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Chapter

Ecological Applications of Enzymes in Plants Based Textile Dyeing

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Abstract

Biotechnology has a foremost role in the textile industry by enhancing ecofriendly, cost-effective, and energy-efficient manufacturing processes. The use of enzymatic biotechnology is one of the sustainable newly developed state-of-the-art processes for textile processing. To reduce the use of toxic and hazardous chemicals, enzymes have been proposed as one of the finest promising alternatives. Many enzymes have been used widely in textile processes such as lipase, laccase, pectinase, cellulase, catalase, amylase, and protease. The enzymatic use in the textile industry is very promising because they produce top-class goods, and give way to the reduction of water, time, and energy. The increasing demand for natural dyes especially with the incorporation of enzymes makes process more sustainable and eco-friendlier to suppress the toxicity of synthetic dyes. In the first part of the chapter, particular attention has been given to the source and extraction of natural dyes. In the second part of the chapter, different enzymes and their possible roles in the textile industry have been discussed. It is expected that this chapter will provide an innovative direction to the academic researchers, the community of textile and traders as well as artisans who are working in the area of biotechnological applications for the betterment of textile processing.

Keywords: bio-processing, bio-bleaching, bio-desizing, bio-mercerization, sustainable coloration

1. Introduction

The textile industry is one of the fundamental requirements of human beings which also contributes significantly to the growing economies of many developing nations. The consumption of textile materials is expanding as a result of a growing population and increased per capita textile demand [1, 2]. However, the traditional approach of textile wet processing needs several processes by using dangerous chemicals, high salt concentrations, and a lot of resource consumption before it produces a completed fabric. This approach is criticized for its negative environmental effects due to its toxicity [3]. Implementation of enzymes in textile wet processing is guided by friendly environmental awareness. Enzymes are used in the textile sector, which appears to establish a perfect balance between commercial requirements and the creation of environmentally beneficial products [4]. Enzymatic procedures are

extensively utilized in the chemical processing of textiles to reduce environmental risks and prevent the excessive use of toxic chemicals. Enzyme biotechnology is a sustainable and effective method that has been utilized in many manufacturing procedures as a favorable alternative to synthetic catalysis with benefits in terms of productivity and sustainability as well as creating top-quality textile fabrics [5]. Enzymatic catalysis is an effective instrument that can be used in the industrial setting and allows for the resolution of most environmental sustainability-related problems, particularly those involving the usage of dangerous chemicals. Enzyme utilization has the potential to significantly lessen the negative effects of industrial processes on the environment [6].

Enzymes are natural and biodegradable proteins that are frequently employed in industries to replace dangerous chemicals since they operate under compassionate circumstances and are durable, safe, and disposable. Enzymes have important roles in reducing contaminants, bio-finishing to improve esthetics, removing fabric fur from the surface, bio-bleaching of cotton to give it an excellent matte texture, and removing leftover hydrogen peroxide after bleaching [7]. Oxidoreductases and hydrolases are two major enzymes used in the textile industry. The hydrolase class comprising catalase, amylase, pectinase, lipase, and cellulase are utilized in desizing, biopolishing, bioscouring, and bleaching processes in textile industries [8]. The oxidoreductase class of enzymes comprises trans-glutaminases, which are used to modify the properties of synthetic fibers whereas laccases are used to decolorize the fabrics. Since synthetic dyes have demonstrated harmful qualities over natural dyes, researchers have drawn towards natural dyes due to their eco-friendliness [9]. The other beneficial aspects of natural dyes over artificial dyes are, the natural dyes are disposable, and the byproducts produced during the dyeing process are less harmful to the environment [10]. Moreover, natural dyes have a calming effect and provide beautiful colors and adorable shades, which make textiles appealing to consumers [11]. The majority of dye-producing plants also exhibit anti-oxidant properties, ultraviolet protection, and antimicrobial potential. Precisely, natural dyes give textiles some additional finishing qualities in addition to color [12].

2. Resources of natural dyes

Various natural sources have been used to create natural colorants, these have been categorized as plants, animals, minerals, and microorganisms [13]. Natural Dye Research and Development Project, in Turkish known as DOBAG, was launched in Turkey in 1981 in collaboration with Polytechnic University, Istanbul, and was very successful in reviving the forgotten craft of naturally colored textiles [14]. Several natural dyeing supplies have now been discovered because of research initiatives by individuals and organizations as well as information exchange at several conventions, festivals, seminars, and articles [15]. There is now a huge amount of knowledge concerning various sources of natural dyes in the literature.

2.1 Plant origin

Several natural colorants have originated from plants in history like an alkane, annatto, madder, chamomile, sappars, coreopsis, etc. Natural dye supplies include a variety of plant components such as leaves, roots, stump branches, core wood, wood shavings, bark, fruits, flowers, hulls, and husks [16]. For example, leaves of *Indigofera*

tinctoria are used to make the well-known natural blue color indigo. Some plant-based dyes are used for other purposes as well, such as food coloring and traditional medicine, and as a result, there is a commercial supply chain for these dyes [17]. The commercial availability of natural dyes has expanded due to a resurgence in interest in them.

2.1.1 Blue dye

The king of natural dyes, Indigo, is the only significant natural dye in blue color which could be extracted from the *I. tinctoria* plant leaves. Since, indigo has been utilized for producing a blue hue since ancient times and is currently the most popular denim material [17]. More precisely, a pale-yellow chemical known as Indican serves as the coloring component, and it is found in the leaves of indigo plants. Interestingly, indigo plants in approximately one-acre area can produce roughly 5000 kg of leaves, which can be converted into 50 kg of pure natural indigo powder. Several plants, besides the *Indigofera* species, may be utilized to make indigo dye such as woad, a plant that naturally produces indigo in Europe. In addition, *Wrightia tinctoria* and Dyer's knotweed (*Polygonum tinctorium*) are two other plants that have historically been used to make indigo [18]. Unfortunately, natural indigo got declined after the production of synthetic indigo in 1987 which attained more preference over natural one.

