Conference Paper

Smartphone-Based Wireless Operated Milliliter-Scale Bioreactor

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Abstract

This paper presents the development of a smartphone-based wireless operated milliliter scale bioreactor. The bioreactor has a working volume of 18 mL and is made of poly (methylmethacrylate) (PMMA) and poly (dimethylsiloxane) (PDMS) polymers using a laser engraving methodology. Temperature and reactor stirring speed are the controlled variables and wireless communication is achieved through a wireless transceiver embedded in a low-cost controller platform. The results attained are very promising as the controller is able to control the desired reactor variables precisely through wireless communications.

Keywords: Minibioreactor, wireless, smart phone, portable, integrated, disposable

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1 Introduction

Microfabrication technology has enabled the possibility of downsizing a laboratory scale bioreactor system for carrying out various biological based reactions. By reducing the size of the reactor, for example from 1.5 liter bioreactor system down to milliliter range scale i.e. a minibioreactor system with working volume of about 15 mL, medium consumption and utility cost will be significantly decreased at least by a factor of 100. Moreover, due to its reduced size and small footprint, a minibioreactor can be scaled out to facilitate multiple reactions in parallel, thus greatly increasing the throughput of the experiment. Research into miniaturization of bioprocessing has shown that this strategy helps minimize the number of expensive large-scale trials required during process development [1].

Monitoring and controlling abioreactor system is a challenging task. This is due to the differences in the real-time measurement technique applied and the execution of various feedback control loops for each of the reactor variables [2]. To meet this requirement, bioreactor operators usually favour an off-the-shelf data acquisition (DAQ) board such as National Instruments (NI) I/O card and Labview software for automation of such a miniature scale bioreactor setup. Maharbiz *et al.* [3] employed a commercially fabricated printed circuit board (PCB) to operate an array of eight 250 μL microbioreactors where various sensors (PT 100, pH ISFET chip and LEDs) were wire-bonded onto the PCB board. Data acquisition were carried out using an on-board BASIC STAMP microcontroller (Parallax, Rocklin, CA), and a PCMCIA data acquisition card (DAQCard-6062E, National Instruments, TX, US). Szita *et al.*. [4] utilized a GPIB interface card (PCI-GPIB, National Instruments, TX, US) for the optical measurement of four 150 μL microbioreactors where multiplexing of the signals for the LEDs and the photodetectors were done using the RS232 ports. Additional I/O card i.e. an analog output card PCI 6722 (National Instruments, TX, US) was used for individual control of reactors stirring speed. As evident, whilst a minibioreactor device is typically small in size, in order to operate it, bulky data acquisition and control apparatus are often required.

In order to tackle this shortcoming, this work presents design and implementation of an integrated smartphone-based wireless operated minibioreactor. Apart from the immediate cost savings from wiring installation, wireless technology offers the possibility for the realization of simpler and compact miniature bioreactor setup. Minibioreactors can for example be designed as a portable (i.e. hand-held) device powered by batteries. If this is achievable, it will make the idea of disposable microbioreactors very attractive. Moreover, multiplexing of minibioreactor setup will also be less complicated as no additional wiring is needed for networking of new reactors. Wired system tends to make a platform containing multiple miniaturized reactors look disordered and complex [5]. By establishing a Wi-Fi connection via wireless router, the bioreactor setup can be assessed/controlled from any wireless device e.g. smartphones, PC tablet, etc. with an internet connection. Such flexibility would also eliminate the need for the presence of laboratory personal when running lengthy bioreactor experiment as the reactor can be monitored merely through a secured internet connection. Literature shows that efforts have been made for wireless control of shake flasks [6] and lab scale bioreactor systems [7] but not for a mini/micro-bioreactor systems.

