Calculations of the Acceleration of Centrifugal Loading on Adherent Cells

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Abstract. Studies have shown that the morphology and function of living cells are greatly affected by the state of different high acceleration. Based on the centrifuge, we designed a centrifugal cell loading machine for the mechanical biology of cells under high acceleration loading. For the machine, the feasibility of the experiment was studied by means of constant acceleration or variable acceleration loading in the Petri dish fixture and/or culture flask. Here we analyzed the distribution of the acceleration of the cells with the change of position and size of the culturing device quantitatively. It is obtained that Petri dish fixture and/or culture flask can be used for constant acceleration loading by experiments; the centripetal acceleration of the adherent cells increases with the increase of the distance between the rotor center of the centrifuge and the fixture of the Petri dish and the size of the fixture. It achieves the idea that the general biology laboratory can conduct the study of mechanical biology at high acceleration. It also provides a basis for more accurate study of the law of high acceleration on mechanobiology of cells.

1. Introduction

With the development of aerospace industry, the performance of fighter aircraft has been greatly improved. As one of the most common and strongest stress factors during the process of fighter flight, the high positive acceleration (forward acceleration, + Gz) produced by the modern high performance fighter has the characteristics of rapid growth rate (6G/s), high +Gz value (+9 Gz) and long acting time (15~45 s), which has exceeded the physical limits of the human body [1]. Research shows that repeated high positive acceleration (+Gz) exposure of the brain can produce similar effects of ischemia and reperfusion, and thus cause brain injury in the continuous high acceleration condition [2]. Li Wenbing et al. [3] used adult male Wistar rats for testing, and the growth rate of +Gz was 1 G/s, the peak values were +6Gz and +10Gz, the peak time was 3 min, 30 min interval, and repeated 5 times. It was found that the liver cells were arranged in disorder, irregular in shape, and the cell’s space was not clear, and the +10Gz group was more significant than the +6Gz group after loading. The effects of shear stress on the morphology of cells are widely founded in organisms. Liu et al. [4] applied shear strength of 1.2 Pa, 1.6 Pa and 1.9 Pa to osteoblasts of adult rats, 1 hour later, they found the morphology of cells was spindle shaped in the two groups of 1.6 Pa and 1.9 Pa. Furthermore, the results of quantitative analysis of cell morphology index also showed that the overall morphology of the cells can be extended along its long axis under suitable shear stress, whose arrangement direction also occurred from the random positioning at the beginning to achieve unity in the end.

Over the last ten years, the biomechanical research has reached the level of cell and molecule, and gradually formed a new research field, "mechanobiology", which is the study of the effects of
mechanical environment on the health, disease and injury of organisms. It studies the mechanism of mechanical signal perception and response to elucidate the relationship between the mechanical processes and biological processes such as growth, reconstruction, adaptive change and repair, then to develop a new therapeutic or diagnostic technology [5].

In this paper, we design and manufacture a kind of centrifugal loading machine, therefore, the current culture conditions and the commonly used culture cell containers in the general laboratory can be used to carry out centrifugal high acceleration loading on the cell. In addition, we can describe the change of the high acceleration with the change of cell’s position by changing the size of the high acceleration loader quantitatively. The above study provides a new idea and theoretical basis for studying the mechanical and biological characteristics of cells.

2. Design and application of centrifugal loading machine
Mechanical factors exist in all the microenvironment of cells, when we construct an engineered tissue in vitro, the lack of mechanical stimulation can lead a result that cells of seed can’t play its function fully, thus, the biological activity of the tissue is low. However, the bioreactor can simulate a variety of mechanical environments easily, so as to promote the function of seed cells in biological materials [6].

Centrifugal high acceleration loading machine, which is a common instrument used in the laboratory to study the high acceleration mechanical environment, and the device is placed indoor, for the device has the advantages of good working environment, small occupied space, accurate control of the dynamic experiment process, and its applicability is very strong.

![Figure 1. Centrifuge rotor.](image1)

![Figure 2. Fixture of Petri dish and Culture flask.](image2)

The centrifugal high acceleration loading machine is mainly composed of a rotary motor, a principal axis, a rotor, an inner shell of a centrifugal loading machine and a casing. The rotor is mainly composed of a rotor body and a rotor cap, which is made of aluminum alloy for aviation. The rotor body is provided with four rectangular grooves, which are mutually corresponding to each other. The grooves are used for placing the Petri dish fixture and the culture flask (Figure 2). The overall structure of the centrifuge rotor is shown in Figure 1. We have to balance the rotor after the processing is completed, which is the so-called centrifugal balancing, and centrifugal balancing refers to the whole rotor in geometry symmetry and mass balance, thus to ensure homogeneous distribution of the rotor load [7]. Then we carry out the loading experiment.

We used grey PVC to manufacture the Petri dish fixtures (Figure 3). There are six grooves in one fixture body (Figure 3A). Six Petri dishes whose specifications are 35mm can be fitted with it accordingly. We sealed the Petri dish with medical tape to prevent the leakage of the culture medium. The sealed Petri dish can be placed in the Petri dish fixture, and thus can ensure that the experiment can be carried out smoothly. Then we cover the lid of Petri dish fixture and put it into the groove of the rotor body.
In this paper, the Petri dish can be loaded on the Petri dish fixture directly; at the same time, it can also be used to test the acceleration of cells under different conditions when the cells were loaded with centrifuge, which makes the study of mechanical biology in the condition of high acceleration more convenient.

3. Study on the centripetal acceleration of cells in different positions

The Petri dish is in the same plane in the fixture, and the distribution of the adherent cells is different in different positions. If the cells are loaded with centrifugal high acceleration at the position of point A in Figure 6, the centripetal acceleration of the cell \((a_{n1})\) was perpendicular to the culture dish. If the cells are loaded at point B in Figure 6, the centripetal acceleration of the cell \((a_{n2})\) can be decomposed into the acceleration along the inner wall of the culture dish \((a_{1})\) and the acceleration perpendicular to the inner wall of the culture dish \((a_{2})\). We can change the size and position of the fixture to describe the change of the acceleration of the cells, which provides a reference for the future study of mechanical biology of cells.

3.1. Change of centripetal acceleration

We can see that the centripetal acceleration of cells is different in different locations (Figure 4). With changing the distance between the center of the rotor and the fixture \((L_1)\) or the distance between A and B \((L_3)\), the centripetal acceleration of the cells in different positions can be calculated quantitatively.

In a rotating state, the magnitude of centripetal acceleration of a point is
We take the range of $L_1$ from 10mm to 50mm. The graph of $a_{n1}$, $a_{n2}$ curve with the change of $L_1$ is shown in Figure 5. We can know that $a_{n1}$ and $a_{n2}$ have corresponding changes with the increase of $L_1$, and $a_{n1}$ is linear increase, $a_{n2}$ is nonlinear increase. With the change of distance, the direction and the magnitude of the acceleration of the cells in different positions are changed, and with the increase of $L_1$, the gap between them has a narrowing trend.

4. Conclusion
The dish fixture and/or the flask could be used for the constant acceleration ((0-40) ×g) or variable acceleration loading of the centrifugal loading machine. The Petri dish and/or the culture flask are in the same plane in the fixture, and the distribution of the adherent cells in different positions is different. The centripetal and tangential acceleration of the cell increases with the increase of the vertical distance between the fixture and the rotating center. In this paper, we describe the centripetal acceleration of the cells with the change of position quantitatively, and this provides the basis for a more accurate study of the mechanical and biological law of high acceleration.

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