2.1.2 Red dye

Red natural dyes can be found in a variety of plant sources. Natural red dyes called madder are made from plants of *Rubia* plant (Hosseinnezhad et al., 2021): "queen of natural dyes". Between 3 and 5 tonnes of roots and 150–200 kg of dye are produced per hectare by the 3-year-old plant [19]. In addition to the roots, the plant also has dye in the stems and other sections like dried root chips or stem pieces after soaking in cold water before being used to extract the dye. Being a mordant dye, it creates insoluble multiplexes with the metal ions existing on mordanted fabric to generate vivid colors. Pink and red hues are frequently produced with alum where's the alum and iron together yield purple hues. A variety of red could also be generated by mixing other mordants with the main metallic salt, alum [20]. The sappan wood often referred to as "Patang," is a tiny tree that produces a red dye often found in India, Malaysia, and Philippines. *Caesalpinia echinate*, the Brazil wood named after the word Braza which means flaming like fire due to the vivid red color of its wood, also contains the same dye.

Another red pigment-producing tree is *Morinda citrifolia* which is found in Sri Lanka and India. The 3–4 years old tree provides a good quantity of coloring matter from its bark and roots. A variety of colors, including chocolate and purple, can be produced by using different mordants [21]. Additionally, an annual herb known as safflower is believed to have come from Afghanistan and has been used to extract the dye. This herb developed an astonishing cheery red shade on silk and cotton. Dried safflower florets are repeatedly washed in acidic water to get rid of all the yellow color water-soluble material before the removal of the dye [22].

2.1.3 Yellow dyes

A well-known source of yellow dye is turmeric which is extracted from turmeric rhizomes, whether they are fresh or dried, and are used to make the color on wool,

silk, and cotton. The coloring material present in turmeric is curcumin, which belongs to the diarylmethane class [23]. To increase the fastness qualities and range of adorable shades, different mordants can be used. Saffron is also a good source of vintage yellow dye from the *Iridaceae* family that is made up of the desiccated stigmas of the *Crocus sativus* plant. By boiling flower stigmas in water, the dye is released and produces a vivid yellow hue on cotton, silk, and wool [24]. *Berberis aristata* commonly known as the barberry plant is also a well-reputed source of yellow dye. The roots, bark, and stems of the barberry plant are used to extract the yellow dye and can be used directly to color silk and wool with average lightfastness and good washing fastness properties [25].

Another source, pomegranate (*Punica granatum*) fruit rinds, which are high in tannin, are used for mordanting. It is also used in conjunction with turmeric to increase the dyed fabrics' light resistance. Myrobolan (*Terminalia chebula*) fruits also have high tannin content. The dried fruit also contains a natural colorant which develops a vivid yellow color for all textile fabrics. Moreover, myrobolan can also be employed as a natural mordant for natural dyes fixing on textile fabrics.

Marigold (*Tagetes spp.*) is also frequently employed to create garlands and floral accents due to its vivid yellow flowers. It comes in a variety of colors, such as yellow, golden yellow, orange, and others [26]. The primary yellow coloring agents are Quercetagetol, a flavonol together with two of its glycosides, and lutein which exerts good fastness characteristics of wool and silk. This dye can be used to quickly create colors on cotton when combined with mordants. Besides, the flame of the forest tree named *Butea monosperma* color all natural fibers. By using correct mordants, the dye extracted from the bright orange flowers of this tree develops vivid yellow, brown, and orange colors. *Mallotus sphillipensis*'s dried fruit capsules, known as Kamala, produce a reddish-orange powder. A vibrant yellow-orange and yellow-golden hue developed onto silk and wool [27]. Similarly, the outside layer of onions, *Allium cepa*, which is typically discarded like trash, be able to be utilized to extract natural yellow colorant. The chemical makeup of the dye is a flavonoid, and it gives wool and silk vibrant colors. A suitable mordant can be used to dye cotton with average washing and light resistance properties.

2.1.4 Black and Brown dye

Oak galls are utilized for mordanting because they are high in tannin and also be employed to achieve a brown hue. Catechu or cutch, which is made from the heartwood of *Acacia catechu* employed to dye cotton, wool, and silk [28]. It has a lot of tannins as well and can be dyed black using iron mordant. By iron mordanting, many yellow and red dyes can also be turned black. Likewise, the heartwood of the *Haematoxylon campechianum* tree, which is located in the West Indies and Mexico, was used to extract the famous logwood black hue, which is quite sharp and has excellent fastness capabilities [29].

Some other research reports the valorization of some natural wastes such as olive wastewater by its use as a possible dye bath for dyeing textile fibers. During olive oil extraction, dark brown to black effluent was produced which was characterized by a high organic load including polyphenols and tannins. Darker brown shades were developed with generally good fastness which demonstrates that protein fibers, cotton, and other synthetic fibers possess a high affinity to this aqueous extract.

3. Natural dyes production and extraction techniques

Natural colors are typically derived from diverse plant parts, distinct from synthetic dyes, which are created from chemical predecessors. These dye-bearing materials typically only have a 0.5–5% dye content and these plant ingredients cannot be used directly for the dyeing process [30]. Additionally, a lot of plant materials, including flowers and fruits, are seasonal and contain a lot of water, making it impossible to store them in their natural state. Therefore, these are put through some processing procedures to make them appropriate for dyeing in textile industry needs and to make them accessible all around the year [31]. To lower their contents of water to about 10–15% or less, collected constituents of plants are desiccated at first, either in the shadow or at a low temperature of 40–50°C in a hot air dryer. To minimize particle size and improve dye extraction, the dried material is subsequently ground into a powder [11]. In most situations, these powdered and dried components can be kept for at least a year in sealed bags or containers and utilized for dyeing whenever necessary. To create pure dye powders, the dye must first be extracted from materials containing dye. Due to the use of several types of machinery and higher energy consumption throughout various processing activities, these refined versions are expensive [32]. Additionally, because dye extraction happens simultaneously with dyeing, its effectiveness is lower when compared to using powdered crude dye-bearing material [33].

