



How can different degumming treatments help the valorization of waste silk for epoxy composite applications?

Tommaso Pini ^{a,b}, Laura Simonini ^c, Giulia Fredi ^c, Maria Paola Bracciale ^{a,b},
Alessandro Pegoretti ^c, Marco Valente ^{a,b}

^a Department of Chemical Engineering, Materials, Environment, Sapienza University of Rome, Rome, 00184, Italy

^b INSTM Reference Laboratory for Engineering of Surface Treatments, UdR Rome, Sapienza University of Rome, Rome, 00184, Italy

^c Department of Industrial Engineering, University of Trento, Via Sommarive 9, Trento, 38123, Italy

ARTICLE INFO

Keywords:

Interfacial strength

Silk waste

Natural fiber composites

Microdebonding test

ABSTRACT

Valorizing silk textile waste as composite reinforcement requires removing the sericin coating (degumming) without damaging the load-bearing fibroin or the fiber/matrix interface. Using floss *Bombyx mori* silk waste, the effect of five degumming protocols, alkaline NaOH and Na₂CO₃ baths and a mild hot water treatment, on the silk/epoxy interface was investigated. The interfacial shear strength (IFSS) was measured by microdebonding, the single-fiber tensile strength and apparent modulus by single-fiber testing with Weibull statistics, and the fiber chemistry and morphology by FT-IR and SEM. Conventionally computed from a circular cross-section, these properties were re-evaluated from the measured, non-circular geometry of the same fibers, which raised the raw stress about 1.65 times. The three properties give a single ranking: the mild hot water treatment, which removes only about 21% of the mass and leaves residual sericin, simultaneously yields the highest IFSS (9.0 MPa), tensile strength ($\sigma_0 = 454$ MPa) and apparent modulus ($E_0 = 11.3$ GPa). It performs best despite, not because of, its residual sericin: gentle degumming preserves the fibroin surface and core, whereas aggressive alkaline routes fully strip sericin but expose and damage the fibroin. Mild hydrothermal degumming thus emerges as a favorable compromise for silk waste epoxy composites.

1. Introduction

Silk has been adopted by mankind for over 5000 years, transitioning, in recent years, from an exclusively luxury textile to an estimated resource in different biomedical and engineering applications [1]. Silk is produced by mulberry silkworms under the form of continuous fibers, and it is the building material for the cocoons that protect the larvae during reproduction stage. A single silk fiber is constituted of two parallel fibroin fibrils, coated by a sericin layer that acts as a binder. Fibroin is a protein with a high content of glycine and alanine; it has a hierarchical structure made of β -sheet nanocrystals and it is responsible for the unique combination of strength and extensibility of silk. On the other hand, sericin is an amorphous water-soluble protein that gives little contribution to the mechanical properties of silk [2–4].

The industrial cycle of processing silk into textiles begins with unraveling the cocoons after sericin is removed by the action of temperature and alkaline solutions. This process is referred to as degumming and its effects on the chemical, physical and mechanical properties was investigated by several scholars [5–8]. Once the sericin is scoured, silk

fibers are further reeled, spun and weaved into fabrics. Due to the strict aesthetic standards of high-end fashion industry, a large fraction of the initial raw material is discarded during the process, downgraded to low values products or becomes waste destined for incineration or land-filling [9]. These practices determine a burden for the environment, especially for the countries with large sericulture practices, and they do not meet the criteria for sustainability and circular economy. On the contrary, recycling and valorization of silk waste into composite materials would be a viable and sustainable option [10–13].

On one hand natural fiber composites address the needs of biodegradability and sustainability but offer, in general, poorer performance than composites containing synthetic fibers such as carbon and glass. Moreover, natural fibers present specific challenges related to the variability of their properties. Tensile strength of silk fibers spans a wide range of values in literature, depending on different factors such as degumming conditions [5,14,15] and the techniques adopted to determine the cross-sectional area of the fibers [16–18]. Often,

* Corresponding author.

E-mail address: tommaso.pini@uniroma1.it (T. Pini).

¹ Present address: Department of Mechanical and Aerospace Engineering, Sapienza University of Rome, 00184 Rome, Italy.

Weibull statistics were used to describe the low reproducibility of the mechanical properties of silk [19,20].

Examples of studies on silk-based composite materials are present in the literature, underlining the high potential of this fiber, especially due to its high specific properties and large deformation at break. Hence, the energy-absorption capability of silk-based composite has been investigated [21–24]. The specific mechanical properties of these materials outperformed glass-based counterparts; in addition, it was found that to fully exploit silk reinforcing and energy-absorbing properties, a critical fiber length and the right coupling with a ductile matrix must be sought. Fiber volume fraction was also found to be a critical aspect, with an effective contribution of the silk fiber obtained only at high fiber content [25–27]. Hamidi et al. [28] investigated silk/epoxy composites prepared via resin transfer molding, finding properties comparable to those of glass/epoxy composites. Their results, however, revealed extended matrix/fiber debonding, underlining the importance of improvement of interfacial adhesion for better performance. Craven et al. [29] studied the interfacial adhesion between silk and epoxy by means of microbonding test, finding a mean interfacial shear strength value of 15 MPa. Their results also pointed out the difficulties of this type of investigation, due to the intrinsic variability of the cross-sectional area and tensile properties of silk fibers.

Studies in the literature have shown that surface treatments, such as sizing or sericin removal, alter the mechanical properties of silk-based composites [5,30–32]. Degumming may enhance the compatibility between the fibroin core and different matrices. Moreover, the roughness of the residual surface and possible mechanical interlocking between fibroin micro-fibrils have also been invoked as contributing factors [33]. At the same time, degumming is known for altering the structural properties of silk, potentially having a detrimental effect on the properties of the final composite. Novel degumming techniques, which rely less on chemical treatments, proved to better preserve the structural integrity of silk fibers [34]. However, to investigate the applicability of silk waste for composite production, degumming treatments commonly adopted by industry should still be considered.

In this work the effects of different degumming methods on the properties of silk fibers and on the silk/epoxy interface were investigated. With the goal of assessing whether silk waste would be a good candidate for composite production, a preliminary screening of the relationship between degumming conditions, interfacial adhesion and fiber mechanical properties was carried out. Tensile strength and apparent modulus of single fibers and the interfacial shear strength of silk/epoxy were measured, corroborating the results with molecular (FT-IR) and morphological (SEM) analyses. A central methodological feature of the present study is that the tensile and interfacial properties, which were originally computed assuming a circular cross-section of fixed diameter, were re-evaluated using the measured cross-sectional geometry of the same fibers reported in a previous work [18].

2. Materials and methods

2.1. Materials

Silk fiber waste was provided by TecnoSeta Srl (Zagarolo, Italy). The material used for this study was the floss silk, or anchoring silk, produced by the *Bombyx mori* silkworm to secure the cocoons to the mulberry trees. Unlike cocoons, this type of silk appears as a loose, fluffy and random web of entangled fibers.

An epoxy base (EC 157.1) and an aminic hardener (W 342) were provided by Elantas Italia S.r.l. (Collecchio, Italy). From the technical datasheet, they had viscosity of 600 mPa s and 30 mPa s, respectively, and density of 1.14 g/cm³ and 0.94 g/cm³, respectively. The two components were mixed at a weight ratio of 100:30 and then cured for 24 h at room temperature and 16 h at 50 °C, following the optimal specific curing process determined in [35]. The following properties of cured epoxy were determined in-house in previous experimental

campaigns: glass transition temperature (T_g) of 84 °C, thermal degradation temperature of 352 °C, elastic modulus of 3.1 ± 0.2 GPa, tensile strength of 61 ± 15 MPa. The large variance of the tensile strength may be attributed to intra-specimens differences in terms of presence of defects and local degree of cross-linking.

NaOH was used in form of pellets and Na₂CO₃ was used in form of powder; both were supplied by Thermo Scientific (Waltham, MA, USA).

2.2. Sample preparation

Silk fibers were degummed using combinations of different solutions, temperatures and durations. Alkaline degumming relies on the hydrolysis and dissolution of sericin promoted by hydroxide ions. Sodium hydroxide (NaOH) is a strong base that raises the bath pH sharply and is an aggressive but effective sericin solvent, whereas sodium carbonate (Na₂CO₃) is a weaker, milder alkali that buffers the bath at a lower pH and is the reagent most commonly used in conventional industrial degumming. Hot pure water removes sericin only by thermal swelling and dissolution, without the chemical assistance of an alkali, and is therefore the gentlest of the routes considered here [8,36].

Four degumming treatments were investigated using three different media; a summary is given in Table 1. Treatments DEG_A, DEG_B, and DEG_E were carried out in NaOH solutions, whereas DEG_D was performed using a Na₂CO₃ solution. An additional treatment (DEG_C) was performed using hot pure water. In all cases, a bundle of raw fibers, weighing approximately 0.10 g, was placed in a beaker containing 200 mL of solution on a heating plate and held for the set time. In all treatments the material-to-liquor ratio was approximately 1:2000 (0.10 g of fibers in 200 mL of bath), corresponding to a fiber concentration of about 0.5 g/L. The alkaline baths were prepared at 0.5 wt% in all cases. The bath pH was not measured analytically; the nominal values were calculated from the reagent concentrations (13.1 for the NaOH baths, 11.5 for the Na₂CO₃ bath and neutral for pure water). The solution was then filtered, the fibers were rinsed with abundant distilled water and left to dry in air 24 h prior to subsequent steps. For each treatment a single bundle was weighed before and after the degumming, on an analytical balance with a resolution of 0.1 mg. The weight loss reported in Table 1 is the relative mass change $(m_i - m_f)/m_i$, where m_i and m_f are the bundle mass before and after treatment. Because a single bundle was processed per condition and the fibers were air dried but not conditioned to a controlled humidity, this value is treated as a semi-quantitative, relative indicator of sericin removal rather than as an exact sericin balance.

2.3. Molecular and morphological characterization

Molecular characterization of silk fibers, both raw and degummed, was performed by Fourier Transform Infrared (FT-IR) spectroscopy. Spectra were collected with a Bruker VERTEX 70 spectrometer equipped with a diamond attenuated total reflectance (ATR) cell. Data were acquired in the mid-infrared region (400–4000 cm⁻¹), averaging 256 scans at a spectral resolution of 2 cm⁻¹. Three spectra were acquired for each sample. The spectra were treated as reported in Appendix A.

The secondary structure composition was evaluated by deconvolution of the amide I band, following the approach proposed by Bucciarelli et al. [36,37]. Each spectrum was cropped to 1580–1720 cm⁻¹ region and a linear baseline (estimated from the mean of four edge points on each side) was subtracted. The curve was then smoothed by 5 points moving average. Fourier self-deconvolution (FSD) and the minima of the second derivative, which were found to be mutually consistent, were used to fit a constrained sum of Gaussians distributions. Although an isolated vibrational transition has a Lorentzian profile, the protein amide I envelope is broadened inhomogeneously by the distribution of hydrogen-bond geometries and backbone conformations, which yields approximately Gaussian component shapes;

Table 1
Sample labeling and weight loss after degumming.

Label	Degumming solution	Nominal pH [-]	Degumming temperature [°C]	Degumming time [min]	Weight loss [%]
RAW	—	—	—	—	—
DEG_A	NaOH 0.5 wt%	13.1	40	50	40.0
DEG_B	NaOH 0.5 wt%	13.1	60	25	42.5
DEG_C	H ₂ O	7.0	90	50	21.4
DEG_D	Na ₂ CO ₃ 0.5 wt%	11.5	90	50	42.6
DEG_E	NaOH 0.5 wt%	13.1	60	50	42.2

Gaussian curve fitting is the established approach for amide I secondary structure analysis [38–40]. The sensitivity of the results to the assumed line shape is examined in Appendix B. The fitted components were assigned to six structural classes (random coil, α -helix, turns, native β -sheet, parallel and antiparallel β -sheet). Each component full width at half maximum was constrained to a physically reasonable value ($\text{FWHM} \leq 15 \text{ cm}^{-1}$). Secondary structure fractions, being area ratios computed within a single amide I band of a single spectrum, are independent of the absolute absorbance and are therefore unaffected by the normalization. The deconvolution was applied independently to each of the three spectra, and the secondary structure fractions are reported as mean $\pm$ SD ($n = 3$).

The fibers were observed by using a scanning electron microscope (SEM) Tescan Mira 3 (Brno, Czech Republic) operating at an accelerating voltage of 10 kV. Prior to the analysis, the samples were gold coated, to enhance electrical conductivity, with an Edwards Sputter Coater S150B (Burgess Hill, UK).

Microdebonding test samples were observed by using a field emission scanning electron microscope (FESEM) Zeiss Supra 40 (Carl Zeiss AG, Oberkochen, Germany), operating at an accelerating voltage of 3.5 kV. Prior to the analysis, the samples were coated with a layer of a platinum/palladium (Pt/Pd) alloy (80:20) with a thickness of about 5 nm.

2.4. Mechanical tests

Single fibers were extracted from the bundles with tweezers, taking great care and discarding those fibers that were too entangled to avoid excessive stresses and pre-tensioning. Typical lengths of the extracted fibers were between 40 and 80 mm. After the fibers were isolated, they were mounted on paper frames with cyanoacrylate glue. The gage length of the frame adopted in this work was 30 mm. The frames, each containing a single fiber, were used for the evaluation of the interfacial adhesion with microdebonding test and for the single fiber tensile test. Microdebonding tests were performed using an Instron 5969 universal testing machine (Instron Inc., Norwood, MA, USA), which was equipped with a 2.5 N load cell. Epoxy droplets were placed on the single fibers mounted on paper frames using micropipettes (Fig. 1) and cured as described in Section 2.3. Droplet lengths were measured from collected optical microscopy pictures (Nikon SMZ25, Nikon Corporation, Tokyo, Japan) using the ImageJ software (version 1.53a) and were between 60 and 120 μm . An example is shown in Fig. 1.

Load data were recorded during the microdebonding test at a displacement rate of 0.75 mm/min. A comprehensive description of the device and methodology employed can be found in Simonini et al. [41–44]. One droplet was debonded per fiber, with a total of five fibers tested per sample.

