CLEANING AND STERILIZATION OF SHEEP WOOL FIBER USING SUPERCRITICAL CARBON DIOXIDE

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by

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LIST OF SYMBOLS

A	High asymptote value (stensby)
N	Viable cell counts after treatment
N_o	Initial cell counts
R^2	Regression coefficients
t_t	Complete inactivation time
λ	Time for Lag phase
k	Maximum cleaning rate (min$_{-1}$)
t	Time
H_2O_2	Hydrogen peroxide
+ve	Gram-positive
-ve	Gram negative
μg	Microgram
O	Oxygen
C	Carbone
N	Nitrogen
S	Sulfur
μm	Micrometre
H2SO4	Sulphuric acid
KBr	Potassium Bromide
Log	Logarithm
CO2	Carbon dioxide
h	Hour
OC	Celsius

LIST OF ABBREVIATIONS

B. cereus	*Bacillus cereus*
B. subtilis	*Bacillus subtilis*
*B. velezensi*s	*Bacillus. Velezensis*
Cal	Calculated values
CCD	Central composite design
EDX	Energy Dispersive X-Ray
CFU	Conoly forming unit
CIE	Commission International Eclairage
CMC	Cell membrane complex
COD	Chemical oxygen demand
CV-I	Crystal violet-iodine
E. cloacae	*Enterobacter cloacae*
Exp:	Experimental data
IWTO	International Wool Textile Organization
K. pneumoniae	*Klebsiella pneumoniae*
min	Minute
MPa	Megapascal
NA	Nutrient agar
rDNA	Ribosomal deoxyribonucleic acid
RSM	Response surface methodology
HDP	High-energy discrete processing
BOD	Biological oxygen demand
SAS	Surface-active substances
FTIR	Fourier Transform Infrared Spectrometer
SEM	Scanning Electron Microscopy
$ScCO_2$	Supercritical carbon dioxide
XRD	X-ray powder diffraction
NCBI	National centre for biotechnology information
PCR	Polymerase chain reaction
RSS	Residual sum of the squares

S. aureus	*Staphylococccus aureus*
$SFCO_2$	Supercritical fluids carbon dioxide
T	Absolute temperature
VM	Vegetable matter
WI	Whiteness index
KHz	Kilohertz
MHz	Megahertz
UV	Ultraviolet
TG	Thermogravimetric
DTG	Differential thermogravimetric
EDTA	Ethylenediaminetetraacetic acid
PED	Pulsed electrohydraulic discharge
ASTM	American Society for Testing and Materials
PLATS	Basic Local Alignment Search Tool

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PEMBERSIHAN DAN PENSTERILAN SERAT BULU BEBIRI MENGGUNAKAN SUPERKRITIKAL KARBON DIOKSIDA

ABSTRAK

Bulu biri-biri digunakan sebagai bahan tekstil untuk pakaian dan fabrik berkualiti tinggi dalam industri tekstil moden. Bulu biri-biri mentah mengandungi kotoran, sut dan lilin. Oleh itu ia memerlukan proses pembersihan yang berkesan untuk membuang bahan cemar. Pemprosesan bulu biri-biri sedia ada memerlukan sejumlah besar air tawar dan penambahan bahan kimia toksik yang mengakibatkan penjanaan efluen toksik dan kerosakan gentian bulu. Selain itu, tercemar dengan bakteria patogen. Oleh itu, teknologi yang berkesan digesa untuk menghapuskan mikroorganisma patogen untuk pengendalian yang selamat. Dalam kajian ini, karbon dioksida superkritikal scCO_2 digunakan untuk merawat gentian bulu biri-biri sebagai alternatif tanpa air dan berkesan kepada pemprosesan bulu biri-biri sedia ada. Bakteria patogen dalam serat bulu biri-biri ditentukan menggunakan pengenalpastian molekul dengan asid ribonukleik ribosom 16sRNA. Pengaruh pensterilan scCO_2 dan pembersihan gentian bulu biri-biri ditentukan dengan tekanan yang berbeza-beza (10-40 MPa), suhu (32-80 ^{o}C), dan masa rawatan (30-90 min). Selepas itu, sifat fizikokimia, termo-mekanikal dan morfologi permukaan scCO_2 yang dirawat dan gentian tidak dirawat telah ditentukan menggunakan pelbagai kaedah analisis. Enam strain bakteria telah diasingkan dalam gentian bulu biri-biri, seperti Bacillus Velezensis, Bacillus amyloliquefaciens, Klebsiella pneumonia, Enterobacter cloacae, Exiguobacterium, dan Bacillus cereus. Didapati bahawa tekanan adalah parameter yang paling berpengaruh, diikuti oleh suhu dalam kedua-dua pensterilan dan pembersihan. Pengaruh scCO_2 untuk penyahaktifan lengkap bakteria dicapai pada tekanan 29 MPa, suhu 55 ^{o}C dan masa 75 min. Indeks keputihan maksimum (25.2

stensby) dicapai pada tekanan 30 MPa, suhu 69 ^{o}C, dan masa 80 minit dicapai. Analisis XRD menunjukkan bahawa indeks kehabluran meningkat daripada 28.1 % kepada 31.6 % pada keadaan eksperimen yang dioptimumkan. Analisis FTIR mendedahkan kehadiran kumpulan berfungsi dalam gentian bulu biri-biri yang tidak dirawat dan dirawat. Mikroskop cahaya dan analisis SEM menunjukkan bahawa scCO_2 berkesan menghilangkan kekotoran. Kestabilan terma gentian bulu bertambah baik dengan pembersihan scCO_2, di mana suhu permulaan 262.17°C dan berat kehilangan adalah kira-kira 25.5 wt%. Sifat mekanikal, seperti kekuatan tegangan, pemanjangan pada putus dan keanjalan modulus gentian bulu biri-biri 2183.8 MPa, 39.54 %, dan 5588.85 MPa, masing-masing meningkat dengan scCO_2. Penggunaan persamaan Gompertz yang diubah suai mendedahkan bahawa pembersihan scCO_2 gentian bulu mengikut lengkung sigmoid. Penemuan kajian ini menunjukkan bahawa scCO_2 adalah teknologi yang berkesan untuk mensteril dan membersihkan gentian bulu biri-biri. Oleh itu, scCO_2, sebagai teknologi pensterilan dan pembersihan tanpa air, boleh dilaksanakan dalam pemprosesan gentian bulu biri-biri untuk meminimumkan kerosakan gentian bulu biri-biri dan meminimumkan penjanaan efluen toksik.

