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Full Paper

Laccase-assisted approach to graft multifunctional materials of interest: keratin-EC based novel composites and their characterisation

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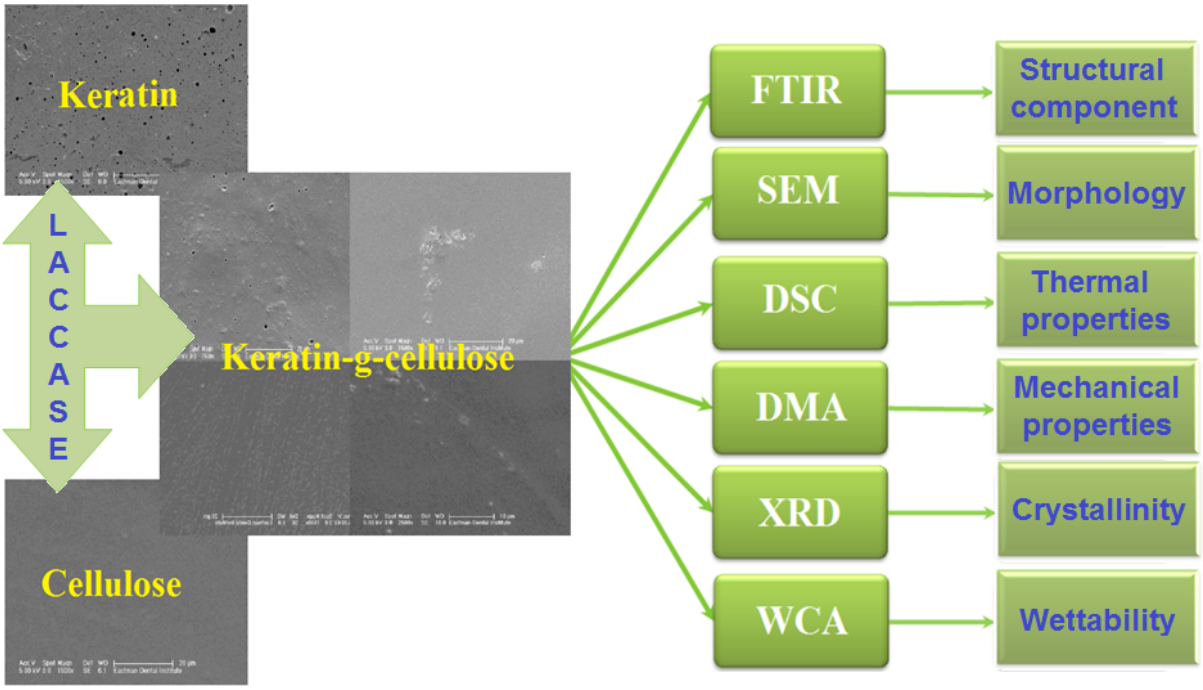
This study focuses on the evaluation of raw keratin as a potential material to develop composites with novel characteristics. Herein, we report a mild and eco-friendly fabrication of in-house extracted feather keratin-based novel enzyme assisted composites consisting of ethyl cellulose (EC) as a backbone material. A range of composites between keratin and EC using different keratin: EC ratios were prepared and characterised. Comparing keratin to the composites, the FT-IR peak at 1,630 cm^{-1} shifted to a lower wavenumber of 1,610 cm^{-1} in keratin-EC which typically indicates the involvement of β-sheet structures of the keratin during the graft formation process. SEM analysis revealed that the uniform dispersion of the keratin increases the area of keratin-EC contact which further contributes to the efficient functionality of the resulting composites. In comparison to the pristine keratin and EC, a clear

shift in the XRD peaks was also observed at the specific region of 2-Theta values of keratin-*g*-EC. The thermo- mechanical properties of the composites reached their highest levels in comparison to the keratin which was too fragile to be measured for its mechanical properties. Considerable improvement in the water contact angle and surface tension properties was also recorded.

FIGURE FOR ToC - ABSTRACT



1. Introduction

Green chemistry needs to overcome many challenges for successful implementation of innovative technologies to accomplish pollution prevention that reduces and/or eliminates the consumption and/or generation of harmful waste materials during the entire production process. In this regard, ever increasing environmental awareness and the demand for sustainable technology have gained substantial consideration by the academia and industry to develop an eco-friendly processing designs.[1,2] To address the aforementioned concerns of global dependence on petroleum-based resources, attention has been directed to the

engineering of composite materials for targeted applications in different industries.[1-4] Research on several proteins, including collagen, fibroin, keratin, and others is in progress for the development of naturally-derived materials. Among the natural materials, keratinous proteins are interesting candidates to prepare keratin-based composites which have a potential for utilisation in a variety of bio- and non-bio-sectors. A large variety of fibrous keratins are available in the form of feather, hair, nail, and horn as bio-waste. These keratin-rich sources are difficult to degrade as the polypeptide in their structure is tightly packed in α-helix (α-keratin) or β-sheet (β-keratin) into super coiled chains which are strongly stabilised by several hydrogen bonds and hydrophobic interactions, in addition to the di-sulphide bonds.[5, 6]