The minibioreactor system presented in this work involves embedded controller, Bluetooth transceiver, a stirrer, temperature sensor, power supply, heater, and a milliliter-scale bioreactor. The minibioreactor is fabricated from poly (methylmethacrylate) (PMMA) and poly (dimethylsiloxane) (PDMS) polymers using a laser engraving methodology. Two experiments were conducted to demonstrate the operation of the minibioreactor through such a wireless connection. In the first experiment, the mixing performance of the device is tested under a number of conditions. In the second experiment, the temperature control feature of the device was evaluated. In addition, the potential of the wireless communication for online monitoring and control of minibioreactors is also discussed.

2 Methodology

2.1 Minibioreactor Design and Fabrication

Figure 1 shows the design and the schematic of the minibioreactor prototype presented in the work. It was realized to have a geometrical configuration that is similar to a commercial laboratory scale bioreactor setup. Briefly, the reactor consisted of two main parts: a 4.5 mm thick top lid and the main body of the reactor. The top lid containing fluid input and output ports (Ø 3 mm), interconnects for the Pt100 temperature probe (Ø 3 mm), space for mounting of a direct-current (DC) gear motor and connection for the heater (Ø 3 mm). It is a multilayer structure formed by a 1 mm thick PMMA polymer and a PDMS polymer. In the final assembly, the PDMS layer self-adhered itself to the PMMA polymer due to the nature of the polymer. The top PMMA layer provided the necessary mechanical support in the final assembly whilst the PDMS polymer forms a fluid seal.

The main body of the reactor contained the reaction chamber and the impeller for mixing. It was fabricated using a transparent PMMA tube with a diameter of 30 mm and a length of 25 mm; resulting in a reactor volume of approximately 18 mL. A circular shape PMMA structure with a thickness of 4 mm was thermally bonded into one end of the tube to form the base of the reactor where else a 'ring-like' structure was bonded to other end of the PMMA tube. This 'ring-like' structure features four through holes (Ø 2.5 mm) for easy assembling with the top lid via M2 screws. All sensors, actuators and fluidic connections were positioned closer to the reactor wall. A gap of about 1 mm is ensured to prevent any undesirable contact with the rotating impeller that rest in the middle of the reaction chamber.

All parts made from PMMA polymer were cut using the using a CO_2 laser engraving/cutting system (Trotec SP500, Australia), and parts made of PDMS polymers were fabricated using a soft lithography procedure. The latter was performed by curing a pre-polymer PDMS liquid solution in a mold made of PMMA at 70°C for 2 hours. The PDMS liquid solution was prepared by mixing 10 parts silicone and 1 part curing agent (Sylgard 184, Dow Corning). After thoroughly mixed and removal of all bubbles, the mixture was poured into the mold. Once cured, the PDMS polymer was left to cool in room temperature before removing it from the mold with great care.

2.2 Minibioreactor Operation and Connectivity

The reactor operation consisted of stirring and temperature control of the reactor content (Figure 1). Mixing for the reactor is achieved by means of a customized 4-blade impeller made of PMMA polymer. The arm of the impeller is 17 mm in length and is held by a stainless steel shaft (Ø 3 mm) that is connected to the DC gear motor on the top lid. Water can be loaded into the reactor using the fluidics interconnects on the top lid.