CLEANING AND STERILIZATION OF SHEEP WOOL FIBRE USING SUPERCRITICAL CARBON DIOXIDE

ABSTRACT

Sheep wool is utilized as a textile material for clothing and high-quality fabric in the modern textile industry. Raw sheep wool contains dirt, suint and wax. Thus it requires an effective cleaning process to remove contaminants. Existing sheep wool processing requires a huge amount of fresh water and the addition of toxic chemicals which results in the generation of toxic effluent and wool fibre damage. Besides, contaminates with pathogenic bacteria. Therefore, it is urged for an effective technology to eliminate pathogenic microorganisms for safe handling. In this study, supercritical carbon dioxide $scCO_2$ was used to treat sheep wool fibre as a waterless and effective alternative to existing sheep wool processing. The pathogenic bacteria in sheep wool fibre were determined using molecular identification with ribosomal ribonucleic acid 16sRNA. The influences of $scCO_2$ sterilization and cleaning of sheep wool fibre were determined with varying pressure (10-40 MPa), temperature (32-80 ^{o}C), and treatment time (30-90 min). Subsequently, the physicochemical, thermo-mechanical properties and surface morphology of $scCO_2$ treated and untreated fibre were determined using various analytical means. Six bacteria strains were isolated in sheep wool fibre, such as *Bacillus Velezensis, Bacillus amyloliquefaciens, Klebsiella pneumonia, Enterobacter cloacae, Exiguobacterium, and Bacillus cereu*s. It was found that pressure is the most influential parameter, followed by temperature in both sterilization and cleaning. The influence of $scCO_2$ for complete inactivation of bacteria was achieved at a pressure of 29 MPa, a temperature of 55 ^{o}C and a time of 75 min. The maximum whiteness index (25.2 stensby) was achieved at a pressure of 30 MPa, a temperature of 69 ^{o}C, and a time of 80 min reached. XRD analyses showed that the crystallinity index increased from 28.1 % to 31.6 % at optimized experimental conditions. FTIR analyses revealed the presence of the functional groups in untreated and treated sheep wool fibre. The light microscope and SEM analyses showed that the scCO_2 effectively removed the impurities. The thermal stability of wool fibre considerably improves with the scCO_2 cleaning, where the onset temperature of 262.17°C and loss weight was approximately 25.5 wt %. Mechanical properties, such as tensile strength, elongation at break and modulus elasticity of sheep wool fibre 2183.8 MPa, 39.54 %, and 5588.85 MPa, respectively increased with scCO_2. The application of the modified Gompertz equation reveals that the scCO_2 cleaning of wool fibre follows the sigmoidal curve. The present study's findings showed that scCO_2 is an effective technology for sterilizing and cleaning sheep wool fibre. Therefore, scCO_2, as waterless sterilization and cleaning technology, could be implemented in sheep wool fibre processing to minimize sheep wool fibre damage and minimize toxic effluent generation.

CHAPTER 1

INTRODUCTION

1.1 Background of research

Sheep wool fibre has been used extensively as a textile material for garments and high-quality texture in modern textile factories (Sabir 2018). The excellent intrinsic properties of this fibre could bring about great marketing potential in textiles and other fields as well (Kumar and Suganya 2017). The wool fibre has unique physical and chemical properties that allow it to be extremely versatile. Chemical features of wool include a high content of pure keratin that contains amino acids, such as glycine, alanine, serine, proline, naline, threoninec, cystine, lucien isomers, aspartic acid glutamic acid, methionine, histidine, hydroxylsine, phenylalanine, arginin, tyrosine, and tryptophane. The structure and chemical composition of the wool fibre is extremely different from other kinds of fibres and illustrate huge diversity and heterogeneity of characteristics, and advantages found in no other natural or manmade fibre (Henry et al., 2019). Wool which is an abundant and renewable proteinous resource is mainly composed of approximately 82 % high-cystine-content keratinous proteins localized in the scale and cortex layers, 17.2 % low-cystine-content non-keratinous proteins and 0.8 % nonprotein materials on the surface of scale layer (Rippon and Evans 2020). Because wool contains highly cross-linked disulfide bonds, especially in the scale and cortex layers (Li 2019). Wool can be warm or cool, it can be casual or formal and it has moisture absorbing properties that allow it to be comfortable to wear in active and passive situations. Wool is a renewable energy resource, where the average sheep produces between 2.3 and 3.6 kg of raw wool annually that must be sheared (Fantilli et al., 2017). From the time wool grows on the sheep, on through the stages of manufacture and wear, the fibres are subject continually to contamination with microorganisms included in this micro-organic flora that may be the pathogenic organisms responsible for the disease. As a result, the effect of microbes on wool may have a significant impact on its use. Oftentimes wool must be sterilized to free it of pathogenic organisms, and of the nonpathogens which damage it. Wool is an important protein fibre and is in high demand due to its excellent mechanical properties such as good flexibility, warmth retention, high hygroscopicity, and soft handling (Erdogan et al., 2020). Raw wool can contain large amounts of surface impurities made up of wool wax and grease. In addition, contains water-soluble materials such as suint (formed from dried perspiration), and inorganic mineral dirt (Madara and Namango, 2014). In the textile industry, wool fibre is subjected to a series of water-based treatment processes and procedures (Alebeid et al., 2020). Wool cleaning is required to prepare raw wool in the textile industry resulting in better high-quality fabric (Li et al 2017).

Besides, sheep wool contaminates with pathogenic bacteria. Therefore, effective sterilization technology is needed to eradicate pathogenic bacteria for safe handling. Supercritical CO_2 has been utilized as good sterilization and cleaning technology in various industries, including food, pharmaceutical, healthcare and materials processing. In the present study, $scCO_2$ was used to treat sheep wool fibre as a waterless and effective alternative to existing sheep wool processing technology (Delma et al., 2020). $ScCO_2$ refers to the carbon dioxide gas in a supercritical state when its temperature and pressure are greater than the critical point. Under standard temperature and pressure conditions, CO_2 behaves like a gas. When the temperature and pressure of CO_2 are increased above its critical points, 31.1 °C, and 7.4 MPa respectively, its properties change to that of neither a gas nor a liquid (Hossain et al., 2016). At this point, CO_2 is referred to as $scCO_2$. The treatment of wool in supercritical carbon dioxide fluid is an environmentally friendly and advantageous solution for cleaner production of wool textiles with water-free and energy preservation, as well as avoiding a large amount of effluent discharge and serious environmental pollution in conventional wet chemical processes (long et al., 2013). In the present work, some basic supports for the applications of supercritical carbon dioxide fluid in the cleaner production of wool were developed at different system pressures. Particularly, in the last decades, a supercritical carbon dioxide fluid was introduced to the textile industry as an alternative to conventional water-based processes, with numerous advantages (Amenaghawon et al., 2020). Waterless techniques using supercritical carbon dioxide fluid to extract lanolin, wool wax, ceramides, and synthetic pyrethroids from raw wool are accessible in the literature due to their ecologically favourable characteristics and economic benefits, such as treatment cost and availability (Long et al 2013; Zakaria El-Sayed et al. 2018).

