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Extraction of sericin freeze dried RSS powder a biodegradable compound from silk coccons

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Abstract
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. Silk fibre is used in many industries, such as as the textile, medical, as well as industrial sectors. Silk is a lengthy, thin, and light fibre. The current work reveals that the shape and structure of sericin freeze-dried RSS powders at different magnifications was examined using a scanning electron microscope. RSS powders are a useful one in numerous aspects and also a biodegradable substance.

Keywords: Silk fibre, sericin, freeze dried RSS powders, biodegradable substance

Introduction
Silk fibre used in several industries including textile, medical, and industrial. A long, delicate, and light fibre is silk. The water absorption, affinity for the process of dyeing thermostoichiometry, insulating qualities, and gloss are widely recognised. This is used to make parachutes, artificial blood vessels, surgical sutures, and precious textiles. The secret to producing designer silk strands with enhanced mechanical and visual properties may be ionic liquid.

The posterior portion of the silk gland lumen's columnar fibroin is composed of numerous spherical clusters containing fibroin fibres. These developed into a homogeneous and compact mass in the anterior silk gland, with greater numbers concentrated at the rear of the centre portion of silk gland. Observations of different regions of the silk gland reveal that the cocoon filament is composed of orientated elementary fibroin fibres, which are formed from fibroin fibres when they exhibit structural change while passing via the lumen of silk gland. The structure and changes of sericin's molecular aggregating 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. However, because hot water treatment makes the orientation unstable, here is a reversal causal connection between non-orientation and orientation. Sericin can be combined with various resins to create biodegradable polymers that are good for the environment [1].

By mixing a admixture similar as a di- butyltin di- laurate, di- isocyanet, polyol, tolylene and trichloro mono fluoro-methane in a proportion of sericin (0.01 to 50 percent w/ w), one can employ sericin to induce polymer flicks, lathers, moulding resins, and fibres. The polyurethane form containing sericin has a humidity immersion/ desorption rate that's between two and five-fold advanced than the control form. also, styles for creating polyurethane with sericin and excellent thermal and mechanical features are presently stated [8].

The separations process is Membrane-based which are used in water desalination, water that is extremely pure manufacturing, bioprocessing, as well as certain chemical treatments (which include dialysis, microfiltration, reverse osmosis and ultra-filtration). Since sericin has a lot of amino acids with neutrality functional groups that are polar, it can be co-polymerized, crosslinked, or combined with other materials easily to form 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. By co-polymerizing particular synthetic polymers, such as acrylonitrile, with sericin including synthetic polymer screen enabling separating water from other organic substances can be created [24, 25].

According to research by [14], a sericin coating placed to a fluid crystalline surface can evenly distribute the molecules of the water-based crystal, resulting in free of distortion, excellent displays. Due to its anti-frosting properties, sericin-coated film is used on the surface of refrigeration gear [16]. A great way to prevent frost in deep freezers, refrigerators and freezers refrigerated cars, as well as in ships is to use coated sericin film. Reduced risk of damage from frost can also be achieved by applying the coated screen on roofs and roadways. Many resistant substances' surfaces can be coated with sericin protein to improve performance [10]. Paint hues and product surface shielding can both be achieved with sericin. The material covering the sericin is extremely porous, weatherproof, and does not distort when exposed to air to dry.

Serin blends well with soluble in water polymers, particularly polyvinyl alcohol (PVA). Sericin, fibroin, and PVA hydrogels are reported to be highly elastic and possess exceptional moisture absorption and release properties [25]. The hydrogel can be utilised in skin care and medical items, as well as to enhance the quality of soil. In order to immobilize enzymes, [12] described using glutaraldehyde as the cross-linking component in an interconnected sericin film. The halted J-glucosidase on the interconnected sericin sheet is more stable and resistant to electro-osmosis than the unbound enzyme.

Silk fibres have demonstrated benefits in several clinical applications over time. However, biological responses to the protein have prompted concerns over biocompatibility. a silk fibroin bandage for wounds that accelerates healing and may be taken off without doing any harm to the newly developed skin [17]. The non-crystalline the fibroin coating of the dressings had an overall thickness of 10–100 m and a water concentration of 3–16%. The wound covering was then created by combining sericin and fibroin [18]. As an alternative to the protein collagen, membranes composed of sericin and fibroin can be employed to encourage the development of sticky mammalian cells.

During the latter phase of larval development, the silkworm B. mori produces a large amount of silk proteins. 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.

