

Enhancement of cell attachment in dynamic culture environment

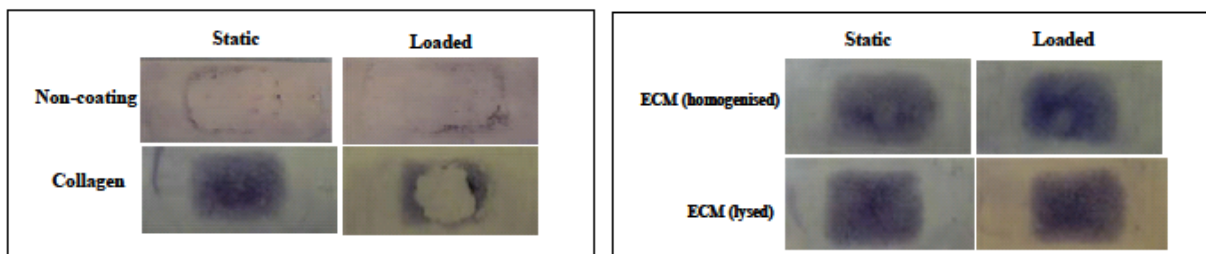
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INTRODUCTION One of the major challenges for tissue engineering is to create a favourable environment for the proliferation and differentiation of cells into functioning tissues. Since the initial interaction between cells and substrate (scaffold) is cell adhesion, the surface properties of the scaffold become a controlling factor in governing the success of tissue engineering. It has been demonstrated that coating the scaffold surface with single extracellular matrix (ECM) molecules such as collagen, fibronectin, laminin, or pre-soaking scaffolds in media improves cell seeding efficiency and spreading. In this study, we take a step further by coating substrates with whole extracellular matrix extracted from autologous cells. The effects of the coatings on bone cells cultured in dynamic environment were investigated.

EXPERIMENTS PLLA films with a thickness of about 10 μm were produced by solvent-casting. The film was then adhered to coverslips by a silicon adhesive. Primary human bone cells were used in the study. Three ECM solution were used to coat the PLLA coverslips: homogenised cellular solution from 0.4×10^6 cell pellet after filtered through a 10 μm sieve; lysed cellular solution from 0.2×10^6 cells by exposing the cells to water / NH_4OH solution; calf collagen type I at $7.5 \mu\text{g}/\text{cm}^2$ density. 0.1×10^6 autologous bone cells were seeded in each coverslip and statically cultured for 2 days then subjected to dynamic culture in a 4-point bending bioreactor with cyclically stretching for 1 hour. After 24 hour post-culture, the cells were fixed, stained by H & E and observed by a light microscope.

RESULTS AND DISCUSSION The bone cells showed dramatically different initial attachment ability to PLLA with various coatings. Using the cell shape as an indicator of adhesion, rounded cells indicating less adhesion and elongated cells indicating more adhesion, the relative degree of cell attachment after 1 hour incubation was as follows: PLLA control < collagen coating < homogenised cell coating \leq lysed cell coating. Strictly, after mechanical loading, the cells on the coating with whole ECM extract solution exhibited a more robust attachment with fewer cells detaching from the surface of the PLLA substrates (Figure 1).



CONCLUSIONS Autologous ECM extraction provides a better coating material for bone cells to adhere to a PLLA substrate. The coating produced tougher adhesion and enabled withstanding of mechanical stimulation at a physiological level, thus safeguarding and enhancing the mechanical signals to the cells.