BIOREACTOR FOR MECHANICAL CELL STIMULATION
Concept and Design

J. G. Rocha, V. Correia
Industrial Electronics Dept., University of Minho, Campus de Azurem, 4800-076 Guimaraes, Portugal

J. L. Gomez Ribelles
Centro de Biomateriales, Universidad Politécnica de Valencia, 46022, Valencia, Spain.
Regenerative Medicine Unit, Centro de Investigación Príncipe Felipe, Autopista del Saler 16, 46013 Valencia, Spain
CIBER en Bioingeniería, Biomateriales y Nanomedicina, Valencia, Spain.

S. Lanceros-Mendez, A. Pitaes
Physics Dept., University of Minho, Campus de Gualtar, 4700-058 Braga, Portugal

Keywords: Bioreactor control, Cell cultivation, Three-dimensional scaffold.

Abstract: Mechanical stimulation plays an important role in improving cell growth in the skeletal system, for example. In this article we describe a bioreactor in which cells in a three-dimensional scaffolds are stimulated by cyclically applied mechanical loads. The objective of this study is to develop a custom-designed bioreactor capable of applying controlled compressive loads to a cell-encapsulating scaffold. Its working principle is based on an innovative design of a feedback controlled electromagnetic actuator, which allows the application of compressive forces to the samples and at the same time, it allows the measurement of the produced displacement.

1 INTRODUCTION

In vivo, skeletal cells such as osteoblasts and chondrocytes are subjected to mechanical stimulation imposed by muscle contraction and body movement (lee, 2009). It has been demonstrated mechanical stimuli play a role in improving cell growth in the skeletal system (Nugent-Derfus, 2007), (Kisidaya, 2004), (Cooper, 2007), (Garvin, 2003). Many research groups have developed bioreactors to stimulate cell-seeded, three-dimensional scaffolds. The mechanical environment influences tissue growth and development. The proper mechanical force that can produce correct bone tissue is a key issue in order to being able to develop bone tissue in vitro. Static constant mechanical loads have little or no effect in cell growth and proliferation, but cyclically applied loads do have profound effects (Meyer, 2001) (Guldberg, 2002).

Several models of bioreactors have been developed for the stimulation of three-dimensional scaffolds of bone and cartilage (Godstein, 2001), (Botchwey, 2001), (Hillsley, 1994). All of these bioreactors are satisfactory for the growth of tissues but do not include the possibility of applying cyclic loads that might be important in the case of skeletal cells.

The main goal of this work is to develop and test a bioreactor in which cells in three-dimensional scaffolds are stimulated by cyclically applied mechanical loads. The objective of this study was to develop a custom-designed bioreactor capable of applying controlled compressive loads to a cell-encapsulating scaffold.

2 BIOREACTOR DESCRIPTION

Figure 1 shows a drawing of the mechanical actuation system of the bioreactor. It is composed by an iron core, where four coils are placed. Two of the coils will produce magnetic fields that displace the

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core, while the other two work as sensors to feedback the core position to the controller.

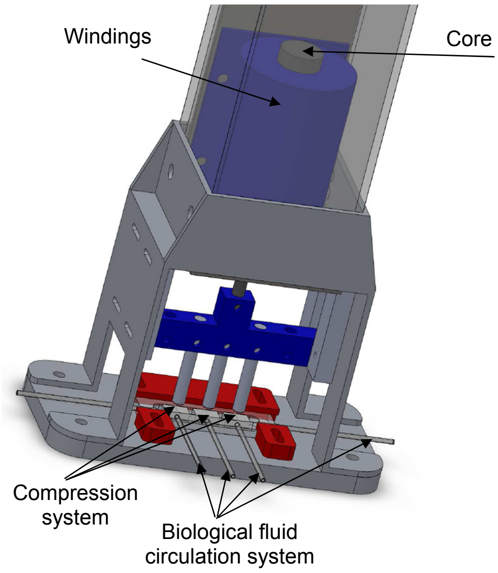

Figure 1: Bioreactor design.

Figure 2 shows a detail of the bioreactor, where the compression and circulation systems are seen in detail.

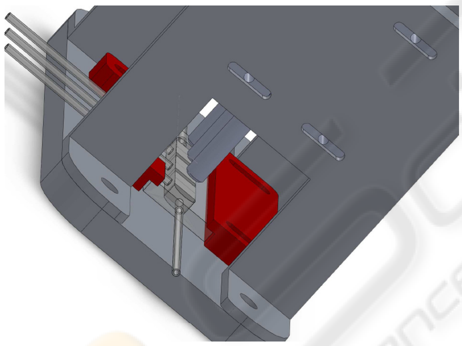

Figure 2: Detail of the actuation system of the bioreactor.

Figure 3 shows the front-view of the bioreactor actuating system.

3 CONTROL ELECTRONICS

The general system architecture is a master-slave structure since there are two control units: a high-level one (master) and a low-level one (slave).

