

利用樟芝(*Taiwanofungus camphoratus* Wu et al.)菌絲體轉換靈芝三萜類及性別遺傳模式應用於育種之研究

Transformation of triterpenoids from *Ganoderma tsugae* by mycelial pellets of *Taiwanofungus camphoratus* and the study on mode of mating types

中文摘要

樟芝(*Taiwanofungus camphoratus* Wu et al.)是台灣特有的真菌，含有高產量之三萜類及其他生理活性物質，價格昂貴，目前難以人工栽培出子實體，是一種高價值的藥材。本研究發現樟芝雙核菌絲單核化的現象，挑選樟芝的雙核菌絲所產生的分節孢子(Arthroconidia)，進行培養後，利用玻片培養及 cotton blue stain 觀察，發現沒有扣子體(clamp-connection)的產生，推測為無性的單核孢子，利用此現象探討樟芝的交配型，分別挑選出節孢子進行培養，將單核無性孢子進行所有可能交配，結果所有的交配菌株皆沒有扣子體的產生。推測可能雙核菌絲形成節孢子時，只會釋出一種性別的核。利用單核孢子的交配於育種，分析無性單核菌絲所含三萜類成份，取含量高者進行交配，發現沒有加乘的作用。利用選種的方法分離出型態及分泌物不同的兩株菌株，分別為菌株 7-1、7-4，其中菌株 7-4 具有一特有成份是菌株 7-1 所稀少或是缺乏的，透過 TLC、PLC、HPLC、及液相層析質譜儀進行純化分析，推測此物質分子量為 488。加入靈芝三萜類於培養基中，觀察樟芝菌絲體的形態並分析三萜類的生成，形態方面，加入少量的靈芝三萜類可以促進菌絲體孔洞的產生，透過光學顯微鏡及電子掃描顯微鏡觀察孔洞，並沒有發現有性世代的證據。三萜類分析方面，25°C 培養 180 天仍然無法將靈芝三萜類轉換成樟芝三萜類。

英文摘要

Taiwanofungus camphoratus Wu et al. contained high triterpenoids and other physiologically active compounds were used as a precious herb and difficult to culture the fruiting body with artificial environment in Taiwan. It was obvious that the clamp-connection was disappeared through the hyphae of the single-aseexual-spore isolates and all the septate hypha containing only one nucleus with HCl-Giemsa stain. It suggested that the isolates were monokaryons. The mating reaction of isolates were now under investigation. It was obvious that no clamp-connection all the mating reaction. It was suggested dikaryon release only one sexual nucleus to form asexual spore and then all the asexual spores had only one sexual spore. The mating reaction was not been observed. Even though, triterpenoid of all the monokaryon was analyzed by TLC, and triterpenoid of all the monokaryon was

different. Triterpenoid content of the after mating reaction with all monokaryon was not increased. Two different types 7-1 and 7-4 had the different appearance and TLC pattern were selected. A component found in mycelium 7-4 was purified with PLC and identified LC/MS/MS with molecular weight 488. *T. camphoratus* was cultured in PDA plate with additional triterpenoid of *Ganoderma tsugae* in 25°C for 180 days. It was obvious that pore formation were observed on the surface of mycelium. However, *T. camphoratus* mycelium could not transfer triterpenoid of *Ganoderma tsugae*.