1.2 Problem statement

Raw wool is a dirty natural fibre, containing a high level of impurities such as wool grease, suint, and dust. Therefore, before being used as a textile material, raw shorn wool involves a long and water-based treatment process and various procedures, such as washing and scouring, fulling and bleaching, carbonization, dyeing, functional and finishing. Furthermore, requires chemicals such as (soap, sodium carbonate, sulphuric acid, sodium, potassium, and ammonia soaps). Involve the use of large volumes of water ranging from (659 l/ kg, to 111 l/ kg) causing the depletion of water fresh resources (Ghaly et al., 2013). Consequently, highly toxic and a large amount of effluents are produced from the conventional wet-chemical treatments with significant pollutant loads of (COD) 25,000 mg/l to 60,000 (Wang et al., 2016; Holkar et al., 2016) and a high-energy consumption is involved which making the production of wool fibre costly and as result in a large quantity of the wool is wasted annually affecting an ecosystem (Mazinani and Rude 2020). In fact, about 75 % of the wool produced by European sheep farms cannot be used by the textile industry. Due to some mechanical treatments, performed to improve the workability, the quality of this wool is generally high (Martin and Herlaar 2021; Cardinale et al., 2017).

The handling and processing of raw sheep wool may pose potential hazards to personnel handling sheep wool. *Bacillus anthrax and Klebsiella pneumoniae* were found in the wool processing factory and are infections of human pathogens that cause some diseases such as bacteraemia, pneumonia, and urinary tract. These bacteria are listed in the human pathogen hazard group (Fan et al. 2018; Giannitti et al., 2018). Certain microbes, on the other hand, may not be pathogenic to sheep, but under conditions, they may rapidly grow and discolour wool fibres with mildew. The activity of various microbes can hamper the utilization of wool. Bacteria damage wool fibres by their metabolites, bond breaks (hydrogen bonds, and disulphide bonds) which provide stability of protein structure, causing deterioration of mechanical properties and reducing breaking load and causing “yellowing” of the wool fibres (Guiamet et al 2016). Annual losses due to microbiological damage to wool fibre reach hundreds of millions of dollars hence; wool must be sterilized to eliminate pathogenic organisms and other microbes that can easily cause damage to it.

1.3 Objectives of research

The main aim of this study is to determine effective processing for the sterilization and cleaning of sheep wool fibre using $scCO_2$. The specific objectives are:

i. To identify the presence of bacteria present in sheep wool fibre using 16 sRNA.

ii. To determine the influence of $scCO_2$ parameters (pressure, temperature, and time) on the sterilization and cleaning of sheep wool.

iii. To illustrate $scCO_2$ cleaning mechanisms by increasing the whiteness in sheep wool using Gompertz modified equation.

iv. To evaluate the $scCO_2$ process by characterizing the physiochemical (crystallinity, chemical structure and organic compounds), thermal (stability) and mechanical (strength, elongation and modulus of elasticity) properties of wool fibre.

1.4 Scope of research

The research emphasizes a new supercritical carbon dioxide ($scCO_2$) sterilization and cleaning technique used for sheep wool fibre processing.

i. In the case of microbes' resistance presence in sheep wool fibre, the supercritical carbon dioxide sterilization technique is used to complete inactivation by optimizing the temperature, pressure and sterilization time.

ii. Sheep wool fibre contained impurities that cause non-white wool; the supercritical carbon dioxide cleaning technique is used for improving the whiteness index by optimizing the temperature, pressure and cleaning time. Gompertz modified equation studies used in supercritical carbon dioxide to illustrate $scCO_2$ cleaning mechanisms by increasing the whiteness in sheep wool using.

iii. Characterization of physicochemical, mechanical, and thermal properties of treated wool fibre to evaluate supercritical carbon dioxide cleaning process effect on the wool fibre.

1.5 Significance of research

Wool an excellent natural protein fibre derived from animals, e.g. sheep, has been widely utilized in the production of clothing materials and technical textiles in the medical, aerospace, automotive, and decoration fields. It is a major raw material in the fabric industry. However, raw sheep wool is a dirty natural fibre because it contains high levels of impurities, such as grease, suint, vegetable matter, and dust which need to be removed through cleaning processes before its use as a textile material. Cleaning methods for sheep wool processing are currently used in the textile sector owing to the enormous volume of toxic effluents generated. A large portion of these effluents comprises toxic volatile organic compounds necessary for the water-based cleaning of sheep wool. Besides the impurities, raw sheep wool may pose potential hazards to personnel handling sheep wool this is because sheep wool fibre is susceptible to microbial contamination. Certain microorganisms can rapidly multiply and stain wool fibres with mildew under favourable conditions and resulting in loss of strength and leading to deterioration. Moreover, most of these organisms exist as spores and are highly resistant to conventional water-based treatment methods. Supercritical carbon dioxide (scCO_2) technology has been extensively utilized in various industrial fields and offers numerous advantages as a cleaner sterilization technology, such as easy separation, shorter processing time, environment-friendliness, waterless nature, high solubility, and treatment of heat-sensitive materials without causing damage.

1.6 Thesis Outline

The thesis has structured into five chapters; the contents of all chapters are summarized as follows:

Chapter 1: This chapter showed the introduction of the research including the background of the study, problem statements with objectives of the research and the scope of the study, which the research attempts to address using supercritical carbon dioxide technology as cleaning and sterilizing sheep wool fibre.

Chapter 2: A literature review. The chapter has discussed the literature review of previous research on supercritical carbon dioxide cleaning, sterilizing and extraction. Furthermore, on sheep wool fibre utilization, structure, impurities of wool fibre and existing sheep wool fibre cleaning technology with potential study of using $scCO_2$ as new sheep wool fibre process technology.