The high-level control unit, central processing unit or simply the master is responsible for supporting the user interface, exchanging and processing the necessary information between the user and The low-level control unit and storing the measurement data. It is based on a computer (PC). The low-level control unit or slave main component is a microcontroller. It is responsible for acquiring the data from the sensor and controlling the stress applied to the samples under test.

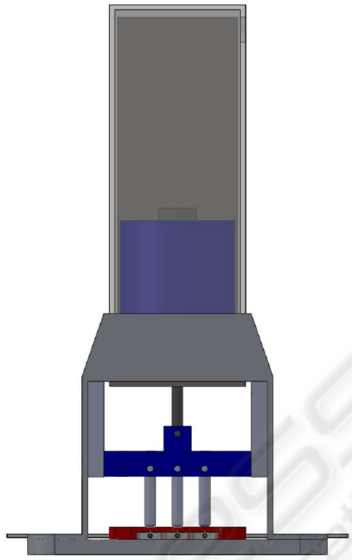

Figure 3: Bioreactor actuation system front view.

Figure 4 shows the system architecture.

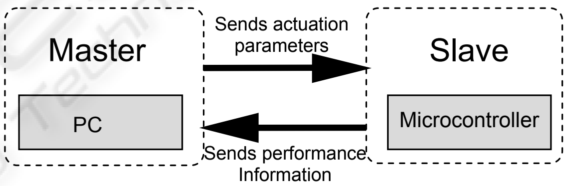

Figure 4: System architecture. The PC operates as master and the microcontroller operates as a slave. The microcontroller functionalities are programmed by the PC, which receives the measured data.

3.1 Master Block

In order to obtain the optimal operation point, that is, when the cellular growth approximates the one that happen in the human body, it is necessary a generic system able to change the operation parameters during the experiments. In this way, in the proposed system, the user is able to control the following parameters:

- Initial date and time;
- Final date and time;
- Shape of the stimulus (on/off, sine wave, pulse, ramp);
- Holding time;
- Active time;
- Displacement;

- Oscillation amplitude and frequency;
- Number of oscillation cycles;
- Standing Time (to generate a sequence);
- Up time

Figure 5 shows the flow chart of the master algorithm.

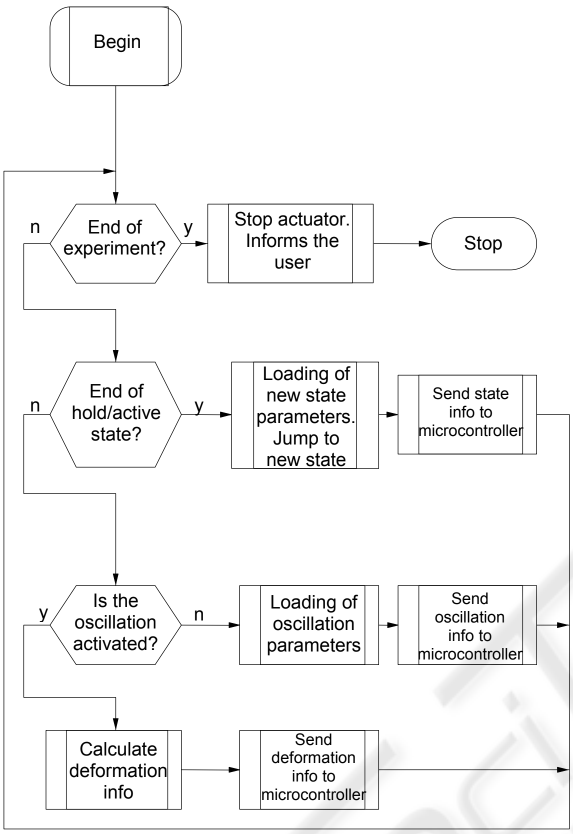

Figure 5: Master control flow chart.

It consists in a main loop where three decisions must be taken. The first one is related to the end of the experiment. If it happens, the actuator must be stopped, the user must be informed and the program stops. The second one is related to the end of a hold or active state. In this case, the new state parameters must be loaded and sent to the slave block. The third one is related with the oscillation of the actuator. If the oscillation is not activated, its parameters must be loaded and sent to the slave block. If the oscillation is activated, the next deformation step must be calculated and sent to the slave block.

Figure 6 shows a picture of the front panel of the control application.

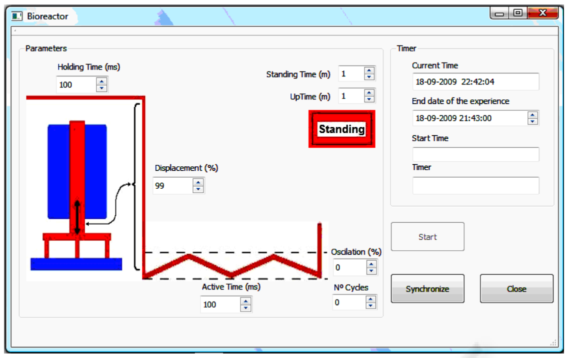

Figure 6: Front panel of the control application.

