A2.2 3L Vertical cylinder reactors

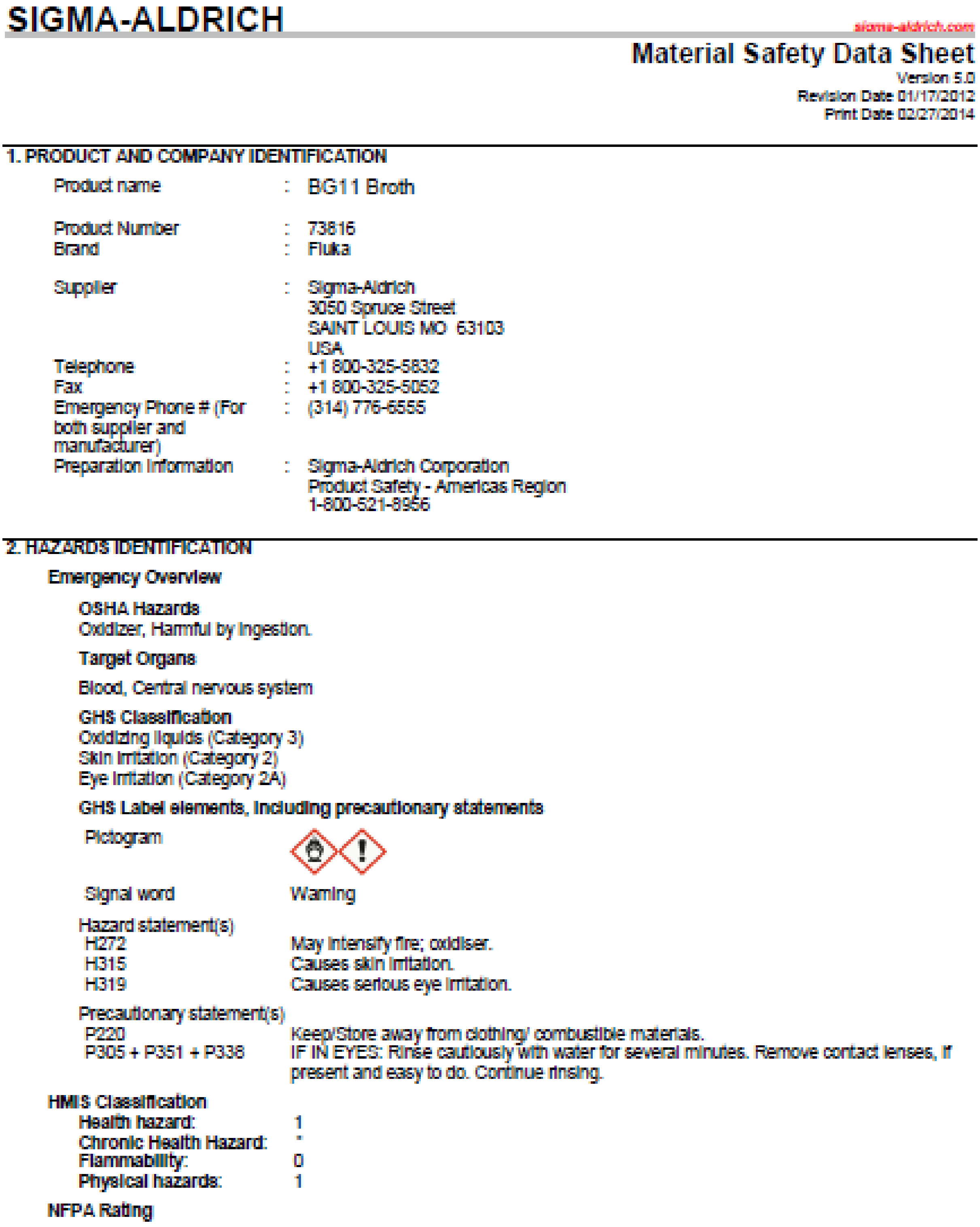




Name: 3L vertical cylinder PBR	ESCALA:1:5	Units: mm

Annex 3. Safety data sheets

A3.1. BG11 Broth



SIGMA-ALDRICH

sigma-aldrich.com

Material Safety Data Sheet

Version 5.0
Revision Date 01/17/2012
Print Date 02/27/2014

1. PRODUCT AND COMPANY IDENTIFICATION

Product name	:	BG11 Broth
Product Number	:	73816
Brand	:	Fluka
Supplier	:	Sigma-Aldrich 3050 Spruce Street SAINT LOUIS MO 63103 USA
Telephone	:	+1 800-325-5832
Fax	:	+1 800-325-5052
Emergency Phone # (For both supplier and manufacturer)	:	(314) 776-6555
Preparation Information	:	Sigma-Aldrich Corporation Product Safety - Americas Region 1-800-521-8956

2. HAZARDS IDENTIFICATION

Emergency Overview

OSHA Hazards
Oxidizer, Harmful by ingestion.

Target Organs

Blood, Central nervous system

GHS Classification
Oxidizing liquids (Category 3)
Skin irritation (Category 2)
Eye irritation (Category 2A)

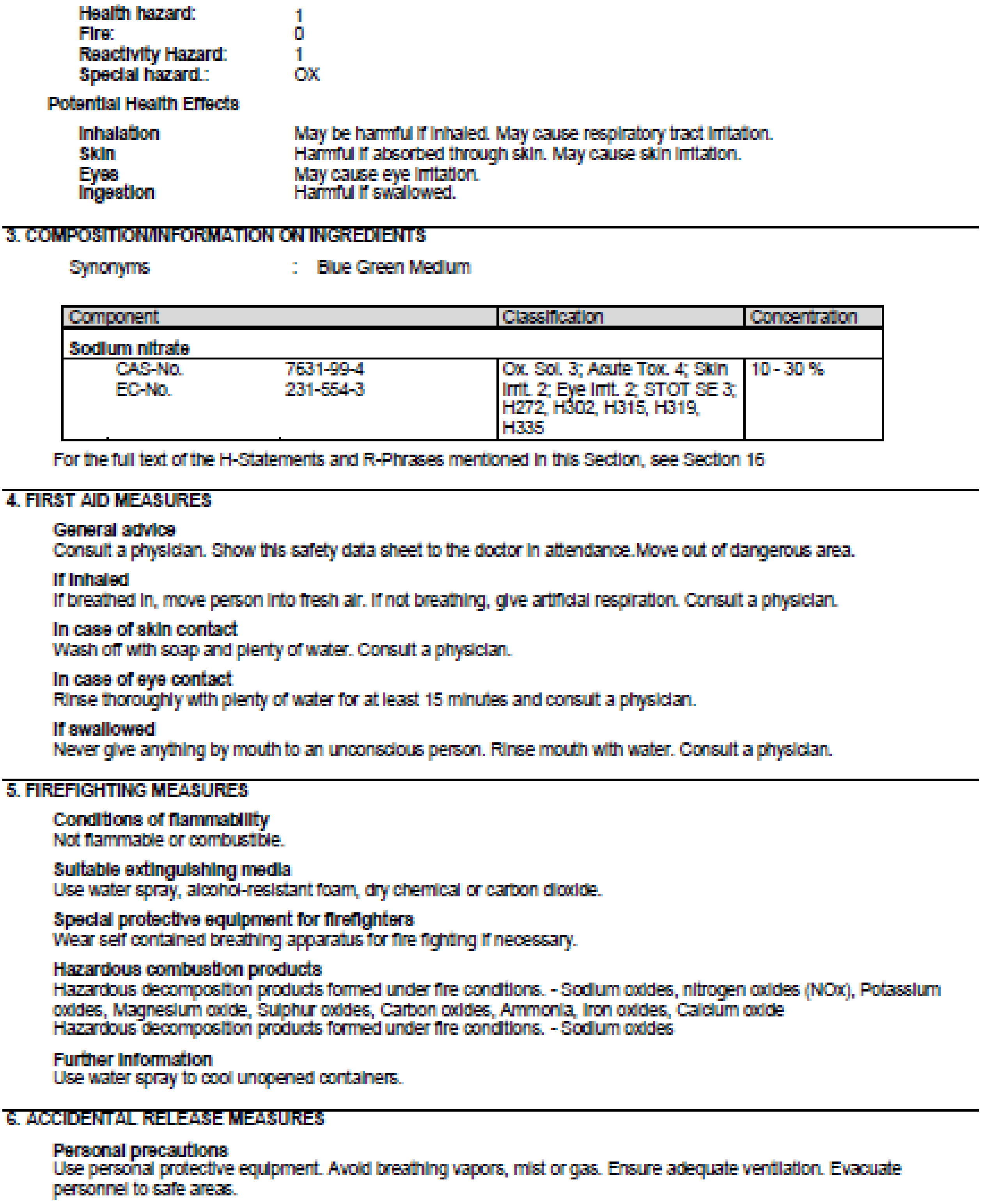
GHS Label elements, including precautionary statements

Pictogram	
Signal word	Warning
Hazard statement(s)	
H272	May intensify fire; oxidiser.
H315	Causes skin irritation.
H319	Causes serious eye irritation.
Precautionary statement(s)	
P220	Keep/Store away from clothing/ combustible materials.
P305 + P351 + P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

HMIS Classification

Health hazard:	1
Chronic Health Hazard:	*
Flammability:	0
Physical hazards:	1

NFPA Rating



Health hazard:	1
Fire:	0
Reactivity Hazard:	1
Special hazard.:	OX

Potential Health Effects

Inhalation	May be harmful if inhaled. May cause respiratory tract irritation.
Skin	Harmful if absorbed through skin. May cause skin irritation.
Eyes	May cause eye irritation.
Ingestion	Harmful if swallowed.

3. COMPOSITION/INFORMATION ON INGREDIENTS

Synonyms : Blue Green Medium

Component		Classification	Concentration
Sodium nitrate			
CAS-No. EC-No.	7631-99-4 231-554-3	Ox. Sol. 3; Acute Tox. 4; Skin Irrit. 2; Eye Irrit. 2; STOT SE 3; H272, H302, H315, H319, H335	10 - 30 %

For the full text of the H-Statements and R-Phrases mentioned in this Section, see Section 16

4. 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
Rinse thoroughly with plenty of water for at least 15 minutes and consult a physician.

If swallowed
Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician.

5. FIREFIGHTING MEASURES

Conditions of flammability
Not flammable or combustible.

Suitable extinguishing media
Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide.

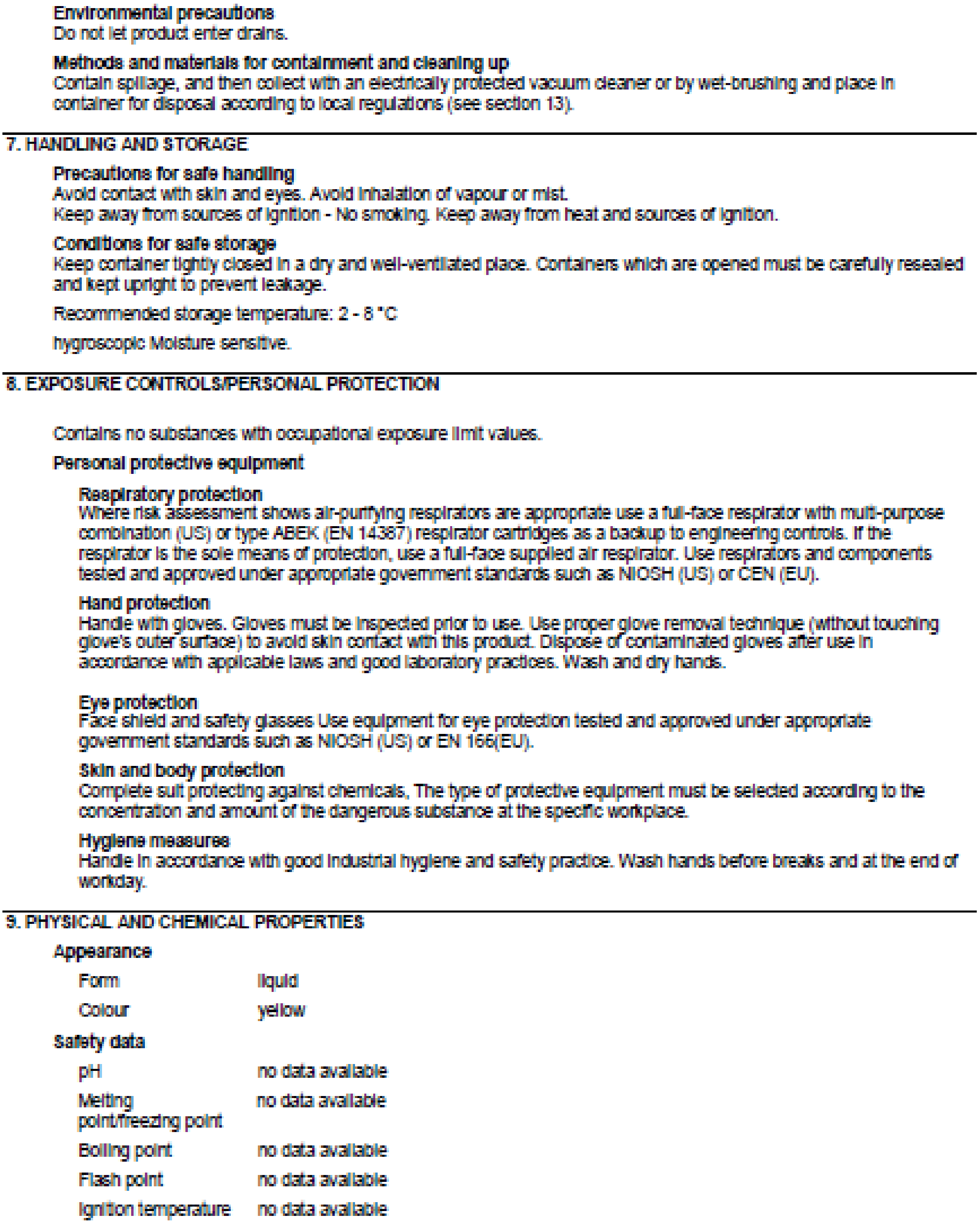
Special protective equipment for firefighters
Wear self contained breathing apparatus for fire fighting if necessary.

Hazardous combustion products
Hazardous decomposition products formed under fire conditions. - Sodium oxides, nitrogen oxides (NOx), Potassium oxides, Magnesium oxide, Sulphur oxides, Carbon oxides, Ammonia, Iron oxides, Calcium oxide
Hazardous decomposition products formed under fire conditions. - Sodium oxides

Further information
Use water spray to cool unopened containers.

6. ACCIDENTAL RELEASE MEASURES

Personal precautions
Use personal protective equipment. Avoid breathing vapors, mist or gas. Ensure adequate ventilation. Evacuate personnel to safe areas.



Environmental precautions
Do not let product enter drains.

Methods and materials for containment and cleaning up
Contain spillage, and then collect with an electrically protected vacuum cleaner or by wet-brushing and place in container for disposal according to local regulations (see section 13).

7. HANDLING AND STORAGE

Precautions for safe handling
Avoid contact with skin and eyes. Avoid inhalation of vapour or mist.
Keep away from sources of ignition - No smoking. Keep away from heat and sources of ignition.

Conditions for safe storage
Keep container tightly closed in a dry and well-ventilated place. Containers which are opened must be carefully resealed and kept upright to prevent leakage.

Recommended storage temperature: 2 - 8 °C

hygroscopic Moisture sensitive.

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Contains no substances with occupational exposure limit values.

Personal protective equipment

Respiratory protection
Where risk assessment shows air-purifying respirators are appropriate use a full-face respirator with multi-purpose combination (US) or type ABEK (EN 14387) 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).

Hand 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.

Eye 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 and 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.

Hygiene measures
Handle in accordance with good industrial hygiene and safety practice. Wash hands before breaks and at the end of workday.

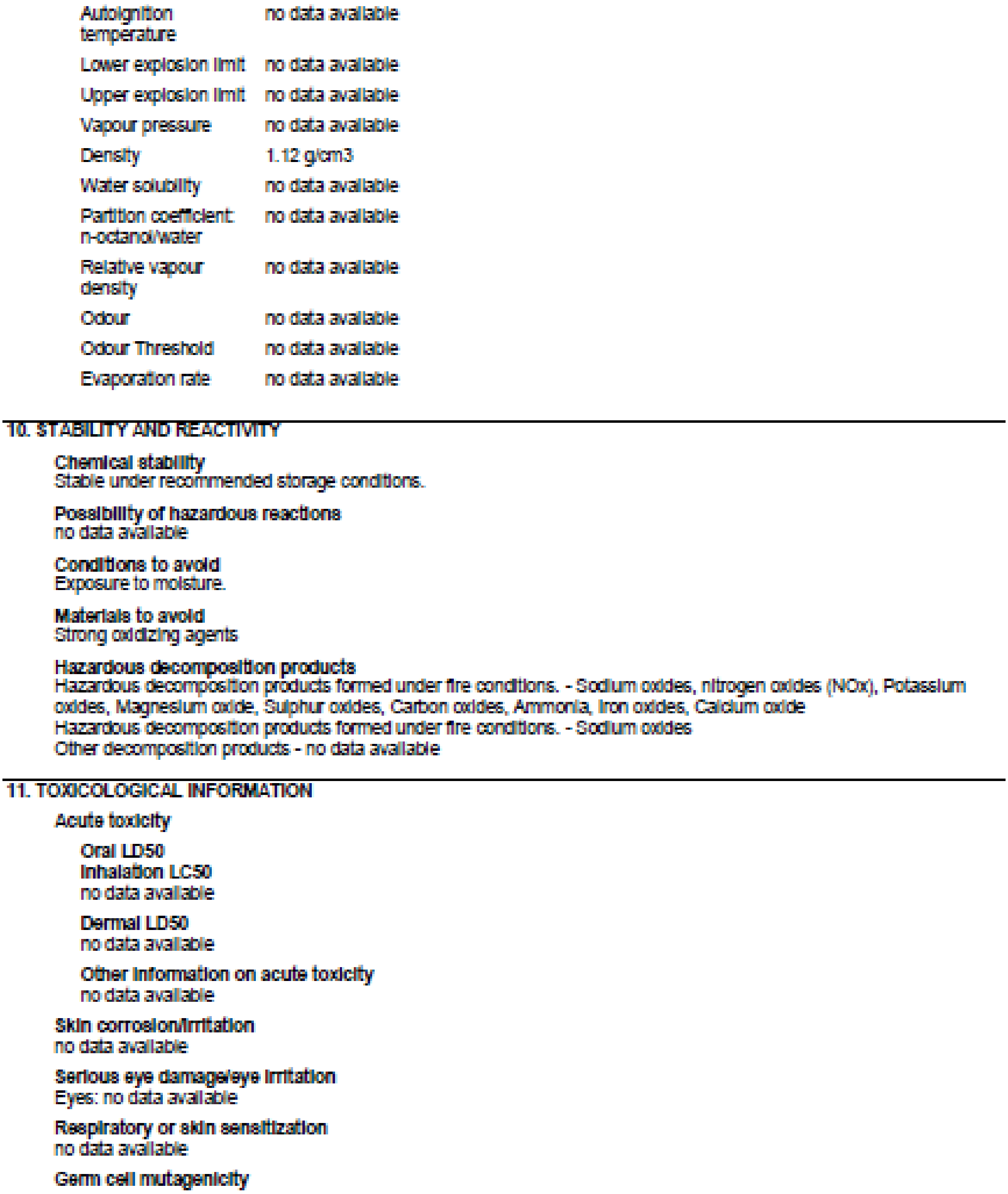
9. PHYSICAL AND CHEMICAL PROPERTIES

Appearance

Form	liquid
Colour	yellow

Safety data

pH	no data available
Melting point/freezing point	no data available
Boiling point	no data available
Flash point	no data available
Ignition temperature	no data available



Autoignition temperature	no data available
Lower explosion limit	no data available
Upper explosion limit	no data available
Vapour pressure	no data available
Density	1.12 g/cm3
Water solubility	no data available
Partition coefficient: n-octanol/water	no data available
Relative vapour density	no data available
Odour	no data available
Odour Threshold	no data available
Evaporation rate	no data available

10. STABILITY AND REACTIVITY

Chemical stability
Stable under recommended storage conditions.

Possibility of hazardous reactions
no data available

Conditions to avoid
Exposure to moisture.

Materials to avoid
Strong oxidizing agents

Hazardous decomposition products
Hazardous decomposition products formed under fire conditions. - Sodium oxides, nitrogen oxides (NOx), Potassium oxides, Magnesium oxide, Sulphur oxides, Carbon oxides, Ammonia, Iron oxides, Calcium oxide
Hazardous decomposition products formed under fire conditions. - Sodium oxides
Other decomposition products - no data available

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Oral LD50
Inhalation LC50
no data available

Dermal LD50
no data available

Other information on acute toxicity
no data available

Skin corrosion/irritation
no data available

Serious eye damage/eye irritation
Eyes: no data available

Respiratory or skin sensitization
no data available

Germ cell mutagenicity

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

no data available

Teratogenicity

no data available

Specific target organ toxicity - single exposure (Globally Harmonized System)
no data available

Specific target organ toxicity - repeated exposure (Globally Harmonized System)
no data available

Aspiration hazard
no data available

Potential health effects

Inhalation	May be harmful if inhaled. May cause respiratory tract irritation.
Ingestion	Harmful if swallowed.
Skin	Harmful if absorbed through skin. May cause skin irritation.
Eyes	May cause eye irritation.

Signs and Symptoms of Exposure
To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.

Synergistic effects
no data available

Additional Information
RTECS: Not available

12. ECOLOGICAL INFORMATION

Toxicity

no data available

Persistence and degradability
no data available

Bioaccumulative potential
no data available

Mobility in soil
no data available

PBT and vPvB assessment
no data available

Other adverse effects

no data available

13. DISPOSAL CONSIDERATIONS

Product
Burn in a chemical incinerator equipped with an afterburner and scrubber but exert extra care in igniting as this material is highly flammable. Offer surplus and non-recyclable solutions to a licensed disposal company. Contact a licensed professional waste disposal service to dispose of this material.

Contaminated packaging
Dispose of as unused product.

14. TRANSPORT INFORMATION

DOT (US)
UN number: 3139 Class: 5.1 Packing group: III
Proper shipping name: Oxidizing liquid, n.o.s. (Sodium nitrate)
Marine pollutant: No
Poison Inhalation Hazard: No

IMDG
UN number: 3139 Class: 5.1 Packing group: III EMS-No: F-A, S-Q
Proper shipping name: OXIDIZING LIQUID, N.O.S. (Sodium nitrate)
Marine pollutant: No

IATA
UN number: 3139 Class: 5.1 Packing group: III
Proper shipping name: Oxidizing liquid, n.o.s. (Sodium nitrate)

15. REGULATORY INFORMATION

OSHA Hazards
Oxidizer, Harmful by ingestion.

SARA 302 Components
SARA 302: No chemicals in this material are subject to the reporting requirements of SARA Title III, Section 302.

SARA 313 Components
SARA 313: 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
Reactivity Hazard, Acute Health Hazard

Massachusetts Right To Know Components

	CAS-No.	Revision Date
Sodium nitrate	7631-99-4	1993-04-24

Pennsylvania Right To Know Components

	CAS-No.	Revision Date
Water	7732-18-5	
Ammonium iron(III) citrate	1185-57-5	1993-04-24
Sodium nitrate	7631-99-4	1993-04-24

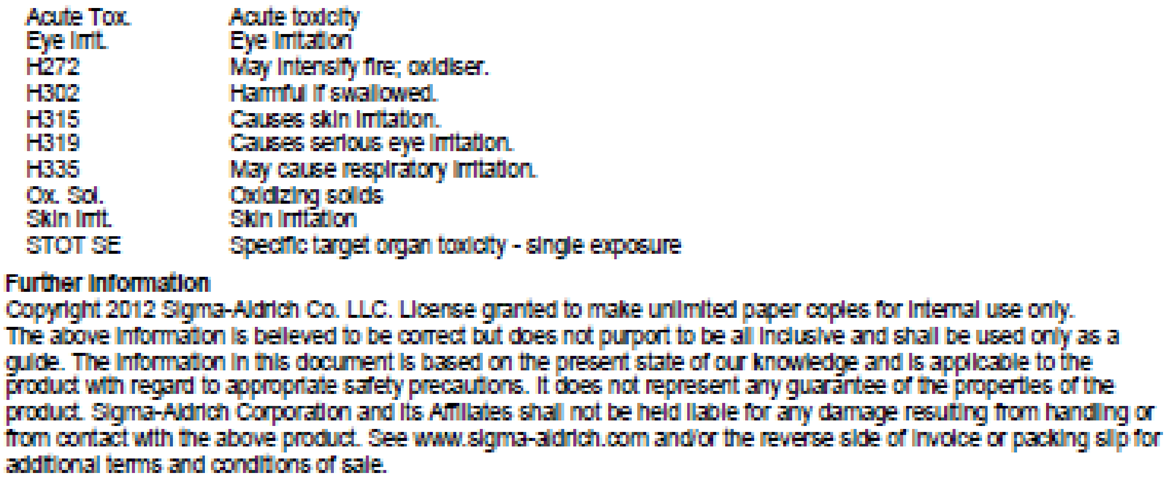
New Jersey Right To Know Components

	CAS-No.	Revision Date
Water	7732-18-5	
Sodium nitrate	7631-99-4	1993-04-24

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

Text of H-code(s) and R-phrase(s) mentioned in Section 3



Acute Tox.	Acute toxicity
Eye Irrit.	Eye irritation
H272	May intensify fire; oxidiser.
H302	Harmful if swallowed.
H315	Causes skin irritation.
H319	Causes serious eye irritation.
H335	May cause respiratory irritation.
Ox. Sol.	Oxidizing solids
Skin Irrit.	Skin irritation
STOT SE	Specific target organ toxicity - single exposure

Further information

A3.2. Potassium dichromate

SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Revision Date 19.10.2016 Version 13.1

SECTION 1. Identification of the substance/mixture and of the company/undertaking

1.1 Product identifier

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure
REACH Registration Number	01-2119454792-32-XXXX
CAS-No.	7778-50-9

1.2 Relevant identified uses of the substance or mixture and uses advised against

Identified uses	Materials for use in technical applications For additional information on uses please refer to the Merck Chemicals portal (www.merckgroup.com).

1.3 Details of the supplier of the safety data sheet

Company	Merck KGaA * 64271 Darmstadt * Germany * Phone:+49 6151 72-0
Responsible Department	EQ-RS * e-mail: prodsafe@merckgroup.com

1.4 Emergency telephone number	**Please contact the regional company representation in your country.**

SECTION 2. Hazards identification

2.1 Classification of the substance or mixture

Classification (REGULATION (EC) No 1272/2008)

SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Oxidizing solid, Category 2, H272

Acute toxicity, Category 3, Oral, H301

Acute toxicity, Category 2, Inhalation, H330

Acute toxicity, Category 4, Dermal, H312

Skin corrosion, Category 1B, H314

Respiratory sensitisation, Category 1, H334

Skin sensitisation, Category 1, H317

Germ cell mutagenicity, Category 1B, H340

Carcinogenicity, Category 1B, H350

Reproductive toxicity, Category 1B, H360FD

Specific target organ toxicity - single exposure, Category 3, Respiratory system, H335

Specific target organ toxicity - repeated exposure, Category 1, H372

Acute aquatic toxicity, Category 1, H400

Chronic aquatic toxicity, Category 1, H410

For the full text of the H-Statements mentioned in this Section, see Section 16.

2.2 Label elements

Labelling (REGULATION (EC) No 1272/2008)

Hazard pictograms

Signal word

Danger

Hazard statements

H340 May cause genetic defects.

H350 May cause cancer.

H360FD May damage fertility. May damage the unborn child.

H272 May intensify fire; oxidizer.

H301 Toxic if swallowed.

H312 Harmful in contact with skin.

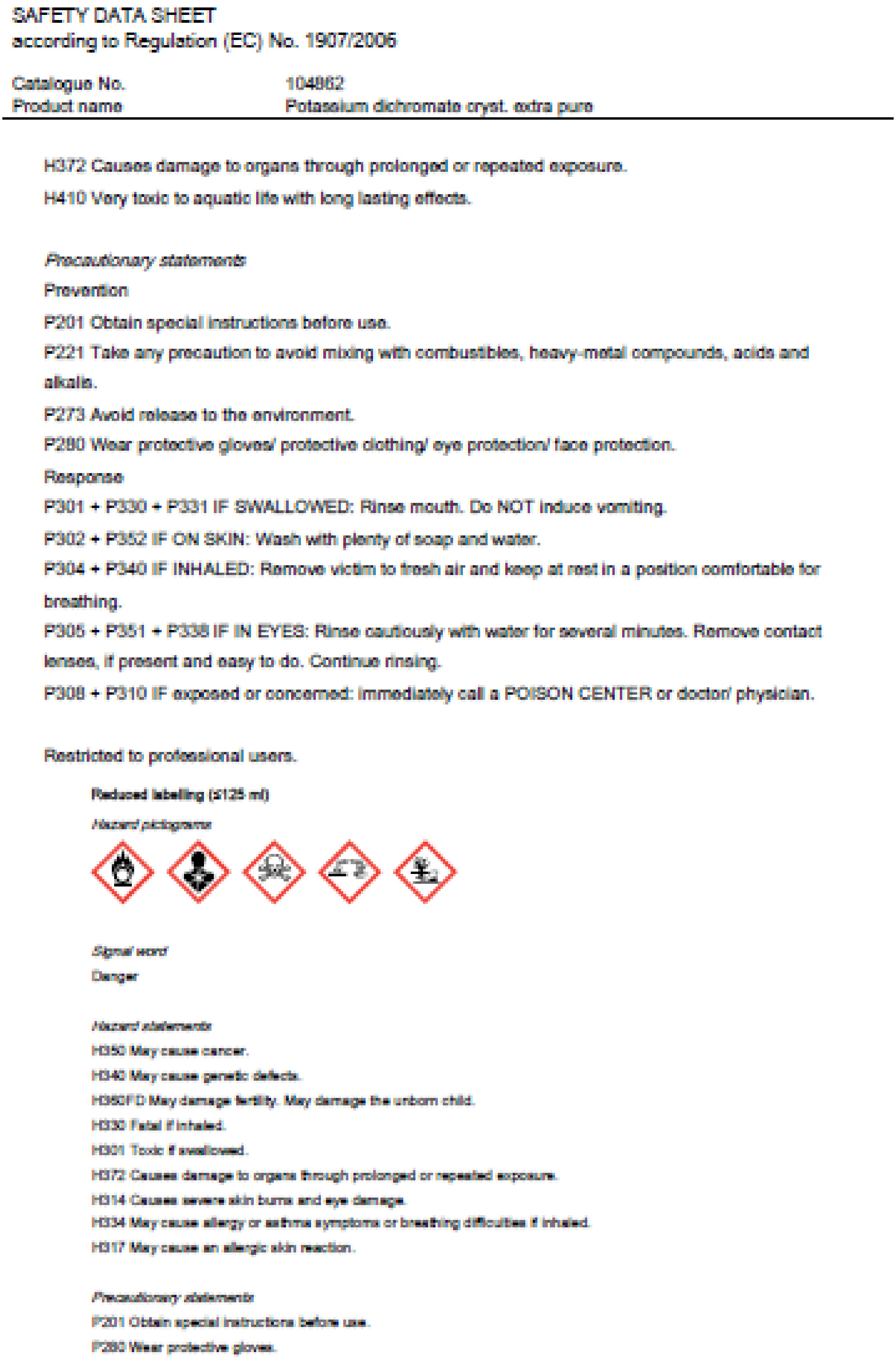
H314 Causes severe skin burns and eye damage.

H317 May cause an allergic skin reaction.

H330 Fatal if inhaled.

H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.

H335 May cause respiratory irritation.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

H372 Causes damage to organs through prolonged or repeated exposure.

H410 Very toxic to aquatic life with long lasting effects.

Precautionary statements

Prevention

P201 Obtain special instructions before use.

P221 Take any precaution to avoid mixing with combustibles, heavy-metal compounds, acids and alkalis.

P273 Avoid release to the environment.

P280 Wear protective gloves/ protective clothing/ eye protection/ face protection.

Response

P301 + P330 + P331 IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.

P302 + P352 IF ON SKIN: Wash with plenty of soap and water.

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.

P308 + P310 IF exposed or concerned: Immediately call a POISON CENTER or doctor/ physician.

Restricted to professional users.

Reduced labelling (≤125 ml)

Hazard pictograms

Signal word

Danger

Hazard statements

H350 May cause cancer.

H340 May cause genetic defects.

H360FD May damage fertility. May damage the unborn child.

H330 Fatal if inhaled.

H301 Toxic if swallowed.

H372 Causes damage to organs through prolonged or repeated exposure.

H314 Causes severe skin burns and eye damage.

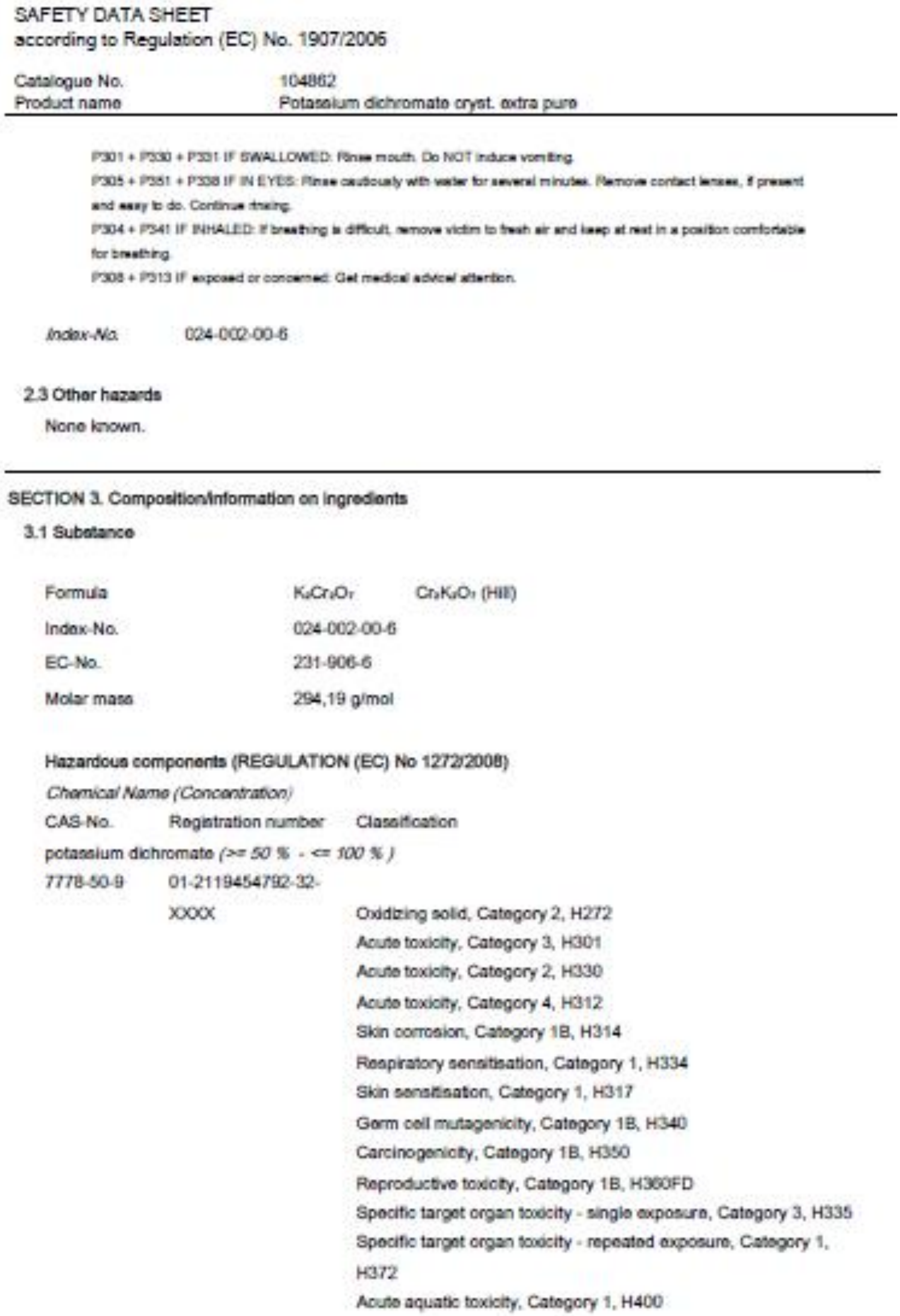
H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.

H317 May cause an allergic skin reaction.

Precautionary statements

P201 Obtain special instructions before use.

P280 Wear protective gloves.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

P301 + P330 + P331 IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.

P305 + P351 + P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

P304 + P341 IF INHALED: If breathing is difficult, remove victim to fresh air and keep at rest in a position comfortable for breathing.

P308 + P313 IF exposed or concerned: Get medical advice/ attention.

Index-No. 024-002-00-6

2.3 Other hazards

None known.

SECTION 3. Composition/information on ingredients

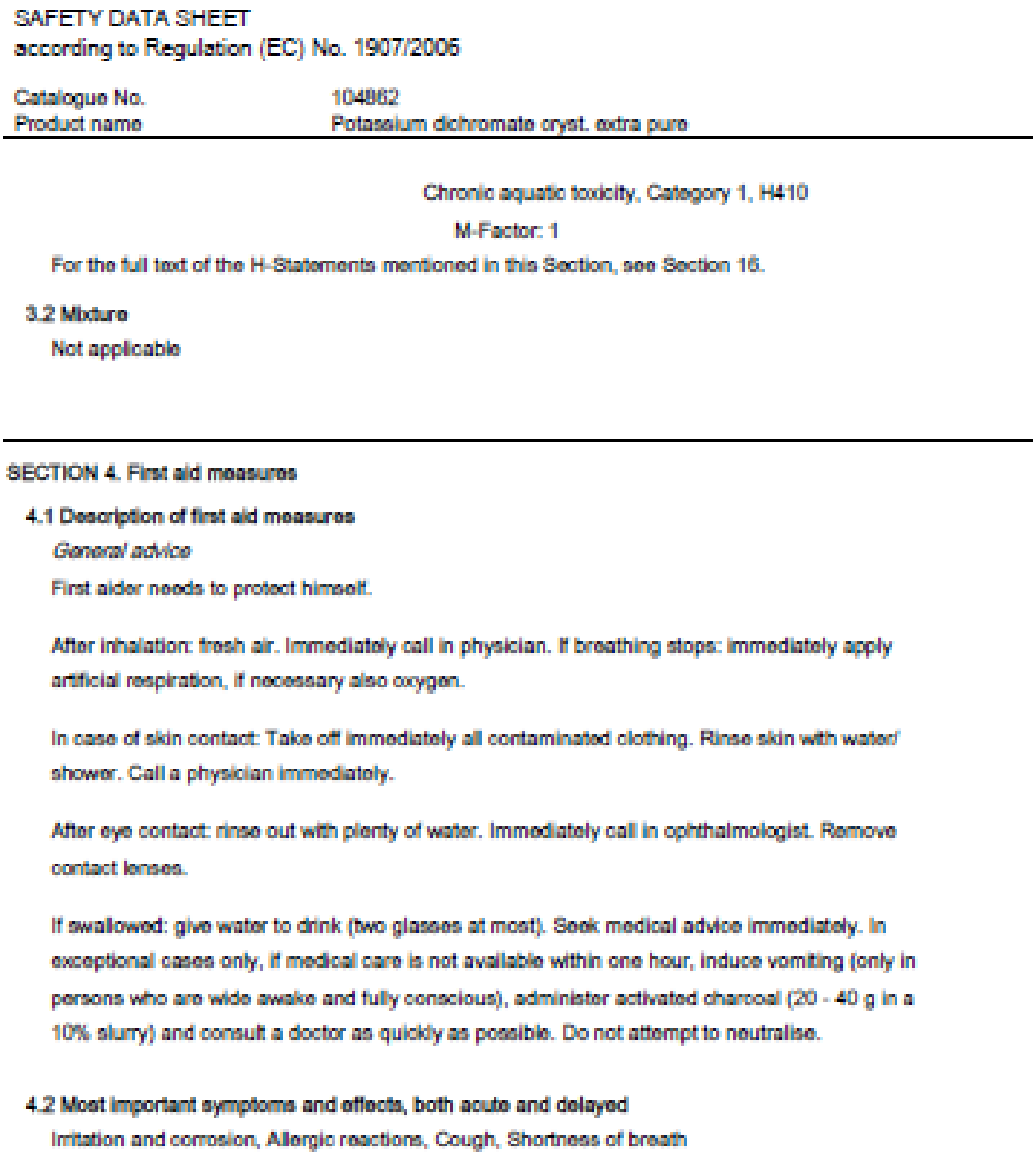
3.1 Substance

Formula	$K_2Cr_2O_7$	$Cr_2K_2O_7$ (Hill)
Index-No.	024-002-00-6	
EC-No.	231-906-6	
Molar mass	294,19 g/mol	

Hazardous components (REGULATION (EC) No 1272/2008)

Chemical Name (Concentration)

CAS-No.	Registration number	Classification
potassium dichromate *(>= 50 % - <= 100 %)*		
7778-50-9	01-2119454792-32-XXXX	Oxidizing solid, Category 2, H272 Acute toxicity, Category 3, H301 Acute toxicity, Category 2, H330 Acute toxicity, Category 4, H312 Skin corrosion, Category 1B, H314 Respiratory sensitisation, Category 1, H334 Skin sensitisation, Category 1, H317 Germ cell mutagenicity, Category 1B, H340 Carcinogenicity, Category 1B, H350 Reproductive toxicity, Category 1B, H360FD Specific target organ toxicity - single exposure, Category 3, H335 Specific target organ toxicity - repeated exposure, Category 1, H372 Acute aquatic toxicity, Category 1, H400



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Chronic aquatic toxicity, Category 1, H410

M-Factor: 1

For the full text of the H-Statements mentioned in this Section, see Section 16.

3.2 Mixture

Not applicable

SECTION 4. First aid measures

4.1 Description of first aid measures

General advice

First aider needs to protect himself.

After inhalation: fresh air. Immediately call in physician. If breathing stops: immediately apply artificial respiration, if necessary also oxygen.

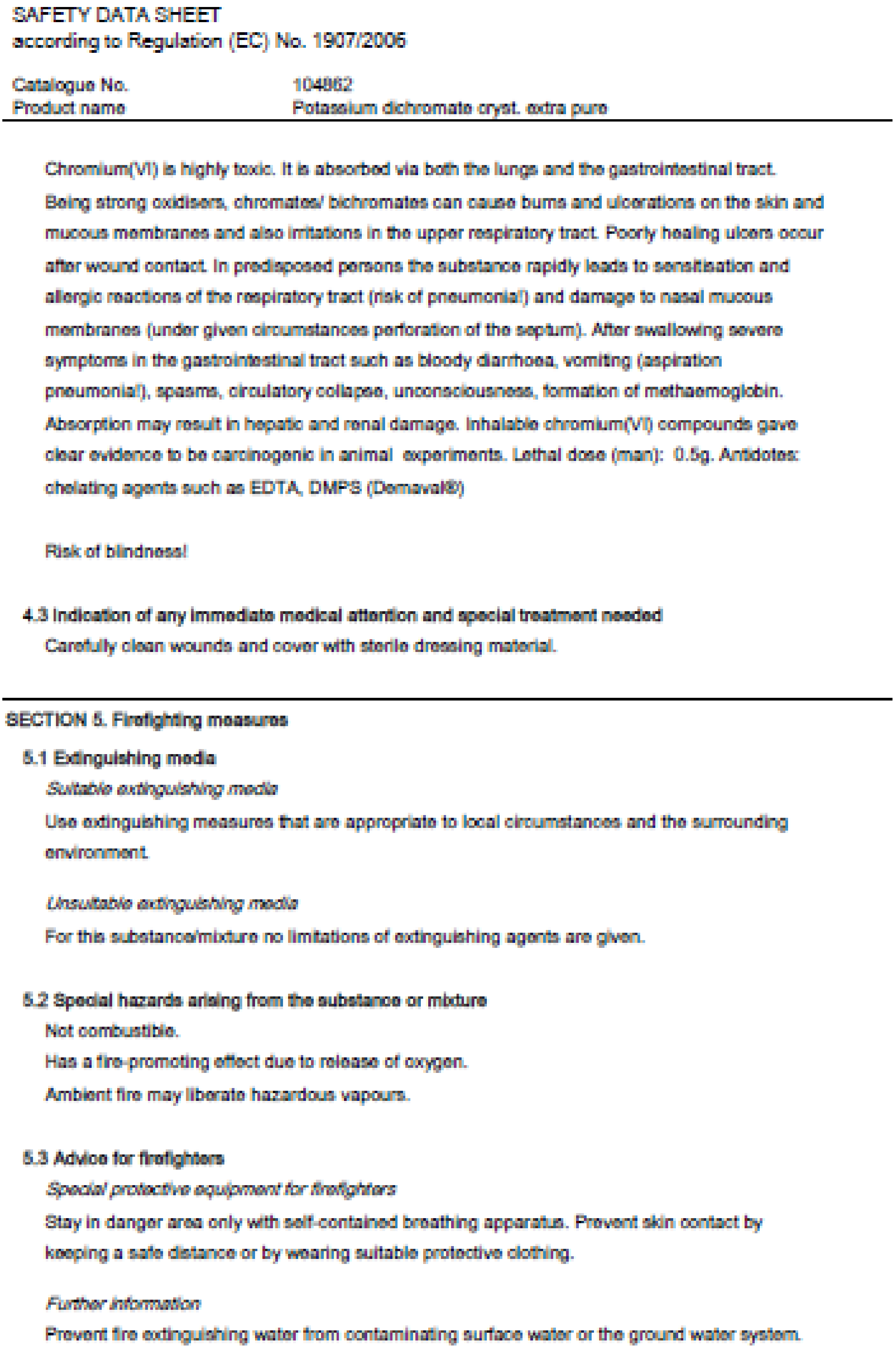
In case of skin contact: Take off immediately all contaminated clothing. Rinse skin with water/ shower. Call a physician immediately.

After eye contact: rinse out with plenty of water. Immediately call in ophthalmologist. Remove contact lenses.

If swallowed: give water to drink (two glasses at most). Seek medical advice immediately. In exceptional cases only, if medical care is not available within one hour, induce vomiting (only in persons who are wide awake and fully conscious), administer activated charcoal (20 - 40 g in a 10% slurry) and consult a doctor as quickly as possible. Do not attempt to neutralise.

4.2 Most important symptoms and effects, both acute and delayed

Irritation and corrosion, Allergic reactions, Cough, Shortness of breath



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Chromium(VI) is highly toxic. It is absorbed via both the lungs and the gastrointestinal tract. Being strong oxidisers, chromates/ bichromates can cause burns and ulcerations on the skin and mucous membranes and also irritations in the upper respiratory tract. Poorly healing ulcers occur after wound contact. In predisposed persons the substance rapidly leads to sensitisation and allergic reactions of the respiratory tract (risk of pneumonia!) and damage to nasal mucous membranes (under given circumstances perforation of the septum). After swallowing severe symptoms in the gastrointestinal tract such as bloody diarrhoea, vomiting (aspiration pneumonia!), spasms, circulatory collapse, unconsciousness, formation of methaemoglobin. Absorption may result in hepatic and renal damage. Inhalable chromium(VI) compounds gave clear evidence to be carcinogenic in animal experiments. Lethal dose (man): 0.5g. Antidotes: chelating agents such as EDTA, DMPS (Dimaval®)

Risk of blindness!

4.3 Indication of any immediate medical attention and special treatment needed

Carefully clean wounds and cover with sterile dressing material.

SECTION 5. Firefighting measures

5.1 Extinguishing media

Suitable extinguishing media

Use extinguishing measures that are appropriate to local circumstances and the surrounding environment.

Unsuitable extinguishing media

For this substance/mixture no limitations of extinguishing agents are given.

5.2 Special hazards arising from the substance or mixture

Not combustible.

Has a fire-promoting effect due to release of oxygen.

Ambient fire may liberate hazardous vapours.

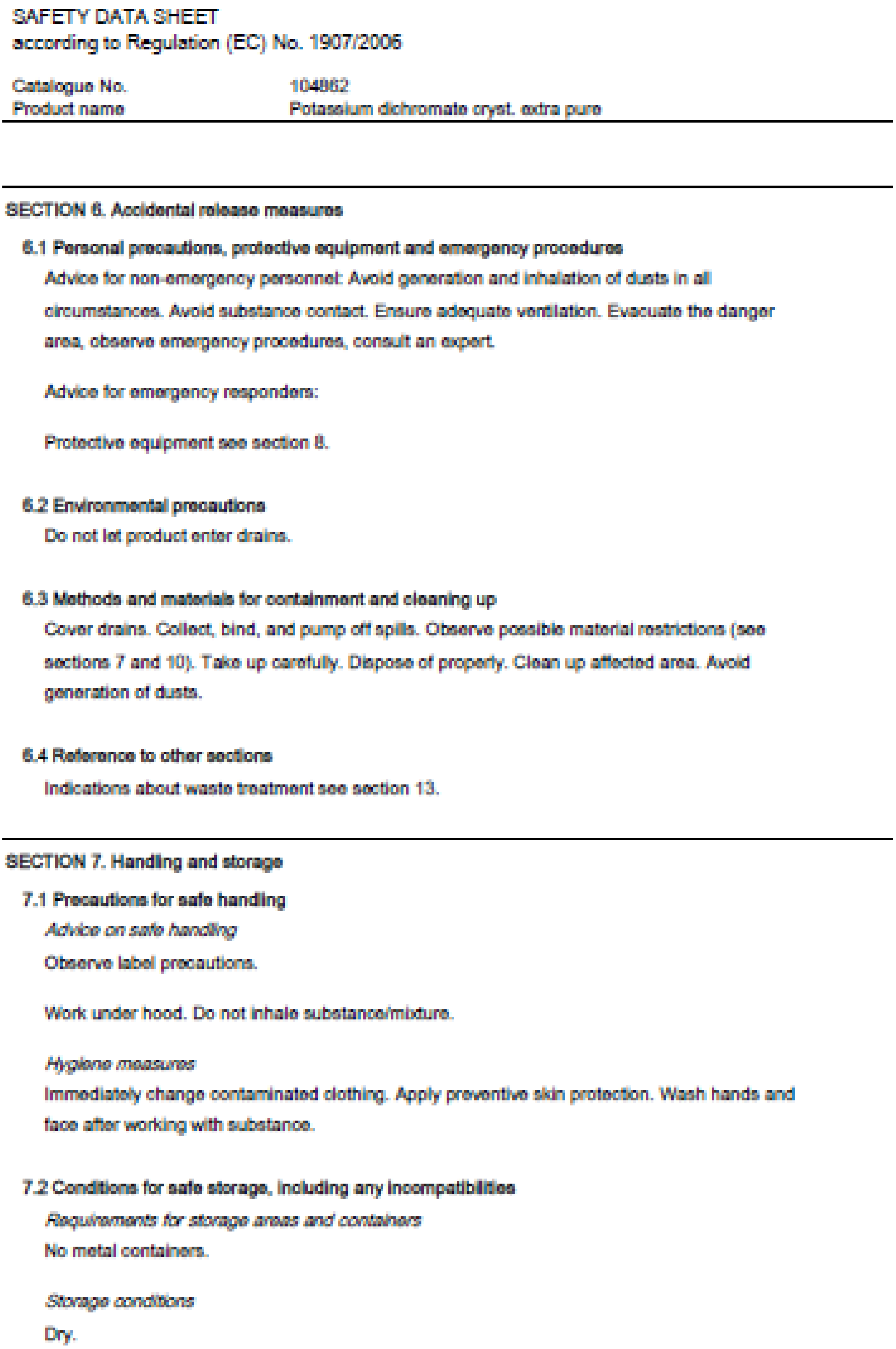
5.3 Advice for firefighters

Special protective equipment for firefighters

Stay in danger area only with self-contained breathing apparatus. Prevent skin contact by keeping a safe distance or by wearing suitable protective clothing.

Further information

Prevent fire extinguishing water from contaminating surface water or the ground water system.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

SECTION 6. Accidental release measures

6.1 Personal precautions, protective equipment and emergency procedures

Advice for non-emergency personnel: Avoid generation and inhalation of dusts in all circumstances. Avoid substance contact. Ensure adequate ventilation. Evacuate the danger area, observe emergency procedures, consult an expert.

Advice for emergency responders:

Protective equipment see section 8.

6.2 Environmental precautions

Do not let product enter drains.

6.3 Methods and materials for containment and cleaning up

Cover drains. Collect, bind, and pump off spills. Observe possible material restrictions (see sections 7 and 10). Take up carefully. Dispose of properly. Clean up affected area. Avoid generation of dusts.

6.4 Reference to other sections

Indications about waste treatment see section 13.

SECTION 7. Handling and storage

7.1 Precautions for safe handling

Advice on safe handling

Observe label precautions.

Work under hood. Do not inhale substance/mixture.

Hygiene measures

Immediately change contaminated clothing. Apply preventive skin protection. Wash hands and face after working with substance.

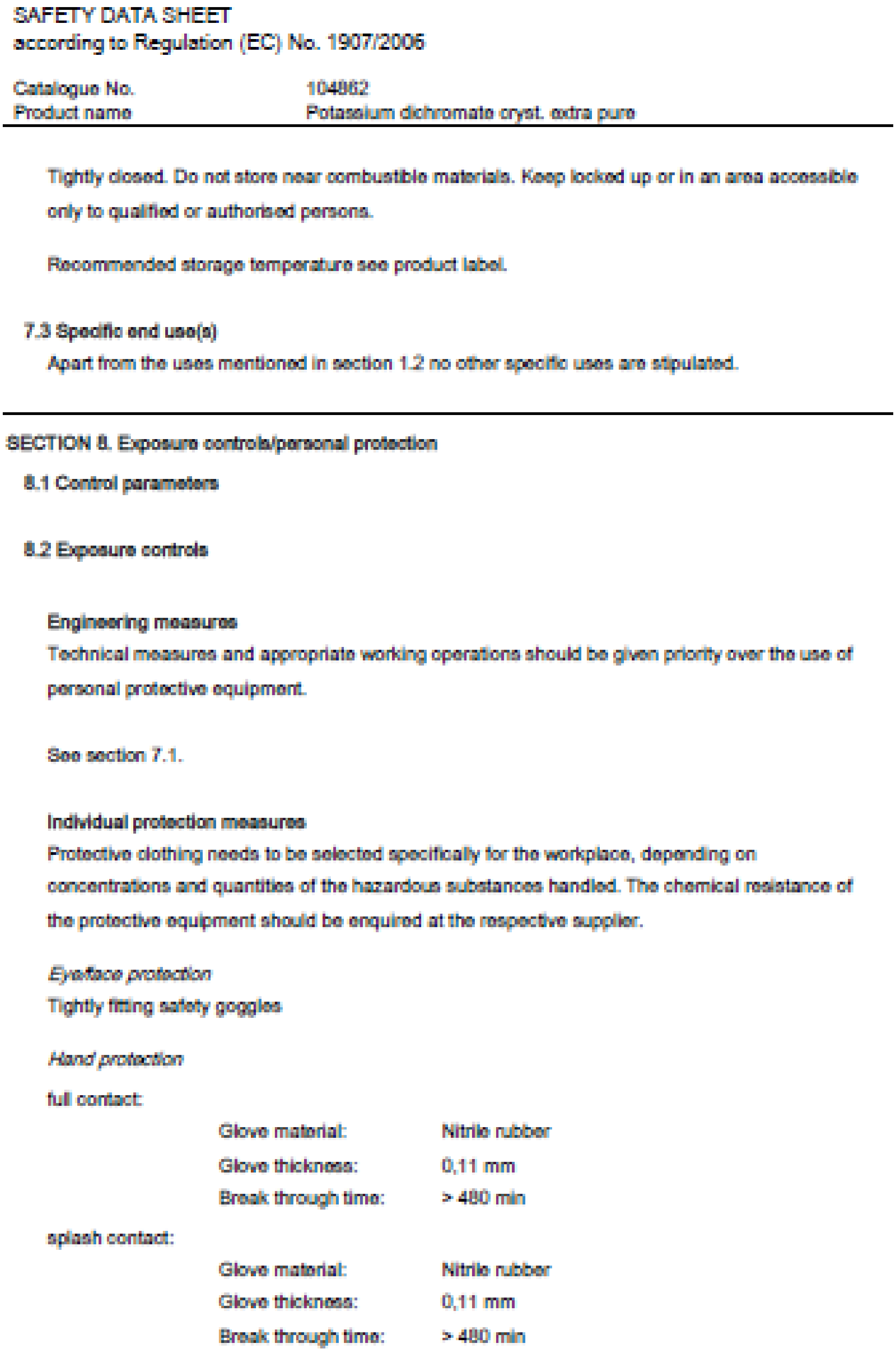
7.2 Conditions for safe storage, including any incompatibilities

Requirements for storage areas and containers

No metal containers.

Storage conditions

Dry.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Tightly closed. Do not store near combustible materials. Keep locked up or in an area accessible only to qualified or authorised persons.

Recommended storage temperature see product label.

7.3 Specific end use(s)

Apart from the uses mentioned in section 1.2 no other specific uses are stipulated.

SECTION 8. Exposure controls/personal protection

8.1 Control parameters

8.2 Exposure controls

Engineering measures

Technical measures and appropriate working operations should be given priority over the use of personal protective equipment.

See section 7.1.

Individual protection measures

Protective clothing needs to be selected specifically for the workplace, depending on concentrations and quantities of the hazardous substances handled. The chemical resistance of the protective equipment should be enquired at the respective supplier.

Eye/face protection

Tightly fitting safety goggles

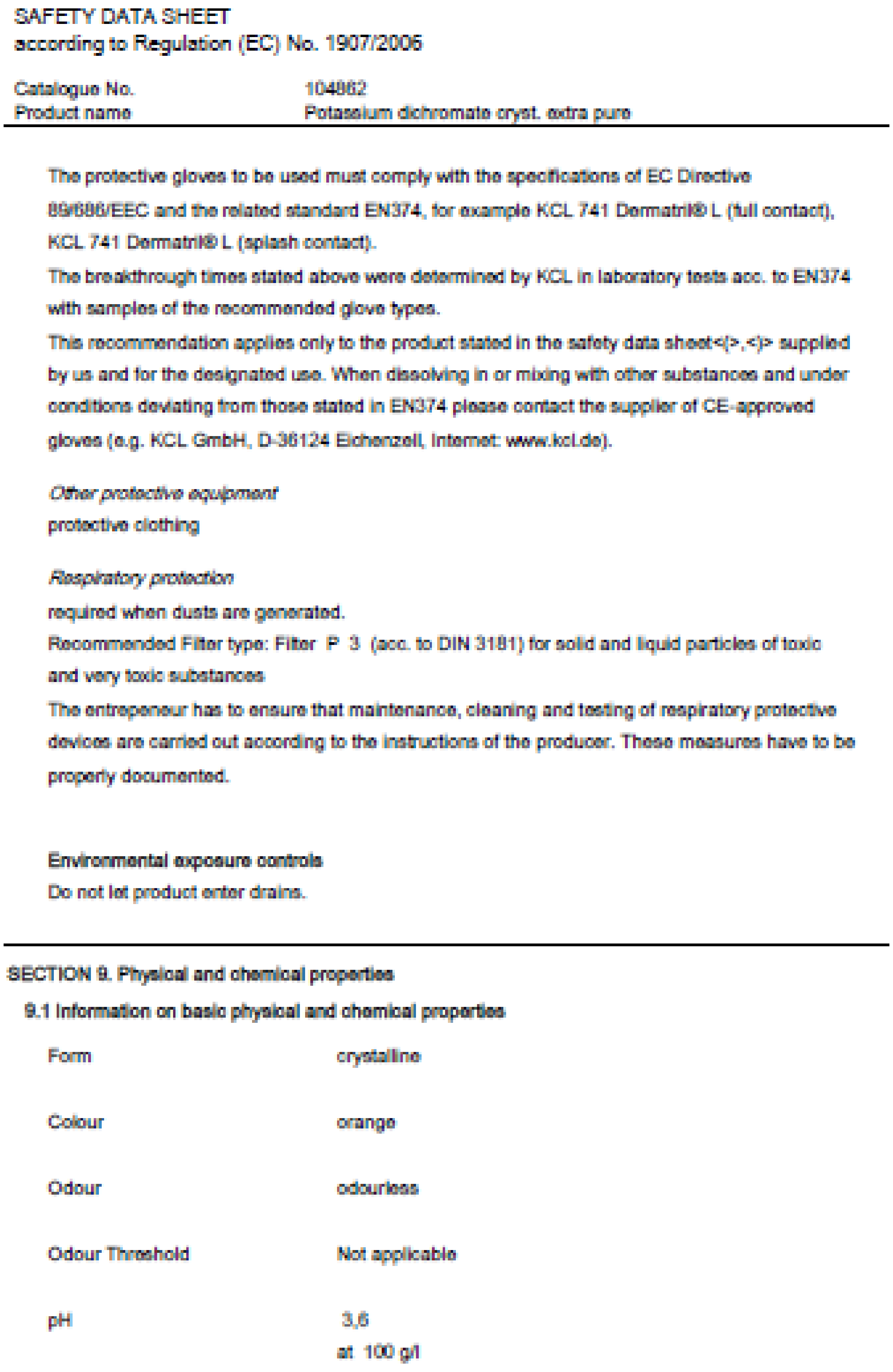
Hand protection

full contact:

Glove material: Nitrile rubber
Glove thickness: 0,11 mm
Break through time: > 480 min

splash contact:

Glove material: Nitrile rubber
Glove thickness: 0,11 mm
Break through time: > 480 min



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

The protective gloves to be used must comply with the specifications of EC Directive 89/686/EEC and the related standard EN374, for example KCL 741 Dermatril® L (full contact), KCL 741 Dermatril® L (splash contact).

The breakthrough times stated above were determined by KCL in laboratory tests acc. to EN374 with samples of the recommended glove types.

This recommendation applies only to the product stated in the safety data sheet<(>,<)> supplied by us and for the designated use. When dissolving in or mixing with other substances and under conditions deviating from those stated in EN374 please contact the supplier of CE-approved gloves (e.g. KCL GmbH, D-36124 Eichenzell, Internet: www.kcl.de).

Other protective equipment
protective clothing

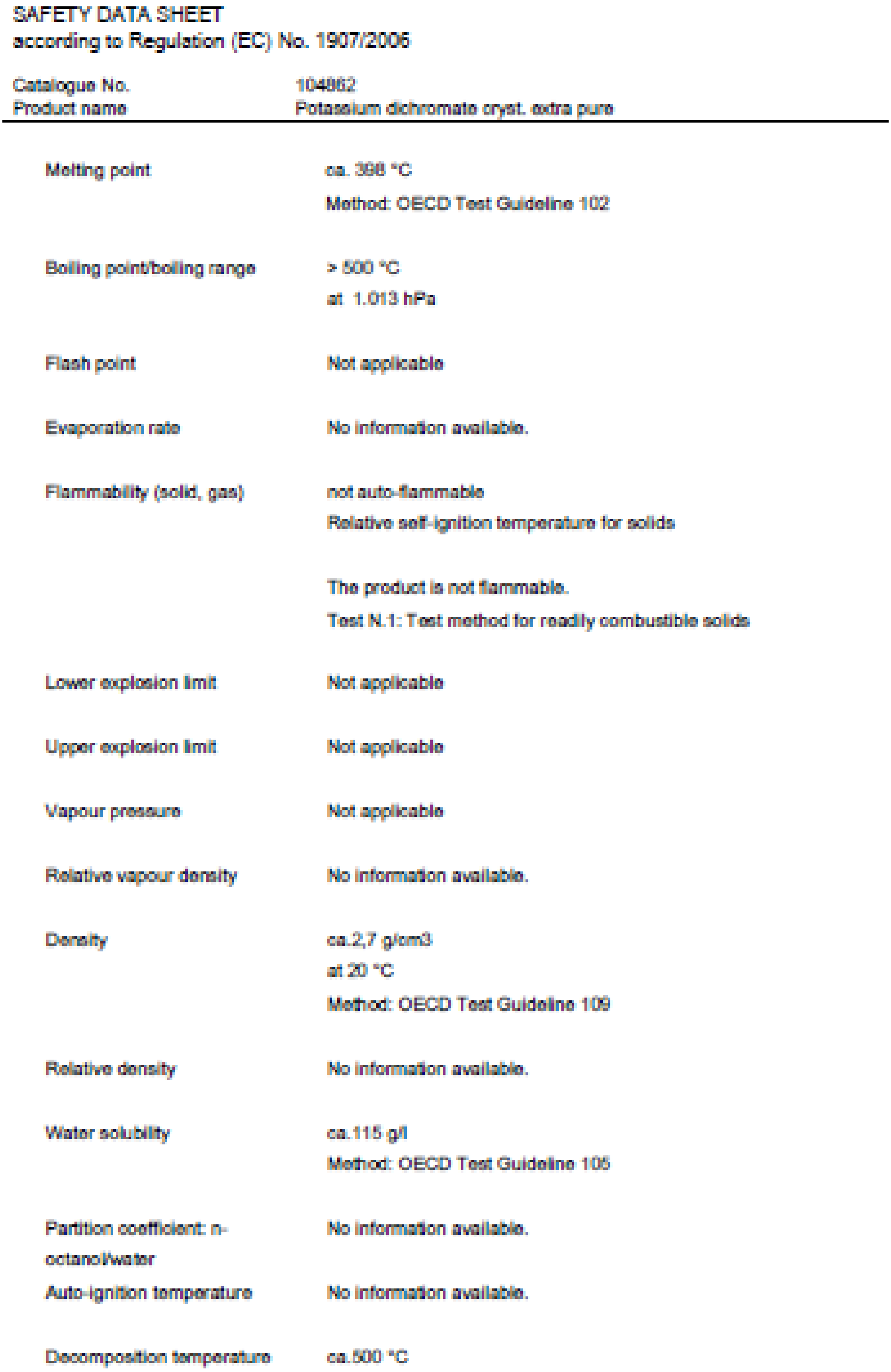
Respiratory protection
required when dusts are generated.
Recommended Filter type: Filter P 3 (acc. to DIN 3181) for solid and liquid particles of toxic and very toxic substances
The entrepeneur has to ensure that maintenance, cleaning and testing of respiratory protective devices are carried out according to the instructions of the producer. These measures have to be properly documented.

Environmental exposure controls
Do not let product enter drains.

SECTION 9. Physical and chemical properties

9.1 Information on basic physical and chemical properties

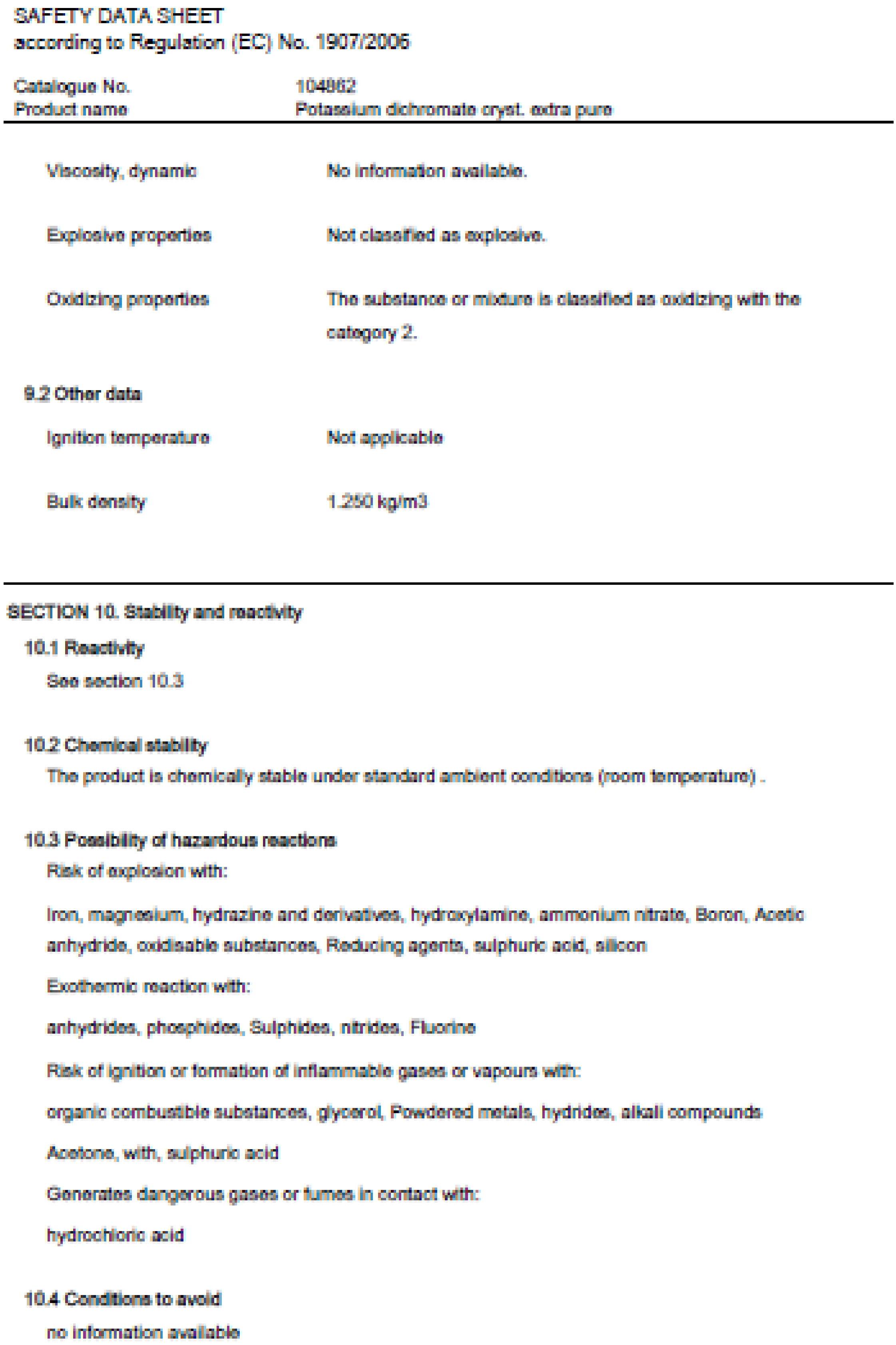
Form	crystalline
Colour	orange
Odour	odourless
Odour Threshold	Not applicable
pH	3,6 at 100 g/l



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Melting point	ca. 398 °C Method: OECD Test Guideline 102
Boiling point/boiling range	> 500 °C at 1.013 hPa
Flash point	Not applicable
Evaporation rate	No information available.
Flammability (solid, gas)	not auto-flammable Relative self-ignition temperature for solids The product is not flammable. Test N.1: Test method for readily combustible solids
Lower explosion limit	Not applicable
Upper explosion limit	Not applicable
Vapour pressure	Not applicable
Relative vapour density	No information available.
Density	ca.2,7 g/cm3 at 20 °C Method: OECD Test Guideline 109
Relative density	No information available.
Water solubility	ca.115 g/l Method: OECD Test Guideline 105
Partition coefficient: n-octanol/water	No information available.
Auto-ignition temperature	No information available.
Decomposition temperature	ca.500 °C



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Viscosity, dynamic	No information available.
Explosive properties	Not classified as explosive.
Oxidizing properties	The substance or mixture is classified as oxidizing with the category 2.