Since the amount of coloring matter or dye present in natural dye-bearing materials is relatively low, along with other plant and animal compounds like water-insoluble fibers, carbohydrates, protein, chlorophyll, and tannins, among others, extraction is a crucial step both in the production of purified natural dyes as well as in the processing of raw dye-bearing materials [34]. Before using an extraction procedure, the type and solubility characteristics of the coloring components must be studied [35]. Many conventional and non-conventional techniques for extracting colored ingredients are described in the following:

3.1 Conventional extraction methods

3.1.1 Aqueous extraction

Plants and other materials were previously utilized to extract colors using aqueous extraction. To increase the effectiveness of the extraction process, the material which is comprising dyes is at first fragmented into tiny bits or powdered before being sieved [36]. It is then immersed in water for a long period typically overnight in earthen, wooden, or metal vessels (ideally copper or stainless steel) to release the cell structure [37]. The dye solution is then extracted and filtered to eliminate any remaining non-dye plant material. To get rid of the non-dye parts, the boiling and filtering operation is repeated, and centrifuges are typically used to separate leftover material. The elimination of tiny plant materials and improved solubility of the purified natural dye can both be achieved by using trickling filters [29].

The extract generated by this procedure may be employed to the constituents of the textile with ease because many dyeing processes are conducted in aqueous solutions. This extraction method has several drawbacks, including a lengthy extraction period, a significant amount of water needed, the usage of elevated temperatures, and a negligible yield of dye because merely the water-soluble

color constituents are removed, even though numerous dyes comprise little water solubility [38]. As well as the dye, other water-soluble materials are also extracted, these materials may need to be eliminated if the extract is to be condensed and turned into a fine powder. Boiling temperature reduces the yield of heat-sensitive dye compounds therefore a low temperature should be optimized for the extraction in these circumstances [39].

3.1.2 Acid and alkali extraction process

Many colors exist as glycosides, they can be removed using diluted acidic or alkaline solutions. Greater extraction and elevated yield of coloring constituents occur from the hydrolysis of glycosides being facilitated by the addition of acid or alkali [40]. Tesu (*B. monosperma*) flower petals are utilized to extract the dye by an acid hydrolysis procedure. To avoid oxidative degradation, some flavone dyes are extracted using acidified water [41].

Alkali and alkaline extraction are appropriate for dyes with phenolic groups as it increases the yield of the color. This method is also used to extract rose-red colorant from petals safflower. One drawback of this procedure is that few dyeing components might be degraded in alkaline environments because some natural dyes are pH-sensitive [42]. The colorants which exist naturally are typically a combination of many biochemical components, altering the pH of the extraction medium by complementing alkali or acid able to cause the extraction of various colorant components, which can result in a range of color outcomes and colorfastness characteristics [43]. To determine the ideal optimization for dye extraction, numerous scholars have investigated natural dye extraction under numerous conditions of pH and equated the fastness and shade attributes of tinted fabrics [44].

3.1.3 Solvent extraction

Depending on their nature, natural coloring substances can also be extracted by utilizing natural solvents like chloroform, methanol, petroleum ether, acetone, ethanol, or mixtures of solvents like ethanol and methanol, water, alcohol, etc. [45]. Both water-soluble and water-insoluble materials can be extracted from plant resources using the water/alcohol extraction method. As a result of the ability to extract a greater variety of chemicals and coloring ingredients than the aqueous approach [46]. Alcoholic solvents may also be used with an acid or alkali to aid in the hydrolysis of glycosides and the release of coloring components. The ability to readily remove and reuse solvents makes it simpler to purify extracted colors. Because extraction is done at a lower temperature, there is less danger of deterioration [47]. The method's drawbacks include the greenhouse effect of the poisonous leftover solvents and requiring an aqueous solution for the dyeing process because the extracted substance is not easily soluble in water. Problems can also result from the co-extraction of compounds like waxy polymers and chlorophylls [41].

3.2 Non-conventional extraction methods

3.2.1 Ultrasonic and microwave extraction

These are ultrasound and microwave-assisted extraction techniques, in which the use of ultrasound or microwaves improves extraction efficiency and reduces the amount of solvent needed as well as the extraction time [48]. Ultrasound induces the

formation of tiny bubbles or cavitation in the liquid when the plant components are preserved with water or some additional solvent. This results in the cavity collapsing or the bubbles bursting, which raises the temperature and pressure [49]. The extraction efficiency is quickly increased when extremely high temperatures and pressures are created. Many studies have lately reported using this extraction approach as the quest for novel dye sources and efforts to improve dye extraction continue [50].

In microwave extraction, the natural sources are processed in the existence of microwave energy sources with the least amount of solvent possible. The procedures are accelerated by the microwave, allowing for faster and more effective extraction [51]. The decrease in temperature of extraction, utilization of solvent, and duration show consequences in less energy utilization. Both extractions ultrasound and microwave may be regarded as green methods [52].