Materials 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 temperature and duration of the extraction process affect the amount of sericin that is extracted. By using this procedure, sericin breaks down, yet it doesn't lose its vital characteristics. 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. The process of degumming is used to achieve this, and the results are disposed of in the effluent. The recovery of sericin from decomposition spirits can reduce the amount of wastewater produced, reducing its impact on the environment and creating a biopolymer with a range of beneficial characteristics.

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. For the examination of the figure of sericin freeze-dried powders at various magnifications, including 2000X, 6000X, 12000X, 18000X, 30000X, and 40000X SEM (Scanning Electron Microscope) was used.

The sizes (1-5 m, 5-20 m), figures (donut-shaped, smooth, straight, wrinkled or a combination of all), and particle size distributions based on photographic micrographs evaluation of the (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.

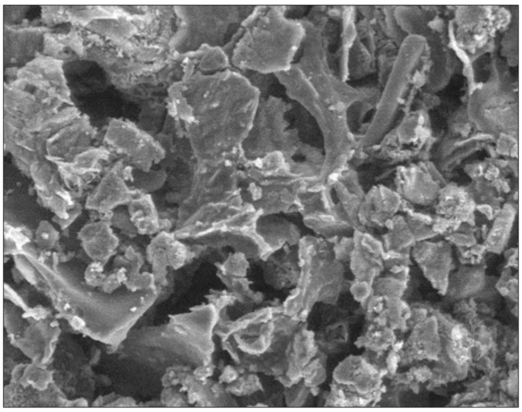

Fig 1: SEM image of sericin samples (Freeze Dried) 2000X magnification

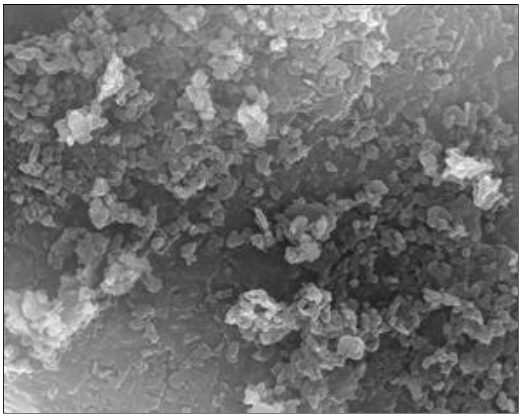

Fig 2: SEM image of sericin samples (Freeze Dried) 6000X magnification

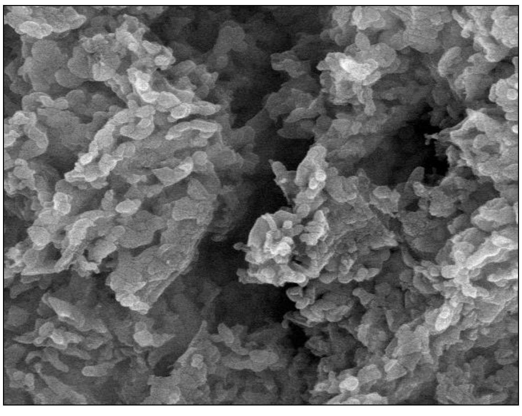

Fig 3: SEM image of sericin samples (Freeze Dried)12000X magnification

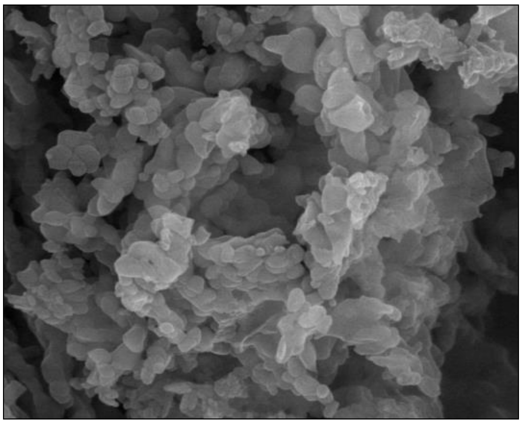

Fig 4: SEM image of sericin samples (Freeze Dried) 18000X magnification

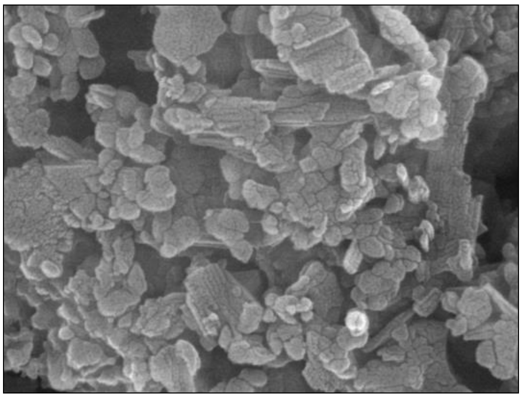

Fig 5: SEM image of sericin samples (Freeze Dried) 30000X magnification

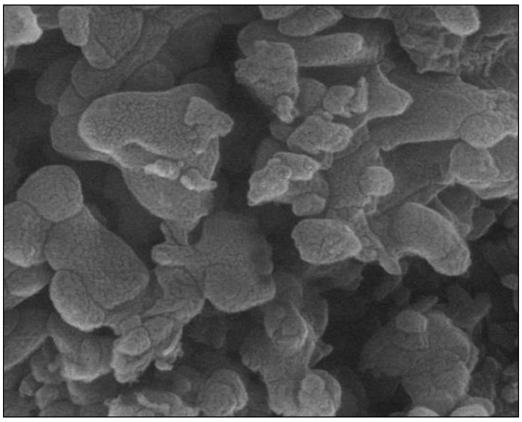

Fig 6: SEM image of sericin samples (Freeze Dried) 40000X magnification

Discussion

Sericin is used in the textile industry for integrating and finishing the surface using polymers and fibres. [2, 5, 25]. Because sericin has potential uses, researchers have created innovative methods for isolating and recovering the compound sericin via silk degumming waste materials (i.e effluent). Several techniques have been documented, including coagulation [12], membrane filtration [3, 6], tray-drying and freezing [19], enzymatic hydrolysis [19, 21] and precipitation [15, 20, 24]. Quaternized chitosan molecules work as coagulating agents to extract sericin from the solution of silk degumming process [10]. A 76% sericin recovery yield was noted applying 1 g/L of 2-hydroxypropyltrimethylammonium chloride chitosan (HACC) under pH 8. The membrane technology yielded a 97% recovery rate; however, the treatment procedure took around 3-5 hours to complete, and the final product comprised both coagulant and sericin [6]. They applied a 3–4 bar pressure to heated up, filtered water while using the degumming method's solution.