9.2 Other data

Ignition temperature	Not applicable
Bulk density	1.250 kg/m3

SECTION 10. Stability and reactivity

10.1 Reactivity

See section 10.3

10.2 Chemical stability

The product is chemically stable under standard ambient conditions (room temperature) .

10.3 Possibility of hazardous reactions

Risk of explosion with:

Iron, magnesium, hydrazine and derivatives, hydroxylamine, ammonium nitrate, Boron, Acetic anhydride, oxidisable substances, Reducing agents, sulphuric acid, silicon

Exothermic reaction with:

anhydrides, phosphides, Sulphides, nitrides, Fluorine

Risk of ignition or formation of inflammable gases or vapours with:

organic combustible substances, glycerol, Powdered metals, hydrides, alkali compounds

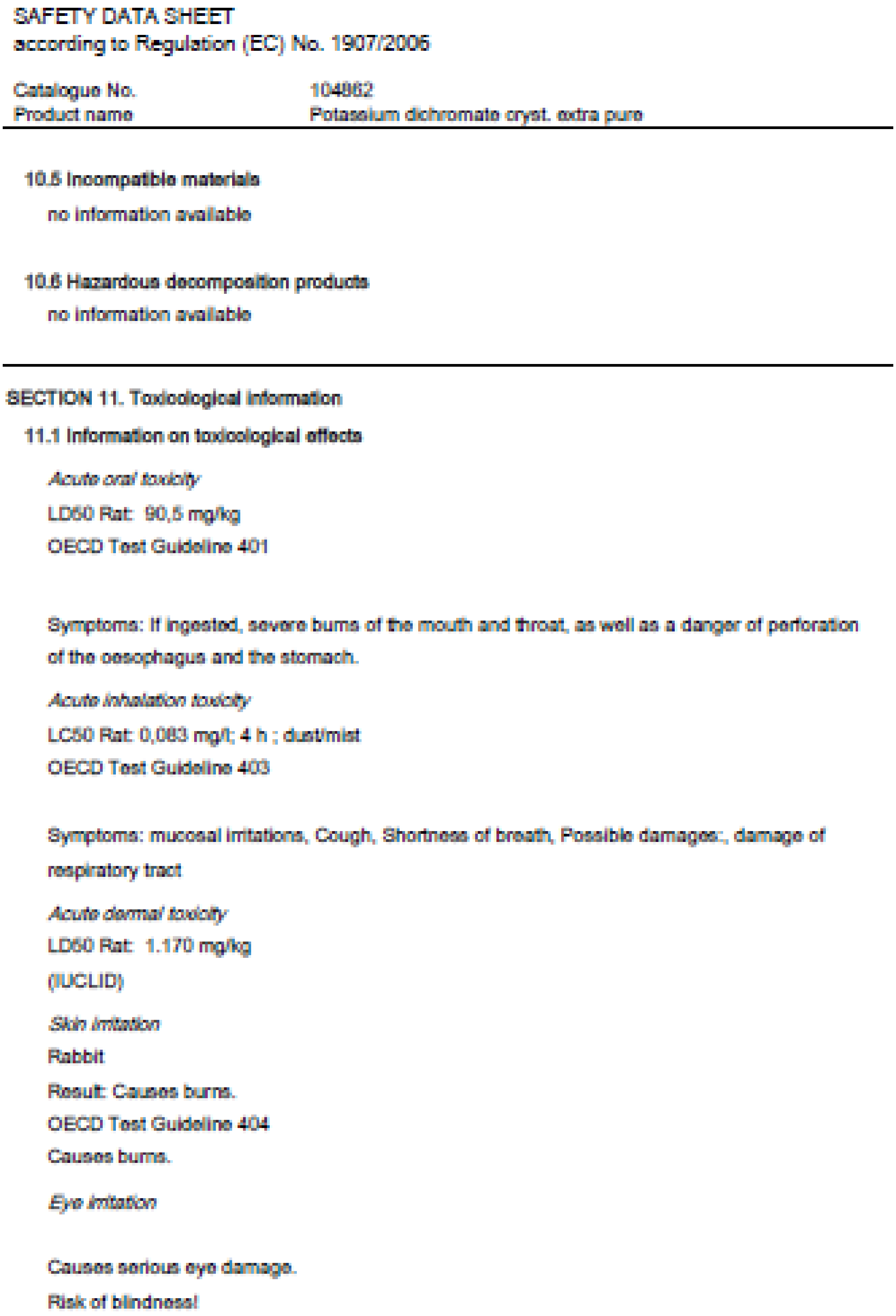
Acetone, with, sulphuric acid

Generates dangerous gases or fumes in contact with:

hydrochloric acid

10.4 Conditions to avoid

no information available



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

10.5 Incompatible materials

no information available

10.6 Hazardous decomposition products

no information available

SECTION 11. Toxicological information

11.1 Information on toxicological effects

Acute oral toxicity

LD50 Rat: 90,5 mg/kg

OECD Test Guideline 401

Symptoms: If ingested, severe burns of the mouth and throat, as well as a danger of perforation of the oesophagus and the stomach.

Acute inhalation toxicity

LC50 Rat: 0,083 mg/l; 4 h ; dust/mist

OECD Test Guideline 403

Symptoms: mucosal irritations, Cough, Shortness of breath, Possible damages:, damage of respiratory tract

Acute dermal toxicity

LD50 Rat: 1.170 mg/kg

(IUCLID)

Skin irritation

Rabbit

Result: Causes burns.

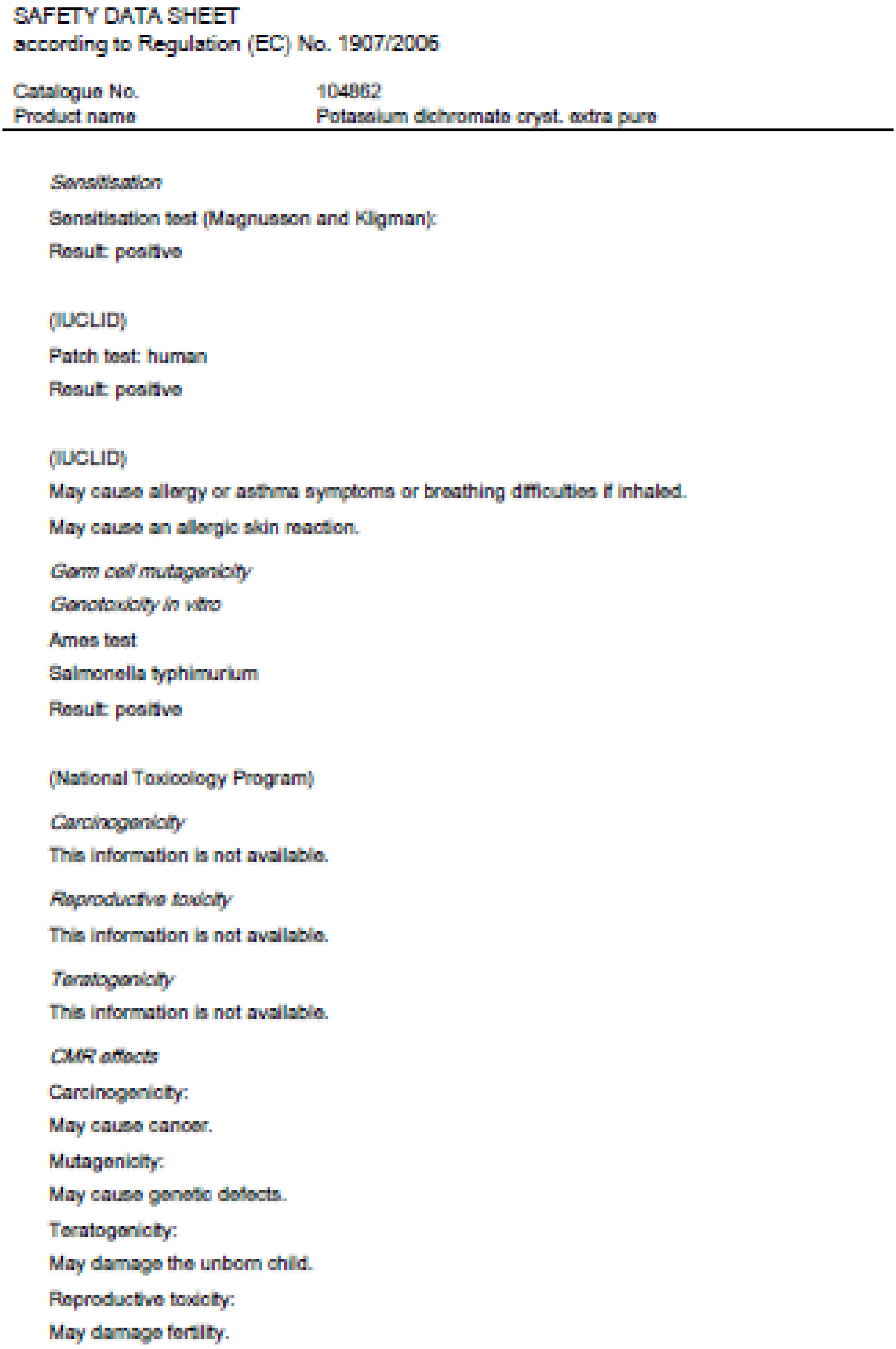
OECD Test Guideline 404

Causes burns.

Eye irritation

Causes serious eye damage.

Risk of blindness!



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Sensitisation

Sensitisation test (Magnusson and Kligman):

Result: positive

(IUCLID)

Patch test: human

Result: positive

(IUCLID)

May cause allergy or asthma symptoms or breathing difficulties if inhaled.

May cause an allergic skin reaction.

Germ cell mutagenicity

Genotoxicity in vitro

Ames test

Salmonella typhimurium

Result: positive

(National Toxicology Program)

Carcinogenicity

This information is not available.

Reproductive toxicity

This information is not available.

Teratogenicity

This information is not available.

CMR effects

Carcinogenicity:

May cause cancer.

Mutagenicity:

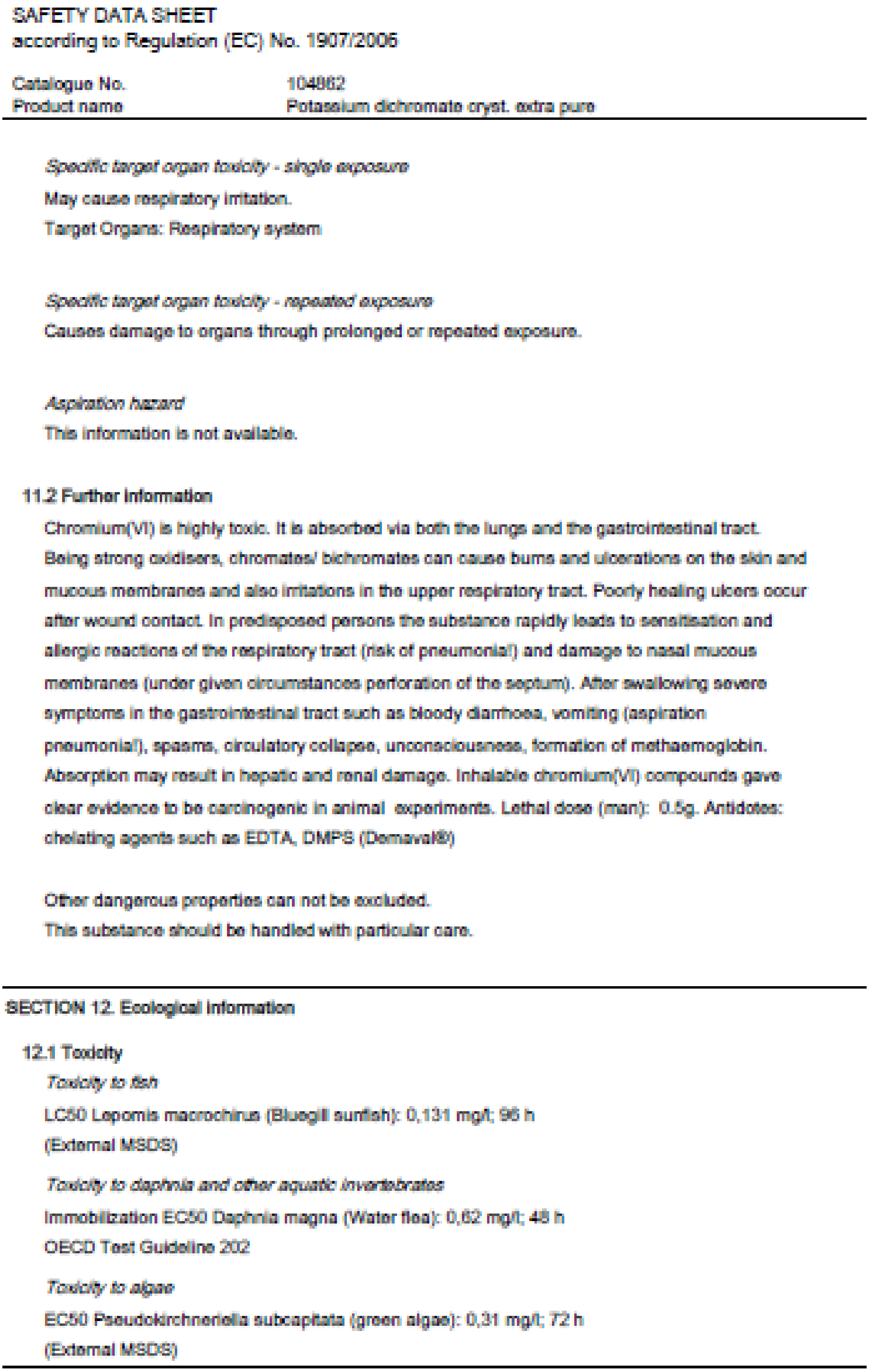
May cause genetic defects.

Teratogenicity:

May damage the unborn child.

Reproductive toxicity:

May damage fertility.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

Specific target organ toxicity - single exposure

May cause respiratory irritation.

Target Organs: Respiratory system

Specific target organ toxicity - repeated exposure

Causes damage to organs through prolonged or repeated exposure.

Aspiration hazard

This information is not available.

11.2 Further information

Chromium(VI) is highly toxic. It is absorbed via both the lungs and the gastrointestinal tract. Being strong oxidisers, chromates/ bichromates can cause burns and ulcerations on the skin and mucous membranes and also irritations in the upper respiratory tract. Poorly healing ulcers occur after wound contact. In predisposed persons the substance rapidly leads to sensitisation and allergic reactions of the respiratory tract (risk of pneumonia!) and damage to nasal mucous membranes (under given circumstances perforation of the septum). After swallowing severe symptoms in the gastrointestinal tract such as bloody diarrhoea, vomiting (aspiration pneumonia!), spasms, circulatory collapse, unconsciousness, formation of methaemoglobin. Absorption may result in hepatic and renal damage. Inhalable chromium(VI) compounds gave clear evidence to be carcinogenic in animal experiments. Lethal dose (man): 0.5g. Antidotes: chelating agents such as EDTA, DMPS (Dimaval®)

Other dangerous properties can not be excluded.

This substance should be handled with particular care.

SECTION 12. Ecological information

12.1 Toxicity

Toxicity to fish

LC50 Lepomis macrochirus (Bluegill sunfish): 0,131 mg/l; 96 h

(External MSDS)

Toxicity to daphnia and other aquatic invertebrates

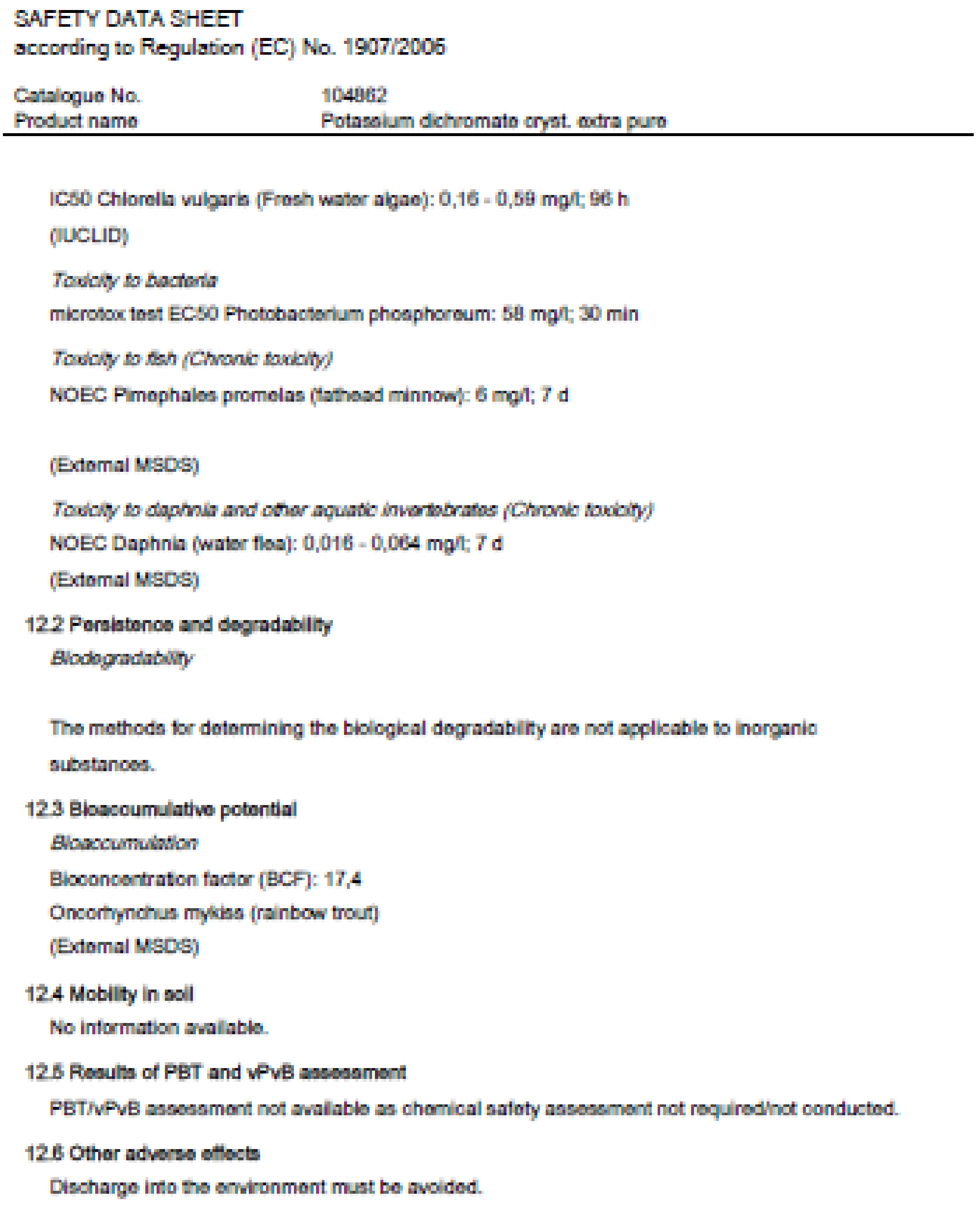
Immobilization EC50 Daphnia magna (Water flea): 0,62 mg/l; 48 h

OECD Test Guideline 202

Toxicity to algae

EC50 Pseudokirchneriella subcapitata (green algae): 0,31 mg/l; 72 h

(External MSDS)



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

IC50 Chlorella vulgaris (Fresh water algae): 0,16 - 0,59 mg/l; 96 h

(IUCLID)

Toxicity to bacteria

microtox test EC50 Photobacterium phosphoreum: 58 mg/l; 30 min

Toxicity to fish (Chronic toxicity)

NOEC Pimephales promelas (fathead minnow): 6 mg/l; 7 d

(External MSDS)

Toxicity to daphnia and other aquatic invertebrates (Chronic toxicity)

NOEC Daphnia (water flea): 0,016 - 0,064 mg/l; 7 d

(External MSDS)

12.2 Persistence and degradability

Biodegradability

The methods for determining the biological degradability are not applicable to inorganic substances.

12.3 Bioaccumulative potential

Bioaccumulation

Bioconcentration factor (BCF): 17,4

Oncorhynchus mykiss (rainbow trout)

(External MSDS)

12.4 Mobility in soil

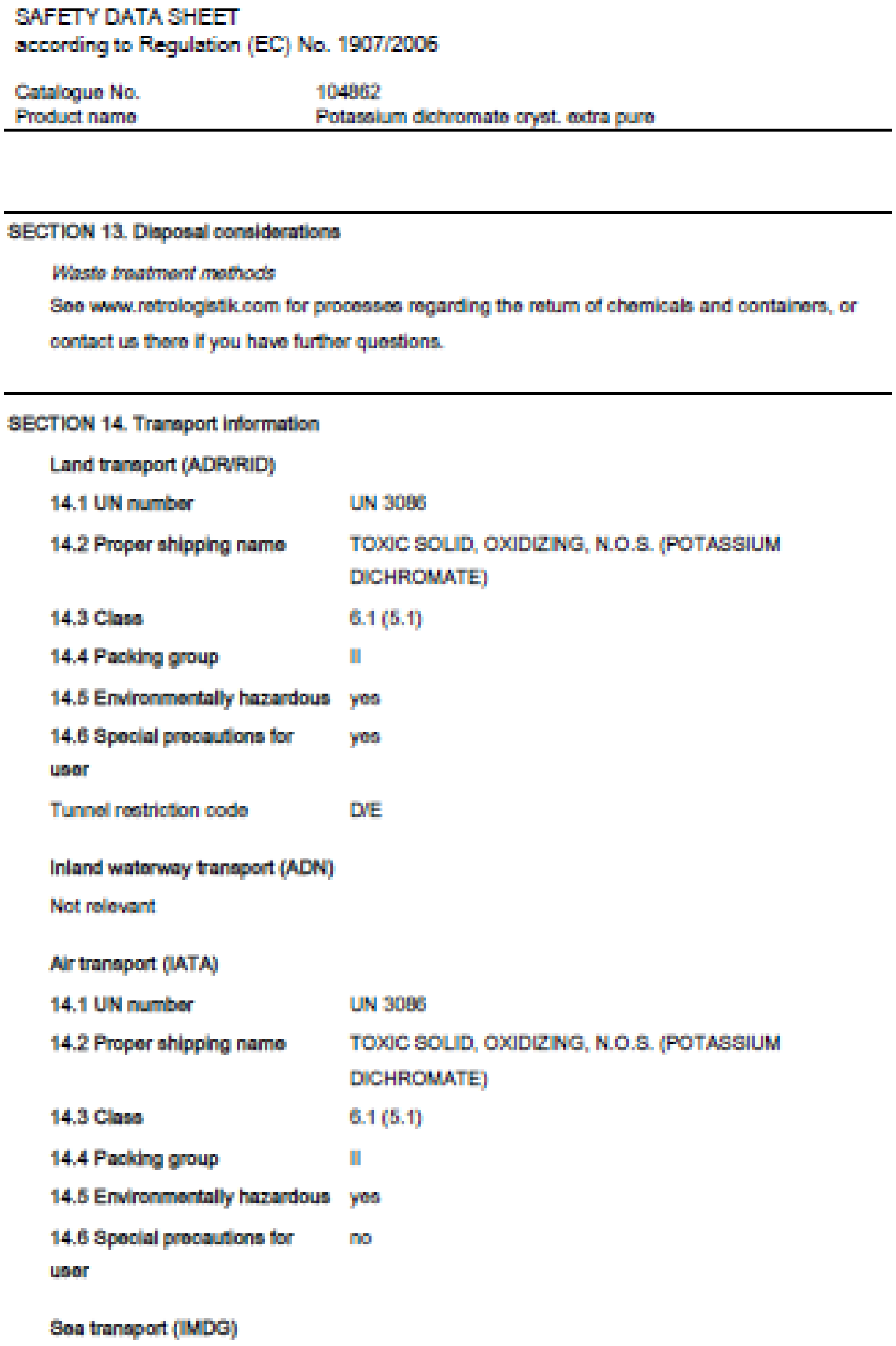
No information available.

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

Discharge into the environment must be avoided.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

SECTION 13. Disposal considerations

Waste treatment methods

See www.retrologistik.com for processes regarding the return of chemicals and containers, or contact us there if you have further questions.

SECTION 14. Transport information

Land transport (ADR/RID)

14.1 UN number	UN 3086
14.2 Proper shipping name	TOXIC SOLID, OXIDIZING, N.O.S. (POTASSIUM DICHROMATE)
14.3 Class	6.1 (5.1)
14.4 Packing group	II
14.5 Environmentally hazardous	yes
14.6 Special precautions for user	yes
Tunnel restriction code	D/E

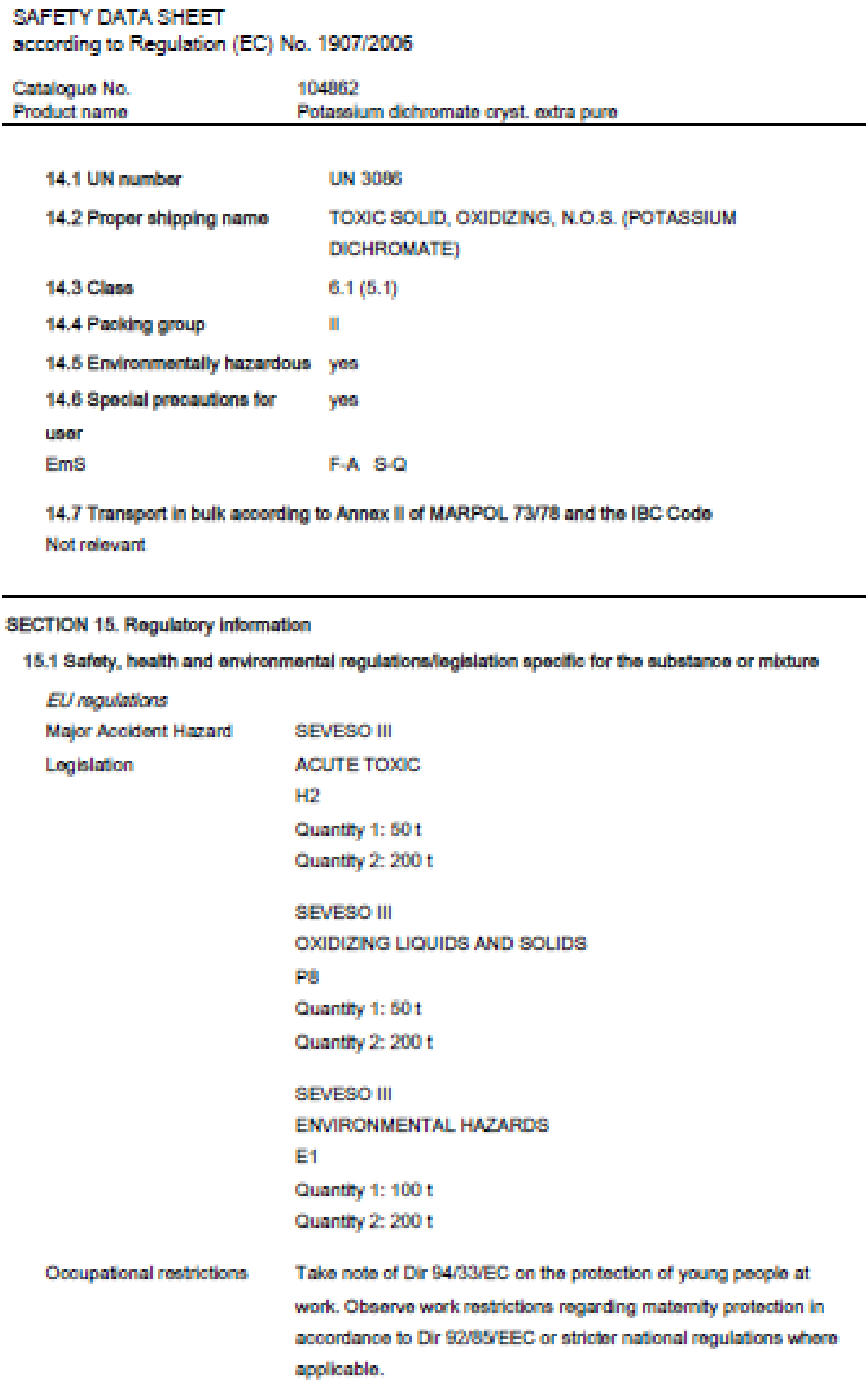
Inland waterway transport (ADN)

Not relevant

Air transport (IATA)

14.1 UN number	UN 3086
14.2 Proper shipping name	TOXIC SOLID, OXIDIZING, N.O.S. (POTASSIUM DICHROMATE)
14.3 Class	6.1 (5.1)
14.4 Packing group	II
14.5 Environmentally hazardous	yes
14.6 Special precautions for user	no

Sea transport (IMDG)



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

14.1 UN number	UN 3086
14.2 Proper shipping name	TOXIC SOLID, OXIDIZING, N.O.S. (POTASSIUM DICHROMATE)
14.3 Class	6.1 (5.1)
14.4 Packing group	II
14.5 Environmentally hazardous	yes
14.6 Special precautions for user	yes
EmS	F-A S-Q

14.7 Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code

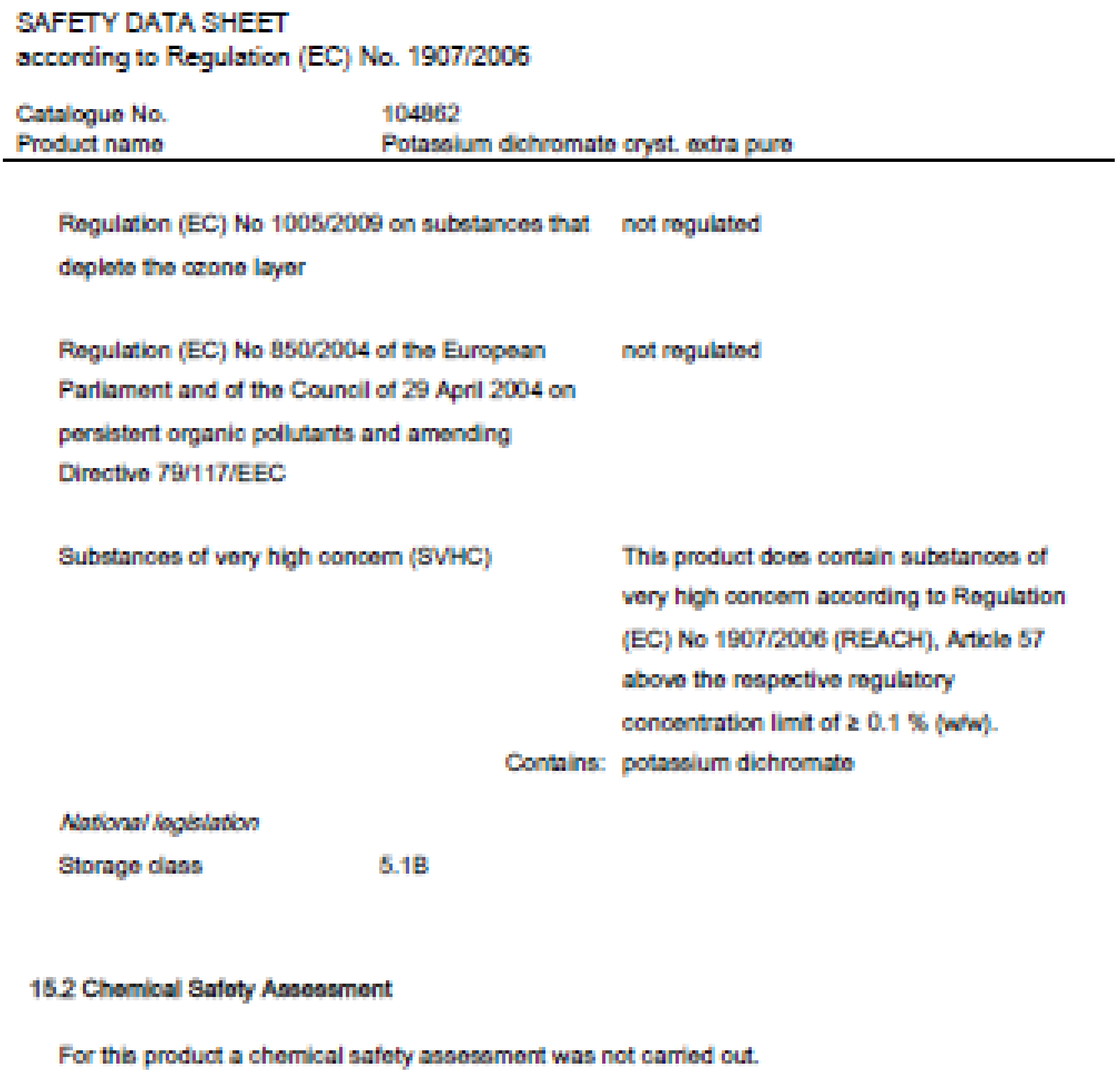
Not relevant

SECTION 15. Regulatory information

15.1 Safety, health and environmental regulations/legislation specific for the substance or mixture

EU regulations

Major Accident Hazard Legislation	SEVESO III ACUTE TOXIC H2 Quantity 1: 50 t Quantity 2: 200 t
	SEVESO III OXIDIZING LIQUIDS AND SOLIDS P8 Quantity 1: 50 t Quantity 2: 200 t
	SEVESO III ENVIRONMENTAL HAZARDS E1 Quantity 1: 100 t Quantity 2: 200 t
Occupational restrictions	Take note of Dir 94/33/EC on the protection of young people at work. Observe work restrictions regarding maternity protection in accordance to Dir 92/85/EEC or stricter national regulations where applicable.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

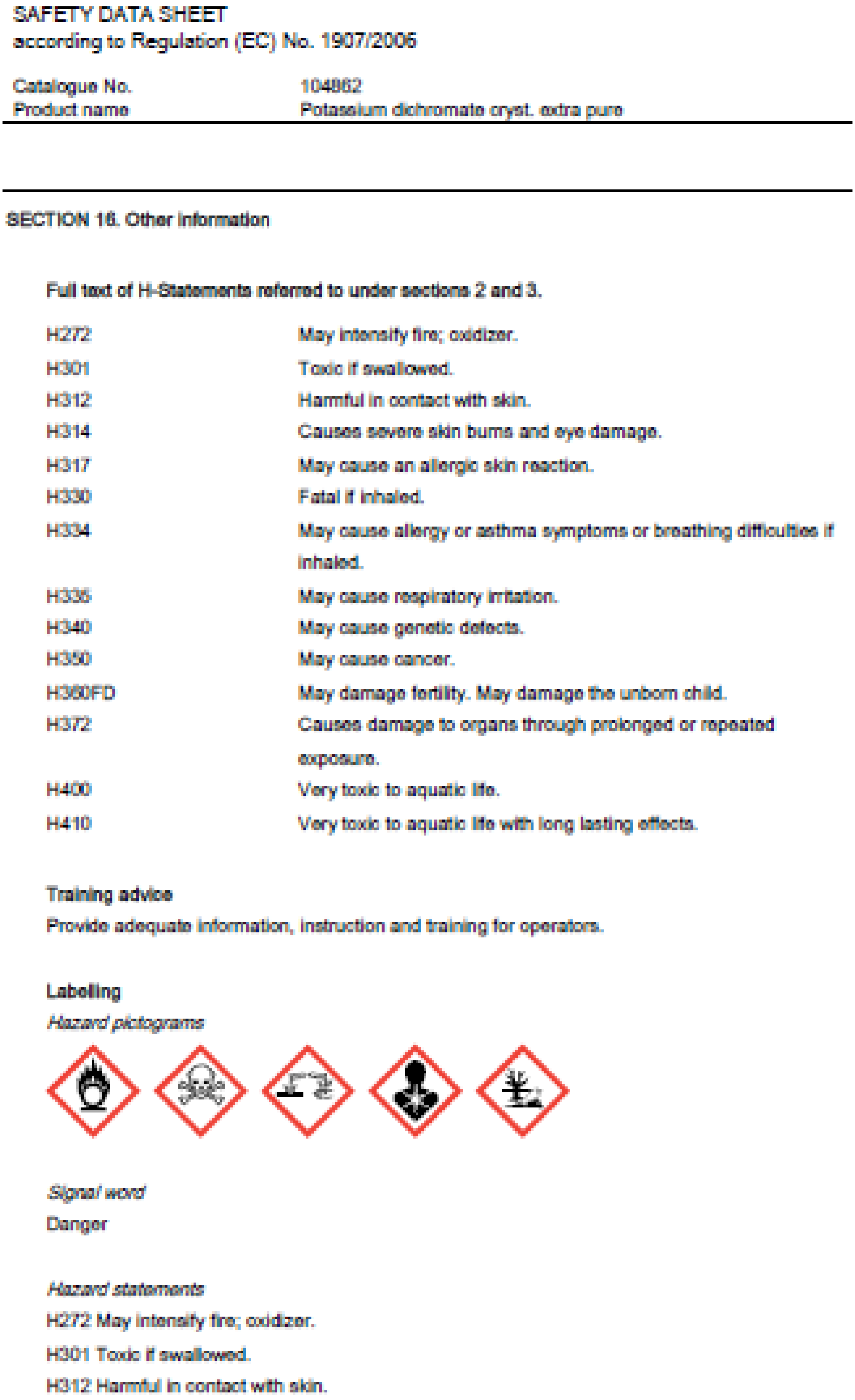
Regulation (EC) No 1005/2009 on substances that deplete the ozone layer	not regulated
Regulation (EC) No 850/2004 of the European Parliament and of the Council of 29 April 2004 on persistent organic pollutants and amending Directive 79/117/EEC	not regulated
Substances of very high concern (SVHC)	This product does contain substances of very high concern according to Regulation (EC) No 1907/2006 (REACH), Article 57 above the respective regulatory concentration limit of ≥ 0.1 % (w/w).
Contains:	potassium dichromate

National legislation

Storage class	5.1B

15.2 Chemical Safety Assessment

For this product a chemical safety assessment was not carried out.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

SECTION 16. Other information

Full text of H-Statements referred to under sections 2 and 3.

H272	May intensify fire; oxidizer.
H301	Toxic if swallowed.
H312	Harmful in contact with skin.
H314	Causes severe skin burns and eye damage.
H317	May cause an allergic skin reaction.
H330	Fatal if inhaled.
H334	May cause allergy or asthma symptoms or breathing difficulties if inhaled.
H335	May cause respiratory irritation.
H340	May cause genetic defects.
H350	May cause cancer.
H360FD	May damage fertility. May damage the unborn child.
H372	Causes damage to organs through prolonged or repeated exposure.
H400	Very toxic to aquatic life.
H410	Very toxic to aquatic life with long lasting effects.

Training advice

Provide adequate information, instruction and training for operators.

Labelling

Hazard pictograms

Signal word

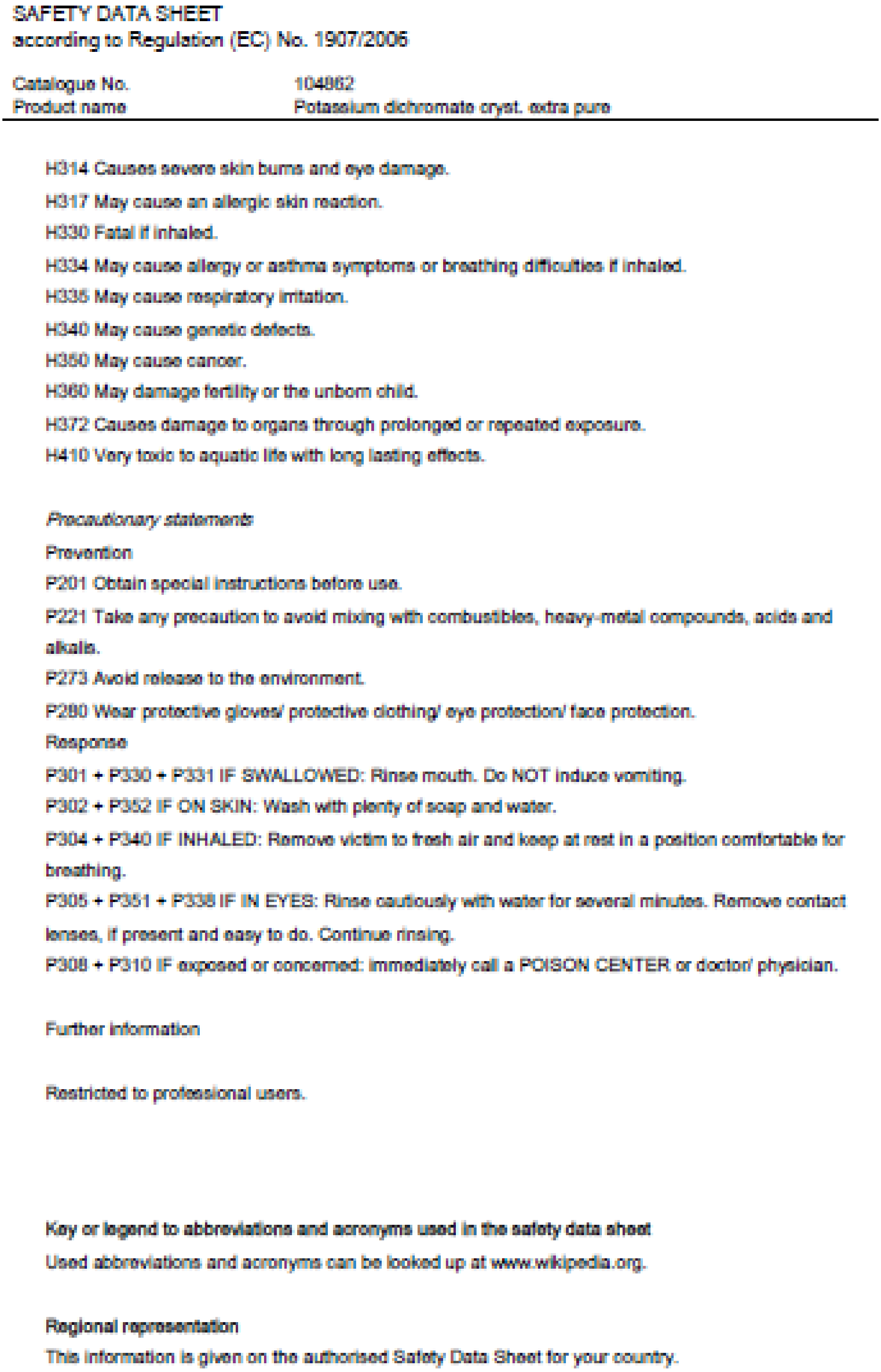
Danger

Hazard statements

H272 May intensify fire; oxidizer.

H301 Toxic if swallowed.

H312 Harmful in contact with skin.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

H314 Causes severe skin burns and eye damage.

H317 May cause an allergic skin reaction.

H330 Fatal if inhaled.

H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.

H335 May cause respiratory irritation.

H340 May cause genetic defects.

H350 May cause cancer.

H360 May damage fertility or the unborn child.

H372 Causes damage to organs through prolonged or repeated exposure.

H410 Very toxic to aquatic life with long lasting effects.

Precautionary statements

Prevention

P201 Obtain special instructions before use.

P221 Take any precaution to avoid mixing with combustibles, heavy-metal compounds, acids and alkalis.

P273 Avoid release to the environment.

P280 Wear protective gloves/ protective clothing/ eye protection/ face protection.

Response

P301 + P330 + P331 IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.

P302 + P352 IF ON SKIN: Wash with plenty of soap and water.

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.

P308 + P310 IF exposed or concerned: immediately call a POISON CENTER or doctor/ physician.

Further information

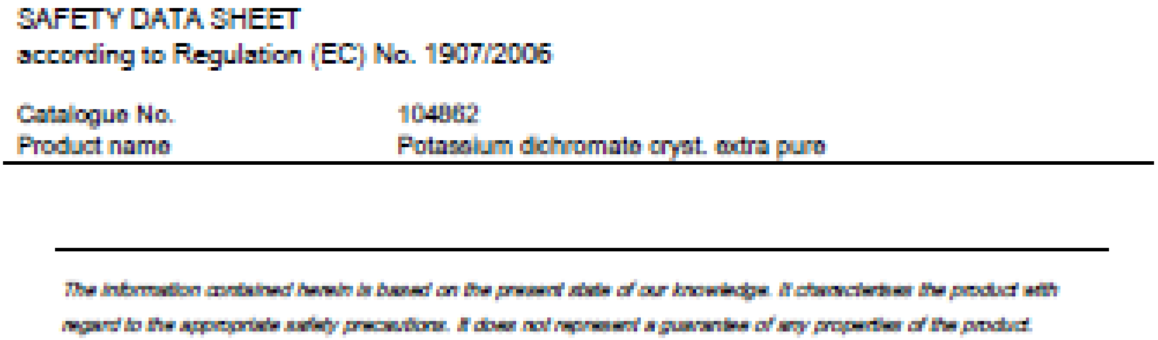
Restricted to professional users.

Key or legend to abbreviations and acronyms used in the safety data sheet

Used abbreviations and acronyms can be looked up at www.wikipedia.org.

Regional representation

This information is given on the authorised Safety Data Sheet for your country.



SAFETY DATA SHEET
according to Regulation (EC) No. 1907/2006

Catalogue No.	104862
Product name	Potassium dichromate cryst. extra pure

The information contained herein is based on the present state of our knowledge. It characterises the product with regard to the appropriate safety precautions. It does not represent a guarantee of any properties of the product.

A3.3. Sulphuric acid

Fisher Scientific

Part of Thermo Fisher Scientific

SAFETY DATA SHEET

Creation Date 12-Nov-2010 **Revision Date** 10-Jan-2017 **Revision Number** 4

1. Identification

Product Name	**Sulfuric Acid (Gerber)**
Cat No. :	**SA176-4**
Synonyms	Hydrogen sulfate; Vitriol brown oil; Oil of vitriol
Recommended Use	Laboratory chemicals.
Uses advised against	No Information available

Details of the supplier of the safety data sheet

Company
Fisher Scientific
One Reagent Lane
Fair Lawn, NJ 07410
Tel: (201) 796-7100

Emergency Telephone Number
CHEMTREC®, Inside the USA: 800-424-9300
CHEMTREC®, Outside the USA: 001-703-527-3887

2. Hazard(s) identification

Classification

This chemical is considered hazardous by the 2012 OSHA Hazard Communication Standard (29 CFR 1910.1200)

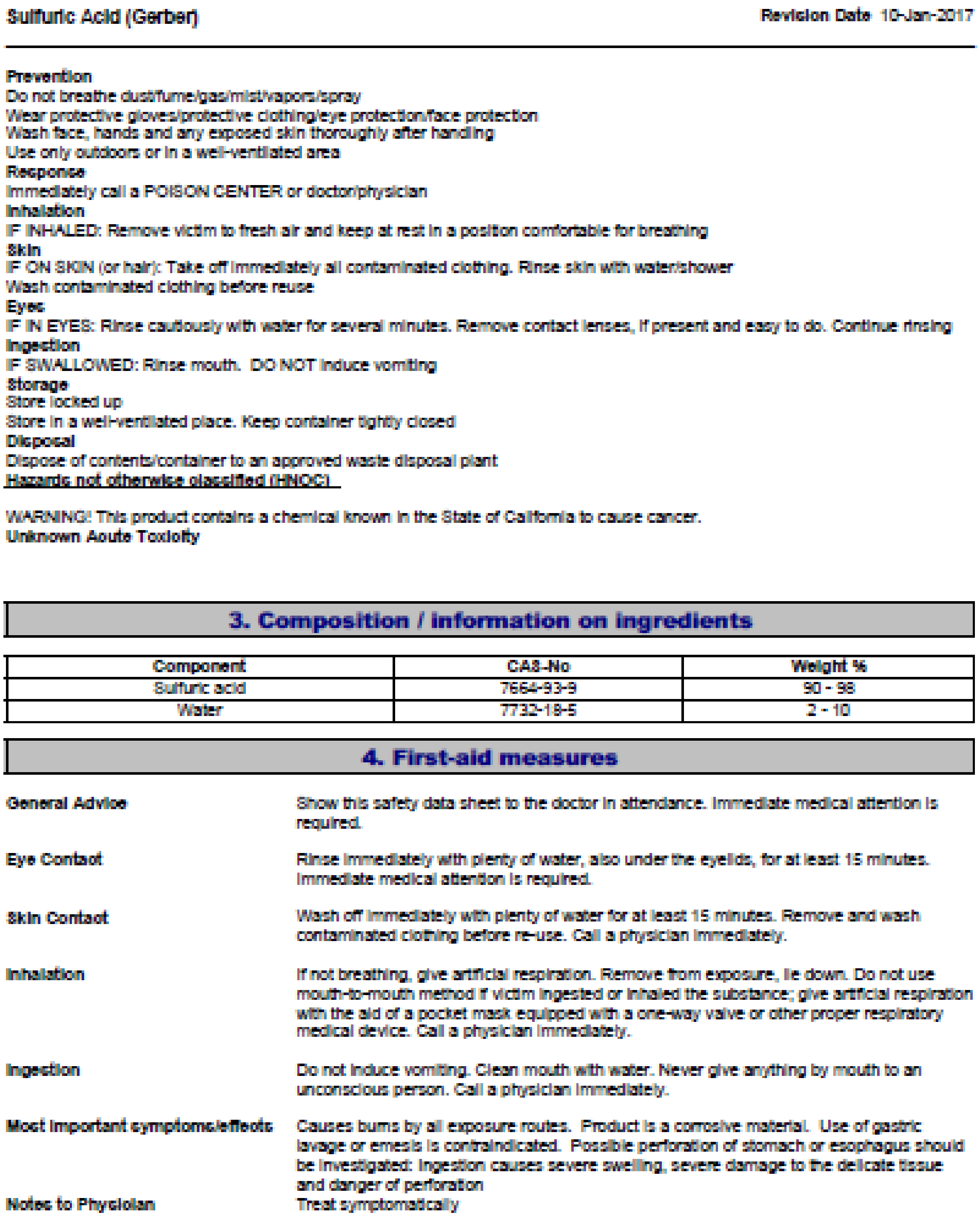
Skin Corrosion/Irritation	Category 1 A
Serious Eye Damage/Eye Irritation	Category 1
Specific target organ toxicity (single exposure)	Category 3
Target Organs - Respiratory system.	

Label Elements

Signal Word
Danger

Hazard Statements
Causes severe skin burns and eye damage
May cause respiratory irritation

Precautionary Statements



Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

Prevention
Do not breathe dust/fume/gas/mist/vapors/spray
Wear protective gloves/protective clothing/eye protection/face protection
Wash face, hands and any exposed skin thoroughly after handling
Use only outdoors or in a well-ventilated area
Response
Immediately call a POISON CENTER or doctor/physician
Inhalation
IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing
Skin
IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water/shower
Wash contaminated clothing before reuse
Eyes
IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing
Ingestion
IF SWALLOWED: Rinse mouth. DO NOT induce vomiting
Storage
Store locked up
Store in a well-ventilated place. Keep container tightly closed
Disposal
Dispose of contents/container to an approved waste disposal plant
Hazards not otherwise classified (HNOC)

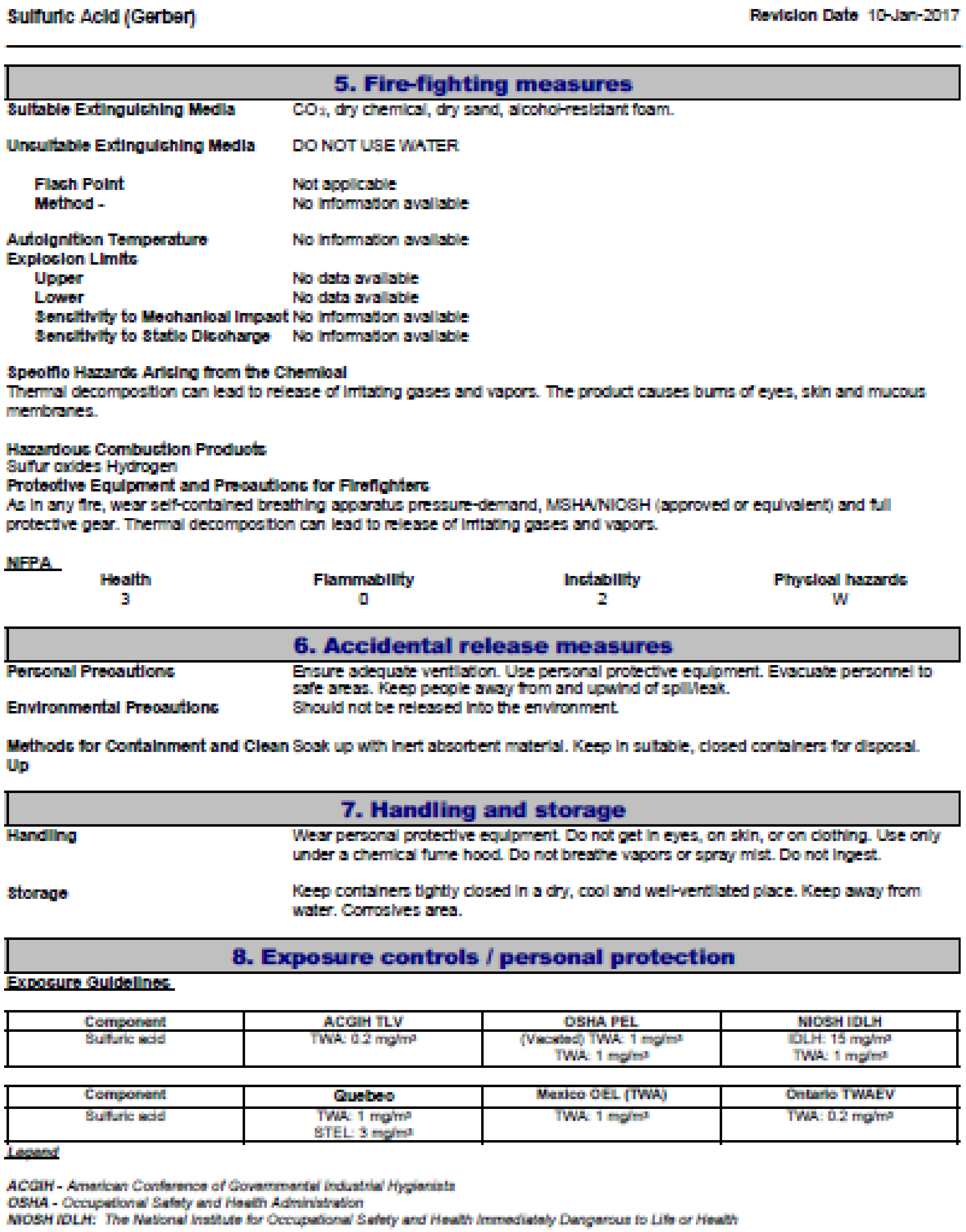
WARNING! This product contains a chemical known in the State of California to cause cancer.
Unknown Acute Toxicity

3. Composition / information on ingredients

Component	CAS-No	Weight %
Sulfuric acid	7664-93-9	90 - 98
Water	7732-18-5	2 - 10

4. First-aid measures

General Advice	Show this safety data sheet to the doctor in attendance. Immediate medical attention is required.
Eye Contact	Rinse immediately with plenty of water, also under the eyelids, for at least 15 minutes. Immediate medical attention is required.
Skin Contact	Wash off immediately with plenty of water for at least 15 minutes. Remove and wash contaminated clothing before re-use. Call a physician immediately.
Inhalation	If not breathing, give artificial respiration. Remove from exposure, lie down. Do not use mouth-to-mouth method if victim ingested or inhaled the substance; give artificial respiration with the aid of a pocket mask equipped with a one-way valve or other proper respiratory medical device. Call a physician immediately.
Ingestion	Do not induce vomiting. Clean mouth with water. Never give anything by mouth to an unconscious person. Call a physician immediately.
Most important symptoms/effects	Causes burns by all exposure routes. Product is a corrosive material. Use of gastric lavage or emesis is contraindicated. Possible perforation of stomach or esophagus should be investigated: Ingestion causes severe swelling, severe damage to the delicate tissue and danger of perforation
Notes to Physician	Treat symptomatically



Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

5. Fire-fighting measures

Suitable Extinguishing Media	CO_2, dry chemical, dry sand, alcohol-resistant foam.
Unsuitable Extinguishing Media	DO NOT USE WATER
Flash Point	Not applicable
Method -	No information available
Autoignition Temperature	No information available
Explosion Limits	
Upper	No data available
Lower	No data available
Sensitivity to Mechanical Impact	No information available
Sensitivity to Static Discharge	No information available

Specific Hazards Arising from the Chemical
Thermal decomposition can lead to release of irritating gases and vapors. The product causes burns of eyes, skin and mucous membranes.

Hazardous Combustion Products
Sulfur oxides Hydrogen
Protective Equipment and Precautions for Firefighters
As in any fire, wear self-contained breathing apparatus pressure-demand, MSHA/NIOSH (approved or equivalent) and full protective gear. Thermal decomposition can lead to release of irritating gases and vapors.

NFPA

Health	Flammability	Instability	Physical hazards
3	0	2	W

6. Accidental release measures

Personal Precautions	Ensure adequate ventilation. Use personal protective equipment. Evacuate personnel to safe areas. Keep people away from and upwind of spill/leak.
Environmental Precautions	Should not be released into the environment.
Methods for Containment and Clean Up	Soak up with inert absorbent material. Keep in suitable, closed containers for disposal.

7. Handling and storage

Handling	Wear personal protective equipment. Do not get in eyes, on skin, or on clothing. Use only under a chemical fume hood. Do not breathe vapors or spray mist. Do not ingest.
Storage	Keep containers tightly closed in a dry, cool and well-ventilated place. Keep away from water. Corrosives area.

8. Exposure controls / personal protection

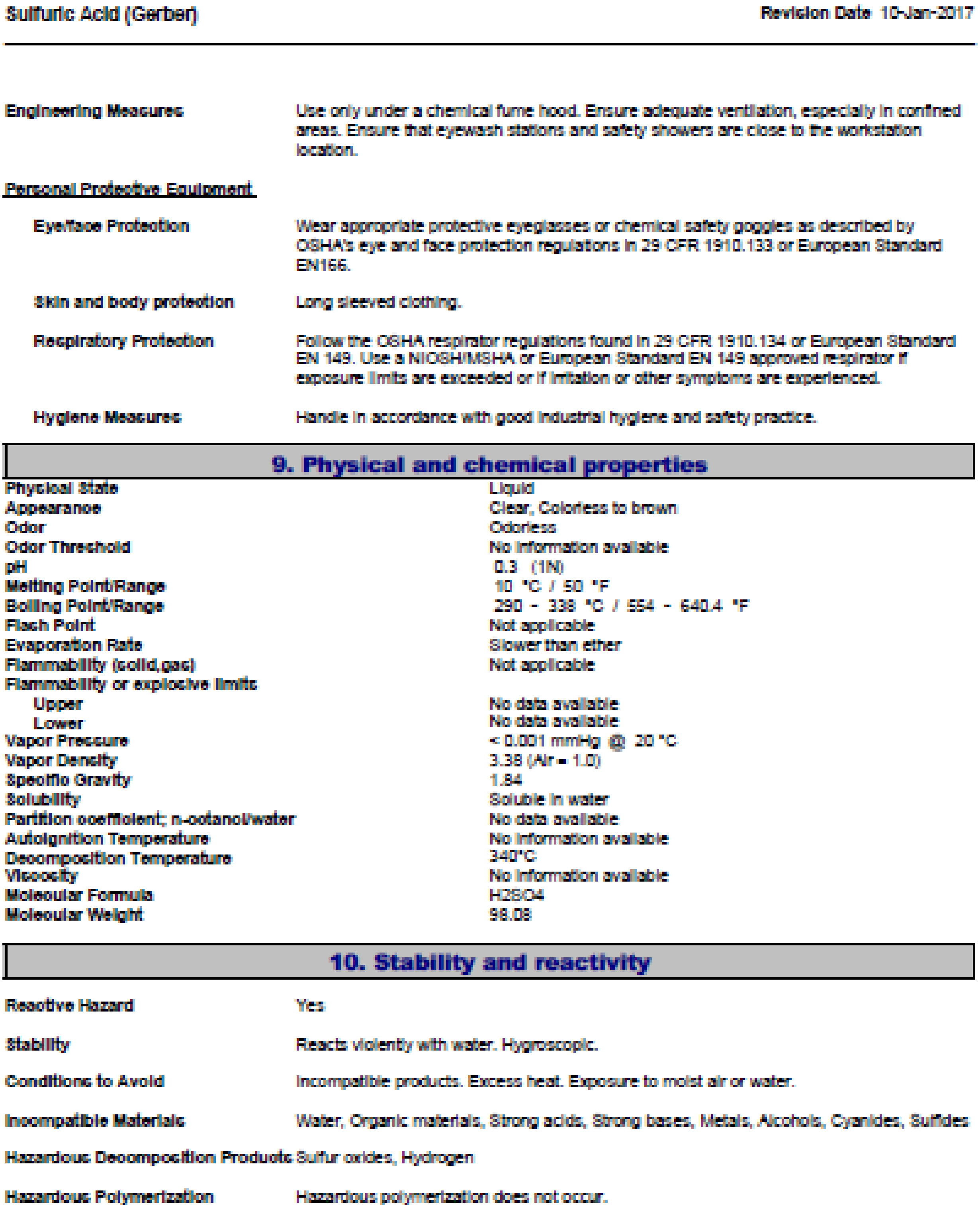
Exposure Guidelines

Component	ACGIH TLV	OSHA PEL	NIOSH IDLH
Sulfuric acid	TWA: 0.2 mg/m³	(Vacated) TWA: 1 mg/m³ TWA: 1 mg/m³	IDLH: 15 mg/m³ TWA: 1 mg/m³

Component	Quebec	Mexico OEL (TWA)	Ontario TWAEV
Sulfuric acid	TWA: 1 mg/m³ STEL: 3 mg/m³	TWA: 1 mg/m³	TWA: 0.2 mg/m³

Legend

ACGIH - American Conference of Governmental Industrial Hygienists
OSHA - Occupational Safety and Health Administration
NIOSH IDLH: The National Institute for Occupational Safety and Health Immediately Dangerous to Life or Health



Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

Engineering Measures	Use only under a chemical fume hood. Ensure adequate ventilation, especially in confined areas. Ensure that eyewash stations and safety showers are close to the workstation location.

Personal Protective Equipment

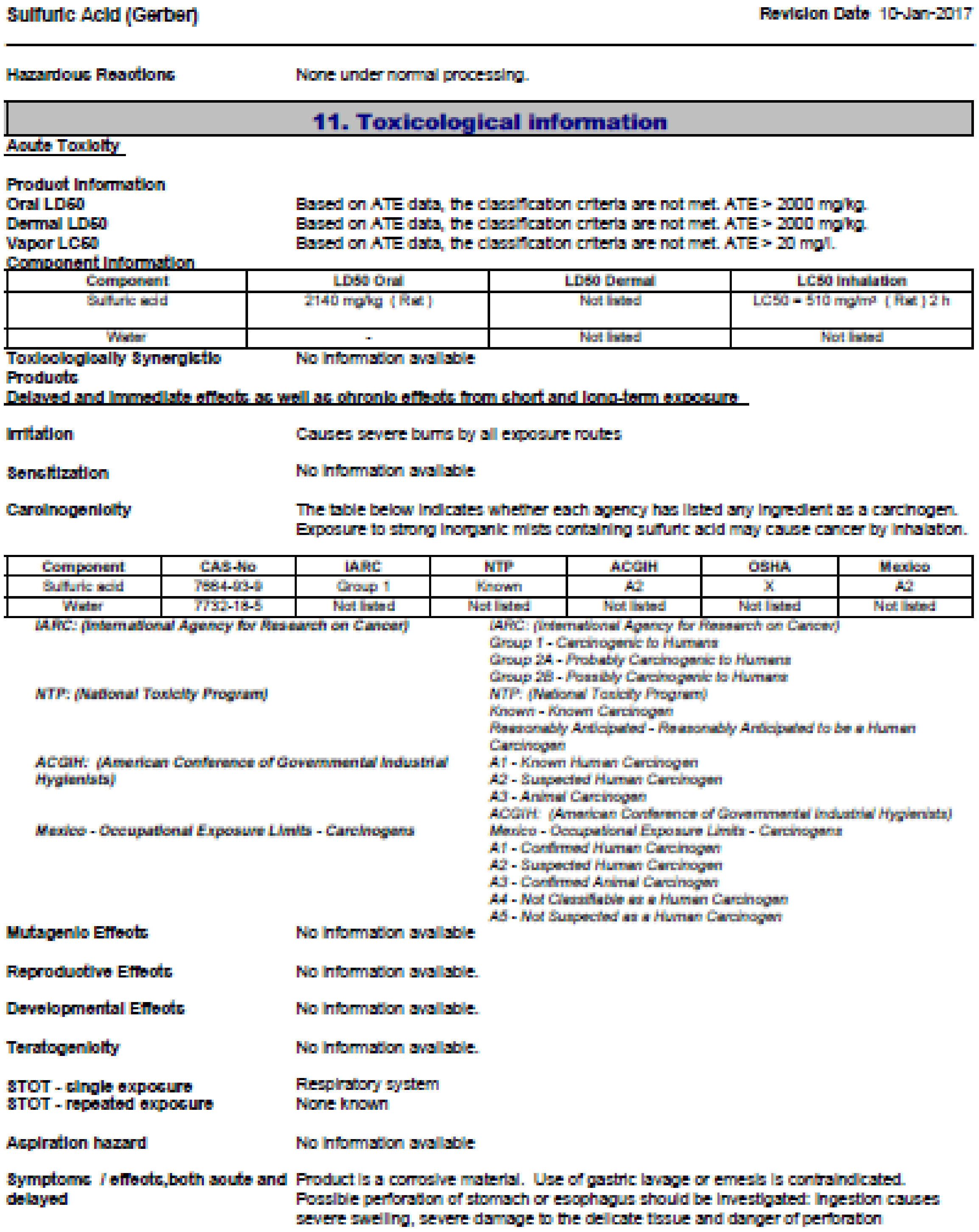
Eye/face Protection	Wear appropriate protective eyeglasses or chemical safety goggles as described by OSHA's eye and face protection regulations in 29 CFR 1910.133 or European Standard EN166.
Skin and body protection	Long sleeved clothing.
Respiratory Protection	Follow the OSHA respirator regulations found in 29 CFR 1910.134 or European Standard EN 149. Use a NIOSH/MSHA or European Standard EN 149 approved respirator if exposure limits are exceeded or if irritation or other symptoms are experienced.
Hygiene Measures	Handle in accordance with good industrial hygiene and safety practice.

9. Physical and chemical properties

Physical State	Liquid
Appearance	Clear, Colorless to brown
Odor	Odorless
Odor Threshold	No information available
pH	0.3 (1N)
Melting Point/Range	10 °C / 50 °F
Boiling Point/Range	290 - 338 °C / 554 - 640.4 °F
Flash Point	Not applicable
Evaporation Rate	Slower than ether
Flammability (solid,gas)	Not applicable
Flammability or explosive limits	
Upper	No data available
Lower	No data available
Vapor Pressure	< 0.001 mmHg @ 20 °C
Vapor Density	3.38 (Air = 1.0)
Specific Gravity	1.84
Solubility	Soluble in water
Partition coefficient; n-octanol/water	No data available
Autoignition Temperature	No information available
Decomposition Temperature	340°C
Viscosity	No information available
Molecular Formula	H_2SO_4
Molecular Weight	98.08

10. Stability and reactivity

Reactive Hazard	Yes
Stability	Reacts violently with water. Hygroscopic.
Conditions to Avoid	Incompatible products. Excess heat. Exposure to moist air or water.
Incompatible Materials	Water, Organic materials, Strong acids, Strong bases, Metals, Alcohols, Cyanides, Sulfides
Hazardous Decomposition Products	Sulfur oxides, Hydrogen
Hazardous Polymerization	Hazardous polymerization does not occur.



Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

Hazardous Reactions	None under normal processing.

11. Toxicological information

Acute Toxicity

Product Information

Oral LD50	Based on ATE data, the classification criteria are not met. ATE > 2000 mg/kg.
Dermal LD50	Based on ATE data, the classification criteria are not met. ATE > 2000 mg/kg.
Vapor LC50	Based on ATE data, the classification criteria are not met. ATE > 20 mg/l.