Keratin-based chicken feathers from butchery account for more than 5 million tons per year worldwide in the form of waste material.[7, 8] Apart from its minor usage in low grade products such as glue, corrugated paper, cardboard, animal feed and fertilisers *etc.*, the landfill disposal of poultry feather poses a significant ecological and environmental threat. On the other hand, from economic and environmental point of view it is also desirable to establish an effective process for the application of such natural sources.[9] Keratin extracted from feathers are small proteins, uniform in size, with a molecular weight between 11-65 kDa.[10,11] The presence of multi-functional groups in keratin, such as di-sulphide, amino, thiol, phenolic and carboxylic, make it reactive under appropriate reaction conditions. Under reducing environment, the amino and other groups mentioned above in keratin make its surface positive, and thus solubilisation takes place.[12] With unique properties of bio-degradability and non-toxic nature, keratin is among versatile biopolymers that can be modified and developed into various products of interests. After modification, keratin composites may find potential applications in bio-medical, pharmaceutical, tissue engineering, and cosmetic industries. [12]

In recent years, much effort has been made to fabricate keratin with other suitable materials. So far most of the work reported deals with blending or grafting of keratin with non-biodegradable synthetic polymers, such as polyethylene (PE), polypropylene (PP), polystyrene, and poly (vinyl chloride).[13-19] To overcome the above said problems many researchers have been proposed to valorise keratinous sources, either through surface grafting or by blending, to prepare innovative graft composites.[7, 10-12] Grafting is preferred over other physiochemical techniques in order to obtain the requisite properties which individual materials fails to demonstrate on their own. Effective modifications should include changes in chemical group functionality, surface charge, biocompatibility and biodegradability.[20]

Enzymatic grafting is quite a new and interesting technique where the enzyme, as an active starting material, offers mild and safe reaction conditions to the current practices in the grafting methods.[2, 20, 21] On the other hand, with respect to health and safety issues linked with other chemical-based procedures, enzymes offer the potential of eliminating the hazards associated with reactive reagents. Moreover, there are several benefits for the use of enzymes in polymeric-based materials synthesis and modifications.[22, 23] Thus, notable amount of information on the characteristics and hydrolysis of keratin has become available where recalcitrant keratinous waste are converted into valuable products.[7, 10, 12] The knowledge on keratin-rich wastes has been robustly increased and their use in cosmetics or in medicines to enhance drug delivery, and production of biodegradable films, are amongst the outstanding and emerging biotechnological and biomedical applications. [23, 24]

This study focuses on the evaluation of raw keratin as a potential material to develop composites with novel characteristics. To the best of our knowledge, in recent literature, laccase-assisted grafting of keratin material onto EC backbone without addition of any plasticiser and/or compatibiliser is not reported. In this work we report that newly grafted composites of keratin and EC with their unique structures under enzymatic environment,

exhibit novel characteristics such as good thermal stability, tensile strength, and hydrophobic/hydrophilic balance.

2. Experimental Section

2.1. Chemicals/reagents

In the present study, a fungal laccase from *Trametes versicolor* with a unit activity of ≥ 10 U/mg was purchased from Sigma-Aldrich Company Ltd., UK and used as received under standard conditions for grafting purposes. All other chemicals and/or reagents used in the current work were of analytical grade and purchased from DIFCO (BD UK Ltd., Oxford, UK), Sigma-Aldrich Company Ltd., UK, and VWR Chemicals (Leicestershire, UK). EC was obtained from Sigma-Aldrich Company Ltd., UK and used as received according to the material safety data sheet provided by the company. Chicken feathers were kindly provided by the Department of Bioengineering, Faculty of Engineering, Ege University, Izmir, Turkey. All of the chemicals and/or reagents were used as received without further purification unless otherwise stated.

2.2. Preliminary processing of the chicken feathers

The chicken feathers were washed three times with hot water and ethylene alcohol to avoid any microbial/dust contamination. Then, they were dried at 60 ^{o}C and cut into small fragments with lengths of 1–2 mm. Clean feathers (10 g) were dissolved in an aqueous solution of 3% wt. NaOH at 50 ^{o}C for 24 h. After the stipulated time the keratin hydrolysate was filtered in order to separate the insoluble parts of feathers, and then was dialysed. After 48 h of dialysis, 2N HCl was added to the keratin hydrolysate in order to extract the keratin in the form of precipitate. The keratin solution was centrifuged at 4000 × g for 10 min, and keratin was washed with distilled water and dried by lyophilisation at -47 °C and at a pressure

of 133 mbar until fully dried. The dried keratin powder was then stored in a desiccated jar to avoid free moisture until used in the subsequent graft synthesis experiments.

2.3. "One-pot" synthesis of composites

Previously extracted keratin from chicken feathers and EC were used to prepare composites with different keratin to EC ratios *i.e.*, keratin: EC; 0:100, 25:75, 50:50, 75:25 and 100:0, respectively. Briefly, the reaction mixture comprised of keratin and EC was prepared using laccase and sodium malonate buffer of pH 4.0 followed by incubation at 120 rpm for 30 min at 25 °C. The above laccase treated mixture was then poured into a sterile labelled petri plate followed by incubation at 50 ^{o}C for 24 h. The newly developed composites were then removed from their respective casting surfaces (petri plate) and designated as keratin-*g*-EC^{A} (prepared using laccase with keratin: EC, 100:0), keratin-*g*-EC^{B} (prepared using laccase with keratin: EC, 75:25), keratin-*g*-EC^{C} (prepared using laccase with keratin: EC, 50:50), keratin-*g*-EC^{D} (prepared using laccase with keratin: EC, 25:75), and keratin-*g*-EC^{E} (prepared using laccase with keratin: EC, 0:100). All of the prepare composites were then characterised using a variety of analytical and imaging techniques as described in the section 2.4.

2.4. Characterisation of composites

2.4.1. Fourier transform infrared spectroscopy (FT-IR)

A Perkin-Elmer System 2000 FT-IR spectrophotometer was used to record the infrared absorption spectra of the grafted composites and their individual counterparts from the wavelength region of 4000-400 cm^{-1}. All spectra were collected with 64 scans and 2 cm^{-1} resolution and assigned peak numbers.