Chapter 3: Materials and methods. This chapter provided an overview of the research, which focused on detecting germs attached to wool fibres, and sterilizing and cleaning wool fibre with $scCO_2$. Using the response surface approach, the experimental settings for $scCO_2$ sterilization and cleaning of sheep wool fibre were optimised based on bacteria inactivation and whiteness index. Further, this chapter has also examined the physiochemical, thermal, and mechanical properties of wool fibre treated by $scCO_2$. The mechanisms of the $scCO_2$ cleaning of sheep wool were assessed with a mathematical model using Gompertz modified equation.

Chapter 4: Results and discussion. This chapter is mainly designed to identify the bacteria in wool fibre and to inactivate it using $scCO_2$ technology. This chapter's major goal is to clean and sterilize wool fibre by measuring the whiteness index and activation rate. RSM is used to create the best cleaning and sterilizing conditions possible. Moreover, the physicochemical, thermo-mechanical properties and surface morphology of $scCO_2$ treated and untreated sheep wool fibre were determined using various analytical means, such as thermogravimetric analyses (TGA), light microscope, X-ray diffraction (XRD), Fourier transform infrared spectroscopy (FTIR), Scanning electron microscopy (SEM) with Energy dispersive X-Ray analysis (EDX).

Chapter 5: Conclusion and recommendations. This chapter described the findings, conclusions, and recommendations based on the analysis results. In this final chapter, the findings are outlined, and the possibilities for further research are indicated.

CHAPTER 2

LITERATURE REVIEW

2.1 Sheep wool fibre

Wool is an outstanding multifunctional natural animal fibre and has a high demand due to its excellent strength, high hygroscopic properties, good insulation, and long lifespan (Niu et al., 2012). Its physical and chemical structure has made it important since the ancient age of humanity. The earliest clear evidence of the use of wool fibre to produce textiles was textiles made of sheep wool found in Shahr-i Sokhta, eastern Iran and Novosvobodnaya in the northern Caucasus (Shishlina et al., 2020). Wool is considered a possible solution. There is archaeological evidence that wool technology has been used for at least 6000 years by human forebears not only for garments but also as an insulative building material and for the storage of fresh foodstuff (Morris and Spilsbury 2016). However, the preparation of wool for final use is difficult and complicated, and every stage of production requires intensive care (Pan et al 2018). Wool is subject to various scanning methods for the manufacture of high-quality finished goods (Zhang et al., 2016).

Raw wool is polluted with impurities that vary depending on the type of sheep breed, the region and the environment in which the sheep are reared (Zhang et al., 2016). Each kg of raw contaminated wool contains approximately 150 g of lanolin, 40 g of suint, 150 g of dirt, 20 g of vegetable matter, and only 640 g of wool fibre yield (Madara and Namango, 2014). Cleaning is the first stage of the processing of wool and is necessary for the determination of the quality of the fibre and the removal of pollutants from the fibre surface (li et al., 2014). The evaluation of the finished product quality is complex and complicated, where the raw wool goes under preparation processes such as carding, dyeing, dirt removal and other realistic methods.

Since raw sheep wool contains various impurities, it must be clean to remove impurities before its use in the manufacture of textile products. Scouring is the most important step in the processing of sheep wool. Although the term "wool scouring" refers to the process of washing and drying wool, in reality, there are several other steps involved in this process, such as blending, mechanical cleaning, and sampling. In addition, several types of chemical treatments are used in conjunction with the scouring process (Li et al., 2017). Determining the quality of fibre cleaning and removal of contaminants from the surface of the fibre depends on factors such as the nature of the contaminants, detergent, temperature and mechanical impact on the surface of the fibre (li et al., 2014; Romanovska and Oseiko., 2017).

2.1.1 Production, properties and structure

Wool is a natural fibre whose specifications are determined by several production variables. According to the statistics reported by the International Sheep Wool Organization, the worldwide production of clean sheep wool was estimated to be approximately 1.155 million kg in 2018 (IWTO, 2019). Generally, sheep wool is collected by trimming wool from sheep once a year during summer or spring. Australia produced the highest amount of clean sheep wool, i.e., 23.4 % of the total worldwide production of sheep wool in 2018. Other major sheep-wool-producing countries are China (15.5 %), Russia (11.4 %), New Zealand (9.1 %), Argentina (2.3 %), South Africa (2.2 %), the UK (2.2 %), and Uruguay (1.6 %). Clean sheep wool can be categorized according to the average diameter of the wool fibre into fine wool (24.5 mm), medium wool (24.6-32.5 mm), and coarse wool (>32.5 mm) fibres. The types of wool obtained from various types of sheep and their characteristics are shown in Table 2.1.

Table 2.1 Sheep wool from various types of sheep and their characteristics

Type of wool	Type of sheep	Fibre diameter (µm)	Staple length (cm)	Spinning count (s)	Mature body weight (lb)	
					Ram	Ewe
Fine sheep wool	Delaine Merino	22-17	2.5 – 4	64-80	190- 240	125-160
	Rambouillet	24-19	2.5 -4	60-80	200-300	140-180
	Deboillet	22-18	3-5	64-80	220-275	125-160
Medium sheep wool	Targhee	25-21	3-5	58-64	200-300	140-200
	Finnsheep	31-24	3-6	48-60	160-200	120-160
	Columbia	30-23	4-6	50-62	250-350	160-240
	Dorset polled	33-26	3-4.5	46-58	225-275	150-200
	Montadale	30-25	3-5	56-58	200-275	160-180
	Corriedale	31-25	3.5-6	50-58	220-275	150-200
	Cheviot	33-27	2.5-4	46-56	160-200	120-160
	Shropshire	33-25	3-4	46-58	225-290	170-200
	Southdown	30-25	2-3	50-58	180-230	120-180
	Suffolk	33-26	2-3.5	46-58	275-400	200-300
	Oxford	34-28	3-5	46-54	225-325	150-200
Coarse sheep wool	Border Leicest	38-30	5-10	36-48	225-300	150-225
	Lincoln	41-34	8-15	36-46	240-300	200-250
	Romney	39-32	5-8	36-48	200-275	150-200

Source; American wool council (www.sheepusa.org)

The classification of sheep wool fibre is determined depending on the breed of sheep. The sources of fine sheep wool are Delaine Merino sheep, Rambouillet sheep, and Deboillet sheep. Fine sheep wool has the shortest staple length and the highest spinning count. Wool is considered a lavish natural fibre owing to its low weight, dirt resistance, durability, and water repellence. Wool can absorb approximately 30 % of its weight in water, which can be released subsequently (Patnaik et al., 2015). Although sheep wool is water repellent but able to absorb moisture. This is because of the cuticle and cortical cell components of sheep wool fibre. The cuticle cells of sheep wool are coated with waxy materials, which makes sheep wool water repellent. Wherein, the cortical cells of sheep wool contain high sulfur proteins that absorb the water molecule. This property makes wool breathable and comfortable to wear. It helps regulate body temperature by absorbing sweat and releasing it as vapour, thereby preventing the clammy, cold feeling experienced when some synthetic materials are worn during hot weather. The ability of wool to retain moisture makes it resistant to static electricity; hence, wool garments have a minimal chance of sparking or clinging to the body (Kim and Kim, 2019). The chemical structure of the wool fibre is shown in Figure 2.1 A (functional group of wool fibre) and Figure 2.1 B (cross-linkage).