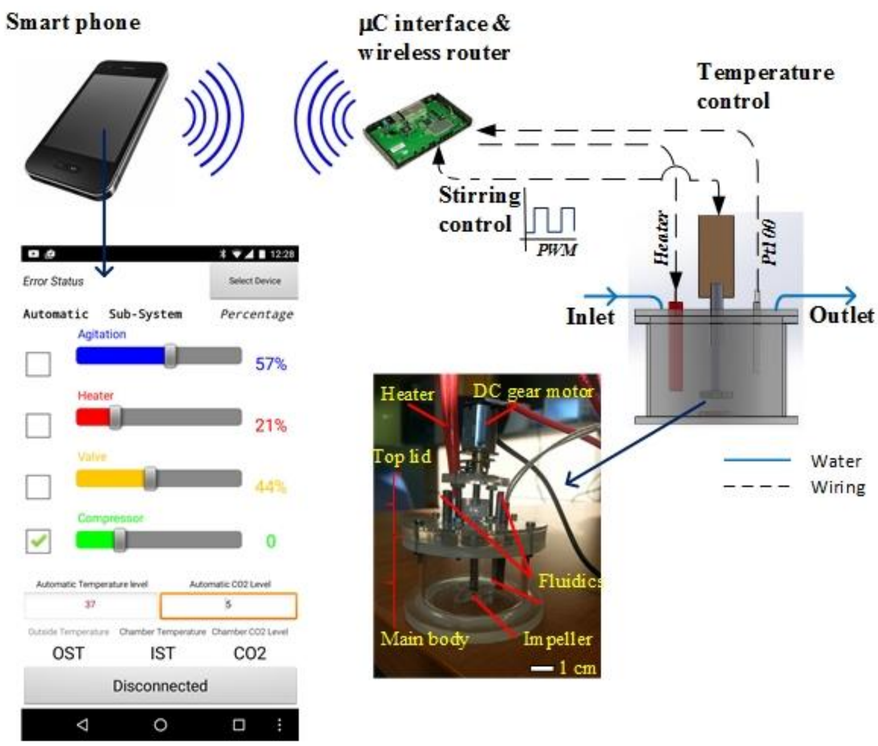

Figure 1: THE MINIBIOREACTOR SYSTEM AND CONNECTIVITY INCLUDING THE IMAGE OF THE MINIBIOREACTOR DESIGN AND THE INTERFACE OF THE APPLICATION DEVELOPED (BOTTOM LEFT CORNER).

Finally, the temperature of the reactor content was measured by using a miniature size Pt 100 probe whilst heating was achieved using a ceramic cartridge heater (12V, 30W). An Android application is developed and then deployed onto a smart phone to control the operation of the minibioreactor system. Wireless communication was achieved through the Bluetooth transceiver embedded on the controller platform.

Details of the wireless communication design and software interface are discussed in the next section. Numbers of experimental work was conducted to validate the reactor operation through wireless communication. These included temperature set-point tracking experiment, mixing efficiency and system stability.

3 Results and Discussions

3.1 Hardware Specifications and Software Interface

A low cost, wireless, miniaturized autonomous circuit, has been designed to control and regulate different aspects of the minibioreactor prototype. Detailed design of the circuit board is shown in Figure 2. The circuit board design includes a programmable microcontroller, 3 motor drivers, power regulators, a high accuracy (±0.3°C) temperature sensor input and the bluetooth module receiver. The ATmega328P 8-bit microcontroller was selected for the reactor operation because it provides high performance, low power consumption with 6 different PWM channels. It also has the capabilities to communicate through serial and I2C. One of the tasks of the microcontroller is to control various PWM signal for the control of the agitation system, heater, valve and compressor. Coding for the control of the entire reactor operation was done through C-language code, whereby the microcontroller autonomously regulates the temperature and the agitation in the minibioreactor into specified conditions designated by the user.

Each component that requires a PWM signal needs a specific voltage and current, therefore different motor drivers were used. The TB6612FNG is a cost effective, high amp rating, small 2 channel motor driver used to amplify the PWM signals from the microcontroller. The main reason the TB6612FNG was used is because it has a variable output voltage level between 3v to 15v, and an average output current of 1.2 amps. This meant all components could use the same type motor diver, keeping the design simple. The 3 power regulators are used to supply the components with the specified voltages. The MC7805 linear power regulator converts 12v to 5v, powering components such as the bluetooth module. The two LF33CDT linear power regulators converts 12v to 3.3v and power components such and the compressor and microcontroller. Two of the LF33CDT are used to split the current draw required for each, creating less heat. The PT100 is a highly accurate, commonly used temperature sensor that offers a large range from -200 to +850°C. The sensor is used in combination with the heater to create a closed loop temperature system.

In order to control the device, a bluetooth HC-06 module wireless receiver was used. Using the HC-06, it offers the capacity for control over the minibioreactor without physically entering the environment where the minibioreactor might be. This is very useful when the minibioreactor is in sterilized environment. Through the HC-06, the minibioreactor systems can be controlled and adjusted using a serial terminal interface on any smartphone with Bluetooth 2.0.