2.4.2. Scanning electron microscopy (SEM)

Scanning electron microscope (Philips, XL-30, FEG SEM; EFI, Netherlands) was used to analyse the surface morphologies of the grafted composites and their individual counterparts in ultra-high vacuum mode at an accelerating voltage of 5 kV. The test composites were prepared using 8 mm diameter aluminium stubs, gold coated for 2 min using the gold spluttering device and then high definition images were recorded.

2.4.3. X-ray diffraction (XRD)

The test composites and their individual counterparts were scanned at 2-Theta values ranging from 10° to 100° with a scan speed of 5 °C/min, using thin film attachment, on a Brüker D-8 Advance X-ray diffractometer equipped with Ni filtered Cu K radiation. An updated MDI/JADE6 software package attached to the Brüker D-8 Advance XRD instrument was used to record the typical XRD diffractograms.

2.4.4. Differential scanning calorimetry (DSC)

To measure the thermal characteristics of the grafted composites and their individual counterparts a Pyris Diamond Differential scanning calorimeter (Perkin-Elmer Instruments, USA) was used. Prior to analyses, test samples were encapsulated in standard aluminium pans to avoid contamination of the ampoules and the thermal profiles were determined using DSC~3-7 mg of sample weight. The temperature range was from -50 ^{o}C to 250 ^{o}C with the scanning rate of 20 ^{o}C/min under nitrogenous atmosphere. DSC analyses were performed in triplicate and results are presented as mean ± S.E. (standard error) of means (n = 3).

2.4.5. Dynamic Mechanical Analyser (DMA)

The mechanical features in terms of Young's modulus, tensile strength and elongation at break point of the composites and individual counterparts were evaluated using a Perkin–Elmer Dynamic Mechanical Analyser. The test composites were cut into a rectangular shape

with 8 mm × 4 mm × 0.25 mm dimensions. The crosshead speed was set at a constant tensile rate of 200 mN min^{-1} with a total range of 1–6000 mN. DMA analyses were performed in triplicate and results are presented as mean ± S.E. (standard error) of means (n = 3).

2.4.6. Water contact angle (WCA)

The hydrophobic/hydrophilic characteristics of the composites and individual counterparts were measured using pendant drop method on a KSV Cam 200 optical contact angle analyser (KSV instruments Ltd., Finland). A Windows-based KSV-Cam software was used to capture the images. Ten independent determinations at different sites of the samples were averaged.

3. Results and Discussion

3.1. Fourier transform infrared spectroscopy (FT-IR)

Figure 1 shows a typical FT-IR spectra of the pure keratin and keratin-EC based composites. The FT-IR spectra shows a peak at 1,630 cm^{-1} and 1,610 cm^{-1} for amide I, 1,530 cm^{-1} with a shoulder at about 1,520 cm^{-1} for amide II, and 1,235 cm^{-1} for amide III, respectively. In particular, the FT-IR spectra (Fig. 1) shows a predominant absorption band at 1,630 cm^{-1} and 1,610 cm^{-1} in case of pure keratin and keratin-EC based composites which typically indicates the involvement of β-sheet structures from the keratin. Based on the literature data, the peak of 1,650 cm^{-1} indicated α-helix structure and the range of 1,631-1,515 cm^{-1} were related to β-sheet structure.[10, 25] Figure 1 illustrates an appearance of new peak at 1,717 cm^{-1} in the grafted composites. Interestingly, relative to the keratin contents in the graft composites a decrease in the intensities of the absorption bands at 1,630 cm^{-1}, 1,610 cm^{-1}, 1,530 cm^{-1}, and 1,235 cm^{-1} was recorded as expected. Evidently, a typical peak at 1,050-1,100 cm^{-1} region is due to the C–O–C stretching which designated to the EC molecules.[26] Furthermore, a high intensity peak at 3,358 cm^{-1} in the graft composite was observed which is linked to the

hydrogen-bonded groups at that distinct band region.[10] The peaks at 2,930 and 2,850 cm^{-1} are characteristic IR bands of aliphatic hydrocarbons of methylene asymmetric C–H stretching and symmetric C–H stretching, respectively. In addition, the positions of these bands indicate the conformations of the protein materials: 1,650 cm^{-1} (α-helix) 1,630 cm^{-1} (β-sheet) for amide I, 1,544 cm^{-1} (α-helix), 1,530 cm^{-1} (random coil) and 1,520 cm^{-1} (β-sheet) for amide II, and 1,230 cm^{-1} (random coil) for amide III).[27] The amino acids chains of keratin present hanging groups such as: amino, hydroxyl, thiol and carboxyl that under a redox system can generate free radicals and thus they can easily react with functional groups of the cellulose.[28] Based on the availability of the functional groups there are different possibilities to propose a mechanism of graft formation between keratin and EC. However, figure 2 is only depicting one potential possibility as a tentative schematic mechanism of graft formation between keratin and EC under laccase-assisted process.

