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Extraction of Sericin Freeze Dried RSS Powder a Biodegradable Compound from Silk Cocoons

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ABSTRACTS
Even the most cutting-edge synthetic polymers cannot compare to the outstanding natural properties of silk fibres, which are produced without the need for harsh processing conditions. During the production of silk, sericin, a substantial component of the fibre is selectively separated from fibroin and discarded. By-products and Seri-waste items are now frequently used to create things with extra value. Fibroin, which is composed of two brins, is the residue left over after degumming. Several industries, including textile, medical, and industrial, use silk fibre. A long, delicate, and light fibre is silk. The current work discloses that a scanning electron microscope was used to examine the morphology of sericin freeze dried RSS powders at various magnifications. RSS powders are a useful one in numerous aspects and also a biodegradable substance.
***Keywords:** Silk fibre, Sericin, freeze dried RSS powders and biodegradable substance.*

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INTRODUCTION

Silk fibre used in several industries including textile, medical, and industrial. A long, delicate, and light fibre is silk. It is well renowned for its water absorption, dyeing affinity, thermostoichiometry, insulating properties, and gloss. It is utilized in the creation of surgical sutures, artificial blood vessels, parachutes, and priceless fabrics. The secret to producing designer silk strands with enhanced mechanical and visual properties may be ionic liquid. The columnar fibroin in the lumen of the posterior silk gland is made up of many spherical aggregates of fibroin fibres. In the anterior silk gland, these became uniform and compact; with a greater concentration in the back of the middle silk gland. Cocoon filament is formed of oriented elementary fibroin fibres, which are derived from fibroin fibres as they undergo structural change while passing through the silk gland lumen, according to observations of various parts of the silk gland. Sericin's molecular aggregating structure and modifications were proposed by [9]. The portion of the middle silk gland's liquid sericin that crystallizes readily after drying is probably composed of short-chain amino acid residues that are folded into a globular matrix composed of longer-chain stretches that crystallize less readily and, after drying, form an unoriented crystal film. After being stretched, dried, and soaked in water, this film takes on the shape of an aligned fibre structure. Yet, there is a reversible link between orientation and non-orientation because hot water treatment renders the orientation unstable. Sericin can be combined with various resins to create biodegradable polymers that are good for the environment [1]. Sericin can be used to create polymer films, foams, moulding resins, and fibres by reacting a composition including a polyol, tolylene, di-isocyanet, di-butyltin di-laurate (catalyst), and trichloro mono fluoro-methane (a blowing agent) in the presence of sericin (0.01 to 50 percent w/w). The moisture absorption/desorption rate of the sericin-containing polyurethane form is two to five times higher than that of the control. There have also been described techniques for producing sericin-containing polyurethane with exceptional mechanical and thermal properties [8]. Water desalination, the production of ultra-pure water, bioprocessing, and some chemical processes all involve membrane-based separations (such as reverse osmosis, dialysis, ultra-filtration, and microfiltration). Sericin contains a large number of amino acids with neutral polar functional groups, making it simple to crosslink, combine, or copolymerize sericin with other substances to make membranes. In separation procedures, sericin and fibroin-derived membranes can be used. The insolubilized silk fibroin barrier can be used to segregate water and alcohol [4]. a thin, across-linked sericin layer that serves as a membrane for separating ethanol and water [13].

Sericin-containing membranes have a high hydrophilicity. A protein-containing synthetic polymer screen for segregating water from organics can be made by copolymerizing specific synthetic polymers, like acrylonitrile, with sericin [24, 25]. A sericin film applied to the surface of a liquid crystal can uniformly orient the liquid crystal molecules, producing distortion-free, high-quality displays, as found by [14]. The surface of refrigeration machinery uses sericin-coated film because of its anti-frosting capabilities [16]. Coated sericin film is a fantastic anti-frosting solution for refrigerators, deep freezers, refrigerated vehicles, and ships. Using the coated film on roads and roofs can also lessen the chance of frost harm. Sericin protein can be coated on the surfaces of many strong materials to enhance performance [10]. Sericin can be used to create paint colours and shield product surfaces. The substance used to coat the sericin is highly weather resistant, permeable, and does not warp on exposure to air for drying. Water-soluble polymers, in especially polyvinyl alcohol, and serin combine well (PVA). The hydrogels made of sericin, fibroin, and PVA are said to be flexible and excellent at absorbing and releasing moisture [25]. The hydrogel can be used to improve soil quality, as well as in medical and skin care products. In order to immobilize enzymes, [12] described using glutaraldehyde as the cross-linking agent in a cross-linked sericin film. In comparison to the free enzyme, the immobilized J-glucosidase on the cross-linked sericin sheet is more stable and resistant to electro-osmosis. The most frequently used silk-like material in biomedical applications, especially sutures, has long been silkworm silk fibres. Over the years, silk fibres have shown to be beneficial in a number of clinical uses. On the other hand, biological reactions to the protein have raised questions about biocompatibility. a silk fibroin wound dressing that promotes healing and can be removed without damaging the freshly formed skin [17]. The non-crystalline fibroin layer of the wound dressings had a water concentration of 3–16% and a thickness of 10–100 m. Then, fibroin and sericin were combined to make the wound dressing [18]. Membranes made of sericin and fibroin are useful for promoting the growth of adherent mammalian cells and can be used as a collagen alternative. The silkworm B. mori makes a lot of silk proteins during the last stage of larval development. The silk gland of the silkworm has lately been used as a bio-factory to synthesize valuable proteins, advancing the science of sericulture. In order to obtain sericin from silk cocoons in the form of RSS (Raw Silk Sericin) powder, the current research was carried out (a biodegradable substance). The study's primary goal is to separate the sericin component from the freeze-dried powder of Bombyx mori silk cocoons.

