Release of chondroitin sulphate from chitosan-based complex hydrogel.

Edvani C. Muniz, Juliana F. Piai, Adriano V. Reis and Adley F. Rubira

Universidade Estadual de Maringá, Departamento de Química,
Grupo de Materiais Poliméricos e Compósitos – Maringá – Paraná - Brazil. - *Corresponding author: ecmuniz@uem.br*

Statement of Purpose:

The Chitosan (CS) is a polysaccharide obtained from deacetylating of chitin that is a naturally occurring mucopolysaccharide and has been found in invertebrates such as crustaceans, fungi, insects, annelids and mollusks. Nowadays, the CS has been applied in biomedical and pharmaceutical field as drugs carrier due to its biodegradability and non-toxicity that provide biocompatibility [1]. The CS is not soluble in pure water but becomes soluble, easily, in mild acid conditions due to the ionization of amine groups along the polymer chains. As the amount of protonated groups is increased higher is the repulsion among them and more water is need for solvation [2]. The chondroitin sulphate (CSulp) is a glycosaminoglycan extracted from bovine aortal. It is a highly water-soluble biopolymer that has been widely used to treat arthritis-related diseases [3]. Also, it is an important component in protein complex found in the extra-cellular matrix conjunctive animal tissue responsible for its good mechanical properties [4]. Due to its cationic nature and high charge density, the CS forms insoluble-water complexes with anionic moieties such as CSulp [5]. In this way, hydrogels based in complexes of CS/CSulp can be synthesized and have been applied as biomaterial. The aim of this work is to investigate the capacity of release of CSulp from the CS / CSulp complexes in water at several pH conditions.

Methods:

Solution A [CS, 2 % w/vplus HCl 0.57 mol L^{-1}, at 65º C] and solution B [CSulp, 10 % w/v, in water] were prepared. Equal volumes (50 mL) of solutions A and B were mixed at room temperature and keep in repose overnight. Hydrogel forms by complexion just by mixing and precipitates in the bottom of recipient. It was filtered and purified in distilled water. The supernatant was renewed at every 24 hours for one week. After, the swollen hydrogel was cut in cubic pieces and dried in vacuum. A requested mass of hydrogel was soaked in 25 mL aqueous solution of HCl (pH 2), of KH_2PO_4/NaOH (pHs 6 or 8) and at constant ionic strength (0.1 mol L^{-1} of KCl) at 37º C. Aliquots were collected at several immersion time and the amount of CSulp in each aliquot was determined through HPLC analysis.

Results/Discussion

In Figure 1 the dependence of CSulp released from hydrogels as a function of time in pH 2, 6 and 8, are presented. The hydrogel used in this study is constituted of 83 % of CSulp and 17 % of CS (wt-%). The sample soaked in aqueous solution of pH 2 presented higher initial rate of release of CSulp when compared to pH 6 and 8. This can be attributed to the ionization degree of chitosan amine-groups in a more acidic condition, i. e., the occurrence of the reaction ($–NH_2 + H^+ \rightarrow –NH_3^+$), and thus the repulsion among them. This allow to a more expansion of the hydrogel. So, in this condition the CSulph entrapped in hydrogel is more easily released. In pH = 8, higher than the pK_a-value of CS (= 6.5), the amine groups remain neutral while the sulphate ($-OSO_3H$) and carboxylic (-COOH) ones of CSulp became deprotonated and negatively charged. The repulsion among these negative charges would rise the initial release rate of CSulp but it was not observed. The barely solubility of CS at pH 8 probably avoid the release of CSulp in the same amount that occurred in pH 2. As the pH value is lower higher is the amount of SC released.

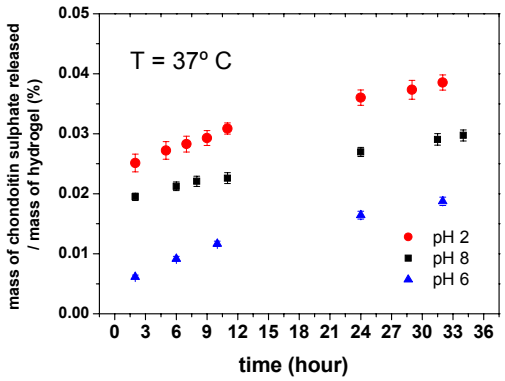

Figure 1 – Amount of CSulp delivered from CS/CSulph complex hydrogel vs. time in three pH values, at 37 ºC.

Conclusions

The chondroitin sulphate (CSulp) can be delivered from hydrogel based in chitosan (CS / CSulp). At 37º C the initial rate as well as the amount of CSulp released are controlled by the pH. The hydrogels investigated on this work present potential to be applied as biomaterial for implants for arthritis treatment.

References

1 - Ravi-Kumar MNV, Muzzarelli RAA, Muzzarelli C, Sashiva H, Domb, AJ *Chem. Rev.* 2004; 104:6017-6084.

2 - Capitani D, Crescenzi V, De Angelis AA, Segre AL *Macromolecules*, 2001;34:4136-4144.

3 - Richy F, Bruyere O, Ethgen O, Cucherat M, Herotin Y, Reginster JY *Arch. Int. Med.* 2003;163:1514-1522.

4 - Chen YL, Chen HC, Lee HP, Chan HY, Hu, YC *Biomaterials*. 2006;27:2222-2232.

5 - Vasiliuasiliu S, Popa M, Rinaudo M. *Eur. Polym. J.* 2005; 41:923-932.

Aknowledgements: CPNq (Proc. 479539/2004-9), CAPES and Fund. Araucária, Paraná (Proc. 5739, Conv. 114/04).