3.2.2 *Fermentation*

Fermentation accelerates the process of extraction by using the enzymes which are generated by microbes found in the environment or natural resources. The most typical instance of this kind of extraction is indigo extraction in which newly collected indigo twigs and leaves are immersed in warm water (about 32°C) [38]. As fermentation progresses, the indimulsin enzyme, which is also present in the leaves, converts the colorful indigo-containing glucoside indican into glucose and indoxyl. In about 10–15 hours, fermentation is finished, and the indoxyl-containing yellow fluid is formally transferred to whipping tanks where indoxyl is became oxidized through the air and turned into the blue color, insoluble indigotin that sinks to the bottom [53]. It is collected, cleaned, and then pressed to remove the extra water. Other colorants, such as annatto, can also be extracted using this method. Except for not requiring high temperatures, the fermentation process is comparable to aqueous extraction [28]. The bacteria naturally break down the chemicals that bind coloring materials. The drawbacks of this method include a lengthy extraction process, the requirement to extract pigments right away after harvesting, a bad odor brought on by microbial activity, and others [54].

3.2.3 *Supercritical fluid extraction*

Supercritical fluid extraction is a developing field in the purification and extraction of natural products. Above its critical temperature and pressure, a gas behaves as a supercritical fluid. A fluid like this has physical characteristics that fall midway between a liquid and a gas [55]. They have substantially lower surface tension than liquids, which allows them to spread out along a surface more quickly [56]. As a result of their low viscosity and excellent diffusivity, they interact with the substrate more effectively. The ability to dissolve the matter in every solvent is increased at elevated pressures and temperatures, and these circumstances are required to sustain a gas in supercritical conditions.

Carbon dioxide (CO₂) supercritical fluid extraction is a viable substitute for solvent extraction since it is inexpensive, simple to use, non-toxic, and residue-free. CO₂ supercritical extractions normally take place between 32 and 49°C between 1070 and 3500 psi of pressure [57]. The procedure has acquired popularity in the extraction of purely natural ingredients for culinary and medicinal uses because the extract is off-loaded from leftover traces of solvent, and heavy-weight metals, and is bright colored because of the lack of polar polymerizing chemicals. The method's drawbacks include expensive equipment costs and polar chemical extraction [58].

3.2.4 Enzymatic extraction

Commercially accessible enzymes like pectinase, cellulase, and amylase have been utilized by certain scholars to loosen the nearby component, allowing the extraction of dye molecules under more benign situations [59]. This is because plant tissues comprise cellulose, starches, and pectins as fixing components. This method might be useful for getting dye out of tough plant components like bark, roots, and the like [60].

4. Microbial-origin enzymes in textile industry

The use of enzymes in the textile industry is an example of white/industrial biotechnology, which allows the development of environmentally friendly technologies in fiber processing and strategies to improve the final product quality [61]. The enzymes in the textile industry is an example of white/industrial biotechnology, which allows the development of environmentally friendly technologies in fiber processing and strategies to improve the final product quality. The enzymes utilization is an illustration of white/modern biotechnology that improve the final product quality and permits the expansion of technologies that are approachable to the environment [62].

Various microbial enzymes are utilized in the clothing industry at various stages on behalf of finishing and waste degradation purposes. Because of its less harmful technology and extremely low waste production, the enzyme in textiles is responsible for a big profit worldwide [63]. Amylases are frequently utilized for the desizing process in the preliminary finishing area, and cellulases are frequently used for softening, bio-stoning, and lowering the pilling tendency for cotton items in the finishing area [64]. Microorganisms are employed for enzyme manufacturing because they have a rapid capacity to adapt to any condition and can create a wide variety of enzymes. *Bacillus amyloliquefaciens*-derived α -amylase worked at pH 6.5 and 60°C for one hour with 100% desizing efficiency [65]. Amylase extracted from *Aspergillus niger* and *Aspergillus flavus* has increased desizing efficiency (*A. niger* 96%, *A. flavus* 90%), and its absorbency and controllable impurities have significantly improved [66]. Chitosan reduces the amount of enzyme needed by two-thirds while improving the desizing effect when combined with mesophilic amylase is thermally stable at high-temperature desizing [67]. A thermostable cellulase extracted from *Talaromyces emersonii* is used to treat jute-based fabrics and show greater brightness, handling, and enduring softness. Due to flavonoid oxidation, laccase from *Trametes hirsute* with mediator improves cotton's whiteness [10]. To provide whiteness, the complex enzymes Laccase and Peroxidase effectively break down and eliminate lignin from flax fabrics [68]. Another key benefit of adopting microbial enzymes is that the microorganisms are easily biotechnologically altered to produce more enzymes.

The two primary classes of enzymes utilized in the pre-treatment of cotton are oxidoreductase and hydrolase. Pectinase, a crucial enzyme extracted from the widespread bacterium *Bacillus subtilis*, is utilized to increase the scouring impact of cotton fibers whereas for the processing of cotton, catalase from *Aspergillus flavus* is utilized. *Bacillus* appears to be a relatively widespread microbe that produces a variety of enzymes that are extremely useful in the finishing process for textiles. Applications for enzymes include the fading of both denim and non-denim, bio-scouring, bio-polishing, finishing wool, removing peroxide, decolorizing dyestuff, etc. [64] (**Table 1**).