The interfacial shear strength (IFSS) was obtained as:

$$IFSS = \frac{F_{\max}}{Pl} \quad (1)$$

where $F_{\max}$ represents the maximum load registered during the microdebonding test, l is the droplet length and P is the perimeter of the cross-section of the fiber. Under the assumption of a cylindrical fiber, P was taken as the cylindrical perimeter πd , with the equivalent diameter d assigned as 20 μm and 10 μm for raw and degummed fibers,

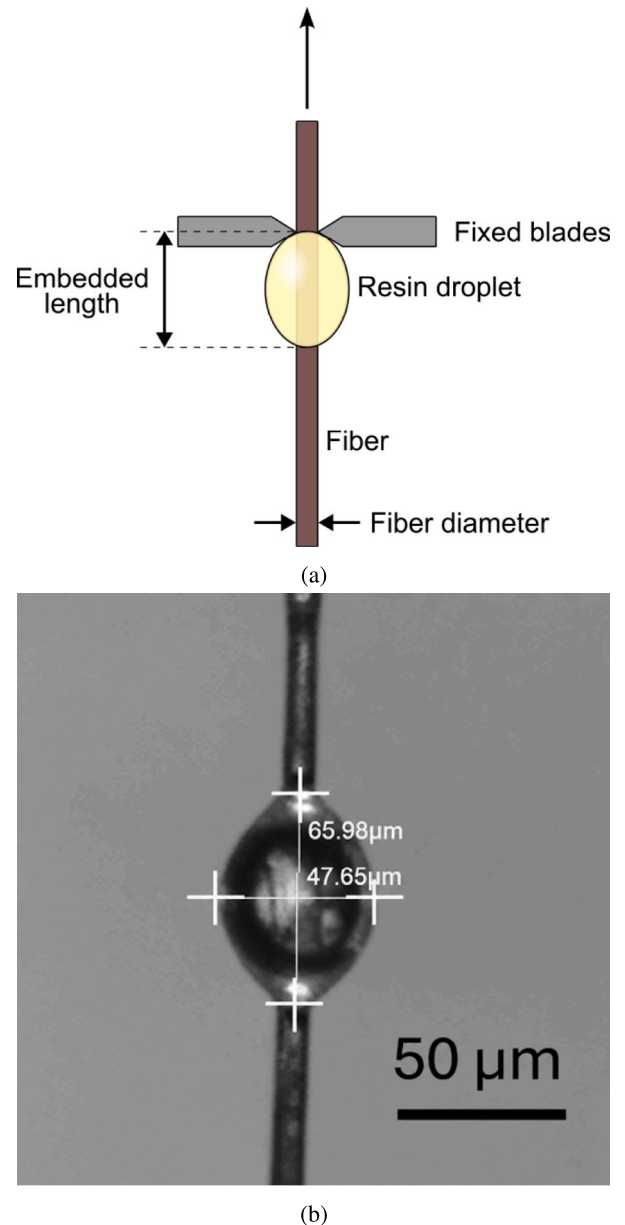


Fig. 1. (a) Microdebonding test schematic representation and (b) OM micrograph of a prepared sample.

respectively. As detailed in Section 2.5, this cylindrical assumption is replaced by the actual measured perimeter.

Single fiber tensile tests were performed by using an Instron 5969 universal testing machine (Instron Inc., Norwood, MA, USA) equipped with a 100 N load cell. Tests were performed at crosshead speed of 2 mm/min. Because the present campaign was intended as a comparative screening, the effect of the gage length was not investigated

Table 2
Measured [18] and nominal cross-sectional geometry of raw and degummed fibers.

Quantity	Raw – measured	Raw – circular	Degummed – measured	Degummed – circular
Area [μm^2]	190 ± 42	314.2	68 ± 14	78.5
Perimeter [μm]	58 ± 9	62.8	32 ± 3	31.4

and a single 30 mm gage length was used. Weibull distribution was used to assess the Weibull scale parameter (σ_0) and the Weibull shape parameter (m). The parameters were obtained from the linearized Weibull relation reported in Eq. (2):

$$\ln \left[\ln \left(\frac{1}{1 - P_f} \right) \right] = m \ln \sigma - m \ln \sigma_0 \quad (2)$$

where P_f is the probability of failure. Moreover, the elastic modulus (E) of the fibers was determined from the tensile stress–strain curves. In particular, it was calculated from the steepest portion within the initial elastic region of the curve using a local linear fitting. This method was adopted to minimize the influence of initial settling effects, fiber alignment, or nonlinearities. Because no system-compliance calibration (e.g. ASTM C1557, based on multiple gage lengths) was performed in this campaign, the reported values are referred to throughout as apparent moduli: the area correction (Section 2.5) rescales the stress axis only and cannot remove the contribution of machine compliance and grip slack to the measured strain.

2.5. Cross-sectional geometry correction and statistical approach

Tensile stress and IFSS were originally computed assuming a circular cross-section and a fixed, assigned diameter (20 μm raw, 10 μm degummed). No per-fiber geometry was measured in this work. This assumption is acceptable for the nearly circular degummed brins but is a large approximation for the raw fiber, whose cross-section is closer to an ellipse or bean shape. The cross-section parameters were corrected with the approach proposed in [18], where the same raw and degummed fibers (DEG_E from the present work coincides with DEG from [18]). Cross-section images exist only for the raw and DEG_E conditions (from the previous campaign). We assume that the degummed geometry (DEG_E) is representative of all degumming protocols, i.e., that any sub-optical-resolution surface effect specific to a given protocol does not change the measured area or perimeter of the brin significantly. Accordingly, DEG_A–E share the same correction parameters. In addition to the cross-sectional areas analyzed previously, the perimeters were extracted from the same image set. Two corrections follow (Table 2, Fig. 2): the cross-sectional area correction raises the tensile raw stress by a factor of ~ 1.65 and the degummed one by ~ 1.16 . On the other hand, the perimeter correction is rather small in magnitude. The area correction is large while the perimeter correction is small because the area scales quadratically with the shape distortion of the section, whereas the perimeter scales approximately linearly. Strain is geometry-independent and is therefore unaffected by either correction. The effect of sharing correction parameters is limited: the correction enters a single multiplicative factor for each group, one for the raw and one for the degummed, so it rescales their relative values, but it cannot reorder degummed values among themselves, i.e. the DEG_A–E ranking is independent of the assumed geometry. Moreover, the perimeter correction that enters IFSS calculation is below 1% for the degummed fibers, so the interfacial results are basically insensitive to any difference between degumming protocols, while the larger area correction that enters the tensile stress already carries its uncertainty through the measured degummed area distribution ($68 \pm 14 \mu\text{m}^2$) in the Monte Carlo procedure reported below.

The sample sizes available in this campaign were small: $n = 5$ – 7 per condition for single fiber tensile tests and $n = 4$ – 6 for IFSS. Three distinct statistical operations were applied:

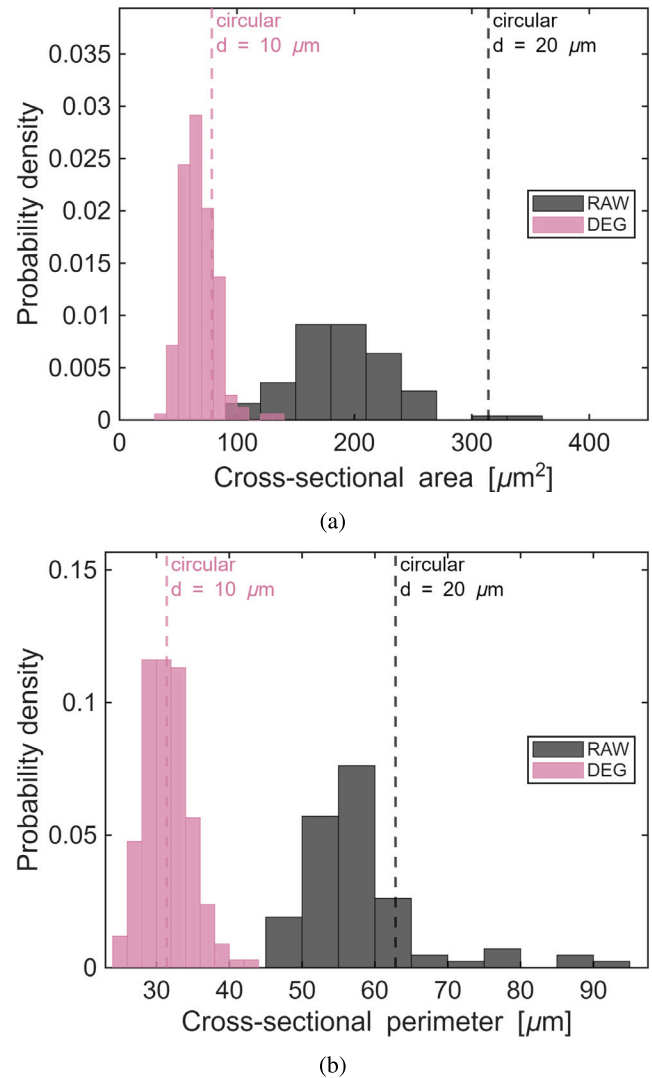


Fig. 2. Empirical distributions of the measured (a) cross-sectional area and (b) perimeter for raw and degummed fibers (from [18]). Dashed lines mark the assigned cylindrical values.

- Weibull small sample bias correction: strength and apparent modulus are described by two-parameter Weibull distributions (Eq. (2)). At small n , the maximum-likelihood estimate of the shape parameter m is biased upward [46]; it is therefore multiplied by the standard unbiasing factor $b(n)$ (≈ 0.67 at $n = 5$, ≈ 0.75 at $n = 7$, ≈ 0.91 at $n = 22$). This bias correction affects only the shape parameter.
- Bootstrap confidence intervals: the bootstrap estimates how much the fitted Weibull parameters would change if the experiment were repeated, by re-drawing new samples from the data already collected. To quantify the sampling uncertainty of the Weibull estimates at small n , the measured values of each condition were resampled with replacement to the same size. Then, Weibull distributions were re-fitted and the procedure was repeated 1500

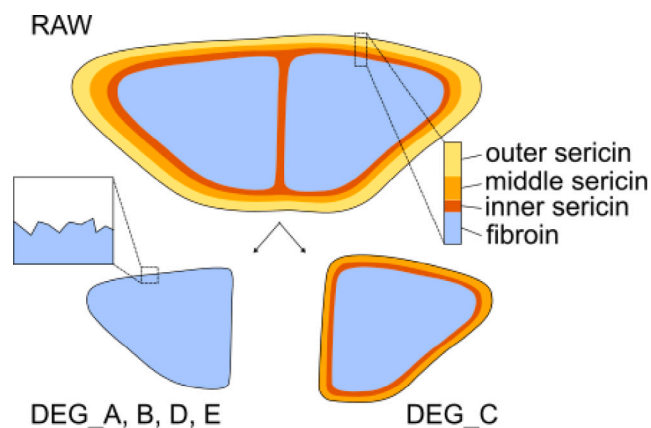


Fig. 3. Schematic of the fiber structure; RAW fiber shows the two fibroin brins and the layered sericin; the outer, middle and inner layers account for about 50, 35 and 15% of the sericin mass [45] and are drawn with their areas in this ratio. The degummed brins, showing completely (DEG_A, B, D, E) and partially (DEG_C) removed sericin are shown at the bottom. The irregular outline of the alkaline-degummed brin hints the degradation of the fibroin surface possibly occurring at harsher conditions.

times, finally obtaining the 95% confidence intervals. As expected for small n , the intervals are wide and asymmetric.

- Monte Carlo propagation of the measured geometry: the corrected stress and IFSS each divide a force-like quantity by a geometric quantity (area or perimeter) that is itself a measured distribution rather than a single value. Instead of assuming a form for the distribution, the geometry was resampled directly from the per-fiber measurements of [18] (84 and 168 cross-sections for raw and degummed, respectively). For each tested fiber the force quantity was recovered, a measured area (for stress) or perimeter (for IFSS) was drawn at random with replacement, and the corrected quantity was computed. The process was repeated 20,000 times, yielding the corrected average value and its 95% confidence interval. Empirical resampling is preferable here because the measured distributions are right-skewed, so a normal model would misrepresent them and would bias the corrected value. Resampling makes no distributional assumption and requires no truncation of non-physical values.

Because the IFSS sample sizes ($n = 4\text{--}6$) are too small for a meaningful Weibull shape estimate, the IFSS is reported as mean $\pm$ SD together with the Monte Carlo confidence interval.

3. Results and discussion

3.1. Fiber morphology and molecular structure

The weight loss after degumming (Table 1) provides a first, macroscopic measure of sericin removal. It cannot be taken exactly as the sericin content, since the mass change also reflects broken fibers leaving the bundle, impurities, uncontrolled moisture, and possible removal or degradation of the fibroin itself under the harsher treatments. It is therefore used here as a relative indicator, so the reading below rests on the ratio between conditions rather than on the absolute value. Nevertheless, the four alkaline treatments (DEG_A, DEG_B, DEG_D and DEG_E) all removed a comparable amount of material, 40%–43% of the initial bundle weight, somewhat above the sericin fraction usually reported for *Bombyx mori* silk (about 20%–30%), the difference being attributable to the source of errors noted above, indicating essentially complete degumming under these conditions. The weight loss for DEG_C, by contrast, was only about 21%, suggesting that pure water at

90 °C acts as a far gentler scouring medium than the alkaline baths and leaves a substantial residual sericin fraction on the fiber. This graded response to the two media can be read in terms of the layered organization of the sericin coat (shown schematically in Fig. 3). Sericin is not homogeneous but arranged in three sublayers of decreasing water solubility from the outside inward: an outer layer soluble in near-boiling water, a middle layer that requires higher temperature and pressure, and an inner layer, adjacent to the fibroin, that is removed only with alkali treatments [45,47]. Since the weight loss also includes impurities and broken fibers, it is used here as a relative rather than an absolute measure; on that basis, the DEG_C loss (about 21%) is close to one half of the alkaline loss (~42%), consistent with hot water removing essentially the outer layer, reported to represent about half of the sericin mass, while leaving the deeper, fibroin-bound layers in place [45].

The picture inferred from the weight loss can be observed directly in the fiber morphology by SEM (Fig. 4). The raw fiber is covered in a continuous sericin layer that binds the two fibroin brins together and gives the fiber a smooth, occasionally cracked, outer surface in which the individual brins are not resolved. After alkaline degumming the sericin layer is removed and the individual fibroin brins are exposed, revealing the texture characteristic of the fibroin core. The DEG_C fiber is intermediate: the brins are largely separated, but the surface retains thin, patchy sericin residues that appear as partially detached, layered sheets, suggesting that the mild hot water treatment exfoliates the outer sericin while leaving the deeper layers in place. This offers a direct visual counterpart to the layered organization of the sericin coat inferred above from the weight loss and is consistent with the low weight loss of DEG_C and, as shown below, with its elevated residual sericin marker in the FT-IR spectra. The aggressive alkaline routes (in particular the NaOH solutions, DEG_A, DEG_B and DEG_E) are expected to be the most likely to etch or fibrillate the fibroin surface, creating the kind of surface irregularities that can act as stress concentrators in a tensile test [14,48]. The gentler DEG_C treatment, which preserves part of the sericin and exposes less of the fibroin to the bath, would be expected to leave the smoothest, least-etched surface. This morphological picture is fully consistent with the mechanical ranking reported below and is discussed in Section 3.4.

Fibroin and sericin differ significantly in amino acid composition and structural organization, leading to distinct IR spectral features. Therefore, FT-IR spectroscopy can be adopted to qualitatively compare the degumming treatments and evaluate their effectiveness. Fibroin is characterized by a highly ordered crystalline structure, predominantly composed of stacked β -sheet configurations. This translates into prominent IR absorption bands, particularly in the amide I (1620 cm^{-1}) and amide II (1511 cm^{-1}) regions of the peptide backbone. Other absorptions associated with fibroin can be found at 1695, 1442, 996, and 975 cm^{-1} . These bands are reliable indicators of fibroin purity or of the presence of coatings above it, as they are associated with fibroin's antiparallel β -sheet structure and its specific amino acid sequences, such as glycine–glycine and glycine–alanine motifs. Sericin, on the other hand, exhibits a predominantly amorphous and disordered structure, resulting in broader IR absorption bands. Moreover, a slight shift toward higher wavenumbers is present; hence, the amide I and II bands for sericin can typically be found around $\sim 1640\text{ cm}^{-1}$ and $\sim 1540\text{ cm}^{-1}$, respectively. A further distinguishing feature is the absorption band near 1400 cm^{-1} , attributed to its higher content of amino acids bearing polar side chains, such as carboxylic acid and hydroxyl groups [6,49]. A summary of the bands associated with sericin and fibroin is given in Table 3.

