INTRODUCTION

Articular cartilage injuries cause pain and disability and lead into early onset osteoarthritis. Treatment of articular cartilage injuries has progressed significantly with the introduction of autologous chondrocytes implantation (ACI). ACI demonstrated long-term clinical benefit to patients for as long as 9 years of follow-up. Clinical application of ACI is laborious requiring arthroscopy of the knee, harvest of a periosteal flap from a secondary surgery site and suturing over the cartilage lesion. Use of the periosteal flap often leads to tissue hypertrophy requiring an arthroscopic intervention to remove the excess tissue. Delivery of the cells within a matrix eliminates the need for a periosteal flap and enables implantation by a minimally invasive procedure thus significantly simplifying surgery and reducing rehabilitation time.

BioCart™II is a new matrix-assisted autologous chondrocyte implant. The autologous cells, propagated with a unique growth factor variant, are delivered within a biocompatible and biodegradable scaffold made of human fibrin and hyaluronic acid. The scaffold is a porous open channel structure that enables immediate three dimensional distribution of the cells and thus contributing to its capacity to promote full thickness repair. The autologous cells cultured in the presence of the engineered growth factor variant that have an increased proliferation rate while maintaining their chondrogenic potential.

The combination of high quality autologous chondrocytes embedded within a three dimensional scaffold made of only natural components provides a novel, safe and effective therapeutic approach for articular cartilage repair.

METHODS

Fibrin hyaluronic acid scaffolds were prepared by mixing human fibrinogen solution with hyaluronic acid and thrombin to produce a clot. The clot is freeze dried to produce an open channel porous scaffold into which cells can be seeded. Human and porcine chondrocytes were obtained from cartilage tissue using enzymatic degradation by collagenase. The cells were then expanded in culture using human serum supplemented with a fibroblast growth factor (FGF) variant. The FGF variant was generated by rational design and site directed mutagenesis of the native FGF and the protein produced by bacterial fermentation. Chondrogenic potential of the cultured chondrocytes was determined by in vitro high density pellet culture. The pellet cultures were analyzed for Chondrogenic potential of the cultured chondrocytes was determined by the native FGF and the protein produced by bacterial fermentation.

RESULTS

Cells cultured in the presence of the FGF variant exhibit an increased proliferation rate compared with untreated cells. After 10 days in culture in a medium containing 10% human serum the number of cells obtained with the growth factor is 10 times the number of cells cultured without the FGF variant. Moreover, the number of cells obtained is 30 times more than culture using fetal bovine serum (FBS). Thus the time to obtain the required number of cells for implantation is significantly reduced compared to standard culture conditions using FBS. The chondrogenic potential of cells cultured for 4, 7, and 10 days in the presence of the growth factor were tested by pellet culture. Cells cultured for 4 days did not form a hyaline-like pellet, while cells cultured for 7 and 10 days formed pellets with hyaline like structure which express proteoglycans and collagen type II. Expression of collagen type I (Col-I) and type II (Col-II) were determined by real time PCR analysis. Col-II expression was significantly increased compared with Col-I in pellet culture indicating the regeneration of hyaline cartilage phenotype in the pellet culture. Pellet culture formed from chondrocytes cultured in the presence of the growth factor formed a much larger pellet and expressed more proteoglycans than pellet from cells cultured without the growth factor (Fig. 1).

Chondrocytes cultured in-vitro were seeded into the fibrin-hyaluronic acid scaffold to create tissue-like implants. Histological analysis of implants immediately post seeding demonstrate the chondrocytes are distributed throughout the scaffold. Expression of Col. II but not that of Coll. I was observed within the scaffold by IHC (Fig. 2).

DISCUSSION

We present a new articular cartilage repair implant composed of autologous cells embedded within a fibrin and hyaluronic acid scaffold. The rationale for the development of BioCartII is to create a safe and efficacious implant by using all natural components scaffold with cells of high chondrogenic potential well distributed within the scaffold.

Fibrin which is the natural scaffold for wound healing is used as the scaffolding material for BioCartII thereby mimicking the body’s natural healing process. The open channel and porous structure of the scaffold allows for an immediate three-dimensional distribution of the cells within the scaffold so to guarantee full thickness repair. Use of the FGF variant increases chondrocytes proliferation rate while maintaining their chondrogenic potential. This allows implantation of BioCartII within two to three weeks from retrieval of the cartilage biopsy and increases the regenerative potential of the implant.

BioCartII is now being tested in clinical studies for the treatment of femoral chondyle cartilage injuries.