

Diamagnetically Induced Structure Changes in Cellular Assembly and Cytoskeletons

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We investigated the effect of a high intensity magnetic field of 14 Tesla on the morphology of smooth muscle cell assemblies, and observed that the shape of cell colonies extended along the direction of the magnetic flux. The phenomenon was most significant under magnetic fields of more than 10 Tesla, and an ellipsoidal pattern of the smooth muscle cell colony was clearly observed. The ellipticity of the cell colony pattern with a 14 Tesla magnetic field was 1.3, while that of 0 T ~ 8 T was close to a circle at about 1.0. The evidence that smooth muscle cells detect high density magnetic flux and thus change its cell orientation, was shown as a visible pattern of cellular colony. The speculated mechanism is a diamagnetic torque force acting on cytoskeleton fibers, which are dynamically polymerizing/depolymerizing during the cell division and cell migration.

Key Words: diamagnetism, living cell, cellular assembly, cyto-skeleton, diamagnetic anisotropy

1. INTRODUCTION

The biological systems and materials are composed of weak-magnetic materials, and it is difficult to prove an effect of magnetic fields on living systems. However, some effects on diamagnetic proteins were shown as a real effect of magnetic fields. An example of this phenomenon is the magnetic orientation, where protein polymers were oriented along the magnetic flux direction [1][2].

The present study is planned to observe the cell alignment under magnetic fields excluding the effects of boundary condition, which is set by the walls of cell culture flask.

2. METHODS

We prepare a condition for cells to form a circle colony pattern, and the contour of colony is observed to evaluate the primal mechanism for cell

alignment induced by external magnetic fields. The experiments are carried out with a 14 T superconducting magnet, and a dependence of cellular colony pattern on a magnetic flux density is shown.

The smooth muscle cells (A7r5, thoracic aorta; rat embryo) were purchased from Dainippon Pharmaceutical Co. Ltd.. The cell was cultured in Dulbecco's modified eagles medium (D-MEM) (Sigma Chemical Co. D6046) containing 10% fetal bovine serum (FBS) (Sigma Chemical Co. F9423) was kept in a CO₂ incubator. After four days of incubation in the CO₂ incubator, the cells were peeled from the culture dish and were suspended in the medium for the following cell colony settings.

50μl of cell suspension was gently put on the surface of the polystyrene flask (NUNC Co., Ltd., SlideFlask 170920) by using a micro-pipette. The suspension formed a circle pattern due to the surface

tension, and during static keeping, cells adhered on the surface of the bottom of the flask within one hour and formed a circle colony pattern, which was 6 mm in diameter. After one hour of static placement in a clean bench, the cell culture medium was flowed into the flask, and the flask was filled nearly 90% with the medium. The reason why the medium was so abundantly utilized was to avoid the parting water by magnetic fields. The cells proliferated and migrated, and the boundary of the circle colony showed an isotropic expansion in the case of a control experiment without magnetic field exposure. The colony patterns were observed after three days of incubations with and without the magnetic field exposure.

3. RESULTS AND DISCUSSION

Several hours after the cell adhesion on the bottom surface of the polystyrene flask, the smooth muscle cells of rats (A7r5) became spindle-like in shape. During one day of incubation, there were vacancies between the cells. In both cases with and without the magnetic field exposure, each of the cells oriented randomly. In 30 to 50 hours, most of the cells contacted each other and formed a local domain where the cells directed its long axis to a specific direction. The directions of the local orientation of cells were observed to be a random distribution or isotropic. The cell orientation directions in the flask were random during 50 hours of incubation both with and without the magnetic field exposures, however, all of the cells in the flask obtained a coherent alignment parallel to the magnetic fields within 60 hours under 14 T.

We formed a circle colony of cells in a polystyrene flask, and the cells were exposed to magnetic fields of up to 14 T for 3 days. The outline of the spot, which was exposed to a 14 T magnetic field for 60 hours, was an ellipsoidal pattern whose long axis was parallel to the magnetic field direction, while the cell assembly of the spot without magnetic exposure was a circle pattern.

The magnification of the magnetic field exposed spot (14 T) showed an aligned cell assembly. The cells in the control spot were randomly oriented. The cells in the flask reached a confluent state after 60 hours of incubation in a medium at 37 degrees centigrade. Control cells formed a micro domain of unidirectional oriented cells, however, all of the micro domains in the flask oriented randomly. It was also observed that the chain of micro domains formed a vortex pattern. In the flask exposed to

the magnetic field, the pattern of micro domains changed to a stream pattern, which was parallel to the magnetic field direction. The marked difference between the control and the magnetic field exposed spot was that the vortex pattern of the cells, which was observed in the control, was quenched under a 14 T magnetic field. In other words, the 14 T magnetic fields changed the randomness in the cells into an ordered assembly.

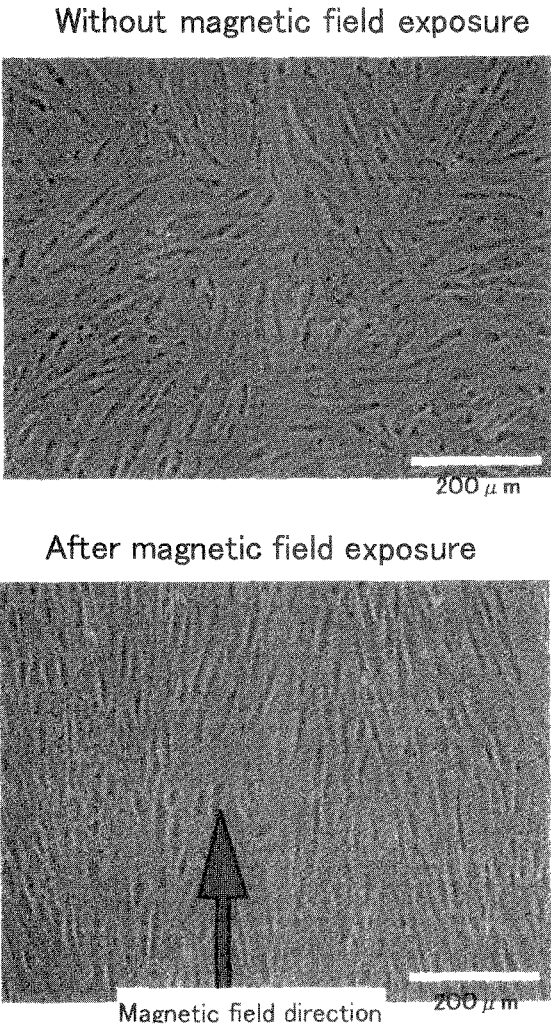


Fig. 1 Smooth muscle cell orientation in the flask after 50 hours of incubation with and without the magnetic field exposure at 14 T.

The assembly of smooth muscle cells has a self organizing process, where the cells contact each other and synergically form a mono-axial domain. This phenomenon is similar to the single magnetic domain formation by spin-spin interactions in ferromagnetic materials. Applied external magnetic fields B can induce cell domain-cell domain interactions and control the orientation of domains which have an accumulated magnetic moment M and $\Delta\chi B$ in strong magnetic materials and in diamagnetic cells with a diamagnetic anisotropy $\Delta\chi$, respectively.

The mechanism for the changing cell spot profile was both the orientation of cells and the result of cells moving toward the field direction. It was speculated that the cell components inside and in cell membranes obtained diamagnetic anisotropy and directed the cell orientation, changing the cell spot profile to an ellipsoidal pattern during cell proliferations.

References

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