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The Characteristics of Self-assembled Tussah Silk Fibroin Nanoparticles for Controlled Drug Release

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Abstract

Tussah silk fibroin is a promising material for drug release because of protein component, biocompatibility, aqueous processability, and biodegradability. Tussah silk fibroin nanoparticles were self-assembled and prepared in this paper. Their appearances and sizes were modified by altering the volume ratios between tussah silk fibroin and ethanol. When 9 mL of ethanol was added, the average microsphere size was 184.52 nm and the scattered index was 0.2903. The tussah silk fibroin nanoparticles for controlled drug release was studied and the result showed that the time of drug release was significantly longer. The process of drug release was divided into rapid release in early stage and slow release in late stage. The target drug delivery can reduce the negative effects and improve the efficiency of the drug. As a result, it favors deeper investigation of tussah silk fibroin nanoparticles as drug carriers for controlled release of sensitive biologicals.

Keywords: Tussah Silk Fibroin; Nanoparticle; Drug Release; Self-assembly

1 Introduction

Silk fibroin is a fibrous protein manufactured by insects or spiders. With its unique properties like processability, mechanical strength, long-term degradability and biocompatibility, it is currently being used in several biomedical fields [1]. Foremost, silk fibroin is used in great potential fields, such as target drug delivery and controlled release system, because of strong mechanical stability, unique protein component and slow degradability, compared with other synthetic and natural polymers, such as alginate, polyanhydrides, polyorthoesters, chitosan and polyesters etc [2]. Therefore, design and characteristic of devices composed by silk fibroin have been reported, including microspheres [3-5] and scaffolds [6-8]. For biomedical purposes, the silk fibroin is a successful material that fulfills most necessary requirements.

The applications of silk fibroin are mostly about the silkworm fibroin. The reports about the developments and applications of tussah silk are few except some studies of tussah silk fibroin membrane [9, 10]. Compared with silkworm fibroin, Tussah Silk Fibroin (TSF) with special three

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peptide sequence of Arginine-Glycine-Aspartate acid (RGD) [11], contributes to cell adhesion which was studied by Minoura et al [12]. The property of affinity of tussah silk fibroin was better than silkworm fibroin because the tussah silk fibroin contained a certain amount of amino acids with positive charges. Cells adhesion property and value-added rate in tussah silk fibroin membrane were much higher than silkworm fibroin membrane which was investigated by Altman et al [13]. However, there was no investigation of tussah silk fibroin self-assembly nanoparticles for drug release.

Above all, tussah silk fibroin is a natural protein, which can be used as biomaterials, e.g. surgical suture, scaffold for tissue engineering, wound covering material, soft contact lens and controlled release carrier, because it has good biocompatibility and biodegradability. Tussah silk fibroin contains 18 kinds of essential amino acids, which can be decomposed by specific enzyme to non-toxic products for the body, not easy to cause inflammation and immune rejection. In view of these characteristics, the nanoparticles as drug carriers and controlled release have good application in medicine. Therefore, the preparation and properties of self-assembly tussah silk fibroin nanoparticles were characterized and the application in controlled release was investigated in this paper.

2 Materials and Methods

2.1 Materials

Cocoons of tussah was provided by Nanyang, Henan Province, China. Sodium carbonate, lithium hydroxide, n-butanol, ammonium thiocyanate, polyethylene glycol (20000 Da), dialysis bag, 5-Fu and other chemicals were purchased from Sinopharm Chemical Reagent Co., Ltd.

2.2 Tussah Silk Purification

Tussah cocoons were boiled 4 times for 30 min in aqueous solution of 0.02 M Na_2CO_3, and thoroughly rinsed with distilled water to remove the sericin completely. After air drying, the extracted tussah silk fibroin was dissolved in 9 M LiSCN solution at 55 °C for 2 h, yielding a 20% (w/v) solution. The solution was dialyzed against distilled water using dialysis bags (MWCO 8000-14000 Da) for 4 days. Silk aggregates as well as debris were removed by filter papers. The solution was concentrated by polyethylene glycol. The final concentration of tussah silk fibroin aqueous solution was approximately 3% (w/v). The 3% tussah silk fibroin solution was kept at 4 °C for futher application.

2.3 Preparation of Tussah Silk Nanoparticles

The concentration of anhydrous ethanol and tussah silk were adjusted to different concentrations (w/v), typically a 3% solution was used for this experiment. 10 mL tussah silk fibroin solution and 0.5 mL, 1 mL, 1.5 mL, 2 mL, 2.5 mL, 3 mL, 3.5 mL, 4 mL, 4.5 mL, 5 mL anhydrous ethanol were mixed separately, and fully blended on a magnetic stirrer at low temperature and low speed for 5 min. Briefly the volume ratio of ethanol to tussah silk fibroin solution was adjusted from 1:20 to 10:20, to investigate the best proportion of ethanol in total solution. Before thawing and