

论文

桦褐孔菌野生菌丝体和培养菌丝体的甾体类化合物组成

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摘要:

甾体类化合物是桦褐孔菌治疗疾病的有效成分之一。该菌的野生菌丝体中有含量很高的多种甾体类化合物。然而人工培养的桦褐孔菌菌丝体中很少积累甾体类化合物。为了分析导致野生菌丝体和培养菌丝体甾体类成分差异的原因, 本研究采用80%乙醇在室温下对菌丝体进行提取, 用硅胶柱色谱制备总甾体类化合物, 并以GC-MS和波谱学方法进行鉴定。与此同时, 桦褐孔菌用基本培养基(葡萄糖2%, 酵母膏0.5%, KH_2PO_4 0.01%, $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ 0.05%, pH 6.5)或在基本培养基中加入不同浓度的 AgNO_3 进行培养。结果显示桦褐孔菌野生菌丝体甾体类化合物以羊毛甾醇和桦褐孔菌醇为主要成分, 分别占45.47%和25.36%。另有10种次要成分, 共占总甾体类化合物的30.17%, 其中包括24-甲基二氢羊毛甾醇、4, 4-二甲基粪甾醇、4-甲基粪甾醇、粪甾醇以及表甾醇。同时, 从柱色谱分离还得到了羊毛甾醇、桦褐孔菌醇、木栓酸、桦褐孔菌醇B和1种新的甾醇类化合物桦褐孔菌醇D。相比之下, 桦褐孔菌的培养菌丝体中仅含有3种甾醇类化合物, 其中麦角甾醇占82.20%, 桦褐孔菌醇占14.12%, 而羊毛甾醇仅有3.68%。在基本培养基中加入 $0.28 \mu\text{mol} \cdot \text{L}^{-1}$ 的 Ag^+ 可将羊毛甾醇的含量提高到56.81%, 使麦角甾醇的含量下降到18.5%。与此同时, 还检测到麦角甾醇合成途径中的中间体。这些结果表明, 野生菌丝体甾醇类化合物种类多样性的原因可能与麦角甾醇生物合成受到抑制有关。同时苛刻的野生生长环境如温差变化和紫外线照射是造成野生菌丝体甾醇类化合物组成多样性的主要原因。

关键词: 桦褐孔菌 甾醇类化合物组成 野生菌丝体 培养菌丝体

ZHENG Wei-fa, et al: Sterol composition in field-grown and cultured mycelia of *Inonotus obliquus* Sterol composition in field-grown and cultured mycelia of *Inonotus obliquus*

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Abstract:

Sterols are one of the active classes of compounds in *Inonotus obliquus* for their effective therapy of many diseases. In field environment, this fungus accumulates large amount of sterols. In cultured mycelia, however, this class of compounds is less accumulated. For analyzing the factors responsible for differing sterol composition, the field-grown and cultured mycelia were extracted with 80% ethanol at room temperature and total sterols were prepared using silicon gel column chromatography followed by identification using either GC-MS or spectroscopic methods. For culturing *Inonotus obliquus*, the seed culture was grown either in basic medium consisting of glucose (2%), yeast extract (0.5%), KH_2PO_4 (0.01%), $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ (0.05%) and distilled water at pH 6.5, or the basic medium supplemented with serial concentrations of AgNO_3 . The results indicated that field-grown mycelia contained lanosterol and inotodiol (comprised 45.47% and 25.36% of the total sterols, respectively) and other 10 sterols (comprising the remaining 30.17%) including ergosterol biosynthetic intermediates such as 24-methylene dihydrolanosterol, 4,4-dimethylfecosterol, 4-methyl fecosterol, fecosterol and episterol. Column chromatography also led to the isolation of lanosterol, Inotodiol, trametenolic acid, foscoparianol B and a new triterpenoid foscoparianol D in field-grown mycelia. In comparison, the cultured mycelia only contained three sterols with ergosterol as the predominant one (82.20%). Lanosterol only accounted for 3.68%. Supplementing Ag^+ into the culture at $0.28 \mu\text{mol} \cdot \text{L}^{-1}$ greatly enhanced content of lanosterol (accounting for 56.81%) and decreased the content of ergosterol (18.5%) together with the presence of intermediates for ergosterol biosynthesis. These results suggested that the sterol composition in mycelia of the fungus can be diversified by supplementing substances inhibiting enzymatic process towards the synthesis of ergosterol. Harsh growth conditions in field environment (i.e. temperature variation, UV irradiation etc.) can delay the synthesis of ergosterol and hereby diversify the sterol composition in the mycelia of *Inonotus obliquus*.

Keywords: sterol composition field-grown mycelia cultured mycelia *Inonotus obliquus*

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