MATERIAL AND METHODS

In Velliyanallur hamlet, Cheyyar Block, Thiruvannamalai District, silk cocoons were gathered. Velliyanallur hamlet is situated 12 kilometres east of Cheyyar, 86 kilometres east of the Thiruvannamalai District Headquarters, and 98 kilometres east of Chennai, the State Capital. The most popular sericin extraction technique is hot-water extraction because it has the advantages stated above. Without using any other additives, silk is boiled in hot distilled water. Sericin can be eliminated by separating it from the fibroin component of silk. Small portions of the silk cocoons (1.5g) were cut up and degummed in 100mL of sodium carbonate solution (0.02 M). The solution was dialyzed for three days against deionized water to obtain RSS powder, which was then freeze-dried (-40° C). The extracted quantity of sericin is influenced by the time and temperature of extraction. Although sericin degrades as a result of this method, sericin still maintains its essential properties. Many scholars opt to use hot-water extraction due to the technique's simplicity. Sericin can be eliminated by separating it from the fibroin component of silk. Since the silk industry only uses the fibroin portion of silk, sericin must be removed. This is accomplished through a degumming procedure, which is then discarded in the effluent. Sericin recovery from degumming spirits can lower effluent load, minimising environmental effect and producing a biopolymer with a variety of advantageous properties.

RESULTS

SEM image of Sericin samples (Liquid Dried):

Sericin freeze-dried granules' morphology was examined under a scanning electron microscope at magnifications of 2000X, 6000X, 12000X, 18000X, 30000X, and 40000X. Figures 1, 2, 3, 4, 5, and 6 provide the SEM pictures of the powder created by freeze drying. Particle formation is a consequence of the drying process, as evidenced by their presence. The micrographs of the sericin powder at each pass revealed a variety of particle sizes, shapes, and size distributions. A scanning electron microscope was used to examine the shape of sericin freeze-dried powders at various magnifications, including 2000X, 6000X, 12000X, 18000X, 30000X, and 40000X. The sizes (1-5 m, 5-20 m), shapes (wrinkled, smooth, donut-shaped, straight, or a mixture of all), and particle size distributions based on visual evaluation of the micrographs (mono disperse or poly disperse) showing small particles with a diameter of 1-5 m and large particles with a diameter of 5 m to 20 m were used to classify the particles into six groups.