3.2 Slave Block

The Slave block is constituted by a microcontroller, a power switching circuit based on a H-bridge, two actuators (solenoids) and a measurement system whose working principle is based on a LVDT (Linear Variable Differential Transformer). Fig. 7 shows the block diagram of the whole system.

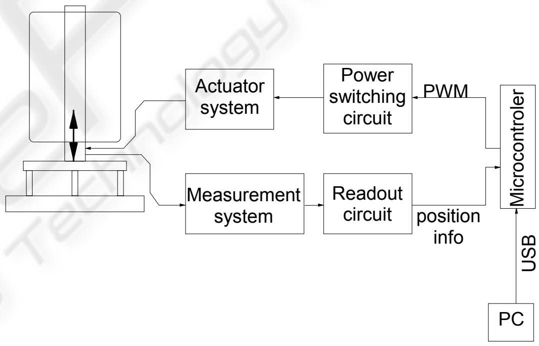

Figure 7: Control system of the mechanical movements of the bioreactor.

The main goal of this system is to generate a set of mechanical stimulus in a cellular cultivation region. The microcontroller receives the desired stimulus parameters from the computer and reads the actual displacement of the actuator from the measurement circuit. With that information, it calculates the duty-cycle of the PWM and sends it to the power switching circuit, in order to produce the correct voltage to the actuator.

The displacement is the most important parameter of the system, but the mechanical actuator only allows the control of the mechanical stress applied to the cultivation cell. In this way, the control of the displacement is made in a feedback loop, where a displacement sensing mechanism is introduced.

The displacement actuator is then constituted by four windings: two primary windings with a small number of turns and two secondary windings with a larger number of turns. The primary windings are excited by the power switching circuit in order to produce the displacement of the core. The secondary windings use to advantage the switching frequency of the PWM in order to measure the displacement, as it will be described in the following paragraphs.

A block diagram of the displacement transducer and respective signal conditioner is shown in Figure 8. The circuit consists of a PWM wave generator to drive the primary windings, a conditioner circuit for each secondary winding, a differential amplifier that determines the difference between the voltages of the secondary windings and a summing amplifier, which determines the sum of the secondary winding voltages.

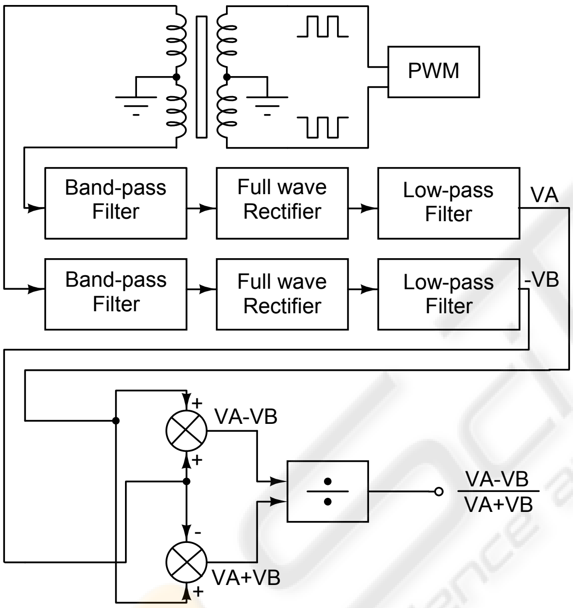

Figure 8: block diagram of the displacement transducer and signal conditioner.

The PWM wave that is applied to the primary windings, where besides producing the displacement of the core, it induces in the secondary windings a voltage whose magnitude difference is proportional to the core position. These two secondary voltages are band-pass filtered for the fundamental frequency of the PWM generator. So, the output from the band-pass filters consists of a pair of sine waves whose amplitude difference, (VA–VB), is proportional to the core position.

In order to operate in a ratiometric principle and thus eliminating the errors associated to non-constant amplitude of the PWM signal, the circuit computes (VA-VB)/(VA+VB).

The signals VA and VB are firstly rectified and low-pass filtered. A signal with precise frequency is not necessary because the inputs are rectified and only the sine wave carrier magnitude is processed. There is also no sensitivity to phase shifts between the primary excitation and the secondary outputs because synchronous detection is not employed. Then, the signals are applied to the differential amplifier and to the summing amplifier. The ratio (VA-VB)/(VA+VB) is performed by the A/D (analog to digital) converter. The (VA-VB) signal is applied to its input and the (VA+VB) signal is used as reference voltage for the A/D conversion. Finally, The digital signal is read by the microcontroller.

4 CONCLUSIONS

In this article, it is described the design of a bioreactor to apply controlled mechanical stimulation to cell cultures. The bioreactor consists on a mechanical loading actuator, experimental chamber, and control system. The actuator is based on an innovative design of a feedback controlled electromagnetic actuator, which allows the measurement of its own position. The control system is based on a master-slave architecture, where the master (computer) receives the user commands and sends the actuation parameters to the slave (microcontroller). This last one reads and feedback controls the actuator position.

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