The baseline-corrected and normalized FT-IR spectra of raw and degummed fibers are presented in pairs (Figs. 5–9), where the curves relevant to each degumming treatment are overlapped to the one from raw fibers. The curves relevant to degummed fibers, with the exception of DEG_C, present common features: (i) the characteristic peaks of fibroin, mentioned above, appear to be more pronounced; (ii)

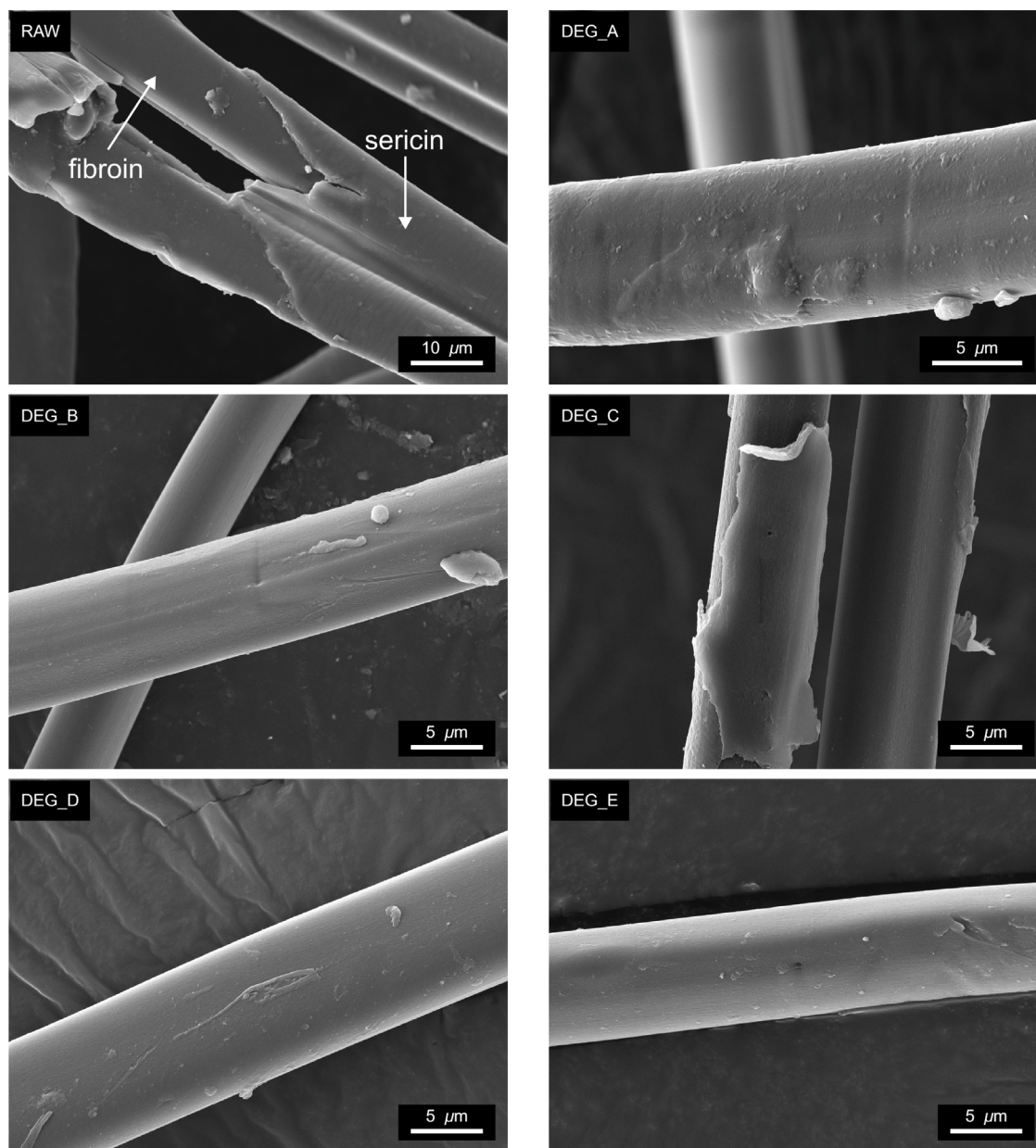


Fig. 4. SEM micrographs of RAW and DEG_A-E samples.

the peak located at 3274 cm^{-1} becomes narrower; (iii) a shoulder at 1262 cm^{-1} appears on the peak located at 1233 cm^{-1} , which appears higher; (iv) the bands associated with sericin (1640 and 1400 cm^{-1}) appear less intense. The presence of these features suggests a complete, or a high degree of sericin removal and they find confirmation in literature [16,40,52,53,55,56]. As previously stated, DEG_C shows a larger retaining of the features of the raw silk. This result, in agreement with the smaller mass loss registered, confirms that the degumming treatment in pure water is not completely effective in removing the sericin coating.

These observations, and in particular the narrowing of the 3274 cm^{-1} band, reflect the nature of the amide A region ($3400\text{--}3270\text{ cm}^{-1}$), where the backbone N–H stretching overlaps the O–H stretching of hydroxyl side chains and bound water [51]. Sericin is much richer than fibroin in polar, hydroxyl-bearing residues (serine alone accounting for about 30% of sericin against 15% of fibroin) so the sericin-coated raw fiber is more hydroxyl-rich and binds more water [51]. The raw fiber therefore shows a broad envelope extending

toward 3400 cm^{-1} , dominated by O–H stretching contributions from bound water and sericin-associated components. Degumming reduces this contribution, resulting in a narrower band mainly centered around 3274 cm^{-1} amide A N–H stretching vibration of the crystalline fibroin structure. Consequently, the higher wavenumber side of the envelope can be considered a sensitive indicator of residual sericin. The latter was evaluated through intensity ratios of sericin-associated bands and adjacent fibroin bands, following the approach reported in [50] and extending it to the amide A envelope. On the baseline-corrected spectra, four intensity ratios were evaluated: the amide A envelope (I_{3400}/I_{3274}), the amide I random coil to β -sheet ratio (I_{1650}/I_{1620}), I_{1400}/I_{1442} (carboxylate + serine over alanine) and I_{1066}/I_{1165} (serine C–O over tyrosine). Each ratio was normalized to the corresponding value of the raw fiber, resulting in a relative residual sericin index from 0 to 1 (RAW = 1). Each ratio is reported separately as mean $\pm$ SD over the three spectra. The four ratios are not averaged into a single index: they probe different moieties and have very different dynamic ranges, so an unweighted mean would be dominated by the most sensitive ones.

Table 3
FT-IR bands specific to sericin and fibroin selected for qualitative analysis.

This work [cm ⁻¹]	Literature values [cm ⁻¹]	Assignment	Molecular origin
<i>Sericin marker bands (weaken/disappear on degumming)</i>			
3400	3400 [50,51]	O–H/N–H stretch	Bound water + ser/tyr hydroxyl side chains, weakly H-bonded N–H overlap
1650	1640 [6]; 1642 [49]; 1650 [50–52]	Amide I, random coil/disordered	Disordered backbone
1540	1547 [49]; 1530 [50]; 1545 [51]	Amide II, disordered	Disordered backbone
1400	1400 [6,50,51,53]; 1395 [49]; 1398 [52]; 1402–1404 [54]	$\nu_s(\text{COO}^-) + \delta(\text{CH}_2, \text{OH})/\nu(\text{C–O})$	Asp/Glu carboxylate + serine side chains
1120	1120–1253 (region) [54]	$\nu(\text{C–O})$ side-chain envelope (whole region drops on degumming)	Serine + Asp/Glu carboxyl C–O
1066	1052–1058, 1068–1075 [49]; 1070 [50]; 1071 [51]; 1053 [52]	C–O/C–OH stretch	Serine + carboxyl side chains
<i>Fibroin marker bands (sharpen/intensify on degumming)</i>			
3274	3270 [50]; 3274 [51,53]	Amide A, N–H stretch	Intermolecular hydrogen-bonded N–H of the fibroin backbone
1695	1699 [49]; 1698 [51]; 1697 [52]; 1693 [40]	Amide I, antiparallel β -sheet/ β -turn (high- ν component)	Crystalline backbone
1620	1624 [2]; 1620 [49]; 1625 [50]; 1628–1616 [51]; 1618 [53]; 1616 [40]	Amide I, β -sheet	Crystalline backbone
1511	1515 [6]; 1508, 1516 [49]; 1515 [51]; 1516–1518 [54]	Amide II, β -sheet (overlaps Tyr ring mode)	Backbone + tyrosine
1442	1440 [6]; 1443 [49]; 1445 [50]; 1445 [54]	CH_3 antisymmetric deformation	Alanine in $(\text{A})_n/(\text{AG})_n$ β -sheet
1262	1260 [6,51]; 1262 [53]	Amide III, antiparallel β -sheet (shoulder on the 1229–1233 band)	Crystalline backbone
1233	1234 [6]; 1237–1245 [49]; 1230 [51]; 1233 [53]	Amide III, main backbone band	Backbone (β -sheet + random-coil sub-bands); Tyr/Asp-Glu overlap
1165	1165 [49,50]; 1170–1179 [54]	Tyr $\delta(\text{CH})$ side chain (alt.: $\nu(\text{N–C}\alpha)$ backbone)	Tyrosine
996/975	998, 975 [49]; 1000, 975 [50,51,53]	(Gly-Gly)/(Gly-Ala) backbone rocking, β -sheet	Glycine-glycine/glycine-alanine

All treatment decreased the four ratios relative to RAW fiber, confirming sericin removal (Fig. 10). The four alkaline treatments cluster tightly and low in every ratio, consistent with almost complete sericin removal. The hot water treatment DEG_C retains markedly higher values in all four: I_{3400}/I_{3274} 0.73 ± 0.05 against 0.27 – 0.35 for the alkaline fibers, I_{1650}/I_{1620} 0.92 ± 0.03 against 0.75 – 0.80 , I_{1400}/I_{1442} 0.67 ± 0.03 against 0.51 – 0.55 and I_{1066}/I_{1165} 0.46 ± 0.03 against 0.28 – 0.30 . Since the difference is reproduced independently by four ratios probing different moieties and, in each case, exceeds the replicate scatter by a wide margin, the result does not depend on any single spectral feature or on how the ratios are combined. This strengthens the hypothesis of DEG_C being a partial sericin removal case.

To move beyond a qualitative band assignment, the amide I envelope was deconvolved into its secondary structure components (Section 2.3 and Appendix B). The resulting relative secondary structure composition is reported in Table 4 and showed in Fig. 10. Two robust trends emerge. First, all degumming treatments increased the relative β -sheet fraction probed by FT-IR, from 40.2% for the raw fiber to values between 44 and 51% for the degummed fibers. Second, among the degummed fibers DEG_C is the least β -sheet-rich ($44.0 \pm 0.5\%$) and is therefore the closest to raw one, whereas the fully degummed alkaline fibers clustered at the higher end of the distribution (DEG_B 50.8 ± 2.9 , DEG_D 50.0 ± 0.7 , DEG_A 48.9 ± 1.1 and DEG_E $48.9 \pm 1.1\%$), which are indistinguishable from one another within the replicate scatter.

3.2. Interfacial properties

The interfacial shear strength of the silk/epoxy systems was measured by the microdebonding test. Load–displacement curves from

microdebonding tests on untreated and treated fibers are reported in Fig. 11(a). The nominal IFSS values (computed with the cylindrical perimeter πd) are summarized in Table 5, together with the values corrected for the measured fiber perimeter and the associated Monte Carlo 95% confidence interval. The comparison is shown graphically in Fig. 11(b). The perimeter correction is rather small: for the degummed fibers the correction is negligible, while for the raw fibers IFSS is raised by about 11%.

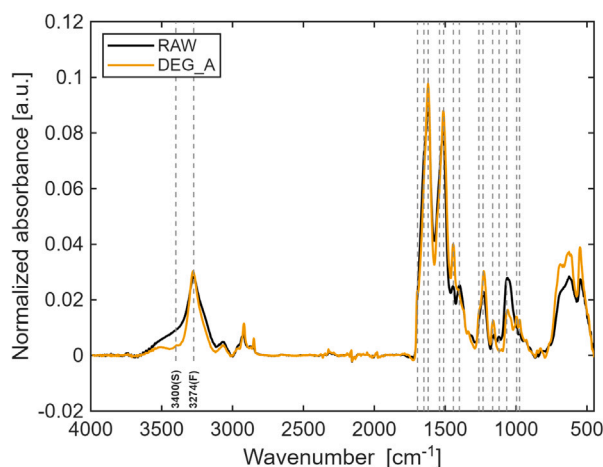
The load–displacement curves in Fig. 11(a) differ from the ideal microdebonding response reported for stiffer synthetic fibers. Each curve is considered from the moment the test engages the fiber/matrix interface. The initial portion, associated solely with the deformation of the silk fiber within the setup is not included. From the onset of interfacial loading, the curves exhibit a quasi-linear increase associated with elastic deformation of the interface. Then, a region of progressively decreasing slope that reflects the initiation of interfacial damage and stress redistribution along the embedded length. The load then reaches a maximum value associated with the onset, and sudden propagation, of interfacial debonding. No stable post-peak frictional plateau is observed; the load drops abruptly instead. This is attributed to the high compliance and extensibility of the silk fiber, which stores a large amount of elastic energy during loading. Once debonding occurs, the energy is released abruptly and the fiber recoils, causing the droplet to lose stable contact with the shearing plates and the test to end prematurely without controlled frictional sliding. The peak load was nonetheless used to compute the IFSS.

Overall, the measured IFSS values (approximately ~ 6.6 – 9.0 MPa) are consistent with those reported in the literature for natural fiber/epoxy

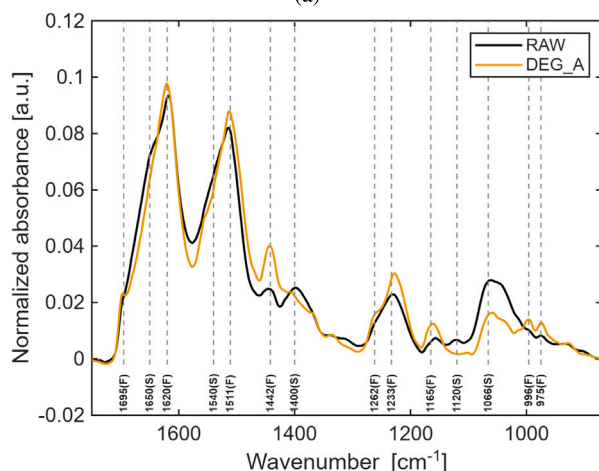
Table 4

Amide I secondary structure composition of raw and degummed fibers obtained by constrained Gaussian deconvolution. Σ β -sheet is the sum of the native, parallel and antiparallel β -sheet components. Mean $\pm$ SD over three spectra per condition. $R^2 \geq 0.996$ for every individual deconvolution (see Appendix B).

