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灰树花多糖硫酸酯化衍生物的制备与抑制HMEC迁移的作用

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Preparation of Grifola frondosa polysaccharide sulfated derivatives and their inhibitory effects on HMEC migration

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摘要 灰树花(Grifola frondosa)高浓缩精粉经热水提取,95%乙醇沉淀,获得水溶性灰树花粗多糖GFP.GFP依次经DEAE-cellulose阴离子交换树脂及Sephadex G-100葡聚糖凝胶分离纯化得到GFP1-F, GFP1-M及GFP1-L 3种不含蛋白质的葡聚糖纯品,其分子量依次为 1.09×10^5 , 1.93×10^4 和 2.76×10^3 Da.采用吡啶-氯磺酸法对其进行硫酸酯化修饰,硫酸酯化衍生物GFP1-FS, GFP1-MS及GFP1-LS的红外光谱分析表明,3个样品均在 1236.90 cm^{-1} 和 811.81 cm^{-1} 有硫酸酯键的特征吸收峰,13C-NMR证明C-6上的羟基被酯化.并且GFP1-FS的硫酸酯化程度最高,其取代度DS为1.07;GFP1-MS与GFP1-LS的硫酸酯化程度相当,DS分别为0.66和0.61.划痕法实验结果表明,经 $1000 \mu\text{g/mL}$ 的GFP1-FS, GFP1-MS及GFP1-LS处理24 h后向划痕区迁移的细胞数明显减少,分别为对照组的73.33%, 34.17%和67.21%,均具有抑制人微血管内皮细胞(HMEC)迁移的活性,其中GFP1-MS的效果最为显著,这可能与GFP1-MS所具有的复杂分支结构有关.

关键词: 灰树花 葡聚糖 硫酸酯化衍生物 人微血管内皮细胞 迁移 灰树花 葡聚糖 硫酸酯化衍生物 人微血管内皮细胞 迁移

Abstract: Polysaccharides from Grifola frondosa (GFP) were extracted by hot water and precipitated by 95% EtOH. GFP1-F, GFP1-M and GFP1-L were further purified by DEAE-cellulose and Sephadex G-100 subsequently from GFP. GFP1-F, GFP1-M and GFP1-L were glucan with molecular weight 1.09×10^5 , 1.93×10^4 and 2.76×10^3 Da. GFP1-FS, GFP1-MS and GFP1-LS were polysaccharide sulfates obtained from GFP1-F, GFP1-M and GFP1-L with chlorosulfonic acid pyridine. The IR spectrum of GFP1-FS, GFP1-MS and GFP1-LS showed the characteristic absorptions of sulfate ester bond at 1230 cm^{-1} and 810 cm^{-1} . The $^{13}\text{C-NMR}$ results indicated the modification mainly occurred at C-6 of the polysaccharide sulfates. GFP1-MS had the greatest sulfated degree with DS 1.07, while DSs of GFP1-FS and GFP1-LS were 0.66 and 0.61 respectively. The scratching assay suggested that 73.33%, 34.17% and 67.21% cells migrated to scratching area compared with control after 24 hour treatment of $1000 \mu\text{g/mL}$ GFP1-FS, GFP1-MS and GFP1-LS respectively. So that all the three sulfated derivatives had inhibitory effects on HMEC migration, especially GFP1-MS had the strongest activity which may be related with its complicated branch structure.

Key words: glucan sulfated derivative HMEC migration Grifola frondosa glucan sulfated derivative HMEC migration

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