Component Information

Component	LD50 Oral	LD50 Dermal	LC50 Inhalation
Sulfuric acid	2140 mg/kg (Rat)	Not listed	LC50 = 510 mg/m^3 (Rat) 2 h
Water	-	Not listed	Not listed

Toxicologically Synergistic Products	No information available

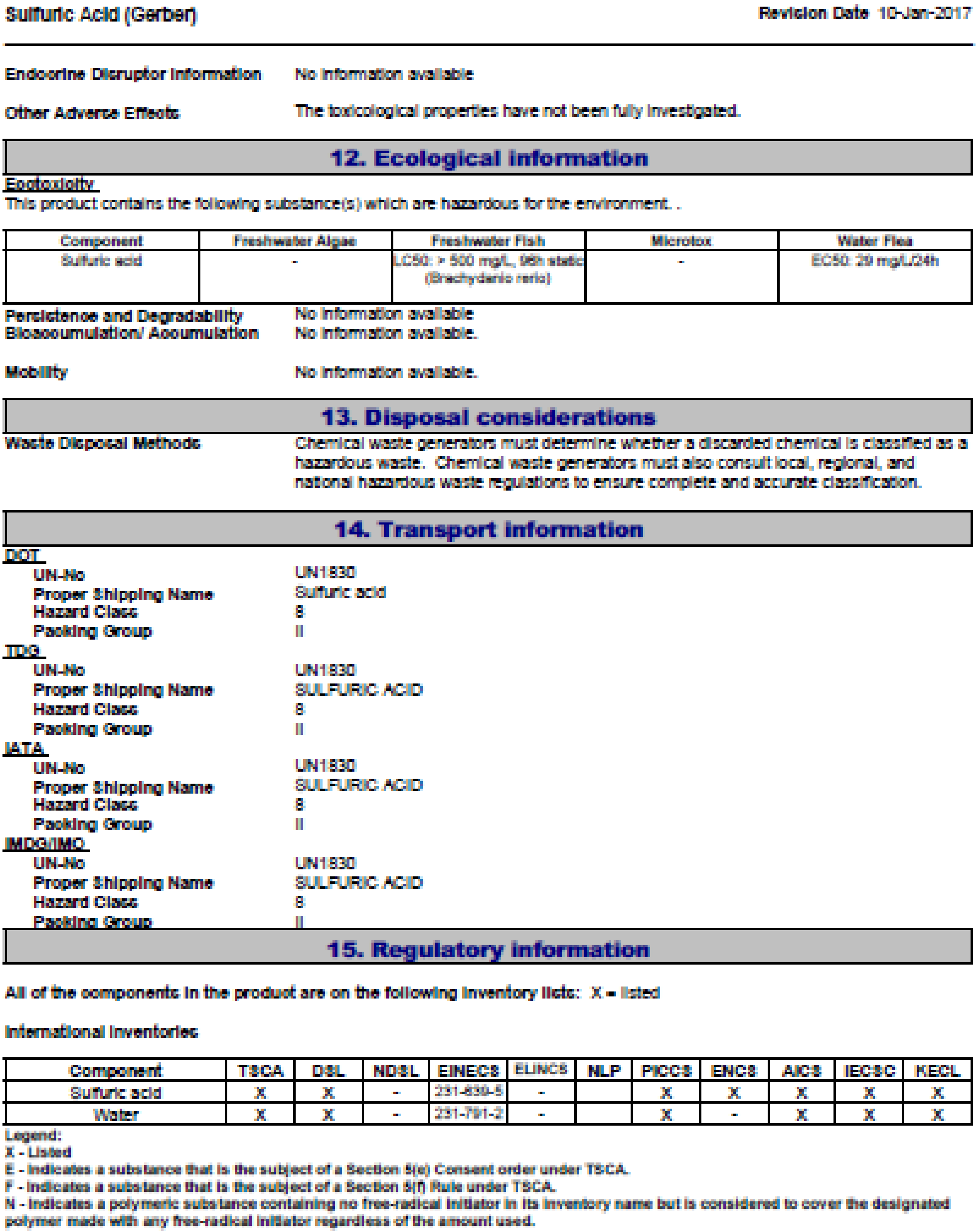
Delayed and immediate effects as well as chronic effects from short and long-term exposure

Irritation	Causes severe burns by all exposure routes
Sensitization	No information available
Carcinogenicity	The table below indicates whether each agency has listed any ingredient as a carcinogen. Exposure to strong inorganic mists containing sulfuric acid may cause cancer by inhalation.

Component	CAS-No	IARC	NTP	ACGIH	OSHA	Mexico
Sulfuric acid	7664-93-9	Group 1	Known	A2	X	A2
Water	7732-18-5	Not listed	Not listed	Not listed	Not listed	Not listed

IARC: (International Agency for Research on Cancer)	*IARC: (International Agency for Research on Cancer)* *Group 1 - Carcinogenic to Humans* *Group 2A - Probably Carcinogenic to Humans* *Group 2B - Possibly Carcinogenic to Humans*
NTP: (National Toxicity Program)	*NTP: (National Toxicity Program)* *Known - Known Carcinogen* *Reasonably Anticipated - Reasonably Anticipated to be a Human Carcinogen*
ACGIH: (American Conference of Governmental Industrial Hygienists)	*A1 - Known Human Carcinogen* *A2 - Suspected Human Carcinogen* *A3 - Animal Carcinogen* *ACGIH: (American Conference of Governmental Industrial Hygienists)*
Mexico - Occupational Exposure Limits - Carcinogens	*Mexico - Occupational Exposure Limits - Carcinogens* *A1 - Confirmed Human Carcinogen* *A2 - Suspected Human Carcinogen* *A3 - Confirmed Animal Carcinogen* *A4 - Not Classifiable as a Human Carcinogen* *A5 - Not Suspected as a Human Carcinogen*

Mutagenic Effects	No information available
Reproductive Effects	No information available.
Developmental Effects	No information available.
Teratogenicity	No information available.
STOT - single exposure	Respiratory system
STOT - repeated exposure	None known
Aspiration hazard	No information available
Symptoms / effects,both acute and delayed	Product is a corrosive material. Use of gastric lavage or emesis is contraindicated. Possible perforation of stomach or esophagus should be investigated: Ingestion causes severe swelling, severe damage to the delicate tissue and danger of perforation



Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

Endocrine Disruptor Information No information available

Other Adverse Effects The toxicological properties have not been fully investigated.

12. Ecological information

Ecotoxicity

This product contains the following substance(s) which are hazardous for the environment. .

Component	Freshwater Algae	Freshwater Fish	Microtox	Water Flea
Sulfuric acid	-	LC50: > 500 mg/L, 96h static (Brachydanio rerio)	-	EC50: 29 mg/L/24h

Persistence and Degradability No information available

Bioaccumulation/ Accumulation No information available.

Mobility No information available.

13. Disposal considerations

Waste Disposal Methods Chemical waste generators must determine whether a discarded chemical is classified as a hazardous waste. Chemical waste generators must also consult local, regional, and national hazardous waste regulations to ensure complete and accurate classification.

14. Transport information

DOT

UN-No	UN1830
Proper Shipping Name	Sulfuric acid
Hazard Class	8
Packing Group	II

TDG

UN-No	UN1830
Proper Shipping Name	SULFURIC ACID
Hazard Class	8
Packing Group	II

IATA

UN-No	UN1830
Proper Shipping Name	SULFURIC ACID
Hazard Class	8
Packing Group	II

IMDG/IMO

UN-No	UN1830
Proper Shipping Name	SULFURIC ACID
Hazard Class	8
Packing Group	II

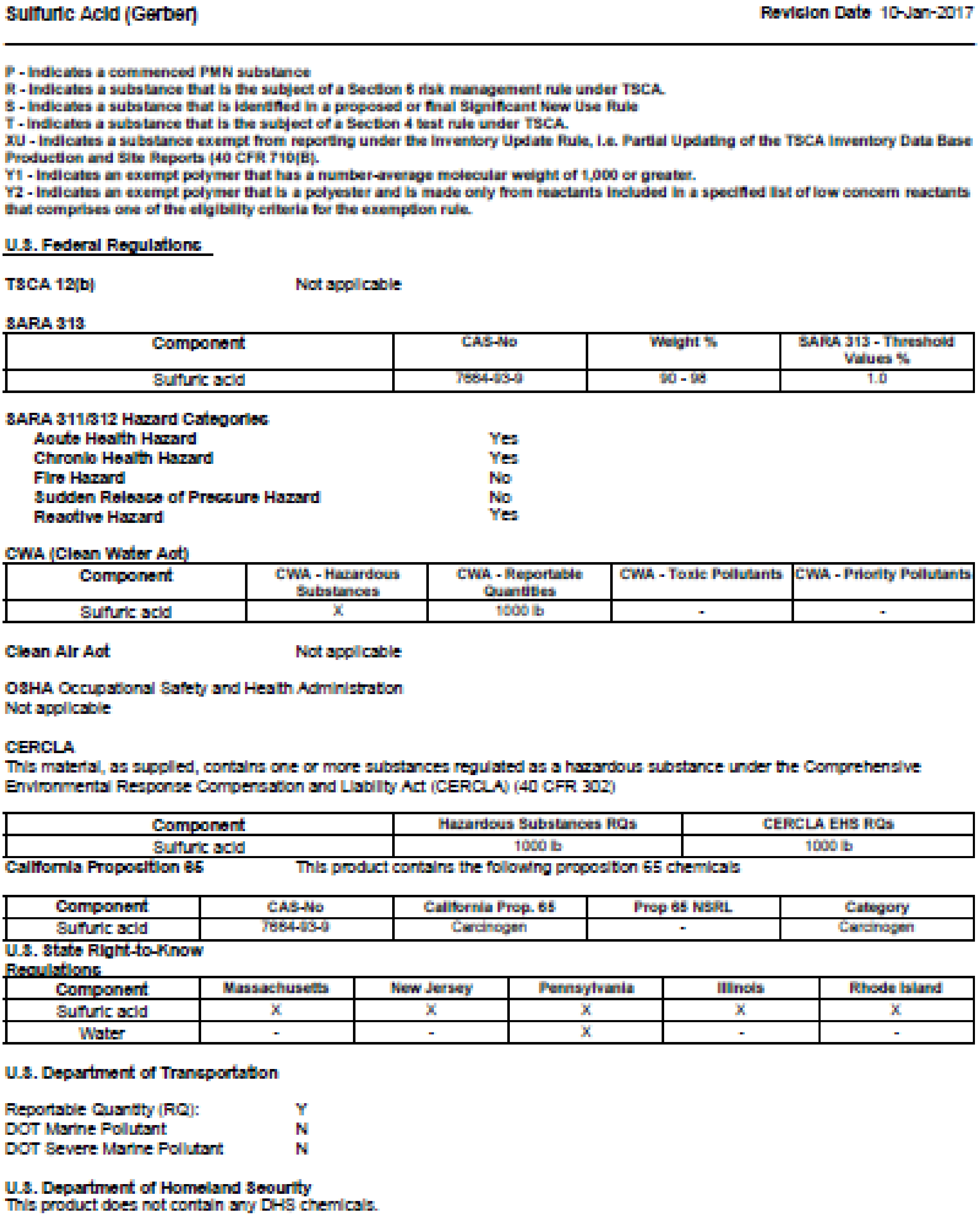
15. Regulatory information

All of the components in the product are on the following Inventory lists: X = listed

International Inventories

Component	TSCA	DSL	NDSL	EINECS	ELINCS	NLP	PICCS	ENCS	AICS	IECSC	KECL
Sulfuric acid	X	X	-	231-639-5	-		X	X	X	X	X
Water	X	X	-	231-791-2	-		X	-	X	X	X

Legend:
X - Listed
E - Indicates a substance that is the subject of a Section 5(e) Consent order under TSCA.
F - Indicates a substance that is the subject of a Section 5(f) Rule under TSCA.
N - Indicates a polymeric substance containing no free-radical initiator in its inventory name but is considered to cover the designated polymer made with any free-radical initiator regardless of the amount used.



Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

P - Indicates a commenced PMN substance
R - Indicates a substance that is the subject of a Section 6 risk management rule under TSCA.
S - Indicates a substance that is identified in a proposed or final Significant New Use Rule
T - Indicates a substance that is the subject of a Section 4 test rule under TSCA.
XU - Indicates a substance exempt from reporting under the Inventory Update Rule, i.e. Partial Updating of the TSCA Inventory Data Base Production and Site Reports (40 CFR 710(B).
Y1 - Indicates an exempt polymer that has a number-average molecular weight of 1,000 or greater.
Y2 - Indicates an exempt polymer that is a polyester and is made only from reactants included in a specified list of low concern reactants that comprises one of the eligibility criteria for the exemption rule.

U.S. Federal Regulations

TSCA 12(b) Not applicable

SARA 313

Component	CAS-No	Weight %	SARA 313 - Threshold Values %
Sulfuric acid	7664-93-9	90 - 98	1.0

SARA 311/312 Hazard Categories

Acute Health Hazard	Yes
Chronic Health Hazard	Yes
Fire Hazard	No
Sudden Release of Pressure Hazard	No
Reactive Hazard	Yes

CWA (Clean Water Act)

Component	CWA - Hazardous Substances	CWA - Reportable Quantities	CWA - Toxic Pollutants	CWA - Priority Pollutants
Sulfuric acid	X	1000 lb	-	-

Clean Air Act Not applicable

OSHA Occupational Safety and Health Administration
Not applicable

CERCLA
This material, as supplied, contains one or more substances regulated as a hazardous substance under the Comprehensive Environmental Response Compensation and Liability Act (CERCLA) (40 CFR 302)

Component	Hazardous Substances RQs	CERCLA EHS RQs
Sulfuric acid	1000 lb	1000 lb

California Proposition 65 This product contains the following proposition 65 chemicals

Component	CAS-No	California Prop. 65	Prop 65 NSRL	Category
Sulfuric acid	7664-93-9	Carcinogen	-	Carcinogen

U.S. State Right-to-Know Regulations

Component	Massachusetts	New Jersey	Pennsylvania	Illinois	Rhode Island
Sulfuric acid	X	X	X	X	X
Water	-	-	X	-	-

U.S. Department of Transportation

Reportable Quantity (RQ):	Y
DOT Marine Pollutant	N
DOT Severe Marine Pollutant	N

U.S. Department of Homeland Security
This product does not contain any DHS chemicals.

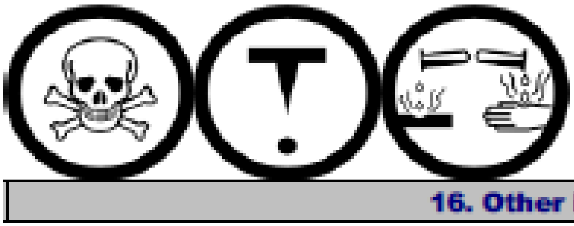
Sulfuric Acid (Gerber) **Revision Date** 10-Jan-2017

Other International Regulations

Mexico - Grade	No information available

Canada
This product has been classified in accordance with the hazard criteria of the Controlled Products Regulations (CPR) and the MSDS contains all the information required by the CPR

WHMIS Hazard Class	D1A Very toxic materials E Corrosive material D2A Very toxic materials

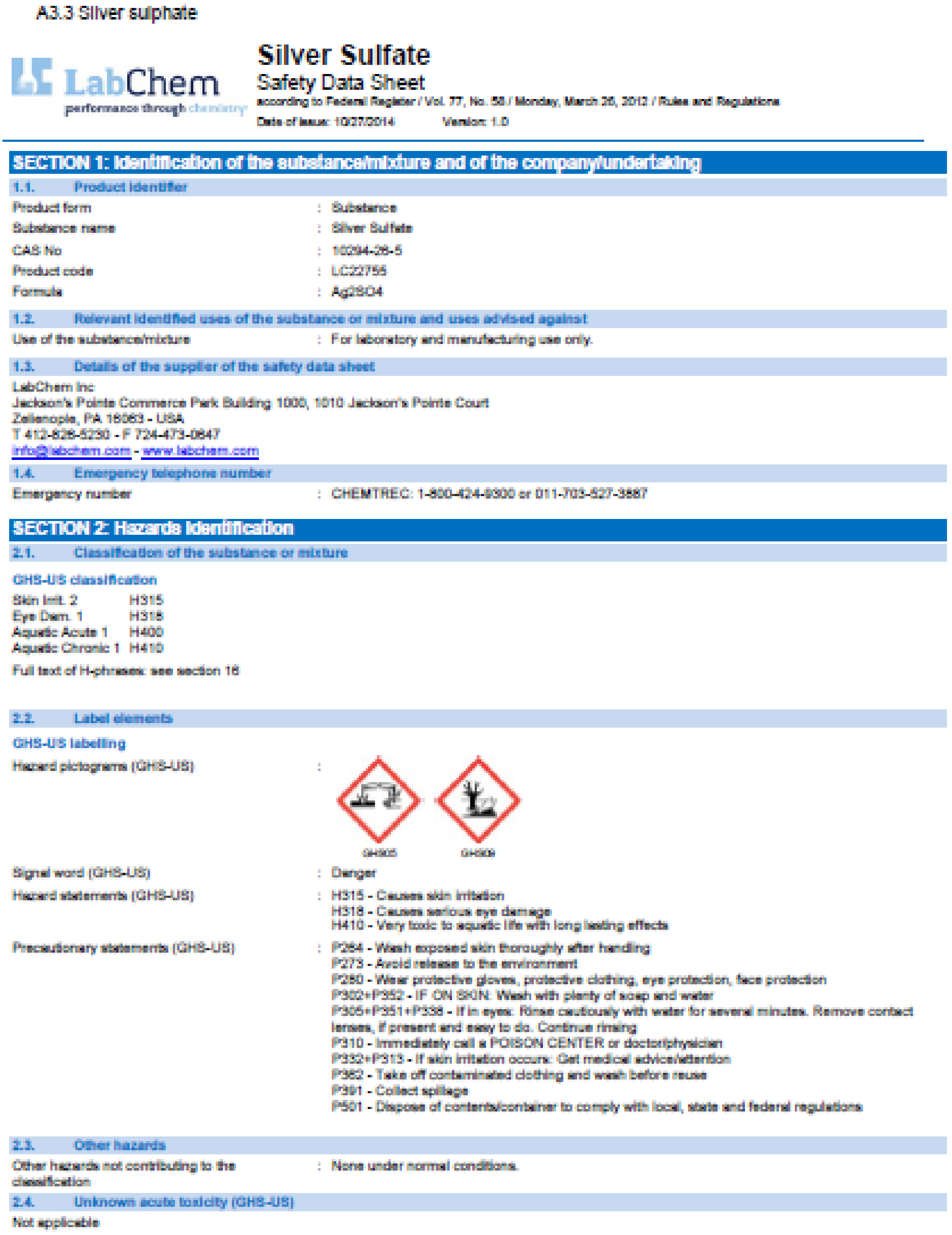


16. Other information

Prepared By	Regulatory Affairs Thermo Fisher Scientific Email: EMSDS.RA@thermofisher.com
Creation Date	12-Nov-2010
Revision Date	10-Jan-2017
Print Date	10-Jan-2017
Revision Summary	SDS sections updated; 2

Disclaimer
The information provided in this Safety Data Sheet is correct to the best of our knowledge, information and belief at the date of its publication. The information given is designed only as a guidance for safe handling, use, processing, storage, transportation, disposal and release and is not to be considered a warranty or quality specification. The information relates only to the specific material designated and may not be valid for such material used in combination with any other materials or in any process, unless specified in the text

A3.4. Silver sulphate



A3.3 Silver sulphate

LabChem
performance through chemistry

Silver Sulfate
Safety Data Sheet
according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations
Date of issue: 10/27/2014 Version: 1.0

SECTION 1: Identification of the substance/mixture and of the company/undertaking

1.1. Product identifier

Product form : Substance
Substance name : Silver Sulfate
CAS No : 10294-26-5
Product code : LC22755
Formula : Ag2SO4

1.2. Relevant identified uses of the substance or mixture and uses advised against

Use of the substance/mixture : For laboratory and manufacturing use only.

1.3. Details of the supplier of the safety data sheet

LabChem Inc
Jackson's Pointe Commerce Park Building 1000, 1010 Jackson's Pointe Court
Zelienople, PA 16063 - USA
T 412-826-5230 - F 724-473-0647
info@labchem.com - www.labchem.com

1.4. Emergency telephone number

Emergency number : CHEMTREC: 1-800-424-9300 or 011-703-527-3887

SECTION 2: Hazards identification

2.1. Classification of the substance or mixture

GHS-US classification

Skin Irrit. 2 H315
Eye Dam. 1 H318
Aquatic Acute 1 H400
Aquatic Chronic 1 H410

Full text of H-phrases: see section 16

2.2. Label elements

GHS-US labelling

Hazard pictograms (GHS-US) :

Signal word (GHS-US) : Danger

Hazard statements (GHS-US) : H315 - Causes skin irritation
H318 - Causes serious eye damage
H410 - Very toxic to aquatic life with long lasting effects

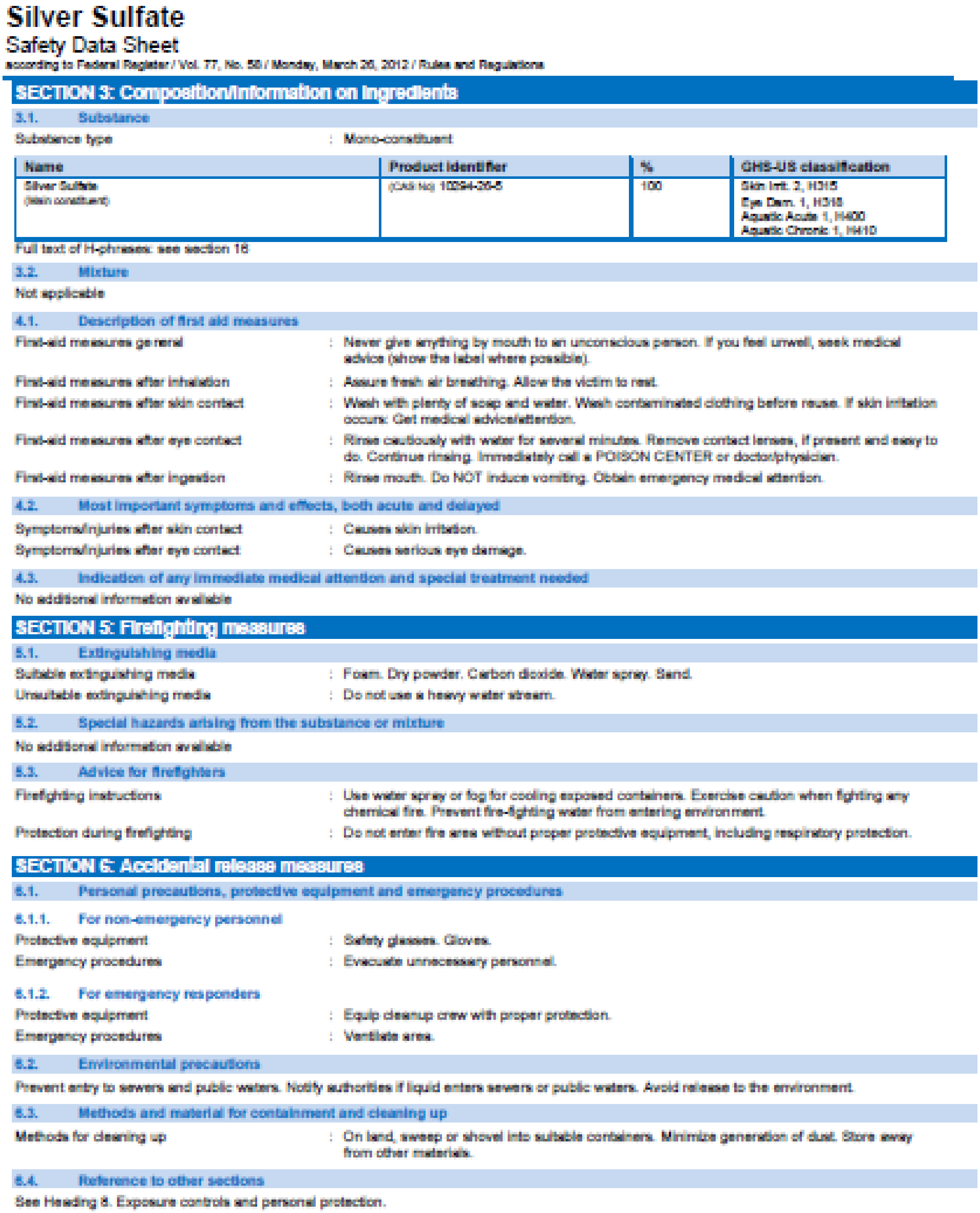
Precautionary statements (GHS-US) : P264 - Wash exposed skin thoroughly after handling
P273 - Avoid release to the environment
P280 - Wear protective gloves, protective clothing, eye protection, face protection
P302+P352 - IF ON SKIN: Wash with plenty of soap and water
P305+P351+P338 - If in eyes: Rinse 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
P332+P313 - If skin irritation occurs: Get medical advice/attention
P362 - Take off contaminated clothing and wash before reuse
P391 - Collect spillage
P501 - Dispose of contents/container to comply with local, state and federal regulations

2.3. Other hazards

Other hazards not contributing to the classification : None under normal conditions.

2.4. Unknown acute toxicity (GHS-US)

Not applicable



Silver Sulfate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

SECTION 3: Composition/information on ingredients

3.1. Substance

Substance type : Mono-constituent

Name	Product identifier	%	GHS-US classification
Silver Sulfate (Main constituent)	(CAS No) 10294-26-5	100	Skin Irrit. 2, H315 Eye Dam. 1, H318 Aquatic Acute 1, H400 Aquatic Chronic 1, H410

Full text of H-phrases: see section 16

3.2. Mixture

Not applicable

4.1. Description of first aid measures

First-aid measures general : Never give anything by mouth to an unconscious person. If you feel unwell, seek medical advice (show the label where possible).

First-aid measures after inhalation : Assure fresh air breathing. Allow the victim to rest.

First-aid measures after skin contact : Wash with plenty of soap and water. Wash contaminated clothing before reuse. If skin irritation occurs: Get medical advice/attention.

First-aid measures after eye contact : Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER or doctor/physician.

First-aid measures after ingestion : Rinse mouth. Do NOT induce vomiting. Obtain emergency medical attention.

4.2. Most important symptoms and effects, both acute and delayed

Symptoms/injuries after skin contact : Causes skin irritation.

Symptoms/injuries after eye contact : Causes serious eye damage.

4.3. Indication of any immediate medical attention and special treatment needed

No additional information available

SECTION 5: Firefighting measures

5.1. Extinguishing media

Suitable extinguishing media : Foam. Dry powder. Carbon dioxide. Water spray. Sand.

Unsuitable extinguishing media : Do not use a heavy water stream.

5.2. Special hazards arising from the substance or mixture

No additional information available

5.3. Advice for firefighters

Firefighting instructions : Use water spray or fog for cooling exposed containers. Exercise caution when fighting any chemical fire. Prevent fire-fighting water from entering environment.

Protection during firefighting : Do not enter fire area without proper protective equipment, including respiratory protection.

SECTION 6: Accidental release measures

6.1. Personal precautions, protective equipment and emergency procedures

6.1.1. For non-emergency personnel

Protective equipment : Safety glasses. Gloves.

Emergency procedures : Evacuate unnecessary personnel.

6.1.2. For emergency responders

Protective equipment : Equip cleanup crew with proper protection.

Emergency procedures : Ventilate area.

6.2. Environmental precautions

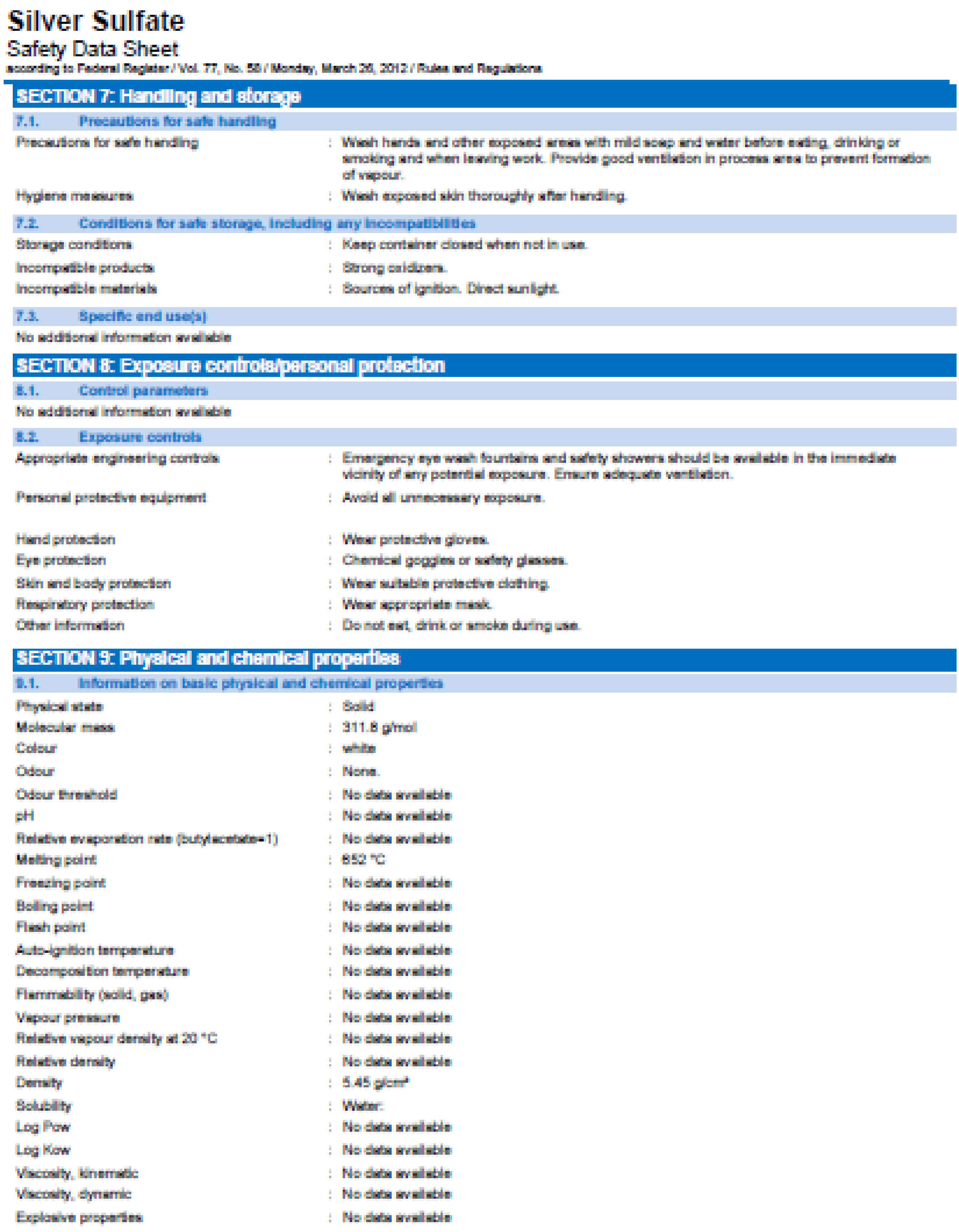
Prevent entry to sewers and public waters. Notify authorities if liquid enters sewers or public waters. Avoid release to the environment.

6.3. Methods and material for containment and cleaning up

Methods for cleaning up : On land, sweep or shovel into suitable containers. Minimize generation of dust. Store away from other materials.

6.4. Reference to other sections

See Heading 8. Exposure controls and personal protection.



Silver Sulfate

Safety Data Sheet

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SECTION 7: Handling and storage

7.1. Precautions for safe handling

Precautions for safe handling	:	Wash hands and other exposed areas with mild soap and water before eating, drinking or smoking and when leaving work. Provide good ventilation in process area to prevent formation of vapour.
Hygiene measures	:	Wash exposed skin thoroughly after handling.

7.2. Conditions for safe storage, including any incompatibilities

Storage conditions	:	Keep container closed when not in use.
Incompatible products	:	Strong oxidizers.
Incompatible materials	:	Sources of ignition. Direct sunlight.

7.3. Specific end use(s)

No additional information available

SECTION 8: Exposure controls/personal protection

8.1. Control parameters

No additional information available

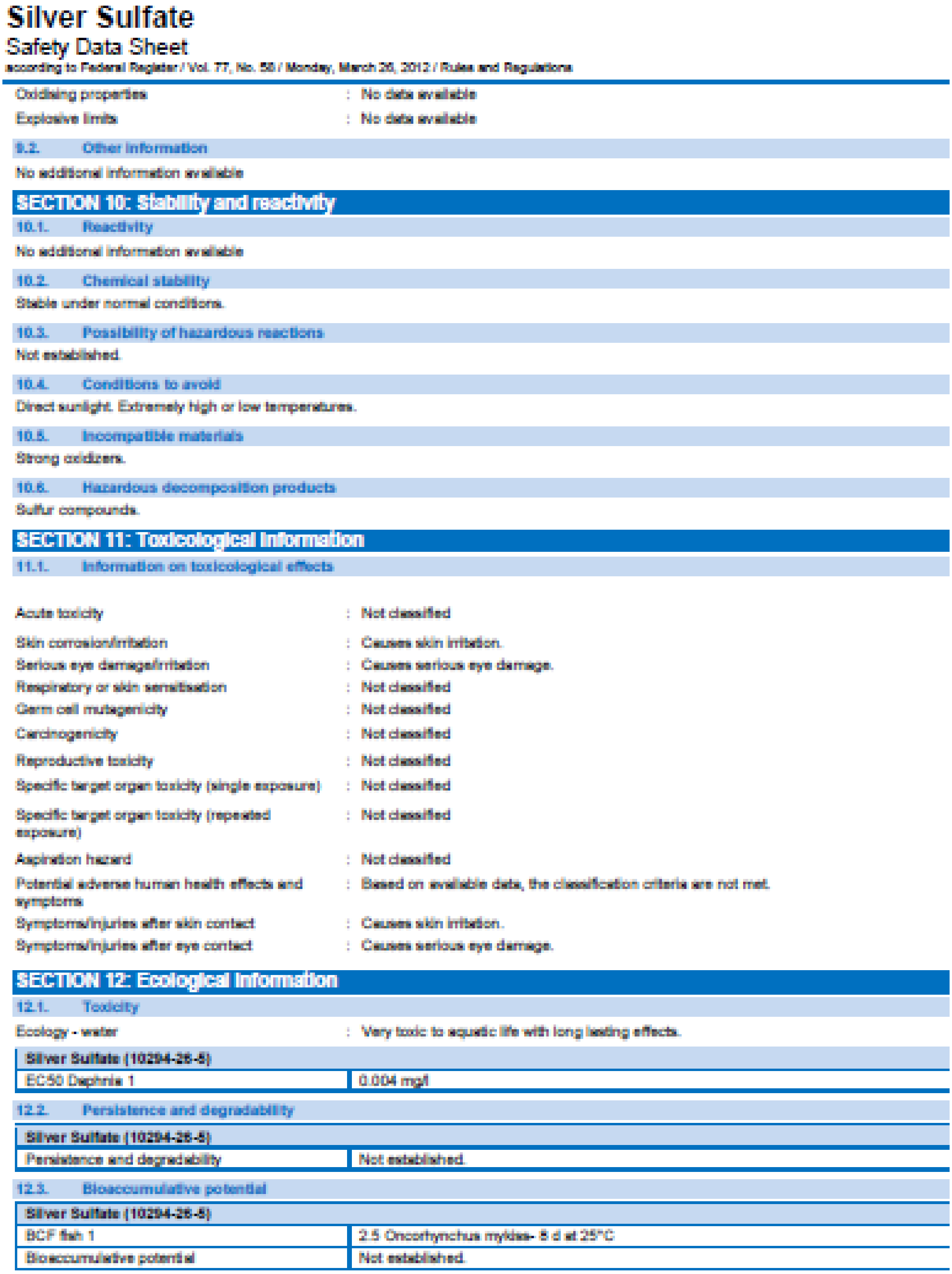
8.2. Exposure controls

Appropriate engineering controls	:	Emergency eye wash fountains and safety showers should be available in the immediate vicinity of any potential exposure. Ensure adequate ventilation.
Personal protective equipment	:	Avoid all unnecessary exposure.
Hand protection	:	Wear protective gloves.
Eye protection	:	Chemical goggles or safety glasses.
Skin and body protection	:	Wear suitable protective clothing.
Respiratory protection	:	Wear appropriate mask.
Other information	:	Do not eat, drink or smoke during use.

SECTION 9: Physical and chemical properties

9.1. Information on basic physical and chemical properties

Physical state	:	Solid
Molecular mass	:	311.8 g/mol
Colour	:	white
Odour	:	None.
Odour threshold	:	No data available
pH	:	No data available
Relative evaporation rate (butylacetate=1)	:	No data available
Melting point	:	652 °C
Freezing point	:	No data available
Boiling point	:	No data available
Flash point	:	No data available
Auto-ignition temperature	:	No data available
Decomposition temperature	:	No data available
Flammability (solid, gas)	:	No data available
Vapour pressure	:	No data available
Relative vapour density at 20 °C	:	No data available
Relative density	:	No data available
Density	:	5.45 g/cm³
Solubility	:	Water:
Log Pow	:	No data available
Log Kow	:	No data available
Viscosity, kinematic	:	No data available
Viscosity, dynamic	:	No data available
Explosive properties	:	No data available



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Oxidising properties : No data available

Explosive limits : No data available

9.2. Other information

No additional information available

SECTION 10: Stability and reactivity

10.1. Reactivity

No additional information available

10.2. Chemical stability

Stable under normal conditions.

10.3. Possibility of hazardous reactions

Not established.

10.4. Conditions to avoid

Direct sunlight. Extremely high or low temperatures.

10.5. Incompatible materials

Strong oxidizers.

10.6. Hazardous decomposition products

Sulfur compounds.

SECTION 11: Toxicological information

11.1. Information on toxicological effects

Acute toxicity : Not classified

Skin corrosion/irritation : Causes skin irritation.

Serious eye damage/irritation : Causes serious eye damage.

Respiratory or skin sensitisation : Not classified

Germ cell mutagenicity : Not classified

Carcinogenicity : Not classified

Reproductive toxicity : Not classified

Specific target organ toxicity (single exposure) : Not classified

Specific target organ toxicity (repeated exposure) : Not classified

Aspiration hazard : Not classified

Potential adverse human health effects and symptoms : Based on available data, the classification criteria are not met.

Symptoms/injuries after skin contact : Causes skin irritation.

Symptoms/injuries after eye contact : Causes serious eye damage.

SECTION 12: Ecological information

12.1. Toxicity

Ecology - water : Very toxic to aquatic life with long lasting effects.

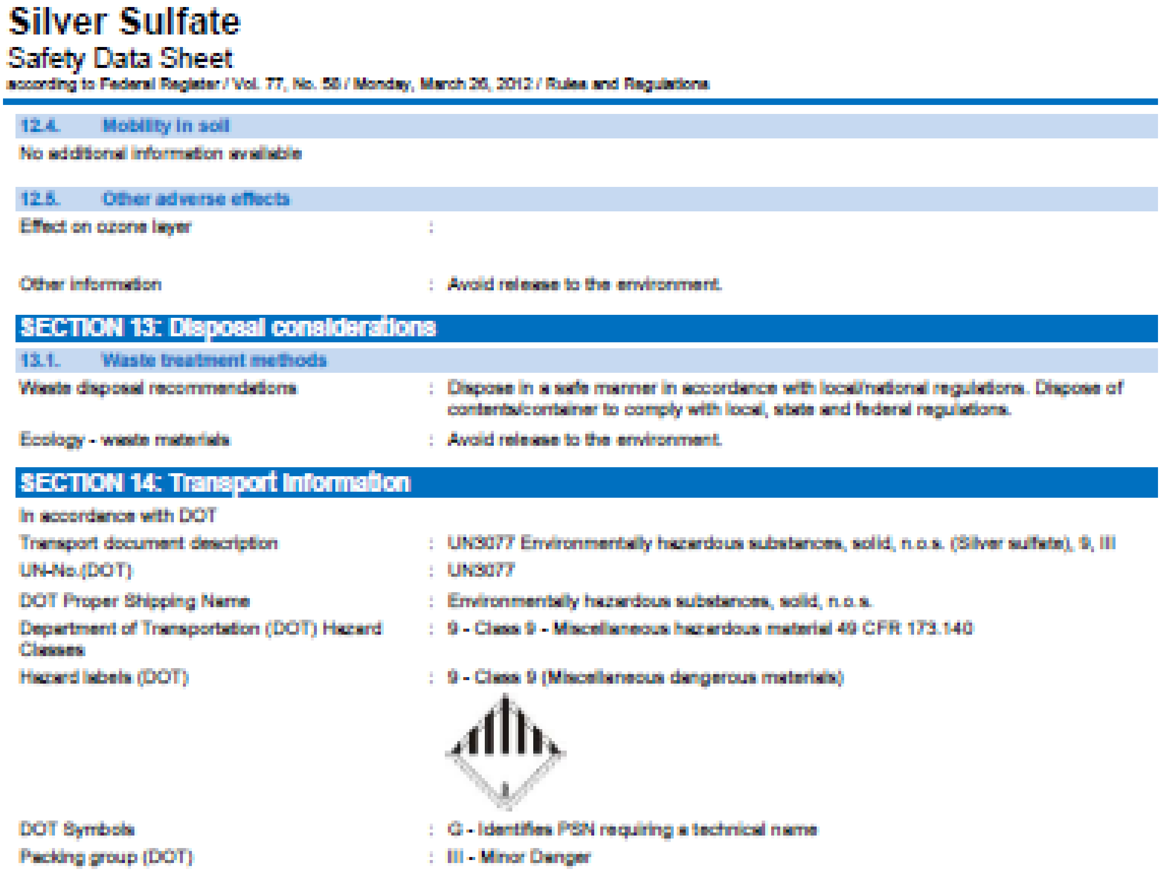
Silver Sulfate (10294-26-5)	
EC50 Daphnia 1	0.004 mg/l

12.2. Persistence and degradability

Silver Sulfate (10294-26-5)	
Persistence and degradability	Not established.

12.3. Bioaccumulative potential

Silver Sulfate (10294-26-5)	
BCF fish 1	2.5 Oncorhynchus mykiss- 8 d at 25°C
Bioaccumulative potential	Not established.



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12.4. Mobility in soil

No additional information available

12.5. Other adverse effects

Effect on ozone layer :

Other information : Avoid release to the environment.

SECTION 13: Disposal considerations

13.1. Waste treatment methods

Waste disposal recommendations : Dispose in a safe manner in accordance with local/national regulations. Dispose of contents/container to comply with local, state and federal regulations.

Ecology - waste materials : Avoid release to the environment.

SECTION 14: Transport information

In accordance with DOT

Transport document description : UN3077 Environmentally hazardous substances, solid, n.o.s. (Silver sulfate), 9, III

UN-No.(DOT) : UN3077

DOT Proper Shipping Name : Environmentally hazardous substances, solid, n.o.s.

Department of Transportation (DOT) Hazard Classes : 9 - Class 9 - Miscellaneous hazardous material 49 CFR 173.140

Hazard labels (DOT) : 9 - Class 9 (Miscellaneous dangerous materials)

DOT Symbols : G - Identifies PSN requiring a technical name

Packing group (DOT) : III - Minor Danger

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DOT Special Provisions (49 CFR 172.102)	: **8 -** A hazardous substance that is not a hazardous waste may be shipped under the shipping description "Other regulated substances, liquid or solid, n.o.s.", as appropriate. In addition, for solid materials, special provision B54 applies. **146 -** This description may be used for a material that poses a hazard to the environment but does not meet the definition for a hazardous waste or a hazardous substance, as defined in 171.8 of this subchapter, or any hazard class as defined in Part 173 of this subchapter, if it is designated as environmentally hazardous by the Competent Authority of the country of origin, transit or destination. **335 -** Mixtures of solids that are not subject to this subchapter and environmentally hazardous liquids or solids may be classified as "Environmentally hazardous substances, solid, n.o.s," UN3077 and may be transported under this entry, provided there is no free liquid visible at the time the material is loaded or at the time the packaging or transport unit is closed. Each transport unit must be leakproof when used as bulk packaging. **A112 -** Notwithstanding the quantity limits shown in Column (9A) and (9B) for this entry, the following IBCs are authorized for transportation aboard passenger and cargo-only aircraft. Each IBC may not exceed a maximum net quantity of 1,000 kg: a. Metal: 11A, 11B, 11N, 21A, 21B and 21N b. Rigid plastics: 11H1, 11H2, 21H1 and 21H2 c. Composite with plastic inner receptacle: 11HZ1, 11HZ2, 21HZ1 and 21HZ2 d. Fiberboard: 11G e. Wooden: 11C, 11D and 11F (with inner liners) f. Flexible: 13H2, 13H3, 13H4, 13H5, 13L2, 13L3, 13L4, 13M1 and 13M2 (flexible IBCs must be sift-proof and water resistant or must be fitted with a sift-proof and water resistant liner). **B54 -** Open-top, sift-proof rail cars are also authorized. **IB8 -** Authorized IBCs: Metal (11A, 11B, 11N, 21A, 21B, 21N, 31A, 31B and 31N); Rigid plastics (11H1, 11H2, 21H1, 21H2, 31H1 and 31H2); Composite (11HZ1, 11HZ2, 21HZ1, 21HZ2, 31HZ1 and 31HZ2); Fiberboard (11G); Wooden (11C, 11D and 11F); Flexible (13H1, 13H2, 13H3, 13H4, 13H5, 13L1, 13L2, 13L3, 13L4, 13M1 or 13M2). **IP3 -** Flexible IBCs must be sift-proof and water-resistant or must be fitted with a sift-proof and water-resistant liner. **N20 -** A 5M1 multi-wall paper bag is authorized if transported in a closed transport vehicle. **T1 -** 1.5 178.274(d)(2) Normal............. 178.275(d)(2) **TP33 -** The portable tank instruction assigned for this substance applies for granular and powdered solids and for solids which are filled and discharged at temperatures above their melting point which are cooled and transported as a solid mass. Solid substances transported or offered for transport above their melting point are authorized for transportation in portable tanks conforming to the provisions of portable tank instruction T4 for solid substances of packing group III or T7 for solid substances of packing group II, unless a tank with more stringent requirements for minimum shell thickness, maximum allowable working pressure, pressure-relief devices or bottom outlets are assigned in which case the more stringent tank instruction and special provisions shall apply. Filling limits must be in accordance with portable tank special provision TP3. Solids meeting the definition of an elevated temperature material must be transported in accordance with the applicable requirements of this subchapter.
DOT Packaging Exceptions (49 CFR 173.xxx)	: 155
DOT Packaging Non Bulk (49 CFR 173.xxx)	: 213
DOT Packaging Bulk (49 CFR 173.xxx)	: 240
DOT Quantity Limitations Passenger aircraft/rail (49 CFR 173.27)	: No limit
DOT Quantity Limitations Cargo aircraft only (49 CFR 175.75)	: No limit
DOT Vessel Stowage Location	: **A -** The material may be stowed "on deck" or "under deck" on a cargo vessel and on a passenger vessel.

Additional information	
Other information	: No supplementary information available.

ADR

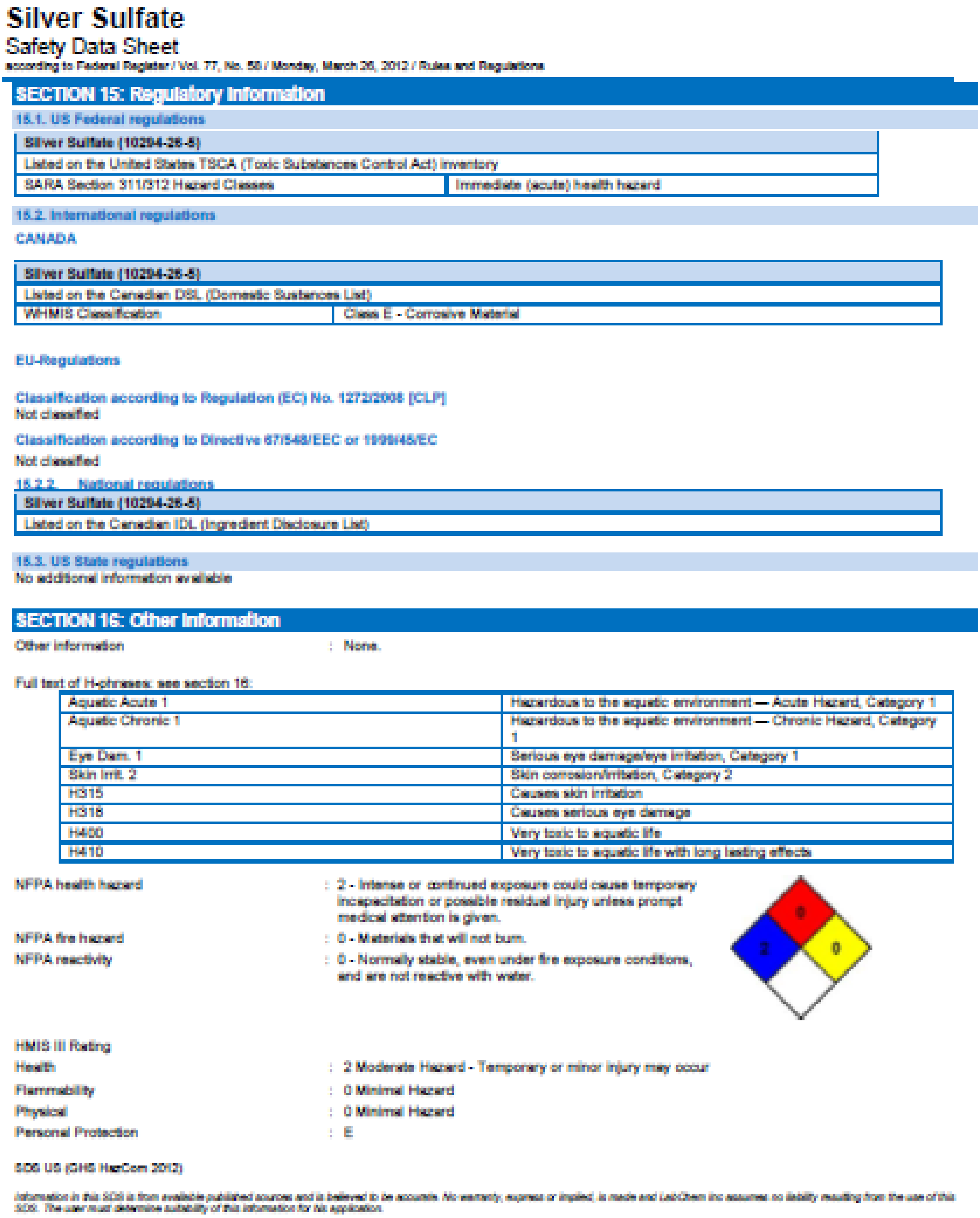
No additional information available

Transport by sea

No additional information available

Air transport

No additional information available



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SECTION 15: Regulatory Information

15.1. US Federal regulations

Silver Sulfate (10294-26-5)	
Listed on the United States TSCA (Toxic Substances Control Act) inventory	
SARA Section 311/312 Hazard Classes	Immediate (acute) health hazard

15.2. International regulations

CANADA

Silver Sulfate (10294-26-5)	
Listed on the Canadian DSL (Domestic Sustances List)	
WHMIS Classification	Class E - Corrosive Material

EU-Regulations

Classification according to Regulation (EC) No. 1272/2008 [CLP]

Not classified

Classification according to Directive 67/548/EEC or 1999/45/EC

Not classified

15.2.2. National regulations

Silver Sulfate (10294-26-5)
Listed on the Canadian IDL (Ingredient Disclosure List)

15.3. US State regulations

No additional information available

SECTION 16: Other Information

Other information : None.

Full text of H-phrases: see section 16:

Aquatic Acute 1	Hazardous to the aquatic environment — Acute Hazard, Category 1
Aquatic Chronic 1	Hazardous to the aquatic environment — Chronic Hazard, Category 1
Eye Dam. 1	Serious eye damage/eye irritation, Category 1
Skin Irrit. 2	Skin corrosion/irritation, Category 2
H315	Causes skin irritation
H318	Causes serious eye damage
H400	Very toxic to aquatic life
H410	Very toxic to aquatic life with long lasting effects

NFPA health hazard : 2 - Intense or continued exposure could cause temporary incapacitation or possible residual injury unless prompt medical attention is given.

NFPA fire hazard : 0 - Materials that will not burn.

NFPA reactivity : 0 - Normally stable, even under fire exposure conditions, and are not reactive with water.

HMIS III Rating

Health : 2 Moderate Hazard - Temporary or minor injury may occur

Flammability : 0 Minimal Hazard

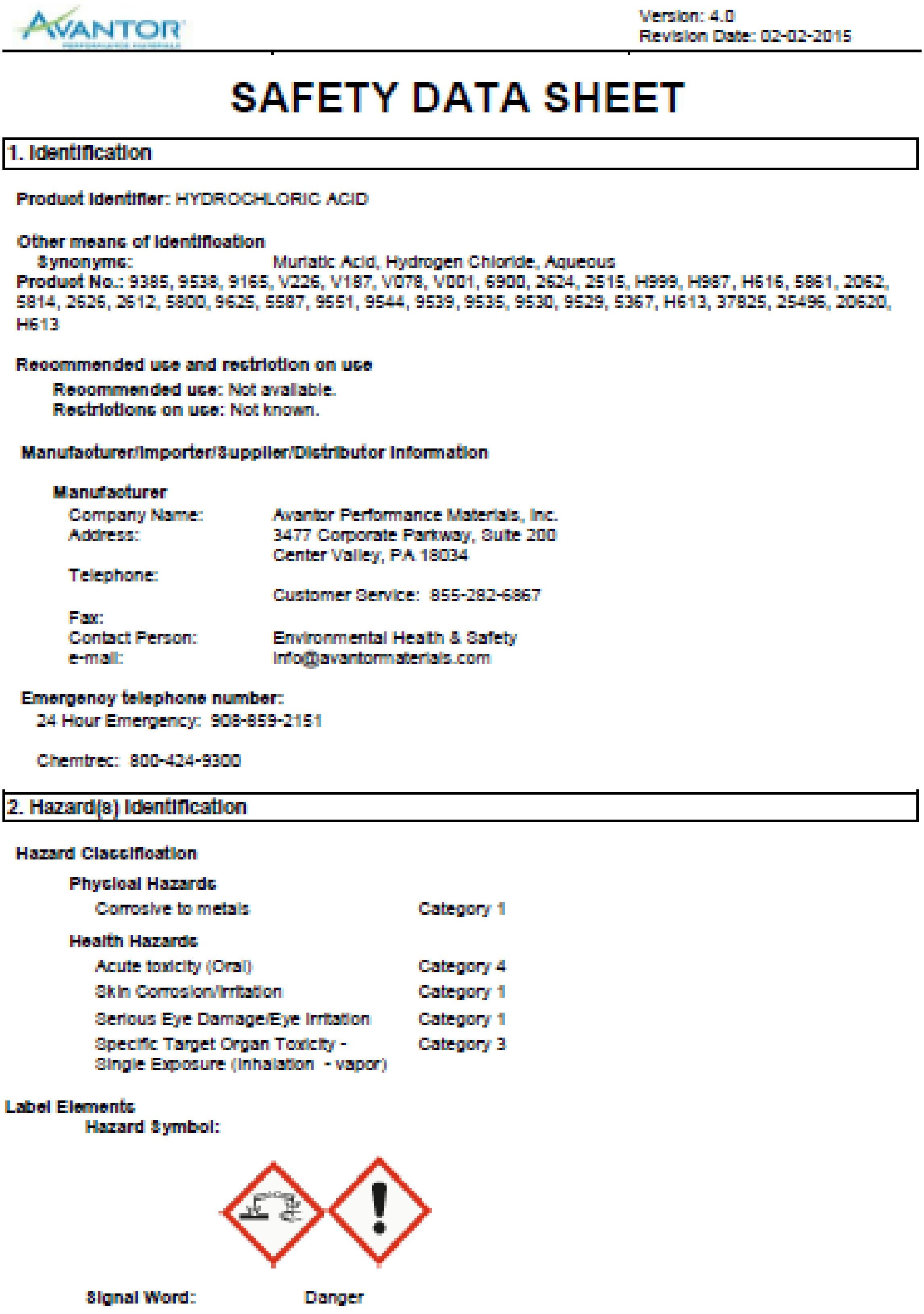
Physical : 0 Minimal Hazard

Personal Protection : E

SDS US (GHS HazCom 2012)

Information in this SDS is from available published sources and is believed to be accurate. No warranty, express or implied, is made and LabChem Inc assumes no liability resulting from the use of this SDS. The user must determine suitability of this information for his application.

A3.5. Chlorhydric acid



AVANTOR

Version: 4.0
Revision Date: 02-02-2015

SAFETY DATA SHEET

1. Identification

Product identifier: HYDROCHLORIC ACID

Other means of identification
Synonyms: Muriatic Acid, Hydrogen Chloride, Aqueous
Product No.: 9385, 9538, 9165, V226, V187, V078, V001, 6900, 2624, 2515, H999, H987, H616, 5861, 2062, 5814, 2626, 2612, 5800, 9625, 5587, 9551, 9544, 9539, 9535, 9530, 9529, 5367, H613, 37825, 25496, 20620, H613

Recommended use and restriction on use
Recommended use: Not available.
Restrictions on use: Not known.

Manufacturer/Importer/Supplier/Distributor Information

Manufacturer
Company Name: Avantor Performance Materials, Inc.
Address: 3477 Corporate Parkway, Suite 200
Center Valley, PA 18034
Telephone:
Customer Service: 855-282-6867
Fax:
Contact Person: Environmental Health & Safety
e-mail: info@avantormaterials.com

Emergency telephone number:
24 Hour Emergency: 908-859-2151

Chemtrec: 800-424-9300

2. Hazard(s) identification

Hazard Classification

Physical Hazards

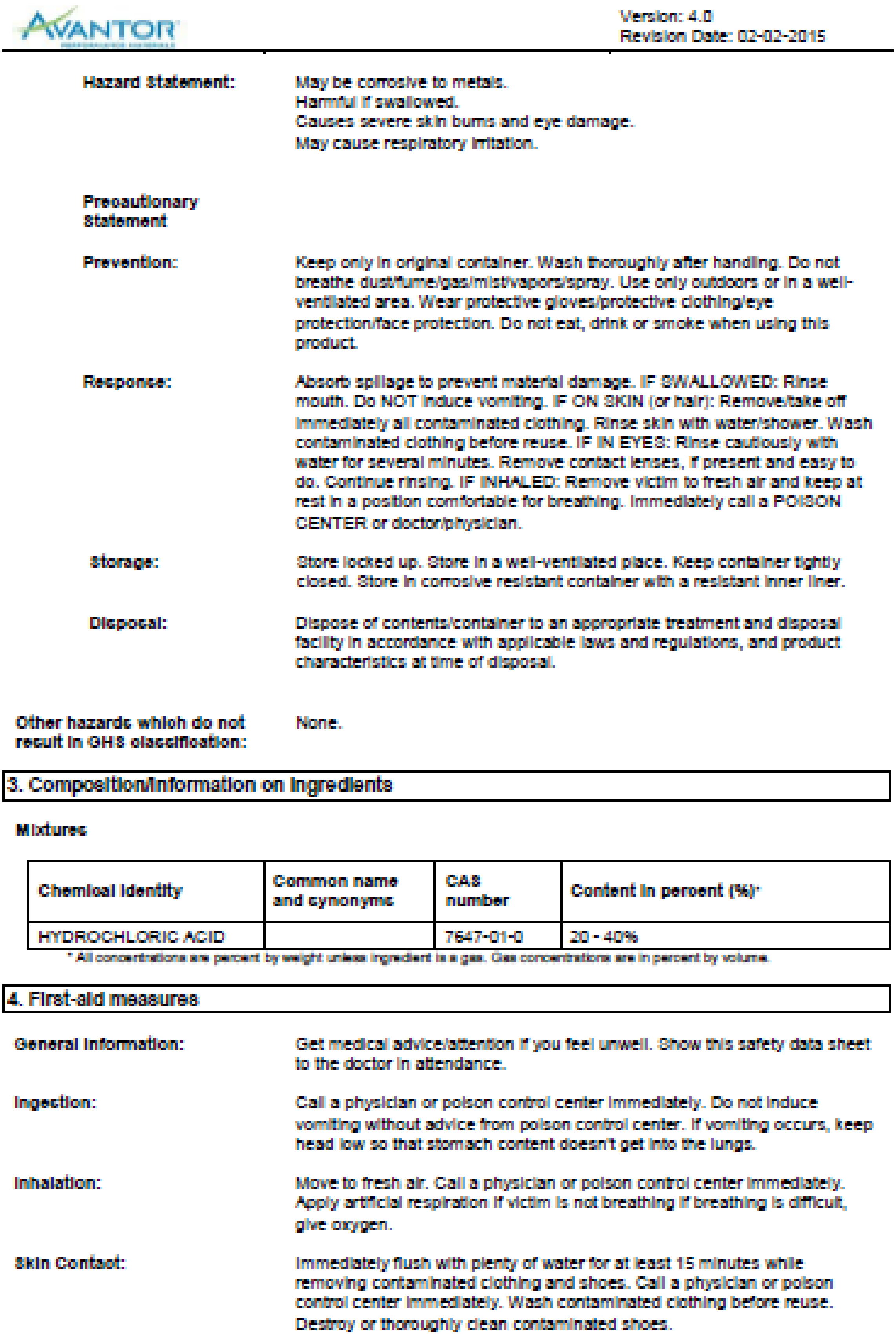
Corrosive to metals	Category 1

Health Hazards

Acute toxicity (Oral)	Category 4
Skin Corrosion/Irritation	Category 1
Serious Eye Damage/Eye Irritation	Category 1
Specific Target Organ Toxicity - Single Exposure (Inhalation - vapor)	Category 3

Label Elements
Hazard Symbol:

Signal Word: Danger



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Hazard Statement: May be corrosive to metals.
Harmful if swallowed.
Causes severe skin burns and eye damage.
May cause respiratory irritation.

Precautionary Statement

Prevention: Keep only in original container. Wash thoroughly after handling. Do not breathe dust/fume/gas/mist/vapors/spray. Use only outdoors or in a well-ventilated area. Wear protective gloves/protective clothing/eye protection/face protection. Do not eat, drink or smoke when using this product.

Response: Absorb spillage to prevent material damage. IF SWALLOWED: Rinse mouth. Do NOT induce vomiting. IF ON SKIN (or hair): Remove/take off immediately all contaminated clothing. Rinse skin with water/shower. Wash contaminated clothing before reuse. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing. Immediately call a POISON CENTER or doctor/physician.

Storage: Store locked up. Store in a well-ventilated place. Keep container tightly closed. Store in corrosive resistant container with a resistant inner liner.

Disposal: Dispose of contents/container to an appropriate treatment and disposal facility in accordance with applicable laws and regulations, and product characteristics at time of disposal.

Other hazards which do not result in GHS classification: None.

3. Composition/information on ingredients

Mixtures

Chemical Identity	Common name and synonyms	CAS number	Content in percent (%)*
HYDROCHLORIC ACID		7647-01-0	20 - 40%

* All concentrations are percent by weight unless ingredient is a gas. Gas concentrations are in percent by volume.

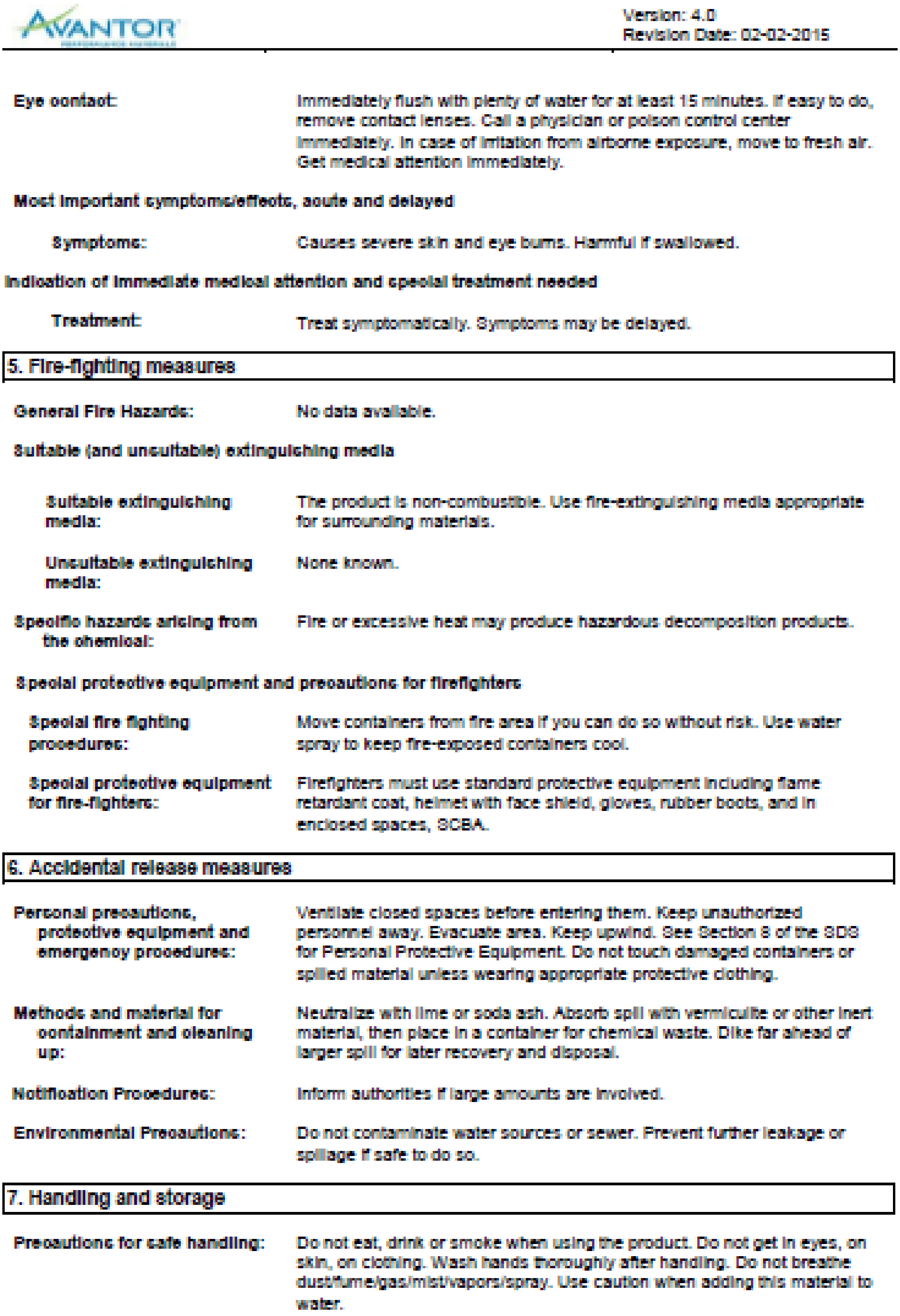
4. First-aid measures

General information: Get medical advice/attention if you feel unwell. Show this safety data sheet to the doctor in attendance.

Ingestion: Call a physician or poison control center immediately. Do not induce vomiting without advice from poison control center. If vomiting occurs, keep head low so that stomach content doesn't get into the lungs.

Inhalation: Move to fresh air. Call a physician or poison control center immediately. Apply artificial respiration if victim is not breathing if breathing is difficult, give oxygen.

Skin Contact: Immediately flush with plenty of water for at least 15 minutes while removing contaminated clothing and shoes. Call a physician or poison control center immediately. Wash contaminated clothing before reuse. Destroy or thoroughly clean contaminated shoes.



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Eye contact:	Immediately flush with plenty of water for at least 15 minutes. If easy to do, remove contact lenses. Call a physician or poison control center immediately. In case of irritation from airborne exposure, move to fresh air. Get medical attention immediately.

Most important symptoms/effects, acute and delayed

Symptoms:	Causes severe skin and eye burns. Harmful if swallowed.

Indication of immediate medical attention and special treatment needed

Treatment:	Treat symptomatically. Symptoms may be delayed.

5. Fire-fighting measures

General Fire Hazards:	No data available.

Suitable (and unsuitable) extinguishing media

Suitable extinguishing media:	The product is non-combustible. Use fire-extinguishing media appropriate for surrounding materials.
Unsuitable extinguishing media:	None known.
Specific hazards arising from the chemical:	Fire or excessive heat may produce hazardous decomposition products.

Special protective equipment and precautions for firefighters

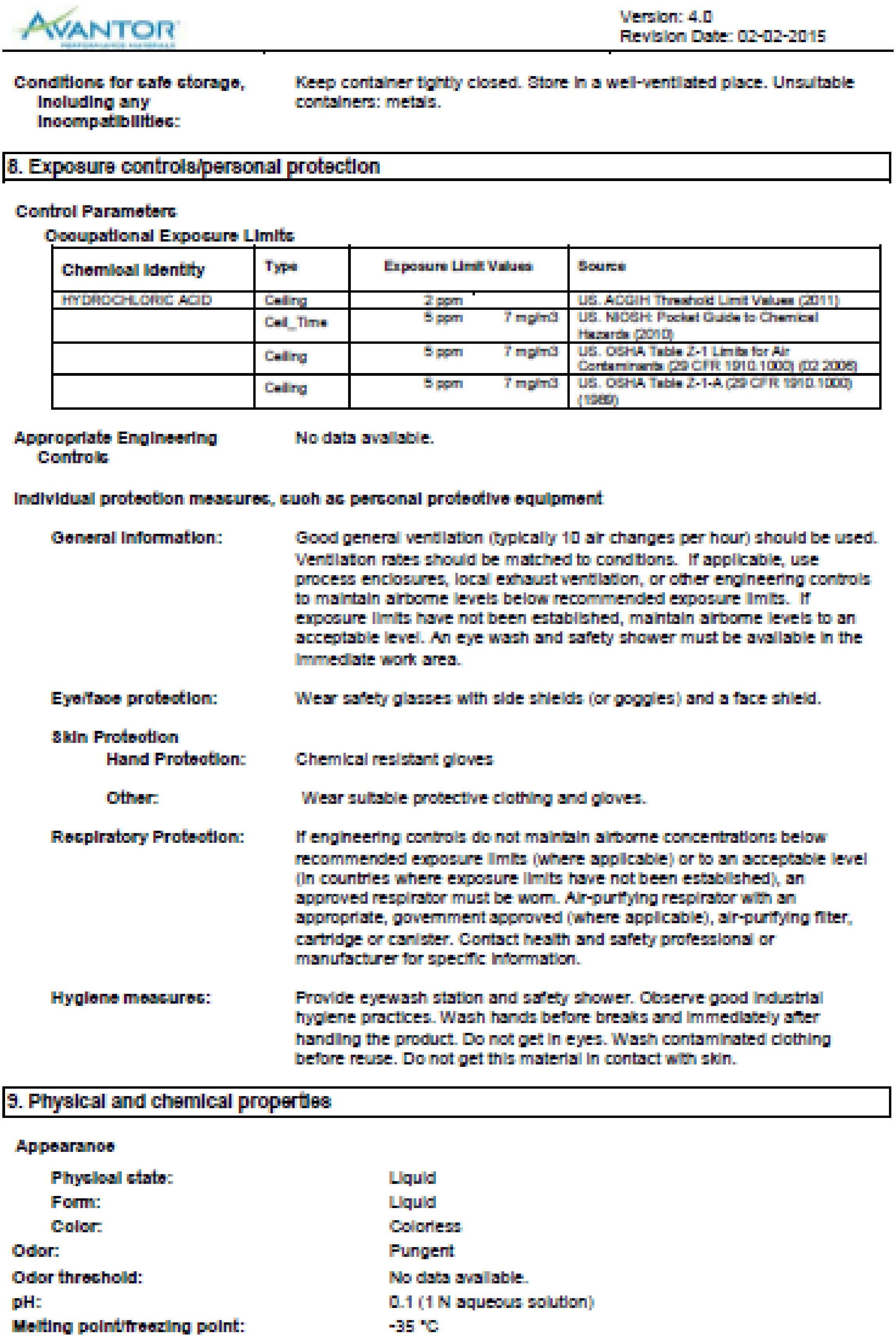
Special fire fighting procedures:	Move containers from fire area if you can do so without risk. Use water spray to keep fire-exposed containers cool.
Special protective equipment for fire-fighters:	Firefighters must use standard protective equipment including flame retardant coat, helmet with face shield, gloves, rubber boots, and in enclosed spaces, SCBA.