Enzyme	Source	Use in textile	References
Cellulase	<i>Bacillus sp.</i> , <i>Streptomyces albaduncus</i> , <i>Aspergillus oryzae</i> , <i>Trichoderma reesei</i> , <i>Chaetomium globosum</i> , <i>Hypocrea jecorina</i> , <i>Trichoderma viride</i> G, <i>Aspergillus nidulans</i> AJ SU04	Bio-stoning, bio-polishing, and softening of denim	[69]
Catalase	<i>Micrococcus luteus</i> , <i>Bacillus sp.</i> , <i>Bacillus cereus</i> , <i>Flavobacterium sp.</i> , <i>Bacillus pumilus</i> .	Used after bleaching for cotton processing, Biostone washing, Bio polishing of cotton fabrics	[70]
Amylase	<i>Bacillus sp.</i> , <i>S. albaduncus</i> , <i>S. albaduncus</i> , <i>Bacillus sp.</i> SI-136, <i>Bacillus sp.</i> SI-136, <i>B. cereus</i> , <i>Aspergillus niger</i> , <i>Aspergillus flavus</i> , <i>Aspergillus niger</i> SH-2, <i>Penenzim HSE</i> , <i>Bacillus subtilis</i> MTCC 121, <i>Aspergillus tamari</i> , <i>Aspergillus tamari</i> .	Remove starchy layer, De-sizing of cotton fabrics	[71]
Protease	<i>Bacillus licheniformis</i> , <i>Arthrobacter</i> , <i>Streptomyces</i> , <i>Flavobacterium sp.</i> , <i>Bacillus sp.</i>	Prevent decolonization of denim, antifelting finishing treatment on wool and silk fabrics	[72]
Pectinase	<i>B. subtilis</i> , <i>Paecilomyces variotii</i> , <i>B. pumilus</i> AJK, <i>Streptomyces griseus</i> , <i>Candida</i> .	Hydrolysis of pectin in cotton fiber preparation, scouring of cotton	[73]
Lipase	<i>Aspergillus niger</i> , <i>Candida cylindracea</i> , <i>Candida rugose</i> , <i>Streptomyces acrimycini</i> NGP 1, <i>S. albogriseolus</i> NGP 2, <i>S. variabilis</i> NGP, <i>B. licheniformis</i> , <i>Pseudomonas fluorescens</i> , <i>Bacillus sonorensis</i> , <i>Thermomyces lanuginosus</i> .	Modification of Polyester fabrics, Bio-scouring of cotton fabrics, Surface modification of Polyethylenetere-phthalate fibers	[74]

Table 1.
 Microorganism-based enzymes and their role in textile.

5. Enzymes in textile

Due to its nontoxic and eco-friendly qualities, enzyme utilization in the textile sector is increasingly getting attention around the world. They also have the advantage of being able to work on specified substrates [75]. Some of the main enzymes used in processing textiles are discussed in the following.

5.1 Amylases

The most widely utilized enzyme in the clothing industry is amylase which works by breaking the starch molecules to make various compounds, for instance, dextrans and ever-smaller polymers made of glucose units [76]. The two types of starch-hydrolyzing enzymes, α -amylases, and β -amylases are categorized based on the kind of sugars they create. Various fungi, yeasts, and bacteria produce amylases, however the enzymes most frequently employed in the industry are from filamentous fungi and bacteria. Most fungi and bacteria's α -amylases are constant throughout a varied pH scale, from 4 to 11 [77]. Typically, it serves as a desizing agent by eliminating the sizing components (which are starch and its derivatives) without harming the fabric. The α -amylases enzyme hydrolyzes the α -1 \rightarrow 4 glycosidic linkages more quickly than the β -amylases, that's why it is also known as alpha-enzyme [78]. Amylases are occasionally added including other enzymes (pectinases and cellulases) during processes like bio-scouring to lower the operating costs.

Traditionally, size materials have been put into the warp yarns to ensure seamless weaving. The majority of the ingredients used to scale the warp yarns are starch-based. Before bleaching and coloring, the sizing protecting layers from the yarns must be eliminated [79]. De-sizing refers to the procedure of eradicating the size components from the warp yarns. Traditionally, desizing is accomplished by applying chemicals (acid, alkali, oxidizing agents) to the woven cloth at higher temperatures. Furthermore, the starch cannot be adequately removed using these traditional methods, which results in inconsistent coloring. The chemicals used in the traditional procedure degrade cotton fabric as well, which results in cotton cloth losing its natural feel. After the procedure, the leftover chemicals are released into the environment, which seriously pollutes the ecosystem [80]. Enzymatic desizing is regarded as the industry's first commercialized use of biotechnology in the textile industry. The enzyme amylase is applied to the woven cloth to break down the starch, which then cleans the warp thread. The α -amylase is one of these three amylases that are best suited for the hydrolysis of starch [81]. These enzymes create dextrin, which is easily removed by water, by breaking the connection between both the glucose molecules in the starch polymer. The use of chemicals will be reduced due to the lower treatment temperature and the gentler pH condition. Although a wide range of yeasts, fungi, and bacteria are employed to manufacture α -amylases [79]. According to a study, using ultrasonic waves increases the efficiency of amylase enzymes during the de-sizing process. Amylase can be used with hydrogen peroxide or cellulase to increase the pretreated fabric absorbency, dye uptake, and rigidity. Novozyme invented a commercial alkali stable enzyme that can be employed in a wide pH (6–10) and temperature (30–90°C) range [82]. It has also been found that immobilized amylase, is less susceptible to pH, chemicals, and temperature [83]. The immobilized amylase has potential industrial applications using the magnetic cross-linked enzyme aggregation technique. For prolonged usage, this immobilized enzyme is simply detached under a magnetic field [84].

5.2 Pectinases

The middle lamella of plant cell walls contains complex polysaccharides like pectin and other compounds. The complex group of enzymes known as pectinases is responsible for the breakdown of these compounds. In nature, saprophytes and plant pathogens (bacteria and fungi) largely create them for the breakdown of plant cell walls. Pectin degrading enzymes fall into three categories: polygalacturonases (PGs), pectin esterases (PEs), and polygalacturonate lyases (PGLs) [85]. In addition to bacteria and fungi, pectin esterases are primarily produced by plants like tomatoes, citrus fruits, and bananas. Most microbial and plant PEs have a molecular weight of 3050 kDa where the ideal pH for their activity ranges from 4.0 to 7.0. Polygalacturonases are a class of enzymes that hydrolyze α -1,4 glycosidic bonds in pectin by utilizing mutually endo- and exo-splitting methods [77]. These enzymes are frequently found with molecular weights of 3080 kDa and work at an ideal temperature between 30 and 50°C, and their ideal pH range from 2.5 to 6. Although PGL from *Bacillus licheniformis* remained ideal in the pH range between 8.0 and 7.0. Although PGL from thermophiles has an optimal range between 50 to 75°C, the optimal range for PGL activity is often between 30 and 40°C [76].

