京都大学	博士（　工　学）	氏名	Ian Torao Hoffecker
論文題目	Decoupling Interdependent Cytoskeletal Processes to Control Cell Adhesion Dynamics （互いに密接に関連する細胞内外の機構の個別操作による細胞接着挙動の制御）		

（論文内容の要旨）

This thesis addresses the issue of controlling cell behavior through its cytoskeleton. One might wish to control one of several behaviors such as migration, adhesion, or traction force alone, and it may be desired to control that particular variable without affecting the others. Three different approaches for achieving selective control of a particular cell behavior: cytoskeletal response-constraining patterned substrates to separate traction forces from rigidity sensing, membrane chemical modification with ssDNA-PEG-lipids to separate integrin-based adhesion from physical attachment to substrates, and pharmacological inhibitors for eliciting changes in relative adhesion between two different cell types. The thesis is divided into 6 chapters, the first of which is a general introduction.

The first chapter provides an introduction to the basic components of the cytoskeleton, their interactions, and their role in higher order cell mechanical behavior. Also discussed are challenges inherent in attempting to study a single aspect of the cytoskeleton when few variables can be modified independently of one another. Finally, the various tools and techniques that have been developed for controlling and studying cytoskeletal processes by isolating and selectively modulating individual components are discussed.

In the second chapter, an elastic gel substrate cell traction force examination is designed with discrete rigid islands, thus separating the behaviors of cells exerting traction forces positively reinforced by the sensing of cell-scale resistance from a cell that could hypothetically respond to the intrinsic micro-scale rigidity of the material. The findings support the notion that cells sense rigidity by exerting cell-scale forces and not by sensing intrinsic rigidity.

In the third chapter, mesenchymal stem cell (MSC)-islet-cell multicellular spheroids are formed and collective sorting into minimally contacting spheroidal compartments of MSCs and islet cells was observed. When the spheroids were formed in the presence of ROCK inhibitor, a drug that disrupts cytoskeletal force generation while simultaneously stabilizing cadherins, the spheroids sorted into core-shell shapes.

In the fourth chapter, glass substrates are patterned with ssDNA and then patterned with ssDNA'-bearing cells using an inkjet printer. The glass substrates permit microscopic observation which showed that adherent-type cells migrated away from points of DNA attachment by establishing integrin connections with adsorbed proteins.

京都大学	博士（　工　学）	氏名	Ian Torao Hoffecker

In the fifth chapter, restriction enzymes are used to sever the connections between substrates and cellular aggregates tethered by ssDNA-PEG-lipids. Both sequence specific restriction enzymes and nonspecific nucleases were used.

In the final chapter, the Hertz contact model is used to estimate the strength of ssDNA-PEG-lipid adhesion between cells. The estimation works by measuring the deformation of cell membranes when adhesive contacts are formed, and by knowing the elastic properties of cells, the adhesion energy can be inferred.

In summary, the thesis explores three types of cytoskeletal manipulation which can be used to constrain the behavior of cells in different contexts. By separating normally coupled cytoskeletal variables such as adhesion vs migration or traction force vs rigidity sensing, a variety of unique forms of cellular behavior can be controlled.