6. Accidental release measures

Personal precautions, protective equipment and emergency procedures:	Ventilate closed spaces before entering them. Keep unauthorized personnel away. Evacuate area. Keep upwind. See Section 8 of the SDS for Personal Protective Equipment. Do not touch damaged containers or spilled material unless wearing appropriate protective clothing.
Methods and material for containment and cleaning up:	Neutralize with lime or soda ash. Absorb spill with vermiculite or other inert material, then place in a container for chemical waste. Dike far ahead of larger spill for later recovery and disposal.
Notification Procedures:	Inform authorities if large amounts are involved.
Environmental Precautions:	Do not contaminate water sources or sewer. Prevent further leakage or spillage if safe to do so.

7. Handling and storage

Precautions for safe handling:	Do not eat, drink or smoke when using the product. Do not get in eyes, on skin, on clothing. Wash hands thoroughly after handling. Do not breathe dust/fume/gas/mist/vapors/spray. Use caution when adding this material to water.



Conditions for safe storage, including any incompatibilities: Keep container tightly closed. Store in a well-ventilated place. Unsuitable containers: metals.

8. Exposure controls/personal protection

Control Parameters

Occupational Exposure Limits

Chemical Identity	Type	Exposure Limit Values		Source
HYDROCHLORIC ACID	Ceiling	2 ppm		US. ACGIH Threshold Limit Values (2011)
	Ceil_Time	5 ppm	7 mg/m3	US. NIOSH: Pocket Guide to Chemical Hazards (2010)
	Ceiling	5 ppm	7 mg/m3	US. OSHA Table Z-1 Limits for Air Contaminants (29 CFR 1910.1000) (02 2006)
	Ceiling	5 ppm	7 mg/m3	US. OSHA Table Z-1-A (29 CFR 1910.1000) (1989)

Appropriate Engineering Controls No data available.

Individual protection measures, such as personal protective equipment

General information: Good general ventilation (typically 10 air changes per hour) should be used. Ventilation rates should be matched to conditions. If applicable, use process enclosures, local exhaust ventilation, or other engineering controls to maintain airborne levels below recommended exposure limits. If exposure limits have not been established, maintain airborne levels to an acceptable level. An eye wash and safety shower must be available in the immediate work area.

Eye/face protection: Wear safety glasses with side shields (or goggles) and a face shield.

Skin Protection

Hand Protection: Chemical resistant gloves

Other: Wear suitable protective clothing and gloves.

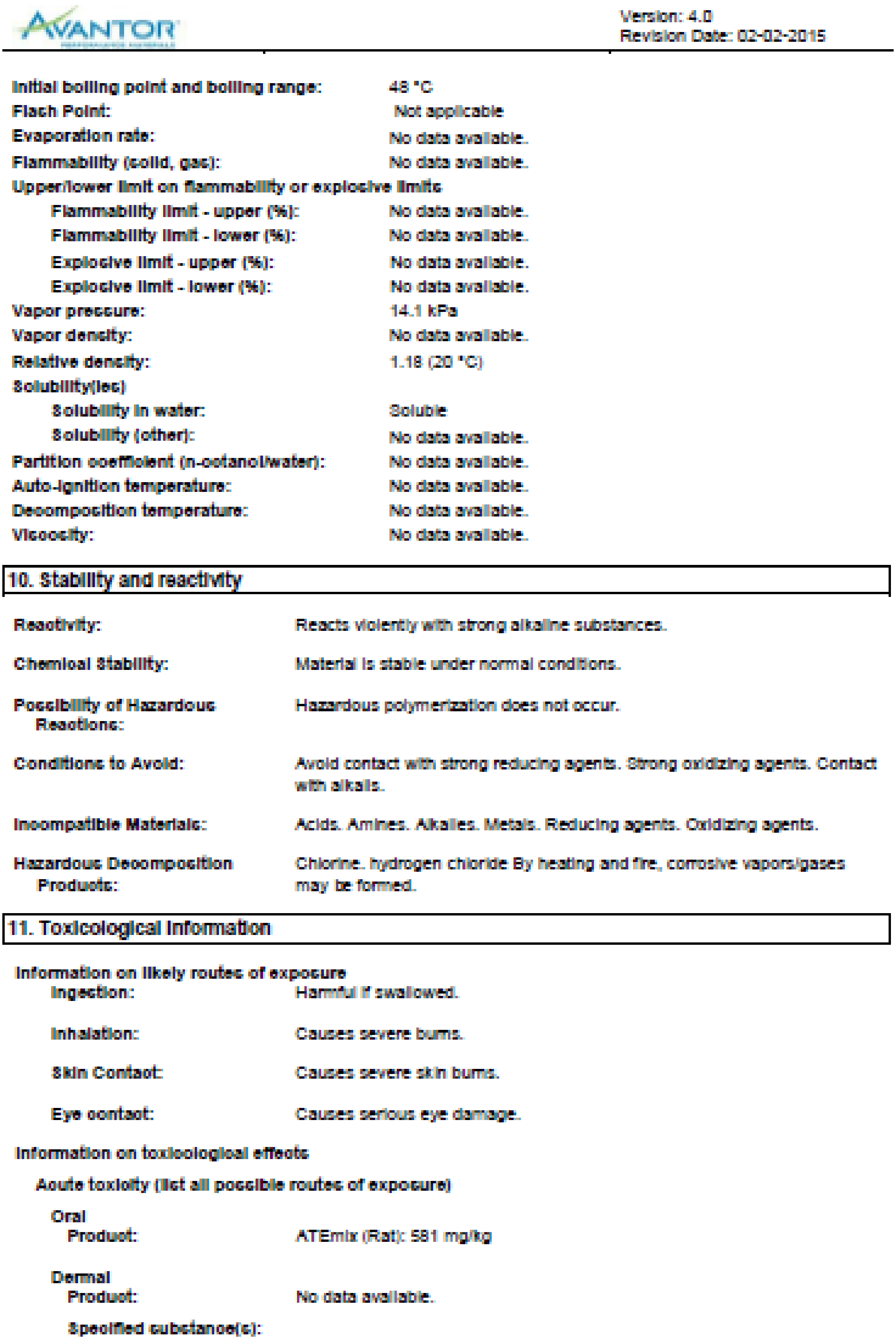
Respiratory Protection: If engineering controls do not maintain airborne concentrations below recommended exposure limits (where applicable) or to an acceptable level (in countries where exposure limits have not been established), an approved respirator must be worn. Air-purifying respirator with an appropriate, government approved (where applicable), air-purifying filter, cartridge or canister. Contact health and safety professional or manufacturer for specific information.

Hygiene measures: Provide eyewash station and safety shower. Observe good industrial hygiene practices. Wash hands before breaks and immediately after handling the product. Do not get in eyes. Wash contaminated clothing before reuse. Do not get this material in contact with skin.

9. Physical and chemical properties

Appearance

Physical state:	Liquid
Form:	Liquid
Color:	Colorless
Odor:	Pungent
Odor threshold:	No data available.
pH:	0.1 (1 N aqueous solution)
Melting point/freezing point:	-35 °C



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Initial boiling point and boiling range:	48 °C
Flash Point:	Not applicable
Evaporation rate:	No data available.
Flammability (solid, gas):	No data available.
Upper/lower limit on flammability or explosive limits	
Flammability limit - upper (%):	No data available.
Flammability limit - lower (%):	No data available.
Explosive limit - upper (%):	No data available.
Explosive limit - lower (%):	No data available.
Vapor pressure:	14.1 kPa
Vapor density:	No data available.
Relative density:	1.18 (20 °C)
Solubility(ies)	
Solubility in water:	Soluble
Solubility (other):	No data available.
Partition coefficient (n-octanol/water):	No data available.
Auto-ignition temperature:	No data available.
Decomposition temperature:	No data available.
Viscosity:	No data available.

10. Stability and reactivity

Reactivity:	Reacts violently with strong alkaline substances.
Chemical Stability:	Material is stable under normal conditions.
Possibility of Hazardous Reactions:	Hazardous polymerization does not occur.
Conditions to Avoid:	Avoid contact with strong reducing agents. Strong oxidizing agents. Contact with alkalis.
Incompatible Materials:	Acids. Amines. Alkalies. Metals. Reducing agents. Oxidizing agents.
Hazardous Decomposition Products:	Chlorine. hydrogen chloride By heating and fire, corrosive vapors/gases may be formed.

11. Toxicological Information

Information on likely routes of exposure

Ingestion:	Harmful if swallowed.
Inhalation:	Causes severe burns.
Skin Contact:	Causes severe skin burns.
Eye contact:	Causes serious eye damage.

Information on toxicological effects

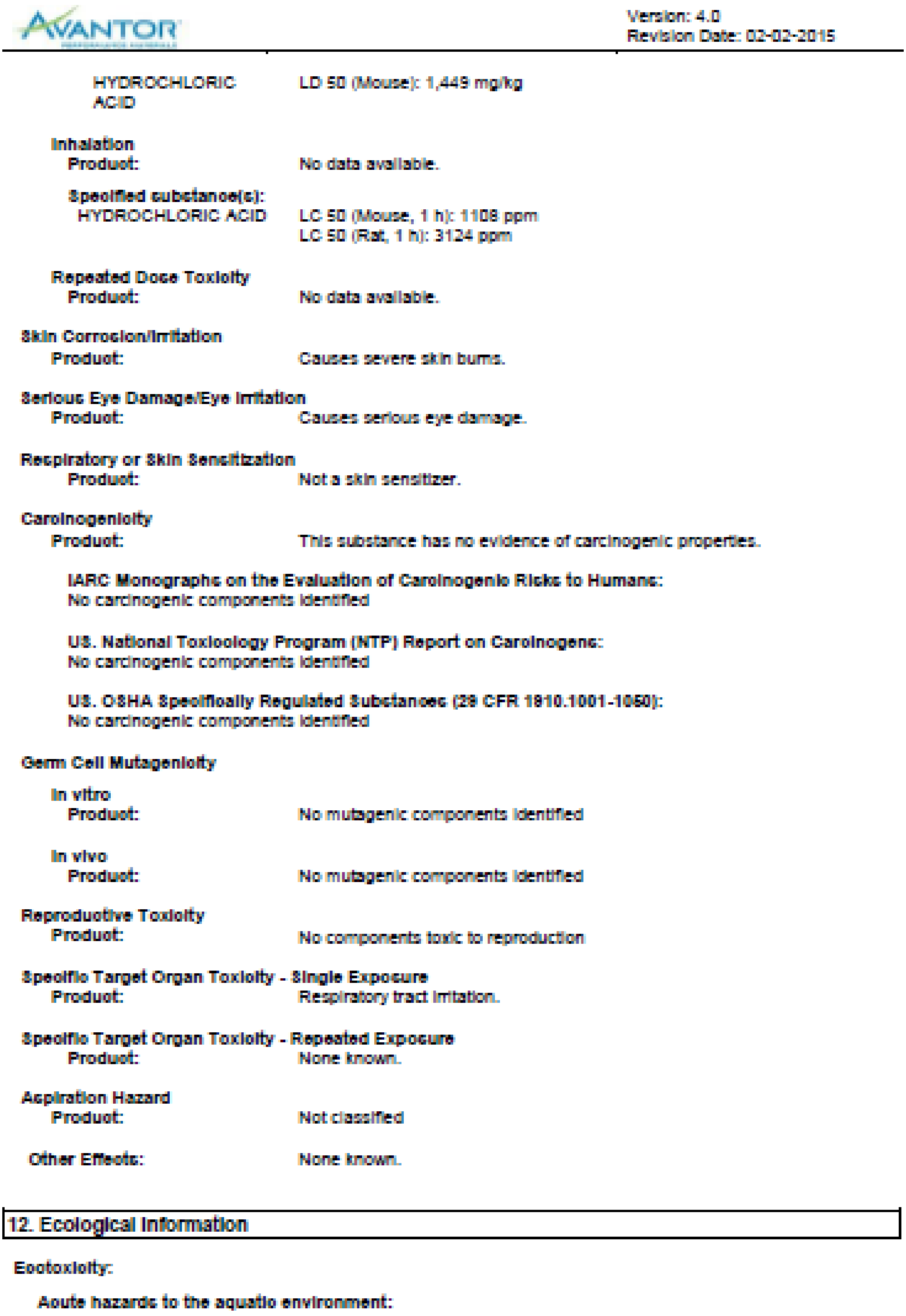
Acute toxicity (list all possible routes of exposure)

Oral

Product:	ATEmix (Rat): 581 mg/kg

Dermal

Product:	No data available.
Specified substance(s):	



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HYDROCHLORIC ACID — LD 50 (Mouse): 1,449 mg/kg

Inhalation

Product: No data available.

Specified substance(s):
HYDROCHLORIC ACID — LC 50 (Mouse, 1 h): 1108 ppm
LC 50 (Rat, 1 h): 3124 ppm

Repeated Dose Toxicity

Product: No data available.

Skin Corrosion/Irritation

Product: Causes severe skin burns.

Serious Eye Damage/Eye Irritation

Product: Causes serious eye damage.

Respiratory or Skin Sensitization

Product: Not a skin sensitizer.

Carcinogenicity

Product: This substance has no evidence of carcinogenic properties.

IARC Monographs on the Evaluation of Carcinogenic Risks to Humans:
No carcinogenic components identified

US. National Toxicology Program (NTP) Report on Carcinogens:
No carcinogenic components identified

US. OSHA Specifically Regulated Substances (29 CFR 1910.1001-1050):
No carcinogenic components identified

Germ Cell Mutagenicity

In vitro

Product: No mutagenic components identified

In vivo

Product: No mutagenic components identified

Reproductive Toxicity

Product: No components toxic to reproduction

Specific Target Organ Toxicity - Single Exposure

Product: Respiratory tract irritation.

Specific Target Organ Toxicity - Repeated Exposure

Product: None known.

Aspiration Hazard

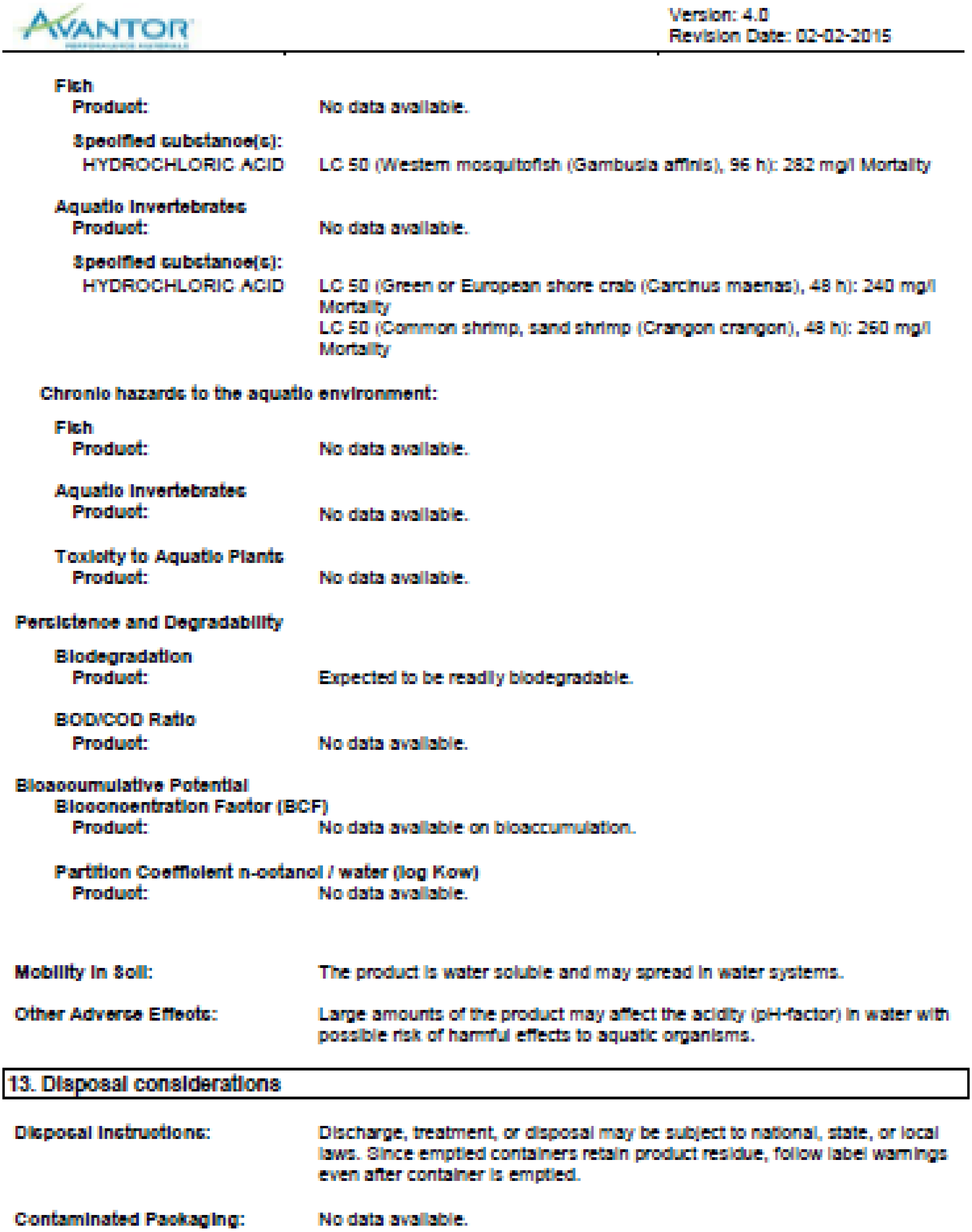
Product: Not classified

Other Effects: None known.

12. Ecological Information

Ecotoxicity:

Acute hazards to the aquatic environment:



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Fish
Product: No data available.

Specified substance(s):
HYDROCHLORIC ACID: LC 50 (Western mosquitofish (Gambusia affinis), 96 h): 282 mg/l Mortality

Aquatic Invertebrates
Product: No data available.

Specified substance(s):
HYDROCHLORIC ACID: LC 50 (Green or European shore crab (Carcinus maenas), 48 h): 240 mg/l Mortality
LC 50 (Common shrimp, sand shrimp (Crangon crangon), 48 h): 260 mg/l Mortality

Chronic hazards to the aquatic environment:

Fish
Product: No data available.

Aquatic Invertebrates
Product: No data available.

Toxicity to Aquatic Plants
Product: No data available.

Persistence and Degradability

Biodegradation
Product: Expected to be readily biodegradable.

BOD/COD Ratio
Product: No data available.

Bioaccumulative Potential
Bioconcentration Factor (BCF)
Product: No data available on bioaccumulation.

Partition Coefficient n-octanol / water (log Kow)
Product: No data available.

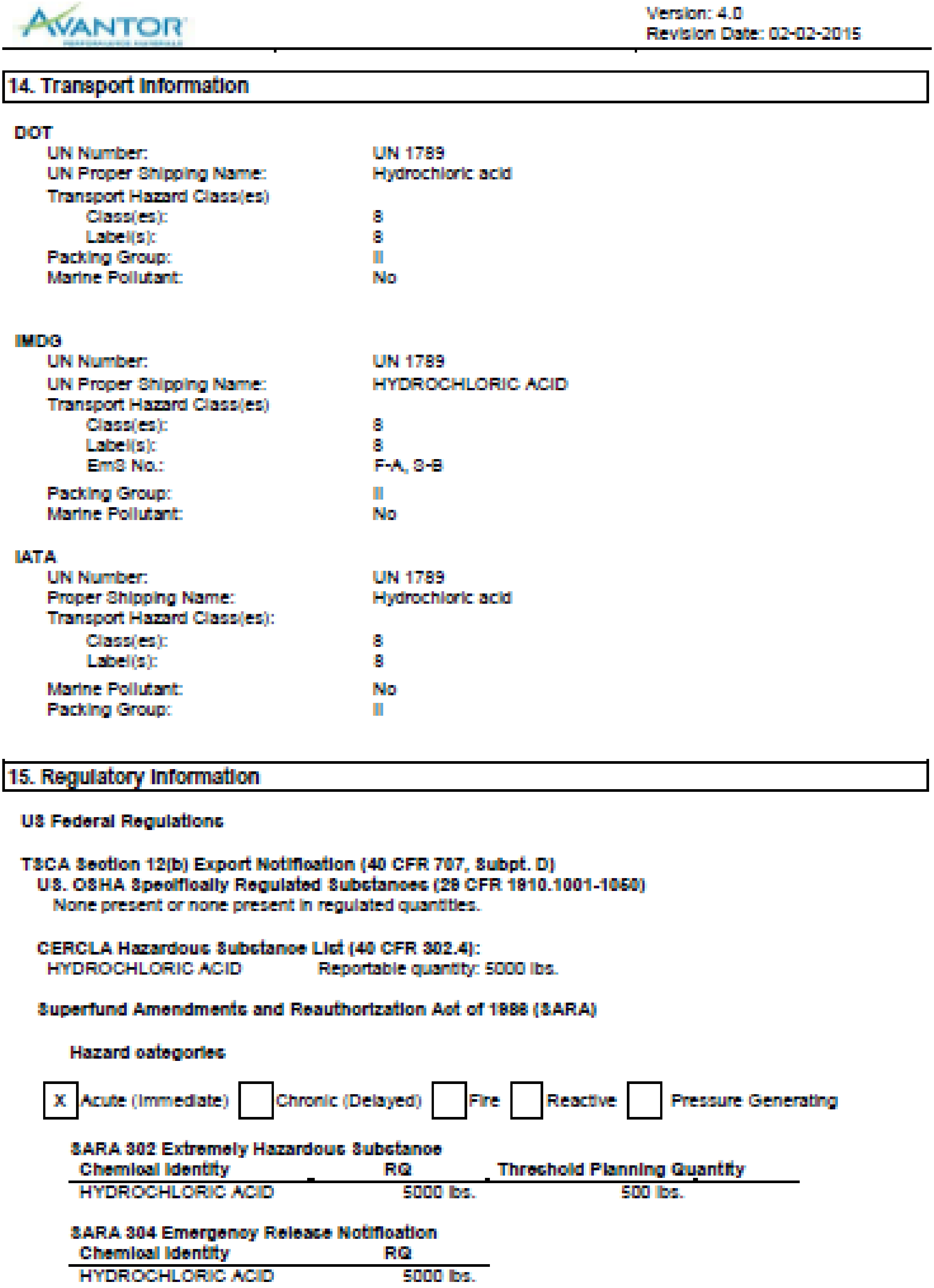
Mobility in Soil: The product is water soluble and may spread in water systems.

Other Adverse Effects: Large amounts of the product may affect the acidity (pH-factor) in water with possible risk of harmful effects to aquatic organisms.

13. Disposal considerations

Disposal Instructions: Discharge, treatment, or disposal may be subject to national, state, or local laws. Since emptied containers retain product residue, follow label warnings even after container is emptied.

Contaminated Packaging: No data available.



AVANTOR

Version: 4.0
Revision Date: 02-02-2015

14. Transport Information

DOT

UN Number:	UN 1789
UN Proper Shipping Name:	Hydrochloric acid
Transport Hazard Class(es)	
Class(es):	8
Label(s):	8
Packing Group:	II
Marine Pollutant:	No

IMDG

UN Number:	UN 1789
UN Proper Shipping Name:	HYDROCHLORIC ACID
Transport Hazard Class(es)	
Class(es):	8
Label(s):	8
EmS No.:	F-A, S-B
Packing Group:	II
Marine Pollutant:	No

IATA

UN Number:	UN 1789
Proper Shipping Name:	Hydrochloric acid
Transport Hazard Class(es):	
Class(es):	8
Label(s):	8
Marine Pollutant:	No
Packing Group:	II

15. Regulatory Information

US Federal Regulations

TSCA Section 12(b) Export Notification (40 CFR 707, Subpt. D)

US. OSHA Specifically Regulated Substances (29 CFR 1910.1001-1050)

None present or none present in regulated quantities.

CERCLA Hazardous Substance List (40 CFR 302.4):

HYDROCHLORIC ACID Reportable quantity: 5000 lbs.

Superfund Amendments and Reauthorization Act of 1986 (SARA)

Hazard categories

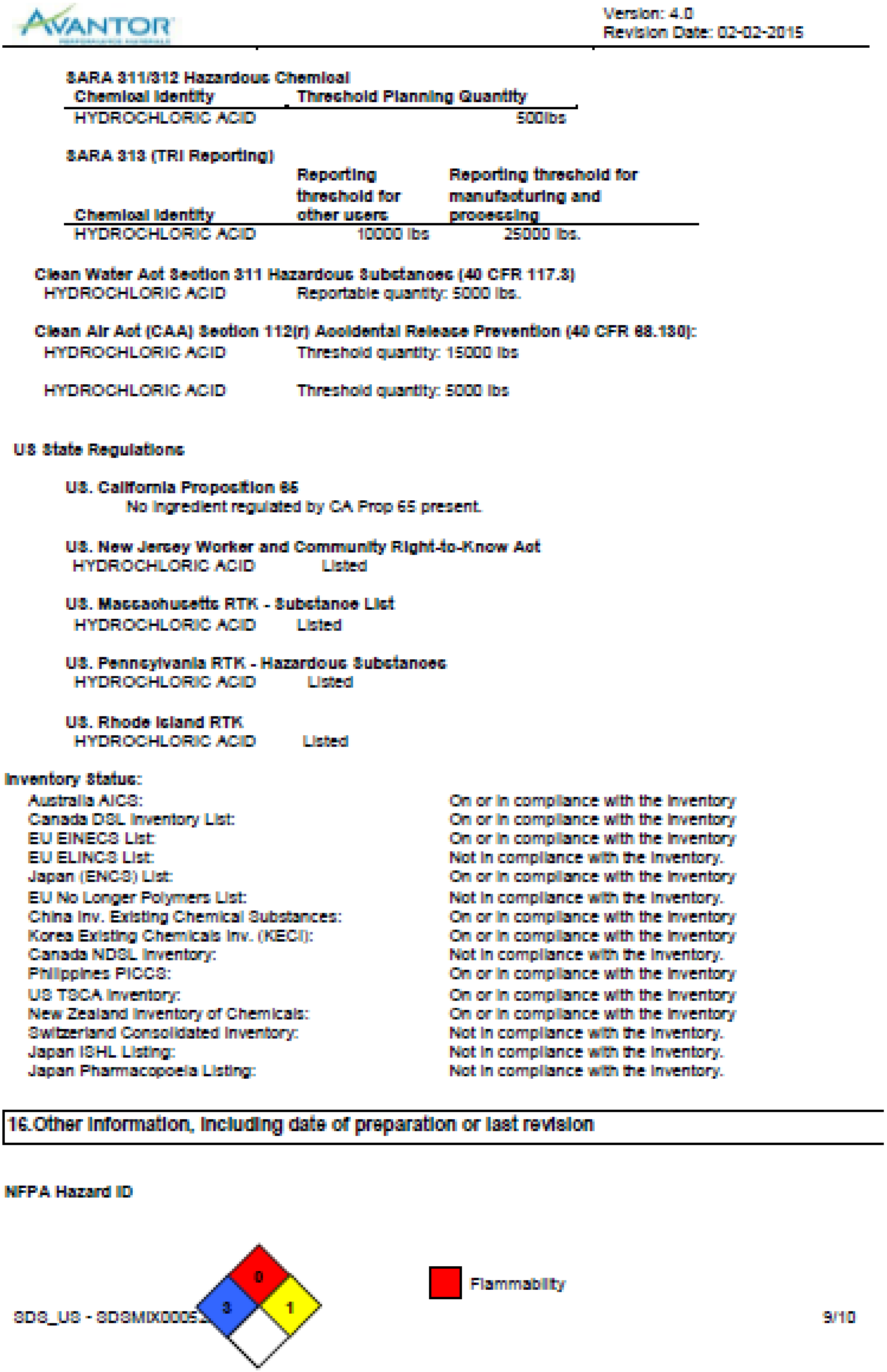
☒ Acute (Immediate) ☐ Chronic (Delayed) ☐ Fire ☐ Reactive ☐ Pressure Generating

SARA 302 Extremely Hazardous Substance

Chemical Identity	RQ	Threshold Planning Quantity
HYDROCHLORIC ACID	5000 lbs.	500 lbs.

SARA 304 Emergency Release Notification

Chemical Identity	RQ
HYDROCHLORIC ACID	5000 lbs.



SARA 311/312 Hazardous Chemical

Chemical Identity	Threshold Planning Quantity
HYDROCHLORIC ACID	500lbs

SARA 313 (TRI Reporting)

Chemical Identity	Reporting threshold for other users	Reporting threshold for manufacturing and processing
HYDROCHLORIC ACID	10000 lbs	25000 lbs.

Clean Water Act Section 311 Hazardous Substances (40 CFR 117.3)
HYDROCHLORIC ACID Reportable quantity: 5000 lbs.

Clean Air Act (CAA) Section 112(r) Accidental Release Prevention (40 CFR 68.130):
HYDROCHLORIC ACID Threshold quantity: 15000 lbs

HYDROCHLORIC ACID Threshold quantity: 5000 lbs

US State Regulations

US. California Proposition 65
No ingredient regulated by CA Prop 65 present.

US. New Jersey Worker and Community Right-to-Know Act
HYDROCHLORIC ACID Listed

US. Massachusetts RTK - Substance List
HYDROCHLORIC ACID Listed

US. Pennsylvania RTK - Hazardous Substances
HYDROCHLORIC ACID Listed

US. Rhode Island RTK
HYDROCHLORIC ACID Listed

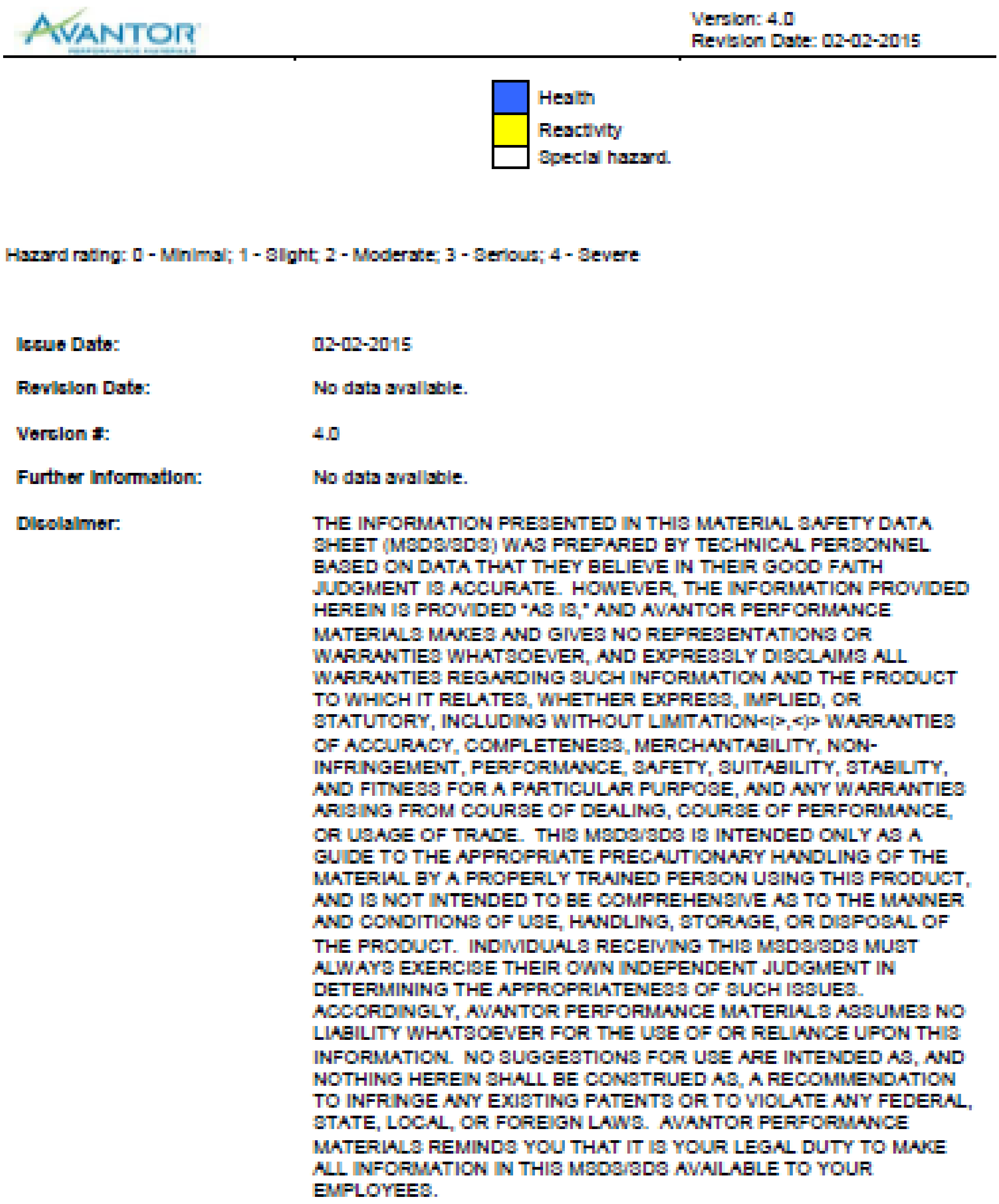
Inventory Status:

Australia AICS:	On or in compliance with the inventory
Canada DSL Inventory List:	On or in compliance with the inventory
EU EINECS List:	On or in compliance with the inventory
EU ELINCS List:	Not in compliance with the inventory.
Japan (ENCS) List:	On or in compliance with the inventory
EU No Longer Polymers List:	Not in compliance with the inventory.
China Inv. Existing Chemical Substances:	On or in compliance with the inventory
Korea Existing Chemicals Inv. (KECI):	On or in compliance with the inventory
Canada NDSL Inventory:	Not in compliance with the inventory.
Philippines PICCS:	On or in compliance with the inventory
US TSCA Inventory:	On or in compliance with the inventory
New Zealand Inventory of Chemicals:	On or in compliance with the inventory
Switzerland Consolidated Inventory:	Not in compliance with the inventory.
Japan ISHL Listing:	Not in compliance with the inventory.
Japan Pharmacopoeia Listing:	Not in compliance with the inventory.

16.Other information, including date of preparation or last revision

NFPA Hazard ID

🟥 Flammability



AVANTOR

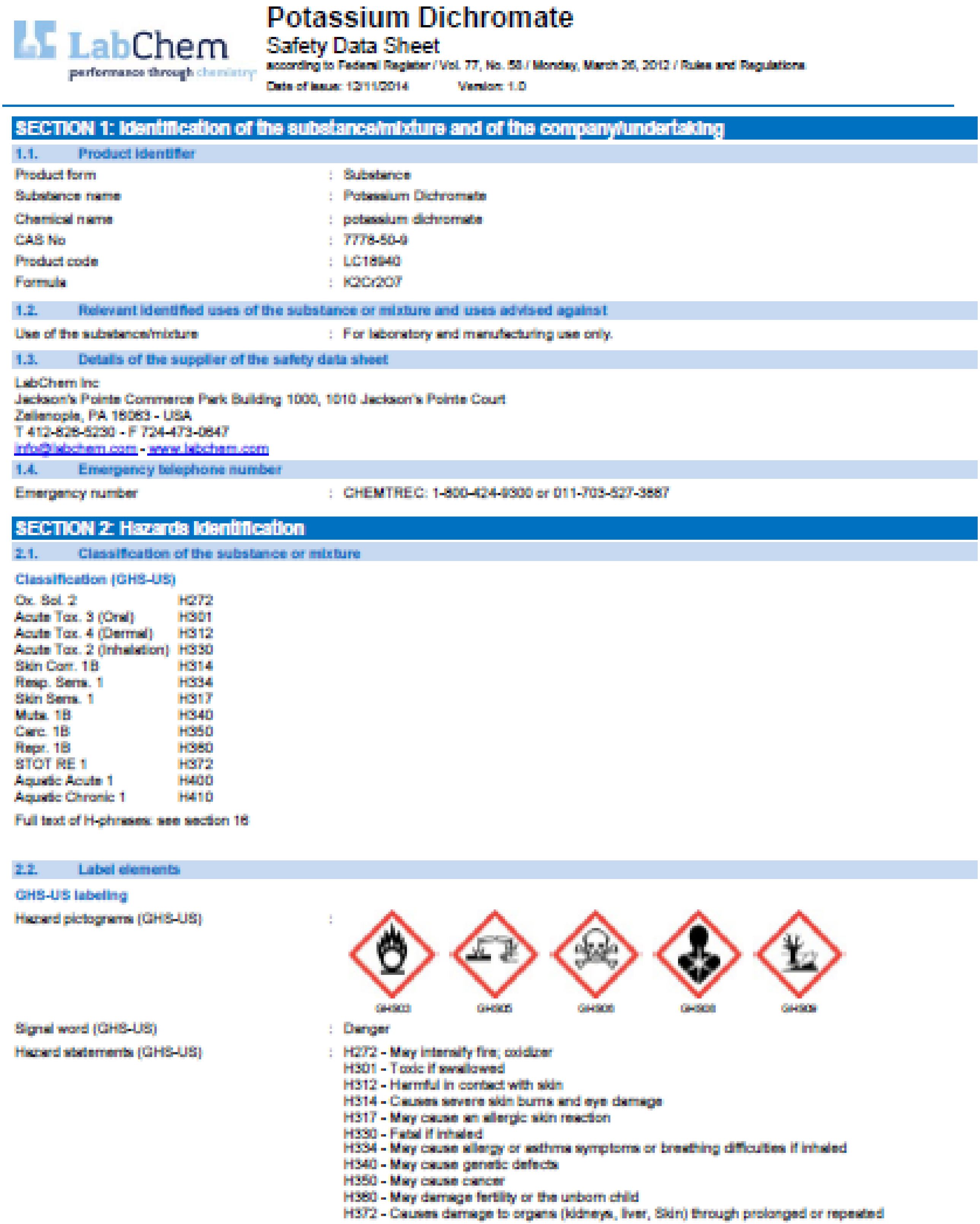
Version: 4.0
Revision Date: 02-02-2015

🟦 Health
🟨 Reactivity
⬜ Special hazard.

Hazard rating: 0 - Minimal; 1 - Slight; 2 - Moderate; 3 - Serious; 4 - Severe

Issue Date:	02-02-2015
Revision Date:	No data available.
Version #:	4.0
Further Information:	No data available.
Disclaimer:	THE INFORMATION PRESENTED IN THIS MATERIAL SAFETY DATA SHEET (MSDS/SDS) WAS PREPARED BY TECHNICAL PERSONNEL BASED ON DATA THAT THEY BELIEVE IN THEIR GOOD FAITH JUDGMENT IS ACCURATE. HOWEVER, THE INFORMATION PROVIDED HEREIN IS PROVIDED "AS IS," AND AVANTOR PERFORMANCE MATERIALS MAKES AND GIVES NO REPRESENTATIONS OR WARRANTIES WHATSOEVER, AND EXPRESSLY DISCLAIMS ALL WARRANTIES REGARDING SUCH INFORMATION AND THE PRODUCT TO WHICH IT RELATES, WHETHER EXPRESS, IMPLIED, OR STATUTORY, INCLUDING WITHOUT LIMITATION<(>,<)> WARRANTIES OF ACCURACY, COMPLETENESS, MERCHANTABILITY, NON-INFRINGEMENT, PERFORMANCE, SAFETY, SUITABILITY, STABILITY, AND FITNESS FOR A PARTICULAR PURPOSE, AND ANY WARRANTIES ARISING FROM COURSE OF DEALING, COURSE OF PERFORMANCE, OR USAGE OF TRADE. THIS MSDS/SDS IS INTENDED ONLY AS A GUIDE TO THE APPROPRIATE PRECAUTIONARY HANDLING OF THE MATERIAL BY A PROPERLY TRAINED PERSON USING THIS PRODUCT, AND IS NOT INTENDED TO BE COMPREHENSIVE AS TO THE MANNER AND CONDITIONS OF USE, HANDLING, STORAGE, OR DISPOSAL OF THE PRODUCT. INDIVIDUALS RECEIVING THIS MSDS/SDS MUST ALWAYS EXERCISE THEIR OWN INDEPENDENT JUDGMENT IN DETERMINING THE APPROPRIATENESS OF SUCH ISSUES. ACCORDINGLY, AVANTOR PERFORMANCE MATERIALS ASSUMES NO LIABILITY WHATSOEVER FOR THE USE OF OR RELIANCE UPON THIS INFORMATION. NO SUGGESTIONS FOR USE ARE INTENDED AS, AND NOTHING HEREIN SHALL BE CONSTRUED AS, A RECOMMENDATION TO INFRINGE ANY EXISTING PATENTS OR TO VIOLATE ANY FEDERAL, STATE, LOCAL, OR FOREIGN LAWS. AVANTOR PERFORMANCE MATERIALS REMINDS YOU THAT IT IS YOUR LEGAL DUTY TO MAKE ALL INFORMATION IN THIS MSDS/SDS AVAILABLE TO YOUR EMPLOYEES.

A3.6. Potassium dichromate



LabChem
performance through chemistry

Potassium Dichromate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Date of issue: 12/11/2014 Version: 1.0

SECTION 1: Identification of the substance/mixture and of the company/undertaking

1.1. Product identifier

Product form : Substance
Substance name : Potassium Dichromate
Chemical name : potassium dichromate
CAS No : 7778-50-9
Product code : LC18940
Formula : K2Cr2O7

1.2. Relevant identified uses of the substance or mixture and uses advised against

Use of the substance/mixture : For laboratory and manufacturing use only.

1.3. Details of the supplier of the safety data sheet

LabChem Inc
Jackson's Pointe Commerce Park Building 1000, 1010 Jackson's Pointe Court
Zelienople, PA 16063 - USA
T 412-826-5230 - F 724-473-0647
info@labchem.com - www.labchem.com

1.4. Emergency telephone number

Emergency number : CHEMTREC: 1-800-424-9300 or 011-703-527-3887

SECTION 2: Hazards identification

2.1. Classification of the substance or mixture

Classification (GHS-US)

Ox. Sol. 2	H272
Acute Tox. 3 (Oral)	H301
Acute Tox. 4 (Dermal)	H312
Acute Tox. 2 (Inhalation)	H330
Skin Corr. 1B	H314
Resp. Sens. 1	H334
Skin Sens. 1	H317
Muta. 1B	H340
Carc. 1B	H350
Repr. 1B	H360
STOT RE 1	H372
Aquatic Acute 1	H400
Aquatic Chronic 1	H410

Full text of H-phrases: see section 16

2.2. Label elements

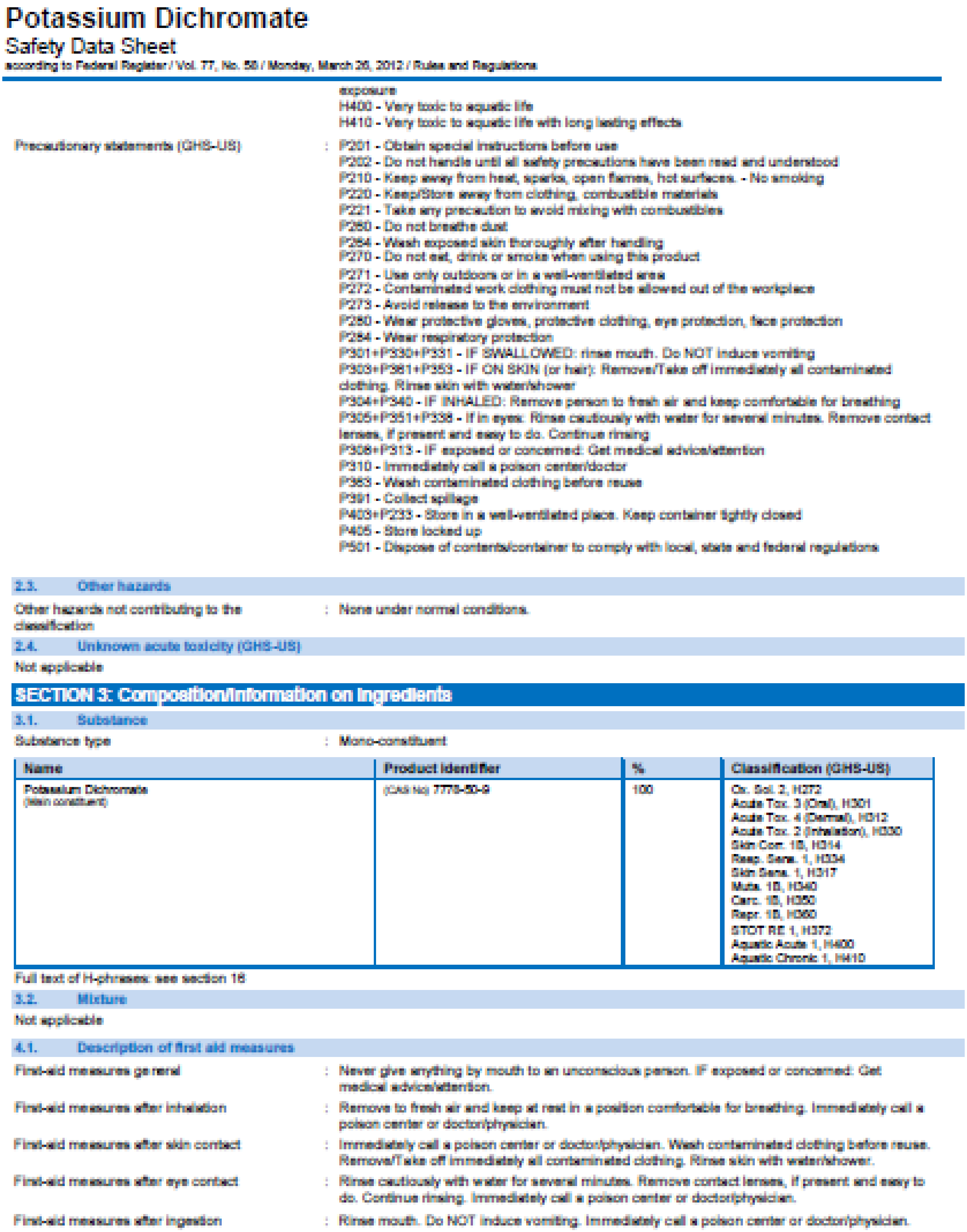
GHS-US labeling

Hazard pictograms (GHS-US) :

GHS03 GHS05 GHS06 GHS08 GHS09

Signal word (GHS-US) : Danger

Hazard statements (GHS-US) : H272 - May intensify fire; oxidizer
H301 - Toxic if swallowed
H312 - Harmful in contact with skin
H314 - Causes severe skin burns and eye damage
H317 - May cause an allergic skin reaction
H330 - Fatal if inhaled
H334 - May cause allergy or asthma symptoms or breathing difficulties if inhaled
H340 - May cause genetic defects
H350 - May cause cancer
H360 - May damage fertility or the unborn child
H372 - Causes damage to organs (kidneys, liver, Skin) through prolonged or repeated



Potassium Dichromate

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according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

exposure
H400 - Very toxic to aquatic life
H410 - Very toxic to aquatic life with long lasting effects

Precautionary statements (GHS-US) : P201 - Obtain special instructions before use
P202 - Do not handle until all safety precautions have been read and understood
P210 - Keep away from heat, sparks, open flames, hot surfaces. - No smoking
P220 - Keep/Store away from clothing, combustible materials
P221 - Take any precaution to avoid mixing with combustibles
P260 - Do not breathe dust
P264 - Wash exposed skin thoroughly after handling
P270 - Do not eat, drink or smoke when using this product
P271 - Use only outdoors or in a well-ventilated area
P272 - Contaminated work clothing must not be allowed out of the workplace
P273 - Avoid release to the environment
P280 - Wear protective gloves, protective clothing, eye protection, face protection
P284 - Wear respiratory protection
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 person to fresh air and keep 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
P308+P313 - IF exposed or concerned: Get medical advice/attention
P310 - Immediately call a poison center/doctor
P363 - Wash contaminated clothing before reuse
P391 - Collect spillage
P403+P233 - Store in a well-ventilated place. Keep container tightly closed
P405 - Store locked up
P501 - Dispose of contents/container to comply with local, state and federal regulations

2.3. Other hazards

Other hazards not contributing to the classification : None under normal conditions.

2.4. Unknown acute toxicity (GHS-US)

Not applicable

SECTION 3: Composition/information on ingredients

3.1. Substance

Substance type : Mono-constituent

Name	Product identifier	%	Classification (GHS-US)
Potassium Dichromate (Main constituent)	(CAS No) 7778-50-9	100	Ox. Sol. 2, H272 Acute Tox. 3 (Oral), H301 Acute Tox. 4 (Dermal), H312 Acute Tox. 2 (Inhalation), H330 Skin Corr. 1B, H314 Resp. Sens. 1, H334 Skin Sens. 1, H317 Muta. 1B, H340 Carc. 1B, H350 Repr. 1B, H360 STOT RE 1, H372 Aquatic Acute 1, H400 Aquatic Chronic 1, H410

Full text of H-phrases: see section 16

3.2. Mixture

Not applicable

4.1. Description of first aid measures

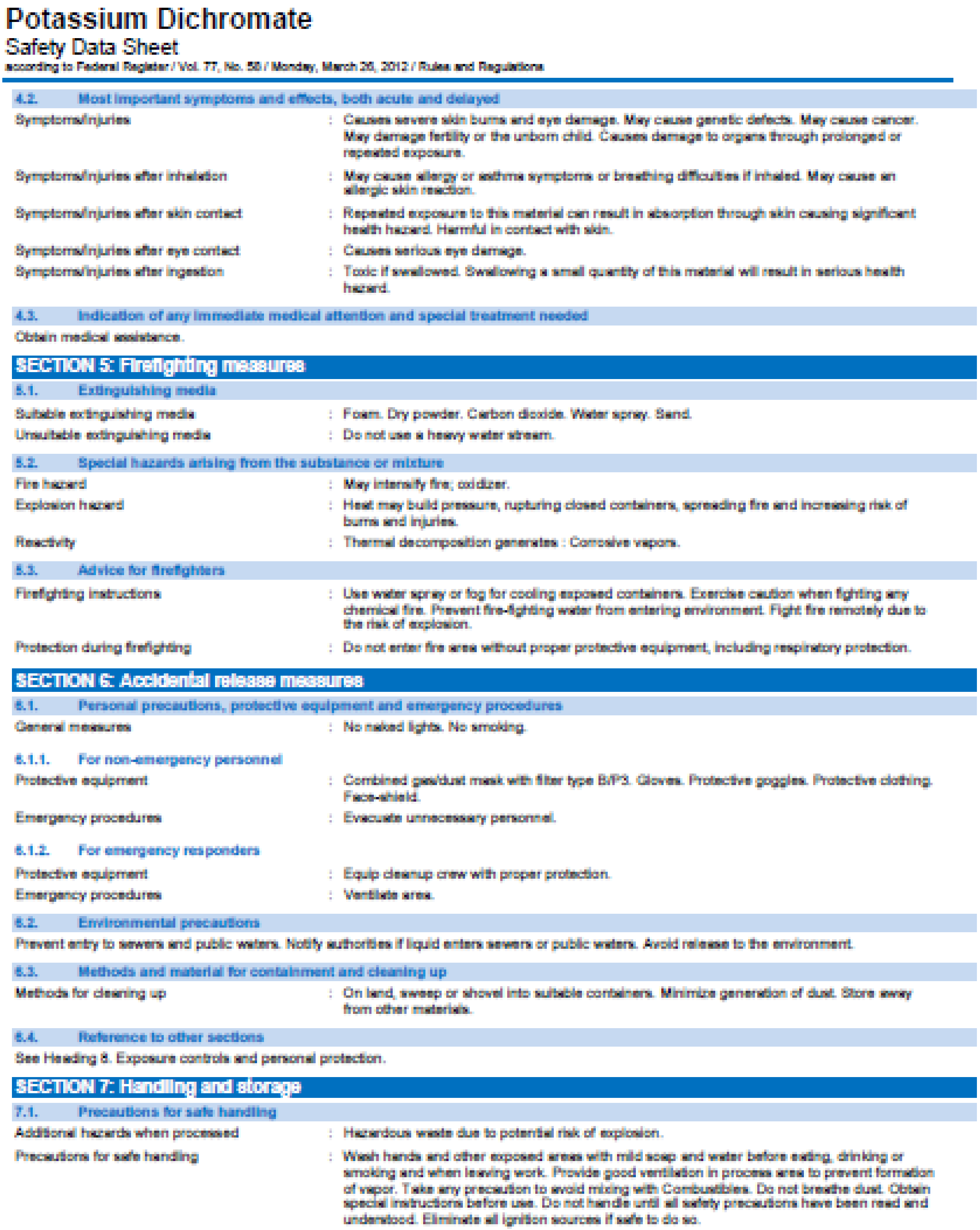
First-aid measures general : Never give anything by mouth to an unconscious person. IF exposed or concerned: Get medical advice/attention.

First-aid measures after inhalation : Remove to fresh air and keep at rest in a position comfortable for breathing. Immediately call a poison center or doctor/physician.

First-aid measures after skin contact : Immediately call a poison center or doctor/physician. Wash contaminated clothing before reuse. Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower.

First-aid measures after eye contact : Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a poison center or doctor/physician.

First-aid measures after ingestion : Rinse mouth. Do NOT induce vomiting. Immediately call a poison center or doctor/physician.



Potassium Dichromate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

4.2. Most important symptoms and effects, both acute and delayed

Symptoms/injuries	: Causes severe skin burns and eye damage. May cause genetic defects. May cause cancer. May damage fertility or the unborn child. Causes damage to organs through prolonged or repeated exposure.
Symptoms/injuries after inhalation	: May cause allergy or asthma symptoms or breathing difficulties if inhaled. May cause an allergic skin reaction.
Symptoms/injuries after skin contact	: Repeated exposure to this material can result in absorption through skin causing significant health hazard. Harmful in contact with skin.
Symptoms/injuries after eye contact	: Causes serious eye damage.
Symptoms/injuries after ingestion	: Toxic if swallowed. Swallowing a small quantity of this material will result in serious health hazard.

4.3. Indication of any immediate medical attention and special treatment needed

Obtain medical assistance.

SECTION 5: Firefighting measures

5.1. Extinguishing media

Suitable extinguishing media	: Foam. Dry powder. Carbon dioxide. Water spray. Sand.
Unsuitable extinguishing media	: Do not use a heavy water stream.

5.2. Special hazards arising from the substance or mixture

Fire hazard	: May intensify fire; oxidizer.
Explosion hazard	: Heat may build pressure, rupturing closed containers, spreading fire and increasing risk of burns and injuries.
Reactivity	: Thermal decomposition generates : Corrosive vapors.

5.3. Advice for firefighters

Firefighting instructions	: Use water spray or fog for cooling exposed containers. Exercise caution when fighting any chemical fire. Prevent fire-fighting water from entering environment. Fight fire remotely due to the risk of explosion.
Protection during firefighting	: Do not enter fire area without proper protective equipment, including respiratory protection.

SECTION 6: Accidental release measures

6.1. Personal precautions, protective equipment and emergency procedures

General measures	: No naked lights. No smoking.

6.1.1. For non-emergency personnel

Protective equipment	: Combined gas/dust mask with filter type B/P3. Gloves. Protective goggles. Protective clothing. Face-shield.
Emergency procedures	: Evacuate unnecessary personnel.

6.1.2. For emergency responders

Protective equipment	: Equip cleanup crew with proper protection.
Emergency procedures	: Ventilate area.

6.2. Environmental precautions

Prevent entry to sewers and public waters. Notify authorities if liquid enters sewers or public waters. Avoid release to the environment.

6.3. Methods and material for containment and cleaning up

Methods for cleaning up	: On land, sweep or shovel into suitable containers. Minimize generation of dust. Store away from other materials.

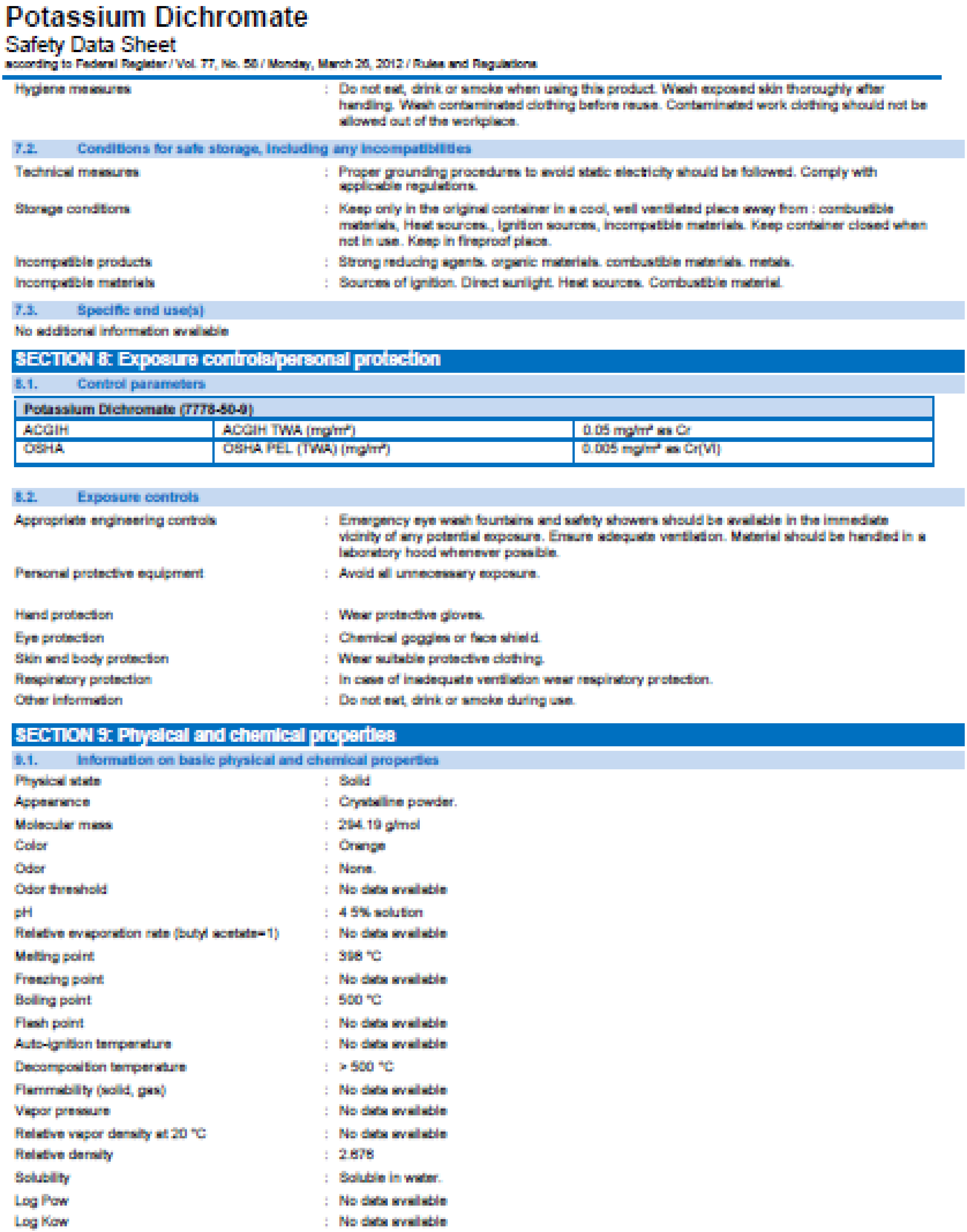
6.4. Reference to other sections

See Heading 8. Exposure controls and personal protection.

SECTION 7: Handling and storage

7.1. Precautions for safe handling

Additional hazards when processed	: Hazardous waste due to potential risk of explosion.
Precautions for safe handling	: Wash hands and other exposed areas with mild soap and water before eating, drinking or smoking and when leaving work. Provide good ventilation in process area to prevent formation of vapor. Take any precaution to avoid mixing with Combustibles. Do not breathe dust. Obtain special instructions before use. Do not handle until all safety precautions have been read and understood. Eliminate all ignition sources if safe to do so.



Potassium Dichromate

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Hygiene measures : Do not eat, drink or smoke when using this product. Wash exposed skin thoroughly after handling. Wash contaminated clothing before reuse. Contaminated work clothing should not be allowed out of the workplace.

7.2. Conditions for safe storage, including any incompatibilities

Technical measures : Proper grounding procedures to avoid static electricity should be followed. Comply with applicable regulations.

Storage conditions : Keep only in the original container in a cool, well ventilated place away from : combustible materials, Heat sources., Ignition sources, incompatible materials. Keep container closed when not in use. Keep in fireproof place.

Incompatible products : Strong reducing agents. organic materials. combustible materials. metals.

Incompatible materials : Sources of ignition. Direct sunlight. Heat sources. Combustible material.

7.3. Specific end use(s)

No additional information available

SECTION 8: Exposure controls/personal protection

8.1. Control parameters

Potassium Dichromate (7778-50-9)		
ACGIH	ACGIH TWA (mg/m³)	0.05 mg/m³ as Cr
OSHA	OSHA PEL (TWA) (mg/m³)	0.005 mg/m³ as Cr(VI)

8.2. Exposure controls

Appropriate engineering controls : Emergency eye wash fountains and safety showers should be available in the immediate vicinity of any potential exposure. Ensure adequate ventilation. Material should be handled in a laboratory hood whenever possible.

Personal protective equipment : Avoid all unnecessary exposure.

Hand protection : Wear protective gloves.

Eye protection : Chemical goggles or face shield.

Skin and body protection : Wear suitable protective clothing.

Respiratory protection : In case of inadequate ventilation wear respiratory protection.

Other information : Do not eat, drink or smoke during use.

SECTION 9: Physical and chemical properties

9.1. Information on basic physical and chemical properties

Physical state : Solid

Appearance : Crystalline powder.

Molecular mass : 294.19 g/mol

Color : Orange

Odor : None.

Odor threshold : No data available

pH : 4 5% solution

Relative evaporation rate (butyl acetate=1) : No data available

Melting point : 398 °C

Freezing point : No data available

Boiling point : 500 °C

Flash point : No data available

Auto-ignition temperature : No data available

Decomposition temperature : > 500 °C

Flammability (solid, gas) : No data available

Vapor pressure : No data available

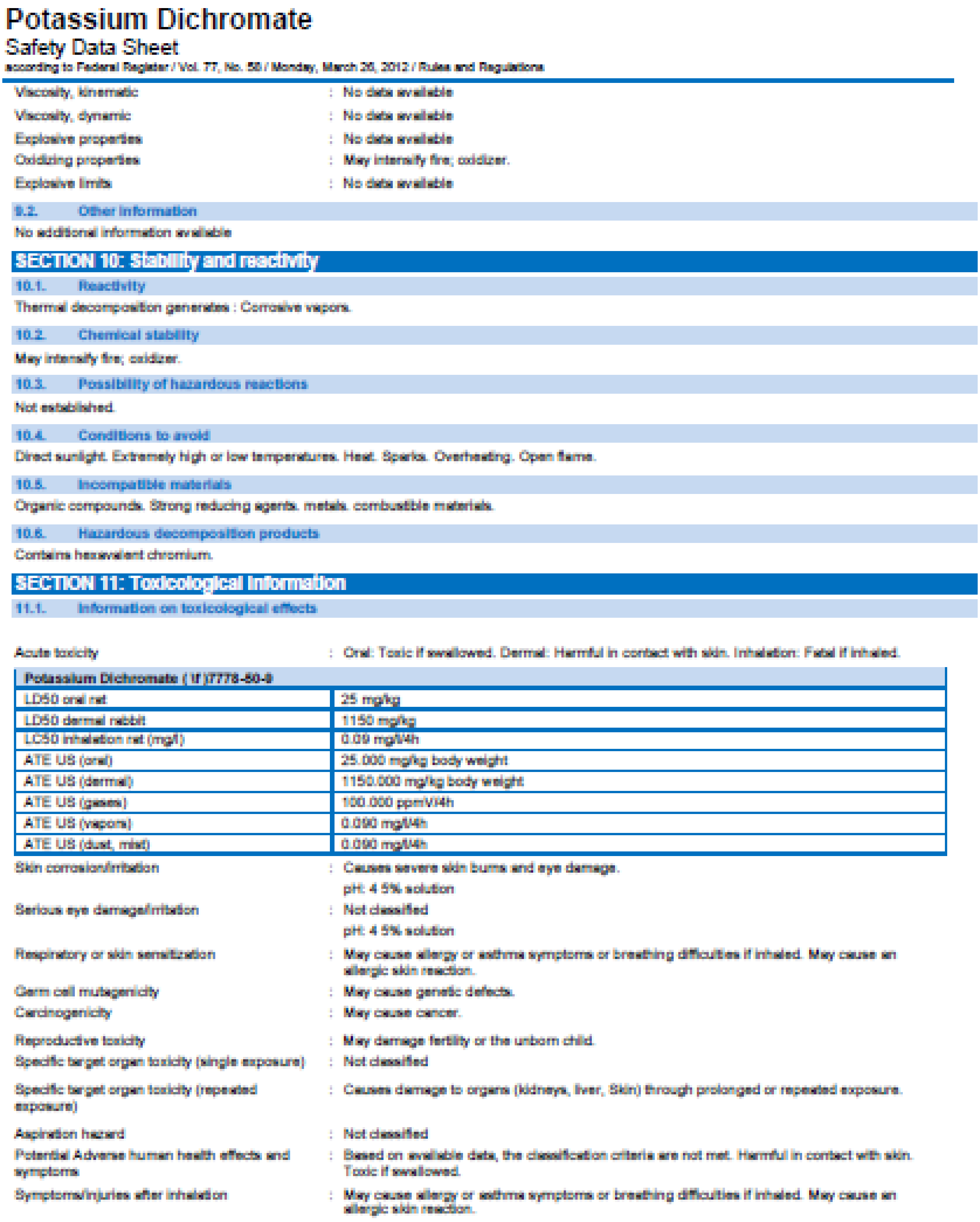
Relative vapor density at 20 °C : No data available

Relative density : 2.676

Solubility : Soluble in water.

Log Pow : No data available

Log Kow : No data available



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Viscosity, kinematic : No data available

Viscosity, dynamic : No data available

Explosive properties : No data available

Oxidizing properties : May intensify fire; oxidizer.

Explosive limits : No data available

9.2. Other information

No additional information available

SECTION 10: Stability and reactivity

10.1. Reactivity

Thermal decomposition generates : Corrosive vapors.

10.2. Chemical stability

May intensify fire; oxidizer.

10.3. Possibility of hazardous reactions

Not established.

10.4. Conditions to avoid

Direct sunlight. Extremely high or low temperatures. Heat. Sparks. Overheating. Open flame.

10.5. Incompatible materials

Organic compounds. Strong reducing agents. metals. combustible materials.

10.6. Hazardous decomposition products

Contains hexavalent chromium.

SECTION 11: Toxicological Information

11.1. Information on toxicological effects

Acute toxicity : Oral: Toxic if swallowed. Dermal: Harmful in contact with skin. Inhalation: Fatal if inhaled.

Potassium Dichromate (\f)7778-50-9	
LD50 oral rat	25 mg/kg
LD50 dermal rabbit	1150 mg/kg
LC50 inhalation rat (mg/l)	0.09 mg/l/4h
ATE US (oral)	25.000 mg/kg body weight
ATE US (dermal)	1150.000 mg/kg body weight
ATE US (gases)	100.000 ppmV/4h
ATE US (vapors)	0.090 mg/l/4h
ATE US (dust, mist)	0.090 mg/l/4h

Skin corrosion/irritation : Causes severe skin burns and eye damage.
pH: 4 5% solution

Serious eye damage/irritation : Not classified
pH: 4 5% solution

Respiratory or skin sensitization : May cause allergy or asthma symptoms or breathing difficulties if inhaled. May cause an allergic skin reaction.

Germ cell mutagenicity : May cause genetic defects.

Carcinogenicity : May cause cancer.

Reproductive toxicity : May damage fertility or the unborn child.

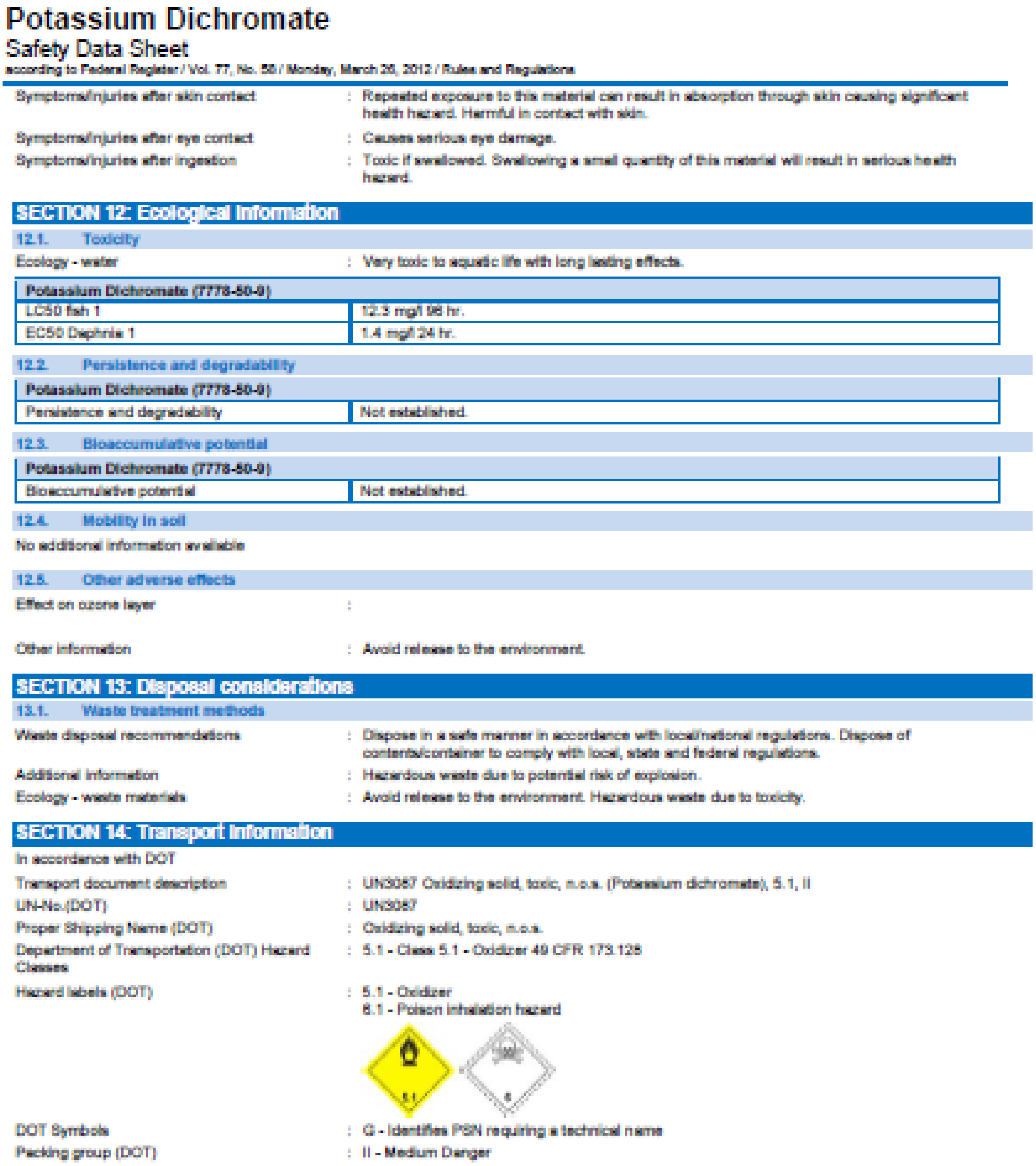
Specific target organ toxicity (single exposure) : Not classified

Specific target organ toxicity (repeated exposure) : Causes damage to organs (kidneys, liver, Skin) through prolonged or repeated exposure.

