Tissue Bioreactor

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Abstract

In order to effectively study vocal fold tissue outside of the human body, a bioreactor has been constructed that appropriately stimulates vocal fold tissue to behave as naturally as possible. Such stimuli include vibration, tensile stress, and changing angles between the cell-seeded strips. Previous bioreactors have been created, both by researchers and a previous biomedical engineering student team, but did not sufficiently mimic the stimuli provided by the human body. To further mimic in vitro conditions, improved cellular substrates were created to simulate human tissue. Cells in this new substrate will be subjected to multiple stimuli.

Motivation

The client is interested in using stimuli, allowing the alteration of cell growth, in order to perform research on injured and healthy human vocal folds. The previous bioreactor simulated the vocal fold environment, but did so poorly. By subjecting the cells to more stimuli, controlling the stimuli with a computer, and employing a better cellular substrate, the bioreactor will much more accurately simulate the human vocal fold environment.

Design Criteria

The bioreactor needs to fit into a standard-sized incubator and have sterile disposable parts and/or permanent parts that can be sterilized. The angle between each pair of strips needs to have the ability to change, and each pair of strips needs to vibrate and stretch as well. The motors need to be programmable and controlled by a computer. In addition, an improvement upon the previously used cellular substrate, Tecoflex, needs to be obtained and tested for its ability to help re-create the *in vivo* vocal fold environment.

Background

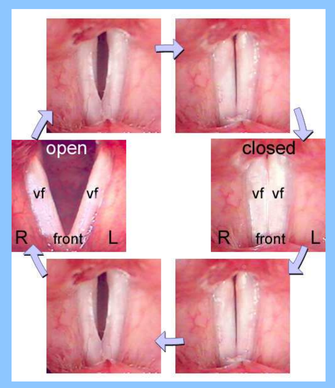

Figure 1: Photographs illustrating the movement of healthy vocal folds when speech is occurring.

A bioreactor is a system or device that supports a biological environment by using appropriate stimulation. The environment that will be simulated is that of the human vocal folds. The vocal folds are a pair of elastic tissue found horizontally inside the larynx (Figure 1). When air is exhaled through the lungs and reaches the closed vocal folds, the folds open and close many times per second, with vibrations that can be manipulated by the throat, mouth, and lips into speech. This environment will be simulated by the multiple stimuli being employed in the bioreactor.

Problem Statement

The previous bioreactor design was able to vibrate two pairs of cell-seeded strips under tensile stress, but had design flaws that needed improvement. The new design addressed these flaws by keeping the bioreactor leak-proof, subjecting the cells to more stimuli, and allowing the equipment providing the stimuli to be controlled by a computer. The goals include finishing the design and fabrication of the new model, obtaining a substitute for the cellular substrate, and testing the bioreactor and cellular substrate for optimal design and operating conditions.

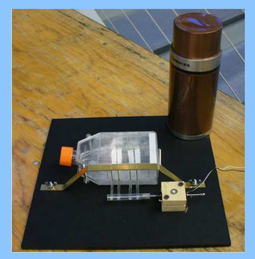

Figure 2 displays the previous bioreactor. The cell seeded strips can be seen inside the T-flask, as well as the actuator encased in the wooden block outside the T-flask.

Final Design

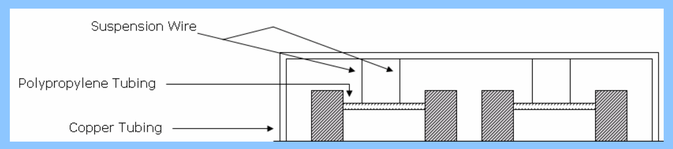

Figure 3

Figure 3 illustrates the design used to counteract the moment reaction caused by the weight of the aluminum arms connected to the vibration bar. By mounting a bar across the length of the bioreactor, wires were extended down from the bar to hold the polystyrene tubing up.

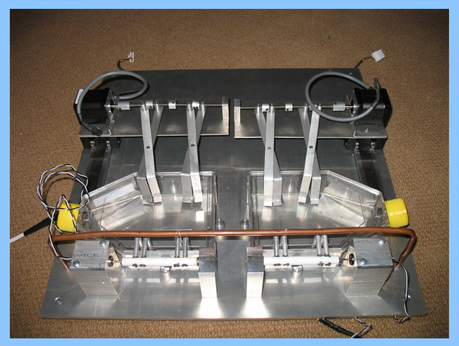

Figure 4

Figure 4 shows the completed working prototype. The actuators, encased by the aluminum blocks, drive the tubing, producing vibrations. The linear stepper motors, attached to the long rails in back, stretch the cellular substrate, located in the T-flask. The rotary stepper motors, located on top of the linear motors, cause the angle to change, producing a wave motion when the strips come in contact.

Final Design: Cellular Substrate

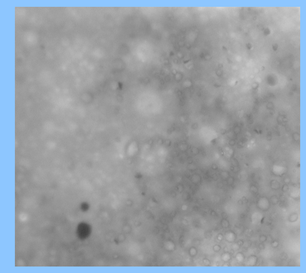

Figure 6 displays 3T3 NIH fibroblasts seeded onto hydrogel layered on top of Tecoflex

In order to accurately imitate the vocal fold, a material that has similar properties needed to be made. We used Tecoflex SG-80A, a very elastic material, in conjunction with hydrogels obtained from the University of Utah to mimic the vocal folds themselves. The Tecoflex is cut into a thin strip (approximately 10 mm wide x 100 mm long x 2 mm thick) and acts as the elastic substrate while the hydrogel is poured on top. The hydrogel is then seeded with cells and the strip is placed in the bioreactor.

Testing

In order to see if cells would grow on the material, hydrogel was put on top of a Tecoflex strip and cells were seeded on top of this layered structure. After incubation of about 15 hours there still were living cells, which shows that cells can survive on the material. Testing was also conducted on the ability of the bioreactor to vibrate the Tecoflex strips. By using a high speed camera and a stroboscope, it was demonstrated that in air at about 180 Hz the vibrated Tecoflex strips could be made to touch and produce a wave similar to that seen in human vocal folds. Also, the test was conducted with the strips under water and similar results were obtained, except there was a decrease in vibration due to drag.

Future Work

Future goals include incorporating a pressure stimulus into the design. Work will continue on the fabrication of the cellular substrate and its installation in the T-flask. Feedback from the client will be used to make any other improvements.

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