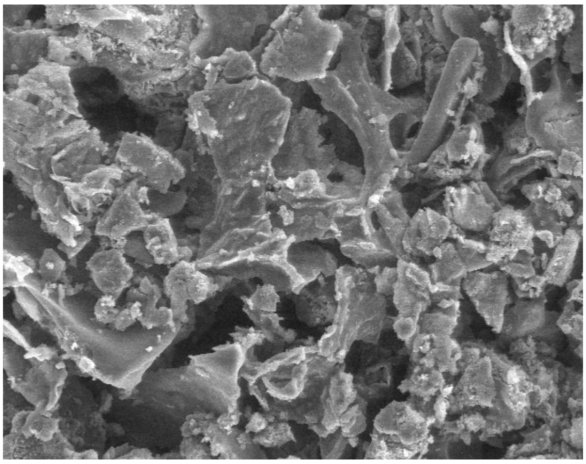

Fig 1 : SEM image of sericin samples (Freeze Dried) 2000X magnification

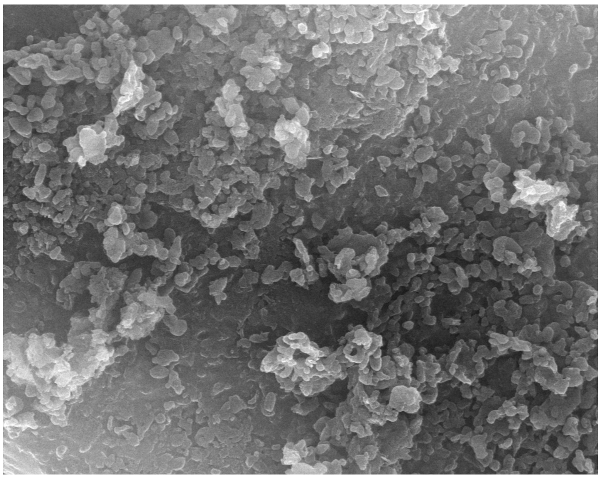

Fig 2: SEM image of sericin samples (Freeze Dried) 6000X magnification

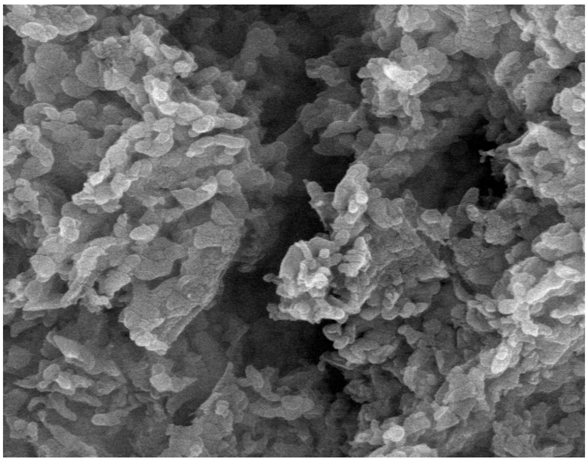

Fig 3: SEM image of sericin samples (Freeze Dried) 12000X magnification

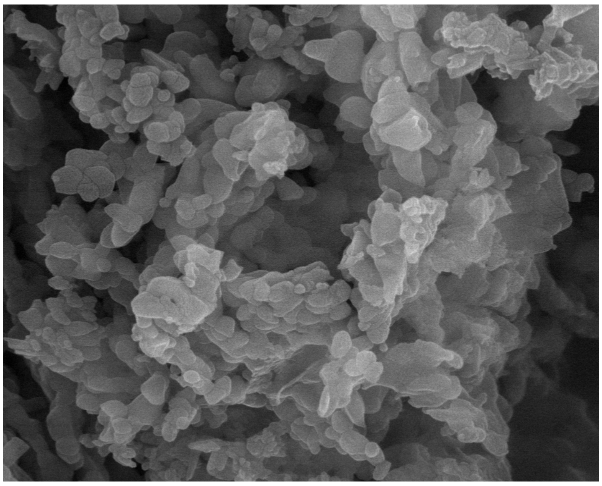

Fig 4: SEM image of sericin samples (Freeze Dried) 18000X magnification

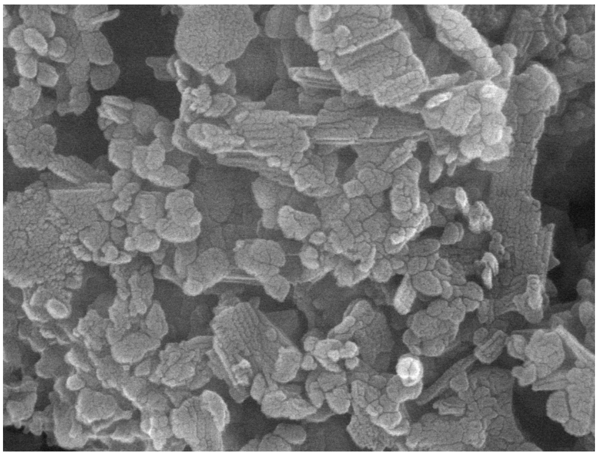

Fig 5: SEM image of sericin samples (Freeze Dried) 30000X magnification

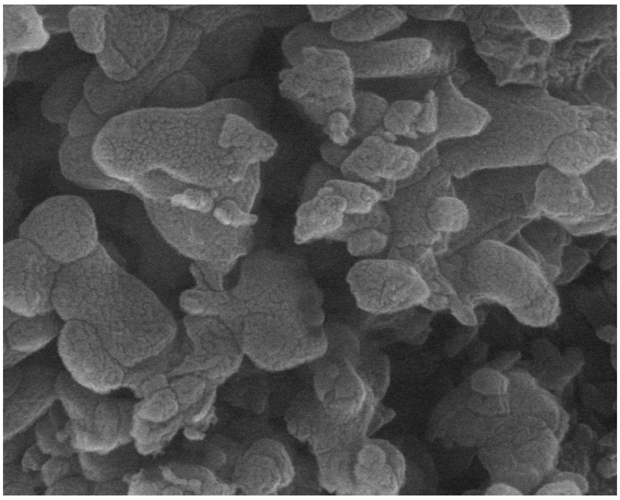

Fig 6: SEM image of sericin samples (Freeze Dried) 40000X magnification

DISCUSSION

In the textile business, sericin is helpful for surface finishing or blending with fibres and polymers [2, 5, 25]. Scientists have developed novel techniques to separate and recover sericin from the silk degumming effluents due to the possible applications of sericin. Coagulation [12], membrane filtration [3,6], freeze and tray-drying [19], precipitation [15, 20, 24], and enzymatic hydrolysis are a few of the methods that have been reported [19, 21]. Chitosan molecules that have been quaternized act as coagulants to draw sericin out of the silk degumming solution [10]. They investigated how pH and coagulants, quantity and settling time in sericin elimination. Using 1 g/L of 2-hydroxypropyltrimethylammonium chloride chitosan (HACC) at pH 8, sericin recovery yield of 76% was observed. The recovery rate using membrane technique was 97%, but the treatment process using their method required about 3–4 hours, and the product obtained contained both sericin and coagulant [6]. They used the degumming method' solution in hot, purified water with a 3–4 bar pressure. The recovery yields of 37–60% and 94–95%, respectively, of high weight molecular sericin from the degumming solution were then obtained by using ultra filtration and nanofiltration techniques. They also noted that the primary obstacle to sericin separation was a severe membrane flux decline (58–75%). The separation of sericin from the fatty acids using centrifugation, low temperature crystallisation, and ultrafiltration was the subject of a different research [3]. They made the effluent more acidic and separated the soap solution's free fatty acid from the salt. Once the free fatty acids had crystallised, the solution's temperature was lowered to 4 °C to create the sericin precipitate. To extract sericin from the degumming solution, [19] used freeze-drying and then membrane filtration. Alkalise enzyme was used to convert the retrieved sericin into short-chain peptides or sericin hydrolysate. However, the sericin that was recovered using this technique had a