Aspiration hazard : Not classified

Potential Adverse human health effects and symptoms : Based on available data, the classification criteria are not met. Harmful in contact with skin. Toxic if swallowed.

Symptoms/injuries after inhalation : May cause allergy or asthma symptoms or breathing difficulties if inhaled. May cause an allergic skin reaction.



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Symptoms/injuries after skin contact : Repeated exposure to this material can result in absorption through skin causing significant health hazard. Harmful in contact with skin.

Symptoms/injuries after eye contact : Causes serious eye damage.

Symptoms/injuries after ingestion : Toxic if swallowed. Swallowing a small quantity of this material will result in serious health hazard.

SECTION 12: Ecological Information

12.1. Toxicity

Ecology - water : Very toxic to aquatic life with long lasting effects.

Potassium Dichromate (7778-50-9)	
LC50 fish 1	12.3 mg/l 96 hr.
EC50 Daphnia 1	1.4 mg/l 24 hr.

12.2. Persistence and degradability

Potassium Dichromate (7778-50-9)	
Persistence and degradability	Not established.

12.3. Bioaccumulative potential

Potassium Dichromate (7778-50-9)	
Bioaccumulative potential	Not established.

12.4. Mobility in soil

No additional information available

12.5. Other adverse effects

Effect on ozone layer :

Other information : Avoid release to the environment.

SECTION 13: Disposal considerations

13.1. Waste treatment methods

Waste disposal recommendations : Dispose in a safe manner in accordance with local/national regulations. Dispose of contents/container to comply with local, state and federal regulations.

Additional information : Hazardous waste due to potential risk of explosion.

Ecology - waste materials : Avoid release to the environment. Hazardous waste due to toxicity.

SECTION 14: Transport information

In accordance with DOT

Transport document description : UN3087 Oxidizing solid, toxic, n.o.s. (Potassium dichromate), 5.1, II

UN-No.(DOT) : UN3087

Proper Shipping Name (DOT) : Oxidizing solid, toxic, n.o.s.

Department of Transportation (DOT) Hazard Classes : 5.1 - Class 5.1 - Oxidizer 49 CFR 173.128

Hazard labels (DOT) : 5.1 - Oxidizer
6.1 - Poison inhalation hazard

DOT Symbols : G - Identifies PSN requiring a technical name

Packing group (DOT) : II - Medium Danger

Potassium Dichromate

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DOT Special Provisions (49 CFR 172.102)	:	62 - Oxygen generators (see §171.8 of this subchapter) are not authorized for transportation under this entry. IB8 - Authorized IBCs: Metal (11A, 11B, 11N, 21A, 21B, 21N, 31A, 31B and 31N); Rigid plastics (11H1, 11H2, 21H1, 21H2, 31H1 and 31H2); Composite (11HZ1, 11HZ2, 21HZ1, 21HZ2, 31HZ1 and 31HZ2). Additional Requirement: Composite IBCs 11HZ2 and 21HZ2 may not be used when the hazardous materials being transported may become liquid during transport. IP2 - When IBCs other than metal or rigid plastics IBCs are used, they must be offered for transportation in a closed freight container or a closed transport vehicle. T3 - 2.65 178.274(d)(2) Normal........... 178.275(d)(2) TP33 - The portable tank instruction assigned for this substance applies for granular and powdered solids and for solids which are filled and discharged at temperatures above their melting point which are cooled and transported as a solid mass. Solid substances transported or offered for transport above their melting point are authorized for transportation in portable tanks conforming to the provisions of portable tank instruction T4 for solid substances of packing group III or T7 for solid substances of packing group II, unless a tank with more stringent requirements for minimum shell thickness, maximum allowable working pressure, pressure-relief devices or bottom outlets are assigned in which case the more stringent tank instruction and special provisions shall apply. Filling limits must be in accordance with portable tank special provision TP3. Solids meeting the definition of an elevated temperature material must be transported in accordance with the applicable requirements of this subchapter.
DOT Packaging Exceptions (49 CFR 173.xxx)	:	152
DOT Packaging Non Bulk (49 CFR 173.xxx)	:	212
DOT Packaging Bulk (49 CFR 173.xxx)	:	242
DOT Quantity Limitations Passenger aircraft/rail (49 CFR 173.27)	:	5 kg
DOT Quantity Limitations Cargo aircraft only (49 CFR 175.75)	:	25 kg
DOT Vessel Stowage Location	:	B - (i) The material may be stowed "on deck" or "under deck" on a cargo vessel and on a passenger vessel carrying a number of passengers limited to not more than the larger of 25 passengers, or one passenger per each 3 m of overall vessel length; and (ii) "On deck only" on passenger vessels in which the number of passengers specified in paragraph (k)(2)(i) of this section is exceeded.
DOT Vessel Stowage Other	:	56 - Stow "separated from" ammonium compounds,58 - Stow "separated from" cyanides,95 - Stow "separated from" foodstuffs,106 - Stow "separated from" powdered metal

Additional information

Other information	:	No supplementary information available.

ADR

No additional information available

Transport by sea

UN-No. (IMDG)	:	3087
Proper Shipping Name (IMDG)	:	OXIDIZING SOLID, TOXIC, N.O.S.
Class (IMDG)	:	5.1 - Oxidizer
Packing group (IMDG)	:	II - substances presenting medium danger

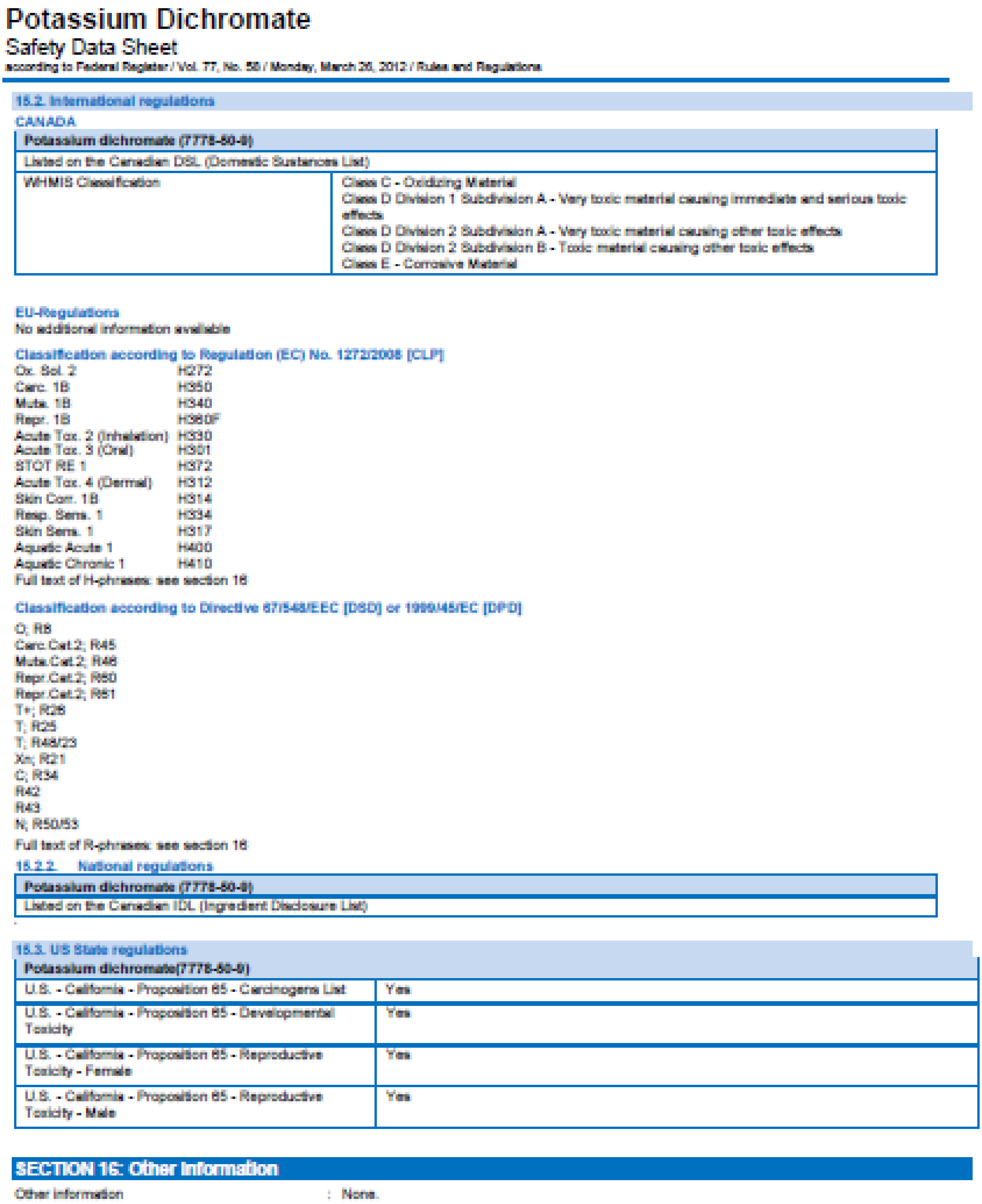
Air transport

UN-No.(IATA)	:	3087
Proper Shipping Name (IATA)	:	Oxidizing solid, toxic, n.o.s.
Class (IATA)	:	5.1 - Oxidizing Substances
Packing group (IATA)	:	II - Medium Danger

SECTION 15: Regulatory information

15.1. US Federal regulations

Potassium dichromate (7778-50-9)	
Listed on the United States TSCA (Toxic Substances Control Act) inventory Listed on United States SARA Section 313	
RQ (Reportable quantity, section 304 of EPA's List of Lists)	10 lb
SARA Section 311/312 Hazard Classes	Immediate (acute) health hazard Delayed (chronic) health hazard Reactive hazard



Potassium Dichromate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

15.2. International regulations

CANADA

Potassium dichromate (7778-50-9)	
Listed on the Canadian DSL (Domestic Sustances List)	
WHMIS Classification	Class C - Oxidizing Material Class D Division 1 Subdivision A - Very toxic material causing immediate and serious toxic effects Class D Division 2 Subdivision A - Very toxic material causing other toxic effects Class D Division 2 Subdivision B - Toxic material causing other toxic effects Class E - Corrosive Material

EU-Regulations

No additional information available

Classification according to Regulation (EC) No. 1272/2008 [CLP]

Ox. Sol. 2	H272
Carc. 1B	H350
Muta. 1B	H340
Repr. 1B	H360F
Acute Tox. 2 (Inhalation)	H330
Acute Tox. 3 (Oral)	H301
STOT RE 1	H372
Acute Tox. 4 (Dermal)	H312
Skin Corr. 1B	H314
Resp. Sens. 1	H334
Skin Sens. 1	H317
Aquatic Acute 1	H400
Aquatic Chronic 1	H410

Full text of H-phrases: see section 16

Classification according to Directive 67/548/EEC [DSD] or 1999/45/EC [DPD]

O; R8
Carc.Cat.2; R45
Muta.Cat.2; R46
Repr.Cat.2; R60
Repr.Cat.2; R61
T+; R26
T; R25
T; R48/23
Xn; R21
C; R34
R42
R43
N; R50/53

Full text of R-phrases: see section 16

15.2.2. National regulations

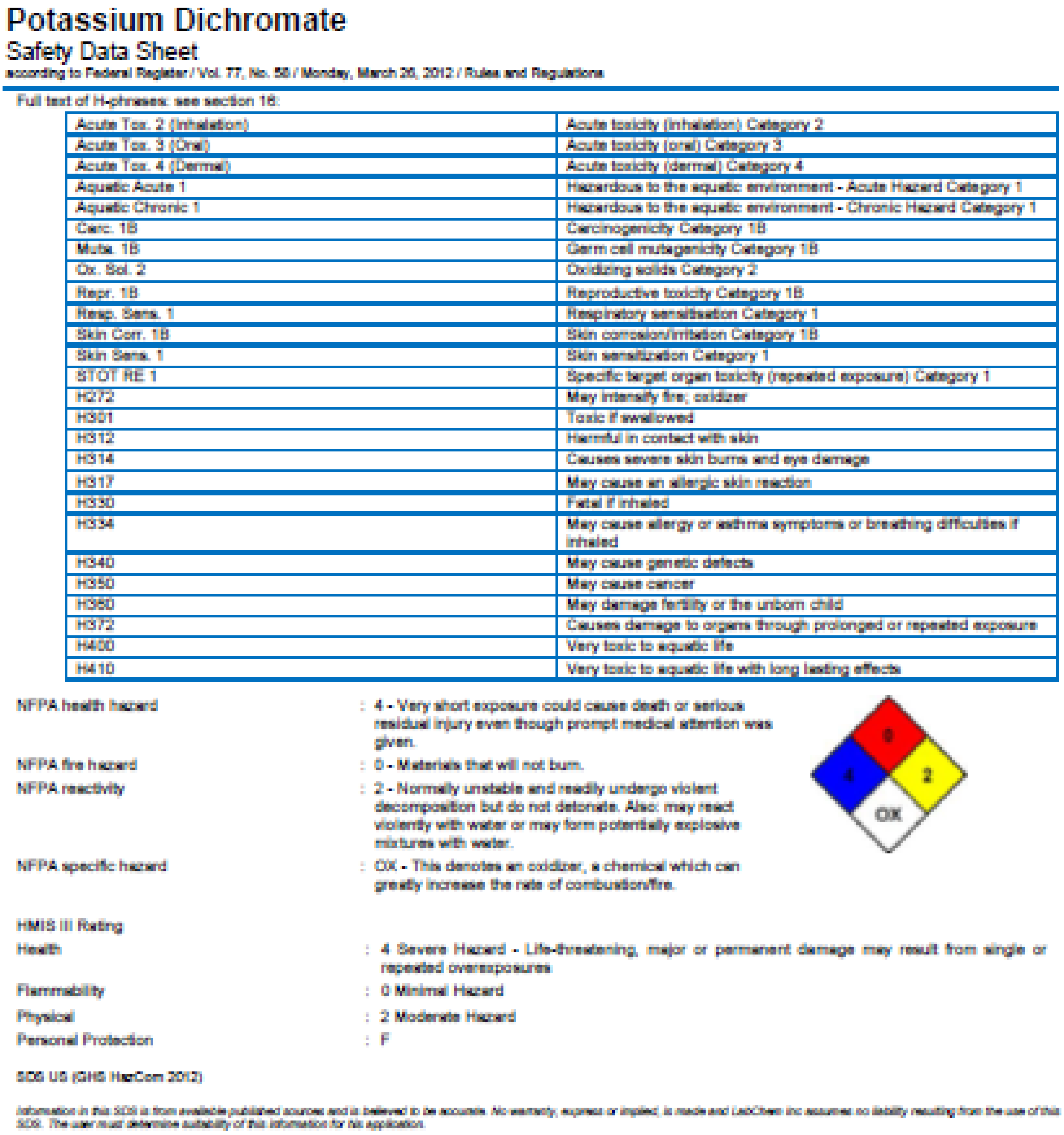
Potassium dichromate (7778-50-9)
Listed on the Canadian IDL (Ingredient Disclosure List)

15.3. US State regulations

Potassium dichromate(7778-50-9)	
U.S. - California - Proposition 65 - Carcinogens List	Yes
U.S. - California - Proposition 65 - Developmental Toxicity	Yes
U.S. - California - Proposition 65 - Reproductive Toxicity - Female	Yes
U.S. - California - Proposition 65 - Reproductive Toxicity - Male	Yes

SECTION 16: Other information

Other information : None.



Potassium Dichromate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Full text of H-phrases: see section 16:

Acute Tox. 2 (Inhalation)	Acute toxicity (inhalation) Category 2
Acute Tox. 3 (Oral)	Acute toxicity (oral) Category 3
Acute Tox. 4 (Dermal)	Acute toxicity (dermal) Category 4
Aquatic Acute 1	Hazardous to the aquatic environment - Acute Hazard Category 1
Aquatic Chronic 1	Hazardous to the aquatic environment - Chronic Hazard Category 1
Carc. 1B	Carcinogenicity Category 1B
Muta. 1B	Germ cell mutagenicity Category 1B
Ox. Sol. 2	Oxidizing solids Category 2
Repr. 1B	Reproductive toxicity Category 1B
Resp. Sens. 1	Respiratory sensitisation Category 1
Skin Corr. 1B	Skin corrosion/irritation Category 1B
Skin Sens. 1	Skin sensitization Category 1
STOT RE 1	Specific target organ toxicity (repeated exposure) Category 1
H272	May intensify fire; oxidizer
H301	Toxic if swallowed
H312	Harmful in contact with skin
H314	Causes severe skin burns and eye damage
H317	May cause an allergic skin reaction
H330	Fatal if inhaled
H334	May cause allergy or asthma symptoms or breathing difficulties if inhaled
H340	May cause genetic defects
H350	May cause cancer
H360	May damage fertility or the unborn child
H372	Causes damage to organs through prolonged or repeated exposure
H400	Very toxic to aquatic life
H410	Very toxic to aquatic life with long lasting effects

NFPA health hazard : 4 - Very short exposure could cause death or serious residual injury even though prompt medical attention was given.

NFPA fire hazard : 0 - Materials that will not burn.

NFPA reactivity : 2 - Normally unstable and readily undergo violent decomposition but do not detonate. Also: may react violently with water or may form potentially explosive mixtures with water.

NFPA specific hazard : OX - This denotes an oxidizer, a chemical which can greatly increase the rate of combustion/fire.

HMIS III Rating

Health : 4 Severe Hazard - Life-threatening, major or permanent damage may result from single or repeated overexposures

Flammability : 0 Minimal Hazard

Physical : 2 Moderate Hazard

Personal Protection : F

SDS US (GHS HazCom 2012)

Information in this SDS is from available published sources and is believed to be accurate. No warranty, express or implied, is made and LabChem Inc assumes no liability resulting from the use of this SDS. The user must determine suitability of this information for his application.

A3.7. Sodium Nitroprusside

LabChem
performance through chemistry

Sodium Nitroprusside

Safety Data Sheet
according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations
Date of issue: 05/14/2014 Version: 1.0

SECTION 1: Identification of the substance/mixture and of the company/undertaking

1.1. Product identifier

Product form : Substance
Substance name : Sodium Nitroprusside
Chemical name : Sodium nitroferricyanide
CAS No : 13755-38-9
Product code : LC24590
Formula : Na2(Fe(CN)5NO)·2H2O

1.2. Relevant identified uses of the substance or mixture and uses advised against

Use of the substance/mixture : For laboratory and manufacturing use only.

1.3. Details of the supplier of the safety data sheet

LabChem Inc
Jackson's Pointe Commerce Park Building 1000, 1010 Jackson's Pointe Court
Zelienople, PA 16063 - USA
T 412-826-5230 - F 724-473-0647
info@labchem.com - www.labchem.com

1.4. Emergency telephone number

Emergency number : CHEMTREC: 1-800-424-9300 or 011-703-527-3887

SECTION 2: Hazards identification

2.1. Classification of the substance or mixture

GHS-US classification

Acute Tox. 3 (Oral) H301

2.2. Label elements

GHS-US labelling

Hazard pictograms (GHS-US) :

GHS06

Signal word (GHS-US) : Danger
Hazard statements (GHS-US) : H301 - Toxic if swallowed
Precautionary statements (GHS-US) : P264 - Wash exposed skin thoroughly after handling
P270 - Do not eat, drink or smoke when using this product
P301 + P310 - IF SWALLOWED: Immediately call a POISON CENTER or doctor/physician
P330 - If swallowed, rinse mouth
P405 - Store locked up
P501 - Dispose of contents/container to comply with local, state and federal regulations

2.3. Other hazards

Other hazards not contributing to the classification : None under normal conditions.

2.4. Unknown acute toxicity (GHS-US)

No data available

SECTION 3: Composition/information on ingredients

3.1. Substance

Substance type : Mono-constituent
Name : Sodium Nitroprusside
CAS No : 13755-38-9

Sodium Nitroprusside

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Name	Product identifier	%	GHS-US classification
Sodium Nitroprusside (Main constituent)	(CAS No) 13755-38-9	100	Acute Tox. 3 (Oral), H301

3.2. Mixture

Not applicable

SECTION 4: First aid measures

4.1. Description of first aid measures

First-aid measures general : Never give anything by mouth to an unconscious person. If you feel unwell, seek medical advice (show the label where possible).

First-aid measures after inhalation : Assure fresh air breathing. Allow the victim to rest.

First-aid measures after skin contact : Remove affected clothing and wash all exposed skin area with mild soap and water, followed by warm water rinse.

First-aid measures after eye contact : Rinse immediately with plenty of water. Obtain medical attention if pain, blinking or redness persist.

First-aid measures after ingestion : Rinse mouth. Do NOT induce vomiting. Obtain emergency medical attention. Immediately call a POISON CENTER or doctor/physician.

4.2. Most important symptoms and effects, both acute and delayed

Symptoms/injuries after ingestion : Toxic if swallowed. Swallowing a small quantity of this material will result in serious health hazard.

4.3. Indication of any immediate medical attention and special treatment needed

Obtain medical assistance.

SECTION 5: Firefighting measures

5.1. Extinguishing media

Suitable extinguishing media : Foam. Dry powder. Carbon dioxide. Water spray. Sand.

Unsuitable extinguishing media : Do not use a heavy water stream.

5.2. Special hazards arising from the substance or mixture

No additional information available

5.3. Advice for firefighters

Firefighting instructions : Use water spray or fog for cooling exposed containers. Exercise caution when fighting any chemical fire. Prevent fire-fighting water from entering environment.

Protection during firefighting : Do not enter fire area without proper protective equipment, including respiratory protection.

SECTION 6: Accidental release measures

6.1. Personal precautions, protective equipment and emergency procedures

6.1.1. For non-emergency personnel

Protective equipment : Safety glasses. Gloves. Dust mask.

Emergency procedures : Evacuate unnecessary personnel.

6.1.2. For emergency responders

Protective equipment : Equip cleanup crew with proper protection.

Emergency procedures : Ventilate area.

6.2. Environmental precautions

Prevent entry to sewers and public waters. Notify authorities if liquid enters sewers or public waters.

6.3. Methods and material for containment and cleaning up

Methods for cleaning up : On land, sweep or shovel into suitable containers. Minimize generation of dust. Store away from other materials.

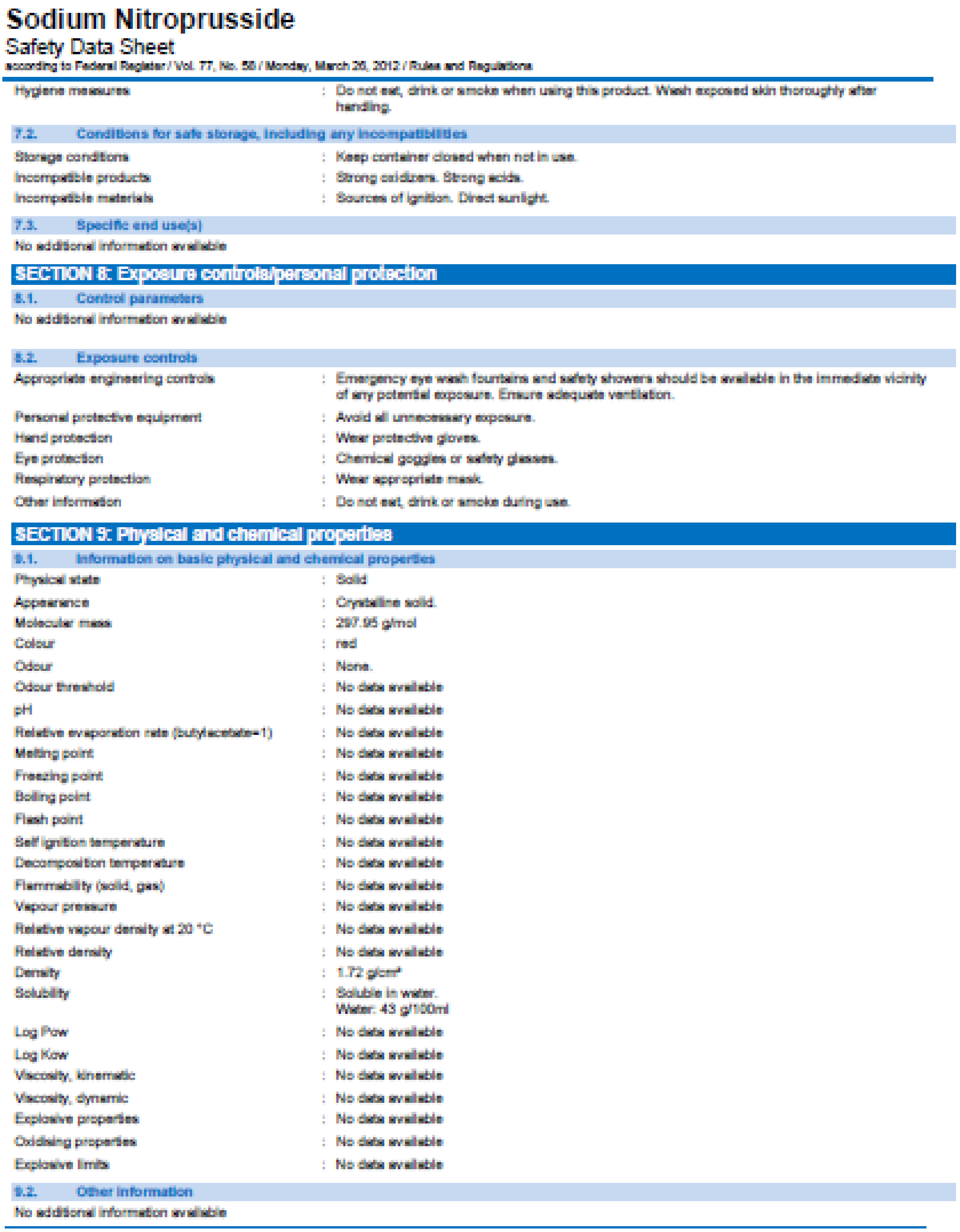
6.4. Reference to other sections

See Heading 8. Exposure controls and personal protection.

SECTION 7: Handling and storage

7.1. Precautions for safe handling

Precautions for safe handling : Wash hands and other exposed areas with mild soap and water before eating, drinking or smoking and when leaving work. Provide good ventilation in process area to prevent formation of vapour.



Sodium Nitroprusside

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Hygiene measures : Do not eat, drink or smoke when using this product. Wash exposed skin thoroughly after handling.

7.2. Conditions for safe storage, including any incompatibilities

Storage conditions : Keep container closed when not in use.
Incompatible products : Strong oxidizers. Strong acids.
Incompatible materials : Sources of ignition. Direct sunlight.

7.3. Specific end use(s)

No additional information available

SECTION 8: Exposure controls/personal protection

8.1. Control parameters

No additional information available

8.2. Exposure controls

Appropriate engineering controls : Emergency eye wash fountains and safety showers should be available in the immediate vicinity of any potential exposure. Ensure adequate ventilation.
Personal protective equipment : Avoid all unnecessary exposure.
Hand protection : Wear protective gloves.
Eye protection : Chemical goggles or safety glasses.
Respiratory protection : Wear appropriate mask.
Other information : Do not eat, drink or smoke during use.

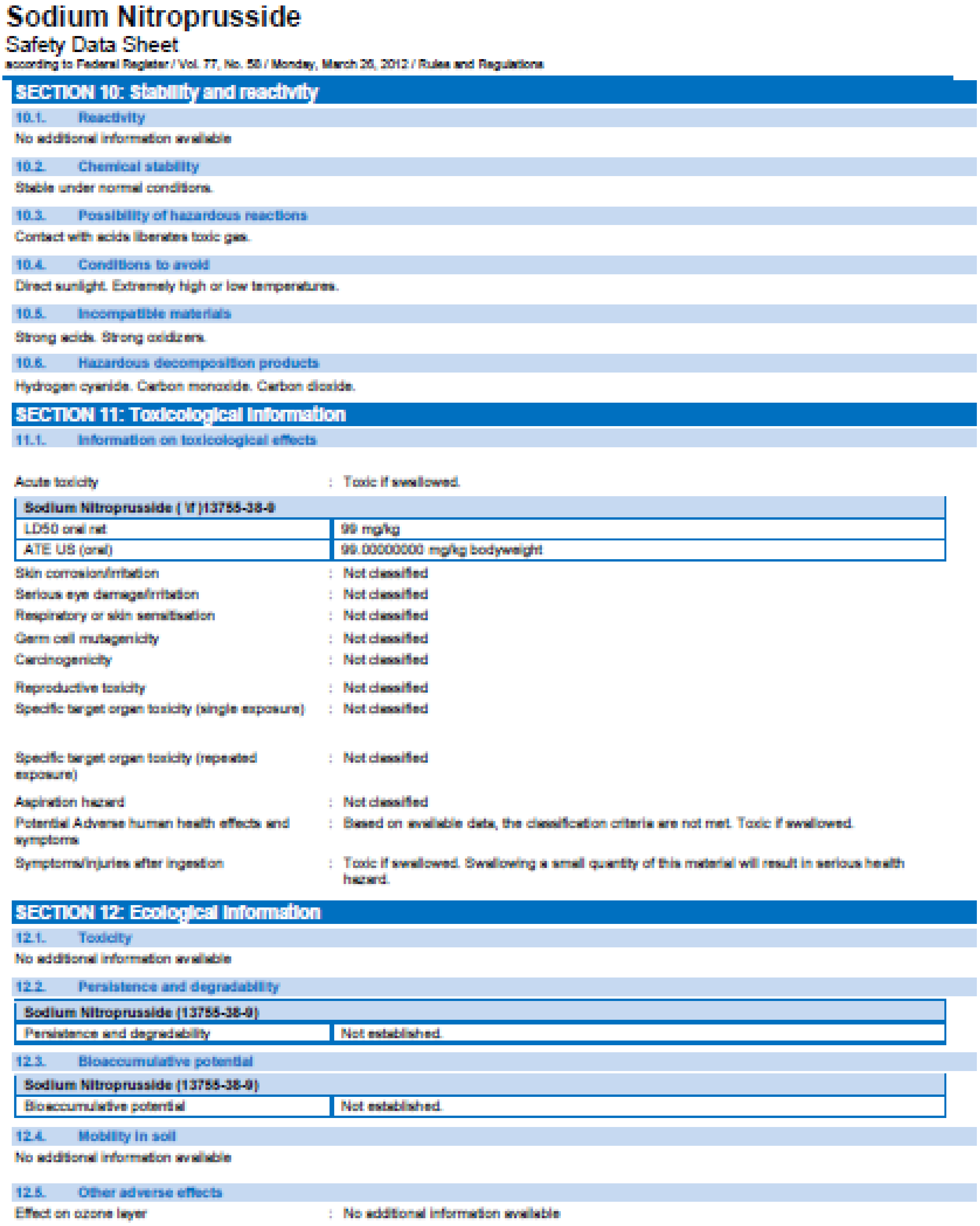
SECTION 9: Physical and chemical properties

9.1. Information on basic physical and chemical properties

Physical state : Solid
Appearance : Crystalline solid.
Molecular mass : 297.95 g/mol
Colour : red
Odour : None.
Odour threshold : No data available
pH : No data available
Relative evaporation rate (butylacetate=1) : No data available
Melting point : No data available
Freezing point : No data available
Boiling point : No data available
Flash point : No data available
Self ignition temperature : No data available
Decomposition temperature : No data available
Flammability (solid, gas) : No data available
Vapour pressure : No data available
Relative vapour density at 20 °C : No data available
Relative density : No data available
Density : 1.72 g/cm^3
Solubility : Soluble in water.
Water: 43 g/100ml
Log Pow : No data available
Log Kow : No data available
Viscosity, kinematic : No data available
Viscosity, dynamic : No data available
Explosive properties : No data available
Oxidising properties : No data available
Explosive limits : No data available

9.2. Other information

No additional information available



Sodium Nitroprusside

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

SECTION 10: Stability and reactivity

10.1. Reactivity

No additional information available

10.2. Chemical stability

Stable under normal conditions.

10.3. Possibility of hazardous reactions

Contact with acids liberates toxic gas.

10.4. Conditions to avoid

Direct sunlight. Extremely high or low temperatures.

10.5. Incompatible materials

Strong acids. Strong oxidizers.

10.6. Hazardous decomposition products

Hydrogen cyanide. Carbon monoxide. Carbon dioxide.

SECTION 11: Toxicological information

11.1. Information on toxicological effects

Acute toxicity : Toxic if swallowed.

Sodium Nitroprusside (\f)13755-38-9	
LD50 oral rat	99 mg/kg
ATE US (oral)	99.00000000 mg/kg bodyweight

Skin corrosion/irritation : Not classified

Serious eye damage/irritation : Not classified

Respiratory or skin sensitisation : Not classified

Germ cell mutagenicity : Not classified

Carcinogenicity : Not classified

Reproductive toxicity : Not classified

Specific target organ toxicity (single exposure) : Not classified

Specific target organ toxicity (repeated exposure) : Not classified

Aspiration hazard : Not classified

Potential Adverse human health effects and symptoms : Based on available data, the classification criteria are not met. Toxic if swallowed.

Symptoms/injuries after ingestion : Toxic if swallowed. Swallowing a small quantity of this material will result in serious health hazard.

SECTION 12: Ecological information

12.1. Toxicity

No additional information available

12.2. Persistence and degradability

Sodium Nitroprusside (13755-38-9)	
Persistence and degradability	Not established.

12.3. Bioaccumulative potential

Sodium Nitroprusside (13755-38-9)	
Bioaccumulative potential	Not established.

12.4. Mobility in soil

No additional information available

12.5. Other adverse effects

Effect on ozone layer : No additional information available

Other information : Avoid release to the environment.

SECTION 13: Disposal considerations

13.1. Waste treatment methods

Waste disposal recommendations : Dispose in a safe manner in accordance with local/national regulations. Dispose of contents/container to comply with local, state and federal regulations.

Ecology - waste materials : Avoid release to the environment. Hazardous waste due to toxicity.

SECTION 14: Transport information

In accordance with DOT

Transport document description : UN1588 Cyanides, inorganic, solid, n.o.s. Sodium nitroferricyanide, 6.1, III

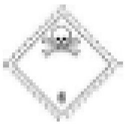
UN-No.(DOT) : 1588

DOT NA no. : UN1588

DOT Proper Shipping Name : Cyanides, inorganic, solid, n.o.s.
Sodium nitroferricyanide

Department of Transportation (DOT) Hazard Classes : 6.1 - Class 6.1 - Poisonous materials 49 CFR 173.132

Hazard labels (DOT) : 6.1 - Poison inhalation hazard



Packing group (DOT) : III - Minor Danger

DOT Special Provisions (49 CFR 172.102) : IB8 - Authorized IBCs: Metal (11A, 11B, 11N, 21A, 21B, 21N, 31A, 31B and 31N); Rigid plastics (11H1, 11H2, 21H1, 21H2, 31H1 and 31H2); Composite (11HZ1, 11HZ2, 21HZ1, 21HZ2, 31HZ1 and 31HZ2); Fiberboard (11G); Wooden (11C, 11D and 11F); Flexible (13H1, 13H2, 13H3, 13H4, 13H5, 13L1, 13L2, 13L3, 13L4, 13M1 or 13M2).
IP3 - Flexible IBCs must be sift-proof and water-resistant or must be fitted with a sift-proof and water-resistant liner.
N74 - Packages consisting of tightly closed inner containers of glass, earthenware, metal or polyethylene, capacity not over 0.5 kg (1.1 pounds) securely cushioned and packed in outer wooden barrels or wooden or fiberboard boxes, not over 15 kg (33 pounds) net weight, are authorized and need not conform to the requirements of part 178 of this subchapter.
N75 - Packages consisting of tightly closed inner packagings of glass, earthenware or metal, securely cushioned and packed in outer wooden barrels or wooden or fiberboard boxes, capacity not over 2.5 kg (5.5 pounds) net weight, are authorized and need not conform to the requirements of part 178 of this subchapter.
T1 - 1.5 178.274(d)(2) Normal........... 178.275(d)(2)
TP33 - The portable tank instruction assigned for this substance applies for granular and powdered solids and for solids which are filled and discharged at temperatures above their melting point which are cooled and transported as a solid mass. Solid substances transported or offered for transport above their melting point are authorized for transportation in portable tanks conforming to the provisions of portable tank instruction T4 for solid substances of packing group III or T7 for solid substances of packing group II, unless a tank with more stringent requirements for minimum shell thickness, maximum allowable working pressure, pressure-relief devices or bottom outlets are assigned in which case the more stringent tank instruction and special provisions shall apply. Filling limits must be in accordance with portable tank special provision TP3. Solids meeting the definition of an elevated temperature material must be transported in accordance with the applicable requirements of this subchapter.

DOT Packaging Exceptions (49 CFR 173.xxx) : 153

DOT Packaging Non Bulk (49 CFR 173.xxx) : 213

DOT Packaging Bulk (49 CFR 173.xxx) : 240

DOT Quantity Limitations Passenger aircraft/rail (49 CFR 173.27) : 100 kg

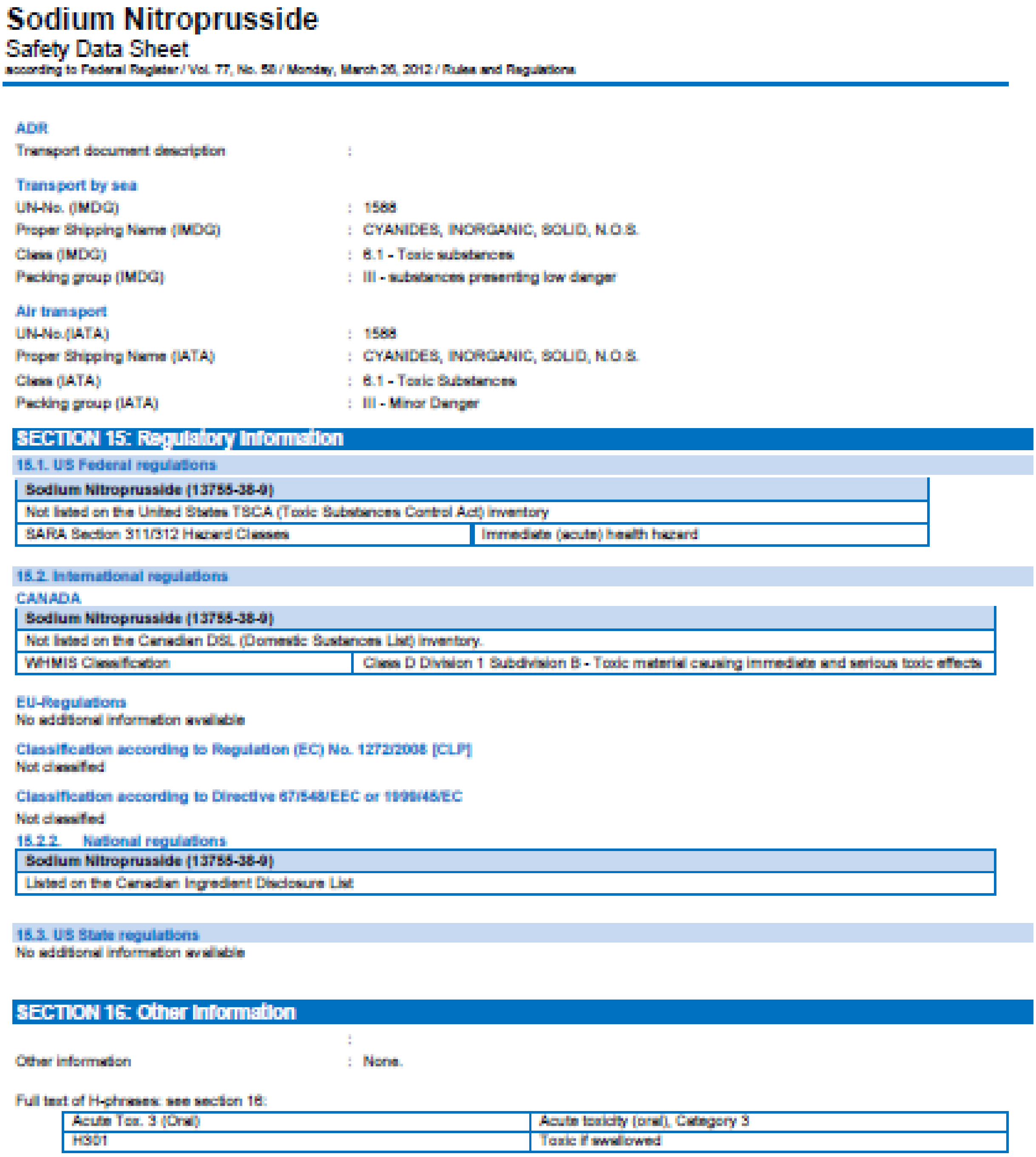
DOT Quantity Limitations Cargo aircraft only (49 CFR 175.75) : 200 kg

DOT Vessel Stowage Location : A - The material may be stowed "on deck" or "under deck" on a cargo vessel and on a passenger vessel.

DOT Vessel Stowage Other : 52 - Stow "separated from" acids

Additional information

Other information : No supplementary information available.



Sodium Nitroprusside

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

ADR

Transport document description :

Transport by sea

UN-No. (IMDG)	: 1588
Proper Shipping Name (IMDG)	: CYANIDES, INORGANIC, SOLID, N.O.S.
Class (IMDG)	: 6.1 - Toxic substances
Packing group (IMDG)	: III - substances presenting low danger

Air transport

UN-No.(IATA)	: 1588
Proper Shipping Name (IATA)	: CYANIDES, INORGANIC, SOLID, N.O.S.
Class (IATA)	: 6.1 - Toxic Substances
Packing group (IATA)	: III - Minor Danger

SECTION 15: Regulatory information

15.1. US Federal regulations

Sodium Nitroprusside (13755-38-9)	
Not listed on the United States TSCA (Toxic Substances Control Act) inventory	
SARA Section 311/312 Hazard Classes	Immediate (acute) health hazard

15.2. International regulations

CANADA

Sodium Nitroprusside (13755-38-9)	
Not listed on the Canadian DSL (Domestic Sustances List) inventory.	
WHMIS Classification	Class D Division 1 Subdivision B - Toxic material causing immediate and serious toxic effects

EU-Regulations

No additional information available

Classification according to Regulation (EC) No. 1272/2008 [CLP]

Not classified

Classification according to Directive 67/548/EEC or 1999/45/EC

Not classified

15.2.2. National regulations

Sodium Nitroprusside (13755-38-9)
Listed on the Canadian Ingredient Disclosure List

15.3. US State regulations

No additional information available

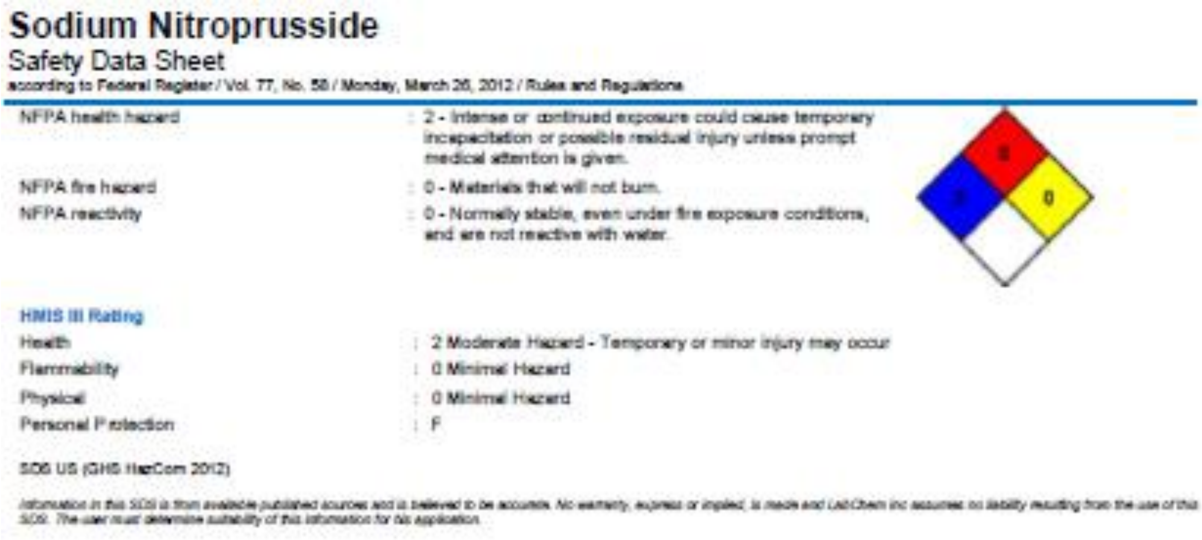
SECTION 16: Other information

:

Other information : None.

Full text of H-phrases: see section 16:

Acute Tox. 3 (Oral)	Acute toxicity (oral), Category 3
H301	Toxic if swallowed



Sodium Nitroprusside

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

NFPA health hazard	: 2 - Intense or continued exposure could cause temporary incapacitation or possible residual injury unless prompt medical attention is given.
NFPA fire hazard	: 0 - Materials that will not burn.
NFPA reactivity	: 0 - Normally stable, even under fire exposure conditions, and are not reactive with water.

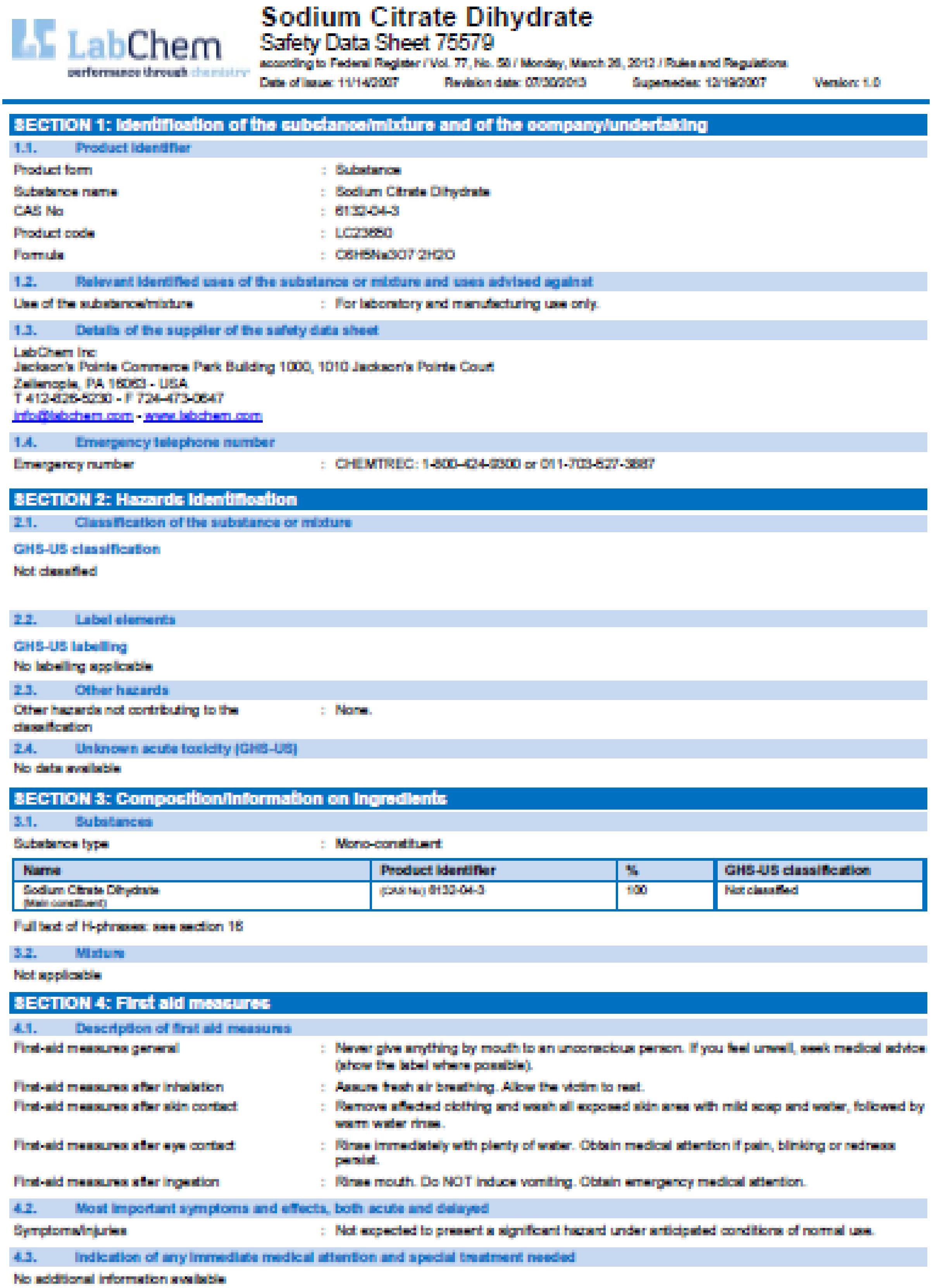
HMIS III Rating

Health	: 2 Moderate Hazard - Temporary or minor injury may occur
Flammability	: 0 Minimal Hazard
Physical	: 0 Minimal Hazard
Personal Protection	: F

SDS US (GHS HazCom 2012)

Information in this SDS is from available published sources and is believed to be accurate. No warranty, express or implied, is made and LabChem Inc assumes no liability resulting from the use of this SDS. The user must determine suitability of this information for his application.

A3.8. Sodium Citrate Dihydrated



LabChem performance through chemistry

Sodium Citrate Dihydrate

Safety Data Sheet 75579

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Date of issue: 11/14/2007 Revision date: 07/30/2013 Supersedes: 12/19/2007 Version: 1.0

SECTION 1: Identification of the substance/mixture and of the company/undertaking

1.1. Product identifier

Product form : Substance
Substance name : Sodium Citrate Dihydrate
CAS No : 6132-04-3
Product code : LC23650
Formula : C6H5Na3O7·2H2O

1.2. Relevant identified uses of the substance or mixture and uses advised against

Use of the substance/mixture : For laboratory and manufacturing use only.

1.3. Details of the supplier of the safety data sheet

LabChem Inc
Jackson's Pointe Commerce Park Building 1000, 1010 Jackson's Pointe Court
Zelienople, PA 16063 - USA
T 412-826-5230 - F 724-473-0647
info@labchem.com - www.labchem.com

1.4. Emergency telephone number

Emergency number : CHEMTREC: 1-800-424-9300 or 011-703-527-3887

SECTION 2: Hazards Identification

2.1. Classification of the substance or mixture

GHS-US classification

Not classified

2.2. Label elements

GHS-US labelling

No labelling applicable

2.3. Other hazards

Other hazards not contributing to the classification : None.

2.4. Unknown acute toxicity (GHS-US)

No data available

SECTION 3: Composition/information on ingredients

3.1. Substances

Substance type : Mono-constituent

Name	Product identifier	%	GHS-US classification
Sodium Citrate Dihydrate (Main constituent)	(CAS No) 6132-04-3	100	Not classified

Full text of H-phrases: see section 16

3.2. Mixture

Not applicable

SECTION 4: First aid measures

4.1. Description of first aid measures

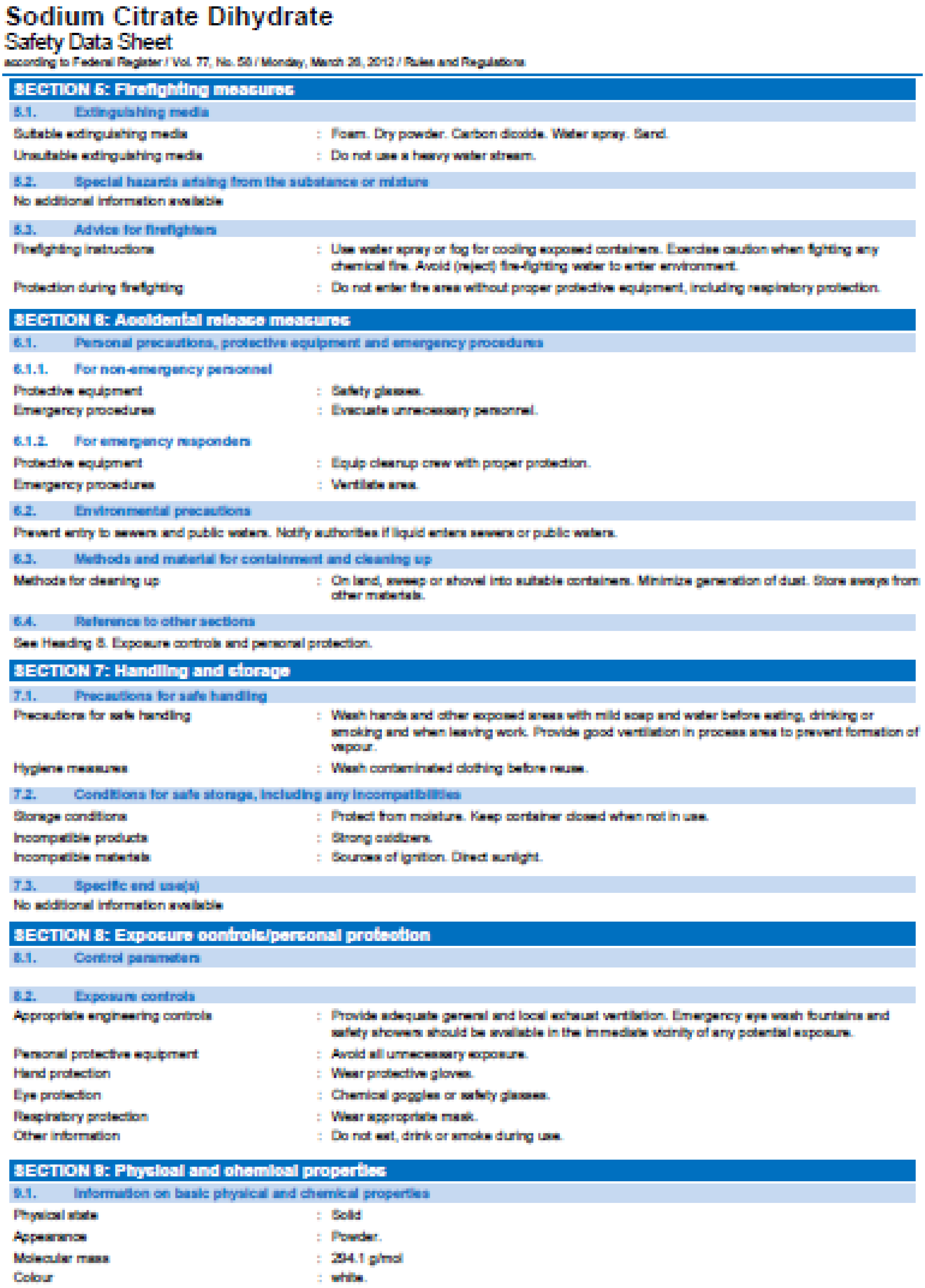
First-aid measures general : Never give anything by mouth to an unconscious person. If you feel unwell, seek medical advice (show the label where possible).
First-aid measures after inhalation : Assure fresh air breathing. Allow the victim to rest.
First-aid measures after skin contact : Remove affected clothing and wash all exposed skin area with mild soap and water, followed by warm water rinse.
First-aid measures after eye contact : Rinse immediately with plenty of water. Obtain medical attention if pain, blinking or redness persist.
First-aid measures after ingestion : Rinse mouth. Do NOT induce vomiting. Obtain emergency medical attention.

4.2. Most important symptoms and effects, both acute and delayed

Symptoms/injuries : Not expected to present a significant hazard under anticipated conditions of normal use.

4.3. Indication of any immediate medical attention and special treatment needed

No additional information available



Sodium Citrate Dihydrate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

SECTION 5: Firefighting measures

5.1. Extinguishing media

Suitable extinguishing media : Foam. Dry powder. Carbon dioxide. Water spray. Sand.

Unsuitable extinguishing media : Do not use a heavy water stream.

5.2. Special hazards arising from the substance or mixture

No additional information available

5.3. Advice for firefighters

Firefighting instructions : Use water spray or fog for cooling exposed containers. Exercise caution when fighting any chemical fire. Avoid (reject) fire-fighting water to enter environment.

Protection during firefighting : Do not enter fire area without proper protective equipment, including respiratory protection.

SECTION 6: Accidental release measures

6.1. Personal precautions, protective equipment and emergency procedures

6.1.1. For non-emergency personnel

Protective equipment : Safety glasses.

Emergency procedures : Evacuate unnecessary personnel.

6.1.2. For emergency responders

Protective equipment : Equip cleanup crew with proper protection.

Emergency procedures : Ventilate area.

6.2. Environmental precautions

Prevent entry to sewers and public waters. Notify authorities if liquid enters sewers or public waters.

6.3. Methods and material for containment and cleaning up

Methods for cleaning up : On land, sweep or shovel into suitable containers. Minimize generation of dust. Store away from other materials.

6.4. Reference to other sections

See Heading 8. Exposure controls and personal protection.

SECTION 7: Handling and storage

7.1. Precautions for safe handling

Precautions for safe handling : Wash hands and other exposed areas with mild soap and water before eating, drinking or smoking and when leaving work. Provide good ventilation in process area to prevent formation of vapour.

Hygiene measures : Wash contaminated clothing before reuse.

7.2. Conditions for safe storage, including any incompatibilities

Storage conditions : Protect from moisture. Keep container closed when not in use.

Incompatible products : Strong oxidizers.

Incompatible materials : Sources of ignition. Direct sunlight.

7.3. Specific end use(s)

No additional information available

SECTION 8: Exposure controls/personal protection

8.1. Control parameters

8.2. Exposure controls

Appropriate engineering controls : Provide adequate general and local exhaust ventilation. Emergency eye wash fountains and safety showers should be available in the immediate vicinity of any potential exposure.

Personal protective equipment : Avoid all unnecessary exposure.

Hand protection : Wear protective gloves.

Eye protection : Chemical goggles or safety glasses.

Respiratory protection : Wear appropriate mask.

Other information : Do not eat, drink or smoke during use.

SECTION 9: Physical and chemical properties

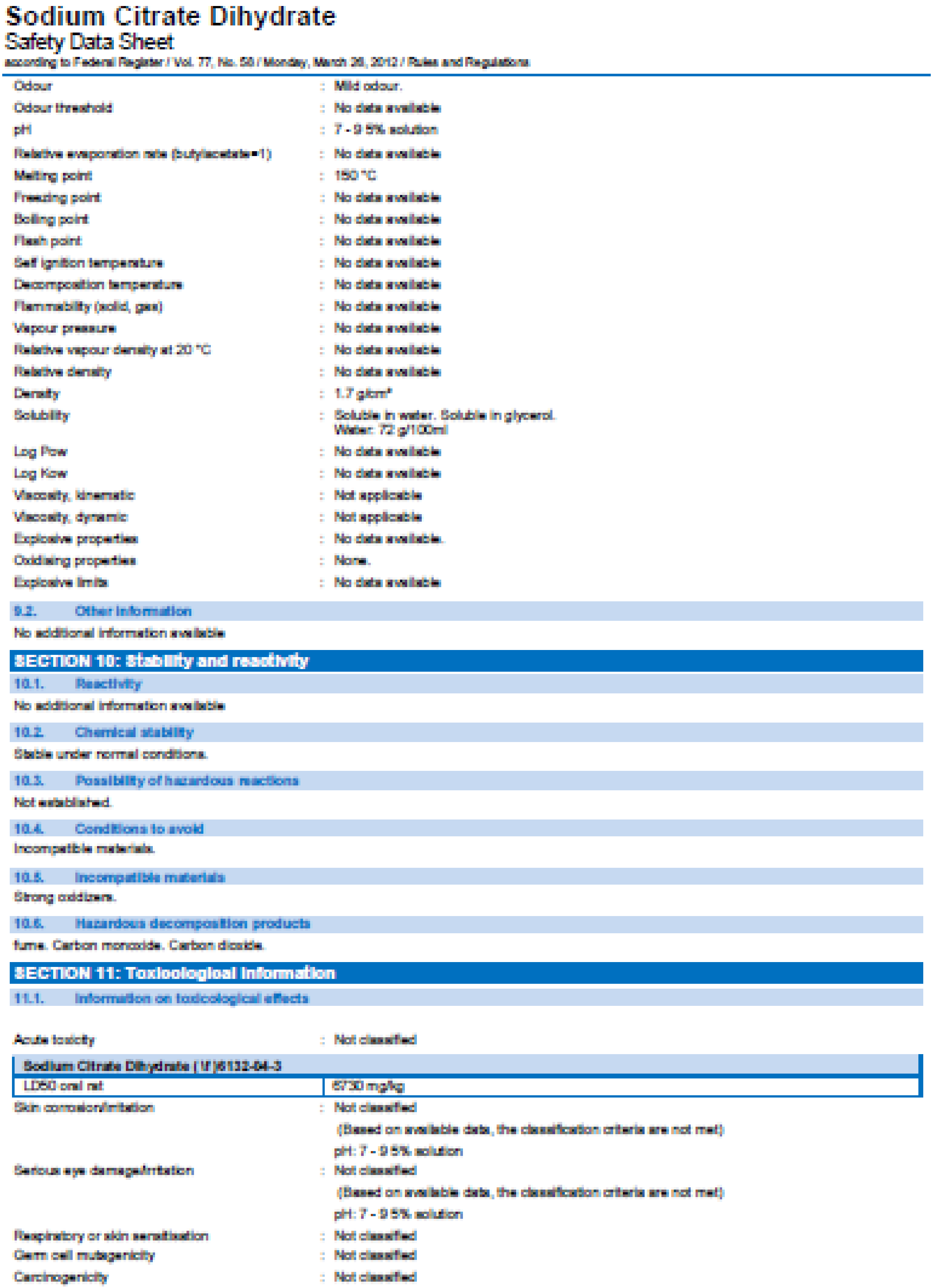
9.1. Information on basic physical and chemical properties

Physical state : Solid

Appearance : Powder.

Molecular mass : 294.1 g/mol

Colour : white.



Sodium Citrate Dihydrate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Odour : Mild odour.
Odour threshold : No data available
pH : 7 - 9 5% solution
Relative evaporation rate (butylacetate=1) : No data available
Melting point : 150 °C
Freezing point : No data available
Boiling point : No data available
Flash point : No data available
Self ignition temperature : No data available
Decomposition temperature : No data available
Flammability (solid, gas) : No data available
Vapour pressure : No data available
Relative vapour density at 20 °C : No data available
Relative density : No data available
Density : 1.7 g/cm³
Solubility : Soluble in water. Soluble in glycerol.
Water: 72 g/100ml
Log Pow : No data available
Log Kow : No data available
Viscosity, kinematic : Not applicable
Viscosity, dynamic : Not applicable
Explosive properties : No data available.
Oxidising properties : None.
Explosive limits : No data available

9.2. Other information

No additional information available

SECTION 10: Stability and reactivity

10.1. Reactivity

No additional information available

10.2. Chemical stability

Stable under normal conditions.

10.3. Possibility of hazardous reactions

Not established.

10.4. Conditions to avoid

Incompatible materials.

10.5. Incompatible materials

Strong oxidizers.

10.6. Hazardous decomposition products

fume. Carbon monoxide. Carbon dioxide.

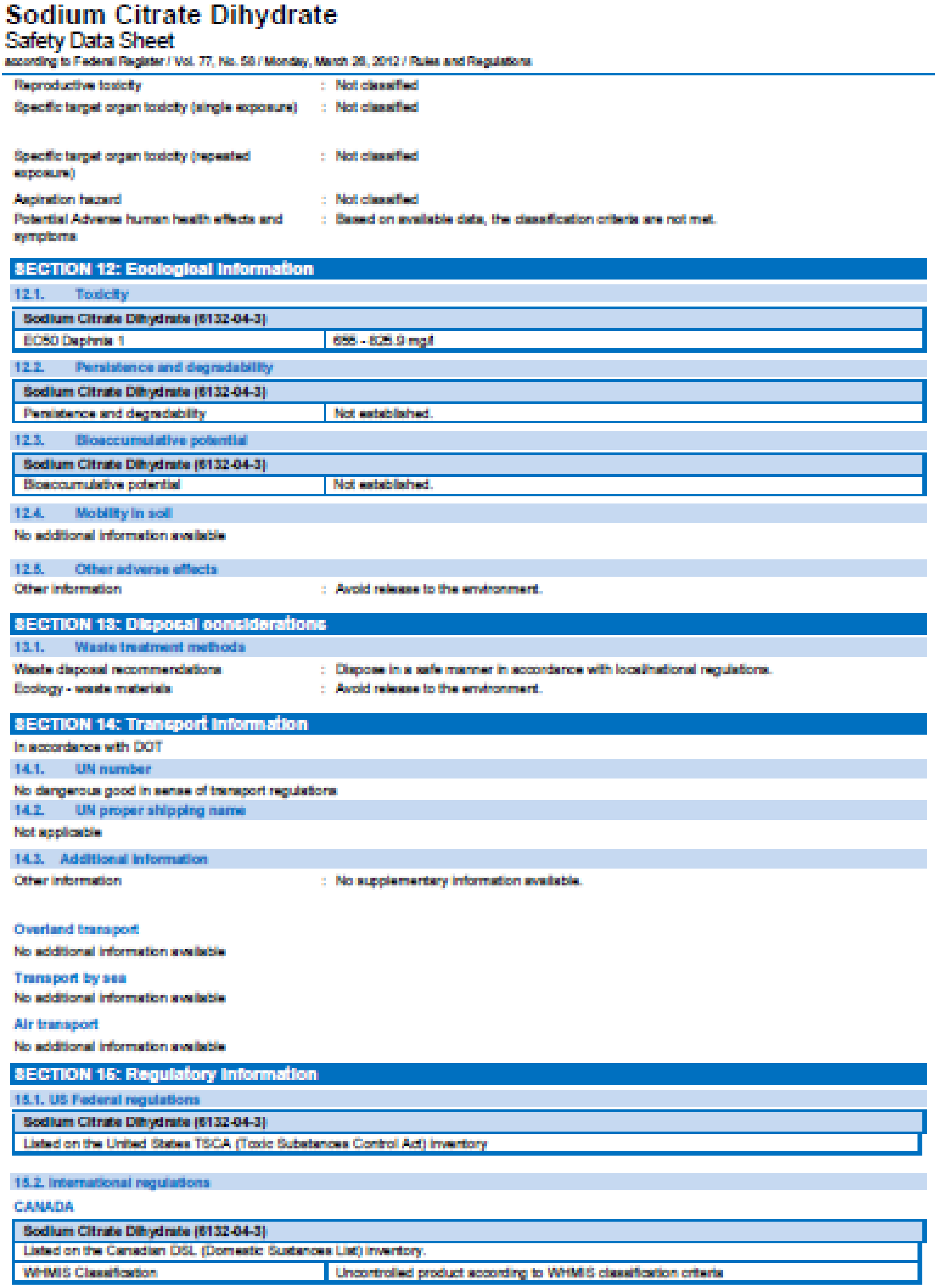
SECTION 11: Toxicological information

11.1. Information on toxicological effects

Acute toxicity : Not classified

Sodium Citrate Dihydrate (\f)6132-04-3	
LD50 oral rat	6730 mg/kg

Skin corrosion/irritation : Not classified
(Based on available data, the classification criteria are not met)
pH: 7 - 9 5% solution
Serious eye damage/irritation : Not classified
(Based on available data, the classification criteria are not met)
pH: 7 - 9 5% solution
Respiratory or skin sensitisation : Not classified
Germ cell mutagenicity : Not classified
Carcinogenicity : Not classified



Sodium Citrate Dihydrate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

Reproductive toxicity : Not classified

Specific target organ toxicity (single exposure) : Not classified

Specific target organ toxicity (repeated exposure) : Not classified

Aspiration hazard : Not classified

Potential Adverse human health effects and symptoms : Based on available data, the classification criteria are not met.

SECTION 12: Ecological Information

12.1. Toxicity

Sodium Citrate Dihydrate (6132-04-3)	
EC50 Daphnia 1	655 - 825.9 mg/l

12.2. Persistence and degradability

Sodium Citrate Dihydrate (6132-04-3)	
Persistence and degradability	Not established.

12.3. Bioaccumulative potential

Sodium Citrate Dihydrate (6132-04-3)	
Bioaccumulative potential	Not established.

12.4. Mobility in soil

No additional information available

12.5. Other adverse effects

Other information : Avoid release to the environment.

SECTION 13: Disposal considerations

13.1. Waste treatment methods

Waste disposal recommendations : Dispose in a safe manner in accordance with local/national regulations.

Ecology - waste materials : Avoid release to the environment.

SECTION 14: Transport information

In accordance with DOT

14.1. UN number

No dangerous good in sense of transport regulations

14.2. UN proper shipping name

Not applicable

14.3. Additional information

Other information : No supplementary information available.

