

DERMATAN SULFATE FROM ASCIDIANS OF ORDERS STOLIDOBRANCHIA AND PHLEBOBRANCHIA (UROCHORDATA:ASCIDIACEA) HAVE A DIFFERENT SULFATE PATTERN THAT IS CORRELATED WITH THE BIOLOGIC ACTIVITY

Eliene O. Kozłowski, Paula Lima and Mauro S. G. Pavão

From Laboratório de Tecido Conjuntivo, HUCFF and IBqM, UFRJ

Dermatan sulfate (DS) is a glycosaminoglycan composed by uronic acid and N-acetylgalactosamine that can be sulfated in different positions. In this work, we aimed to correlate the sulfation pattern of DSs obtained from two different orders of ascidians with its phylogenetic position and anticoagulant activity. DS from *Herdmania momus*, *Halocynthia roretzi* (Order Stolidobranchia) and *Ciona intestinalis* (Order Phlebobranchia) was purified by ionic-exchange chromatography on a Mono Q/FPLC column. Disaccharide analysis were performed to verify the sulfation pattern of the DSs and revealed a characteristic sulfate distribution in the DS chains of order. DS from the order Stolidobranchia is formed almost exclusively by uronic acid(2S)-N-acetylgalactosamine(4S) units, while Phlebobranch ascidians contain DS chains rich in uronic acid(2S)-N-acetylgalactosamine(6S). To study the anticoagulant activity, the direct measurement of inhibition of thrombin by heparin cofactor II was performed in the presence of ascidian and mammalian DS. DS from Stolidobranch ascidians have has a higher anticoagulant activity ($IC_{50}= 0.2-0.6 \mu\text{g/mL}$) than mammalian DS ($IC_{50}= 2.1 \mu\text{g/mL}$). This effect is proportional to the content of 2,4-sulfated disaccharides. DS from Phlebobranch ascidians were not active as an anticoagulant molecule. Taken together these results show an interesting change in the sulfation position of DS during the chordate evolution and a correlation of this feature in the biologic activity of this macromolecules.

Key words: ascidian, anticoagulant, chordate, evolution, Urochordata

Financial support: CAPES, CNPq, FAPERJ, NIH-FIC