SYNTHESIS AND BIOCOMPATIBILITY OF POROUS COLLAGEN/CHONDROITIN SULFATE/HYDROXYAPATITE COMPOSITES FOR BONE TISSUE ENGINEERING

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INTRODUCTION: In recent years, biomaterials based on porous composites have proved to be effective scaffolds for bone regeneration. These scaffolds, built from synthetic or natural materials, serve as temporary substitutes for the native extracellular matrix and guide the proliferation and spread of seeded cells in vitro and in vivo [1-3]. The aim of our study was to develop porous bioactive materials composed of type I collagen (COL) , chondroitin sulfate (CS) and hydroxyapatite (HA) and to investigate their in vitro biocompatibility in a human dermal fibroblast culture system.

METHODS: In order to obtain porous composites, a type I COL solution (0.8%) was mixed with a CS solution (1.0%) and HA (Merck) in different weight ratios (1:0.5:1 (I); 1:0.5:2 (II); 1:0.5:4 (III)) and conditioned by lyophilization and sterilized by UV exposure. Materials obtained only from type I COL were used as controls.

Their ultrastructure was studied by scanning electron microscopy (SEM). Physico-chemical properties were assessed by porosity and collagenase degradation measurements. Fibroblasts isolated from human dermis (1.2 x 10^6 cells/cm^2) were cultured into porous composites for 48 h and 72 h. Cell viability and proliferation was assayed by measuring the mitochondrial dehydrogenase activity (MTT test). Cell morphology was evaluated by light microscopy after staining with Hematoxylin-eosin.

RESULTS: SEM micrograph of the COL-CS-HA composites showed a highly porous structure with small white aggregates of HA, non-homogenous scattered in the COL framework without any preferential orientation (fig. 1, left).

The average porosity of composite variants ranged from 84.75 % to 96.1 % and for COL scaffold was 97.33 %. In vitro biodegradability test demonstrated that the UV exposure increased the resistance of porous composites to collagenase digestion.

The biological test indicated a high cell viability for COL-CS-HA variant I (139.6%), followed by variant II (126.8%) and III (96.9%). Light micrographs showed that small cell groups adhered at the periphery or were included into the scaffolds (fig. 1, right). Cells showed a normal morphology characterized by a round or oval nucleus containing granular chromatin. The cell proliferation was superior in composite variants I and II compared to the control at both periods of time.

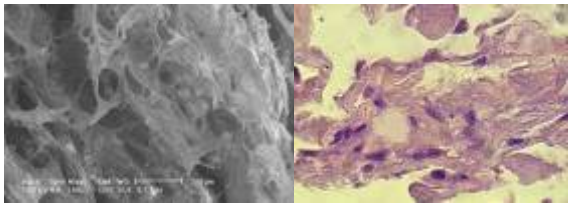

Fig. 1: SEM micrograph of COL-CS-HA variant I porous material (left) and light micrograph of cells migrated into the scaffold (right).

DISCUSSION & CONCLUSIONS: Porous COL-CS-HA composites with good physico-chemical properties were developed through integrating HA powder in a mixture of COL-CS. All three variants were biocompatible for human dermal fibroblasts in vitro. The high porosity of I and II variants allowed cell penetration into the composite materials. These porous composites can be used as potential bone regeneration scaffolds.

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