Overland transport

No additional information available

Transport by sea

No additional information available

Air transport

No additional information available

SECTION 15: Regulatory information

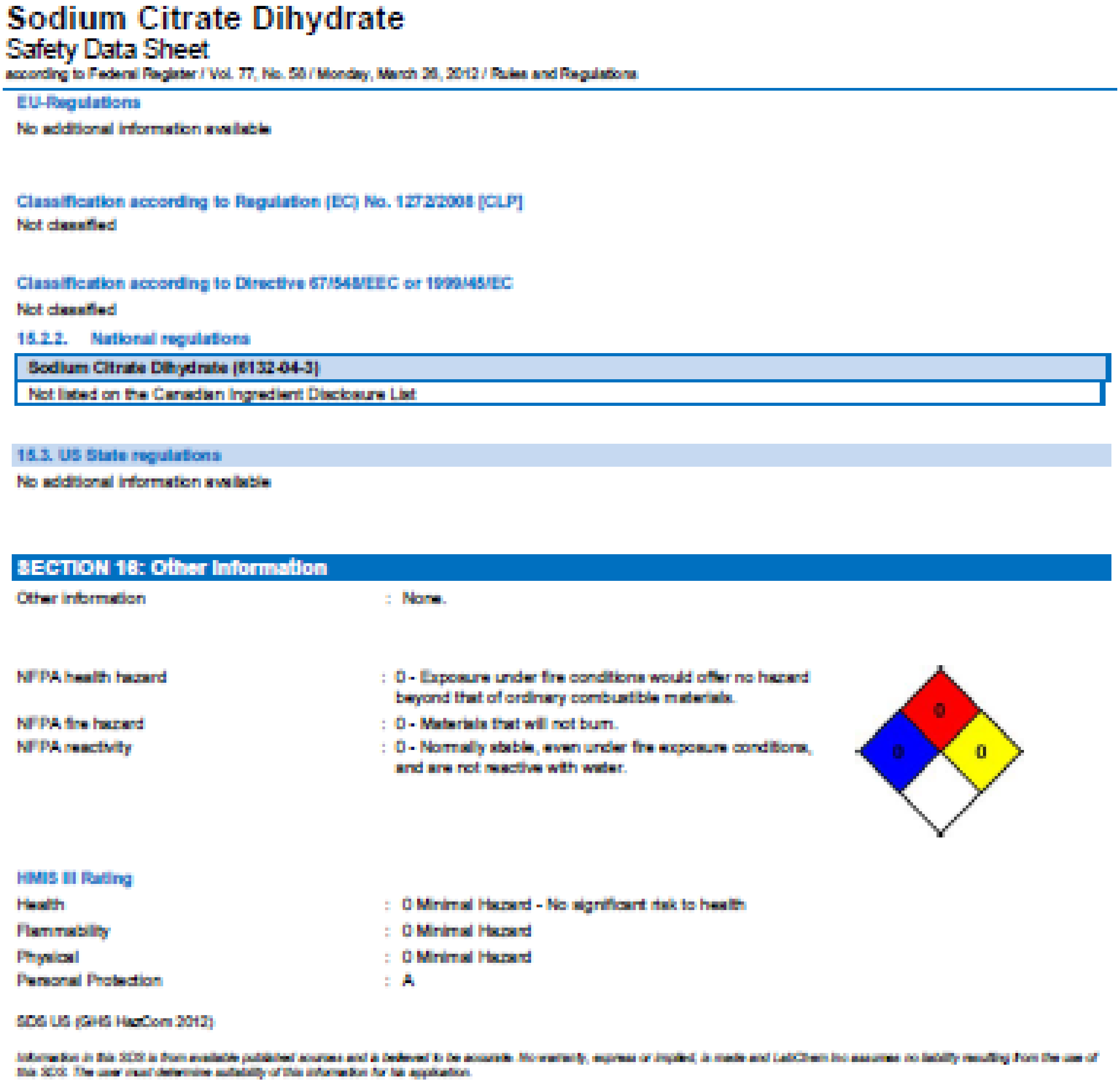
15.1. US Federal regulations

Sodium Citrate Dihydrate (6132-04-3)
Listed on the United States TSCA (Toxic Substances Control Act) inventory

15.2. International regulations

CANADA

Sodium Citrate Dihydrate (6132-04-3)	
Listed on the Canadian DSL (Domestic Substances List) inventory.	
WHMIS Classification	Uncontrolled product according to WHMIS classification criteria



Sodium Citrate Dihydrate

Safety Data Sheet

according to Federal Register / Vol. 77, No. 58 / Monday, March 26, 2012 / Rules and Regulations

EU-Regulations

No additional information available

Classification according to Regulation (EC) No. 1272/2008 [CLP]

Not classified

Classification according to Directive 67/548/EEC or 1999/45/EC

Not classified

15.2.2. National regulations

Sodium Citrate Dihydrate (6132-04-3)
Not listed on the Canadian Ingredient Disclosure List

15.3. US State regulations

No additional information available

SECTION 16: Other information

Other information : None.

NFPA health hazard : 0 - Exposure under fire conditions would offer no hazard beyond that of ordinary combustible materials.

NFPA fire hazard : 0 - Materials that will not burn.

NFPA reactivity : 0 - Normally stable, even under fire exposure conditions, and are not reactive with water.

HMIS III Rating

Health : 0 Minimal Hazard - No significant risk to health

Flammability : 0 Minimal Hazard

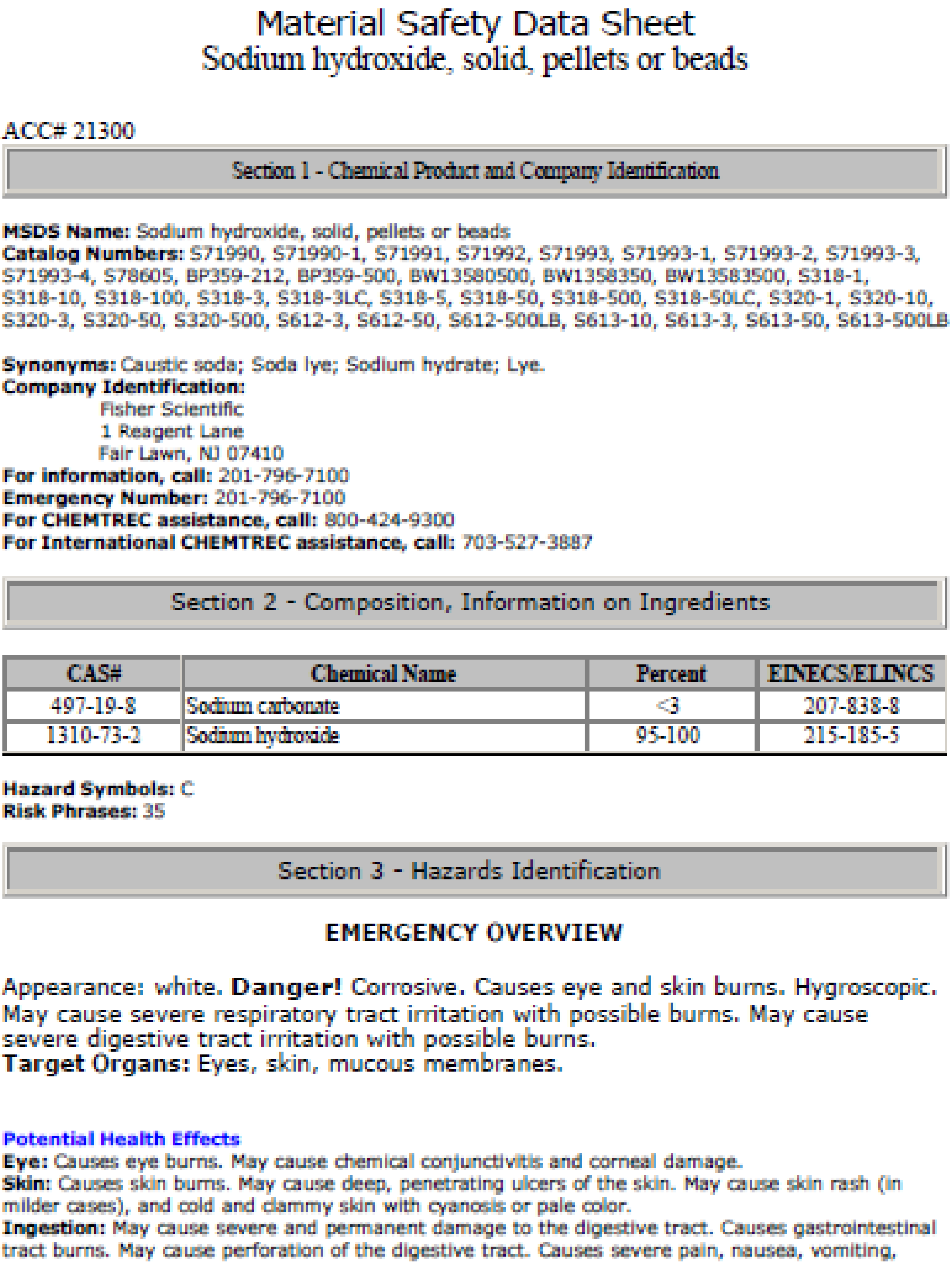
Physical : 0 Minimal Hazard

Personal Protection : A

SDS US (GHS HazCom 2012)

Information in this SDS is from available published sources and is believed to be accurate. No warranty, express or implied, is made and LabChem Inc assumes no liability resulting from the use of this SDS. The user must determine suitability of this information for his application.

A3.9. Sodium hydroxide



Material Safety Data Sheet
Sodium hydroxide, solid, pellets or beads

ACC# 21300

Section 1 - Chemical Product and Company Identification

MSDS Name: Sodium hydroxide, solid, pellets or beads
Catalog Numbers: S71990, S71990-1, S71991, S71992, S71993, S71993-1, S71993-2, S71993-3, S71993-4, S78605, BP359-212, BP359-500, BW13580500, BW1358350, BW13583500, S318-1, S318-10, S318-100, S318-3, S318-3LC, S318-5, S318-50, S318-500, S318-50LC, S320-1, S320-10, S320-3, S320-50, S320-500, S612-3, S612-50, S612-500LB, S613-10, S613-3, S613-50, S613-500LB

Synonyms: Caustic soda; Soda lye; Sodium hydrate; Lye.
Company Identification:
Fisher Scientific
1 Reagent Lane
Fair Lawn, NJ 07410
For information, call: 201-796-7100
Emergency Number: 201-796-7100
For CHEMTREC assistance, call: 800-424-9300
For International CHEMTREC assistance, call: 703-527-3887

Section 2 - Composition, Information on Ingredients

CAS#	Chemical Name	Percent	EINECS/ELINCS
497-19-8	Sodium carbonate	<3	207-838-8
1310-73-2	Sodium hydroxide	95-100	215-185-5

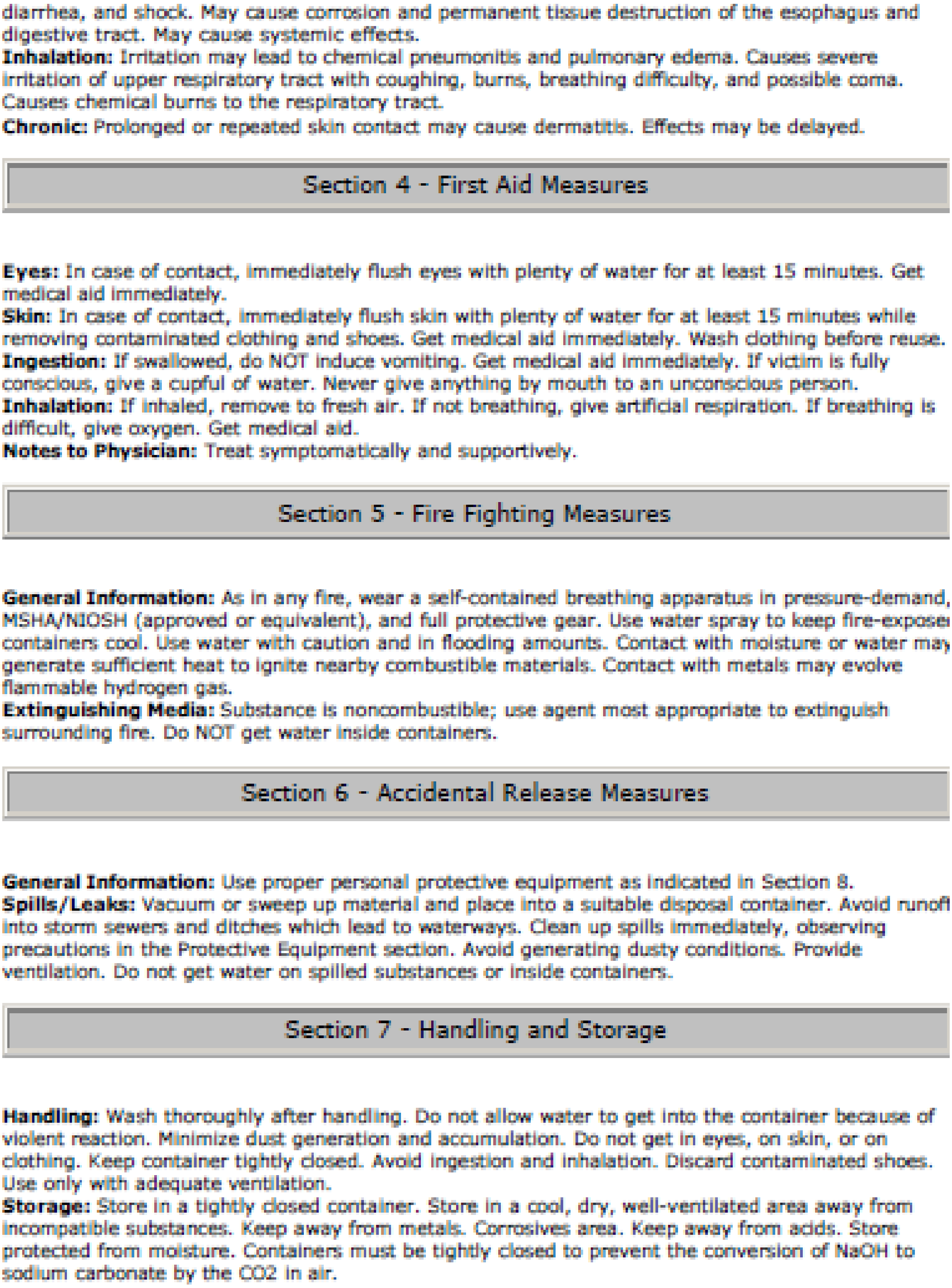
Hazard Symbols: C
Risk Phrases: 35

Section 3 - Hazards Identification

EMERGENCY OVERVIEW

Appearance: white. **Danger!** Corrosive. Causes eye and skin burns. Hygroscopic. May cause severe respiratory tract irritation with possible burns. May cause severe digestive tract irritation with possible burns.
Target Organs: Eyes, skin, mucous membranes.

Potential Health Effects
Eye: Causes eye burns. May cause chemical conjunctivitis and corneal damage.
Skin: Causes skin burns. May cause deep, penetrating ulcers of the skin. May cause skin rash (in milder cases), and cold and clammy skin with cyanosis or pale color.
Ingestion: May cause severe and permanent damage to the digestive tract. Causes gastrointestinal tract burns. May cause perforation of the digestive tract. Causes severe pain, nausea, vomiting,



diarrhea, and shock. May cause corrosion and permanent tissue destruction of the esophagus and digestive tract. May cause systemic effects.
Inhalation: Irritation may lead to chemical pneumonitis and pulmonary edema. Causes severe irritation of upper respiratory tract with coughing, burns, breathing difficulty, and possible coma. Causes chemical burns to the respiratory tract.
Chronic: Prolonged or repeated skin contact may cause dermatitis. Effects may be delayed.

Section 4 - First Aid Measures

Eyes: In case of contact, immediately flush eyes with plenty of water for at least 15 minutes. Get medical aid immediately.
Skin: In case of contact, immediately flush skin with plenty of water for at least 15 minutes while removing contaminated clothing and shoes. Get medical aid immediately. Wash clothing before reuse.
Ingestion: If swallowed, do NOT induce vomiting. Get medical aid immediately. If victim is fully conscious, give a cupful of water. Never give anything by mouth to an unconscious person.
Inhalation: If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. Get medical aid.
Notes to Physician: Treat symptomatically and supportively.

Section 5 - Fire Fighting Measures

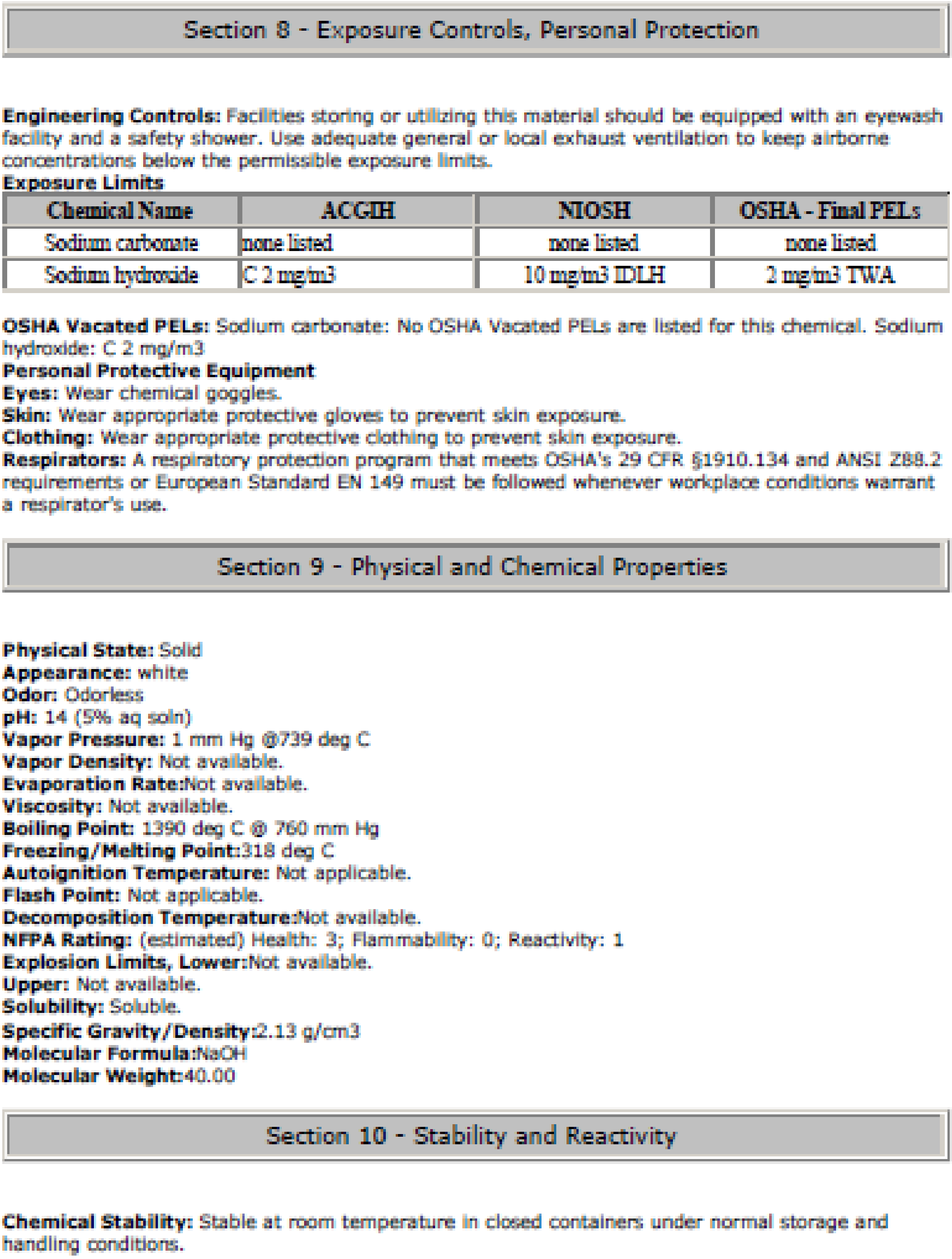
General Information: As in any fire, wear a self-contained breathing apparatus in pressure-demand, MSHA/NIOSH (approved or equivalent), and full protective gear. Use water spray to keep fire-expose containers cool. Use water with caution and in flooding amounts. Contact with moisture or water may generate sufficient heat to ignite nearby combustible materials. Contact with metals may evolve flammable hydrogen gas.
Extinguishing Media: Substance is noncombustible; use agent most appropriate to extinguish surrounding fire. Do NOT get water inside containers.

Section 6 - Accidental Release Measures

General Information: Use proper personal protective equipment as indicated in Section 8.
Spills/Leaks: Vacuum or sweep up material and place into a suitable disposal container. Avoid runoff into storm sewers and ditches which lead to waterways. Clean up spills immediately, observing precautions in the Protective Equipment section. Avoid generating dusty conditions. Provide ventilation. Do not get water on spilled substances or inside containers.

Section 7 - Handling and Storage

Handling: Wash thoroughly after handling. Do not allow water to get into the container because of violent reaction. Minimize dust generation and accumulation. Do not get in eyes, on skin, or on clothing. Keep container tightly closed. Avoid ingestion and inhalation. Discard contaminated shoes. Use only with adequate ventilation.
Storage: Store in a tightly closed container. Store in a cool, dry, well-ventilated area away from incompatible substances. Keep away from metals. Corrosives area. Keep away from acids. Store protected from moisture. Containers must be tightly closed to prevent the conversion of NaOH to sodium carbonate by the CO2 in air.



Section 8 - Exposure Controls, Personal Protection

Engineering Controls: Facilities storing or utilizing this material should be equipped with an eyewash facility and a safety shower. Use adequate general or local exhaust ventilation to keep airborne concentrations below the permissible exposure limits.

Exposure Limits

Chemical Name	ACGIH	NIOSH	OSHA - Final PELs
Sodium carbonate	none listed	none listed	none listed
Sodium hydroxide	C 2 mg/m3	10 mg/m3 IDLH	2 mg/m3 TWA

OSHA Vacated PELs: Sodium carbonate: No OSHA Vacated PELs are listed for this chemical. Sodium hydroxide: C 2 mg/m3

Personal Protective Equipment

Eyes: Wear chemical goggles.

Skin: Wear appropriate protective gloves to prevent skin exposure.

Clothing: Wear appropriate protective clothing to prevent skin exposure.

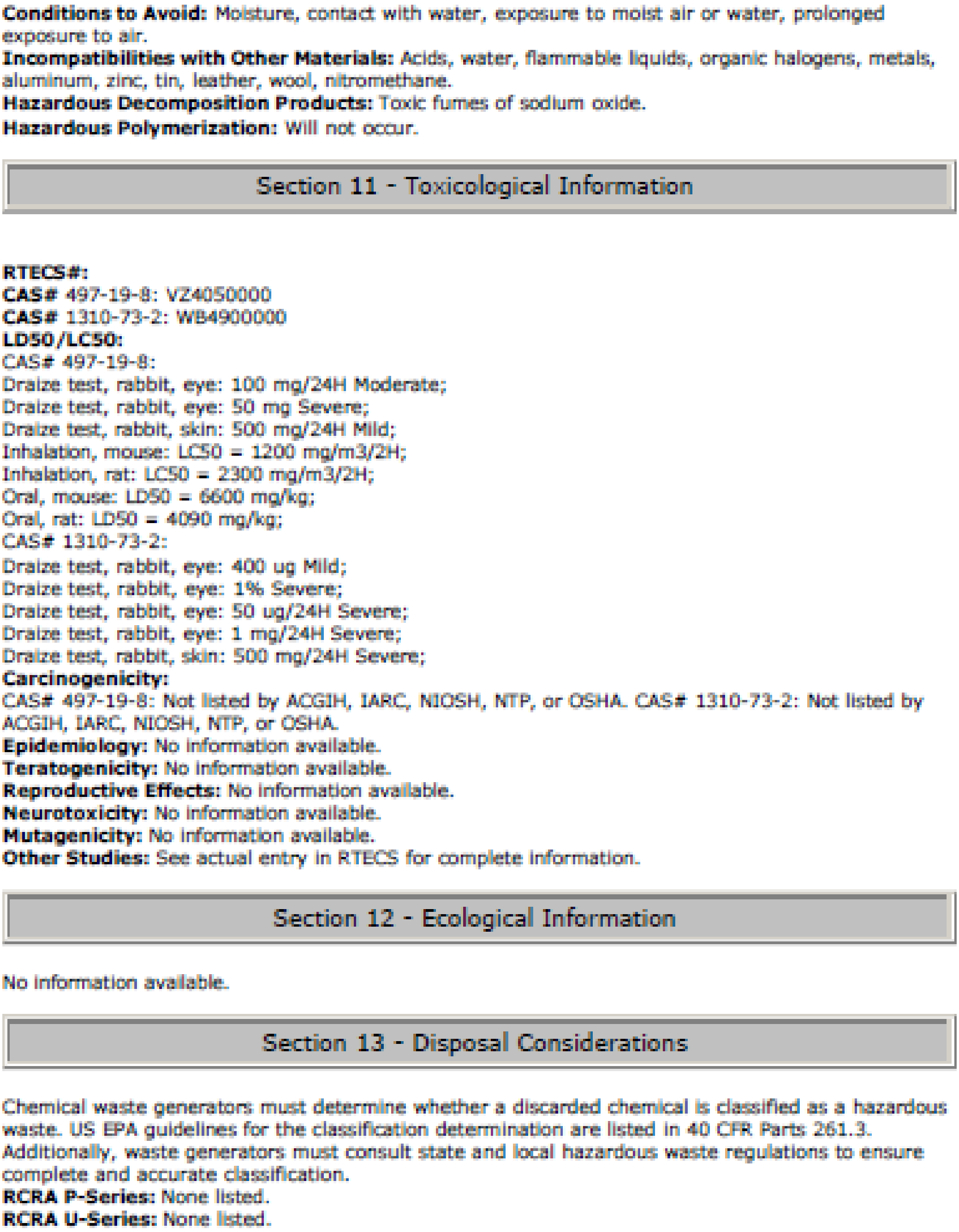
Respirators: A respiratory protection program that meets OSHA's 29 CFR §1910.134 and ANSI Z88.2 requirements or European Standard EN 149 must be followed whenever workplace conditions warrant a respirator's use.

Section 9 - Physical and Chemical Properties

Physical State: Solid
Appearance: white
Odor: Odorless
pH: 14 (5% aq soln)
Vapor Pressure: 1 mm Hg @739 deg C
Vapor Density: Not available.
Evaporation Rate: Not available.
Viscosity: Not available.
Boiling Point: 1390 deg C @ 760 mm Hg
Freezing/Melting Point:318 deg C
Autoignition Temperature: Not applicable.
Flash Point: Not applicable.
Decomposition Temperature: Not available.
NFPA Rating: (estimated) Health: 3; Flammability: 0; Reactivity: 1
Explosion Limits, Lower: Not available.
Upper: Not available.
Solubility: Soluble.
Specific Gravity/Density:2.13 g/cm3
Molecular Formula: NaOH
Molecular Weight:40.00

Section 10 - Stability and Reactivity

Chemical Stability: Stable at room temperature in closed containers under normal storage and handling conditions.



Conditions to Avoid: Moisture, contact with water, exposure to moist air or water, prolonged exposure to air.
Incompatibilities with Other Materials: Acids, water, flammable liquids, organic halogens, metals, aluminum, zinc, tin, leather, wool, nitromethane.
Hazardous Decomposition Products: Toxic fumes of sodium oxide.
Hazardous Polymerization: Will not occur.

Section 11 - Toxicological Information

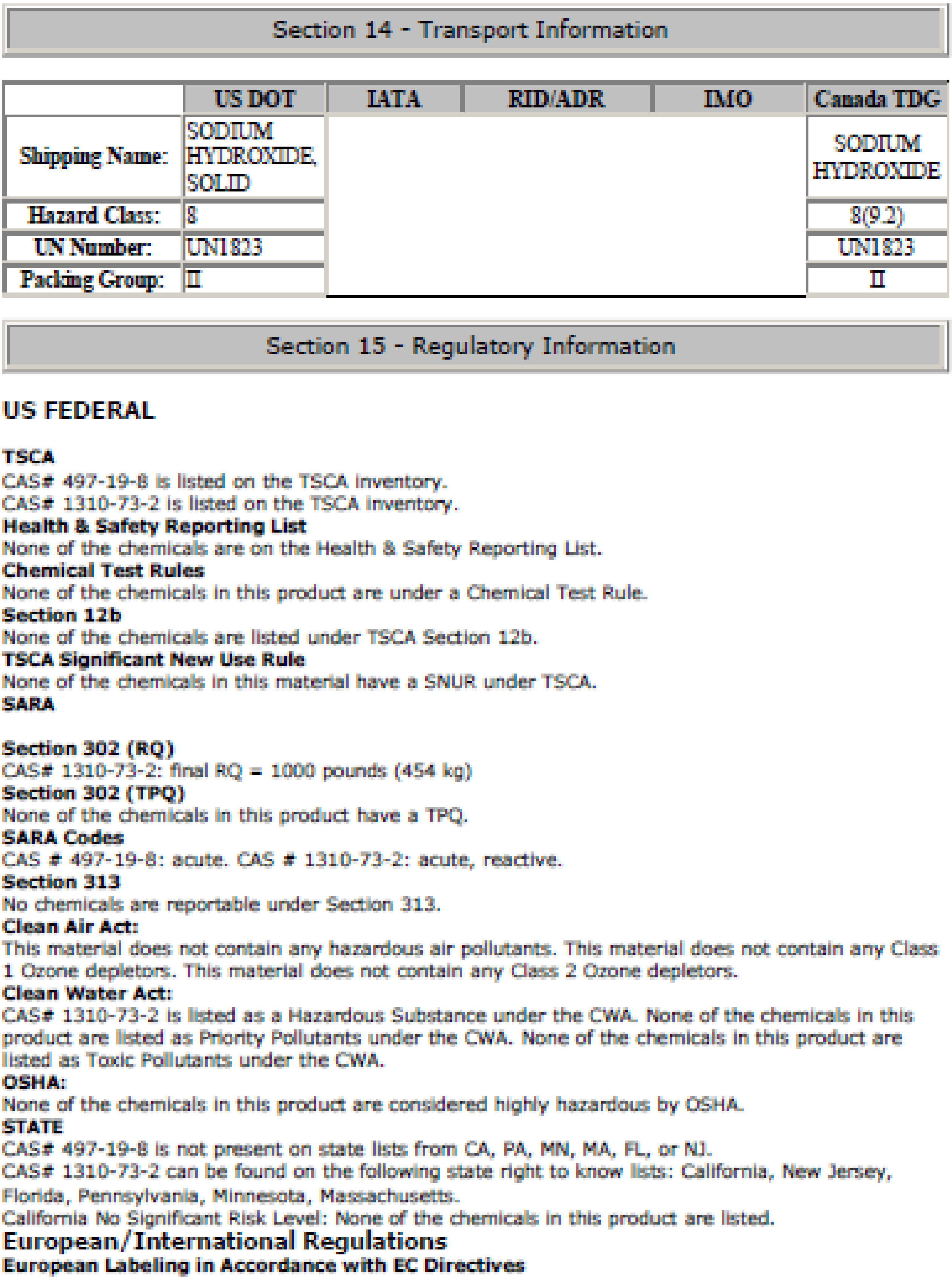
RTECS#:
CAS# 497-19-8: VZ4050000
CAS# 1310-73-2: WB4900000
LD50/LC50:
CAS# 497-19-8:
Draize test, rabbit, eye: 100 mg/24H Moderate;
Draize test, rabbit, eye: 50 mg Severe;
Draize test, rabbit, skin: 500 mg/24H Mild;
Inhalation, mouse: LC50 = 1200 mg/m3/2H;
Inhalation, rat: LC50 = 2300 mg/m3/2H;
Oral, mouse: LD50 = 6600 mg/kg;
Oral, rat: LD50 = 4090 mg/kg;
CAS# 1310-73-2:
Draize test, rabbit, eye: 400 ug Mild;
Draize test, rabbit, eye: 1% Severe;
Draize test, rabbit, eye: 50 ug/24H Severe;
Draize test, rabbit, eye: 1 mg/24H Severe;
Draize test, rabbit, skin: 500 mg/24H Severe;
Carcinogenicity:
CAS# 497-19-8: Not listed by ACGIH, IARC, NIOSH, NTP, or OSHA. CAS# 1310-73-2: Not listed by ACGIH, IARC, NIOSH, NTP, or OSHA.
Epidemiology: No information available.
Teratogenicity: No information available.
Reproductive Effects: No information available.
Neurotoxicity: No information available.
Mutagenicity: No information available.
Other Studies: See actual entry in RTECS for complete information.

Section 12 - Ecological Information

No information available.

Section 13 - Disposal Considerations

Chemical waste generators must determine whether a discarded chemical is classified as a hazardous waste. US EPA guidelines for the classification determination are listed in 40 CFR Parts 261.3. Additionally, waste generators must consult state and local hazardous waste regulations to ensure complete and accurate classification.
RCRA P-Series: None listed.
RCRA U-Series: None listed.



Section 14 - Transport Information

	US DOT	IATA	RID/ADR	IMO	Canada TDG
Shipping Name:	SODIUM HYDROXIDE, SOLID				SODIUM HYDROXIDE
Hazard Class:	8				8(9.2)
UN Number:	UN1823				UN1823
Packing Group:	II				II

Section 15 - Regulatory Information

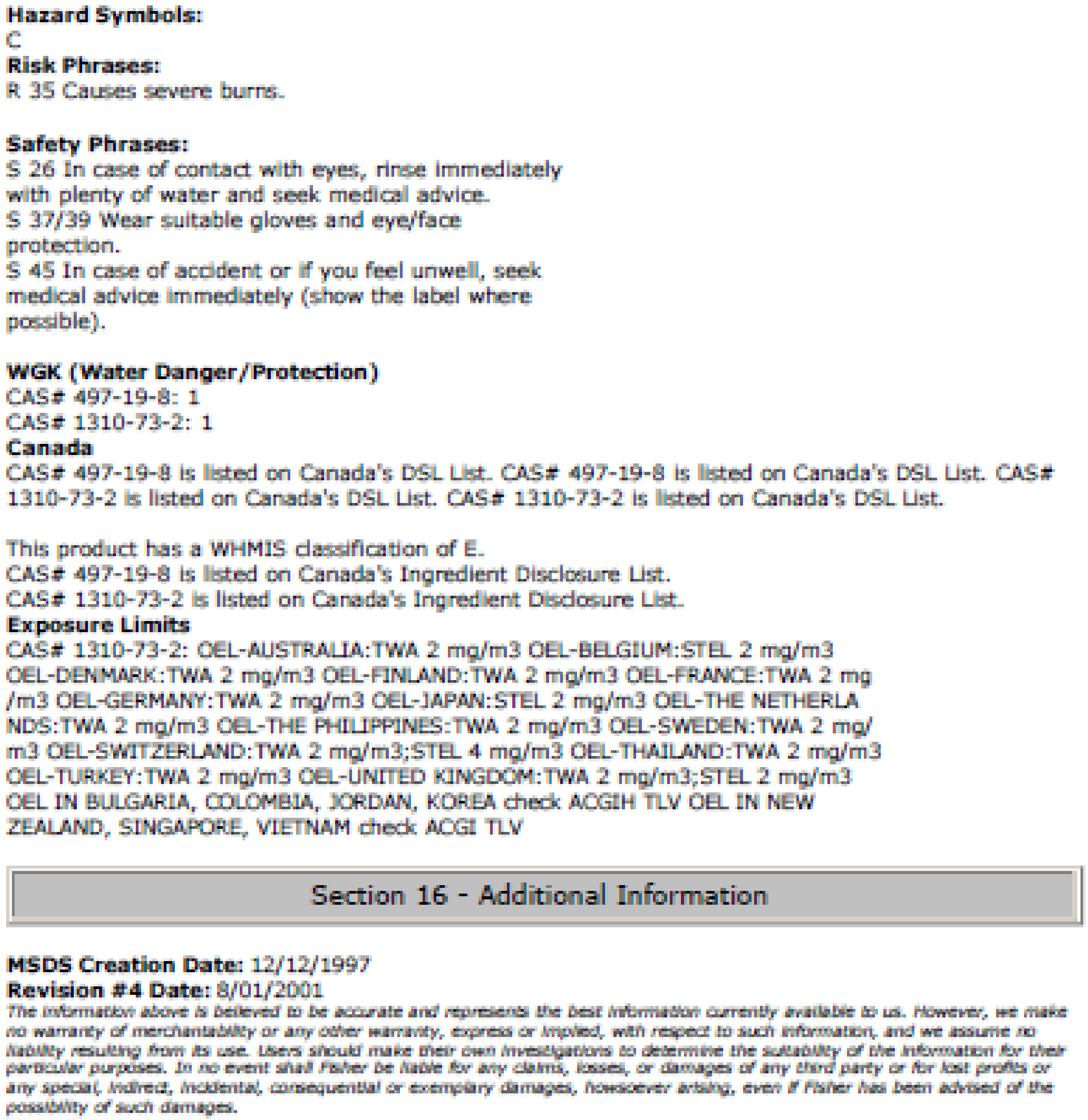
US FEDERAL

TSCA
CAS# 497-19-8 is listed on the TSCA inventory.
CAS# 1310-73-2 is listed on the TSCA inventory.
Health & Safety Reporting List
None of the chemicals are on the Health & Safety Reporting List.
Chemical Test Rules
None of the chemicals in this product are under a Chemical Test Rule.
Section 12b
None of the chemicals are listed under TSCA Section 12b.
TSCA Significant New Use Rule
None of the chemicals in this material have a SNUR under TSCA.
SARA

Section 302 (RQ)
CAS# 1310-73-2: final RQ = 1000 pounds (454 kg)
Section 302 (TPQ)
None of the chemicals in this product have a TPQ.
SARA Codes
CAS # 497-19-8: acute. CAS # 1310-73-2: acute, reactive.
Section 313
No chemicals are reportable under Section 313.
Clean Air Act:
This material does not contain any hazardous air pollutants. This material does not contain any Class 1 Ozone depletors. This material does not contain any Class 2 Ozone depletors.
Clean Water Act:
CAS# 1310-73-2 is listed as a Hazardous Substance under the CWA. None of the chemicals in this product are listed as Priority Pollutants under the CWA. None of the chemicals in this product are listed as Toxic Pollutants under the CWA.
OSHA:
None of the chemicals in this product are considered highly hazardous by OSHA.
STATE
CAS# 497-19-8 is not present on state lists from CA, PA, MN, MA, FL, or NJ.
CAS# 1310-73-2 can be found on the following state right to know lists: California, New Jersey, Florida, Pennsylvania, Minnesota, Massachusetts.
California No Significant Risk Level: None of the chemicals in this product are listed.

European/International Regulations

European Labeling in Accordance with EC Directives



Hazard Symbols:
C
Risk Phrases:
R 35 Causes severe burns.

Safety Phrases:
S 26 In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
S 37/39 Wear suitable gloves and eye/face protection.
S 45 In case of accident or if you feel unwell, seek medical advice immediately (show the label where possible).

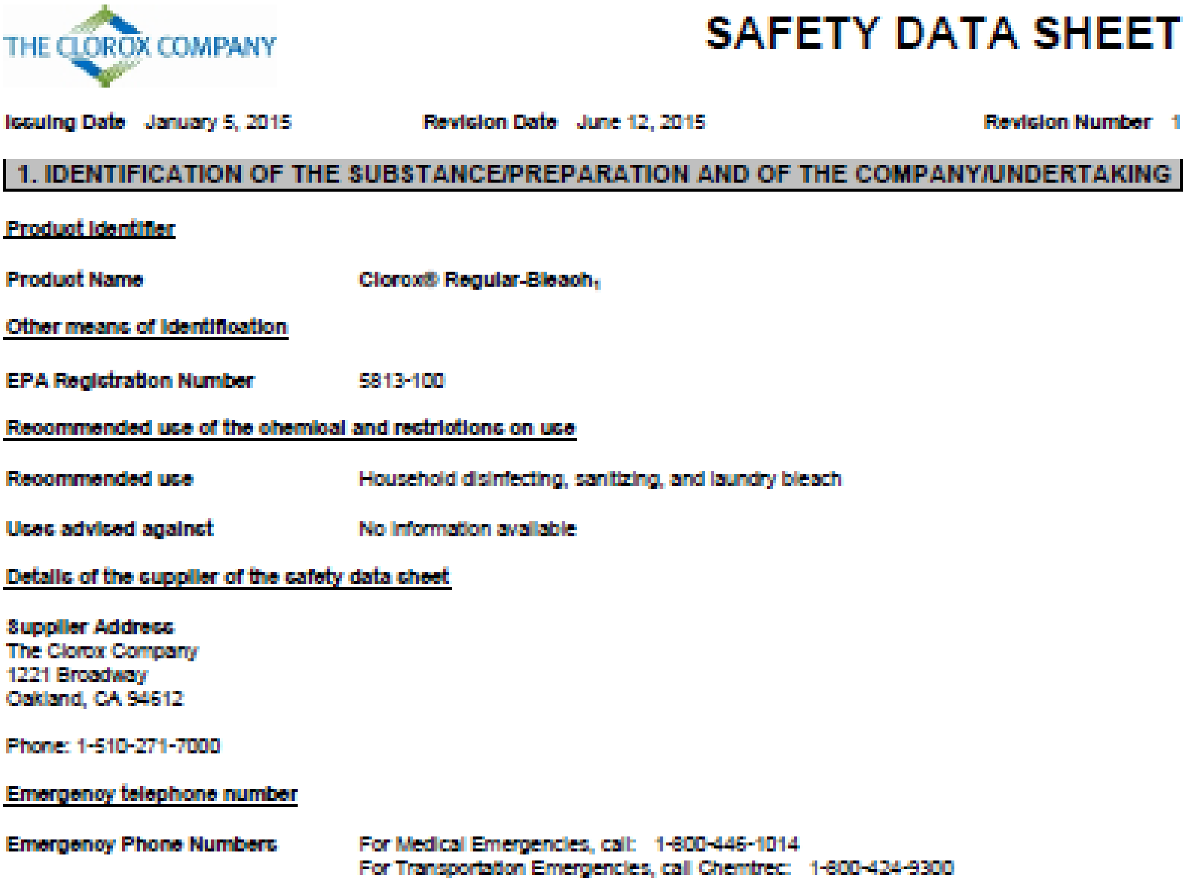
WGK (Water Danger/Protection)
CAS# 497-19-8: 1
CAS# 1310-73-2: 1
Canada
CAS# 497-19-8 is listed on Canada's DSL List. CAS# 497-19-8 is listed on Canada's DSL List. CAS# 1310-73-2 is listed on Canada's DSL List. CAS# 1310-73-2 is listed on Canada's DSL List.

This product has a WHMIS classification of E.
CAS# 497-19-8 is listed on Canada's Ingredient Disclosure List.
CAS# 1310-73-2 is listed on Canada's Ingredient Disclosure List.
Exposure Limits
CAS# 1310-73-2: OEL-AUSTRALIA:TWA 2 mg/m3 OEL-BELGIUM:STEL 2 mg/m3 OEL-DENMARK:TWA 2 mg/m3 OEL-FINLAND:TWA 2 mg/m3 OEL-FRANCE:TWA 2 mg/m3 OEL-GERMANY:TWA 2 mg/m3 OEL-JAPAN:STEL 2 mg/m3 OEL-THE NETHERLANDS:TWA 2 mg/m3 OEL-THE PHILIPPINES:TWA 2 mg/m3 OEL-SWEDEN:TWA 2 mg/m3 OEL-SWITZERLAND:TWA 2 mg/m3;STEL 4 mg/m3 OEL-THAILAND:TWA 2 mg/m3 OEL-TURKEY:TWA 2 mg/m3 OEL-UNITED KINGDOM:TWA 2 mg/m3;STEL 2 mg/m3 OEL IN BULGARIA, COLOMBIA, JORDAN, KOREA check ACGIH TLV OEL IN NEW ZEALAND, SINGAPORE, VIETNAM check ACGI TLV

Section 16 - Additional Information

MSDS Creation Date: 12/12/1997
Revision #4 Date: 8/01/2001

A3.10. Bleach



THE CLOROX COMPANY

SAFETY DATA SHEET

Issuing Date January 5, 2015 **Revision Date** June 12, 2015 **Revision Number** 1

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND OF THE COMPANY/UNDERTAKING

Product identifier

Product Name	**Clorox® Regular-Bleach$_1$**

Other means of identification

EPA Registration Number	5813-100

Recommended use of the chemical and restrictions on use

Recommended use	Household disinfecting, sanitizing, and laundry bleach
Uses advised against	No information available

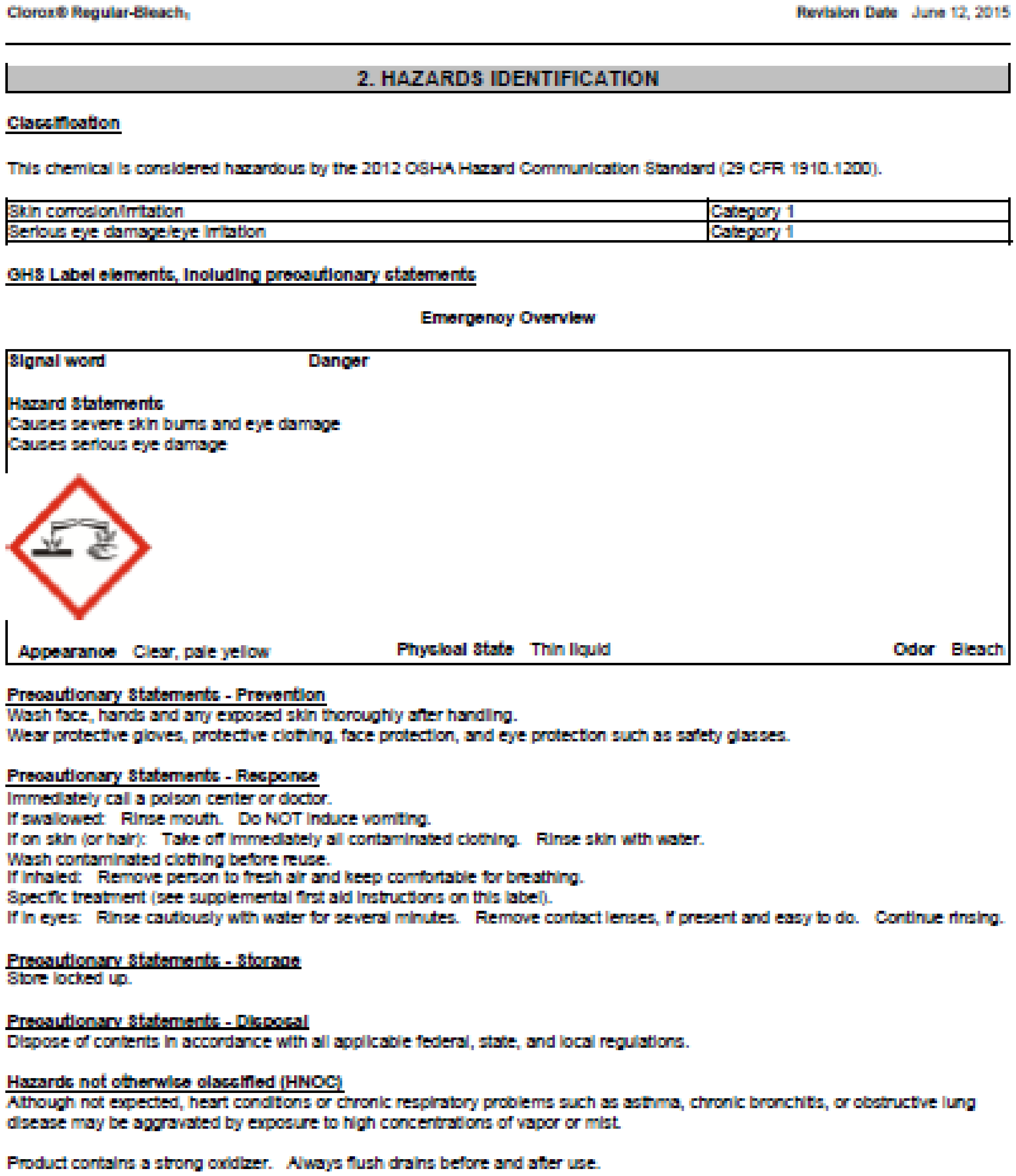
Details of the supplier of the safety data sheet

Supplier Address
The Clorox Company
1221 Broadway
Oakland, CA 94612

Phone: 1-510-271-7000

Emergency telephone number

Emergency Phone Numbers	For Medical Emergencies, call: 1-800-446-1014 For Transportation Emergencies, call Chemtrec: 1-800-424-9300



Clorox® Regular-Bleach₁ **Revision Date** June 12, 2015

2. HAZARDS IDENTIFICATION

Classification

This chemical is considered hazardous by the 2012 OSHA Hazard Communication Standard (29 CFR 1910.1200).

Skin corrosion/irritation	Category 1
Serious eye damage/eye irritation	Category 1

GHS Label elements, including precautionary statements

Emergency Overview

Signal word **Danger**

Hazard Statements
Causes severe skin burns and eye damage
Causes serious eye damage

Appearance Clear, pale yellow **Physical State** Thin liquid **Odor** Bleach

Precautionary Statements - Prevention
Wash face, hands and any exposed skin thoroughly after handling.
Wear protective gloves, protective clothing, face protection, and eye protection such as safety glasses.

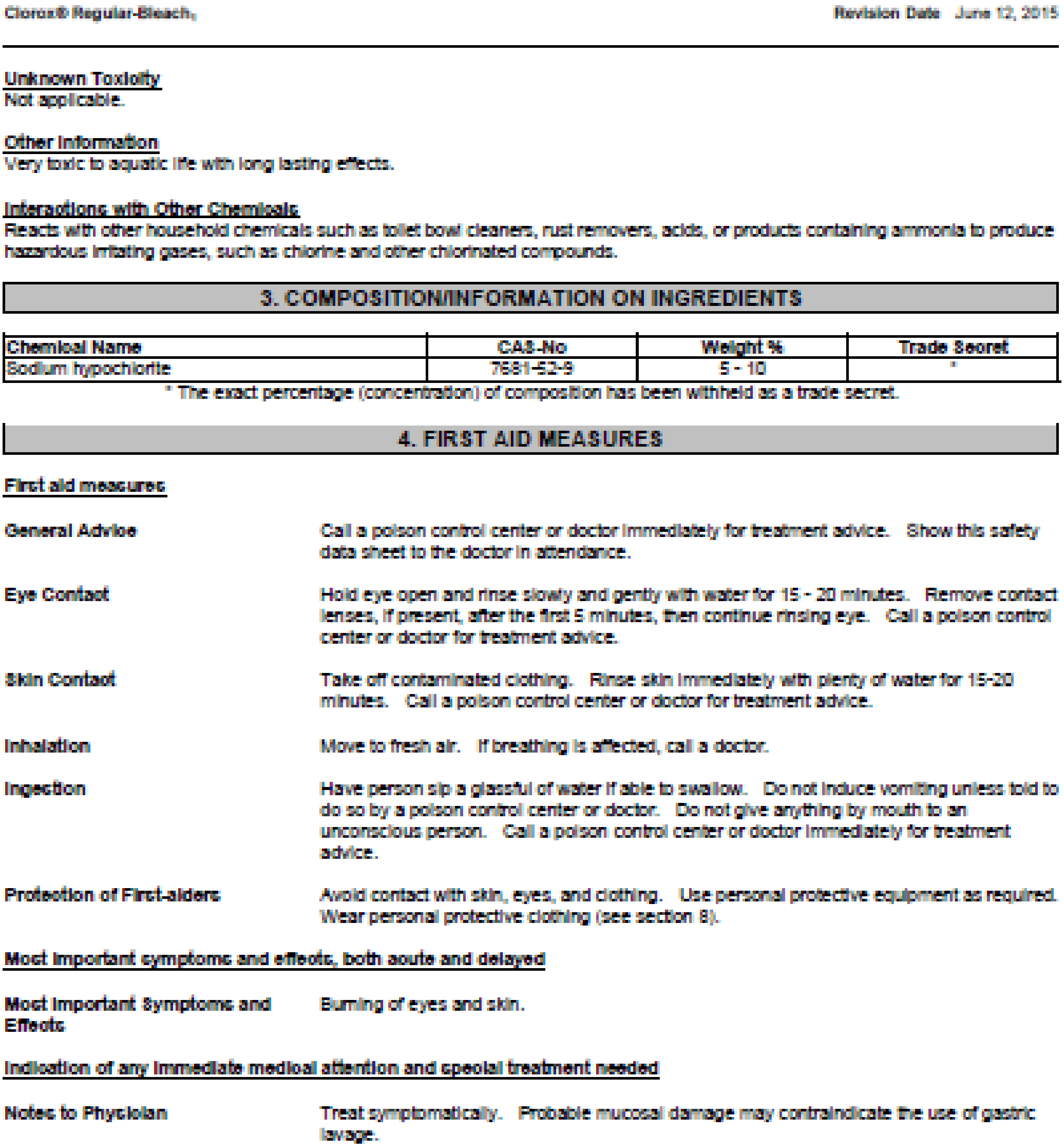
Precautionary Statements - Response
Immediately call a poison center or doctor.
If swallowed: Rinse mouth. Do NOT induce vomiting.
If on skin (or hair): Take off immediately all contaminated clothing. Rinse skin with water.
Wash contaminated clothing before reuse.
If inhaled: Remove person to fresh air and keep comfortable for breathing.
Specific treatment (see supplemental first aid instructions on this label).
If in eyes: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

Precautionary Statements - Storage
Store locked up.

Precautionary Statements - Disposal
Dispose of contents in accordance with all applicable federal, state, and local regulations.

Hazards not otherwise classified (HNOC)
Although not expected, heart conditions or chronic respiratory problems such as asthma, chronic bronchitis, or obstructive lung disease may be aggravated by exposure to high concentrations of vapor or mist.

Product contains a strong oxidizer. Always flush drains before and after use.



Clorox® Regular-Bleach₁ Revision Date June 12, 2015

Unknown Toxicity

Not applicable.

Other Information

Very toxic to aquatic life with long lasting effects.

Interactions with Other Chemicals

Reacts with other household chemicals such as toilet bowl cleaners, rust removers, acids, or products containing ammonia to produce hazardous irritating gases, such as chlorine and other chlorinated compounds.

3. COMPOSITION/INFORMATION ON INGREDIENTS

Chemical Name	CAS-No	Weight %	Trade Secret
Sodium hypochlorite	7681-52-9	5 - 10	*

* The exact percentage (concentration) of composition has been withheld as a trade secret.

4. FIRST AID MEASURES

First aid measures

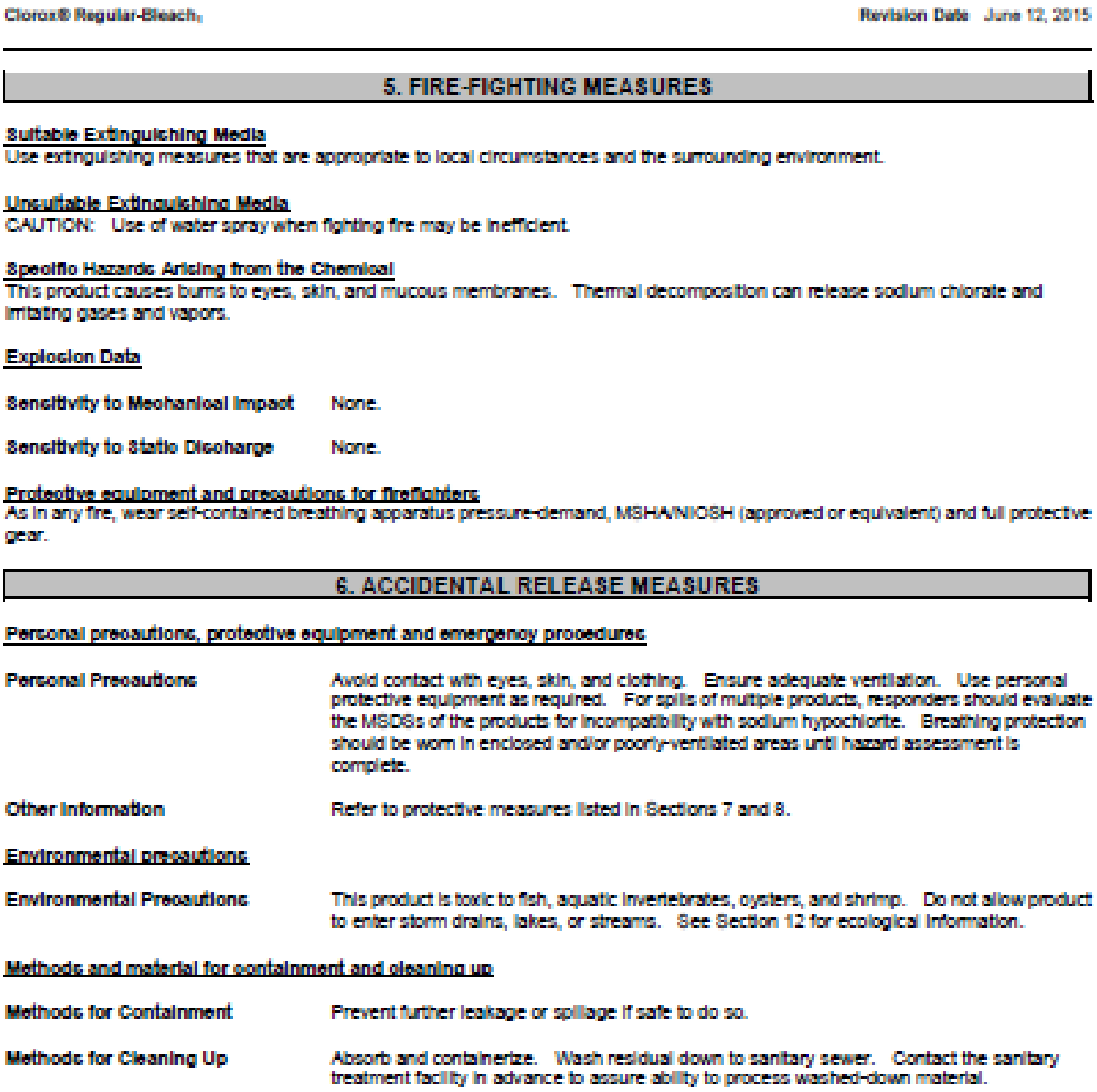
General Advice	Call a poison control center or doctor immediately for treatment advice. Show this safety data sheet to the doctor in attendance.
Eye Contact	Hold eye open and rinse slowly and gently with water for 15 - 20 minutes. Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye. Call a poison control center or doctor for treatment advice.
Skin Contact	Take off contaminated clothing. Rinse skin immediately with plenty of water for 15-20 minutes. Call a poison control center or doctor for treatment advice.
Inhalation	Move to fresh air. If breathing is affected, call a doctor.
Ingestion	Have person sip a glassful of water if able to swallow. Do not induce vomiting unless told to do so by a poison control center or doctor. Do not give anything by mouth to an unconscious person. Call a poison control center or doctor immediately for treatment advice.
Protection of First-aiders	Avoid contact with skin, eyes, and clothing. Use personal protective equipment as required. Wear personal protective clothing (see section 8).

Most important symptoms and effects, both acute and delayed

Most Important Symptoms and Effects	Burning of eyes and skin.

Indication of any immediate medical attention and special treatment needed

Notes to Physician	Treat symptomatically. Probable mucosal damage may contraindicate the use of gastric lavage.



Clorox® Regular-Bleach$_1$ Revision Date June 12, 2015

5. FIRE-FIGHTING MEASURES

Suitable Extinguishing Media
Use extinguishing measures that are appropriate to local circumstances and the surrounding environment.

Unsuitable Extinguishing Media
CAUTION: Use of water spray when fighting fire may be inefficient.

Specific Hazards Arising from the Chemical
This product causes burns to eyes, skin, and mucous membranes. Thermal decomposition can release sodium chlorate and irritating gases and vapors.

Explosion Data

Sensitivity to Mechanical Impact	None.
Sensitivity to Static Discharge	None.

Protective equipment and precautions for firefighters
As in any fire, wear self-contained breathing apparatus pressure-demand, MSHA/NIOSH (approved or equivalent) and full protective gear.

6. ACCIDENTAL RELEASE MEASURES

Personal precautions, protective equipment and emergency procedures

Personal Precautions	Avoid contact with eyes, skin, and clothing. Ensure adequate ventilation. Use personal protective equipment as required. For spills of multiple products, responders should evaluate the MSDSs of the products for incompatibility with sodium hypochlorite. Breathing protection should be worn in enclosed and/or poorly-ventilated areas until hazard assessment is complete.
Other Information	Refer to protective measures listed in Sections 7 and 8.

Environmental precautions

Environmental Precautions	This product is toxic to fish, aquatic invertebrates, oysters, and shrimp. Do not allow product to enter storm drains, lakes, or streams. See Section 12 for ecological information.

Methods and material for containment and cleaning up

Methods for Containment	Prevent further leakage or spillage if safe to do so.
Methods for Cleaning Up	Absorb and containerize. Wash residual down to sanitary sewer. Contact the sanitary treatment facility in advance to assure ability to process washed-down material.

7. HANDLING AND STORAGE

Precautions for safe handling

Handling Handle in accordance with good industrial hygiene and safety practice. Avoid contact with skin, eyes, and clothing. Do not eat, drink, or smoke when using this product.

Conditions for safe storage, including any incompatibilities

Storage Store away from children. Reclose cap tightly after each use. Store this product upright in a cool, dry area, away from direct sunlight and heat to avoid deterioration. Do not contaminate food or feed by storage of this product.

Incompatible Products Toilet bowl cleaners, rust removers, acids, and products containing ammonia.

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Control parameters

Exposure Guidelines

Chemical Name	ACGIH TLV	OSHA PEL	NIOSH IDLH
Sodium hypochlorite 7681-52-9	None	None	None

ACGIH TLV: American Conference of Governmental Industrial Hygienists - Threshold Limit Value. OSHA PEL: Occupational Safety and Health Administration - Permissible Exposure Limits. NIOSH IDLH: Immediately Dangerous to Life or Health.

Appropriate engineering controls

Engineering Measures
Showers
Eyewash stations
Ventilation systems

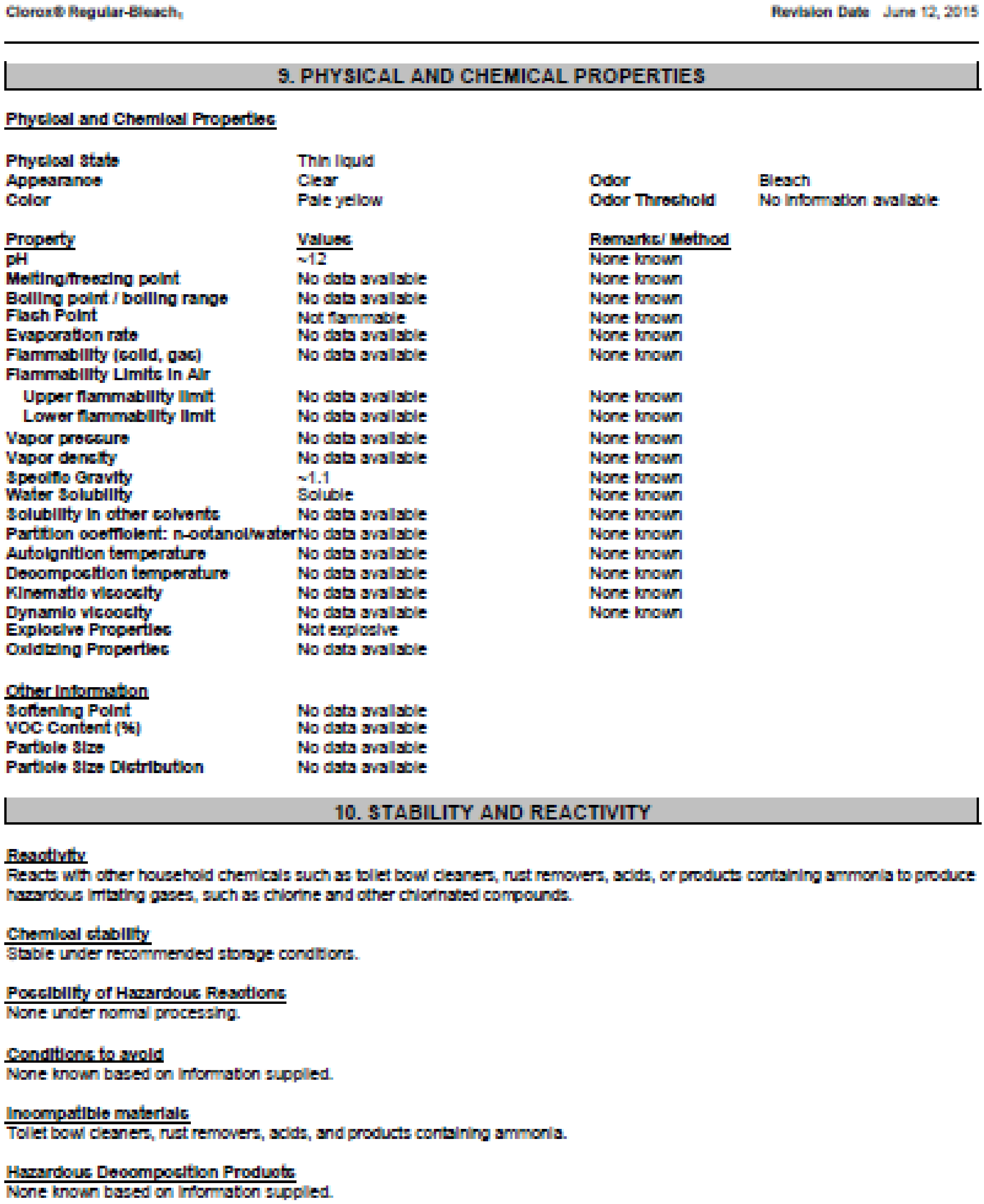
Individual protection measures, such as personal protective equipment

Eye/Face Protection If splashes are likely to occur: Wear safety glasses with side shields (or goggles) or face shield.

Skin and Body Protection Wear rubber or neoprene gloves and protective clothing such as long-sleeved shirt.

Respiratory Protection If irritation is experienced, NIOSH/MSHA approved respiratory protection should be worn. Positive-pressure supplied air respirators may be required for high airborne contaminant concentrations. Respiratory protection must be provided in accordance with current local regulations.

Hygiene Measures Handle in accordance with good industrial hygiene and safety practice. Wash hands after direct contact. Do not wear product-contaminated clothing for prolonged periods. Remove and wash contaminated clothing before re-use. Do not eat, drink, or smoke when using this product.



Clorox® Regular-Bleach1 **Revision Date** June 12, 2015

9. PHYSICAL AND CHEMICAL PROPERTIES

Physical and Chemical Properties

Physical State	Thin liquid		
Appearance	Clear	**Odor**	Bleach
Color	Pale yellow	**Odor Threshold**	No information available

Property	**Values**	**Remarks/ Method**
pH	~12	None known
Melting/freezing point	No data available	None known
Boiling point / boiling range	No data available	None known
Flash Point	Not flammable	None known
Evaporation rate	No data available	None known
Flammability (solid, gas)	No data available	None known
Flammability Limits in Air		
Upper flammability limit	No data available	None known
Lower flammability limit	No data available	None known
Vapor pressure	No data available	None known
Vapor density	No data available	None known
Specific Gravity	~1.1	None known
Water Solubility	Soluble	None known
Solubility in other solvents	No data available	None known
Partition coefficient: n-octanol/water	No data available	None known
Autoignition temperature	No data available	None known
Decomposition temperature	No data available	None known
Kinematic viscosity	No data available	None known
Dynamic viscosity	No data available	None known
Explosive Properties	Not explosive	
Oxidizing Properties	No data available	

Other Information

Softening Point	No data available
VOC Content (%)	No data available
Particle Size	No data available
Particle Size Distribution	No data available

10. STABILITY AND REACTIVITY

Reactivity

Reacts with other household chemicals such as toilet bowl cleaners, rust removers, acids, or products containing ammonia to produce hazardous irritating gases, such as chlorine and other chlorinated compounds.

Chemical stability

Stable under recommended storage conditions.

Possibility of Hazardous Reactions

None under normal processing.

Conditions to avoid

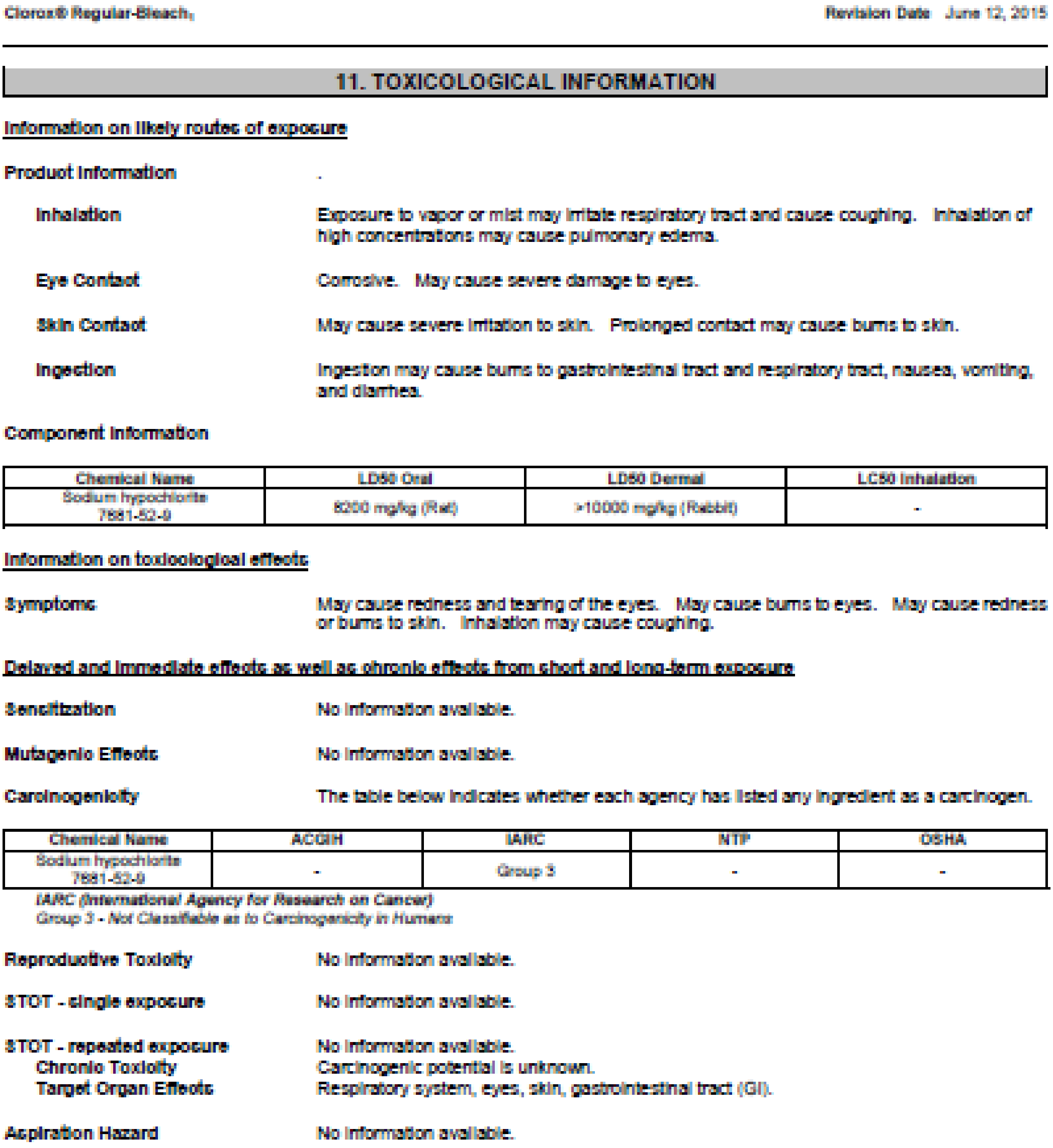
None known based on information supplied.