high ash content and required a lot of energy to dry. They emphasised the significance of removing the alkali ions and reducing the amount of the solution before the drying stage. They found that the ultrafiltration method with membrane (20,000-30,000 Da cut-off) recovered 94% of sericin with a molecular weight of 2427-9863 Da [24]. According to reports, the ultrafiltration procedure is necessary to remove impurities from the degumming fluid. However, they found no evidence of a decline in membrane flux efficiency in their study. Using ethanol at various concentrations, [20] isolated high molecular sericin from the silk degumming solution. According to their results, a sericin extraction yield of 71% (w/w) was obtained by adding 90% (v/v) of ethanol to the solution. Although a high percentage of sericin was extracted, their technique does not appear to be environmentally friendly because it uses a lot of ethanol and is not appropriate for industrial size applications. In order to precipitate the surfactant, sodium oleate, [24] added calcium chloride to the degumming solution. They then retrieved the sericin from the supernatant solution. To separate the sericin solid components, the sediment underwent dialysation and freezing [24]. UV-vis spectra with a high intensity at 280 nm were used to determine the concentration of sericin in the solution. They claimed that by adding 10% calcium chloride to the fluid, sericin was recovered to a level of 75.6%. However, owing to sericin degradation in the precipitation phase or sericin removal during centrifugation, this method was unable to recover the high molecular weight sericin residue. As a result, the presence of particles in the liquid dried compound and RSS powder produced by the freeze-drying technique in this study's SEM images suggests that the drying process itself is what gives rise to the particles. The micrographs of the sericin powder at each pass revealed different particle types, sizes (1–5 m, 5–20 m), and size distributions. A scanning electron microscope was used to examine the morphology of sericin freeze dried RSS granules at various magnifications. RSS granules are a useful substance in many ways and a biodegradable substance.

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CONFLICT OF INTEREST
All authors must declare no conflict of interest or any affiliation or involvement in any organization whether it is academic, commercial, financial, personal and professionally relevant to the work under consideration to avoid the potential for bias and accept responsibility for what is said in the manuscript.

AUTHOR'S CONTRIBUTION
Each author contribution should be clearly mentioned.

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