3.2. Scanning electron microscopy (SEM)

The surface morphologies were investigated by SEM, and the respective micrographs are shown in Figure 3 (A-F). It can be observed that in case of the grafted composites the keratin appear homogeneously distributed within the EC backbone. Nevertheless, in the case of the pure keratin film the adhesion seems quite poor in comparison to the keratin film prepared in the presence of the laccase (Fig. 3) and (Fig. 3B) respectively, where large number of pores were observed in the pristine keratin film. Similar results with porous appearance were observed by Selmin et al. [29] during the membrane formation between regenerated keratin and ceramides (CERs). The presence of these pores suggests poor adhesion between CERs and keratin. [29] It has also been reported in literature that the graft copolymerisation modified the surface morphology; physical, chemical, and microstructural characteristics of grafted materials.[2, 30] Better results were obtained in the grafted composites by increasing EC ratios as testified by micrographs (Fig. 3D), and (Fig. 3E) where the keratin material seems more

adherent to the EC backbone with keratin to EC ratios, 50:50 and 25:75 wt. %. With the addition of EC, the distribution of pores was reduced on the surfaces of the grafted composites and finally disappeared as compared to the pristine keratin. On the other hand, the presence of aggregates provides an evidence of the poor dispersion of the material of interests within the polymeric matrix and this behaviour is a consequence of the surface chemical modification that confers a non-polar character to the cellulose surface. [31]

3.3. X-ray diffraction (XRD)

Figure 4 illustrate XRD patterns obtained for pure keratin and keratin-*g*-EC composites (with keratin: EC, 100:0; 75:25; 50:50; 25:75 and 0:100). The pure keratin showed a very broad peak at 2-Theta value of 20^{o}, which specifically corresponds to the □-sheet structure.[32] XRD pattern showed crystallinity in the pure keratin from the substantial 2-Theta peak values with sharp appearance at about 26^{o}, 31^{o}, 45^{o} and 56 ^{o}. In fact, the wider peaks appeared in the keratin film prepared using laccase in comparison to the peaks at 26^{o}, 31 ^{o} 45 ^{o} and 56^{o} obtained for pure keratin which is possibly due to the laccase action. This significant shift in peaks clearly indicates the regeneration and breakdown of amorphous and some crystalline domains at that specific region of 2-Theta values, respectively. However, this phenomenon is more dominant in the case of the graft with keratin: EC ratio 50:50 as compare to the other grafts. The keratin-*g*-EC composites with keratin: EC ratios, 75:25, 50:50 and 25:75 showed a significant peak shift between 20^{o} to 25^{o} (2-Theta). This is due to the occurrence of strong intermolecular and intramolecular bonding interactions which are typically indicate the involvement of β-sheet structures from the keratin as evident by the FT-IR spectra. If there was no or weak interaction between keratin and EC molecules in the graft, each component would have its own individual region and the XRD patterns would be expressed as the simple mixture of keratin and EC. On the other hand, EC had broader diffraction with a broad peak at 20^{o} in the XRD-patterns that represents the highly amorphous nature of EC.[4, 33]

3.4. Differential Scanning Calorimetry (DSC)

Thermal properties such as glass transition temperature (T_g), melting temperature (T_m), crystallisation temperature (T_c) and melting enthalpy (ΔH_m J/g) obtained from the DSC studies are summarised in Table 1. For each specimen an endo-thermal peak was observed during the temperature scan. The first endothermic peak for pure keratin and keratin only film was visible at low temperature about 42 °C and 34 °C, respectively, which is in turn associated with the amount of hydrogen-bound molecules (sometimes referred to as the denaturation peak or glass transition temperature). The results showed a slight change in the glass transition temperature (T_g) from 42 ^oC to 34 ^oC and melting temperature (T_m) from 232 ^oC to 210 ^oC of the keratin film prepared using laccase as compared to the pristine keratin film. Based on the data reported in the literature T_m of the polymers is linked to the size of the polymers and/or dependent on the crystalline domains of the polymers.[34] This initial reduction in the T_m of the keratin film prepared using laccase is an indication of the clear change or disruption in the size of the crystalline domains during the reaction phase. The DSC results are well in support with the observations recorded through XRD studies where the wider peaks appeared in the graft composites as compare to the sharp peaks of the pure keratin at 26^o, 31^o, 45^o and 56^o. However, the introduction of the keratin into the EC determines a shift of the melting temperatures to higher values, indicating an increase of the thermal stability. On high temperature, keratin can contribute to the formation of a protective barrier, on the material surface. This layer delays the heat transfer to the system and diminishes the release of the gaseous products from the system.[35]

3.5. Dynamic Mechanical Analyser (DMA)

Table 2 provides the results of mechanical characteristics of the composites and individual counterparts. The keratin film prepared using laccase was too fragile to permit measurement

of its mechanical characteristics compared to the pure untreated keratin film. However, the composite film prepared using keratin and EC with keratin: EC ratio 75:25 was fairly flexible and strong judged by the tensile strength (22 MPa), elongation at break (37%) and Young's modulus (1.1 GPa). The further increase of EC content ratio resulted in little additional change in the ultimate strength and elongation. However, an increased Young's modulus, suggests that further increase of EC content up to 75% make the film stiffer. As it can be seen in Table 2, keratin film incorporated with 50% wt. EC showed a considerable improvements in the tensile strength (55 MPa), elongation (18%) and Young's modulus (2.4 GPa) in comparison to the pristine keratin. From these results, laccase-assisted grafting of keratin and EC was found to increase mainly the strength and flexibility of the keratin. Based on the literature, appropriate addition of polymers like cellulose, chitosan and glycerol, could control the mechanical properties of keratin films and keratin containing composites.[36] As shown in Table 2, the grafted composites with the "higher" keratin (obtained from chicken feathers) have lower tensile strength. This has also been reported by other researchers,[34] and can be ascribed to the fact that the strength of hydrolysed chicken feathers is insufficient to test for mechanical characteristics. Our tests showed that laccase-assisted grafting brings a slight increase in the tensile strength in the resulting composites relative to the pristine keratin. Recently, it has been reported that laccase treatments can significantly improve the existing and/or impart new physicochemical characteristics including dry and wet tensile strength through various cross-linking reactions.[2, 10, 37]

