

# 應用毛細管線性聚丙烯醯胺凝膠電泳來分離木糖寡糖

研究生:張恩璋

指導教授:謝有容

國立交通大學應用化學所

## 摘要

本研究利用毛細管線性聚丙烯醯胺凝膠電泳來進行木糖寡糖的分析，旨在於改善傳統的固態毛細管凝膠電泳所產生的製備不易、再現性不佳及管柱壽命短的問題。醣類近年來在醫藥、食品、化妝品上有卓越的貢獻與應用。然而醣類質荷比相近，於傳統毛細管電泳上不易分析。本實驗採用塗佈管柱，藉由緩衝溶液添加線性聚丙烯醯胺凝膠，可以提高凝膠電泳的再現性與穩定性，壽命可達 3 週以上。此外，凝膠分離系統俱大小篩分的功能，可大大提升分離效率，於高聚合度醣類之解析度也有優異表現，更能有效地提升所分離的木糖寡糖聚合度。分析物樣品製備採用加酸加熱水解的方式，將木糖聚醣切成聚合度不等的木糖寡糖，並以 3-aminonaphthalene-2,7-disulfonic acid (3-ANDA) 為紫外光吸收衍生試劑進行還原胺化 (reductive amination) 反應，其吸收波長為 254 nm。本研究中探討以下實驗參數進行對分離效率之影響：控制木糖水解所需的加熱時間、背景緩衝溶液的酸鹼值、離子強度、線性聚丙烯醯胺凝膠的濃度、及凝膠的起始劑濃度等。結果顯示採用加熱酸水解 10 分鐘之後，木糖寡糖進行衍生反應，經由 2% 線性凝膠電泳，pH 5.0 濃度為 25 mM 的 citric acid/sodium citrate 緩衝溶液下，可依序分離出聚合度 1 至 20 糖，單糖出現在 5 分鐘以內，九糖、十糖的解析度可高達 6

以上。並計畫將此技術應用於其他類型的醣類，如幾丁醣等，開發其應用的潛力。



# Analysis 3-ANDA-Xylan Oligosaccharides by Capillary Gel Electrophoresis

Student : En-Wei Chang

Advisor : You-Zong Hsieh

## Abstract

In this study, we improve the disadvantages of traditional capillary gel electrophoresis (CGE), including complicated preparation process, poor reproducibility, and limited life-time. Using low-viscosity linear polyacrylamide in the electrolyte provides the sieving effect to separate xylan-oligosaccharides. Xylan-oligosaccharides by acid hydrolysis were derivatized with 3-aminonaphthalene-2,7-disulfonic acid (3-ANDA) via reductive amination. The derivatized xylan-oligosaccharides were separated by 2% (w/w) linear polyacrylamide mobile gel with high resolving power. Peaks additional to the regular oligomers can be observed that are assumed to be conformers of this oligomer. The effect of the time of acid hydrolysis treatment, acid-base study, the concentration of the linear polyacrylamide, electric field strengths and the ion strength of the buffer on the separation are discussed. Finally, the 3-ANDA derivatized xylan oligosaccharides with degree of polymerization 1-20 were successfully separated by linear polyacrylamide in a 25 mM citric acid/sodium citrate buffer.