The cuticle and primary wall of a raw cotton fiber include natural contaminants or noncellulosic substances (oil, proteins, waxes, pectin, lipids). To create absorbency cotton fibers, non-cellulosic elements are eliminated by the process of scouring which is boiling the gray cotton fabric in a 3% alkaline solution [86]. This conventional

method has several drawbacks, including using large amounts of chemical compounds, necessitating rapid rise after scouring to make the fabric neutral, reducing the fabric weight by 6%, weakening cotton fabrics due to the formation of oxycellulose in the presence of oxygen, and usually requires more energy due to boiling temperatures [87]. The wastewater that results also has a high salt content, a great chemical oxygen demand (COD), and a high biological oxygen demand (BOD). Enzymatic scouring has been looked into in this situation as a potential replacement for conventional alkaline-based scouring [88]. In comparison to the conventional method, bioscouring has many benefits, including less water usage and energy-saving treatment temperatures. Since enzymes do not destroy cotton, unlike conventional scouring methods, there is no loss of weight or strength. Pectinases are frequently employed for scrubbing cotton as it works by breaking down the pectin in cotton's main cell wall [89]. The noncellulosic material is kept linked to the cellulose by pectin, as a result, the other contaminants can be easily separated if the pectin is eliminated. Polygalacturonases, pectin esterases, and polygalacturonate lyases are the three groups of pectinases [90]. The optimum temperature and pH range of 30–50°C and 05–08 has been reported for pectin esterases to catalyze the hydrolysis of pectin methyl esters and convert them into pectin acid. Glycosidic links in pectin are hydrolyzed by polygalacturonases which optimally work in a temperature range of 40–60°C and acidic pH 03–07 [91]. Pectinases and cellulases are usually used in combination for bio-scouring, pectinases break down pectin in this conjunction whereas cellulases demolish the cuticle by rupturing the main wall. The outcomes demonstrated that the immobilized enzymes were as effective as the combination of pectinase and cellulase and this approach is more efficient than aqueous bio-scouring [92]. High temperatures and alkaline environments are required to increase the activity and stability of pectinase. *Bacillus pumilus* BK2 has been reported as a novel source of pectate lyase, with optimal activity at pH 8.5 and a temperature of 70°C to evaluate the bio-scouring of cotton fabric [87]. To well comprehend the procedure of bioscouring and its impacts on textile materials, it is crucial to characterize the biochemical and physical surface modifications of clothes following bio-scouring and to identify effective methodologies for surface characterization [4].

5.3 Laccases

Multicopper enzymes called laccases catalyze the oxidation of a variety of phenolic and non-phenolic substances by reducing molecular oxygen by four electrons to produce water. Laccases are most common in fungi, though they have been discovered in plants, insects, and bacteria. More than 60 fungi species have been shown to produce laccase having a molecular weight of 6070 kDa, with an ideal pH range of acidic, and an ideal temperature range of 50 to 70°C [73]. Laccase from *Ganoderma lucidum* is one of the enzymes with optimal temperatures below 35°C, it works at an optimum temperature of 25°C. Laccases have several different roles in the treatment of textiles, including finishing fabric, bleaching, scouring, and dyeing wool, as well as playing a part in water treatment and dye synthesis. The laccases are extensively researched for denim bleaching to substitute stone washing because they can destroy indigo [93]. Research on laccases for the decolorization of textile effluents is extensively used as an eco-friendly method for treating dye wastewater because they have the potential to degrade a wide range of chemical compounds including synthetic dyes [94].

Conventionally, cotton is bleached by discoloring natural pigments, which gives white appearance to cotton fabric. Cotton has natural pigments, primarily flavonoids,

which give its inherent grayness. The procedure of bleaching is used to take out the textile's natural colorants [95]. Hydrogen peroxide is a conventional market bleaching agent that works at pH 11–13 and temperatures up to 130°C to bleach materials. The traditional bleaching technique has significant drawbacks due to high temperatures and alkaline pH which seriously harm the fabric [96]. The bleaching compounds can also reduce cotton's degree of polymerization, which create overall damage to the fabric. Additionally, more water is needed after bleaching to neutralize the fabric and eliminate extra hydrogen peroxide. The use of enzymes can solve the issues with the conventional procedure [97]. Cotton is bleached by laccases which oxidized the flavonoids present in the fabric. These enzymes are utilized at temperatures between 60 and 80°C with an acidic pH, where the use of ultrasonic energy could enhance laccases' bleaching effect [98]. Besides that, glucose oxidases can generate hydrogen peroxide and gluconic acid in an aqueous solution by oxidizing glucose at acidic pH to neutral and lower temperature. It is feasible to reuse the desizing bath as a source of glucose, this biochemical method offers combined desizing and bleaching [99]. The excess hydrogen peroxide is traditionally eliminated after bleaching cotton cloth by using a reductant or by rinsing it with water. Besides this traditional method can be replaced by catalase which converts hydrogen peroxide into water and oxygen [100]. Previous studies reported a laccase-assisted wool dyeing technique that uses low temperatures and no dyeing auxiliaries and prevents the excessive use of water and energy [101]. In a more recent study, laccases were used to catalyze an enzyme process that used natural flavonoids to color cotton [102].