3.6. Water contact angle (WCA)

The hydrophobic/hydrophilic characteristics in terms of water contact angle and the surface tension properties of the newly synthesised graft composites and their individual counterparts are presented in Figure 5. The water contact angle and surface tension values for the pure keratin film prepared in the absence of laccase were 61^{o} and 42 mN/m, respectively, while in

comparison to this an increase in the water contact angle from 61^{o} to 70^{o} and surface tension from 42 mN/m to 48 mN/m was observed for the keratin film prepared in the presence of laccase (Fig. 5). This initial reduction in the hydrophilicity of the pure keratin film is probably because of the laccase action. The water contact angle values for the graft composites prepared with different keratin to EC ratios *i.e.*, keratin: EC; 100:0, 75:25, 50:50, 25:75 and 0:100 were 25^{o}, 70^{o}, 48^{o}, 52^{o}, 49^{o} and 47^{o} respectively. Hence, with the increase of EC content ratio indicates that the hydrophilic property of keratin-*g*-EC composites is much better than that of the pure keratin. It can be expected that the keratin-*g*-EC composite with high hydrophilicity is more suitable for cell adhesion and proliferation than pure keratin film prepared directly from the keratin hydrolysate. It has also been reported in literature that the hydrophilic/hydrophobic balances are among critical factors which affects the platelet adhesion potential and the cytocompatibility features of the materials, and these properties would endow the polymeric materials as a potential candidate for various biomedical type applications.[2, 10]

4. Conclusions

In this study, in-house extracted keratin from waste chicken feathers in combination with EC was used to develop novel composites using different keratin to EC ratios. According to the results obtained and discussed above, the following conclusions can be drawn:

1) A novel laccase-assisted method was developed to manufacture keratin-EC based novel composites.
2) The technology developed in the present investigation enables the re-use of the chicken feather, a very troublesome waste of the poultry industry, which contributes a major role in the current ecological and/or environmental pollution problems.

3) As evidenced by characterisation analyses, considerable improvement in the morphology, thermo-mechanical and wettability features was recorded in the newly synthesised composites that individual material (keratin) fail to demonstrate on its own.
4) The main benefits of laccase-assisted grafting processes are the facile synthesis in the absence of harmful solvents and chemicals, the mild eco-friendly and energy saving reaction conditions.

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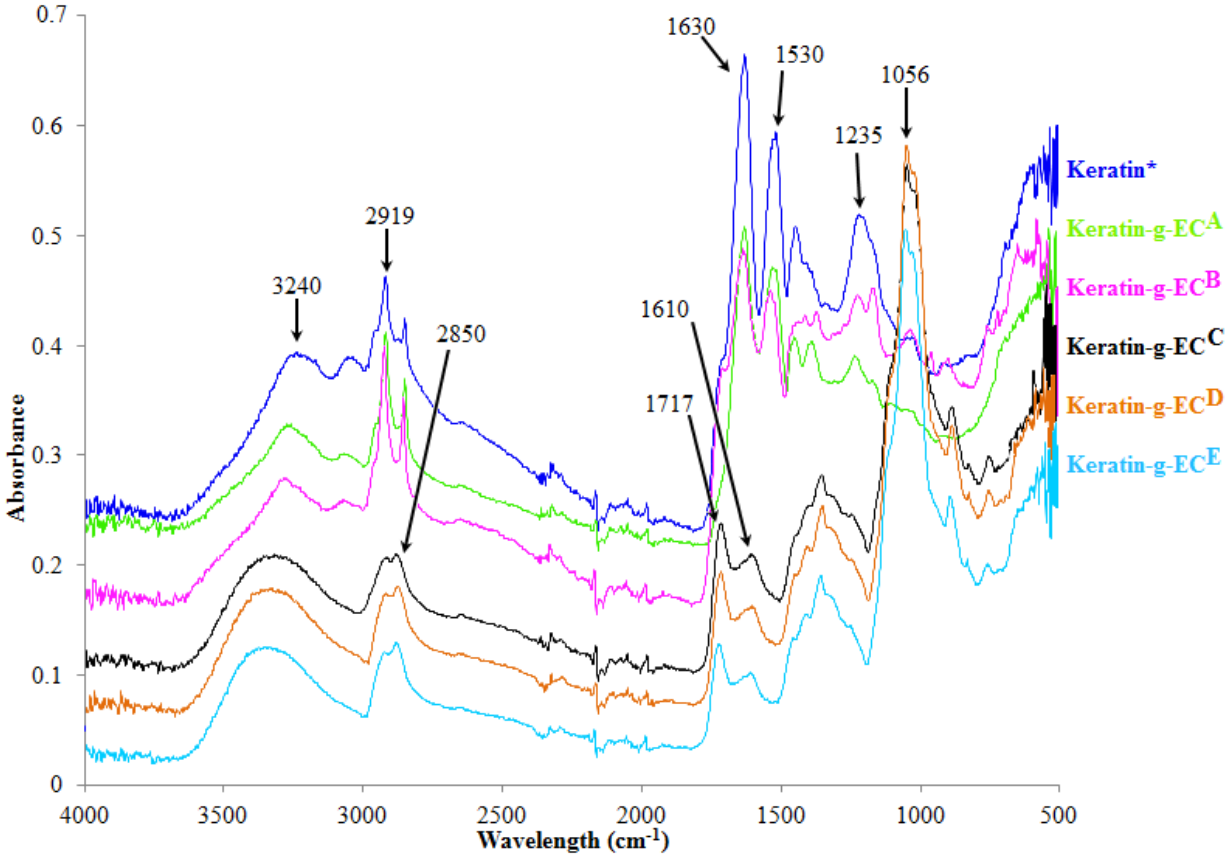