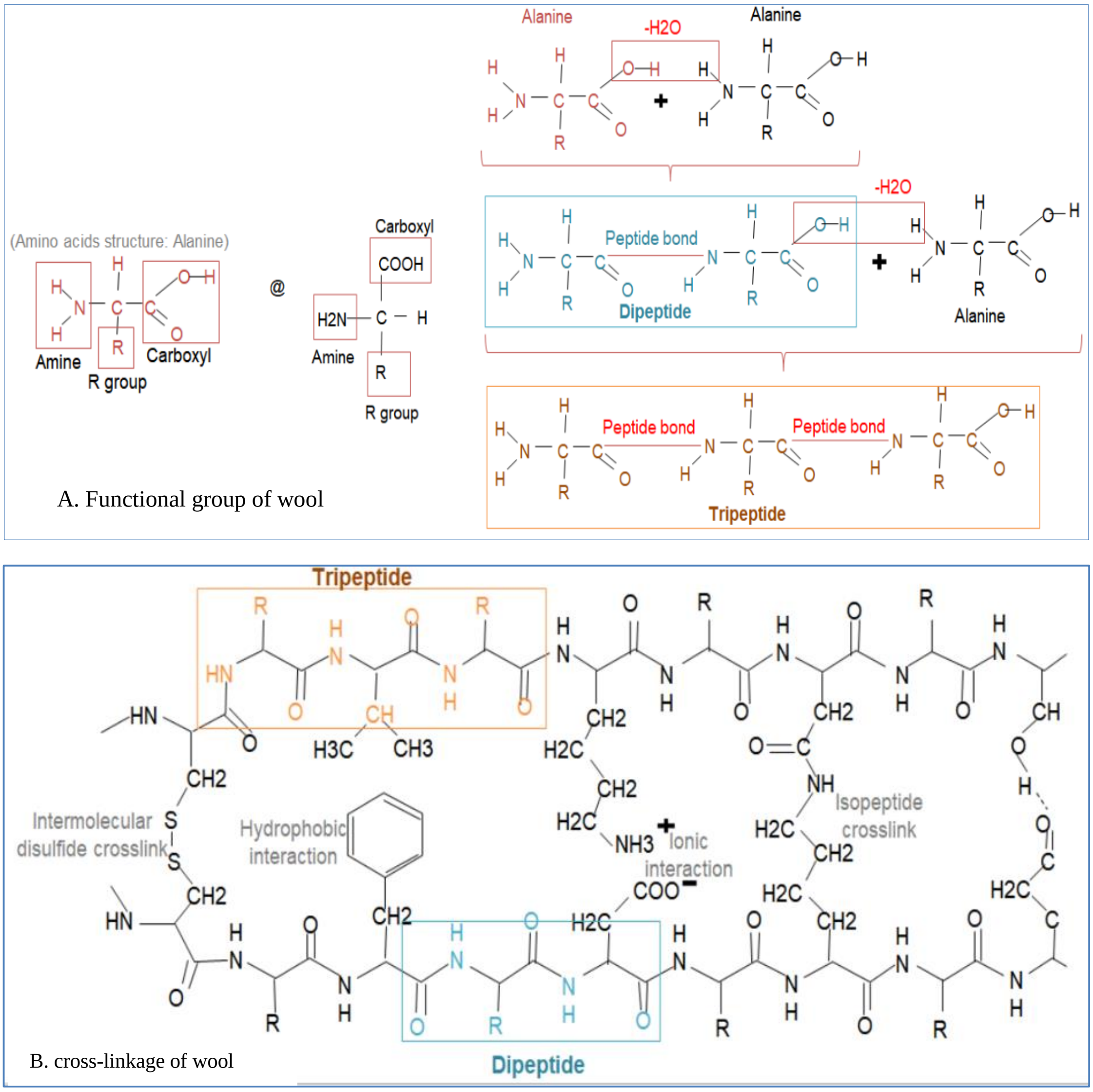

Figure 2.1. The chemical structure of wool A, B

Wool contains keratin, which is strengthened by disulfide bonds from sulfur-containing amino acids. The non-protein fraction consists of an outer lipid hydrophobic layer, which is known as lanolin or wool wax (Fiore et al., 2019; Sabatini et al., 2018). Because of its resistance to wear and tear due to its mechanical and chemical properties, wool fibre is used on flooring and car seats to reduce shock risks when they come into touch with grounded objects. (Bharath et al., 2016). Wool is a biological polymer characterized by the cuticle, cell membrane complex, and cortex (Duman and Küçük 2022). Ninety per cent of the fibre by weight is composed of the cortex. Wool is applied as a filler to produce mortar or plastic, which makes it a renewable resource (Fiore et al., 2019). Wool fibres are hydrophilic swells when it adsorbs moisture, resulting in an increase in weight and size, which also affects their mechanical and electrical properties (Memon et al., 2018). Sheep wool has a diameter in the range of 16-40 μm. However, unlike synthetic fibres, sheep wool has no specific thickness (Del- Rey et al., 2017) and contains 60 % animal protein fibres, 15 % moisture, 10 % fat, 10 % sheep sweat, 5 % impurities and is known to be highly hygroscopic (Zach et al., 2012). Fabrics are bulky compared with other textiles because they crimp and retain air, which makes them good insulators. They help retain warmth in cold weather. However, their insulation capability functions in both ways. For instance, the Bedouins and Tuaregs use clothes made of wool to keep the heat out, as it does not cling to their skin, allowing proper air circulation. Wool fibres are more resistant to tension compared with cotton and rayon; fibres can resist breakage even when bent 20,000 times, whereas cotton and rayon can break when bent just 3000 and 75 times, respectively (Yan 2016).