3.2 Reactor Performance via Wireless Communication

The reactor performance was evaluated by investigating system capacity to control both the temperature and the agitation speed at the desired set-point values. Additionally, mixing quality of the reactor was also assessed by determining the time needed for a concentrated red dye to be completely mixed (i.e. fully dispersed) at various agitation speed. Figure 3 shows the results for the PID (Proportional-Integral-Derivative) temperature set-point tracking experiment. Based on the results attained, it was found that it took nearly 15 minutes to reach a temperature set-point value of 37°C from a room temperature which was measured to be about 26°C. The response time of the controller was relatively quick however due to the small step change applied in the algorithm, the system heated up rather slowly. Nevertheless, no temperature overshoot was detected which justified why a small step changed was selected and the reactor

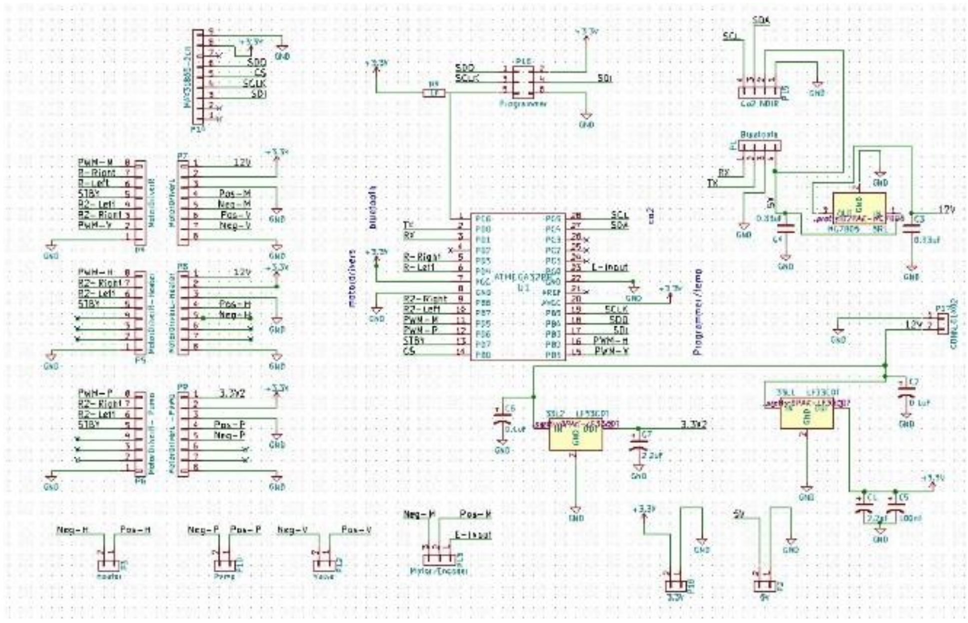

Figure 2: SCHEMATIC OF THE CIRCUIT BOARD FOR THE MINIBIOREACTOR PROTOTYPE.

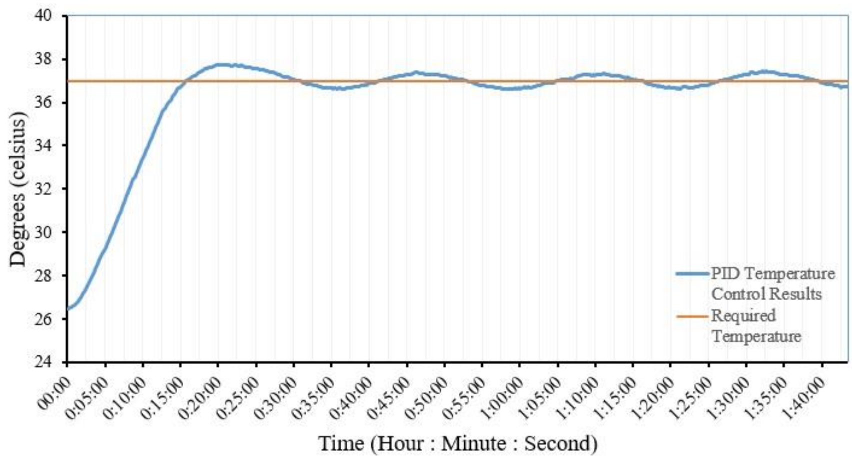

Figure 3: TEMPERATURE PROFILE OBTAINED THROUGH THE SET-POINT TRACKING EXPERIMENT.

