OPTIMIZATION OF THE CHEMICAL SULFATION PROCESS OF A FUCOGALACTAN FROM Agaricus bisporus TO OBTAIN AN ANTICOAGULANT AGENT

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Introduction: Mushrooms have been valued by human kind as an edible and medical resource containing a number of bioactive molecules with therapeutic properties, including polysaccharides. Several studies have shown the anticoagulant activity of sulfated polysaccharides. The common mushroom Agaricus bisporus is a good source of polysaccharides which can be chemically sulfated to present anticoagulant activity.

Objective: To optimize the chemical sulfation of a fucogalactan from A. bisporus in order to obtain an anticoagulant agent.

Materials and methods: The fucogalactan was purified from an aqueous extract of A. bisporus, and characterized by methylation, NMR and HPSEC-MALLS analyses. It was sulfated in function of different factors [time, molar ratio of ClSO₃H to OH of the polysaccharide (ηClSO₃H:ηOH), and weight ratio of polysaccharide to total reaction volume (Wp/VT)]. The Degree of Sulfation (DS) was measured and the anticoagulant activity was evaluated by Activated Partial Thromboplastin Time (APTT) and Prothrombin Time (PT).

Discussion and results: The fucogalactan has a (1→6)-linked-α-D-Galp main-chain, partially methylated at O-3, and partially substituted at O-2 by non-reducing end-units of α-L-Fucp and β-D-Galp, and a Mw of 3.64 x 10⁴ g.moL⁻¹. The best anticoagulant activity was obtained with the sulfated fucogalactan synthesized with a ηClSO₃H:ηOH of 18:1 and a Wp/VT of 1:100 in 6 hours of reaction, named E100. The highest DS value (2.79) was also obtained in these conditions. The results of anticoagulant activity of E100 showed a linear increment of APTT for concentrations of 15 to 45 µg.mL⁻¹, whereas PT was constant between 120 and 160 µg.mL⁻¹. The NMR analyses suggest that non-reducing end-units of α-L-Fucp and β-D-Galp were greatly replaced by sulfate groups.

Conclusions: E100 inhibited especially the intrinsic pathway of blood coagulation. Different sulfation conditions generated different sulfated polysaccharides, which varied at DS and anticoagulant activity.

Keywords: Anticoagulant activity, Chemical sulfation, Fucogalactan

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