From the viewpoint of the morphology of wool, consists of ortho- para- and meso-cortexes, as well as covering scales on the fibre surface. They are composed of two types of cells the internal cells of the cortex and external cuticle cells that form a sheath around the fibre (Lakshmanan 2022). The ortho- and para cortexes are bilaterally arranged in fibre with the former on the outside and the para cortexes on the inside of wool curvature. The covering scales play an important role in fibre's chemical and physical properties, and in protecting the fibre from damage. Cuticle cells (or scales), which overlap as if tiles on a roof; make wool unique among textile fibres (Rippon and Evans 2020). The complex physical structure of cuticle cells is shown in Figure 2.2. An important function of cuticle cells is to anchor wool fibres in the skin of sheep. The exposed edge of each cuticle cell points from the fibre root towards the tip. This gives rise to a larger surface frictional value when a fibre is drawn in the against-scale direction than in the with-scale direction. The frictional difference helps to expel dirt and other contaminants from the fleece, but it is also responsible for wool's property of felting when agitated in water. This unique feature is not shared by any other textile fibre and enables fabrics with very dense structures to be produced, such as blankets, felts and overcoat materials. When felting is regarded as undesirable (for example in knitted garments that will be machine-washed), processes are available to remove the frictional difference and make wool shrink resistant. The fibre surface is also largely responsible for the natural softness of wool and its property as one of the smoothest textile fibres. Even after the natural wool grease is removed by scouring with a detergent, wool fibres are relatively difficult to wet compared with other textile materials. This natural water repellency makes wool fabrics 'shower-proof ' and able to resist water-based stains. The cortex of wool comprises approximately 90 % of the fibre. It consists of overlapping spindle-shaped cells cortical cells, shown schematically in Figure 2.2. Both the cuticle and cortical cells have highly complex substructures, the cell membrane complex (CMC) holds cortical cells together and separates them from cuticle cells. The CMC is a continuous region, containing relatively lightly cross-linked proteins and waxy lipids that extend throughout the whole fibre. Although it comprises only around 5 % of the total fibre mass, it plays an important role in the overall properties of wool. It is a region of relatively low mechanical strength in the fibre composite.

Wool fibres can be elongated by approximately 25 % - 30 % and still return to a natural shape. Fibres have a higher ignition point than cotton and some synthetics. Even when burnt, they do not melt or drip like synthetic materials (Hu et al., 2020). They have the self-extinguishing property when the flame source is extinguished. They generate fewer toxic gases when burned compared with synthetic materials, and they are often recommended for high-safety environments such as aircraft and trains (Millington and Rippon 2017). Wool fibres have the capability to protect from UV (ultraviolet) is much higher than that of cotton and synthetic materials, the presence of both crystalline and amorphous regions makes them one of the most advanced complex and composite structures in nature. Figure 2.2 shows the physical structure of sheep wool fibre. Sheep wool has a complex physical structure consisting of a membrane, cortical cell, cortex, microfibril, matrix, twisted molecular chain, and helical coil (Erdogan et al., 2020).