Figure 1. Typical FT-IR spectra of the pure keratin and keratin-EC based graft composites *i.e.*, keratin-*g*-EC^{A}, keratin-*g*-EC^{B}, keratin-*g*-EC^{C}, keratin-*g*-EC^{D}, and keratin-*g*-EC^{E} prepared using laccase as a model catalyst.
*Where, *: Pure keratin film prepared without laccase treatment*

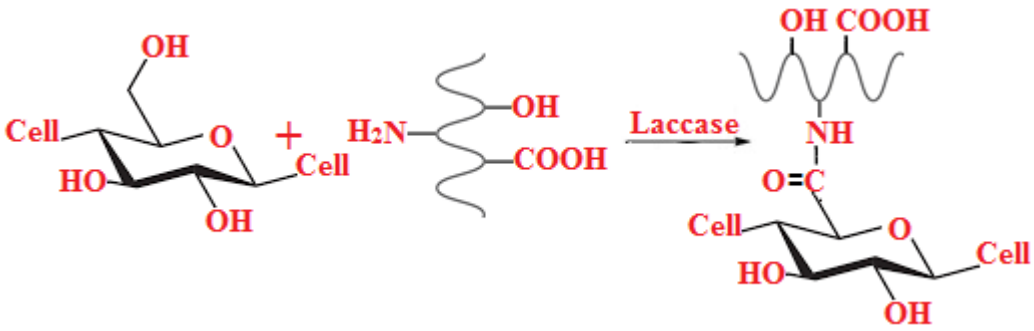

Figure 2. A tentative schematic representation of proposed mechanism of graft formation between keratin and EC under laccase-assisted environment.

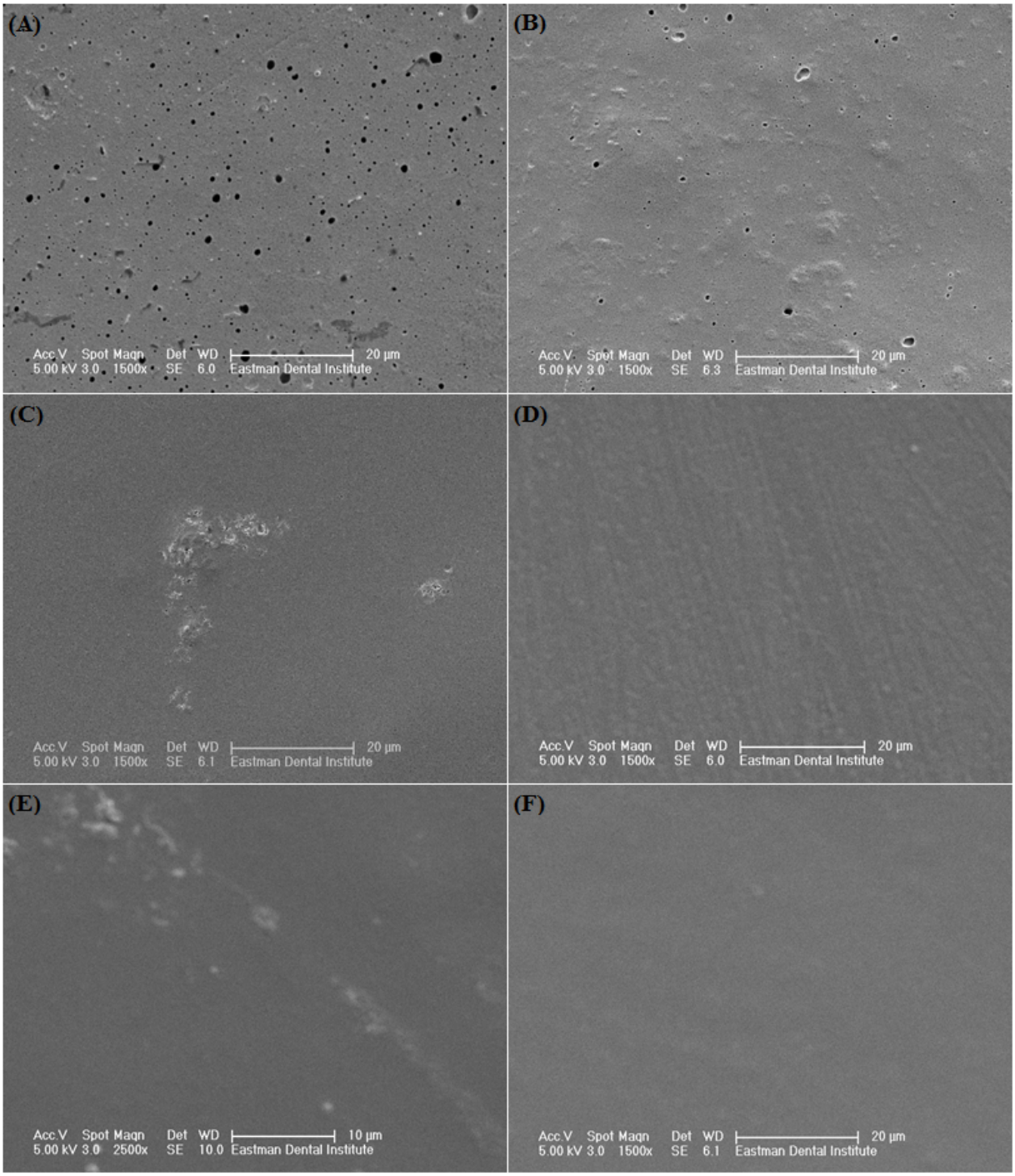

Figure 3. SEM micrograph of the pure keratin (A) and keratin-EC based graft composites *i.e.*, keratin-*g*-EC^{A} (B), keratin-*g*-EC^{B} (C), keratin-*g*-EC^{C} (D), keratin-*g*-EC^{D} (E), and keratin-*g*-EC^{E} (F) prepared using laccase with different keratin to EC ratios.

