EFFECT OF SHEAR FLOW ON CULTURED CELLS

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Introduction

Biological cells are responsive to various environmental factors, such as electric [Hashimoto, 2012], magnetic and mechanical fields [Hashimoto, 2011]. The acceleration technique for orientation and differentiation of cells has been studied to make biological tissue *in vivo* and *in vitro*. Control methodology for orientation and differentiation of cells would be applied to regenerative tissue technology. In the present study, the effect of the shear flow on cultured cells has been studied *in vitro*.

Methods

A donut-shaped open channel system for the cell culture has been designed to apply a vortex flow on cells *in vitro*. A silicone rubber disk is attached on the inner bottom of the culture dish to restrict the space for the flow of the medium. The culture dish is placed on a plate, which inclines at 0.1 rad of the horizontal plane. The plate rotates to generate a swing motion. The motion produces a one-way clockwise vortex flow in the medium in the open channel. The continuously swinging plate is placed in an incubator.

In the other flow test with parallelepiped channel, the channel with the suspension was placed in the incubator for several hours (24 hours for the glass, 3 hours for PDMS) to make cells adhere to the plate of the chamber before the flow test. A constant flow of the medium was applied to adhered cells with the syringe pump. The behavior of cells on the plate of the chamber was observed with a microscope. Variation was made in the flow rate to vary the wall shear stress between 0.2 and 5 Pa calculated with a parabolic velocity profile.

Results

The experimental results with C2C12 (mouse myoblast) of donut shaped canal show that cells adhere adjacent to the inner circle in the donut shape of the canal. The cells proliferate in the vortex flow of the medium. The array of myotubes grows around the silicone disk, and the alignment curves to the radial direction. The longitudinal axes of cells orient to the direction perpendicular to the flow. The results with rat normal cartilage cell and with L929 (fibroblast-like, mouse connective tissue) were similar to that with C2C12. Flow stimulates differentiation of C2C12 to myotubes. The experiment with CS-2P2-C75 (porcine aortic endothelial cell) shows that cells adhere to the bottom of the culture dish adjacent to the inner circle of silicone disk, and that the area of adherence extends to the radial direction. Cells elongate to the spindle shape, of which long axis tilts to the circumferential flow direction. C2C12 are more adhesive to glass than rat cartilage cells, and L929 is more adhesive to polydimethylsiloxane than glass under shear flow in the parallelepiped channel.

Discussion

The experimental results show that cells are responsive to the shear flow, which governs orientation, exfoliation, and differentiation. Endothelial cells orient along the stream line, although myocytes orient perpendicular to the stream line. The mechanical stress is one of the interested points in the environment of cells, because they receive mechanical forces *in vivo*. A transmission point of stress to a specimen is important. In the most of studies, the stress is applied to a scaffold. When fixation between the cell and the scaffold is not enough, the stress is not transmitted to the cell. A flow can be used, on the other hand, to apply a stress field to a specimen. The whole specimen directly receives the shear stress in the shear flow.

References

Hashimoto *et al*, J Systemics Cybernetics Informatics, 9:1-7, 2011.

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