Incompatible materials

Toilet bowl cleaners, rust removers, acids, and products containing ammonia.

Hazardous Decomposition Products

None known based on information supplied.



Clorox® Regular-Bleach$_1$ Revision Date June 12, 2015

11. TOXICOLOGICAL INFORMATION

Information on likely routes of exposure

Product Information	.
Inhalation	Exposure to vapor or mist may irritate respiratory tract and cause coughing. Inhalation of high concentrations may cause pulmonary edema.
Eye Contact	Corrosive. May cause severe damage to eyes.
Skin Contact	May cause severe irritation to skin. Prolonged contact may cause burns to skin.
Ingestion	Ingestion may cause burns to gastrointestinal tract and respiratory tract, nausea, vomiting, and diarrhea.

Component Information

Chemical Name	LD50 Oral	LD50 Dermal	LC50 Inhalation
Sodium hypochlorite 7681-52-9	8200 mg/kg (Rat)	>10000 mg/kg (Rabbit)	-

Information on toxicological effects

Symptoms	May cause redness and tearing of the eyes. May cause burns to eyes. May cause redness or burns to skin. Inhalation may cause coughing.

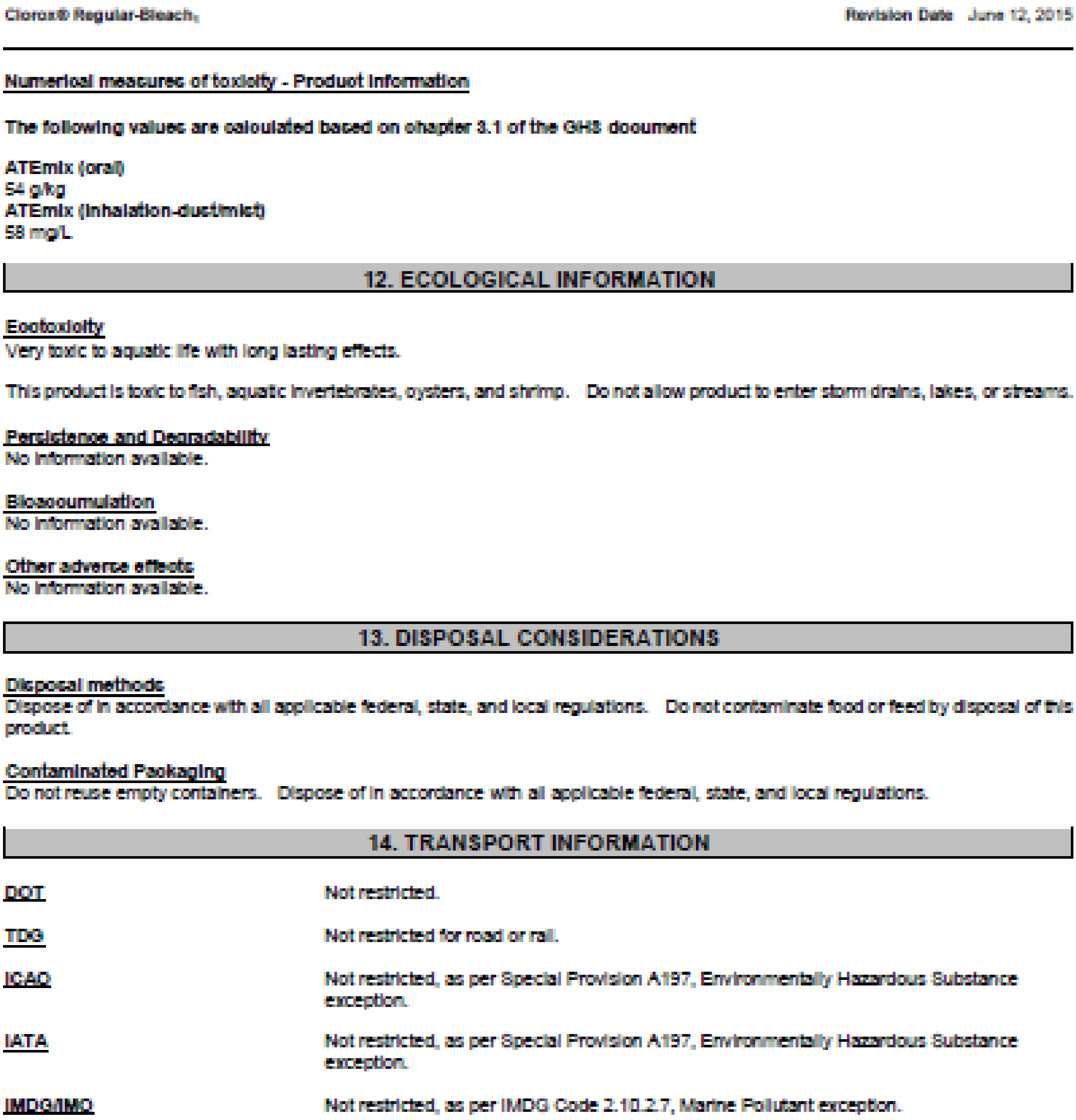
Delayed and immediate effects as well as chronic effects from short and long-term exposure

Sensitization	No information available.
Mutagenic Effects	No information available.
Carcinogenicity	The table below indicates whether each agency has listed any ingredient as a carcinogen.

Chemical Name	ACGIH	IARC	NTP	OSHA
Sodium hypochlorite 7681-52-9	-	Group 3	-	-

IARC (International Agency for Research on Cancer)
Group 3 - Not Classifiable as to Carcinogenicity in Humans

Reproductive Toxicity	No information available.
STOT - single exposure	No information available.
STOT - repeated exposure	No information available.
Chronic Toxicity	Carcinogenic potential is unknown.
Target Organ Effects	Respiratory system, eyes, skin, gastrointestinal tract (GI).
Aspiration Hazard	No information available.



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Numerical measures of toxicity - Product Information

The following values are calculated based on chapter 3.1 of the GHS document

ATEmix (oral)
54 g/kg
ATEmix (inhalation-dust/mist)
58 mg/L

12. ECOLOGICAL INFORMATION

Ecotoxicity
Very toxic to aquatic life with long lasting effects.

This product is toxic to fish, aquatic invertebrates, oysters, and shrimp. Do not allow product to enter storm drains, lakes, or streams.

Persistence and Degradability
No information available.

Bioaccumulation
No information available.

Other adverse effects
No information available.

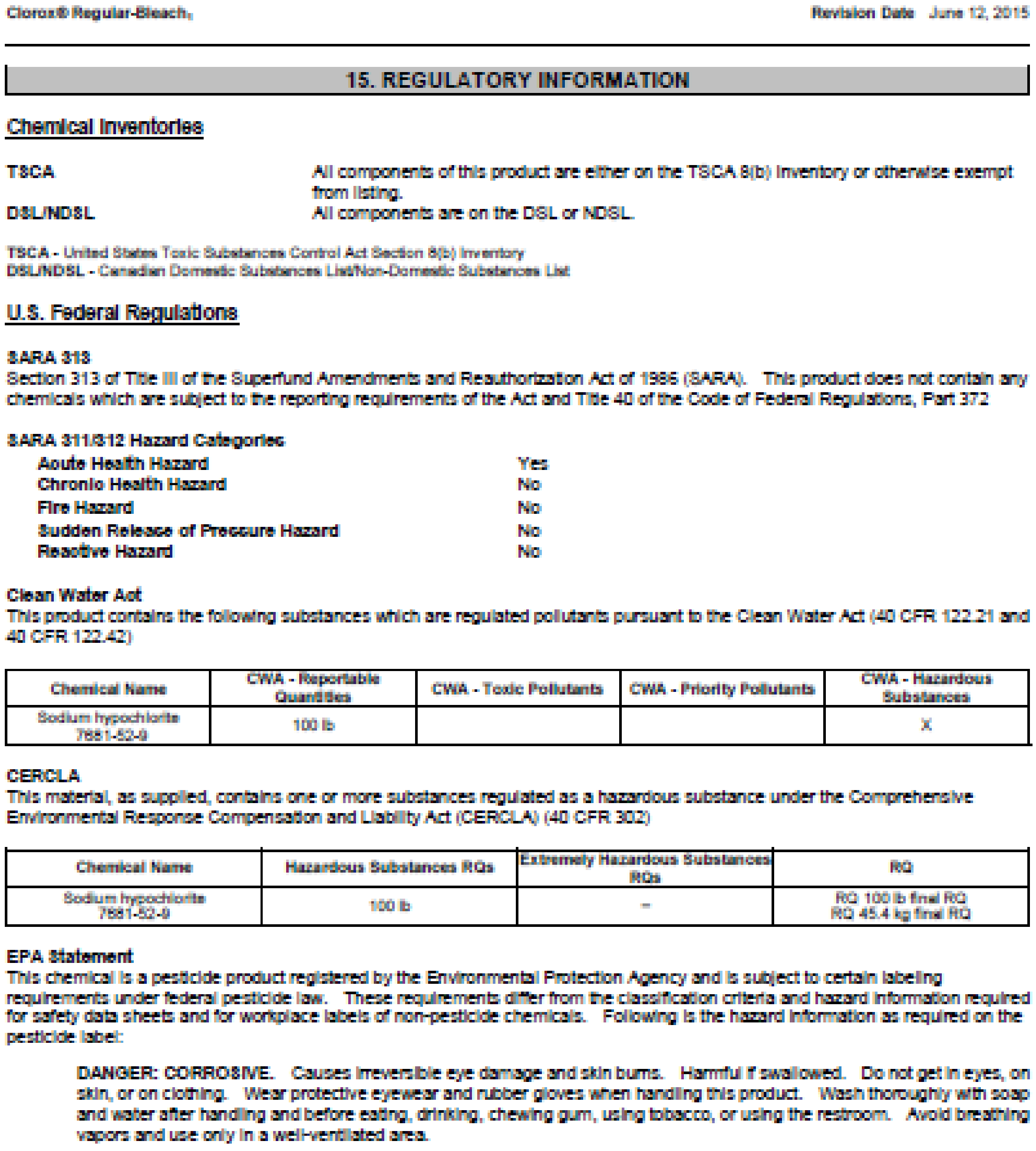
13. DISPOSAL CONSIDERATIONS

Disposal methods
Dispose of in accordance with all applicable federal, state, and local regulations. Do not contaminate food or feed by disposal of this product.

Contaminated Packaging
Do not reuse empty containers. Dispose of in accordance with all applicable federal, state, and local regulations.

14. TRANSPORT INFORMATION

DOT	Not restricted.
TDG	Not restricted for road or rail.
ICAO	Not restricted, as per Special Provision A197, Environmentally Hazardous Substance exception.
IATA	Not restricted, as per Special Provision A197, Environmentally Hazardous Substance exception.
IMDG/IMO	Not restricted, as per IMDG Code 2.10.2.7, Marine Pollutant exception.



15. REGULATORY INFORMATION

Chemical Inventories

TSCA	All components of this product are either on the TSCA 8(b) Inventory or otherwise exempt from listing.
DSL/NDSL	All components are on the DSL or NDSL.

TSCA - United States Toxic Substances Control Act Section 8(b) Inventory
DSL/NDSL - Canadian Domestic Substances List/Non-Domestic Substances List

U.S. Federal Regulations

SARA 313
Section 313 of Title III of the Superfund Amendments and Reauthorization Act of 1986 (SARA). This product does not contain any chemicals which are subject to the reporting requirements of the Act and Title 40 of the Code of Federal Regulations, Part 372

SARA 311/312 Hazard Categories

Acute Health Hazard	Yes
Chronic Health Hazard	No
Fire Hazard	No
Sudden Release of Pressure Hazard	No
Reactive Hazard	No

Clean Water Act
This product contains the following substances which are regulated pollutants pursuant to the Clean Water Act (40 CFR 122.21 and 40 CFR 122.42)

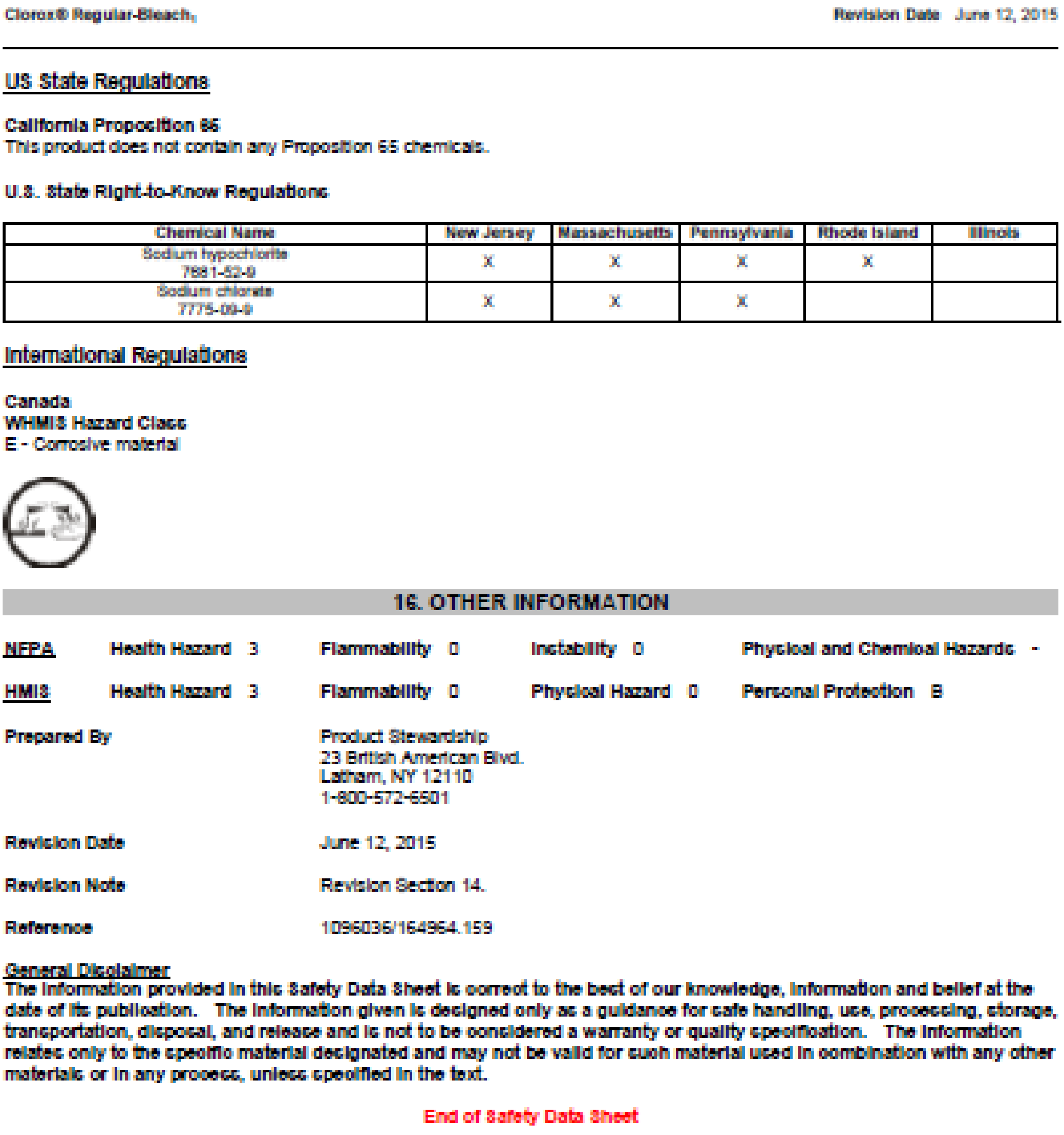
Chemical Name	CWA - Reportable Quantities	CWA - Toxic Pollutants	CWA - Priority Pollutants	CWA - Hazardous Substances
Sodium hypochlorite 7681-52-9	100 lb			X

CERCLA
This material, as supplied, contains one or more substances regulated as a hazardous substance under the Comprehensive Environmental Response Compensation and Liability Act (CERCLA) (40 CFR 302)

Chemical Name	Hazardous Substances RQs	Extremely Hazardous Substances RQs	RQ
Sodium hypochlorite 7681-52-9	100 lb	-	RQ 100 lb final RQ RQ 45.4 kg final RQ

EPA Statement
This chemical is a pesticide product registered by the Environmental Protection Agency and is subject to certain labeling requirements under federal pesticide law. These requirements differ from the classification criteria and hazard information required for safety data sheets and for workplace labels of non-pesticide chemicals. Following is the hazard information as required on the pesticide label:

> **DANGER: CORROSIVE.** Causes irreversible eye damage and skin burns. Harmful if swallowed. Do not get in eyes, on skin, or on clothing. Wear protective eyewear and rubber gloves when handling this product. Wash thoroughly with soap and water after handling and before eating, drinking, chewing gum, using tobacco, or using the restroom. Avoid breathing vapors and use only in a well-ventilated area.



Clorox® Regular-Bleach$_1$ **Revision Date** June 12, 2015

US State Regulations

California Proposition 65
This product does not contain any Proposition 65 chemicals.

U.S. State Right-to-Know Regulations

Chemical Name	New Jersey	Massachusetts	Pennsylvania	Rhode Island	Illinois
Sodium hypochlorite 7681-52-9	X	X	X	X	
Sodium chlorate 7775-09-9	X	X	X		

International Regulations

Canada
WHMIS Hazard Class
E - Corrosive material

16. OTHER INFORMATION

NFPA	**Health Hazard** 3	**Flammability** 0	**Instability** 0	**Physical and Chemical Hazards** -
HMIS	**Health Hazard** 3	**Flammability** 0	**Physical Hazard** 0	**Personal Protection** B

Prepared By Product Stewardship
23 British American Blvd.
Latham, NY 12110
1-800-572-6501

Revision Date June 12, 2015

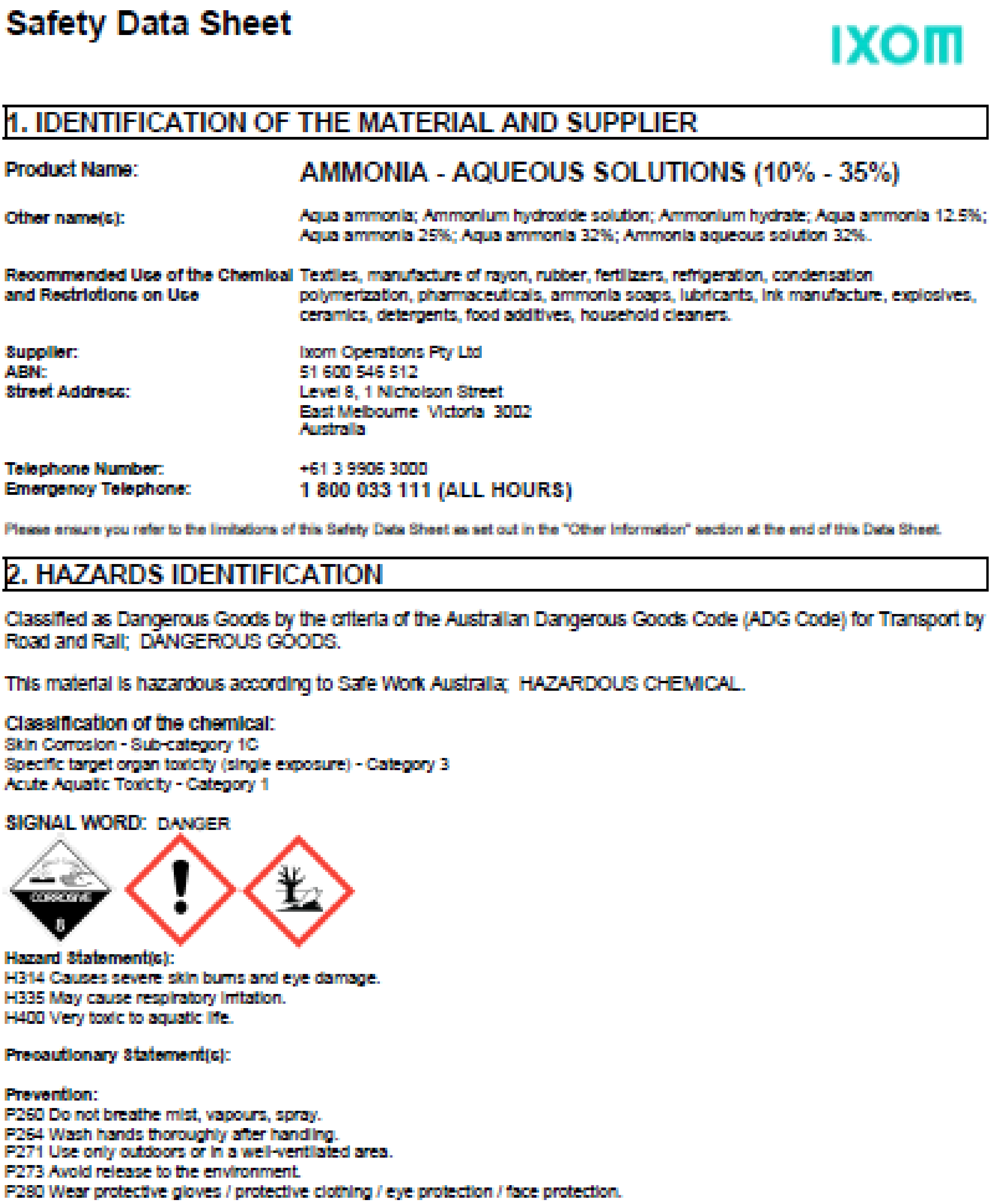
Revision Note Revision Section 14.

Reference 1096036/164964.159

General Disclaimer
The information provided in this Safety Data Sheet is correct to the best of our knowledge, information and belief at the date of its publication. The information given is designed only as a guidance for safe handling, use, processing, storage, transportation, disposal, and release and is not to be considered a warranty or quality specification. The information relates only to the specific material designated and may not be valid for such material used in combination with any other materials or in any process, unless specified in the text.

End of Safety Data Sheet

A3.11. Ammonia



Safety Data Sheet

IXOM

1. IDENTIFICATION OF THE MATERIAL AND SUPPLIER

Product Name:	AMMONIA - AQUEOUS SOLUTIONS (10% - 35%)
Other name(s):	Aqua ammonia; Ammonium hydroxide solution; Ammonium hydrate; Aqua ammonia 12.5%; Aqua ammonia 25%; Aqua ammonia 32%; Ammonia aqueous solution 32%.
Recommended Use of the Chemical and Restrictions on Use	Textiles, manufacture of rayon, rubber, fertilizers, refrigeration, condensation polymerization, pharmaceuticals, ammonia soaps, lubricants, ink manufacture, explosives, ceramics, detergents, food additives, household cleaners.
Supplier:	Ixom Operations Pty Ltd
ABN:	51 600 546 512
Street Address:	Level 8, 1 Nicholson Street East Melbourne Victoria 3002 Australia
Telephone Number:	+61 3 9906 3000
Emergency Telephone:	1 800 033 111 (ALL HOURS)

Please ensure you refer to the limitations of this Safety Data Sheet as set out in the "Other Information" section at the end of this Data Sheet.

2. HAZARDS IDENTIFICATION

Classified as Dangerous Goods by the criteria of the Australian Dangerous Goods Code (ADG Code) for Transport by Road and Rail; DANGEROUS GOODS.

This material is hazardous according to Safe Work Australia; HAZARDOUS CHEMICAL.

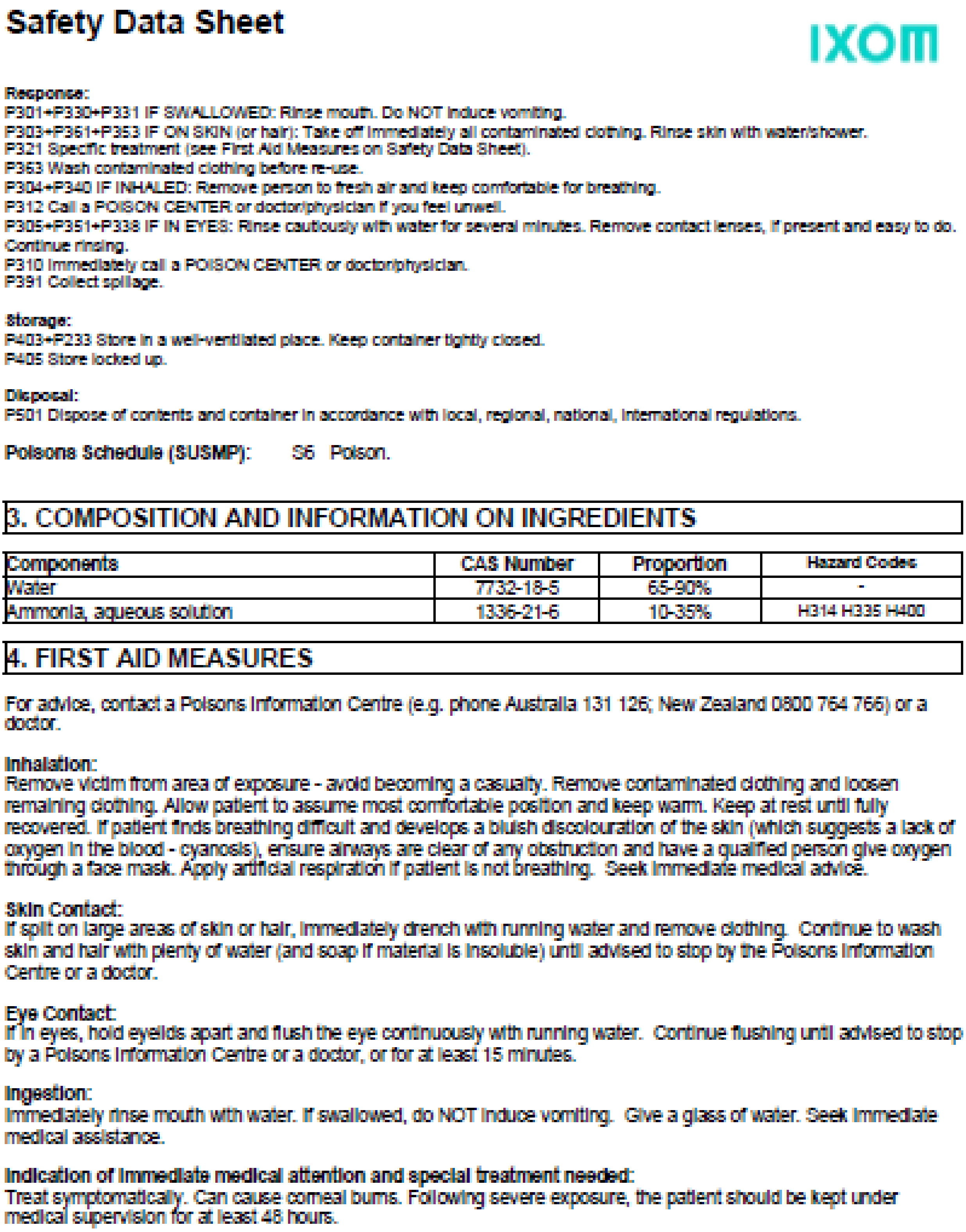
Classification of the chemical:
Skin Corrosion - Sub-category 1C
Specific target organ toxicity (single exposure) - Category 3
Acute Aquatic Toxicity - Category 1

SIGNAL WORD: DANGER

Hazard Statement(s):
H314 Causes severe skin burns and eye damage.
H335 May cause respiratory irritation.
H400 Very toxic to aquatic life.

Precautionary Statement(s):

Prevention:
P260 Do not breathe mist, vapours, spray.
P264 Wash hands thoroughly after handling.
P271 Use only outdoors or in a well-ventilated area.
P273 Avoid release to the environment.
P280 Wear protective gloves / protective clothing / eye protection / face protection.



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Response:
P301+P330+P331 IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.
P303+P361+P353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water/shower.
P321 Specific treatment (see First Aid Measures on Safety Data Sheet).
P363 Wash contaminated clothing before re-use.
P304+P340 IF INHALED: Remove person to fresh air and keep comfortable for breathing.
P312 Call a POISON CENTER or doctor/physician if you feel unwell.
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.
P391 Collect spillage.

Storage:
P403+P233 Store in a well-ventilated place. Keep container tightly closed.
P405 Store locked up.

Disposal:
P501 Dispose of contents and container in accordance with local, regional, national, international regulations.

Poisons Schedule (SUSMP): S6 Poison.

3. COMPOSITION AND INFORMATION ON INGREDIENTS

Components	CAS Number	Proportion	Hazard Codes
Water	7732-18-5	65-90%	-
Ammonia, aqueous solution	1336-21-6	10-35%	H314 H335 H400

4. FIRST AID MEASURES

For advice, contact a Poisons Information Centre (e.g. phone Australia 131 126; New Zealand 0800 764 766) or a doctor.

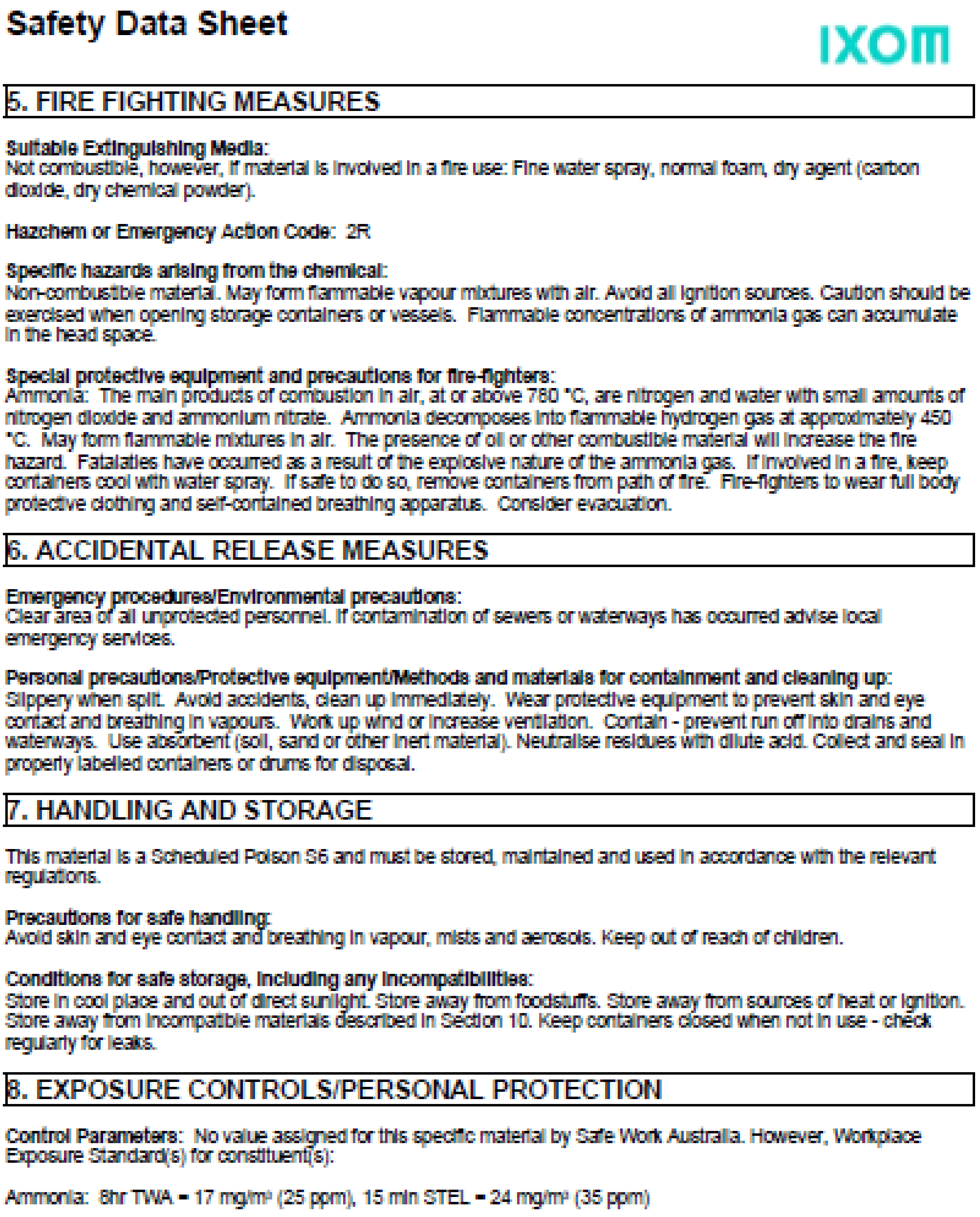
Inhalation:
Remove victim from area of exposure - avoid becoming a casualty. Remove contaminated clothing and loosen remaining clothing. Allow patient to assume most comfortable position and keep warm. Keep at rest until fully recovered. If patient finds breathing difficult and develops a bluish discolouration of the skin (which suggests a lack of oxygen in the blood - cyanosis), ensure airways are clear of any obstruction and have a qualified person give oxygen through a face mask. Apply artificial respiration if patient is not breathing. Seek immediate medical advice.

Skin Contact:
If spilt on large areas of skin or hair, immediately drench with running water and remove clothing. Continue to wash skin and hair with plenty of water (and soap if material is insoluble) until advised to stop by the Poisons Information Centre or a doctor.

Eye Contact:
If in eyes, hold eyelids apart and flush the eye continuously with running water. Continue flushing until advised to stop by a Poisons Information Centre or a doctor, or for at least 15 minutes.

Ingestion:
Immediately rinse mouth with water. If swallowed, do NOT induce vomiting. Give a glass of water. Seek immediate medical assistance.

Indication of immediate medical attention and special treatment needed:
Treat symptomatically. Can cause corneal burns. Following severe exposure, the patient should be kept under medical supervision for at least 48 hours.



Safety Data Sheet

IXOM

5. FIRE FIGHTING MEASURES

Suitable Extinguishing Media:
Not combustible, however, if material is involved in a fire use: Fine water spray, normal foam, dry agent (carbon dioxide, dry chemical powder).

Hazchem or Emergency Action Code: 2R

Specific hazards arising from the chemical:
Non-combustible material. May form flammable vapour mixtures with air. Avoid all ignition sources. Caution should be exercised when opening storage containers or vessels. Flammable concentrations of ammonia gas can accumulate in the head space.

Special protective equipment and precautions for fire-fighters:
Ammonia: The main products of combustion in air, at or above 780 °C, are nitrogen and water with small amounts of nitrogen dioxide and ammonium nitrate. Ammonia decomposes into flammable hydrogen gas at approximately 450 °C. May form flammable mixtures in air. The presence of oil or other combustible material will increase the fire hazard. Fatalaties have occurred as a result of the explosive nature of the ammonia gas. If involved in a fire, keep containers cool with water spray. If safe to do so, remove containers from path of fire. Fire-fighters to wear full body protective clothing and self-contained breathing apparatus. Consider evacuation.

6. ACCIDENTAL RELEASE MEASURES

Emergency procedures/Environmental precautions:
Clear area of all unprotected personnel. If contamination of sewers or waterways has occurred advise local emergency services.

Personal precautions/Protective equipment/Methods and materials for containment and cleaning up:
Slippery when spilt. Avoid accidents, clean up immediately. Wear protective equipment to prevent skin and eye contact and breathing in vapours. Work up wind or increase ventilation. Contain - prevent run off into drains and waterways. Use absorbent (soil, sand or other inert material). Neutralise residues with dilute acid. Collect and seal in properly labelled containers or drums for disposal.

7. HANDLING AND STORAGE

This material is a Scheduled Poison S6 and must be stored, maintained and used in accordance with the relevant regulations.

Precautions for safe handling:
Avoid skin and eye contact and breathing in vapour, mists and aerosols. Keep out of reach of children.

Conditions for safe storage, including any incompatibilities:
Store in cool place and out of direct sunlight. Store away from foodstuffs. Store away from sources of heat or ignition. Store away from incompatible materials described in Section 10. Keep containers closed when not in use - check regularly for leaks.

8. EXPOSURE CONTROLS/PERSONAL PROTECTION

Control Parameters: No value assigned for this specific material by Safe Work Australia. However, Workplace Exposure Standard(s) for constituent(s):

Ammonia: 8hr TWA = 17 mg/m^3 (25 ppm), 15 min STEL = 24 mg/m^3 (35 ppm)

Safety Data Sheet

As published by Safe Work Australia Workplace Exposure Standards for Airborne Contaminants.

TWA - The time-weighted average airborne concentration of a particular substance when calculated over an eight-hour working day, for a five-day working week.

STEL (Short Term Exposure Limit) - the airborne concentration of a particular substance calculated as a time-weighted average over 15 minutes, which should not be exceeded at any time during a normal eight hour work day. According to current knowledge this concentration should neither impair the health of, nor cause undue discomfort to, nearly all workers.

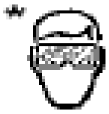
These Workplace Exposure Standards are guides to be used in the control of occupational health hazards. All atmospheric contamination should be kept to as low a level as is workable. These workplace exposure standards should not be used as fine dividing lines between safe and dangerous concentrations of chemicals. They are not a measure of relative toxicity.

Appropriate engineering controls:
Ensure ventilation is adequate and that air concentrations of components are controlled below quoted Workplace Exposure Standards. Keep containers closed when not in use.

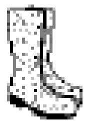
If in the handling and application of this material, safe exposure levels could be exceeded, the use of engineering controls such as local exhaust ventilation must be considered and the results documented. If achieving safe exposure levels does not require engineering controls, then a detailed and documented risk assessment using the relevant Personal Protective Equipment (PPE) (refer to PPE section below) as a basis must be carried out to determine the minimum PPE requirements.

Individual protection measures, such as Personal Protective Equipment (PPE):
The selection of PPE is dependent on a detailed risk assessment. The risk assessment should consider the work situation, the physical form of the chemical, the handling methods, and environmental factors.

OVERALLS, CHEMICAL GOGGLES, RUBBER BOOTS, AIR MASK , GLOVES (Long), APRON.
* Not required if wearing air supplied mask.






Wear overalls, chemical goggles, full face shield, elbow-length impervious gloves, splash apron or equivalent chemical impervious outer garment, and rubber boots. Use with adequate ventilation. If determined by a risk assessment an inhalation risk exists, wear an air-supplied mask meeting the requirements of AS/NZS 1715 and AS/NZS 1716. Always wash hands before smoking, eating, drinking or using the toilet. Wash contaminated clothing and other protective equipment before storage or re-use.

9. PHYSICAL AND CHEMICAL PROPERTIES

Physical state:	Liquid
Colour:	Colourless
Odour:	Sharp , Irritating
Odour Threshold:	0.6-53 ppm (detection); 0.7-55 ppm (recognition).
Solubility:	Miscible in water.
Specific Gravity:	0.88-0.92 @20°C
Relative Vapour Density (air=1):	0.6

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Vapour Pressure (20 °C):	6.9-10.5 psi
Flash Point (°C):	Not applicable
Flammability Limits (%):	16-25
Autoignition Temperature (°C):	Not applicable
% Volatile by Volume:	100
Boiling Point/Range (°C):	18-37
pH:	11.7 (1% aqueous solution)

10. STABILITY AND REACTIVITY

Reactivity:	Reacts violently with acids.
Chemical stability:	May form explosive compounds with mercury, halogens, and hypochlorites. Reacts exothermically with strong mineral acids .
Possibility of hazardous reactions:	Corrosive to copper , nickel , tin , zinc , and their alloys .
Conditions to avoid:	Avoid exposure to heat. Avoid exposure to light.
Incompatible materials:	Incompatible with peroxides , metal salts , acids , and reducing agents .
Hazardous decomposition products:	Hydrogen.

11. TOXICOLOGICAL INFORMATION

No adverse health effects expected if the product is handled in accordance with this Safety Data Sheet and the product label. Symptoms or effects that may arise if the product is mishandled and overexposure occurs are:

Ingestion:	Swallowing can result in nausea, vomiting, diarrhoea, abdominal pain and chemical burns to the gastrointestinal tract.
Eye contact:	A severe eye irritant. Corrosive to eyes; contact can cause corneal burns. Contamination of eyes can result in permanent injury.
Skin contact:	Contact with skin will result in severe irritation. Corrosive to skin - may cause skin burns.
Inhalation:	Breathing in mists or aerosols will produce respiratory irritation. Inhalation of high concentrations may result in shortness of breath, chest pain, severe headache and lung damage including pulmonary oedema. Effects may be delayed.

Acute toxicity:
Oral LD50 (rat): 350 mg/kg (1)

Chronic effects: Chronic exposure to ammonia may cause chemical pneumonitis and kidney damage.

12. ECOLOGICAL INFORMATION

Ecotoxicity	Avoid contaminating waterways.
Aquatic toxicity:	Toxic to aquatic organisms.
96hr LC50 (rainbow trout):	0.53 mg/L (for ammonia) (2)

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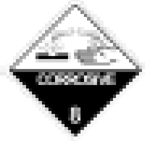
IXOM

13. DISPOSAL CONSIDERATIONS

Disposal methods:
Refer to Waste Management Authority. Dispose of contents and container in accordance with local, regional, national, international regulations.

14. TRANSPORT INFORMATION

Road and Rail Transport
Classified as Dangerous Goods by the criteria of the Australian Dangerous Goods Code (ADG Code) for Transport by Road and Rail; DANGEROUS GOODS.



UN No:	2672
Transport Hazard Class:	8 Corrosive
Packing Group:	III
Proper Shipping Name or Technical Name:	AMMONIA SOLUTION
Hazchem or Emergency Action Code:	2R

Marine Transport
Classified as Dangerous Goods by the criteria of the International Maritime Dangerous Goods Code (IMDG Code) for transport by sea; DANGEROUS GOODS.

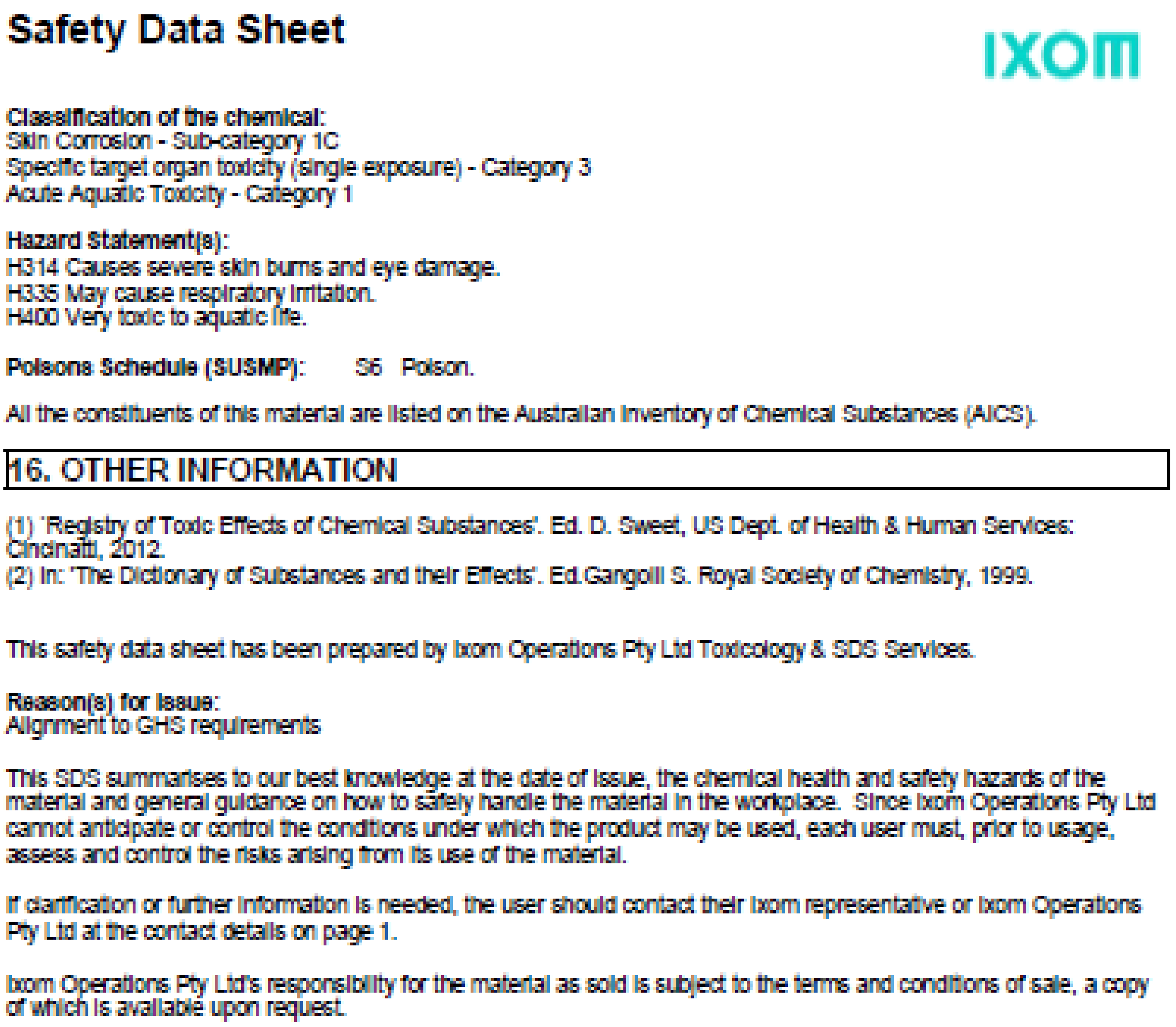
UN No:	2672
Transport Hazard Class:	8 Corrosive
Packing Group:	III
Proper Shipping Name or Technical Name:	AMMONIA SOLUTION
IMDG EMS Fire:	F-A
IMDG EMS Spill:	S-B

Air Transport
Classified as Dangerous Goods by the criteria of the International Air Transport Association (IATA) Dangerous Goods Regulations for transport by air; DANGEROUS GOODS.

UN No:	2672
Transport Hazard Class:	8 Corrosive
Packing Group:	III
Proper Shipping Name or Technical Name:	AMMONIA SOLUTION

15. REGULATORY INFORMATION

Classification:
This material is hazardous according to Safe Work Australia; HAZARDOUS CHEMICAL.



Safety Data Sheet

Classification of the chemical:
Skin Corrosion - Sub-category 1C
Specific target organ toxicity (single exposure) - Category 3
Acute Aquatic Toxicity - Category 1

Hazard Statement(s):
H314 Causes severe skin burns and eye damage.
H335 May cause respiratory irritation.
H400 Very toxic to aquatic life.

Poisons Schedule (SUSMP): S6 Poison.

All the constituents of this material are listed on the Australian Inventory of Chemical Substances (AICS).

16. OTHER INFORMATION

(1) 'Registry of Toxic Effects of Chemical Substances'. Ed. D. Sweet, US Dept. of Health & Human Services: Cincinatti, 2012.
(2) In: 'The Dictionary of Substances and their Effects'. Ed.Gangolli S. Royal Society of Chemistry, 1999.

This safety data sheet has been prepared by Ixom Operations Pty Ltd Toxicology & SDS Services.

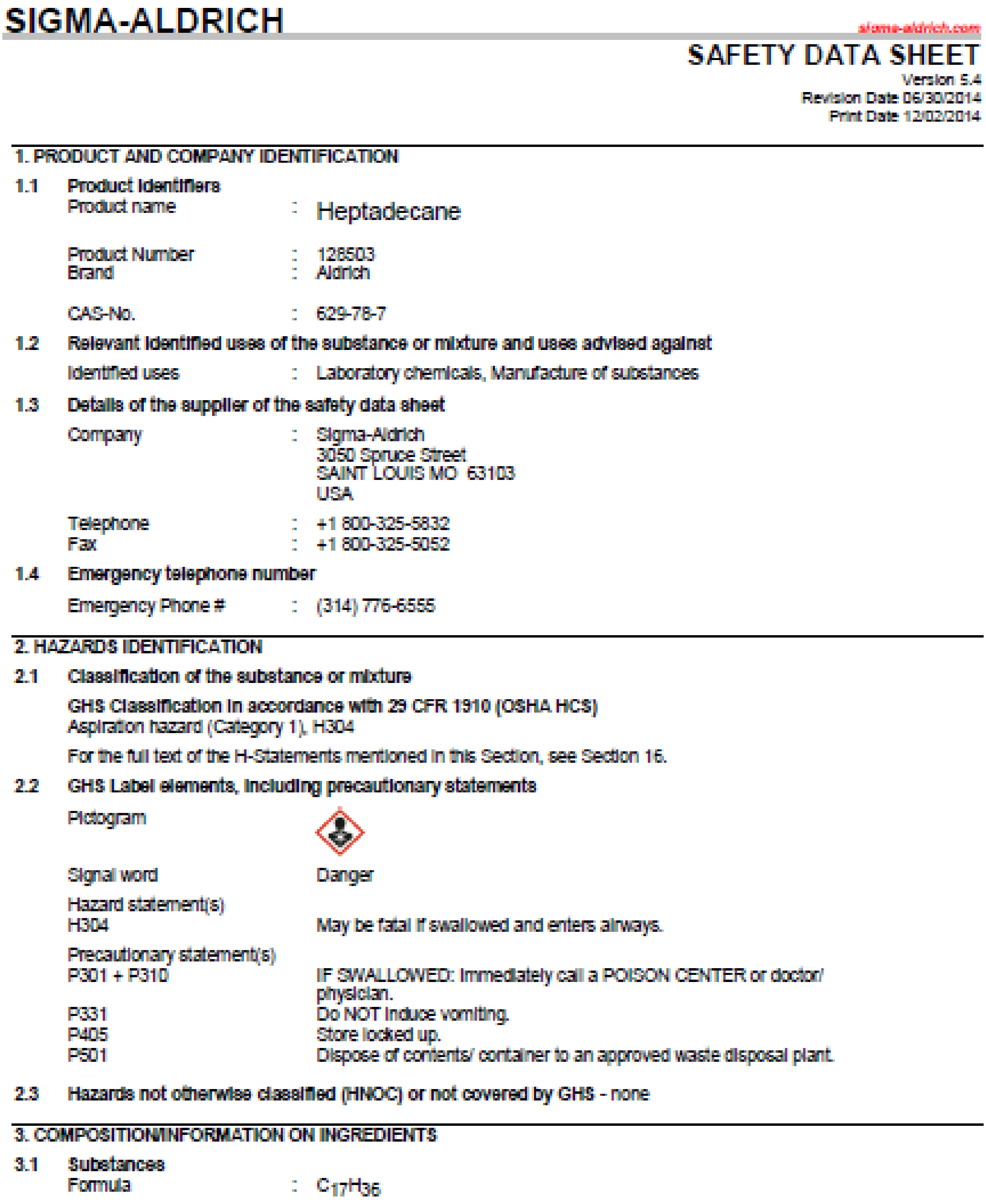
Reason(s) for Issue:
Alignment to GHS requirements

This SDS summarises to our best knowledge at the date of issue, the chemical health and safety hazards of the material and general guidance on how to safely handle the material in the workplace. Since Ixom Operations Pty Ltd cannot anticipate or control the conditions under which the product may be used, each user must, prior to usage, assess and control the risks arising from its use of the material.

If clarification or further information is needed, the user should contact their Ixom representative or Ixom Operations Pty Ltd at the contact details on page 1.

Ixom Operations Pty Ltd's responsibility for the material as sold is subject to the terms and conditions of sale, a copy of which is available upon request.

A3.12. Heptadecane



SIGMA-ALDRICH

sigma-aldrich.com

SAFETY DATA SHEET

Version 5.4
Revision Date 06/30/2014
Print Date 12/02/2014

1. PRODUCT AND COMPANY IDENTIFICATION

1.1 Product identifiers

Product name : Heptadecane

Product Number : 128503
Brand : Aldrich

CAS-No. : 629-78-7

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)
Aspiration hazard (Category 1), H304

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

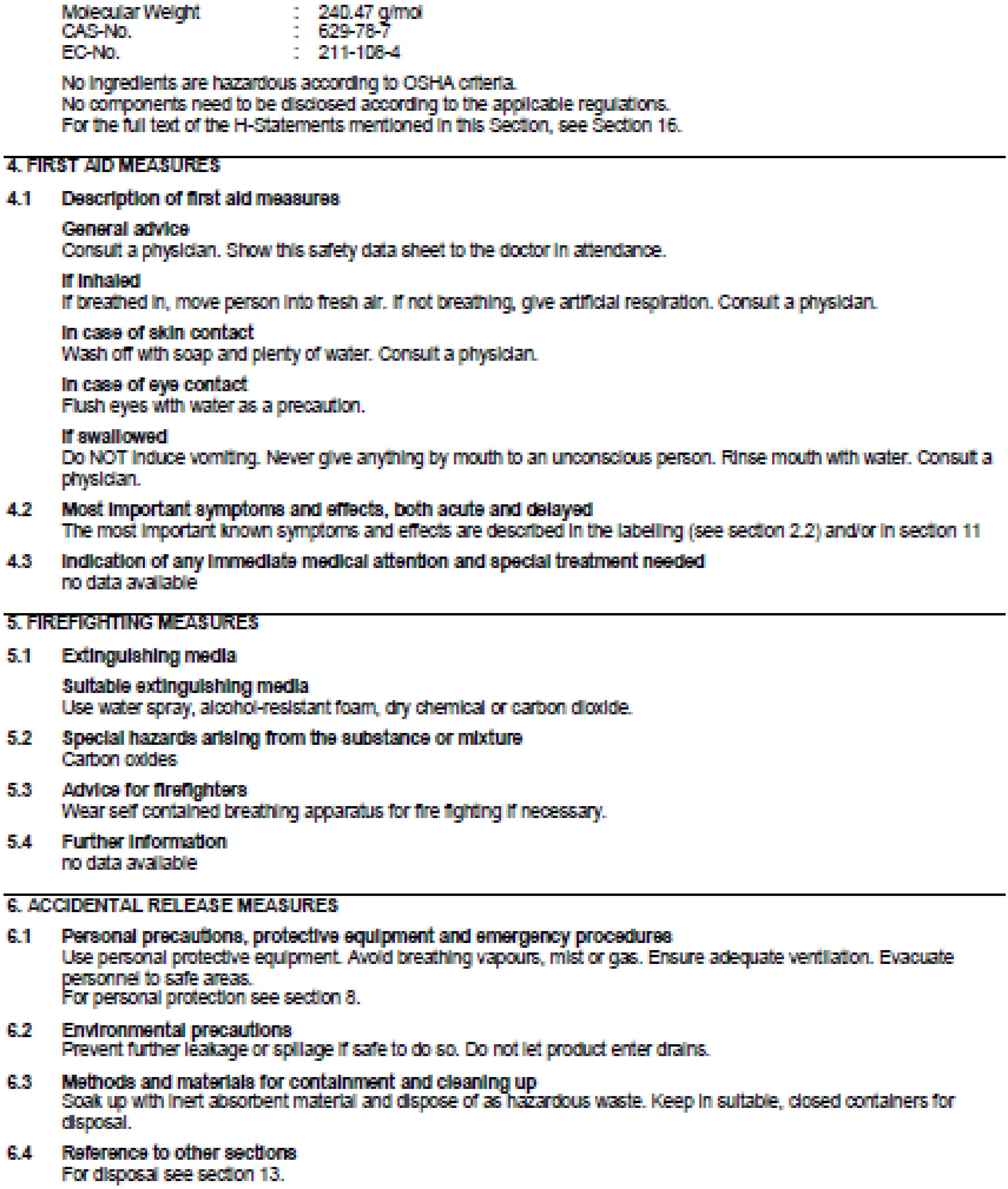
Hazard statement(s)
H304 May be fatal if swallowed and enters airways.

Precautionary statement(s)
P301 + P310 IF SWALLOWED: Immediately call a POISON CENTER or doctor/ physician.
P331 Do NOT induce vomiting.
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
Formula : $C_{17}H_{36}$



Molecular Weight : 240.47 g/mol
CAS-No. : 629-78-7
EC-No. : 211-108-4

No ingredients are hazardous according to OSHA criteria.
No components need to be disclosed according to the applicable regulations.
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.

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
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

5.3 Advice for firefighters

Wear self contained breathing apparatus for fire fighting 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 breathing vapours, mist or gas. Ensure adequate ventilation. Evacuate personnel to safe areas.
For personal protection see section 8.

6.2 Environmental precautions

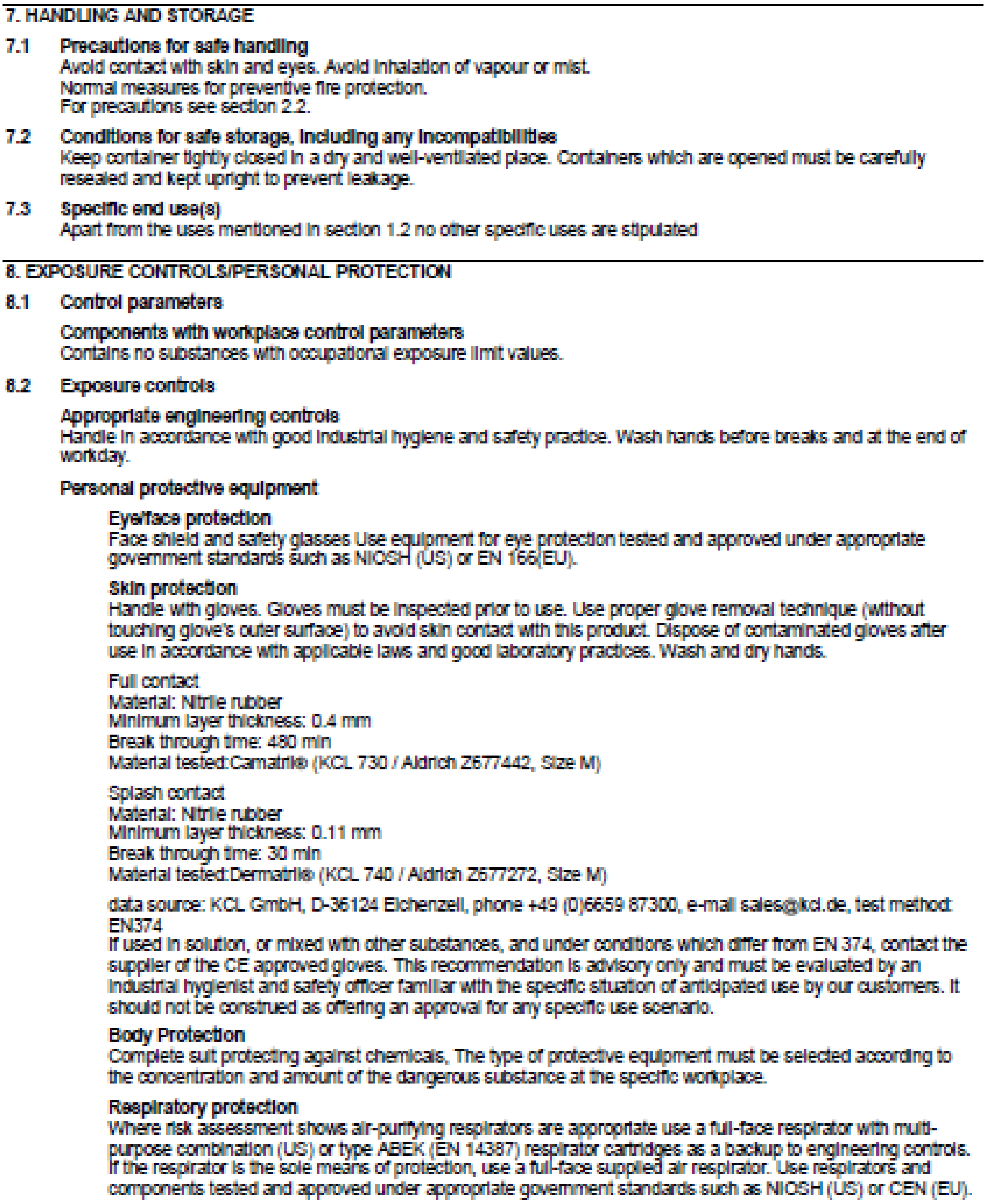
Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

6.3 Methods and materials for containment and cleaning up

Soak up with inert absorbent material and dispose of as hazardous waste. 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 inhalation of vapour or mist.
Normal measures for preventive fire protection.
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. Containers which are opened must be carefully resealed and kept upright to prevent leakage.

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.

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: 0.4 mm
Break through time: 480 min
Material tested:Camatril® (KCL 730 / Aldrich Z677442, Size M)

Splash contact
Material: Nitrile rubber
Minimum layer thickness: 0.11 mm
Break through time: 30 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 respirator with multi-purpose combination (US) or type ABEK (EN 14387) 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: liquid Colour: colourless
b)	Odour	no data available
c)	Odour Threshold	no data available
d)	pH	no data available
e)	Melting point/freezing point	Melting point/range: 20 - 22 °C (68 - 72 °F) - lit.
f)	Initial boiling point and boiling range	302 °C (576 °F) - lit.
g)	Flash point	149 °C (300 °F) - closed cup
h)	Evapouration rate	no data available
i)	Flammability (solid, gas)	no data available
j)	Upper/lower flammability or explosive limits	no data available
k)	Vapour pressure	1 hPa (1 mmHg) at 115 °C (239 °F)
l)	Vapour density	8.3 - (Air = 1.0)
m)	Relative density	0.777 g/mL at 25 °C (77 °F)
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

Relative vapour density 8.3 - (Air = 1.0)

10. STABILITY AND REACTIVITY

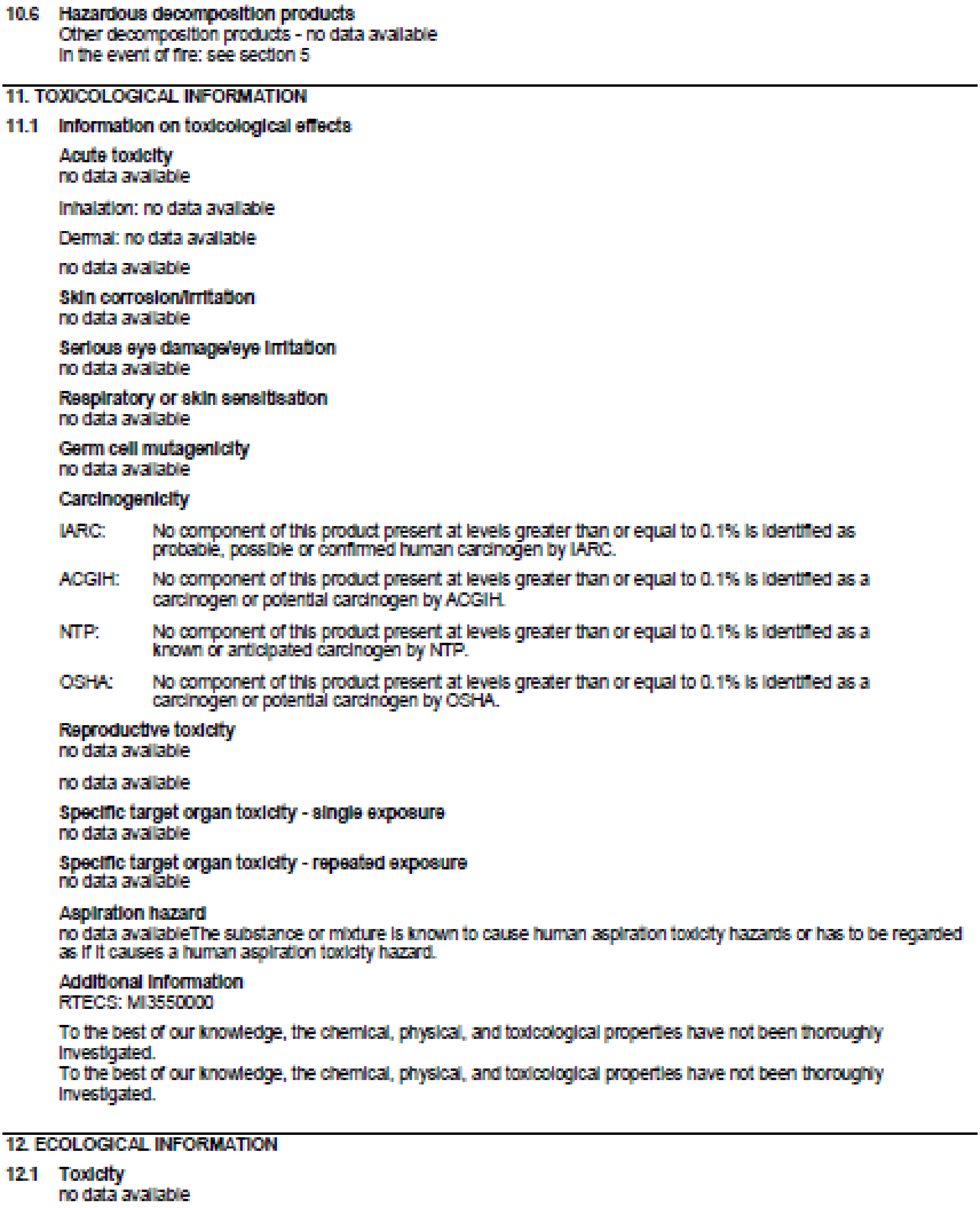
10.1 Reactivity
no data available

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
Strong oxidizing agentsStrong 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:	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
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 availableThe substance or mixture is known to cause human aspiration toxicity hazards or has to be regarded as if it causes a human aspiration toxicity hazard.

Additional Information
RTECS: MI3550000

To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.
To the best of our knowledge, the chemical, physical, and toxicological properties have not been thoroughly investigated.

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.

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
SARA 302: No chemicals in this material are subject to the reporting requirements of SARA Title III, Section 302.

SARA 313 Components
SARA 313: 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
No SARA Hazards

Massachusetts Right To Know Components
No components are subject to the Massachusetts Right to Know Act.

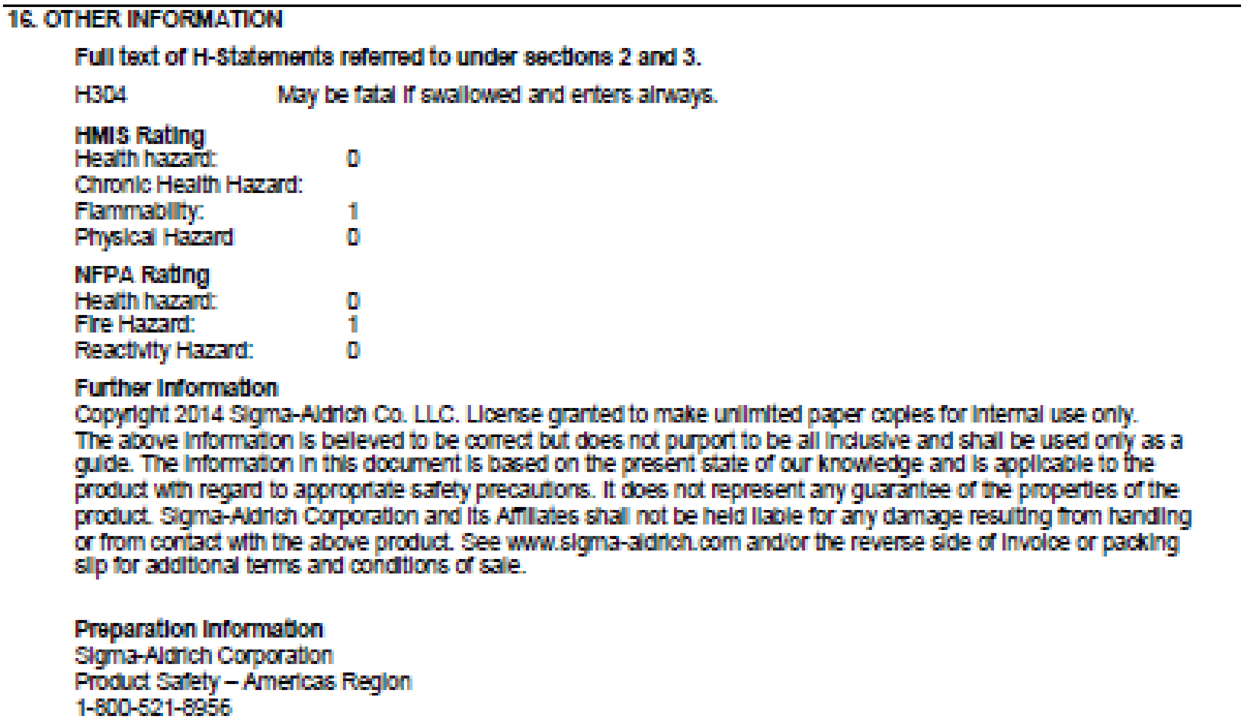
Pennsylvania Right To Know Components

	CAS-No.	Revision Date
N-Heptadecane	629-78-7	

New Jersey Right To Know Components

	CAS-No.	Revision Date
N-Heptadecane	629-78-7	

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.

H304 May be fatal if swallowed and enters airways.