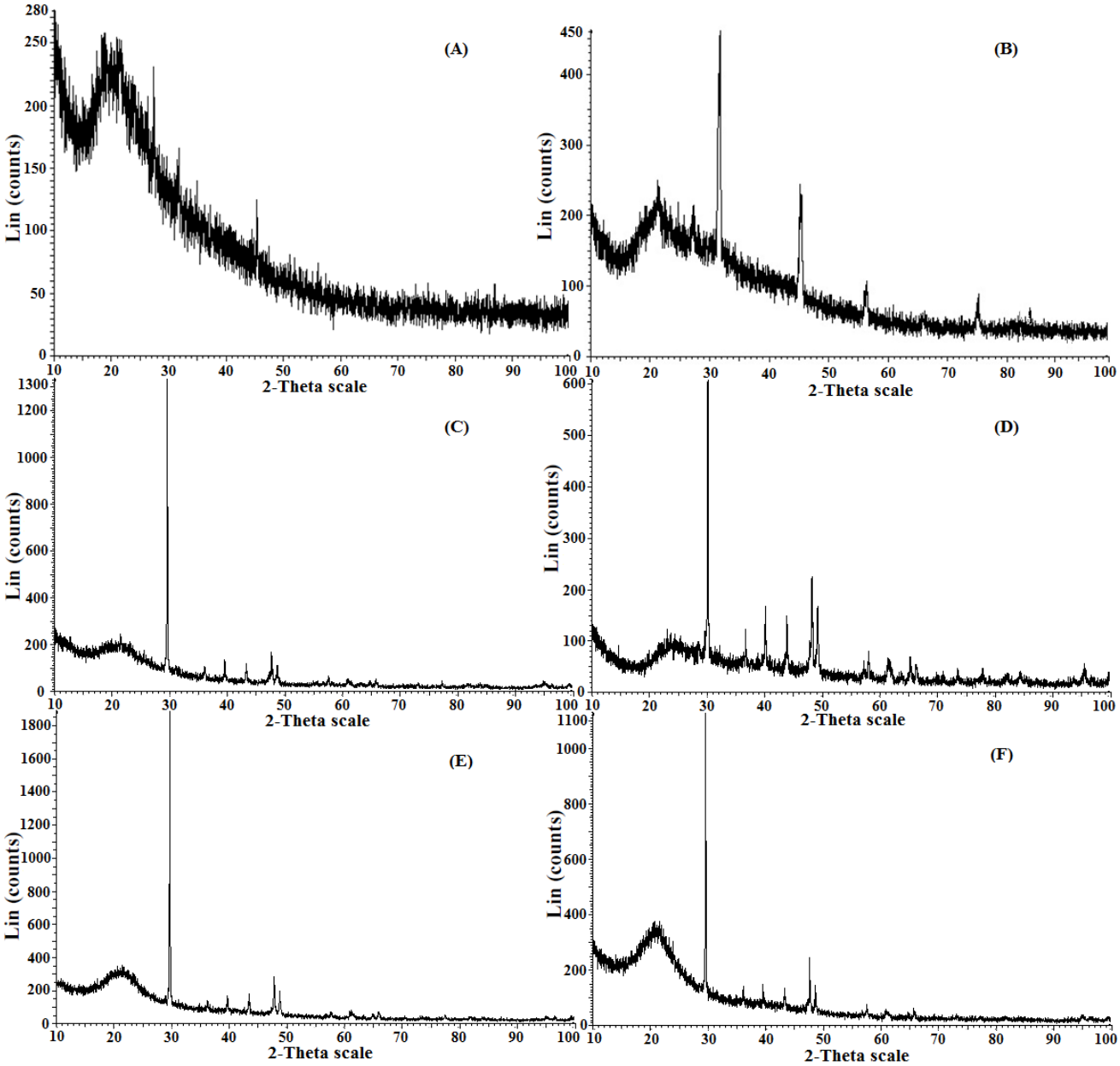

Figure 4. Typical X-ray diffracogram of the pure keratin (A) and keratin-EC based graft composites *i.e.*, keratin-*g*-EC^{A} (B), keratin-*g*-EC^{B} (C), keratin-*g*-EC^{C} (D), keratin-*g*-EC^{D} (E), and keratin-*g*-EC^{E} (F) prepared using laccase with different keratin to EC ratios.