temperature was successfully maintained within ±0.3°C of the set-point values. The workability of the controller was also very stable as the entire operation remained in steady state without any disturbance over a period of 48 hours.

Figure 4 shows the results of the mixing efficiency of the reactor. The experiment demonstrated that the system can operated within a reasonable range i.e. between 30 and 90 rpm. As can be clearly seen that at lower agitation rate, it took approximately 107 seconds to reach a complete mixing. However, as the agitation rate was increased, the mixing time was improved exponentially and a complete mixing is achievable within 6 seconds. It was also found that at low agitation speed, a partial volume of the dye sediments at the bottom of the reactor. Obviously, this is not desirable and can be overcome by increasing the agitation rate. The operating range for the agitation

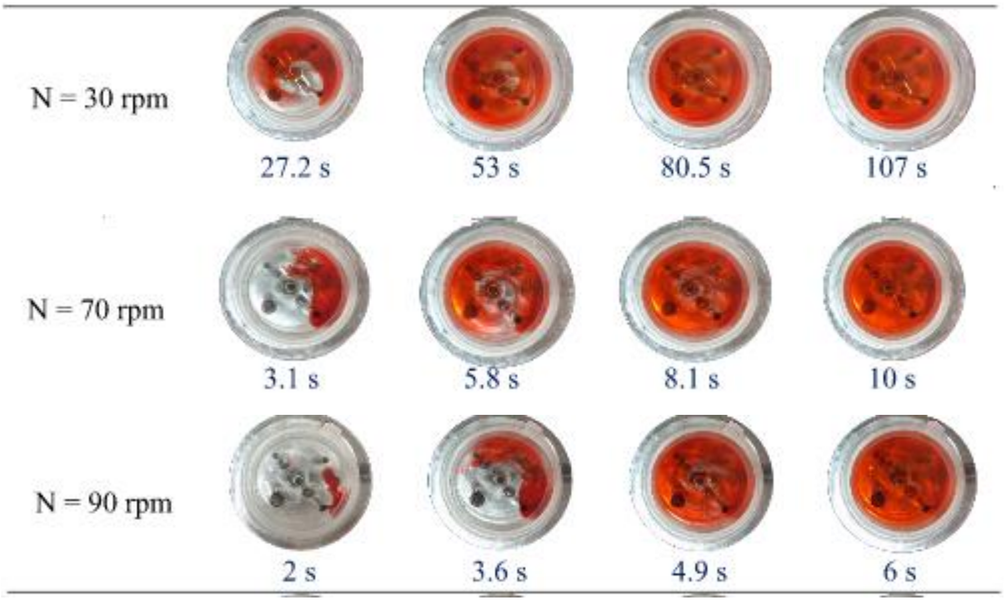

Figure 4: IMAGES ILLUSTRATING THE MIXING QUALITY OF THE REACTOR AT THREE DIFFERENT AGITATION SPEED.

of the reactor can be modified (if necessary) to work at any desirable range simply by changing the DC motor that drives the impeller.

4 Conclusions

A wireless controlled minibioreactor system was successfully designed and demonstrated. The temperature of the reactor can be regulated and controlled within ±0.3°C of the setpoint values and a good mixing of the reactor (indicated by a fast mixing time) was also attained. The minibioreactor system was also able to run up to a period of 48 hours stably without any undesired disturbance. The capacity of controlling a reactor operation via wireless communication is indeed beneficial particularly if ones were running multiple reactors at a time where human supervision is limited.

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