Label	Random coil [%]	α -helix [%]	Turns [%]	β -native [%]	β -parallel [%]	β -antiparallel [%]	Σ β -sheet [%]
RAW	27.2 $\pm$ 0.7	11.3 $\pm$ 0.5	21.2 $\pm$ 1.2	16.5 $\pm$ 0.4	22.3 $\pm$ 1.6	1.4 $\pm$ 0.2	40.2 $\pm$ 1.2
DEG_A	25.0 $\pm$ 1.4	9.0 $\pm$ 0.3	17.1 $\pm$ 0.9	21.1 $\pm$ 0.3	25.0 $\pm$ 1.7	2.7 $\pm$ 0.3	48.9 $\pm$ 1.1
DEG_B	23.4 $\pm$ 0.8	8.8 $\pm$ 0.8	16.9 $\pm$ 1.9	20.8 $\pm$ 0.3	27.4 $\pm$ 3.2	2.7 $\pm$ 0.6	50.8 $\pm$ 2.9
DEG_C	27.0 $\pm$ 1.0	9.4 $\pm$ 1.0	19.6 $\pm$ 0.5	19.1 $\pm$ 0.6	22.8 $\pm$ 1.0	2.2 $\pm$ 0.1	44.0 $\pm$ 0.5
DEG_D	24.8 $\pm$ 0.2	8.8 $\pm$ 0.3	16.4 $\pm$ 0.4	20.9 $\pm$ 0.2	26.0 $\pm$ 1.1	3.2 $\pm$ 0.1	50.0 $\pm$ 0.7
DEG_E	24.2 $\pm$ 1.1	9.5 $\pm$ 0.2	17.4 $\pm$ 0.6	20.9 $\pm$ 0.1	24.8 $\pm$ 1.4	3.2 $\pm$ 0.2	48.9 $\pm$ 1.1



(a)

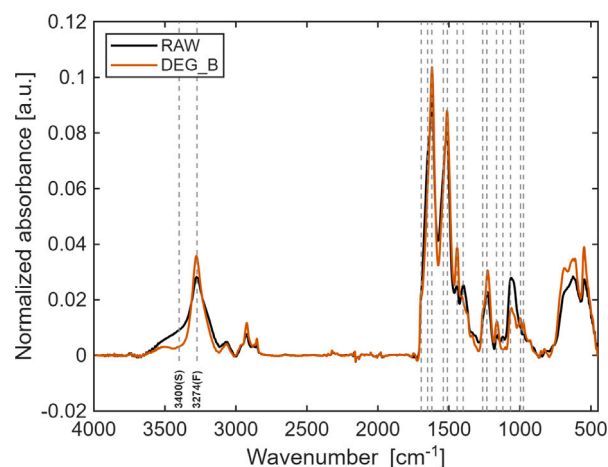


(b)

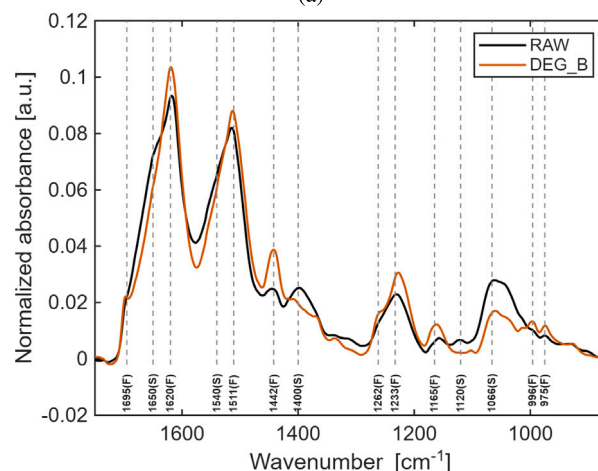
Fig. 5. FT-IR spectra of raw and degummed (NaOH 0.5 wt%, 40 °C, 50 min) silk fibers: (a) full measured range and (b) detail of the region between 1750 and 870 cm^{-1} . Dashed lines indicate the bands of interest (Table 3) relevant to sericin (S) and fibroin (F). The spectra shown are the replicates closest to the family mean spectrum, of the three acquired.

systems (e.g. approximately 11.64 MPa for jute/epoxy, 6.41 MPa for kenaf/epoxy, and 11.83 MPa for flax/epoxy systems) [57] and lie below the 15 MPa value reported by Craven et al. for a silk/epoxy interface [29].

Ranking the corrected values, DEG_C gives the highest interfacial shear strength (9.1 MPa), above the RAW fiber (7.7 MPa, roughly 17% higher) and the other degummed conditions (6.6 to 8.0 MPa). The complete IFSS ranking is DEG_C > DEG_B > RAW > DEG_A > DEG_D > DEG_E. This advantage is consistent and substantial in magnitude. However, given the small number of successful microdebonding tests



(a)



(b)

Fig. 6. FT-IR spectra of raw and degummed (NaOH 0.5 wt%, 60 °C, 25 min) silk fibers: (a) full measured range and (b) detail of the region between 1750 and 870 cm^{-1} . Dashed lines indicate the bands of interest (Table 3) relevant to sericin (S) and fibroin (F). The spectra shown are the replicates closest to the family mean spectrum, of the three acquired.

per condition ($n = 4-6$) and the scatter intrinsic to natural fibers, neither the overall comparison across conditions nor the individual pairwise differences reach formal statistical significance, even where the effect size is large. The DEG_C ranking is read as a consistent trend rather than a set of individually significant contrasts. The superior interfacial performance of the water-treated DEG_C fiber, achieved despite its residual sericin is interpreted in Section 3.4.

The adhesion properties of the RAW fibers can be directly correlated with the SEM observations (Fig. 12(a,b)). Although the meniscus appears regularly shaped before debonding, the epoxy droplet shows

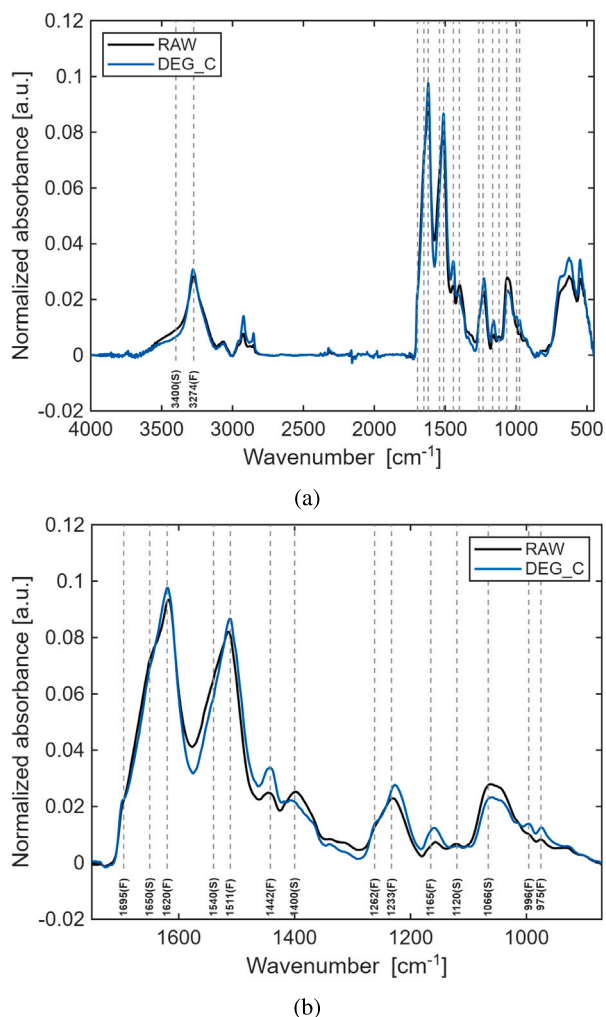


Fig. 7. FT-IR spectra of raw and degummed (H_2O , 90 °C, 50 min) silk fibers: (a) full measured range and (b) detail of the region between 1750 and 870 cm^{-1} . Dashed lines indicate the bands of interest (Table 3) relevant to sericin (S) and fibroin (F). The spectra shown are the replicates closest to the family mean spectrum, of the three acquired.

limited wetting of the fiber surface. This is evident from the non-uniform epoxy distribution immediately beyond the meniscus region that suggests low interfacial compatibility between matrix and fiber. After debonding the droplet appears to be detached and to have slid along the fiber. Slight irregularities are still visible on the fiber surface similar to those observed in Fig. 12(a), indicating that the poor wettability persists also in the debonding region, i.e., the fiber surface that was in direct contact with the droplet prior to debonding. Conversely, SEM micrograph in Fig. 12(c) shows that DEG_C exhibits improved interfacial interaction: the epoxy homogeneously wets the fiber surface, forming a smoother meniscus prior to debonding. After debonding (Fig. 12(d)), the fiber surface appears rougher and partially covered by epoxy residues, suggesting that failure involves some degree of cohesive damage within the matrix, consistent with the highest IFSS value registered. Although these micrographs were acquired at moderate magnification, they consistently show more uniform epoxy coverage before debonding and greater epoxy retention afterward for DEG_C than for the RAW fiber, which provides qualitative support for the higher IFSS measured on DEG_C. It is worth noting that excessively large epoxy droplets were not suitable for this type of fiber, which undergoes significant deformation prior to debonding. Samples such as the one shown in Fig. 12(e) were therefore excluded, as the

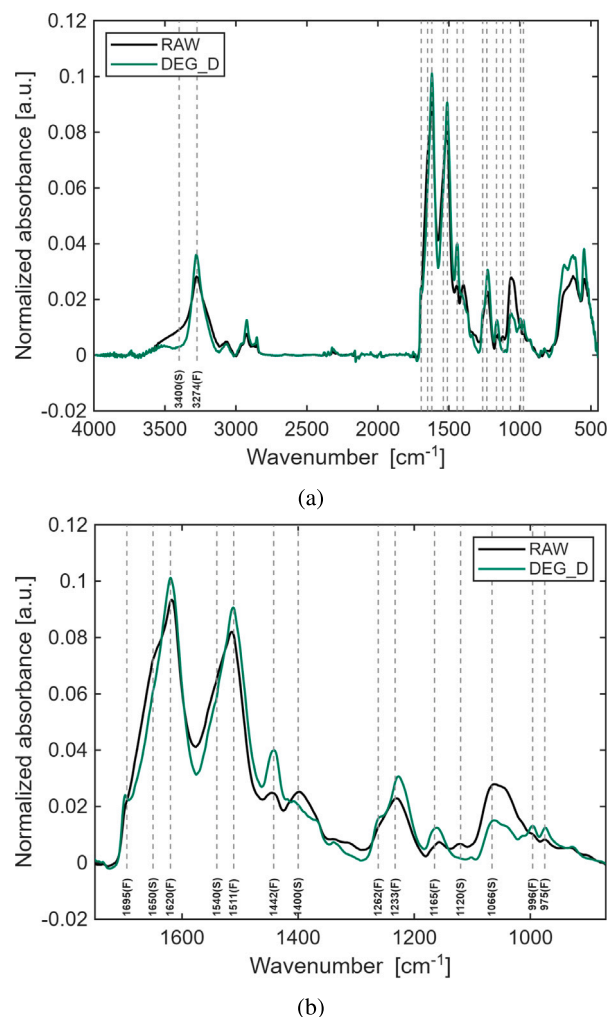


Fig. 8. FT-IR spectra of raw and degummed (Na_2CO_3 , 0.5 wt%, 90 °C, 50 min) silk fibers: (a) full measured range and (b) detail of the region between 1750 and 870 cm^{-1} . Dashed lines indicate the bands of interest (Table 3) relevant to sericin (S) and fibroin (F). The spectra shown are the replicates closest to the family mean spectrum, of the three acquired.

large droplet generates a non-uniform stress distribution during testing, leading to localized fiber damage and premature failure, rather than a representative interfacial debonding process.

3.3. Single-fiber tensile tests

To determine whether the integrity of the fibroin core was retained after the degumming treatments, some of the prepared fibers were selected for further investigation of their tensile mechanical properties. In particular, samples RAW, DEG_B, DEG_C and DEG_E were chosen. Being interested only in a comparative analysis, the effect of the gage length was not investigated. Replacing the assigned cross-sectional area (the value used by the testing machine, computed from the nominal fiber diameter assuming a circular section) with the measured area (obtained from the real, non-circular cross-sections quantified in [18]) raises the tensile stress of every fiber, mostly for the RAW fiber whose measured area ($190 \mu\text{m}^2$) is far smaller than the assigned $314 \mu\text{m}^2$ (a factor of ~ 1.65). For the degummed fibers, the factor is less severe (~ 1.16). Crucially, strain is geometry-independent, so the corrected curves are simply the original curves with a rescaled stress axis: the relative ductility of the fibers is unchanged by the correction. Representative corrected stress-strain curves are shown in Fig. 13. From

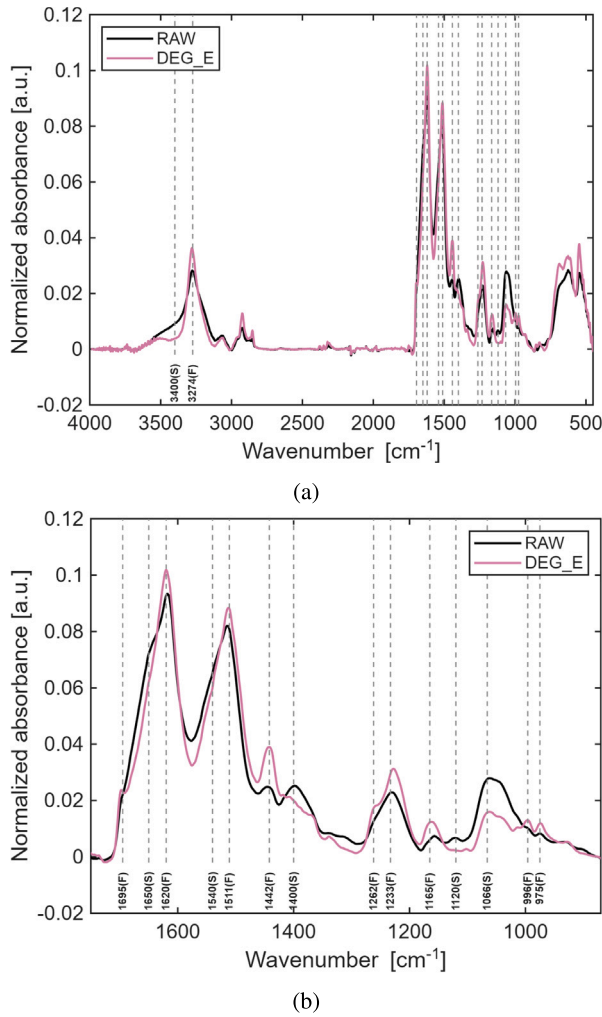


Fig. 9. FT-IR spectra of raw and degummed (NaOH 0.5 wt%, 60 °C, 50 min) silk fibers: (a) full measured range and (b) detail of the region between 1750 and 870 cm^{-1} . Dashed lines indicate the bands of interest (Table 3) relevant to sericin (S) and fibroin (F). The spectra shown are the replicates closest to the family mean spectrum, of the three acquired.

Table 5
Values of IFSS from microbonding tests.

