蠶絲蛋白質薄膜之靜電紡絲製程及其牙齒美白貼片應用研究

中文摘要

本研究之目的在以靜電紡絲(Electrospinning)技術將絲蛋白(Silk Fibroin, SF)製成酵素固定用之再生絲蛋白纖維薄膜，並評估應用於家用型牙齒美白貼片基材之可行性。取蠶繭經脫膠(Degum)處理所得絲蛋白，以($CaCl_2:C_2H_5OH:H_2O$)溶劑系統溶解配成絲蛋白溶液，利用迴轉式流變儀觀察其剪切黏度變化後，以滴定法配合剪切應力建構絲蛋白三成分比例圖，發展出異於傳統透析(Dialysis)方式之剪切應力誘導相分離製程獲得新再生絲蛋白(New Regenerated SF)，並分別使用毛細管黏度計、掃描式電子顯微鏡及 X 光繞射儀，分析再生絲蛋白產物之固有黏度、表面形態及結晶結構。將再生絲蛋白溶於甲酸配成不同濃度之紡絲液，以表面張力儀及拉伸流變儀進行表面張力與拉伸黏度之量測，並經靜電紡絲製成不織布薄膜，其後使用甲醇做表面改質，再分別以掃描式電子顯微鏡、接觸角量測儀及反射式傅立葉紅外線光譜儀，進行薄膜之表面形態、纖維直徑、潤濕性、表面能量及二級結構研究，最後將馬鈴薯萃取酵素 PEA (Potatoes Extract Antioxidant)以冷凍乾燥法固定於薄膜上，利用牙齒樣品進行體外漂白，並以分光測色儀分析漂白前後之牙齒色差。結果顯示，當絲蛋白溶液中之絲蛋白比例低於 1 wt% ，於 35 ℃下配合 6~8krpm 剪切應力誘導，平均可得再生絲蛋白產率為 87.7±3.5 % ，高於傳統透析法(70.8±4.7 %)，但相對分子量較低，結晶結構亦具差異性。另外，再生絲蛋白紡絲液之拉伸黏度隨溶液濃度上升而逐漸增加，並發現濃度在 4~7 wt% 時溶液呈現剪切稀化(Shear Thinning)，當濃度提高至 8~9 wt% ，則溶液之流變特性出現轉折呈現剪切稠化(Shear Thickening)，在此濃度範圍後始可以靜電紡絲紡得具均勻形態之纖維。而本研究以 10 wt% 再生絲蛋白紡絲液進行靜電紡絲，所得纖維直徑為 0.71±0.06 ?慆，收成薄膜經甲醇改質後，表面接觸角由 110.1±1.52°降至 60.1±0.25°，表徵??-Pleated Crystal與 Random Coil二級結構比例之 Crystallinity Index(CI=A1628/A1665)則從 0.7 上升至 2.86，最後固定有 PEA 之薄膜加速老化試驗兩周前後，牙齒漂白色差△E*值從 61 降至 8。本研究發展出可利於再生絲蛋白大量生產之剪切應力誘導相分離法，解決傳統透析所需時間過長及製程不穩定之問題。本研究同時也建立再生絲蛋白紡絲液與溶液拉伸流變特性之關係，值得深入探討。而以新製程生產再生絲蛋白所紡製出之薄膜，展現較傳統透析法再生絲蛋白薄膜優良之抗水溶解性及親水性。最後將 PEA 固定於靜電紡絲再生絲蛋白薄膜上，結合 5.25 wt% H_2O_2 使用，可具有優異之牙齒美白效果，證實其具有應用於牙齒美白貼片之可行性。

英文摘要

The objective of this study is to produce electrospun regenerated silk fibroin (R-SF) membrane for enzyme immobilization and to explore the feasibility of its dental applications as a tooth whitening strip. The SF harvested from degummed cocoons of silkworm was dissolved in ($CaCl_2$:C_2H_5OH:H_2O) solvent. The shear viscosity of SF solution was measured by rotational rheometer. The processing window of R-SF was constructed with shear-induced method and titration test. The R-SF precipitation could be produced by shear-induced method without dialysis. The inherent viscosity, morphology, and crystal structure of R-SF were analyzed by capillary viscometer, scanning electron microscope (SEM), and X-ray diffractiometer. The R-SF was dissolved in formic acid to prepare the spinning dopes. The extensional viscosity of R-SF/formic acid was analyzed by Wilhelmy type of surface tensiometer and capillary breakup extensional rheometer (CaBER®). Non-woven membranes were then produced via electrospinning. The morphology, wettability, surface energy, and secondary structure of methanol treated R-SF membranes were respectively examined by SEM, contact angle measurement, and FTIR-ATR. Finally, the potatoes extract antioxidant (PEA) was immobilized on electrospun R-SF membranes via freeze drying. The tooth whitening strips were prepared by adding 5.25 wt% hydrogen peroxide onto PEA immobilized R-SF membranes and examined whitening efficacy by colorimeter. R-SF was produced by shear-induced precipitation method (6~8k rpm, 35 ℃) when percentage of SF was below 1 wt%. With this new method, the average yield (87.7±3.51 %) was higher than that of dialysis one (70.8±4.72 %). The crystal structures were different between the new method and dialysis. Besides, extensional viscosity of R-SF/formic acid was depended on solution concentration. The characteristic of solution showed shear thinning in the concentration range of 4~7 wt%, however it transformed to shear thickening at higher concentration range of 8~9 wt%. Within this shear thickening range, the uniform fibers were obtained by electrospinning. The 10 wt% R-SF spinning solution was chosen for experiment hereafter. The average diameter of electrospun fibers was 0.71±0.06 ?惛. After treated by methanol, the contact angle of electrospun R-SF membranes decreased from 110.1±1.52 to 60.1±0.25 degrees. The structure transformation from random coil to ??-pleated crystal of SF was expressed by crystallinity index, ratio of A_{1628} cm^{-1}/A_{1665} cm^{-1}, increasing from 0.7 to 2.86. After the treatment of fresh whitening strips, the stained teeth showed a significant color difference (?嫓*) value of 61. But it decreased down to 8 after the whitening strip aged for 2 weeks. A new shear-induced precipitation method for R-SF production is developed with advantage of high productivity. Besides, the relationship between electrospinnability of R-SF/formic acid and solution characteristic about extensional rheology is established. This phenomenon is worth further exploration. Membranes prepare from shear-induced R-SF show superior water-resistance and hydrophilicity than those from dialyzed one.

The concept for a novel tooth whitening system containing hydrogen peroxide/PEA/electrospun R-SF membrane is effective for tooth whitening evidently