Then, employing ultra filtration and nanofiltration procedures, heavy-weight molecular sericin extraction rates of 37–60% and 94–95%, respectively, were achieved through the degumming fluid. They also observed that a considerable membrane flow reduction (58–75%) was the main hindrance to sericin separation. A different study looked at the extraction of sericin from the fatty acids by the process of centrifugation, lower temperatures for crystallisation, and ultrafiltration to remove contaminants [3]. They separated the unbound fatty acids in the soap solution from the sodium chloride and increased the acidity of the effluent. The sericin precipitate was produced by lowering the temperature of the solution to 4 °C after the unbound fatty acids had crystallised.

In order to separate sericin from the degumming solution, [19] employed filtration using membranes followed by freezing and drying. The recovered sericin was transformed into small length chain peptides that or sericin hydrolysate substrate using an alkalise enzyme. But the sericin that was recovered with this method needed a lot of energy to dry and had a significant ash percentage. The need of eliminating the alkali particles and lowering a solution's volume prior to the (drying) dehydration phase was stressed. 94% of sericin with a molecular weight range of 2427–9863 Da was recovered using the ultrafiltration technique with membrane (20,000–30,000 Da the limit), according to their findings [24]. Reportedly, the ultrafiltration process is required to rid the degumming solution of contaminants. In their investigation, they could not discover any proof of a decrease in membrane flux efficiency, though. High molecular sericin was extracted out of the silk degumming liquid by [20] using ethanol at different concentrations.

Increasing 90% (v/v) of ethanol with the solution produced a 71% (w/w) sericin separation yields, in accordance to their findings. Their method does not seem to be environmentally benign because it takes a lot of ethanol and is not suitable for industrial-scale applications, even though a high proportion of sericin was extracted. Calcium chloride was introduced to the degumming solution by [24] in order to precipitate the surfactant, which is sodium oleate. Subsequently, the sericin was extracted via the extract of supernatant. To isolate 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, dimensions (1–5 m, 5–20 m), and dimensions distributions. SEM (scanning electron microscope) was employed for the examination of the sericin morphology, 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
It is mandatory for all authors to disclose any affiliations or i nvolvements in organisations, whether academic, commerci al, financial, personal, or professionally related to the work being reviewed, in order to prevent bias and to take account ability for the content they write.

Author's Contribution
Every author's contribution ought to be acknowledged expli citly.

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