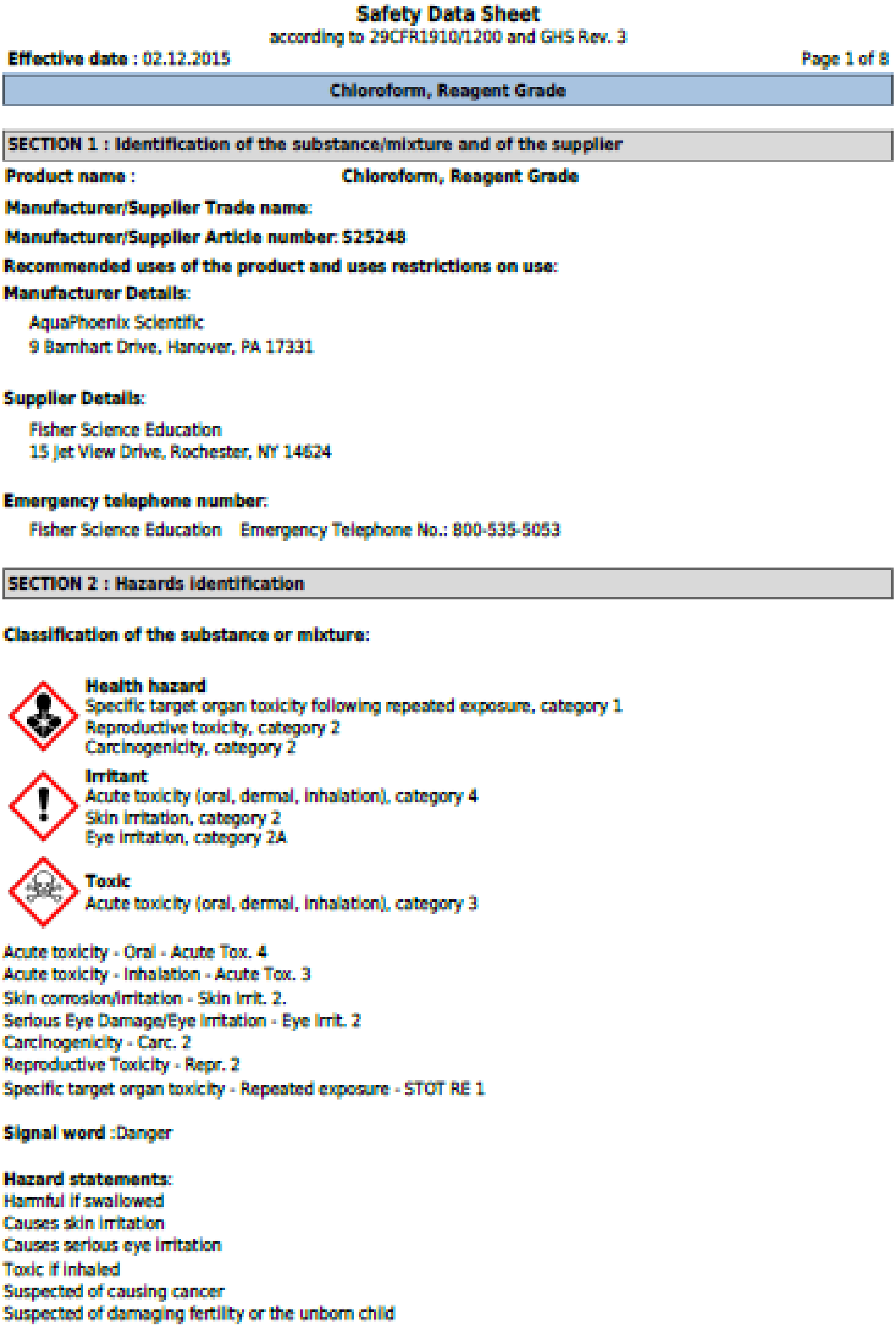
HMIS Rating
Health hazard: 0
Chronic Health Hazard:
Flammability: 1
Physical Hazard 0

NFPA Rating
Health hazard: 0
Fire Hazard: 1
Reactivity Hazard: 0

Further information

Preparation Information
Sigma-Aldrich Corporation
Product Safety – Americas Region
1-800-521-8956

A3.13. Chloroform



Safety Data Sheet
according to 29CFR1910/1200 and GHS Rev. 3

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Chloroform, Reagent Grade

SECTION 1 : Identification of the substance/mixture and of the supplier

Product name : **Chloroform, Reagent Grade**

Manufacturer/Supplier Trade name:

Manufacturer/Supplier Article number: 525248

Recommended uses of the product and uses restrictions on use:

Manufacturer Details:

AquaPhoenix Scientific
9 Barnhart Drive, Hanover, PA 17331

Supplier Details:

Fisher Science Education
15 Jet View Drive, Rochester, NY 14624

Emergency telephone number:

Fisher Science Education Emergency Telephone No.: 800-535-5053

SECTION 2 : Hazards identification

Classification of the substance or mixture:

Health hazard
Specific target organ toxicity following repeated exposure, category 1
Reproductive toxicity, category 2
Carcinogenicity, category 2

Irritant
Acute toxicity (oral, dermal, inhalation), category 4
Skin irritation, category 2
Eye irritation, category 2A

Toxic
Acute toxicity (oral, dermal, inhalation), category 3

Acute toxicity - Oral - Acute Tox. 4
Acute toxicity - Inhalation - Acute Tox. 3
Skin corrosion/irritation - Skin Irrit. 2.
Serious Eye Damage/Eye Irritation - Eye Irrit. 2
Carcinogenicity - Carc. 2
Reproductive Toxicity - Repr. 2
Specific target organ toxicity - Repeated exposure - STOT RE 1

Signal word :Danger

Hazard statements:
Harmful if swallowed
Causes skin irritation
Causes serious eye irritation
Toxic if inhaled
Suspected of causing cancer
Suspected of damaging fertility or the unborn child

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Chloroform, Reagent Grade

Causes damage to organs through prolonged or repeated exposure

Precautionary statements:

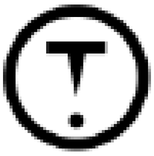
If medical advice is needed, have product container or label at hand
Keep out of reach of children
Read label before use
Do not handle until all safety precautions have been read and understood
Obtain special instructions before use
Avoid breathing dust/fume/gas/mist/vapours/spray
Use only outdoors or in a well-ventilated area
Use personal protective equipment as required
Wash skin thoroughly after handling
Do not eat, drink or smoke when using this product
Wear protective gloves/protective clothing/eye protection/face protection
Call a POISON CENTER or doctor/physician
If skin irritation occurs: Get medical advice/attention
IF SWALLOWED: Call a POISON CENTER or doctor/physician if you feel unwell
IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing
IF exposed or concerned: Get medical advice/attention
Get Medical advice/attention if you feel unwell
Specific treatment (see supplemental first aid instructions on this label)
Rinse mouth
Take off contaminated clothing and wash before reuse
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 advice/attention
IF ON SKIN: Wash with soap and water
Store in a well ventilated place. Keep container tightly closed
Store locked up
Dispose of contents and container as instructed in Section 13

Other Non-GHS Classification:

WHMIS

D1B

D2B



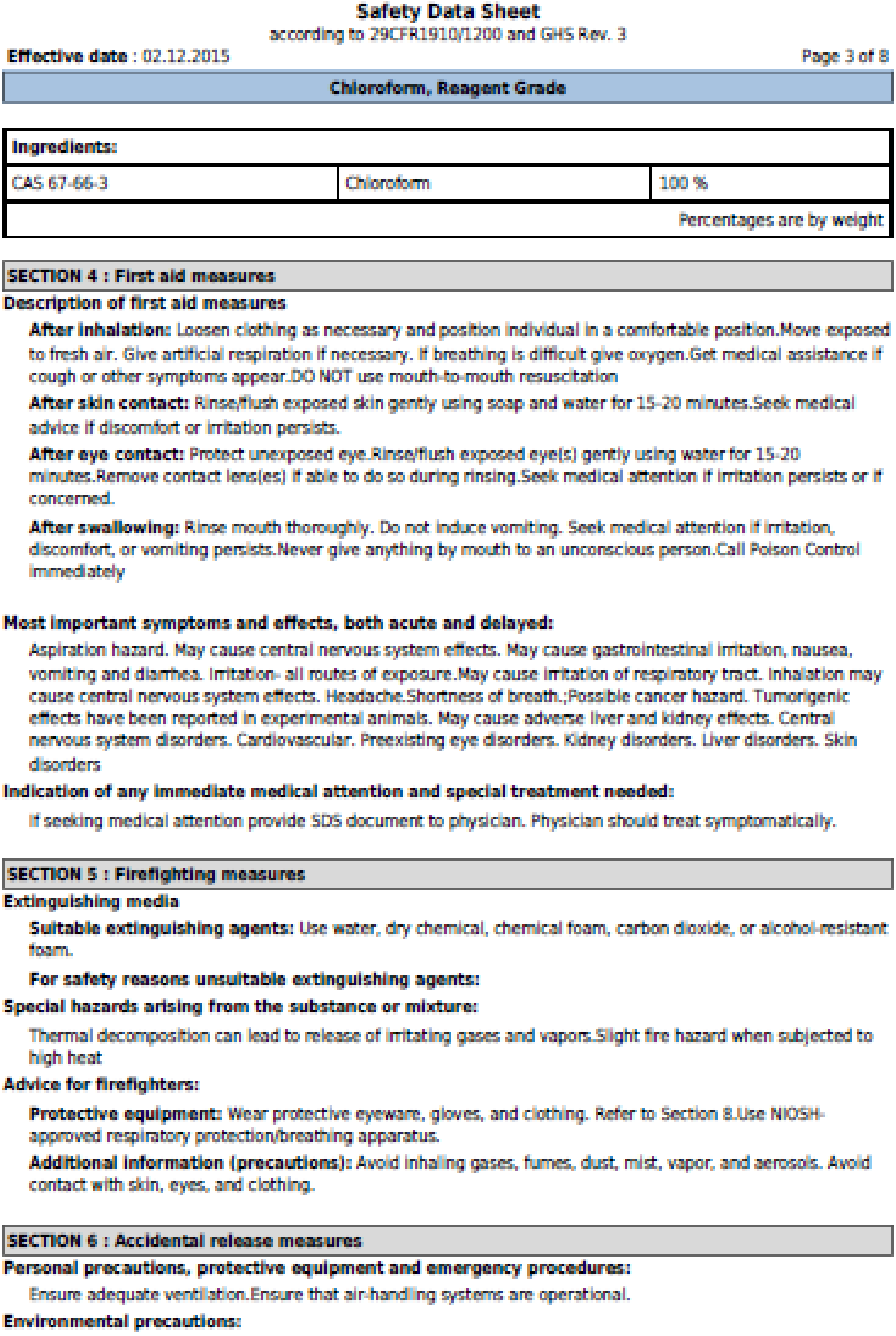
NFPA/HMIS

NFPA SCALE (0-4)

Health	2
Flammability	0
Physical Hazard	0
Personal Protection	X

HMIS RATINGS (0-4)

SECTION 3 : Composition/information on ingredients



Safety Data Sheet

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Chloroform, Reagent Grade

Ingredients:		
CAS 67-66-3	Chloroform	100 %
		Percentages are by weight

SECTION 4 : First aid measures

Description of first aid measures

After inhalation: Loosen clothing as necessary and position individual in a comfortable position.Move exposed to fresh air. Give artificial respiration if necessary. If breathing is difficult give oxygen.Get medical assistance if cough or other symptoms appear.DO NOT use mouth-to-mouth resuscitation

After skin contact: Rinse/flush exposed skin gently using soap and water for 15-20 minutes.Seek medical advice if discomfort or irritation persists.

After eye contact: Protect unexposed eye.Rinse/flush exposed eye(s) gently using water for 15-20 minutes.Remove contact lens(es) if able to do so during rinsing.Seek medical attention if irritation persists or if concerned.

After swallowing: Rinse mouth thoroughly. Do not induce vomiting. Seek medical attention if irritation, discomfort, or vomiting persists.Never give anything by mouth to an unconscious person.Call Poison Control immediately

Most important symptoms and effects, both acute and delayed:

Aspiration hazard. May cause central nervous system effects. May cause gastrointestinal irritation, nausea, vomiting and diarrhea. Irritation- all routes of exposure.May cause irritation of respiratory tract. Inhalation may cause central nervous system effects. Headache.Shortness of breath.;Possible cancer hazard. Tumorigenic effects have been reported in experimental animals. May cause adverse liver and kidney effects. Central nervous system disorders. Cardiovascular. Preexisting eye disorders. Kidney disorders. Liver disorders. Skin disorders

Indication of any immediate medical attention and special treatment needed:

If seeking medical attention provide SDS document to physician. Physician should treat symptomatically.

SECTION 5 : Firefighting measures

Extinguishing media

Suitable extinguishing agents: Use water, dry chemical, chemical foam, carbon dioxide, or alcohol-resistant foam.

For safety reasons unsuitable extinguishing agents:

Special hazards arising from the substance or mixture:

Thermal decomposition can lead to release of irritating gases and vapors.Slight fire hazard when subjected to high heat

Advice for firefighters:

Protective equipment: Wear protective eyeware, gloves, and clothing. Refer to Section 8.Use NIOSH-approved respiratory protection/breathing apparatus.

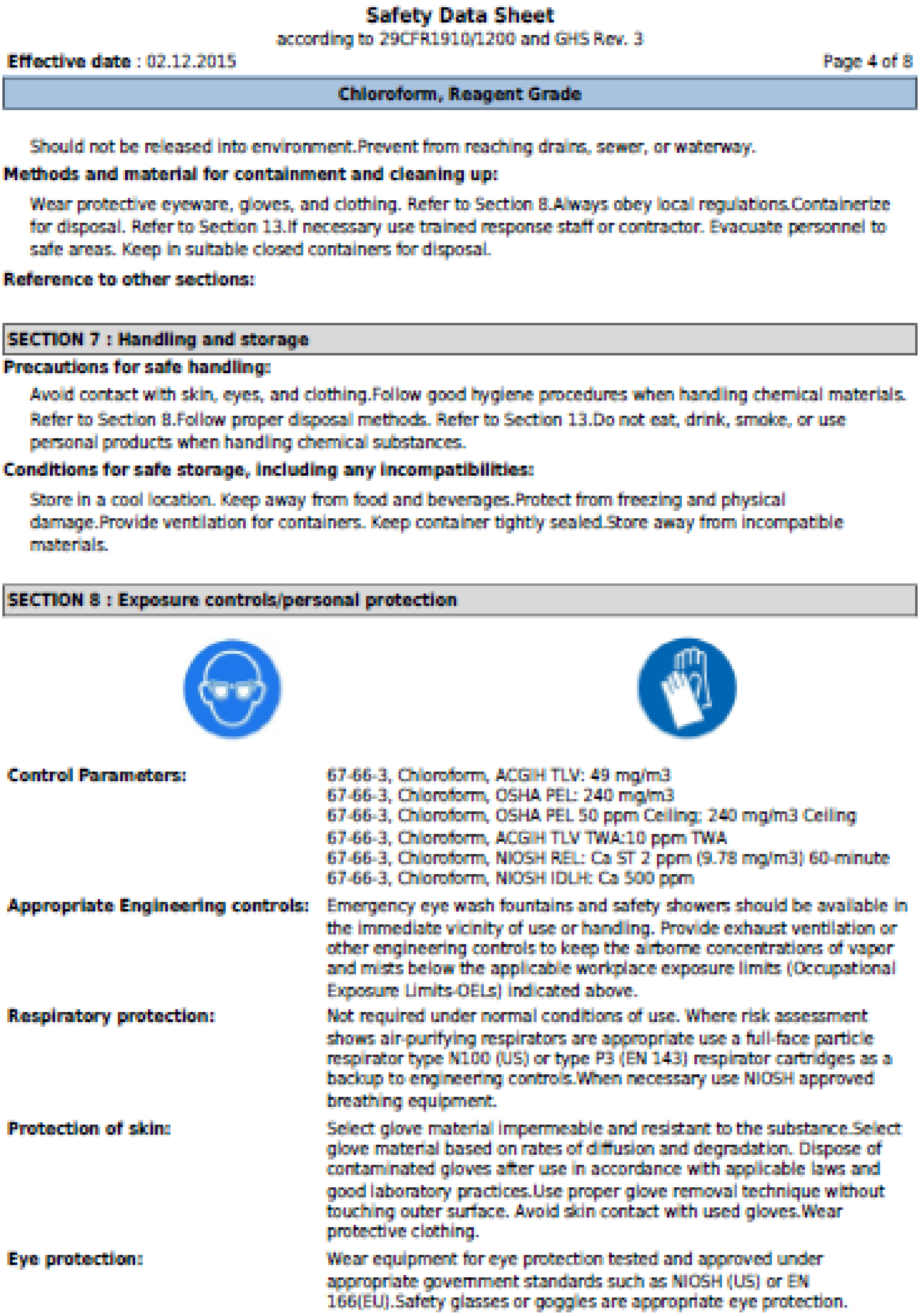
Additional information (precautions): Avoid inhaling gases, fumes, dust, mist, vapor, and aerosols. Avoid contact with skin, eyes, and clothing.

SECTION 6 : Accidental release measures

Personal precautions, protective equipment and emergency procedures:

Ensure adequate ventilation.Ensure that air-handling systems are operational.

Environmental precautions:



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Chloroform, Reagent Grade

Should not be released into environment.Prevent from reaching drains, sewer, or waterway.

Methods and material for containment and cleaning up:

Wear protective eyeware, gloves, and clothing. Refer to Section 8.Always obey local regulations.Containerize for disposal. Refer to Section 13.If necessary use trained response staff or contractor. Evacuate personnel to safe areas. Keep in suitable closed containers for disposal.

Reference to other sections:

SECTION 7 : Handling and storage

Precautions for safe handling:

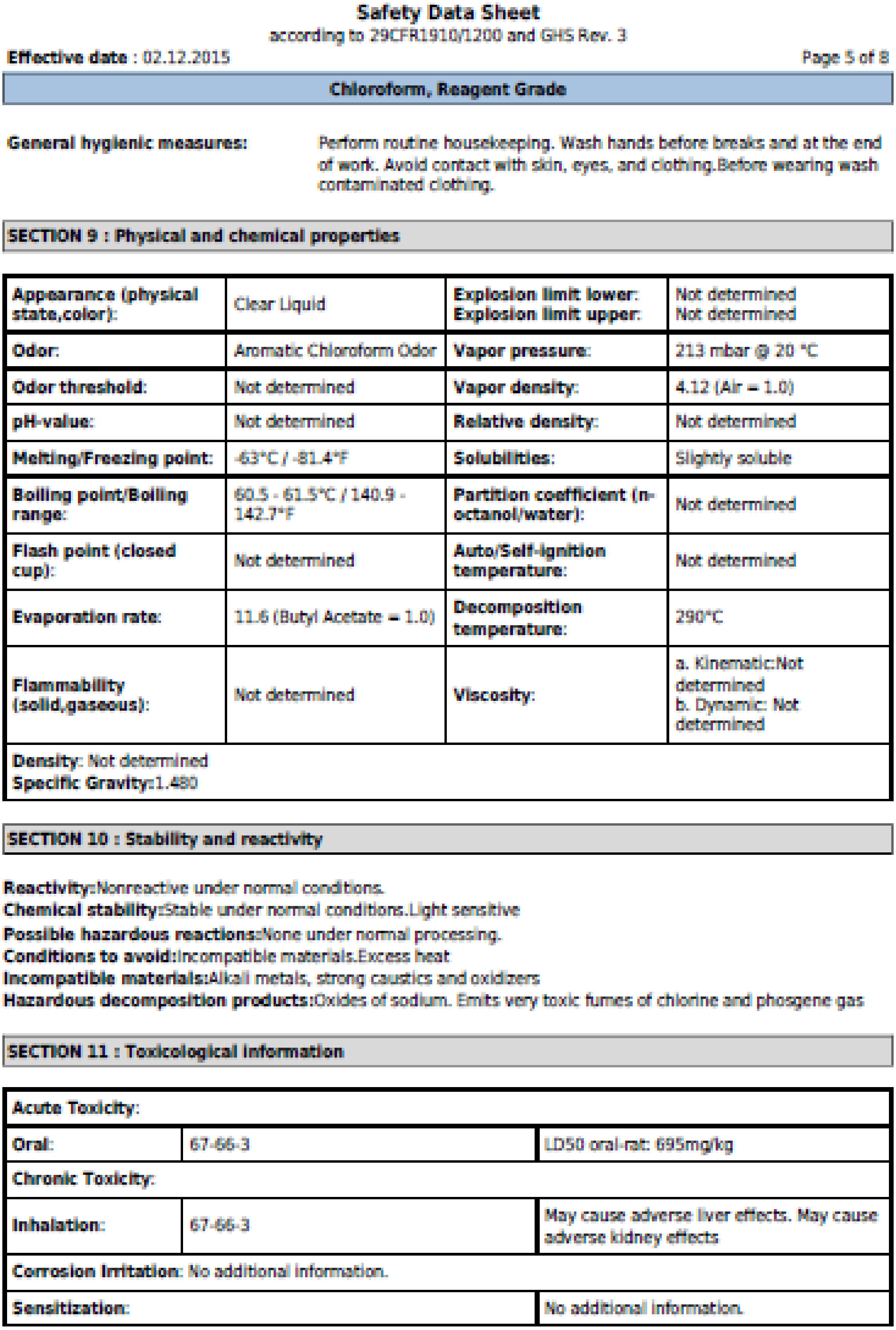
Avoid contact with skin, eyes, and clothing.Follow good hygiene procedures when handling chemical materials. Refer to Section 8.Follow proper disposal methods. Refer to Section 13.Do not eat, drink, smoke, or use personal products when handling chemical substances.

Conditions for safe storage, including any incompatibilities:

Store in a cool location. Keep away from food and beverages.Protect from freezing and physical damage.Provide ventilation for containers. Keep container tightly sealed.Store away from incompatible materials.

SECTION 8 : Exposure controls/personal protection

Control Parameters:	67-66-3, Chloroform, ACGIH TLV: 49 mg/m3 67-66-3, Chloroform, OSHA PEL: 240 mg/m3 67-66-3, Chloroform, OSHA PEL 50 ppm Ceiling; 240 mg/m3 Ceiling 67-66-3, Chloroform, ACGIH TLV TWA:10 ppm TWA 67-66-3, Chloroform, NIOSH REL: Ca ST 2 ppm (9.78 mg/m3) 60-minute 67-66-3, Chloroform, NIOSH IDLH: Ca 500 ppm
Appropriate Engineering controls:	Emergency eye wash fountains and safety showers should be available in the immediate vicinity of use or handling. Provide exhaust ventilation or other engineering controls to keep the airborne concentrations of vapor and mists below the applicable workplace exposure limits (Occupational Exposure Limits-OELs) indicated above.
Respiratory protection:	Not required under normal conditions of use. 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.When necessary use NIOSH approved breathing equipment.
Protection of skin:	Select glove material impermeable and resistant to the substance.Select glove material based on rates of diffusion and degradation. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices.Use proper glove removal technique without touching outer surface. Avoid skin contact with used gloves.Wear protective clothing.
Eye protection:	Wear equipment for eye protection tested and approved under appropriate government standards such as NIOSH (US) or EN 166(EU).Safety glasses or goggles are appropriate eye protection.



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Chloroform, Reagent Grade

General hygienic measures: Perform routine housekeeping. Wash hands before breaks and at the end of work. Avoid contact with skin, eyes, and clothing.Before wearing wash contaminated clothing.

SECTION 9 : Physical and chemical properties

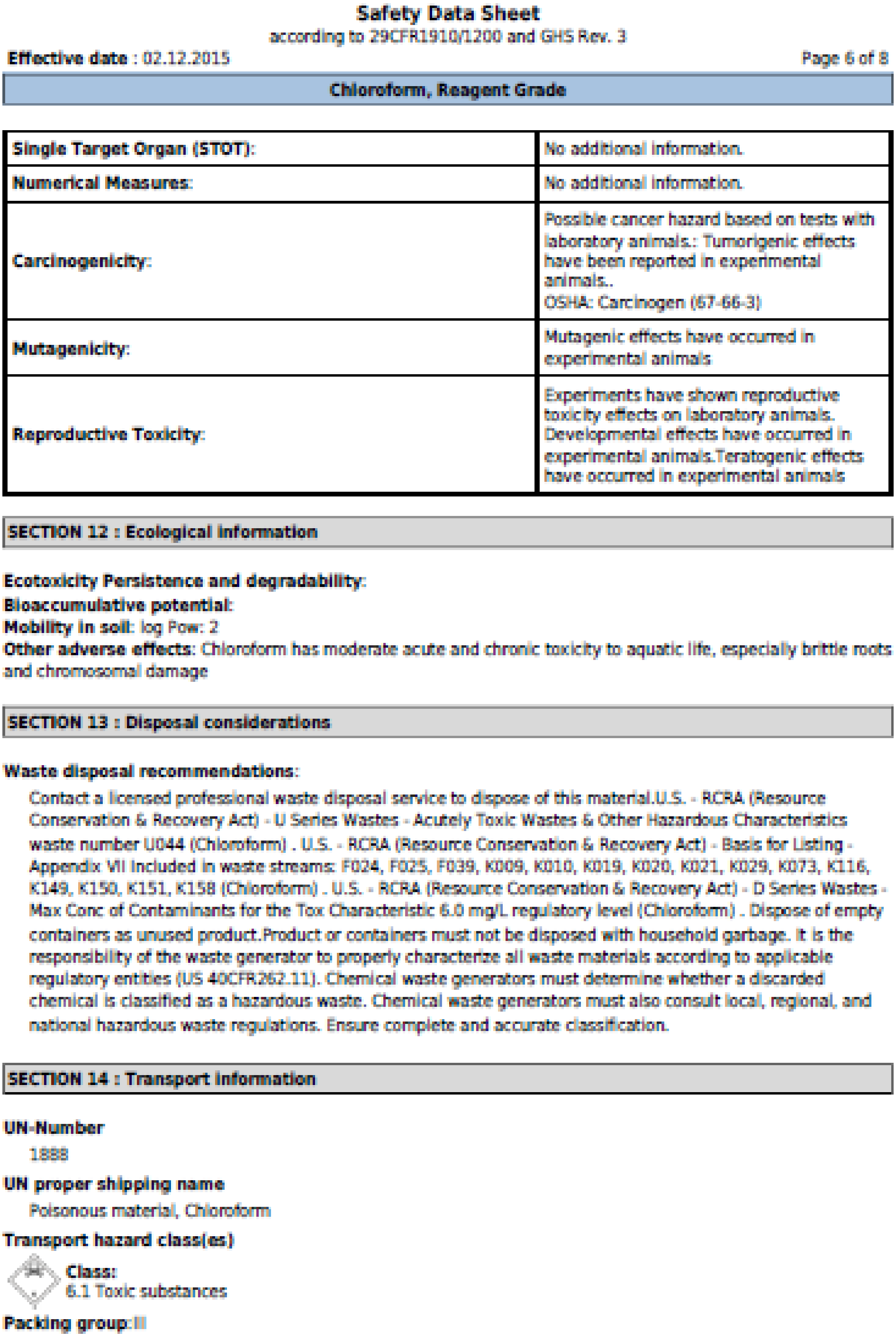
Appearance (physical state,color):	Clear Liquid	**Explosion limit lower:** **Explosion limit upper:**	Not determined Not determined
Odor:	Aromatic Chloroform Odor	**Vapor pressure:**	213 mbar @ 20 °C
Odor threshold:	Not determined	**Vapor density:**	4.12 (Air = 1.0)
pH-value:	Not determined	**Relative density:**	Not determined
Melting/Freezing point:	-63°C / -81.4°F	**Solubilities:**	Slightly soluble
Boiling point/Boiling range:	60.5 - 61.5°C / 140.9 - 142.7°F	**Partition coefficient (n-octanol/water):**	Not determined
Flash point (closed cup):	Not determined	**Auto/Self-ignition temperature:**	Not determined
Evaporation rate:	11.6 (Butyl Acetate = 1.0)	**Decomposition temperature:**	290°C
Flammability (solid,gaseous):	Not determined	**Viscosity:**	a. Kinematic:Not determined b. Dynamic: Not determined
Density: Not determined **Specific Gravity:**1.480			

SECTION 10 : Stability and reactivity

Reactivity: Nonreactive under normal conditions.
Chemical stability: Stable under normal conditions.Light sensitive
Possible hazardous reactions: None under normal processing.
Conditions to avoid: Incompatible materials.Excess heat
Incompatible materials: Alkali metals, strong caustics and oxidizers
Hazardous decomposition products: Oxides of sodium. Emits very toxic fumes of chlorine and phosgene gas

SECTION 11 : Toxicological information

Acute Toxicity:		
Oral:	67-66-3	LD50 oral-rat: 695mg/kg
Chronic Toxicity:		
Inhalation:	67-66-3	May cause adverse liver effects. May cause adverse kidney effects
Corrosion Irritation: No additional information.		
Sensitization:		No additional information.



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Chloroform, Reagent Grade

Single Target Organ (STOT):	No additional information.
Numerical Measures:	No additional information.
Carcinogenicity:	Possible cancer hazard based on tests with laboratory animals.: Tumorigenic effects have been reported in experimental animals.. OSHA: Carcinogen (67-66-3)
Mutagenicity:	Mutagenic effects have occurred in experimental animals
Reproductive Toxicity:	Experiments have shown reproductive toxicity effects on laboratory animals. Developmental effects have occurred in experimental animals.Teratogenic effects have occurred in experimental animals

SECTION 12 : Ecological information

Ecotoxicity Persistence and degradability:

Bioaccumulative potential:

Mobility in soil: log Pow: 2

Other adverse effects: Chloroform has moderate acute and chronic toxicity to aquatic life, especially brittle roots and chromosomal damage

SECTION 13 : Disposal considerations

Waste disposal recommendations:

Contact a licensed professional waste disposal service to dispose of this material.U.S. - RCRA (Resource Conservation & Recovery Act) - U Series Wastes - Acutely Toxic Wastes & Other Hazardous Characteristics waste number U044 (Chloroform) . U.S. - RCRA (Resource Conservation & Recovery Act) - Basis for Listing - Appendix VII Included in waste streams: F024, F025, F039, K009, K010, K019, K020, K021, K029, K073, K116, K149, K150, K151, K158 (Chloroform) . U.S. - RCRA (Resource Conservation & Recovery Act) - D Series Wastes - Max Conc of Contaminants for the Tox Characteristic 6.0 mg/L regulatory level (Chloroform) . Dispose of empty containers as unused product.Product or containers must not be disposed with household garbage. It is the responsibility of the waste generator to properly characterize all waste materials according to applicable regulatory entities (US 40CFR262.11). Chemical waste generators must determine whether a discarded chemical is classified as a hazardous waste. Chemical waste generators must also consult local, regional, and national hazardous waste regulations. Ensure complete and accurate classification.

SECTION 14 : Transport information

UN-Number

1888

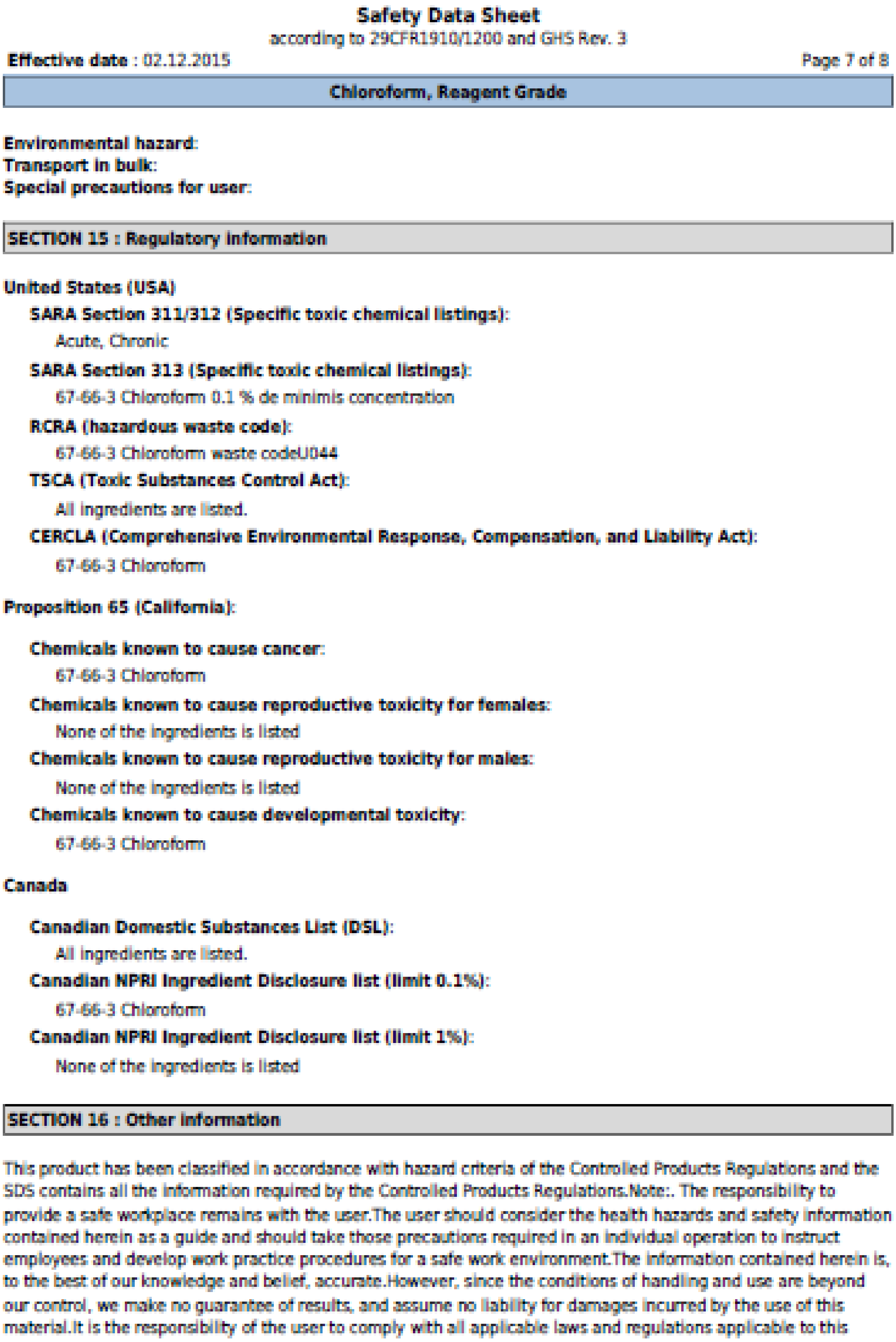
UN proper shipping name

Poisonous material, Chloroform

Transport hazard class(es)

Class:
6.1 Toxic substances

Packing group: III



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according to 29CFR1910/1200 and GHS Rev. 3

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Chloroform, Reagent Grade

Environmental hazard:
Transport in bulk:
Special precautions for user:

SECTION 15 : Regulatory information

United States (USA)

SARA Section 311/312 (Specific toxic chemical listings):

Acute, Chronic

SARA Section 313 (Specific toxic chemical listings):

67-66-3 Chloroform 0.1 % de minimis concentration

RCRA (hazardous waste code):

67-66-3 Chloroform waste codeU044

TSCA (Toxic Substances Control Act):

All ingredients are listed.

CERCLA (Comprehensive Environmental Response, Compensation, and Liability Act):

67-66-3 Chloroform

Proposition 65 (California):

Chemicals known to cause cancer:

67-66-3 Chloroform

Chemicals known to cause reproductive toxicity for females:

None of the ingredients is listed

Chemicals known to cause reproductive toxicity for males:

None of the ingredients is listed

Chemicals known to cause developmental toxicity:

67-66-3 Chloroform

Canada

Canadian Domestic Substances List (DSL):

All ingredients are listed.

Canadian NPRI Ingredient Disclosure list (limit 0.1%):

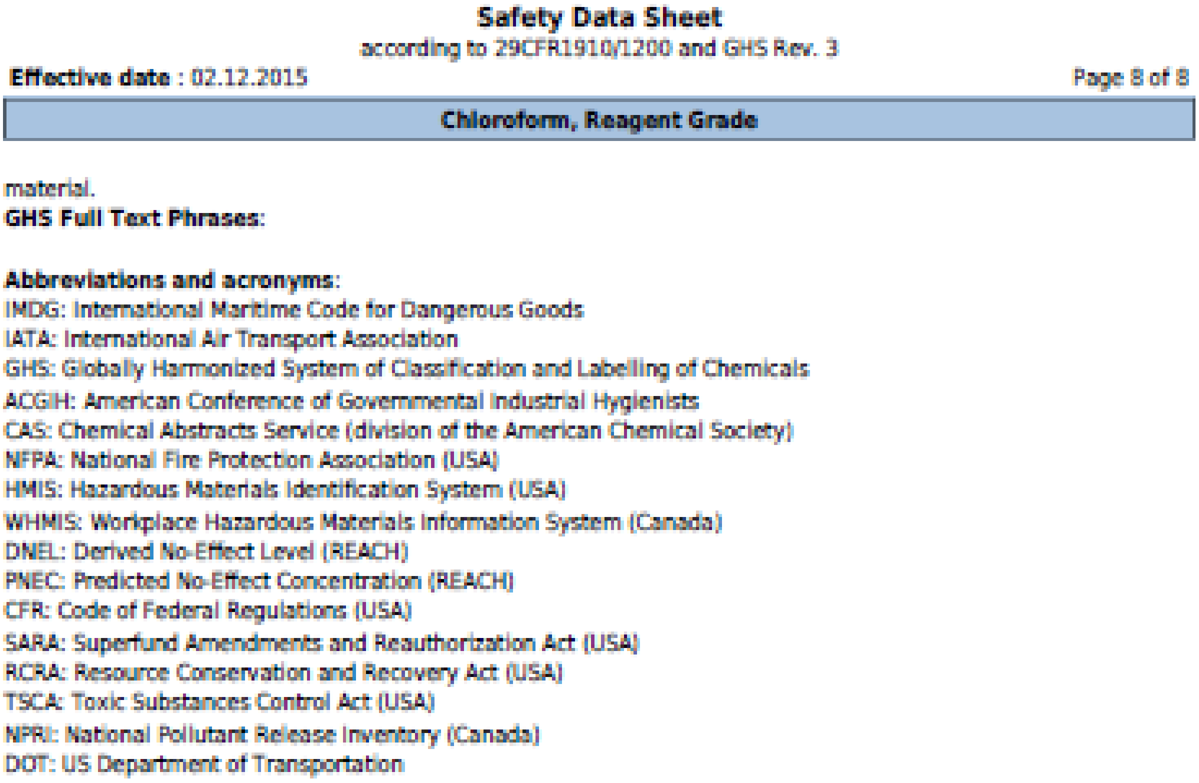
67-66-3 Chloroform

Canadian NPRI Ingredient Disclosure list (limit 1%):

None of the ingredients is listed

SECTION 16 : Other information

This product has been classified in accordance with hazard criteria of the Controlled Products Regulations and the SDS contains all the information required by the Controlled Products Regulations.Note:. The responsibility to provide a safe workplace remains with the user.The user should consider the health hazards and safety information contained herein as a guide and should take those precautions required in an individual operation to instruct employees and develop work practice procedures for a safe work environment.The information contained herein is, to the best of our knowledge and belief, accurate.However, since the conditions of handling and use are beyond our control, we make no guarantee of results, and assume no liability for damages incurred by the use of this material.It is the responsibility of the user to comply with all applicable laws and regulations applicable to this



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Chloroform, Reagent Grade

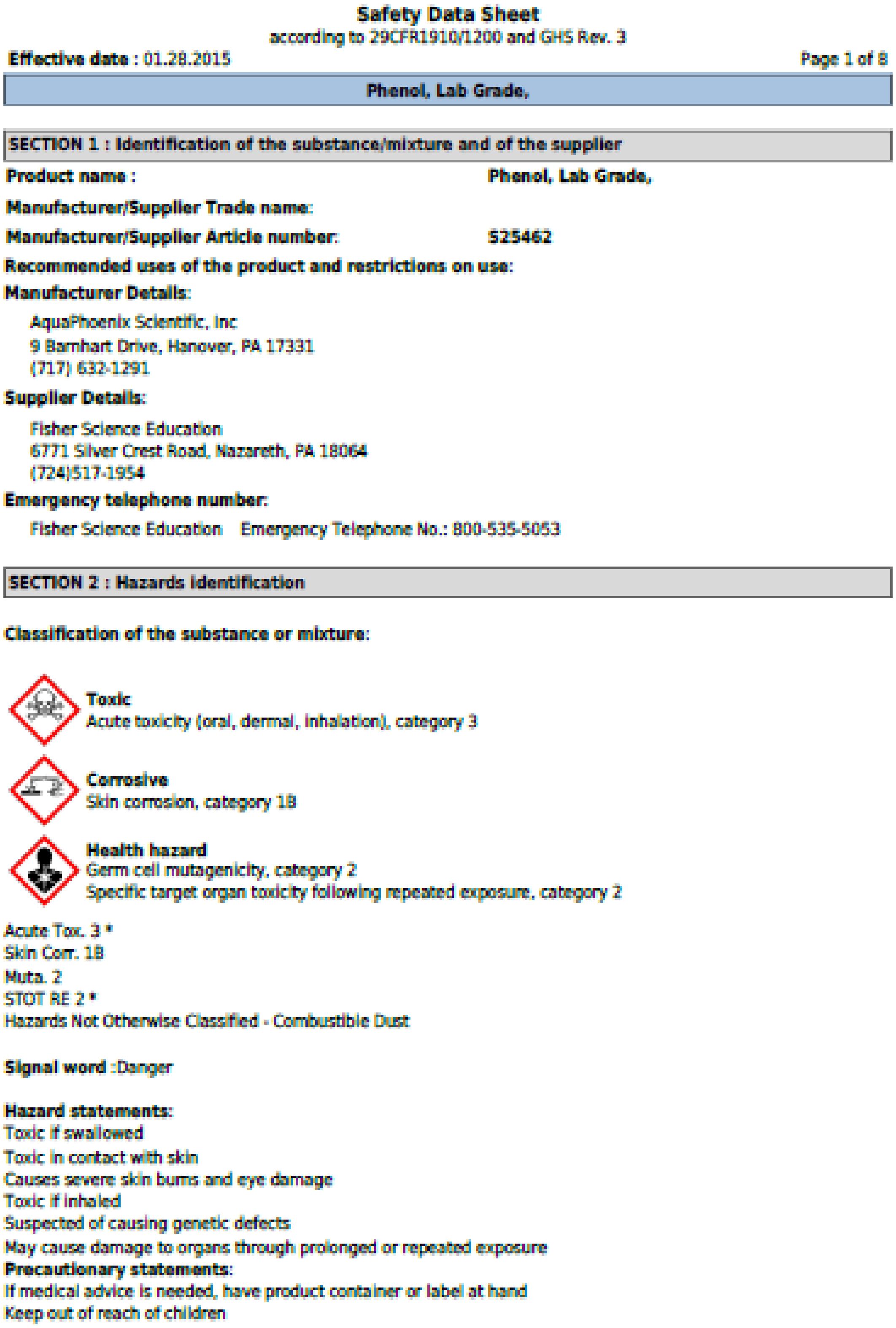
material.

GHS Full Text Phrases:

Abbreviations and acronyms:

IMDG: International Maritime Code for Dangerous Goods
IATA: International Air Transport Association
GHS: Globally Harmonized System of Classification and Labelling of Chemicals
ACGIH: American Conference of Governmental Industrial Hygienists
CAS: Chemical Abstracts Service (division of the American Chemical Society)
NFPA: National Fire Protection Association (USA)
HMIS: Hazardous Materials Identification System (USA)
WHMIS: Workplace Hazardous Materials Information System (Canada)
DNEL: Derived No-Effect Level (REACH)
PNEC: Predicted No-Effect Concentration (REACH)
CFR: Code of Federal Regulations (USA)
SARA: Superfund Amendments and Reauthorization Act (USA)
RCRA: Resource Conservation and Recovery Act (USA)
TSCA: Toxic Substances Control Act (USA)
NPRI: National Pollutant Release Inventory (Canada)
DOT: US Department of Transportation

A3.14. Phenol



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Phenol, Lab Grade,

SECTION 1 : Identification of the substance/mixture and of the supplier

Product name : **Phenol, Lab Grade,**

Manufacturer/Supplier Trade name:

Manufacturer/Supplier Article number: **S25462**

Recommended uses of the product and restrictions on use:

Manufacturer Details:

AquaPhoenix Scientific, Inc
9 Barnhart Drive, Hanover, PA 17331
(717) 632-1291

Supplier Details:

Fisher Science Education
6771 Silver Crest Road, Nazareth, PA 18064
(724)517-1954

Emergency telephone number:

Fisher Science Education Emergency Telephone No.: 800-535-5053

SECTION 2 : Hazards identification

Classification of the substance or mixture:

Toxic
Acute toxicity (oral, dermal, inhalation), category 3

Corrosive
Skin corrosion, category 1B

Health hazard
Germ cell mutagenicity, category 2
Specific target organ toxicity following repeated exposure, category 2

Acute Tox. 3 *
Skin Corr. 1B
Muta. 2
STOT RE 2 *
Hazards Not Otherwise Classified - Combustible Dust

Signal word :Danger

Hazard statements:
Toxic if swallowed
Toxic in contact with skin
Causes severe skin burns and eye damage
Toxic if inhaled
Suspected of causing genetic defects
May cause damage to organs through prolonged or repeated exposure
Precautionary statements:
If medical advice is needed, have product container or label at hand
Keep out of reach of children

Read label before use
Obtain special instructions before use
Do not handle until all safety precautions have been read and understood
Avoid breathing dust/fume/gas/mist/vapours/spray
Wash skin thoroughly after handling
Do not eat, drink or smoke when using this product
Use only outdoors or in a well-ventilated area
Use personal protective equipment as required
Wear protective gloves/protective clothing/eye protection/face protection
Call a POISON CENTER or doctor/physician if you feel unwell
IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing
IF exposed or concerned: Get medical advice/attention
Collect spillage
IF SWALLOWED: Rinse mouth. Do NOT induce vomiting
IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower
IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses if present and easy to do. Continue rinsing
Immediately call a POISON CENTER or doctor/physician
Specific measures (see supplemental first aid instructions on this label)
Remove/Take off immediately all contaminated clothing
Wash contaminated clothing before reuse
IF SWALLOWED: Immediately call a POISON CENTER or doctor/physician
Store locked up
Dispose of contents and container to an approved waste disposal plant

Combustible Dust Hazard: :
May form combustible dust concentrations in air (during processing).

Other Non-GHS Classification:

WHMIS

NFPA/HMIS

NFPA SCALE (0-4)

Health	3
Flammability	2
Physical Hazard	0
Personal Protection	X

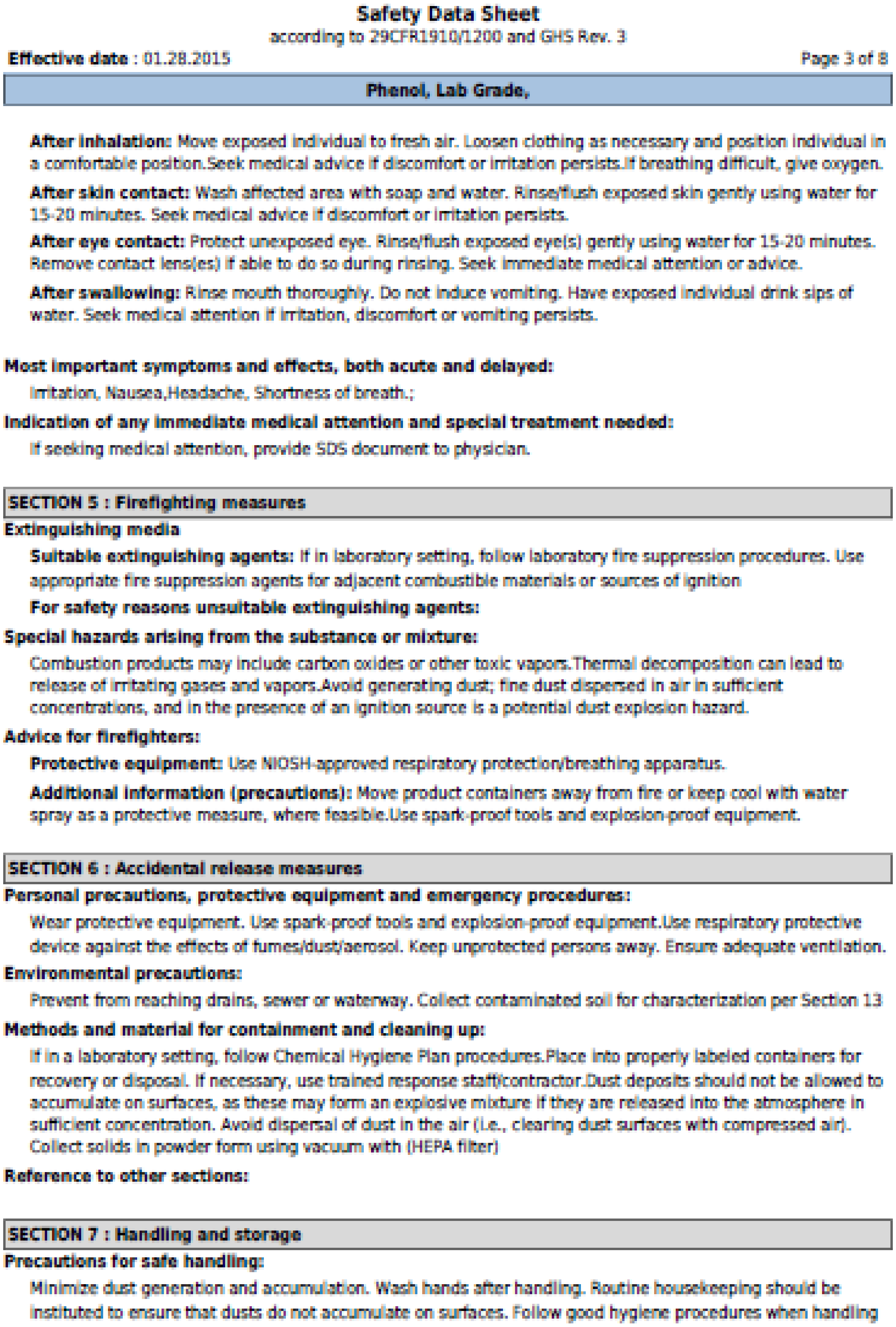
HMIS RATINGS (0-4)

SECTION 3 : Composition/information on ingredients

Ingredients:		
CAS 108-95-2	Phenol	100 %
		Percentages are by weight

SECTION 4 : First aid measures

Description of first aid measures



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Phenol, Lab Grade,

After inhalation: Move exposed individual to fresh air. Loosen clothing as necessary and position individual in a comfortable position.Seek medical advice if discomfort or irritation persists.If breathing difficult, give oxygen.

After skin contact: Wash affected area with soap and water. Rinse/flush exposed skin gently using water for 15-20 minutes. Seek medical advice if discomfort or irritation persists.

After eye contact: Protect unexposed eye. Rinse/flush exposed eye(s) gently using water for 15-20 minutes. Remove contact lens(es) if able to do so during rinsing. Seek immediate medical attention or advice.

After swallowing: Rinse mouth thoroughly. Do not induce vomiting. Have exposed individual drink sips of water. Seek medical attention if irritation, discomfort or vomiting persists.

Most important symptoms and effects, both acute and delayed:

Irritation, Nausea,Headache, Shortness of breath.;

Indication of any immediate medical attention and special treatment needed:

If seeking medical attention, provide SDS document to physician.

SECTION 5 : Firefighting measures

Extinguishing media

Suitable extinguishing agents: If in laboratory setting, follow laboratory fire suppression procedures. Use appropriate fire suppression agents for adjacent combustible materials or sources of ignition

For safety reasons unsuitable extinguishing agents:

Special hazards arising from the substance or mixture:

Combustion products may include carbon oxides or other toxic vapors.Thermal decomposition can lead to release of irritating gases and vapors.Avoid generating dust; fine dust dispersed in air in sufficient concentrations, and in the presence of an ignition source is a potential dust explosion hazard.

Advice for firefighters:

Protective equipment: Use NIOSH-approved respiratory protection/breathing apparatus.

Additional information (precautions): Move product containers away from fire or keep cool with water spray as a protective measure, where feasible.Use spark-proof tools and explosion-proof equipment.

SECTION 6 : Accidental release measures

Personal precautions, protective equipment and emergency procedures:

Wear protective equipment. Use spark-proof tools and explosion-proof equipment.Use respiratory protective device against the effects of fumes/dust/aerosol. Keep unprotected persons away. Ensure adequate ventilation.

Environmental precautions:

Prevent from reaching drains, sewer or waterway. Collect contaminated soil for characterization per Section 13

Methods and material for containment and cleaning up:

If in a laboratory setting, follow Chemical Hygiene Plan procedures.Place into properly labeled containers for recovery or disposal. If necessary, use trained response staff/contractor.Dust deposits should not be allowed to accumulate on surfaces, as these may form an explosive mixture if they are released into the atmosphere in sufficient concentration. Avoid dispersal of dust in the air (i.e., clearing dust surfaces with compressed air). Collect solids in powder form using vacuum with (HEPA filter)

Reference to other sections:

SECTION 7 : Handling and storage

Precautions for safe handling:

Minimize dust generation and accumulation. Wash hands after handling. Routine housekeeping should be instituted to ensure that dusts do not accumulate on surfaces. Follow good hygiene procedures when handling

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Phenol, Lab Grade,

chemical materials. Do not eat, drink, smoke, or use personal products when handling chemical substances. If in a laboratory setting, follow Chemical Hygiene Plan.Use only in well ventilated areas.Avoid contact with eyes, skin, and clothing.

Conditions for safe storage, including any incompatibilities:

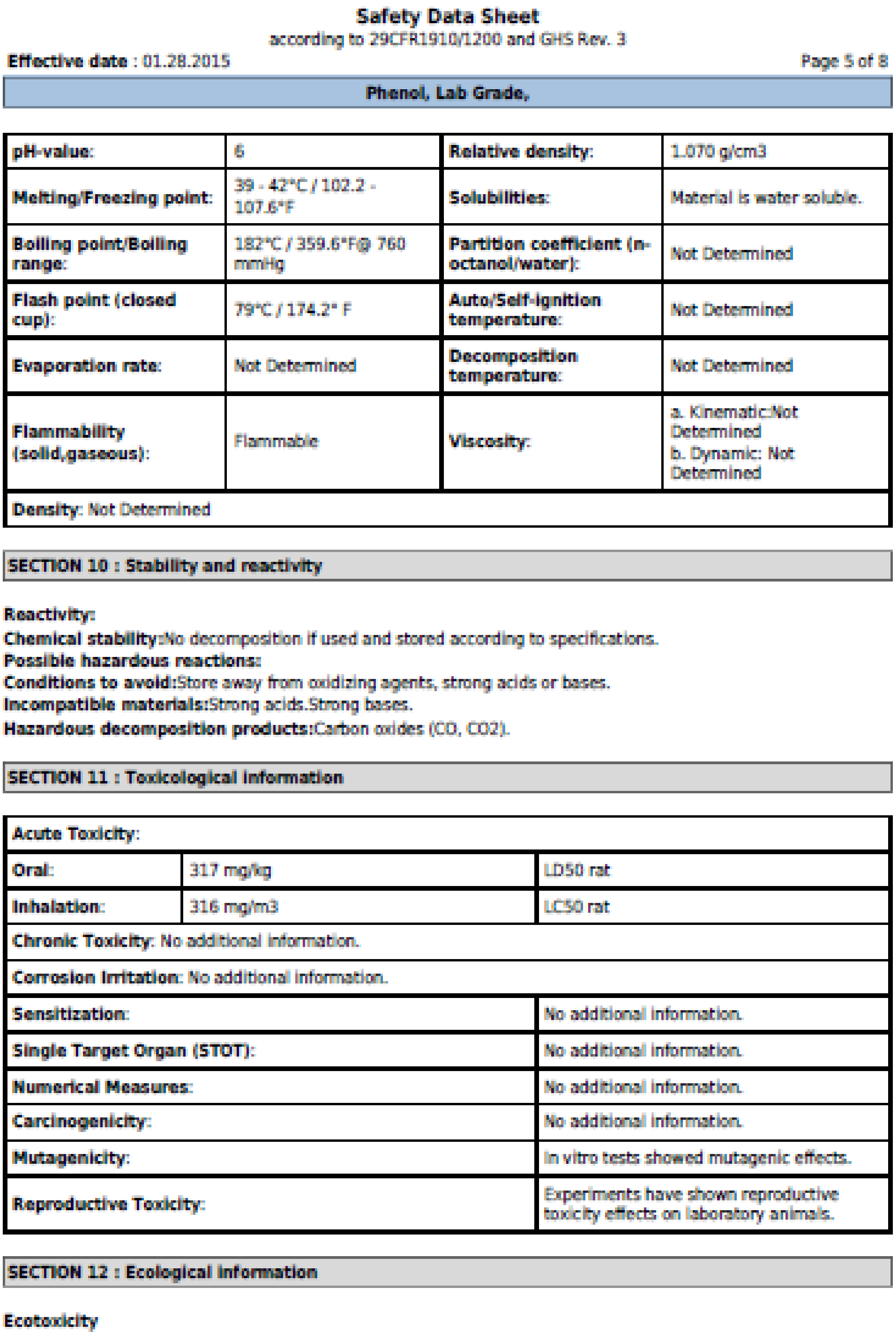
Provide ventilation for containers. Avoid storage near extreme heat, ignition sources or open flame. Store away from foodstuffs. Store away from oxidizing agents.Store in cool, dry conditions in well sealed containers. Store with like hazards

SECTION 8 : Exposure controls/personal protection

Control Parameters:	108-95-2, Phenol, ACGIH TLV TWA 5ppm 108-95-2, Phenol, NIOSH TWA 19mg/m3
Appropriate Engineering controls:	Emergency eye wash fountains and safety showers should be available in the immediate vicinity of use/handling.Provide exhaust ventilation or other engineering controls to keep the airborne concentrations of vapor or dusts (total/respirable) below the applicable workplace exposure limits (Occupational Exposure Limits-OELs) indicated above.Use under a fume hood. It is recommended that all dust control equipment such as local exhaust ventilation and material transport systems involved in handling of this product contain explosion relief vents or an explosion suppression system or an oxygen deficient environment.Ensure that dust-handling systems (such as exhaust ducts, dust collectors, vessels, and processing equipment) are designed in a manner to prevent the escape of dust into the work area (i.e., there is no leakage from the equipment).
Respiratory protection:	Use suitable respiratory protective device when high concentrations are present. Use suitable respiratory protective device when aerosol or mist is formed. For spills, respiratory protection may be advisable.
Protection of skin:	The glove material has to be impermeable and resistant to the product/ the substance/ the preparation being used/handled.Selection of the glove material on consideration of the penetration times, rates of diffusion and the degradation.
Eye protection:	Safety glasses with side shields or goggles.
General hygienic measures:	The usual precautionary measures are to be adhered to when handling chemicals. Keep away from food, beverages and feed sources. Immediately remove all soiled and contaminated clothing. Wash hands before breaks and at the end of work. Do not inhale gases/fumes/dust/mist/vapor/aerosols. Avoid contact with the eyes and skin.

SECTION 9 : Physical and chemical properties

Appearance (physical state,color):	Clear, colorless crystals.	**Explosion limit lower:** **Explosion limit upper:**	Not Determined Not Determined
Odor:	Disinfectant odor	**Vapor pressure:**	0.4 mbar @ 20 °C
Odor threshold:	Not Determined	**Vapor density:**	3.2



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Phenol, Lab Grade,

pH-value:	6	**Relative density:**	1.070 g/cm3
Melting/Freezing point:	39 - 42°C / 102.2 - 107.6°F	**Solubilities:**	Material is water soluble.
Boiling point/Boiling range:	182°C / 359.6°F@ 760 mmHg	**Partition coefficient (n-octanol/water):**	Not Determined
Flash point (closed cup):	79°C / 174.2° F	**Auto/Self-ignition temperature:**	Not Determined
Evaporation rate:	Not Determined	**Decomposition temperature:**	Not Determined
Flammability (solid,gaseous):	Flammable	**Viscosity:**	a. Kinematic:Not Determined b. Dynamic: Not Determined
Density: Not Determined			

SECTION 10 : Stability and reactivity

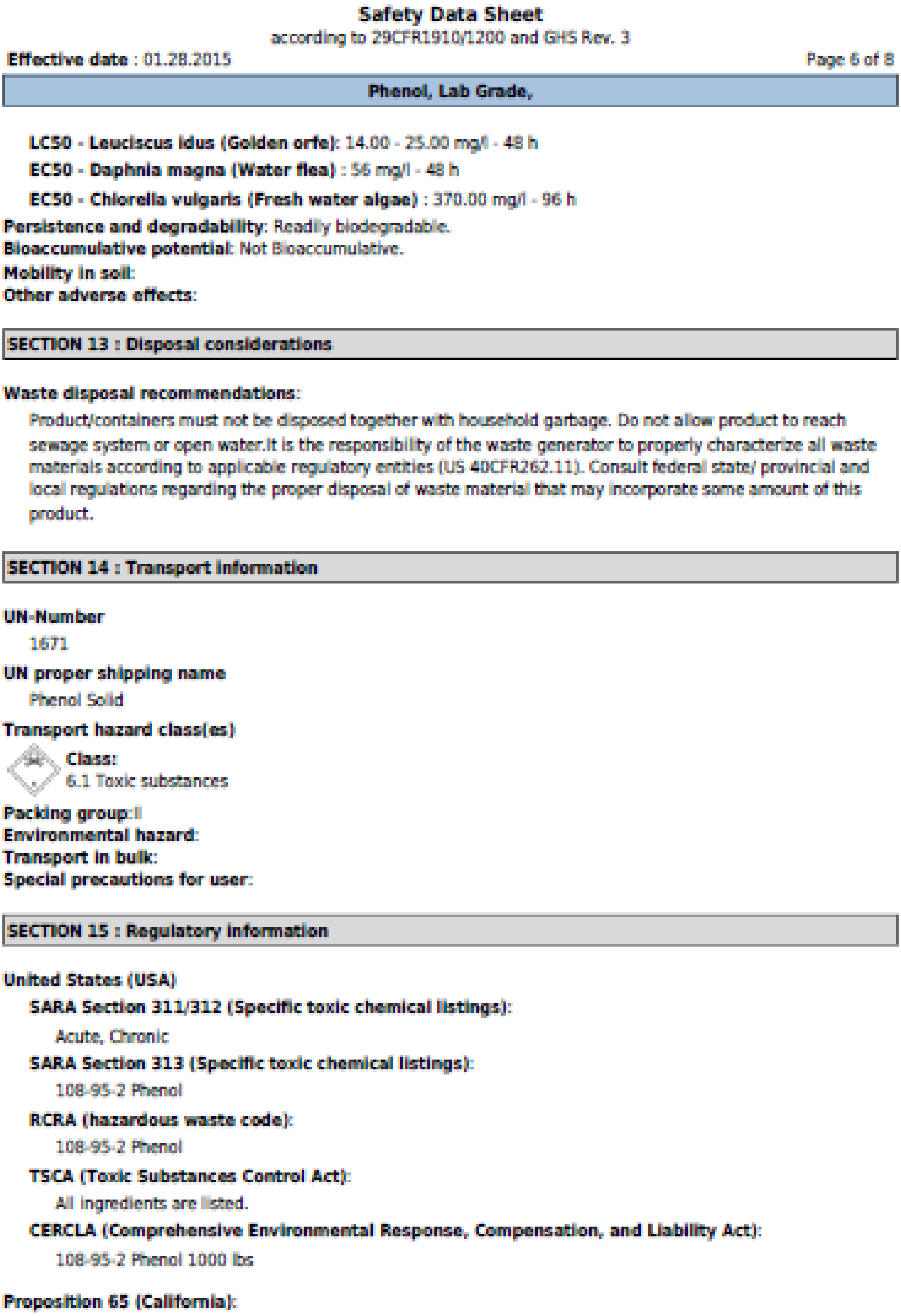
Reactivity:
Chemical stability: No decomposition if used and stored according to specifications.
Possible hazardous reactions:
Conditions to avoid: Store away from oxidizing agents, strong acids or bases.
Incompatible materials: Strong acids.Strong bases.
Hazardous decomposition products: Carbon oxides (CO, CO2).

SECTION 11 : Toxicological information

Acute Toxicity:		
Oral:	317 mg/kg	LD50 rat
Inhalation:	316 mg/m3	LC50 rat
Chronic Toxicity: No additional information.		
Corrosion Irritation: No additional information.		
Sensitization:		No additional information.
Single Target Organ (STOT):		No additional information.
Numerical Measures:		No additional information.
Carcinogenicity:		No additional information.
Mutagenicity:		In vitro tests showed mutagenic effects.
Reproductive Toxicity:		Experiments have shown reproductive toxicity effects on laboratory animals.

SECTION 12 : Ecological information

Ecotoxicity



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Phenol, Lab Grade,

LC50 - Leuciscus idus (Golden orfe): 14.00 - 25.00 mg/l - 48 h

EC50 - Daphnia magna (Water flea) : 56 mg/l - 48 h

EC50 - Chlorella vulgaris (Fresh water algae) : 370.00 mg/l - 96 h

Persistence and degradability: Readily biodegradable.
Bioaccumulative potential: Not Bioaccumulative.
Mobility in soil:
Other adverse effects:

SECTION 13 : Disposal considerations

Waste disposal recommendations:

Product/containers must not be disposed together with household garbage. Do not allow product to reach sewage system or open water.It is the responsibility of the waste generator to properly characterize all waste materials according to applicable regulatory entities (US 40CFR262.11). Consult federal state/ provincial and local regulations regarding the proper disposal of waste material that may incorporate some amount of this product.

SECTION 14 : Transport information

UN-Number

1671

UN proper shipping name

Phenol Solid

Transport hazard class(es)

Class:
6.1 Toxic substances

Packing group: II
Environmental hazard:
Transport in bulk:
Special precautions for user:

SECTION 15 : Regulatory information

United States (USA)

SARA Section 311/312 (Specific toxic chemical listings):

Acute, Chronic

SARA Section 313 (Specific toxic chemical listings):

108-95-2 Phenol

RCRA (hazardous waste code):

108-95-2 Phenol

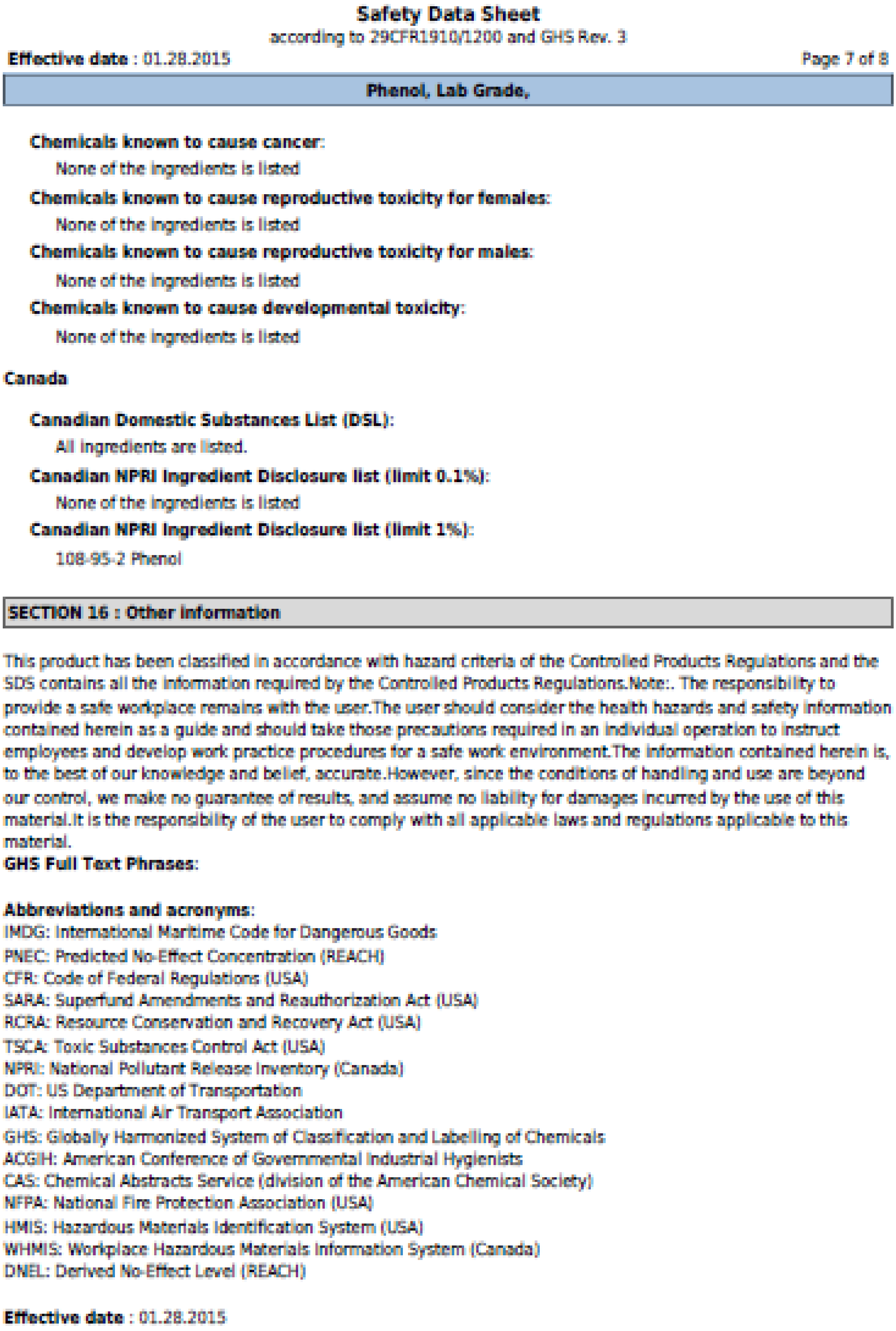
TSCA (Toxic Substances Control Act):

All ingredients are listed.

CERCLA (Comprehensive Environmental Response, Compensation, and Liability Act):

108-95-2 Phenol 1000 lbs

Proposition 65 (California):



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Phenol, Lab Grade,

Chemicals known to cause cancer:

None of the ingredients is listed

Chemicals known to cause reproductive toxicity for females:

None of the ingredients is listed

Chemicals known to cause reproductive toxicity for males:

None of the ingredients is listed

Chemicals known to cause developmental toxicity:

None of the ingredients is listed

Canada

Canadian Domestic Substances List (DSL):

All ingredients are listed.

Canadian NPRI Ingredient Disclosure list (limit 0.1%):

None of the ingredients is listed

Canadian NPRI Ingredient Disclosure list (limit 1%):

108-95-2 Phenol

SECTION 16 : Other information

This product has been classified in accordance with hazard criteria of the Controlled Products Regulations and the SDS contains all the information required by the Controlled Products Regulations.Note:. The responsibility to provide a safe workplace remains with the user.The user should consider the health hazards and safety information contained herein as a guide and should take those precautions required in an individual operation to instruct employees and develop work practice procedures for a safe work environment.The information contained herein is, to the best of our knowledge and belief, accurate.However, since the conditions of handling and use are beyond our control, we make no guarantee of results, and assume no liability for damages incurred by the use of this material.It is the responsibility of the user to comply with all applicable laws and regulations applicable to this material.

GHS Full Text Phrases:

Abbreviations and acronyms:

IMDG: International Maritime Code for Dangerous Goods
PNEC: Predicted No-Effect Concentration (REACH)
CFR: Code of Federal Regulations (USA)
SARA: Superfund Amendments and Reauthorization Act (USA)
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DNEL: Derived No-Effect Level (REACH)

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