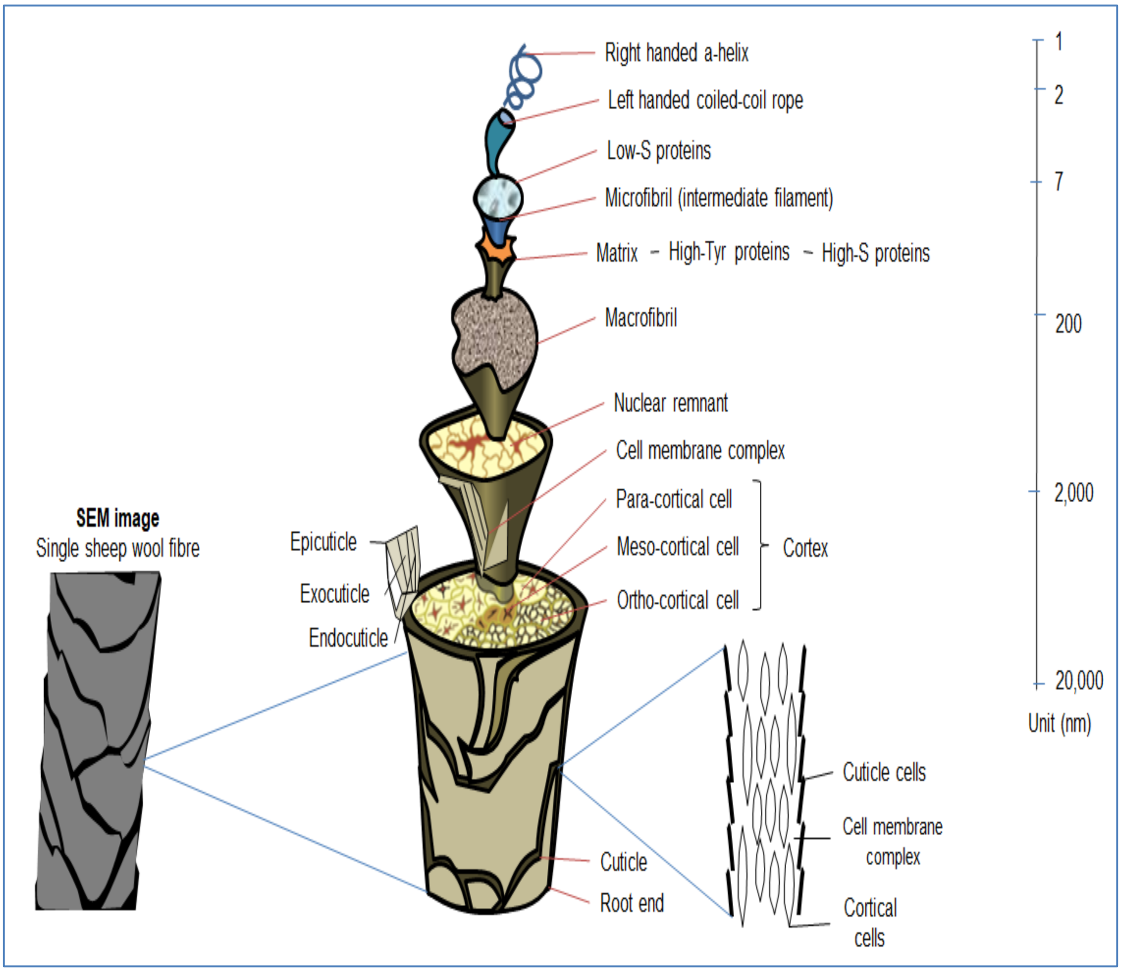

Figure 2.2 Physical structure of sheep wool

Further, sheep wool is coated with the waxy material, lanolin. Therefore, lanolin needs to be removed during the processing of sheep wool. Notably, each wool component has a specific composition (physical and chemical), which plays a significant role in the wool system. Changes in the physical structure due to activities such as unscaling and peeling off may affect the morphology of the wool fibre. Consequently, the cuticle cells might enhance the surface area by weakening the structural bonds within the fibre (Kuzmanova et al., 2018). The cortex cell is surrounded by cuticle scales, which are rich in cysteine, a sulphur-containing amino acid whose disulfide bond confers both chemical and mechanical stability to wool. Even though the cuticle scales are known for their hydrophobicity, the propensity of the cortex to absorb 30 % by weight in moisture indicates hygroscopic characteristics. A combination of these properties provides a unique wool surface (each component possesses desirable properties such as high moisture absorbency and chemical resistance). A wool fibre can be reverse-engineered into its major subcomponents by altering its chemical and physical structures (Caven and Bechtold, 2017).

2.1.2 Fourier transform infrared

The first developmental phase of Fourier transform infrared spectroscopy (FTIR) was initiated by Sir William Herschel who discovered infrared (IR) radiation in the year 1800 (Mayerhofer et al., 2021). Lord Rayleigh made the following significant discovery in the development of FTIR spectroscopy not long after Michelson built the interferometer in 1892. He discovered that it is possible to obtain a spectrum from an interferogram by computing the Fourier transform of the interference pattern.

FTIR spectrophotometry measures the functional group vibrations as a consequence of electromagnetic radiation with a sample to generate a spectrum with fingerprint properties (Yuliani et al., 2018). This technique offers qualitative and quantitative analyses of targeted samples including pharmaceutical products. There are two types of FTIR spectroscopy. Fourier transform near-infrared (FT-NIR) spectroscopy and Fourier transform mid-infrared (FT-MIR) spectroscopy (Masithoh et al., 2020). In FT-NIR spectroscopy, the absorption of the near-infrared range of the electromagnetic spectrum ranges from 600 to 4000 cm^{1}. While FT-MIR spectroscopy is a type of FTIR spectroscopy which uses the mid-infrared wavelength ranging from 4000 to 400 cm^{1} in FTIR to obtain a chemical profile of interest by identifying the fundamental vibrational and rotational stretching of molecular bonds.

It has been reported that FTIR spectroscopy has been applied in many studies to analyse the effect of technologies that have been used on wool fibre as treatment or modification. Wang, et al. (2008) utilized FTIR to determine the functional groups of treated wool with a scanning range from 600 to 4000 $cm^{-1.}$. Long et al. (2013) utilized FTIR with the traditional KBr pellet sampling method. The transmittance of the infrared was recorded from 400.0 to 4000.0 cm^{-1} at a resolution of 2.0 cm^{-1} for infrared spectra.

Wang et al. (2012) analyzed wool on FTIR with attenuated total reflectance and a scanning range from 600 to 4000 cm^{-1} to check amides in wool fibre treated with H2O2. Zhang et al. (2016) Fourier transform infrared (FTIR) spectrophotometer with (ATR) was used to characterize the surface chemical structure of wool to check the crosslink and various functional groups and bonds such as peptide bonds (–CONH–). Jiang et al. (2022) reported that the structure of regenerated keratin from wool was analyzed using the amide I, II, and III bands from second-order derivation Fourier transform infrared spectroscopy.

2.1.3 Utilization of sheep wool fibre

Sheep wool has received considerable attention for use as an apparel fibre and interior fabric because of its good technical properties. The utilization of sheep wool in various industrial applications is presented in Table 2.2. Sheep wool is mainly utilised as a textile material for producing garments, warm clothes, carpets, and curtains. However, the technical application of wool has attracted scant attention compared with that of other synthetic fibres. Successful technical utilization of wool fibre requires a high-value application that can exploit its natural properties or modify it to achieve a particular target performance. The sustainability and biodegradability of wool fibre make it attractive for use as an ideal material for various technical applications. Sheep wool, as a natural fibre, is utilized in civil engineering for the thermal and acoustic insulation of facades and roofs or as reinforcement in composites with polymeric, earthen, or cementations matrices (Zagarella et al., 2014). As well as for heavy metals in wastewater (Hanzlikova 2018).

There are two categories of sheep wool insulation products in the building market. They are soft mats composed of 100 % sheep wool having a thickness of 4-6 cm, mainly used for the insulation of pitched roofs, and semi-rigid panels composed of 70 % - 80 % sheep wool and 20 % - 30 % polyester having a thickness of 5-12 cm (Zach et al., 2012). They are applicable on walls because of the stiffness achieved through the partial fusion of polyester fibres. The insolubility and the resistance of sheep wool are attributed to the disulfide bridges formed in the sheep wool structure by cysteine (Alston et al., 2019). Besides, sheep wool contains resilience and excellent elongation characteristics. Murugappan and Muthadhi (2022) utilized the sheep wool fibre as a reinforcement in soil-alginate composite to enhance bonding within soil particles. Zagarella et al. 2014 isolated a bio composite concrete using lime and sheep wool fibres (20-40 wt %) by following granulometric and a layered envelope system. It was found that the thermal conductivity of the biocomposite concrete was increased with the percentage of sheep wool content. The study reported that sheep wool has the potential to be used in the energy efficiency of a building. Sheep wool shows a sufficiently good acoustical performance for its use as a noise barrier, as a sound absorber in acoustic rooms, or as a vibration insulator. The absorption coefficient of sheep wool panels was determined to be 0.84 at 2000 Hz. (Borlea et al., 2020; Korjenic et al., 2015). Sheep wool panels could be utilized to improve the cavity absorber or vibration insulator maximum of 10 dB and to improve the transmission loss on plasterboard walls by a maximum of 6 dB (Borlea et al. 2020). Wool fibres are a self-extinguishing material and it burns slowly with a slight sputtering in the presence of a flame (Zach et al., 2012). Besides, wool fibres are more hygroscopic and they can absorb moisture in vapour form up to 20-35 wt % without causing a damp feeling (Korjenic et al., 2015). The moisture absorption capability of sheep wool fibres is higher than that of glass wool fibre (0.2%) or polystyrene foam (0.03 % -0.1 %) (Borlea et al., 2020).