Label	Nominal IFSS [MPa]	Corrected IFSS + 95% CI [MPa]	Variation [%]
RAW	7.0 ± 1.1	7.7 [6.9, 8.4]	+10.7
DEG_A	7.3 ± 3.5	7.3 [6.7, 8.0]	+0.5
DEG_B	8.0 ± 1.8	8.0 [7.3, 8.7]	+0.5
DEG_C	9.0 ± 1.8	9.1 [8.2, 9.9]	+0.7
DEG_D	7.0 ± 1.7	7.0 [6.4, 7.7]	+0.6
DEG_E	6.6 ± 2.2	6.6 [6.0, 7.3]	+0.6

the corrected data DEG_C is the strongest fiber by a clear margin: its Weibull scale parameter $\sigma_0 = 454$ MPa exceeds those of DEG_B (285 MPa), RAW (251 MPa) and DEG_E (248 MPa). The same ranking holds for the corrected arithmetic means (Table 6) and is visible directly in the Weibull plot of Fig. 14. The shape parameters, after small-sample bias correction, are in the range 3.1–4.1, thus indicating a broad strength scatter for all conditions, as expected for silk. Unlike the interfacial comparison, the differences in single-fiber tensile strength are statistically significant (one-way ANOVA $F = 16.2$, $p < 0.001$), with DEG_C stronger than every other tested condition and this result is unchanged by the cross-section correction.

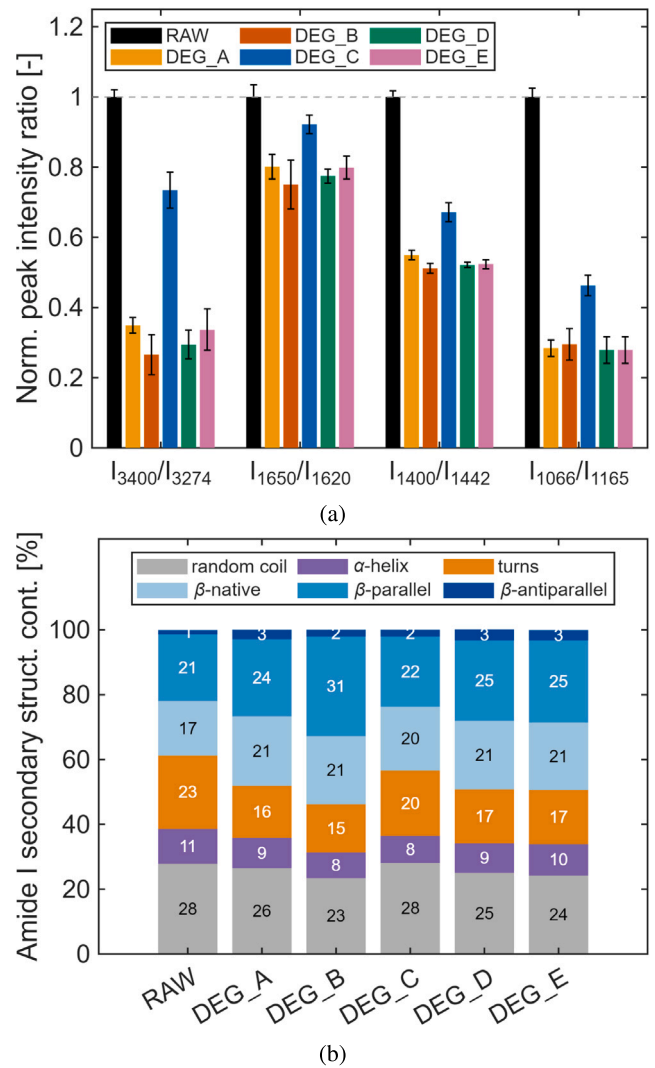


Fig. 10. (a) Relative residual sericin index of raw and degummed silk fibers; each of the four intensity ratios is normalized to the raw value (RAW = 1) and reported as mean $\pm$ SD over three spectra. (b) Amide I secondary-structure composition of raw and degummed silk fibers from the constrained deconvolution; bars show the mean of three spectra, with the corresponding standard deviations given in Table 4.

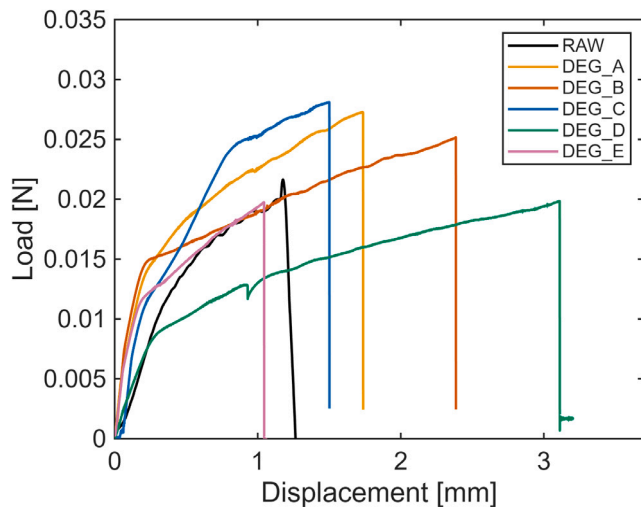
The results suggest that the treatments remove sericin and superficial defects, partially damaging the fiber core in some cases. Non-uniform surface impurities (e.g., residual sericin) and structural heterogeneities can act as stress concentrators that increase the scatter in single-fiber strength [19,58,59]. DEG_C, in line with its superior IFSS, likely benefits from a balanced treatment that preserves structural integrity while enhancing surface energy. In contrast, DEG_E exhibits a reduction in tensile strength and a lower shape parameter, indicating greater variability and a higher defect sensitivity. This behavior is consistent with the microbonding results and supports the hypothesis that the harsher NaOH treatment (longer time at elevated temperature) induces damage to the fiber structure, which compromises its mechanical integrity.

The corrected RAW strength obtained here ($\sigma_0 = 251$ MPa, $m = 4.0$) is in good agreement with the corrected raw value of the previous, geometry-resolved study [18] ($\sigma_0 = 229$ MPa, $m = 5.1$ or 4.6 considering the bias-correction for $n = 22$), which validates both the correction procedure and the present test set-up for the RAW fiber. The degummed reference condition (DEG_E here = “degummed” in [18])

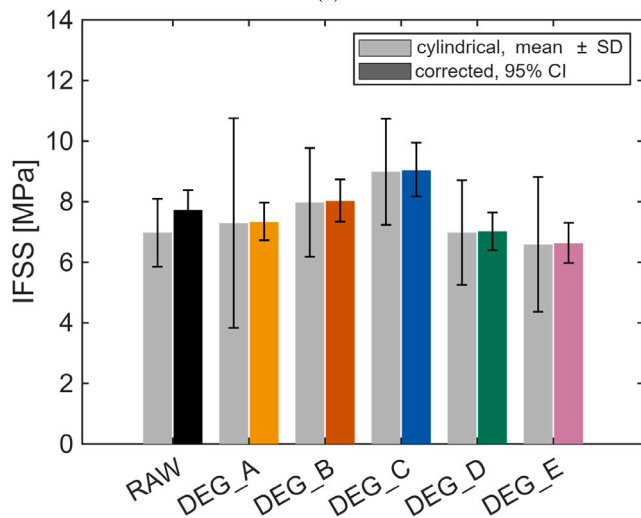
Table 6

Results from single-fiber tensile tests: experimental tensile strength calculated both with the assumption of circular cross section from the measured diameter and with the corrected cross section; Weibull's σ_0 and m , the latter corrected for the small-sample bias.

Label	Circular mean strength [MPa]	Corrected mean strength + 95% CI [MPa]	Weibull σ_0 [MPa]	Weibull m (bias-corrected)
RAW	141 ± 29	245 [204, 300]	251	4.01
DEG_B	228 ± 45	276 [233, 323]	285	4.05
DEG_C	360 ± 86	435 [371, 504]	454	3.89
DEG_E	194 ± 55	235 [195, 280]	248	3.06



(a)



(b)

Fig. 11. (a) Representative load-displacement curves from microbonding tests on untreated and treated fibers. (b) Nominal versus perimeter-corrected IFSS for the six conditions (error bars: Monte Carlo 95% CI).

shows a lower corrected strength than in the previous study (248 versus 347 MPa). We regard this discrepancy as a non-critical limitation rather than a correction error: it is consistent with the small sample size of the present screening ($n = 5-7$ against $n = 22$ previously) and with differences in apparatus and other test parameters between the two campaigns.

The modulus ranking broadly mirrors the strength ranking (Table 6): DEG_C is the stiffest fiber (Weibull $E_0 = 11.3$ GPa, corrected mean 10.9 GPa), ahead of DEG_B (9.9 GPa), DEG_E (9.2 GPa) and the RAW (8.8 GPa); see Table 7 and Fig. 14. Consistent with strength, the

cross-section correction raises every modulus value, in particular for RAW. It should be noted that these are apparent moduli: because no system-compliance calibration (e.g. ASTM C1557, based on multiple gage lengths) was performed in this comparative campaign, machine compliance and grip slack inflate the measured strain and depress the apparent modulus. The proposed correction rescales the stress axis only and cannot remove this contribution. The apparent moduli are nonetheless directly comparable among the conditions of this study, since all were measured under identical conditions at a single 30 mm gage length.

This compliance caveat also frames the comparison with [18], where the corrected raw modulus ($E_0 = 8.0$ GPa) again agrees well with the present raw value (8.8 GPa), while the degummed modulus is lower here than previously (9.2 versus 12.6 GPa). The strength and modulus gaps for the degummed reference are of different origins and should be kept distinct: the strength gap is attributable to sampling and apparatus/operator differences, while the modulus gap is dominated by the absence of a compliance calibration in the present screening. Notably, the corrected DEG_C modulus mean value (10.9 GPa) approaches the fibroin intrinsic modulus that can be evaluated under the hypothesis that the fibroin is the only load-bearing element: adopting the fibroin equivalent cross-section proposed in [18], the fibroin modulus mean value measured from RAW fibers is 11.9 GPa. Therefore, the fiber treated with water only recovers a near-pristine bulk stiffness, while, according to the surface FT-IR analysis, it is the least crystalline of the degummed fibers. This aspect is discussed in detail in Section 3.4.

3.4. Structure-property relationships

Three mechanical properties resulting from this study (interfacial shear strength, tensile strength and apparent modulus) agree that DEG_C ranks first: the hot-water-degummed fiber (DEG_C) is simultaneously the most adhesive, the strongest and the stiffest of the conditions examined. This conclusion comes from three measurements obtained by two different tests (microbonding and tensile), therefore bringing strong, mutually corroborating evidence that the effect is real and not an artifact of any one measurement. In particular, the higher Weibull's σ_0 of DEG_C corroborates its higher IFSS, linking bulk fiber quality to interfacial performance. DEG_C is the mildest treatment and the one that removes the least sericin (~21% weight loss, highest residual-sericin FT-IR marker). Its superiority is therefore best explained as a consequence of gentle, partial sericin removal that preserves the fibroin surface, rather than as a consequence of the residual sericin itself: sericin is an amorphous, mechanically weak protein and would not, by itself, strengthen either the fiber or the fiber/matrix interface. On the other hand, the aggressive alkaline solutions fully strip the sericin but expose the fibroin to a harsher chemical environment, which has been reported to degrade the fibroin molecular structure and mechanical properties [36,48] and may therefore introduce surface flaws that lower the fiber strength and create a more defective interface. In short, DEG_C performs best despite its residual sericin, because the route damages less the load-bearing fibroin and the fiber surface that interacts with the matrix.

At first sight the FT-IR result appears to contradict the mechanics: DEG_C is the least β -sheet-rich of the degummed fibers yet the stiffest

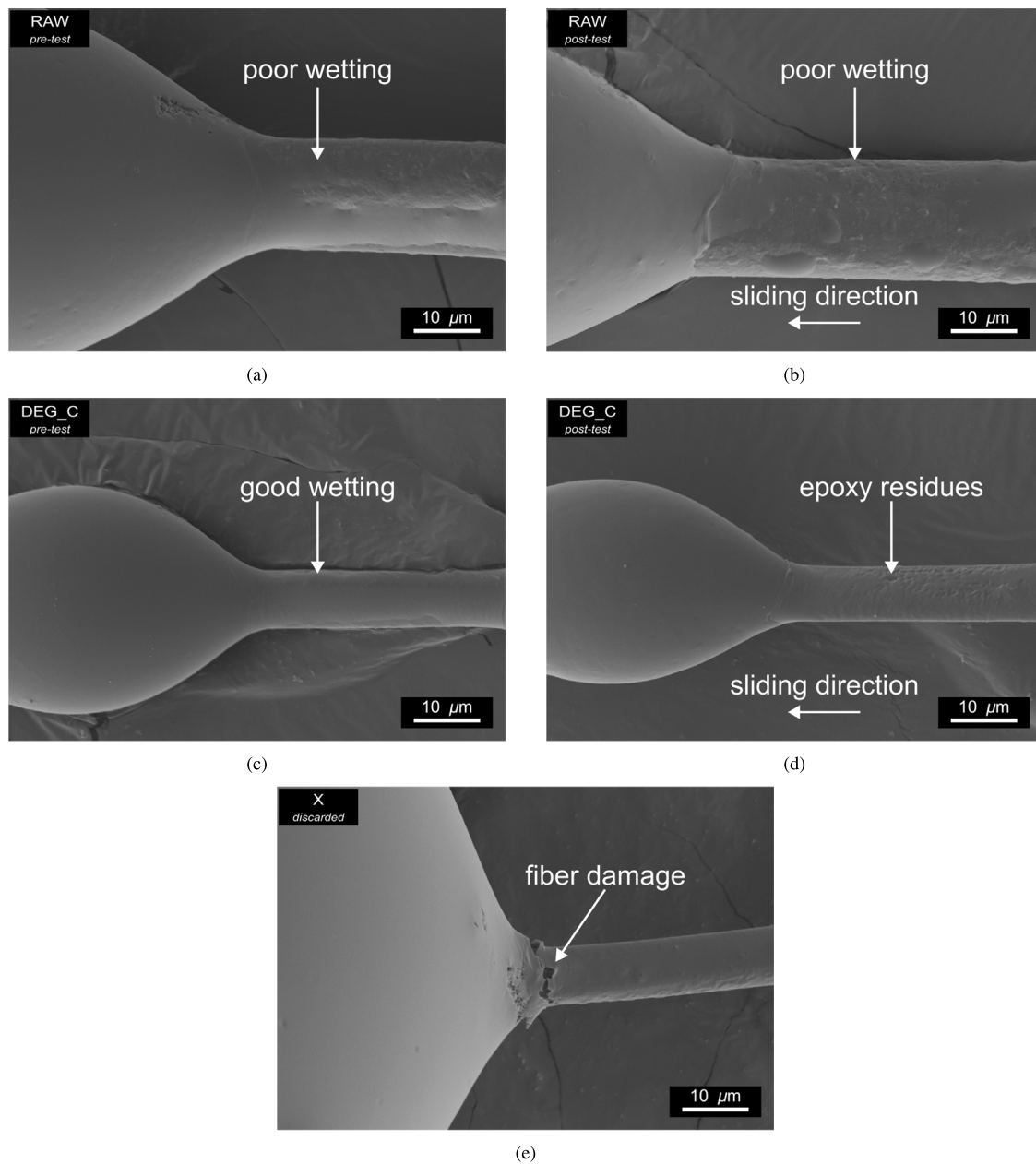


Fig. 12. SEM micrographs from microdebonding tests: (a) RAW before microdebonding, (b) RAW after microdebonding, (c) DEG_C before microdebonding, (d) DEG_C after microdebonding, and (e) example of a discarded tested sample.

