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## ACCEPTED MANUSCRIPT

New composite materials prepared by calcium phosphate precipitation in chitosan/collagen/hyaluronic acid sponge cross-linked by EDC/NHS

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#### **Abstract**

Nowadays, fabrication of composite materials based on biopolymers is a rising field due to potential for bone repair and tissue engineering application. Blending of different biopolymers and incorporation of inorganic particles in the blend can lead to new materials with improved physicochemical properties and biocompatibility. In this work 3D porous structures called scaffolds based on chitosan, collagen and hyaluronic acid were obtained through the lyophilization process. Scaffolds were cross-linked by EDC/NHS. Infrared spectra for the materials were made, the percentage of swelling, scaffolds porosity and density, mechanical parameters, thermal stability were studied. Moreover, the scaffolds were used as matrixes for the calcium phosphate in situ precipitation. SEM images were taken and EDX analysis was carried out for calcium and phosphorous content determination in the scaffold. In addition, the adhesion and proliferation of human osteosarcoma SaOS-2 cells was examined on obtained scaffolds.

The results showed that the properties of 3D composites cross-linked by EDC/NHS were altered after the addition of 1, 2 and 5% hyaluronic acid. Mechanical parameters, thermal stability and porosity of scaffolds were improved. Moreover, calcium and phosphorous were found in each kind of scaffold. SEM images showed that the precipitation

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