Abstract

Recently tissue engineering has escalated much interest in biomedical and biotechnological applications. In this regard, exploration of new and suitable biomaterials is much needed for various tissue engineering applications. Silks are biopolymers that are spun into fibers by several arthropods such as silkworms or spiders, scorpions, mites and flies. Silk consists of mainly two kinds of proteins: hydrophobic, silk fibroin and hydrophilic, silk sericin protein. Silk based materials have been clinically used earlier as sutures and now days are being used in various biomedical and tissue engineering applications. Chitosan is a copolymer of D-glucosamine and N-acetyl-D-glucosamine and widely used for various biomedical applications due to its structural similarity with glycosaminoglycans, one of the main components of cartilage matrix. In this study, silk fibroin protein and chitosan polyelectrolyte complex porous three-dimensional (3D) scaffolds were fabricated and characterized as a potential biomaterial for cartilage tissue engineering applications. The use of cell-scaffold constructs is a promising tissue engineering approach to repair cartilage defects and to study cartilaginous tissue formation. Silk fibroin served as a substrate for cell adhesion and proliferation while chitosan has a structure similar to that of glycosaminoglycans, and shows promise for cartilage repair. These blended 3D scaffolds showed controlled pore size with good interconnectivity, porosity, tunable degradation, mechanical properties and antimicrobial properties. Additionally, these scaffolds showed good growth and attachment of feline fibroblasts. These macroporous blended scaffolds were subsequently tested for *in vitro* cartilage tissue engineering. The formation of cartilaginous tissue in silk fibroin/chitosan blended scaffolds was assessed with primary bovine chondrocytes and rat bone marrow derived mesenchymal stem cells cultured *in vitro* for 2-3 weeks. The blended fibroin/chitosan scaffolds supported good cell attachment and growth, proliferation and differentiation as compared to silk fibroin scaffolds alone. Chondrogenic phenotype was maintained in these scaffolds as indicated by Alcian Blue and Saffranin O histochemistry and relative expression of type II versus type I collagen. Biomechanical studies showed significantly higher static and dynamic stiffness at high frequencies in cell-seeded constructs than non-seeded controls. Chondrogenic differentiation was confirmed by up regulation of chondrogenic marker genes sox-9, col II and aggrecan. These wide ranges of applications using silk fibroin/chitosan blended scaffolds suggest its potential as a tissue engineering biomaterial.

Keywords: Silk fibroin, Chitosan, Scaffolds, Biocompatibility, Stem cells, Biomaterials, Cartilage, Tissue Engineering.