Table 7

Results from single-fiber tensile tests: apparent modulus.

Label	Circular mean apparent modulus [GPa]	Corrected apparent modulus + 95% CI [GPa]	Weibull E_0 [GPa]	Weibull m (bias-corrected)
RAW	4.9 ± 1.1	8.5 [7.1, 10.5]	8.8	4.31
DEG_B	8.0 ± 1.6	9.6 [8.1, 11.3]	9.9	4.67
DEG_C	9.0 ± 2.0	10.9 [9.3, 12.6]	11.3	4.19
DEG_E	7.4 ± 1.6	8.9 [7.4, 10.6]	9.2	3.88

and strongest, whereas a higher β -sheet crystallinity is generally associated with higher fibroin modulus [2,4]. However, the contradiction is only apparent and is resolved by distinguishing surface and bulk properties. The FT-IR β -sheet fraction is a surface quantity, weighted toward the outer 1–2 μm , which for the raw and DEG_C fibers is dominated by the residual sericin coating; the high β -sheet values of

the fully degummed fibers largely reflect the removal of that disordered outer layer rather than a true bulk increase of crystallinity. The tensile modulus, by contrast, is a bulk property of the fibroin core. This is consistent with the layered structure of the sericin coat: the outer sericin that dominates the RAW and DEG_C surface is disordered and random-coil, whereas the inner layer retained after gentle degumming,

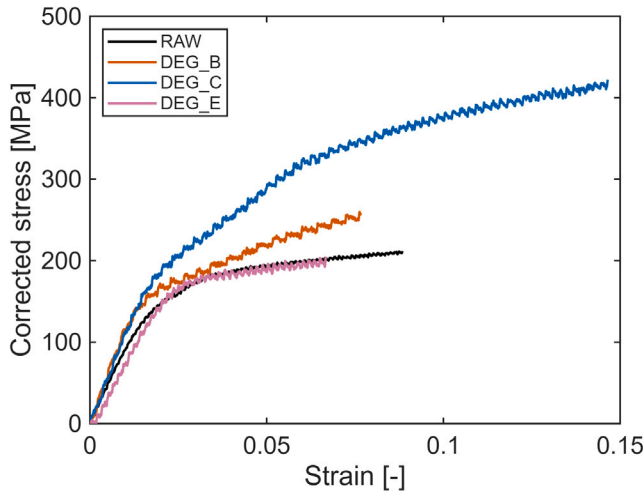


Fig. 13. Single-fiber tensile tests: representative stress-strain curves of the four tested samples.

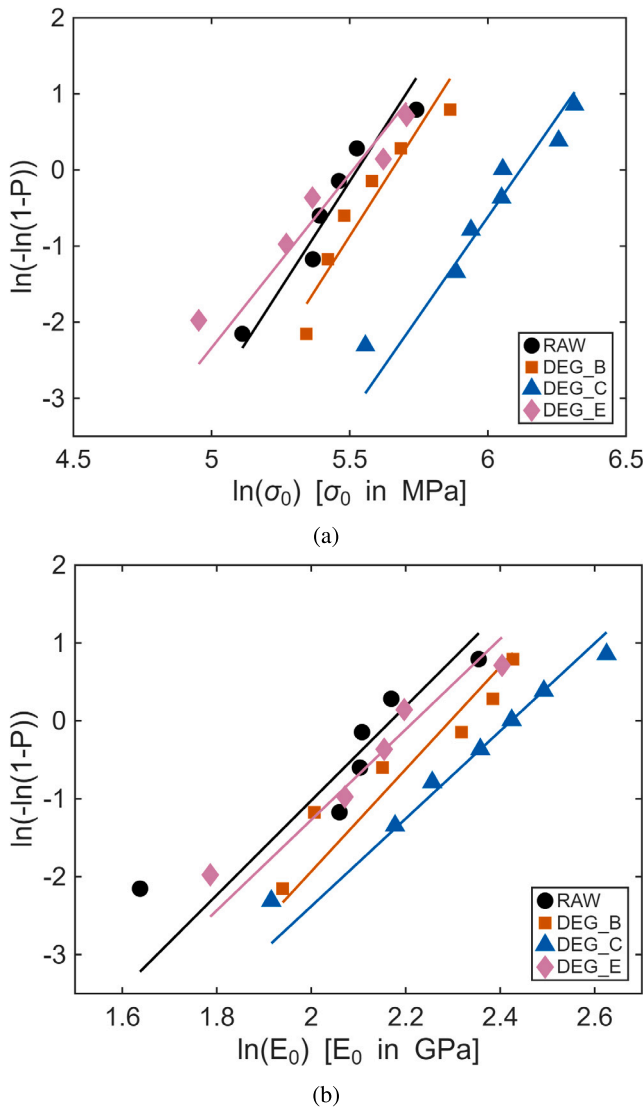


Fig. 14. Single-fiber tensile tests: Weibull distributions of (a) strength and (b) modulus.

together with the fibroin it exposes, is richer in β -sheet [45,60]. The intermediate surface β -sheet of DEG_C, above the raw fiber but below the fully degummed ones, therefore corresponds to partial removal of the outer layer rather than to a change in the fibroin core. From an interfacial-engineering standpoint, these results identify gentle hot water degumming as a favorable compromise for silk-waste composites: it cleans the fiber enough to expose a compatible fibroin surface, yet it is gentle enough to preserve the bulk fibroin and to deliver the best interfacial adhesion of the routes tested.

The properties found are low in terms of values, if compared with those of high-performance plant fibers such as flax [21]. Silk waste presents, however, a different combination of properties: large strain to failure, high toughness and energy-absorption capability, low density, and an origin as a textile by-product that would otherwise be downgraded or discarded [10,21–23]. Its features suit silk to energy-absorbing and impact applications, possibly in hybrid architectures with stiffer fibers. Assessing this potential at the component scale is the natural continuation of the present work: it requires moving from the single fiber to the laminate, where fiber length, fiber volume fraction, the interface and the matrix act together. Further, different matrices could be screened; exploiting the ductility and energy absorption of silk in particular calls for a sufficiently ductile and tough matrix and for an adequate fiber length [24,25], while a matrix that is at the same time more compatible with the fibroin surface and more sustainable would be a further improvement. A direct, depth-resolved characterization of the fiber surface and mechanical tests at the composite scale would be necessary to confirm these expectations.

4. Conclusions

The silk/epoxy interface across different degumming conditions of floss *Bombyx mori* silk was investigated, combining microdebonding tests, single-fiber tensile tests, FT-IR and SEM. A central methodological feature was the replacement of the conventional circular cross-section by the measured cross-sectional area and perimeter of the same fibers from a previous work. The main findings are:

- The area correction is large (raising the raw tensile stress by $\sim 1.65\times$ and the degummed stress by $\sim 1.16\times$), whereas the perimeter correction is small ($< 1\%$ for the degummed fibers and $\sim +11\%$ for the raw IFSS).
- The mild hot water treatment DEG_C, which removes only half of the sericin, gives the highest interfacial shear strength (9.0 MPa), the highest corrected tensile strength (Weibull $\sigma_0 = 454$ MPa) and the highest apparent modulus (Weibull $E_0 = 11.3$ GPa), i.e. a single ranking is confirmed by three independent measurements.
- FT-IR shows that every degumming treatment raises the surface β -sheet fraction (from 40% to 44%–51%) and that DEG_C is the least crystalline of the degummed fibers at the surface. However, these results from surface analysis are decoupled from the bulk mechanical properties.
- DEG_C performs best despite, not because of, its residual sericin: the gentle route preserves the fibroin surface and bulk structure, while aggressive alkaline degumming fully strips the sericin but exposes the fibroin to a harsher environment.

Overall, mild hydrothermal degumming emerges as a favorable compromise for valorizing silk waste in epoxy composites, combining adequate sericin removal with preservation of the fibroin properties and the best interfacial adhesion among the routes tested. Verifying this at the composite scale, where fiber length, matrix ductility and the interface act simultaneously, is the natural continuation of this interfacial study.

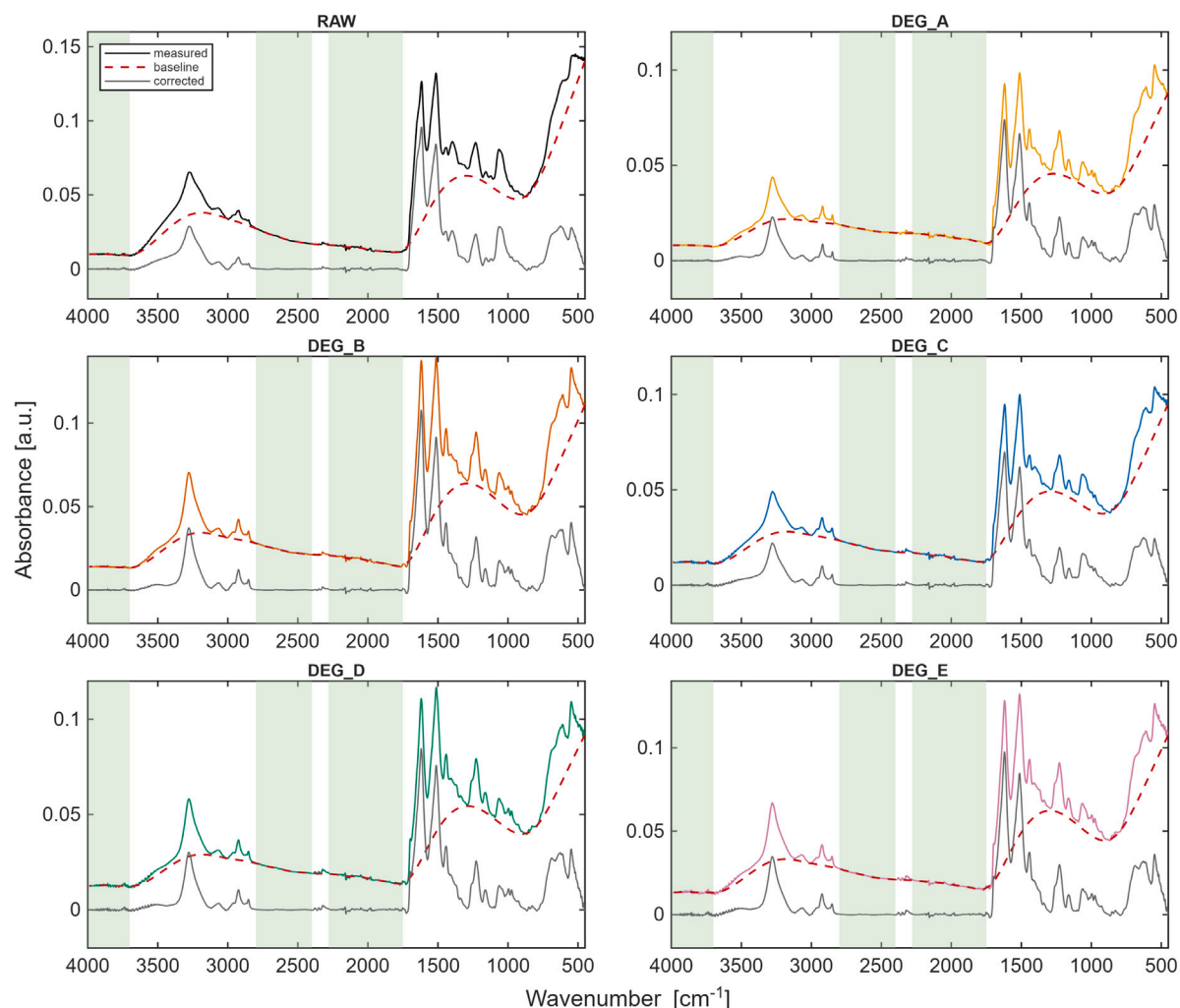


Fig. 15. FT-IR baseline removal. Green regions indicate the anchor points for the fitting procedure.

CRedit authorship contribution statement

Tommaso Pini: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Software, Visualization, Writing – original draft. **Laura Simonini:** Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Writing – original draft. **Giulia Fredi:** Conceptualization, Validation, Writing – review & editing. **Maria Paola Bracciale:** Investigation, Validation, Writing – review & editing. **Alessandro Pegoretti:** Resources, Supervision, Validation, Writing – review & editing. **Marco Valente:** Resources, Supervision, Validation, Writing – review & editing.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Spectra pre-processing

Figs. 15 and 16 illustrate the baseline correction applied to the FT-IR spectra. For each spectrum, a smooth baseline is estimated by asymmetric least-squares (AsLS) [61,62], in which a roughness penalty ($\lambda = 1 \times 10^6$) is balanced against an asymmetric fit that weights points lying below the curve far more heavily than points above it (asymmetry $p = 1 \times 10^{-3}$); the baseline therefore follows the lower envelope of the signal and passes beneath the absorption bands. To keep the baseline from being drawn upward under the broad silk bands, its weights were pinned to unity within three anchor windows that contain no silk-specific absorption: 3700–4000 cm^{-1} (above the O–H/N–H stretching envelope), 2400–2800 cm^{-1} (the gap between the C–H stretching and amide regions), and 1750–2280 cm^{-1} (the silent region just above amide I). These windows are highlighted in green in Fig. 15. In each panel the estimated baseline (red dashed line) is subtracted from the measured spectrum (colored line) to yield the corrected spectrum (gray line), reported separately for the raw fiber and the five degummed conditions. Fig. 15 shows the full measured range (450–4000 cm^{-1}), where the anchor windows are visible. Fig. 16 shows the same correction over the fingerprint region (870–1750 cm^{-1}), where the resolved sericin and fibroin bands used in the analysis are most clearly seen.

