

# 納豆菌生產果糖聚合物之研究

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## 摘要

Levan為fructofuranosidic殘基以  $\beta(2\rightarrow6)$  形式鍵結之果糖聚合物，可以利用微生物來生產，且能夠應用在化妝品、藥品、藥物釋放、農業與食品工業上。雖然已經有許多關於levan之研究被發表，但其所提到關於levan之產量不是過低，就是生產過程中會有其他產物的產生，而造成純化上的困難。因此，使得levan之價格較高，進而使得levan的應用性受到限制。為了更進一步了解利用Bacillus subtilis natto來大量生產levan之可能性，本研究探討各項影響此B. subtilis natto生產levan時的環境因子，並進行產物之純化與鑑定其結構。B. subtilis natto於含有20% (w/v) sucrose之培養基中，經過24 h培養後，可生產40-50 g/L的levan聚果糖。經膠體過濾層析法 (Gel permeation chromatography, GPC) 分析levan產物時，可發現本實驗之產物是具有兩種不同之分子量 ( $2 \times 107$  and  $9 \times 103$  Da)，且容易利用乙醇沈澱法來進行純化，並且將此產物利用FTIR (Fourier transform infrared) 與NMR (Nuclear magnetic resonance) 進行特性分析後，已證實多醣聚合物即為levan。利用無菌水清洗後之B. subtilis natto菌株以各種氮源基質之條件來進行levan的發酵生產，氮源包括yeast extract、peptone、urea、NH<sub>4</sub>Cl、NaNO<sub>3</sub>與玉米漬液。結果顯示B. subtilis natto是能夠代謝無機氮源 (例如，NH<sub>4</sub>Cl與NaNO<sub>3</sub>) 來進行levan生產，但是卻無法有效利用有機氮源(尤其是yeast extract與玉米漬液)。另外，蔗糖濃度也是影響levan產量與levan生產速率 (VL) 的原因之一。實驗結果顯示levan之合成是需以蔗糖作為碳源，並且將B. subtilis natto菌株培養於含有250 g/L蔗糖基質的培養基中，是能得到最大的levan產量 (71 g/L) 與levan生產速率。並且探討影響levan生產之各項環境因子 (例如，初始pH與培養溫度)，結果顯示pH 7與溫度為37 為最適合levan的生產條件。另外，本研究亦探討利用PVA固定化菌體顆粒來進行生產levan的可行性，並發現此固定化菌體顆粒確實是能用來大量生產levan 聚果糖。

關鍵詞：Bacillus subtilis natto；發酵生產；聚果糖；levan；固定化

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