5.4 Cellulases

Cellulases are hydrolytic enzymes that speed up the process of cellulose breaking down into smaller oligosaccharides and then glucose. Cellulase activity signifies the combined action of at least three different types of cellulases in a multicomponent enzyme system [103]. Combinations of all three varieties of enzymes have better activity than the total activities of every enzyme alone because cellobiohydrolases work synergistically with endoglucanases and each other. Exo-cellulases generate cellobiose and soluble oligosaccharides, which are then transformed into glucose by β -4-glucosidase [104]. Numerous fungus cellulases are modular proteins made up of a connecting linker, a carbohydrate-binding domain (CBD), and a catalytic domain (CD). CBD serves as a mediator for the enzyme's attachment to the substrate of insoluble cellulose [105]. Temperatures between 30 to 60°C are the active range for cellulases, but they are categorized as acid-stable (pH 4.55), neutral (pH 6.67), or alkali stable depending on how sensitive to pH (pH 9.10). Cellulases were first time utilized in the textile processing industry for the finishing of denim in the late 1980s. Cellulases are currently utilized to treat cotton and other cellulose-based fabrics in addition to bio stoning [86].

Many clothes are given a washing action to provide them a somewhat worn appearance, such as stone-washing of denim jeans, which causes the blue denim to fade due to the pumice stones. This ancient method has some drawbacks, including difficulties in removing pumice residue from denim clothing, harm to the machinery and clothing, dust in the washing equipment, and ecological damage [106]. The use of pumice stone has decreased or has been entirely removed in the industry of textile due to the introduction of cellulase enzymes. In the business of textile, indigo dyes are typically used to color fabric, whereas cellulase enables the surface of the fabric to hydrolyze, which eliminates some of the indigo dyes from the surface of the fabric

and gives it an aging and faded appearance [107]. Part of the indigo is removed from the fiber's surface through partial hydrolysis, resulting in bright patches. The current research focuses on the prevention or improvement of back staining, which is the redeposition of liberated indigo onto the clothing. According to reports, back staining issues can be reduced by utilizing neutral and endo cellulases at neutral pH. Purified and characterized 20 and 50 kDa endoglucanase, as well as a 50 kDa cellobiohydrolase, were previously reported for their use in textile processing at neutral pH [75]. The 20 kDa endoglucanase performed well during biostoning, and it was feasible to reduce the amount of back staining by joining the 50 kDa cellobiohydrolase or 50 kDa endoglucanase with the 20 kDa endoglucanase [108]. This problem is also solved by using enzyme-based anti-black staining agents which are composed of lipases and proteases. An important area of research has been the optimization of biofinishing procedures to collect the enzyme and reuse it. The preventive measures that have to be adopted for the usage of cellulases are that it should operate with deliberate kinetics so that no impairment appears to the internal composition of fiber and the procedure should be confined to the hydrolysis of only unfastened surface fibrils [103]. This issue can be solved by selecting the right immobilized enzyme, adjusting the concentration and incubation duration, using liquids with varying viscosities, making foam ingredients, and using hydrophobic agents to impregnate clothing.

5.5 Serine proteases: Subtilisins

Alkaline serine proteases belonging to the subtilisin family are typically extracted from numerous *Bacillus* species. They create an intermediate acyl-enzyme that accelerates the hydrolysis of peptide and ester linkages. Subtilisins are made as pre-proteins precursors, where's the active site of the enzyme made of a catalytic triad of aspartate, serine, and histidine [2]. Majorly subtilisins have a molecular weight between 15 and 30 kDa, however, there is a small number of anticipations like *B. subtilis* subtilisin having a molecular weight of 90 kDa. Alkaline proteases have an optimum temperature range of 50 to 70°C, where they function at their best, but they are relatively stable at higher temperatures [109]. Thermos ability of enzyme is increased when one or more calcium binding sites are present. Subtilisins can be effectively inhibited by diisopropyl-fluorophosphate (DFP) and phenyl methyl sulphonyl fluoride (PMSF). Therefore, most subtilisin protein engineering has concentrated on improving thermostability, substrate selectivity, and oxidation resistance [33].

Raw wool is hydrophobic because of the epicuticle surface membranes comprising fatty acids and hydrophobic contaminants such as grease and wax. Alkaline scouring with sodium carbonate and preparation with potassium permanganate, sodium sulfite, or hydrogen peroxide is common methods to remove these contaminants [72]. When wet processed, wool cloth has a propensity to feel and shrink, and several chemical techniques can be used to control the way wool shrinks. The chlorine-Hercosett technique, which has been used for more than 30 years, is the very effective industrial shrink-resistant method now existing [110]. There are some significant drawbacks to this method, despite its advantages (noble anti-felt influence, little destruction, and little loss of weight), including its restricted endurance, inadequate treating features, yellowing of the fibers, challenges with coloring, and influence of the environment from the liberation of absorbable organic halogens [111]. Previous studies have recommended treating wool using safe chemical techniques, like low-temperature plasma. Plasma treatment, which is a dry method, uses electric gas discharges to treat wool fabric, since no chemicals are used in the process and only

the surface characteristics of the wool are altered, it is considered as being environmentally [112]. However, the commercialization of a plasma treatment technique is limited by costs, compatibility, capacity, and the shrink-resist qualities acquired do not impart a machine-washable finish, which is one of the primary goals. A natural polymer, like chitosan, may then be used to enhance the wool's anti-felting or shrink-resistance qualities [113].

Proteases of the subtilisin form have lately been investigated as a substitute for chemically pre-treating wool, primarily for environmental concerns. According to much research, pretreating wool fabric with proteases enhanced its anti-shrinkage qualities, eliminate contaminants, and raised dyeing affinity. The enzyme can, however, enter the fabric cortex because of its small size, which leads to the degradation of the internal structure of the wool fabric [114]. According to several studies, increasing the size of the enzyme through chemical cross-linking with glutaraldehyde or by attaching synthetic polymers like polyethylene glycol might minimize the enzyme penetration, which in turn lowers potency and weight loss [115]. By increasing the cuticle's sensitivity to proteolytic decomposition, hydrogen peroxide pretreatment of wool fabric at an alkaline pH in the existence of elevated salt concentrations also focuses the activity of enzymes on the wool's outer surface [100]. As an alternative to the current proteases, the search for novel protease-producing microorganisms with great cuticle specificity is being researched.