Because ATR absorbances are affected by experimental factors such as the effective fiber/crystal contact area and the wavenumber-dependent penetration depth, which prevent direct comparison of

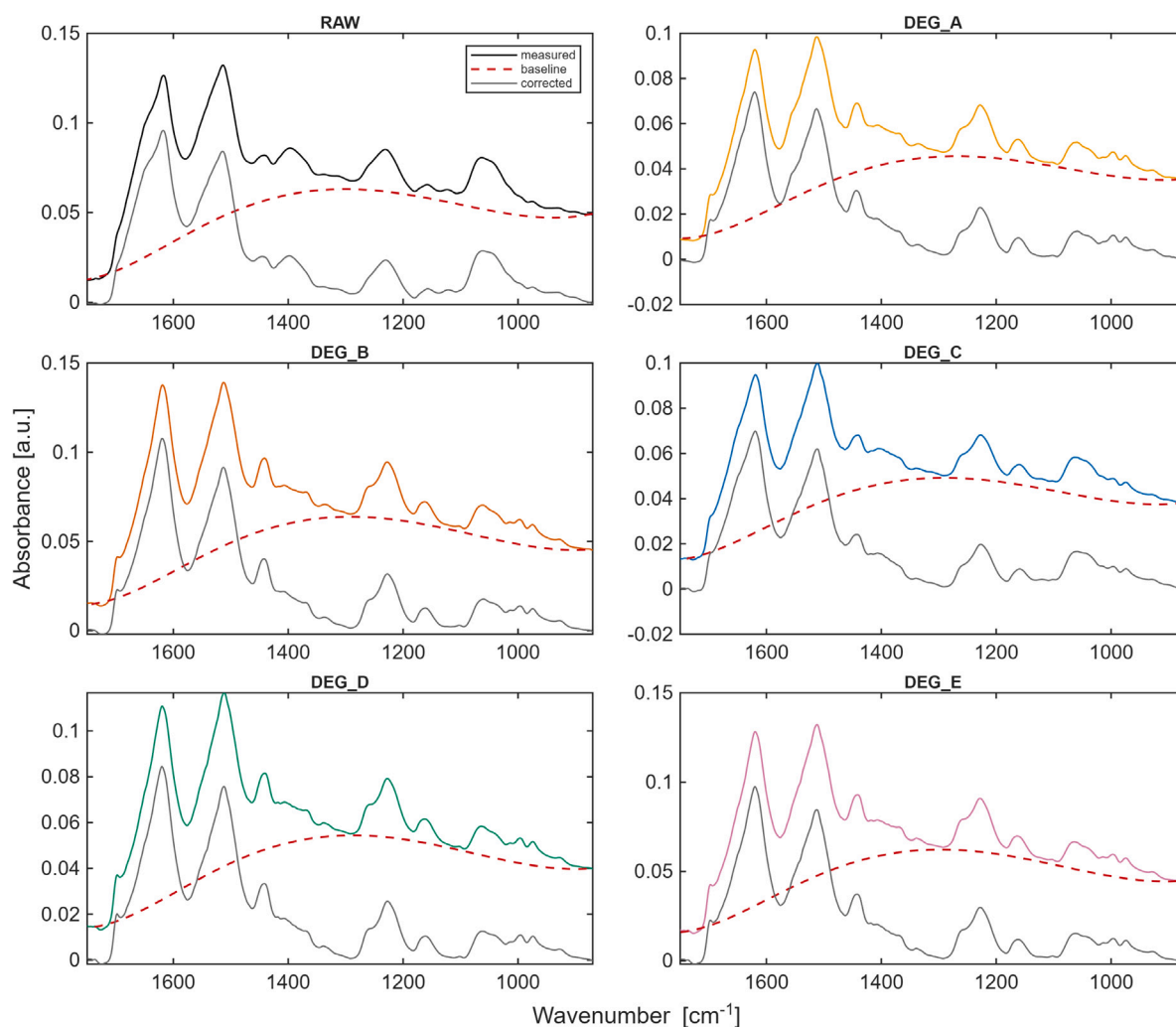


Fig. 16. FT-IR baseline removal, focus on the region between 1750 and 870 cm^{-1} .

absolute intensities among spectra, each baseline-corrected spectrum was scaled by vector normalization over the 870–1750 cm^{-1} region [39, 63].

Appendix B. Amide I deconvolution

Fig. 17 shows amide I deconvolutions for the different formulations. In each case the baseline-subtracted band (points) is reproduced by the constrained sum of Gaussians (solid line) with $R^2 \geq 0.996$ and a $RMSE$ below 0.002 absorbance units. The residual (lower panel) is small and free of sharp unmodeled features; it is largest on the low-wavenumber shoulder (1580–1595 cm^{-1}), where the Gaussian tail decays faster than the measured wing. Because amide I curve fitting is mathematically non-unique, the decomposition was further validated against a model-free area partition of the band (Section 2.3), which agrees with it for every sample.

Although isolated vibrational transitions are Lorentzian, the protein amide I envelope is dominated by inhomogeneous broadening, which produces approximately Gaussian component shapes [38–40]. To confirm that the reported composition does not depend on this

assumption, the deconvolution was repeated with Lorentzian and with mixed Gauss–Lorentz (pseudo-Voigt) components, keeping the same peak positions and width constraints. The comparison was carried out on all three spectra of each sample and the values quoted are means over the replicates. The fitted Lorentzian fraction of the pseudo-Voigt was small for every sample ($\eta = 0.11$ – 0.15), i.e. the bands are predominantly Gaussian, and the total β -sheet content changed by at most 1.3 percentage points relative to the Gaussian fit, leaving the ranking of the treatments unchanged (Fig. 18). The pure-Lorentzian model gave a markedly poorer fit ($RMSE$ about twice the Gaussian value). A pseudo-Voigt shape produced a marginally better fit and was mildly favored by information criteria, but as it did not alter any secondary structure conclusion, the standard Gaussian model was retained for consistency with the established amide I literature.

Data availability

Data will be made available on request.

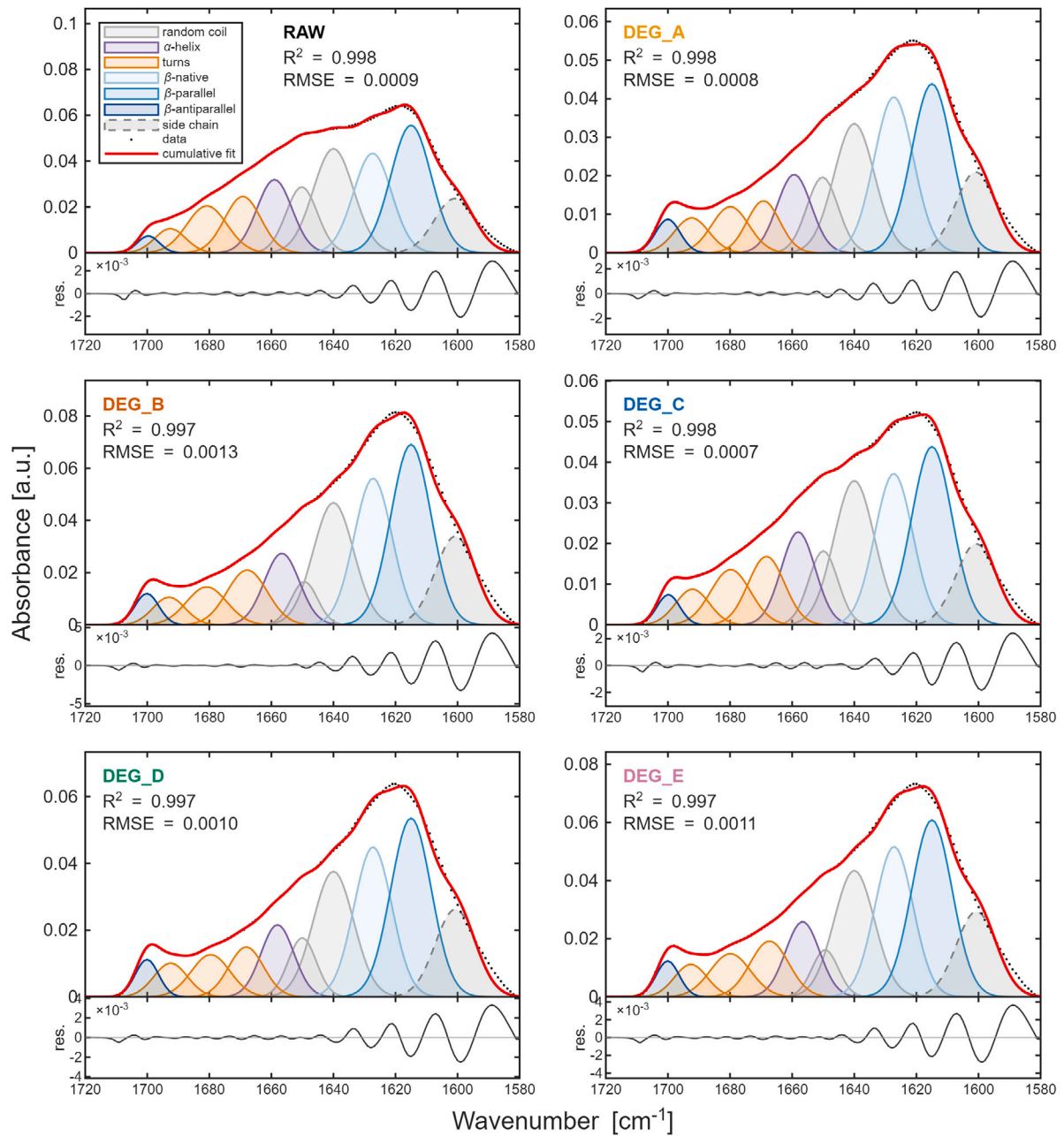


Fig. 17. Amide I band deconvolution of the raw and degummed fibers. In each panel the baseline-subtracted amide I band (1580 to 1720 cm^{-1} , black dots) is fitted with a constrained sum of Gaussians (red line); the shaded curves are the individual components. The strip below each spectrum is the fit residual.

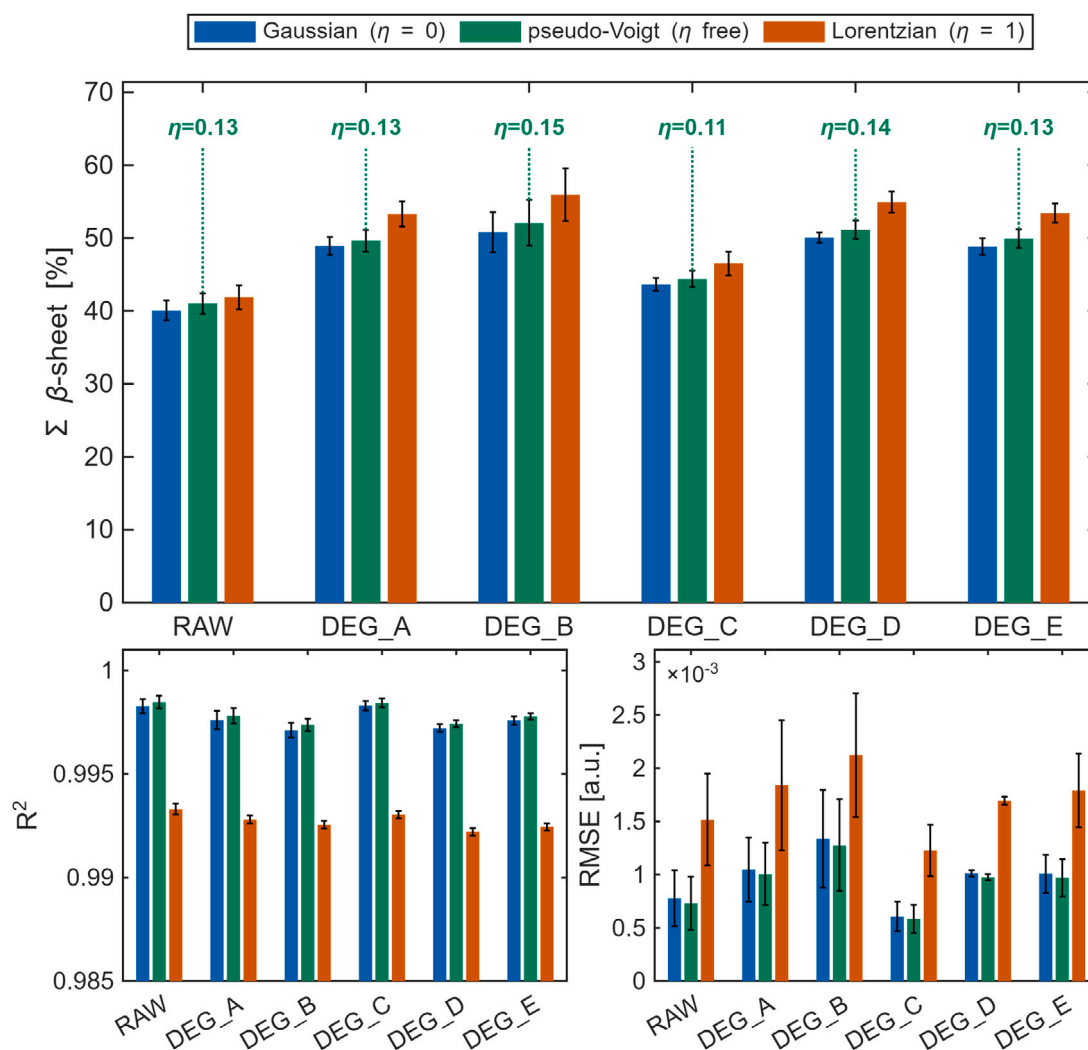


Fig. 18. Effect of the assumed component line shape on the amide I analysis. Top: total β -sheet fraction of each case obtained with Gaussian, pseudo-Voigt and Lorentzian components, keeping the same peak positions and width constraints; the value above each group is the fitted Lorentzian fraction η of the pseudo-Voigt ($\eta = 0$ a pure Gaussian, $\eta = 1$ a pure Lorentzian). Bottom: coefficient of determination (R^2) and root-mean-square residual (RMSE) of the three fits.

References

- [1] Chand S, Chand S, Raula B. Usage of silkworm materials in various ground of science and research. *J Nat Fibers* 2023;20:2139328. <http://dx.doi.org/10.1080/15440478.2022.2139328>.
- [2] Guo C, Zhang J, Jordan JS, Wang X, Henning RW, Yarger JL. Structural comparison of various silkworm silks: An insight into the structure–property relationship. *Biomacromolecules* 2018;19:906–17. <http://dx.doi.org/10.1021/acs.biomac.7b01687>.
- [3] Ude AU, Eshkoo RA, Zulkifili R, Ariffin AK, Dzuraidah AW, Azhari CH. *Bombyx Mori* silk fibre and its composite: A review of contemporary developments. *Mater Des* 2014;57:298–305. <http://dx.doi.org/10.1016/j.matdes.2013.12.052>.
- [4] Guo C, Zhang J, Wang X, Nguyen AT, Liu XY, Kaplan DL. Comparative study of strain-dependent structural changes of silkworm silks: Insight into the structural origin of strain-stiffening. *Small* 2017;13:1702266. <http://dx.doi.org/10.1002/smll.201702266>.
- [5] Bhowmik P, Kant R, Singh H. Effect of degumming duration on the behavior of waste filature silk-reinforced wheat gluten composite for sustainable applications. *ACS Omega* 2023;8:6268–78. <http://dx.doi.org/10.1021/acsomega.2c05963>.
- [6] Gaviria A, Jaramillo-Quiceno N, Motta A, Restrepo-Osorio A. Silk wastes and autoclaved degumming as an alternative for a sustainable silk process. *Sci Rep* 2023;13:15296. <http://dx.doi.org/10.1038/s41598-023-41762-6>.
- [7] Khan MMR, Tsukada M, Gotoh Y, Morikawa H, Freddi G, Shiozaki H. Physical properties and dyeability of silk fibers degummed with citric acid. *Bioresour Technol* 2010;101:8439–45. <http://dx.doi.org/10.1016/j.biortech.2010.05.100>.
- [8] Rastogi S, Kandasubramanian B. Processing trends of silk fibers: Silk degumming, regeneration and physical functionalization. *J Text Inst* 2020;111:1794–810. <http://dx.doi.org/10.1080/00405000.2020.1727269>.
- [9] Sashina ES, Yakovleva OI. The current state and prospects of recycling silk industry waste into nonwoven materials. *Fibers* 2023;11:56. <http://dx.doi.org/10.3390/fib11060056>.
- [10] Pini T, Sambucci M, Valente M. Potential use of silk waste in sustainable thermoplastic composite material applications: A review. *Fibers* 2025;13:6. <http://dx.doi.org/10.3390/fib13010006>.
- [11] Joshi S, Drzal L, Mohanty A, Arora S. Are natural fiber composites environmentally superior to glass fiber reinforced composites? *Compos Part A: Appl Sci Manuf* 2004;35:371–6. <http://dx.doi.org/10.1016/j.compositesa.2003.09.016>.
- [12] Alves DI, Barreiros M, Fanguero R, Ferreira DP. Valorization of textile waste: Non-woven structures and composites. *Front Environ Sci* 2024;12:1365162. <http://dx.doi.org/10.3389/fenvs.2024.1365162>.
- [13] Lu L, Fan W, Ge S, Liew RK, Shi Y, Dou H, Wang S, Lam SS. Progress in recycling and valorization of waste silk. *Sci Total Environ* 2022;830:154812. <http://dx.doi.org/10.1016/j.scitotenv.2022.154812>.
- [14] Jiang P, Liu H, Wang C, Wu L, Huang J, Guo C. Tensile behavior and morphology of differently degummed silkworm (*Bombyx mori*) cocoon silk fibres. *Mater Lett* 2006;60:919–25. <http://dx.doi.org/10.1016/j.matlet.2005.10.056>.
- [15] Prakash N, Jaya, Shanmugarajan D, Wang X, Kandasubramanian B. Enhancement of mechano-structural characteristics of silk fibroin using microwave assisted degumming. *Sustain Chem Pharm* 2022;30:100902. <http://dx.doi.org/10.1016/j.scp.2022.100902>.
- [16] Eliaz D, Kellersztein I, Miali ME, Benyamin D, Brookstein O, Daraio C, Wagner HD, Raviv U, Shimanovich U. Fine structural analysis of degummed fibroin fibers reveals its superior mechanical capabilities. *ChemSusChem* 2025;18:e202401148. <http://dx.doi.org/10.1002/cssc.202401148>.
- [17] Greco G, Schmuck B, Jalali SK, Pugno NM, Rising A. Influence of experimental methods on the mechanical properties of silk fibers: A systematic literature