Table 2.2 Utilization of sheep wool in various industrial applications

Applications	**Role**	**References**
Textile materials	To produce warm clothes, carpet and curtains	Laitala *et al*. (2018)
Civil engineering materials	Thermal and acoustic insulations of facade and roof insulations	Korjenic *et al.*(2015)
Reinforcement material	Enhance mechanical and thermal properties of polymeric, earthen or cementitious matrices	Grădinaru *et al.*(2016)
Acoustical material	Adsorbed sound, noise barrier and as a vibration insulator	Borlea *et al*. (2020) Del *et al.*(2017)
Catalyst	Enhance mass transfer, physical and chemical stability	McNeil *et al* . (2017)
Geotextile	Steep slopes protection of soil from erosion	Broda *et al*. (2018)
Soil nutrient	Cube nutrients for soil fertilization	Abdallah *et al.* (2019)

2.1.4 Impurities of sheep wool fibre

Raw sheep wool is a highly contaminated natural fibre because it contains high levels of impurities, including wax, grease, suint, mineral soil, dust, and vegetable matter (VM) (Millington and Rippon 2017). On average, each tonne of greasy wool contains 150 kg of wool grease, 40 kg of dirt, 150 kg of dirt, 20 kg of vegetable matter and only 640 kg of wool fibre. Therefore, before being used in textile material, raw wool involves a long process of water-based treatment involving washing and scouring (Alebeid et al., 2020), carbonization, and bleaching (El-Sayed et al., 2022). Cleaning is the most important step in the processing of wool, which removes contaminants that may hinder its manufacture in textiles or affect the aesthetics of the finished textile (Pan et al., 2018).

2.1.4(a) Wool grease

Wool grease or wax is a soft, yellow, waxy substance that is excreted by sebaceous glands located in the sheep's skin and provides waterproof properties and protection for wool fibre (Albanell et al., 2018). Chemically, wool wax is a complex mixture of different properties that can be determined by different extraction methods (Albanell et al., 2018). Soxhlet extraction has been applied using dichloromethane as a solvent to extract substances as well as supercritical carbon dioxide, using toluene as a co-solvent applied to extract grease (Valverde and Recasens 2019). Wool grease recovered from wool scouring liquors or by solvent-extraction from greasy wool usually contains pesticide residues and dirt (Zakaria El-Sayed et al., 2018).

Dirt and wool grease recovery are integrated with the wool cleaning process, as the wool fibre contains about 2% of internal lipids and external lipids. External lipid is wool grease that is removed during the scouring process (Koivu-Tikkanen 2022). Conventional aqueous wool grease cleaning consumes time, water and detergents to be removed (Li et al., 2014). Wool grease has different fractionated and refined forms known as lanolin (Millington and Rippon, 2017; Road, 2017). Internal lipids are produced from membranes that surround living cells. At the end of the keratinisation process, lipids are trapped in different locations within the compacted mass of the wool proteins, where the fatty acids alpha-hydroxy and 18-methyl eicosanoic acid (18-MEA) are covalently bound to the surface of the wool fibre.

2.1.4(b) Suint

Suint is produced from sheep-dried sweat in the form of proteins, amino acids, and potassium salts of short-chain fatty acids. Suint helps the cleaning process because it has detergent properties (Jose 2021). Cations that are present in potassium salts of organic acids account for 25–27 % of the dry suint weight. The amount of suint present in wool depends on the sheep type, location and climate of the breed, where crossbreed sheep tend to have more suint than merino (Cottle and Baxter 2015). Wool suint is a water-extractable material likely to originate from the animal's sweat glands and contains small amounts of succinic acid, hippuric acid, lactic acid and urea (Madara and Namango, 2014). Suint and other impurities are responsible for non-white wool colours, thus subjecting wool to a scouring cycle, to attain the original whiteness (Zhang et al., 2016).

2.1.4(c) Vegetable matter

Raw wool is contaminated with different types of vegetable matter VM such as burrs, seeds, twigs, and straw, in different sizes, depending on the place of grazing and the environment in which the sheep are raised. VM should be removed before the tinting operation, which would otherwise appear to be defective (Hassan and Shao, 2015). Vegetable matter is major contamination of wool fibre and requires a carbonising process using an acidic solution of embrittling vegetable matter, adding costs to the classification and processing of wool (Atkinson, 2014).

2.1.4(d) Biological hazard of sheep wool fibre

Organisms such as gram-positive bacilli, gram-negative *pleomorphic rods, coccobacilli, and cocci* are found in the fleece of sheep, although their counts may vary. The common resident floras in the fleece are *Pseudomonas, Proteus, Staphylococcus, and Bacillus. Mycotic* dermatitis or “lumpy wool” is caused by the bacterium *Dermatophilus congolensis* (Saleh et al., 2019). The action of *Mycobacterium avium ss. Paratuberculosis* can result in wool breakage. Derzelle et al., (2016) detected *Bacillus anthrax* spores in a wool fibre-processing factory. *B. anthrax* is an infectious human pathogen. Thus, the presence of microorganisms in sheep wool must be considered before selecting technology for sheep wool processing to ensure complete inactivation of the pathogen to protect the wool fibre from damage and prevent exposure to the infectious pathogen. The biodeterioration of wool leads to odours, staining, or discolouration, a decrease in strength, and stains appear due to the action of exo pigments which are secreted by microbial cells and diffuse into the wool (Harmsen et al., 2021). The most frequently occurring pigment is melamine, which is produced in the mycelium of fungi, and gives the fibre a dark shade. Depending on the group of microorganisms present on the wool, the colour can vary from creamy to black and yellow discolouration (Mowafi et al., 1016).

Microbial growth could lead to fibre degradation, changes in structure, cracking, and fragmentation, thereby causing a reduction in the degree of polymerization, a decrease in tensile strength and elasticity of the wool as well as a decrease in crystallinity index (Harmsen et al., 2021). Table 2.3 show the type of bacteria in raw sheep wool fibre. The signs of biodeterioration of wool are pitting corrosion, cracking, fragmentation, staining, and total decomposition (Guiamet et al., 2016). Yellowness increase of wool fibres affected by microorganisms testifies occurrence of additional colouring centres. To elucidate the mechanism of microorganisms' action on the wool fibres that leads to significant changes in properties and structure of the material, the changes in the amino acid composition of wool keratine proteins being the nutrition source of microorganisms damaging