USE OF A FIBER REINFORCED POROELASTIC MODEL TO PREDICT
THE MECHANICAL PROPERTIES OF TISSUE ENGINEERED CARTILAGE

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INTRODUCTION

Natural articular cartilage is composed of a charged solid matrix phase consisting of charged proteoglycan macromolecules and collagen fibres, an interstitial fluid phase, and an ion phase. The proteoglycan matrix is mainly responsible for the equilibrium compressive stiffness of cartilage, while the collagen fibres influence the instantaneous compressive and tensile response of the tissue. The interstitial fluid phase affects the transient response of cartilage during compression. Articular cartilage is known to have a poor ability to self-repair, partly due to the lack of innervations and vascularization. The use of autologous cartilaginous tissue, engineered in vitro and transplanted in vivo, represents an encouraging novel technique for cartilage repair. A key to success in this approach would be the achievement of an adequate in vitro maturation of the constructs into a fully functional tissue. Characterisation of the mechanical properties of the constructs at various stages of maturation appears necessary for a better evaluation of the biofunctionality of candidate scaffolds and culturing methods. Confined and unconfined compression tests are commonly used to evaluate the Young’s modulus (E), and the aggregate modulus (H), and permeability (k) of the constructs respectively. The role of collagen can be studied analyzing the stress-relaxation transients of unconfined compression tests, by using a fibre reinforced poroelastic model (Li et al, 1999). The model was successfully used to simulate unconfined compression experiments of natural (Li et al 1999) and degenerated cartilage (Korhonen et al, 2003).

The aim of this study was to investigate the ability of the fibre reinforced model to simulate unconfined compression experiments performed on constructs of collagen and chondrocytes at different times of in vitro culture and to identify parameters of difficult experimental evaluation for these constructs, such as the collagen stiffness (E_c) and the permeability.

MATERIALS AND METHODS

Experiments

Swine articular chondrocytes were enzymatically isolated from pig joints and expanded in monolayer culture. When confluence was reached, cells were re-suspended and seeded onto biological collagen type I and III scaffolds in vitro. Samples were retrieved from the culture after 18 and 38 days for further analysis. Samples were biomechanically tested by unconfined compression using an electromagnetic testing machine (Enduratec ELF3200) with a load cell of 22N, under displacement control, to evaluate the stress-strain curve and the Young’s modulus (E).

Model

The experimental stress-relaxation of the constructs was simulated implementing a fibril reinforced model (Li et al 1999) using a commercial FE package (ABAQUS v. 6.4, Hibbit, Karlsson&Sorensen, Inc. Pawtucket, RI, USA). The model was composed of extracellular matrix (ECM) and interstitial fluid. The ECM was supposed to be made of collagen oriented in different directions and proteoglycans. The radially oriented fibers were modeled as non-linear springs which resist tension only. The material parameters were the Young’s modulus and Poisson’s coefficient of the non-fibrillar matrix (E_m, v_m), the Young’s modulus of the fibers (E_f), the initial void ratio (e_0), the permeability (k).

ESSENTIAL RESULTS

The stress relaxation curves for constructs at time 0 (pure membrane), 18 and 38 days of cultures are shown in Fig. 1. The Young’s moduli evaluated from the equilibrium response were respectively 14, 4, and 5 kPa. The Young’s modulus was higher for the pure membrane than the constructs, this may indicate a faster scaffold degradation than the proteoglycans build up by chondrocytes. However the Young’s modulus at 38 days was higher than that at 18 days. The cellular ECM production may be revealed by the increase in peak to equilibrium ratio shown by the constructs both at 18 and 38 days with respect to the pure membrane.

The quantification of such a production in terms of biomechanical parameters (E, k) was evaluated by fitting the experimental stress-relaxation curves to the simulations of the poroelastic model (Fig. 2).

A good fit could be obtained by assigning the following values to the collagen stiffness modulus and the radial permeability: E values were 15, 31, and 67 kPa (0, 18, 38 days), k values were 6x10^-13, 2x10^-13, and 5x10^-13 (0, 18, 38 days). The increase in collagen stiffness and decrease in permeability at 38 days in respect to the pure membrane are consistent with the production of collagen type II and proteoglycans respectively, as confirmed by histological evaluation after immunostaining and Safranin-o staining (results not shown here).

In conclusion, the use of a fibre reinforced poroelastic model in combination with biomechanical experimental testing seems a valuable tool to analyze the biofunctionality of tissue engineered cartilage.

REFERENCES

Li et al, Clinical Biomechanics, 1999.

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