- review and future road map. *Biophys Rev* 2023;4:031301. <http://dx.doi.org/10.1063/5.0155552>.
- [18] Pini T, Pasquini C, Bracciale MP, Sarasini F, Tirillò J, Valente M. An enhanced cross-sectional analysis to evaluate tensile properties of silk waste fibers. *Compos Part A: Appl Sci Manuf* 2025;199:109210. <http://dx.doi.org/10.1016/j.compositesa.2025.109210>.
 - [19] Zhao H-P, Feng X-Q, Shi H-J. Variability in mechanical properties of *Bombyx mori* silk. *Mater Sci Eng: C* 2007;27:675–83. <http://dx.doi.org/10.1016/j.msec.2006.06.031>.
 - [20] Chen S, Liu M, Huang H, Cheng L, Zhao H-P. Mechanical properties of *Bombyx mori* silkworm silk fibre and its corresponding silk fibroin filament: A comparative study. *Mater Des* 2019;181:108077. <http://dx.doi.org/10.1016/j.matdes.2019.108077>.
 - [21] Shah DU, Porter D, Vollrath F. Can silk become an effective reinforcing fibre? A property comparison with flax and glass reinforced composites. *Compos Sci Technol* 2014;101:173–83. <http://dx.doi.org/10.1016/j.compscitech.2014.07.015>.
 - [22] Ataollahi S, Taher S, Eshkoor R, Ariffin A, Azhari C. Energy absorption and failure response of silk/epoxy composite square tubes: Experimental. *Compos Part B: Eng* 2012;43:542–8. <http://dx.doi.org/10.1016/j.compositesb.2011.08.019>.
 - [23] Vuure AW Van, Vanderbeke J, Mosleh Y, Verpoest I, El-Asmar N. Ductile woven silk fibre thermoplastic composites with quasi-isotropic strength. *Compos Part A: Appl Sci Manuf* 2021;147:106442. <http://dx.doi.org/10.1016/j.compositesa.2021.106442>.
 - [24] Vuure AW Van, Mosleh Y, Vanderbeke J, Verpoest I. Highly impact-resistant silk fiber thermoplastic composites. *Adv Eng Mater* 2023;25:2300080. <http://dx.doi.org/10.1002/adem.202300080>.
 - [25] Yang K, Ritchie RO, Gu Y, Wu SJ, Guan J. High volume-fraction silk fabric reinforcements can improve the key mechanical properties of epoxy resin composites. *Mater Des* 2016;108:470–8. <http://dx.doi.org/10.1016/j.matdes.2016.06.128>.
 - [26] Yang K, Wu S, Guan J, Shao Z, Ritchie RO. Enhancing the mechanical toughness of epoxy-resin composites using natural silk reinforcements. *Sci Rep* 2017;7:11939. <http://dx.doi.org/10.1038/s41598-017-11919-1>.
 - [27] Zainudin Z, Yusoff NIS Mohd, Wahit MU. Characteristics of continuous unidirectional silk fibre reinforced epoxy composites. *J Adv Res Mater Sci* 2020;69:16–28. <http://dx.doi.org/10.37934/arms.69.1.1628>.
 - [28] Hamidi YK, Yalcinkaya MA, Guloglu GE, Pishvar M, Amirhosravi M, Altan MC. Silk as a natural reinforcement: Processing and properties of silk/epoxy composite laminates. *Materials* 2018;11:2135. <http://dx.doi.org/10.3390/ma11112135>.
 - [29] Craven J, Cripps R, Viney C. Evaluating the silk/epoxy interface by means of the Microbond Test. *Compos Part A: Appl Sci Manuf* 2000;31:653–60. [http://dx.doi.org/10.1016/S1359-835X\(00\)00042-7](http://dx.doi.org/10.1016/S1359-835X(00)00042-7).
 - [30] p. Ho M, Wang H, t. Lau K, h. Lee J, Hui D. Interfacial bonding and degumming effects on silk fibre/polymer biocomposites. *Compos Part B: Eng* 2012;43:2801–12. <http://dx.doi.org/10.1016/j.compositesb.2012.04.042>.
 - [31] Ranakoti L, Gangil B, Rajesh PK, Singh T, Sharma S, Li C, Ilyas R, Mahmoud O. Effect of surface treatment and fiber loading on the physical, mechanical, sliding wear, and morphological characteristics of tasar silk fiber waste-epoxy composites for multifaceted biomedical and engineering applications: Fabrication and characterizations. *J Mater Res Technol* 2022;19:2863–76. <http://dx.doi.org/10.1016/j.jmrt.2022.06.024>.
 - [32] Zulkifli R, Azhari CH. Effect of silane concentrations on Mode I interlaminar fracture of woven silk/epoxy composites. *Adv Mater Res* 2010;150:1171–5. <http://dx.doi.org/10.4028/www.scientific.net/amr.150-151.1171>.
 - [33] Shi R, Ye D, Ma K, Tian W, Zhao Y, Guo H, Shao Z, Guan J, Ritchie RO. Understanding the interfacial adhesion between natural silk and polycaprolactone for fabrication of continuous silk biocomposites. *ACS Appl Mater Interfaces* 2022;14:46932–44. <http://dx.doi.org/10.1021/acsami.2c11045>.
 - [34] Vyas SK, Shukla SR. Comparative study of degumming of silk varieties by different techniques. *J Text Inst* 2016;107:191–9. <http://dx.doi.org/10.1080/00405000.2015.1020670>.
 - [35] Mahmood H, Simonini L, Dorigato A, Pegoretti A. Graphene deposition on glass fibers by triboelectrification. *Appl Sci* 2021;11:3123. <http://dx.doi.org/10.3390/app11073123>.
 - [36] Bucciarelli A, Greco G, Corridori I, Pugno NM, Motta A. A Design of Experiment Rational Optimization of the Degumming Process and Its Impact on the Silk Fibroin Properties. *ACS Biomater Sci Eng* 2021;7:1374–93. <http://dx.doi.org/10.1021/acsbomaterials.0c01657>.
 - [37] Bucciarelli A, Chiera S, Quaranta A, Yadavalli VK, Motta A, Maniglio D. A thermal-reflow-based low-temperature, high-pressure sintering of lyophilized silk fibroin for the fast fabrication of biosubstrates. *Adv Funct Mater* 2019;29:1901134. <http://dx.doi.org/10.1002/adfm.201901134>.
 - [38] Byler DM, Susi H. Examination of the secondary structure of proteins by deconvolved FTIR spectra. *Biopolymers* 1986;25:469–87. <http://dx.doi.org/10.1002/bip.360250307>.
 - [39] Kong J, Yu S. Fourier transform infrared spectroscopic analysis of protein secondary structures. *Acta Biochim et Biophys Sin* 2007;39:549–59. <http://dx.doi.org/10.1111/j.1745-7270.2007.00320.x>.
 - [40] Belton DJ, Plowright R, Kaplan DL, Perry CC. A robust spectroscopic method for the determination of protein conformational composition- application to the annealing of silk. *Acta Biomater* 2018;73:355–64. <http://dx.doi.org/10.1016/j.actbio.2018.03.058>.
 - [41] Simonini L, Kakkonen M, Dsouza R, Kanerva M, Mahmood H, Dorigato A, Pegoretti A. Tailoring the interfacial properties of glass fiber-epoxy microcomposites through the development of a self-healing poly(ϵ -caprolactone) coating. *Compos Sci Technol* 2025;261:110991. <http://dx.doi.org/10.1016/j.compscitech.2024.110991>.
 - [42] Simonini L, Mahmood H, Dorigato A, Pegoretti A. Evaluation of self-healing capability of a polycaprolactone interphase in epoxy/glass composites. *Compos Part A: Appl Sci Manuf* 2023;169:107539. <http://dx.doi.org/10.1016/j.compositesa.2023.107539>.
 - [43] Simonini L, Canale R, Mahmood H, Dorigato A, Pegoretti A. Multifunctional epoxy/carbon composites with a fully repairable interface. *Polym Compos* 2024;45:2558–68. <http://dx.doi.org/10.1002/pc.27939>.
 - [44] Simonini L, Rigotti D, Pidapa JK Reddy, Pegoretti A. Poly(butylene adipate-co-terephthalate) as a new healing agent for epoxy/basalt composites. *Compos Part A: Appl Sci Manuf* 2025;196:109010. <http://dx.doi.org/10.1016/j.compositesa.2025.109010>.
 - [45] Wang YJ, Zhang YQ. Three-layered sericins around the silk fibroin fiber from *Bombyx mori* cocoon and their amino acid composition. *Adv Mater Res* 2011;175–176:158–63. <http://dx.doi.org/10.4028/www.scientific.net/AMR.175-176.158>.
 - [46] Thoman DR, Bain LJ, Antle CE. Inferences on the parameters of the Weibull distribution. *Technometrics* 1969;11:445–60. <http://dx.doi.org/10.2307/1267019>.
 - [47] Yuan Y, Nasri M, Manayi A, Zhang J, Wu C, Jeon T-J, Kang L. Sericin coats of silk fibres, a degumming waste or future material? *Mater Today Bio* 2024;29:101306. <http://dx.doi.org/10.1016/j.mtbio.2024.101306>.
 - [48] Carissini G, Lozano-Pérez AA, Montalbán MG, Aznar-Cervantes SD, Cenis JL, Villora G. Revealing the influence of the degumming process in the properties of silk fibroin nanoparticles. *Polymers* 2019;11:2045. <http://dx.doi.org/10.3390/polym11122045>.
 - [49] Boulet-Audet M, Vollrath F, Holland C. Identification and classification of silks using infrared spectroscopy. *J Exp Biol* 2015;218:3138–49. <http://dx.doi.org/10.1242/jeb.128306>.
 - [50] Zhang X, Wyeth P. Using FTIR spectroscopy to detect sericin on historic silk. *Sci China Chem* 2010;53:626–31. <http://dx.doi.org/10.1007/s11426-010-0050-y>.
 - [51] Geminiani L, Campione FP, Canevali C, Corti C, Giussani B, Gorla G, Luraschi M, Recchia S, Rampazzi L. Historical silk: A novel method to evaluate degumming with non-invasive infrared spectroscopy and spectral deconvolution. *Materials* 2023;16:1819. <http://dx.doi.org/10.3390/ma16051819>.
 - [52] Teramoto H, Miyazawa M. Analysis of structural properties and formation of sericin fiber by infrared spectroscopy. *J Insect Biotechnol Sericology* 2003;72:157–62. <http://dx.doi.org/10.11416/jibs.72.157>.
 - [53] Baranowska-Korczyn A, Hudecki A, Kamińska I, Cieślak M. Silk powder from cocoons and woven fabric as a potential bio-modifier. *Materials* 2021;14:6919. <http://dx.doi.org/10.3390/ma14226919>.
 - [54] Barth A. The infrared absorption of amino acid side chains. *Prog Biophys Mol Biol* 2000;74:141–73. [http://dx.doi.org/10.1016/S0079-6107\(00\)00021-3](http://dx.doi.org/10.1016/S0079-6107(00)00021-3).
 - [55] Lee H, Park SJ, e. Lee M, m. Choi K, Choi HY, Hasegawa Y, Kim M, Kim KB. Fabrication of nanofibers using fibroin regenerated by recycling waste silk selva. *Polym Bull* 2020;77:3853–62. <http://dx.doi.org/10.1007/s00289-020-03113-7>.
 - [56] Kim HJ, Chung DE, Um IC. Effect of processing conditions on the homogeneity of partially degummed silk evaluated by FTIR spectroscopy. *Int J Ind Entomol* 2013;26:54–60. <http://dx.doi.org/10.7852/IJIE.2013.26.1.054>.
 - [57] Kandemir A, Pozegic TR, Hamerton I, Eichhorn SJ, Longana ML. Characterisation of natural fibres for sustainable discontinuous fibre composite materials. *Materials* 2020;13:2129. <http://dx.doi.org/10.3390/ma13092129>.
 - [58] Poza P, Pérez-Rigueiro J, Elices M, Llorca J. Fractographic analysis of silkworm and spider silk. *Eng Fract Mech* 2002;69:1035–48. [http://dx.doi.org/10.1016/S0013-7944\(01\)00120-5](http://dx.doi.org/10.1016/S0013-7944(01)00120-5).
 - [59] Pérez-Rigueiro J, Elices M, Llorca J, Viney C. Effect of degumming on the tensile properties of silkworm (*Bombyx mori*) silk fiber. *J Appl Polym Sci* 2002;84:1431–7. <http://dx.doi.org/10.1002/app.10366>.
 - [60] Zhao Z-L, Zhang Y-Q. Greener degumming production of layered sericin peptides from a silkworm cocoon and their physicochemical characteristics and bioactivities in vitro. *J Clean Prod* 2020;261:121080. <http://dx.doi.org/10.1016/j.jclepro.2020.121080>.
 - [61] Eilers PHC. A perfect smoother. *Anal Chem* 2003;75:3631–6. <http://dx.doi.org/10.1021/ac034173t>.
 - [62] Baek S-J, Park A, Ahn Y-J, Choo J. Baseline correction using asymmetrically reweighted penalized least squares smoothing. *Analyst* 2014;140:250–7. <http://dx.doi.org/10.1039/C4AN01061B>.
 - [63] Rinnan Å, van den Berg F, Engelsen SB. Review of the most common pre-processing techniques for near-infrared spectra. *TRAC Trends Anal Chem* 2009;28:1201–22. <http://dx.doi.org/10.1016/j.trac.2009.07.007>.