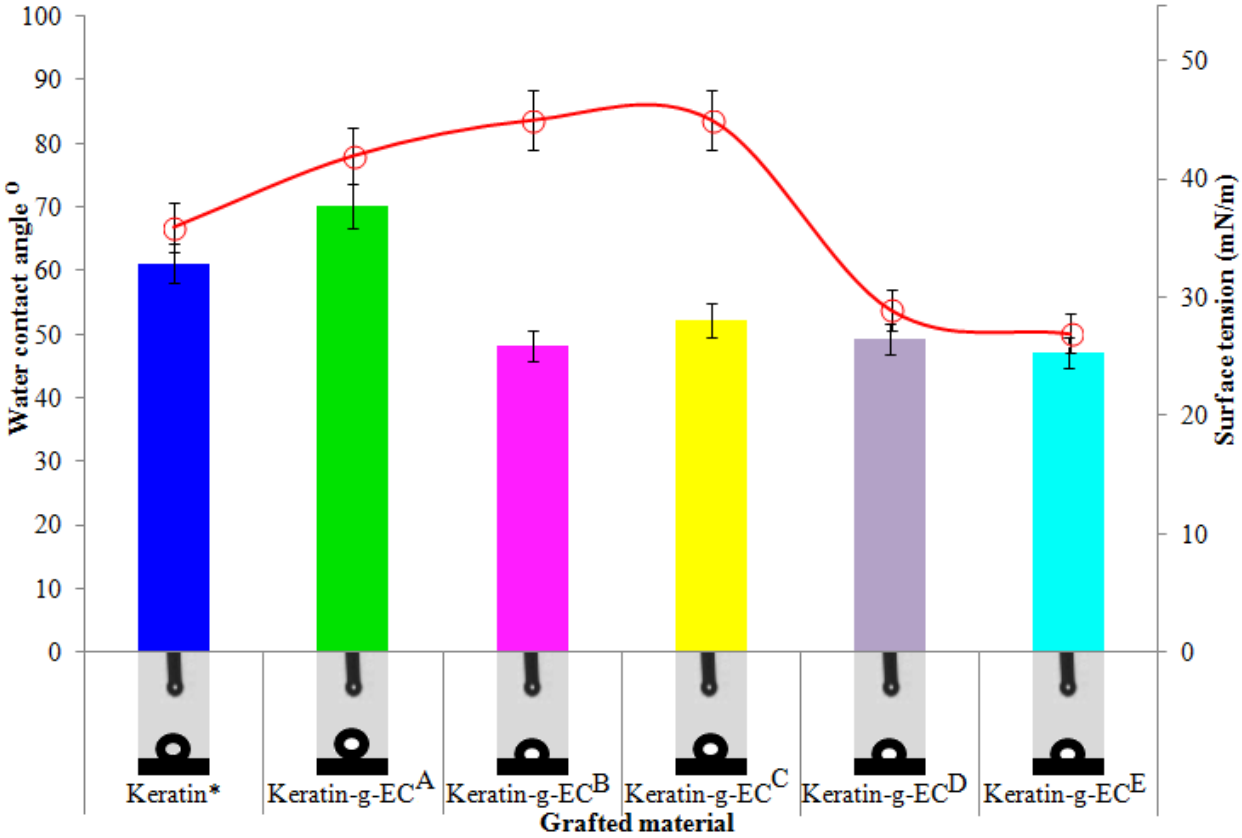

Figure 5. Water contact angle (bars in colour) and surface tension (red line) measurements of the pure keratin and keratin-EC based graft composites *i.e.*, keratin-*g*-EC^{A}, keratin-*g*-EC^{B}, keratin-*g*-EC^{C}, keratin-*g*-EC^{D}, and keratin-*g*-EC^{E} prepared using laccase with different keratin to EC ratios.

Where, *: Pure keratin film prepared without laccase treatment

Table 1. DSC-thermal characteristics of the individual polymers *i.e.*, keratin and EC and their grafted composites *i.e.*, keratin-*g*-EC^{A}, keratin-*g*-EC^{B}, keratin-*g*-EC^{C}, keratin-*g*-EC^{D}, and keratin-*g*-EC^{E} prepared using laccase with different keratin to EC ratios.

Sample ID	**T_g (°C)**	**T_m (°C)**	**T_c (°C)**	**ΔH_m (J/g)**
Keratin[a]	42±0.35	232±2.22	112±1.50	31±1.25
Keratin-g-EC^{A}	34±0.31	210±1.22	177±1.35	24±0.35
Keratin-g-EC^{B}	74±0.20	228±2.65	165±2.10	16±0.08
Keratin-g-EC^{C}	105±0.95	223±1.10	169±1.85	9.5±0.45
Keratin-g-EC^{D}	88±1.10	212±2.20	155±2.21	12±0.12
Keratin-g-EC^{E}	[b]	199±1.65	120±1.32	92±1.11

[a)] Pure keratin film prepared without laccase treatment; [b)] not detected.

Table 2. DMA-mechanical characteristics of the individual polymers *i.e.*, keratin and EC and their grafted composites *i.e.*, keratin-*g*-EC^{A}, keratin-*g*-EC^{B}, keratin-*g*-EC^{C}, keratin-*g*-EC^{D}, and keratin-*g*-EC^{E} prepared using laccase with different keratin to EC ratios.

Sample ID	**Tensile strength (MPa)**	**Young's modulus (GPa)**	**Elongation at break (%)**
Keratin[a]	[b]	[b]	[b]
Keratin-g-EC^{A}	[b]	[b]	[b]
Keratin-g-EC^{B}	22±0.09	1.1±0.31	37±0.33
Keratin-g-EC^{C}	55±0.15	2.4±0.42	18±0.45
Keratin-g-EC^{D}	91±0.21	3.1±0.25	8±0.28
Keratin-g-EC^{E}	122±0.42	3.5±0.65	9±0.15

[a)] Pure keratin film prepared without laccase treatment; [b)] not detected.

Table of Contents

This study focuses on the evaluation of raw keratin as a potential material to develop composites with novel characteristics. Herein, we report a mild and eco-friendly fabrication of in-house extracted feather keratin-based novel composites consisting of ethyl cellulose as a backbone material.