5.6 Nitrilases and nitrile hydratases

About 40 years ago, the nitrile-hydrolyzing enzyme nitrilase was originally identified. After studying the composition and amino acid sequence of nitrilase, 13 divisions make up the nitrilase superfamily. In contrast to the eight or more branches that appear to have evident amidase or amide concentration activities, associates of only a single division are identified to exhibit the actual activity of nitrilase [116]. A small number of fungal species and 3 out of the 21 plant families have this enzyme function, however, it is more frequently found in bacteria. It is known that certain taxa, including *Pseudomonas*, *Klebsiella*, *Nocardia*, and *Rhodococcus*, use nitriles as their only carbon and nitrogen sources [117]. Different bacteria and fungi that can hydrolyze nitriles have been discovered, mostly because of the biotechnological potential of nitrilases. The majority of the isolated nitrilases were composed of one polypeptide with a molecular weight of 3045 kDa, which under certain conditions would assemble to create the active holoenzyme [118]. The enzyme appears to exist most frequently as a big aggregate with 626 subunits, although increased levels of solvents which are organic in nature, temperature, pH, salt, or even the enzyme itself can cause subunit interaction and subsequently stimulation, whereas the majority of enzymes exhibit substrate-dependent activation [119]. The primary enzyme in the enzymatic process for converting nitriles to amides, which are then transformed by amidases to the appropriate acid, is nitrile hydratase (NHase). Several microorganisms with NHase activity have been isolated, and the enzymes have been refined.

Excellent characteristics of polyacrylonitrile fiber (PAN), including their great biochemical conflict, superior flexibility, and ordinary-looking esthetic qualities, have led to a rise in their use; at the moment, they account for 10% of the synthetic fiber market worldwide [3]. However, PAN textiles' hydrophobic characteristic imposes unwanted qualities, making the dyeing and finishing procedure challenging. The surface chemical hydrolysis of PAN fabrics typically causes an irreversible yellowing of the fabric [120]. Selective enzymatic hydrolysis of PAN could therefore

be a fascinating alternative, just like with other synthetic fabrics. Different sources (*Rhodococcus*, *Rhodochrous*, and *Agrobacterium tumefaciens*) of nitrile hydratase and amidase affected the surface of PAN [121]. The fabric's hydrophilicity and dye absorption improved after enzymatic treatment. In previous studies, PAN was treated with nitrile hydratases extracted from *Brevibacterium imperiale*, *Corynebacterium nitrilophilus*, and *Arthrobacter sp.* showed an increased number of amide groups on the PAN outward, increasing its hydrophilicity and dye ability [122]. In different investigations, it was discovered that the *Micrococcus luteus* strain BST20 produces membrane-bounded nitrile hydrolysing enzymes, which were shown to hydrolyze nitrile groups on the PAN surface by determining the NH₃ release from PAN powder and the depth of shade of enzyme-treated fabric following dyeing with a basic dye [123]. Matamá et al. [57] demonstrated the biomodification of acrylic fabric by utilizing a nitrilase rather than nitrile hydratases/amidases. The catalytic efficiency was increased by adding 4% N-N-dimethylacetamide and 1 M sorbitol to the treatment solution. Previous findings show that the enzymatic action of PAN will improve the characteristics of treated fabric while also saving energy and reducing pollutants, even though there is not an industrial application for it yet.

5.7 Lipase/esterases

Acyl-hydrolase enzymes sometimes referred to as lipases found in a vast variety of animals, plants, and microorganisms, accelerate the breakdown of triacyl glycerol into fatty acids and glycerol. These enzymes exhibit excellent regio- and stereo specificity a broad substrate tolerance, and other properties that make them desirable biocatalysts for the synthesis of fine chemicals and the creation of optically pure molecules [124]. They are often quite stable, do not require cofactors, and even function in organic solvents [96]. For lipases and esterases, the mechanism for ester hydrolysis or production consists of four steps in which the substrate is attached to the active serine, resulting in a tetrahedral intermediate that is stabilized by the catalytic His and Asp residues [125]. A tetrahedral intermediate is formed by the attack of a nucleophile, which following resolution produces the desired product (an acid or an ester) and free enzyme. The interfacial activation phenomenon allows lipases to be separated from esterases (which is only observed for lipases) [126]. A hydrophobic domain (lid) shielding the active site of lipase is responsible for this interfacial activation, according to structure elucidation. This lid will only open in the existence of the least concentration of substrate, such as a hydrophobic solvent that is organic in nature or a triglyceride phase, allowing access to the active site [127]. Esterases and lipases were among the main enzymes to be investigated for stability and activity in organic solvents; however, lipases exhibit this property more clearly. In the clothing industry, lipases are mostly utilized for bio-scouring, desizing fabrics, and surface functionalization of synthetic fibers. They may also be used in conjunction with other enzymes including protease, and xylanases [128]. Polyester fabrics' ability to absorb color is improved by the lipase enzyme, which also causes less surface abrasion and weight loss.

The fabrics made of PET (polyethylene terephthalate), PAN (polyacrylonitrile), and PA (polyamide) exhibit exceptional qualities such as good potency, great chemical endurance, minimal abrasion, and minimal shrinkage [129]. Extraordinary hydrophobicity and crystallinity, which impair wearing comfort (making these fabrics less suitable to be in contact with human skin), as well as the processing of fabrics (preventing the usage of finishing chemicals and dyeing representatives), are major drawbacks of synthetic fabrics [130]. Major finishing procedures/agents depend on water, hence

increasing the hydrophilicity of the fabric surface is necessary. Currently, sodium hydroxide-based chemical treatments are utilized to increase the hydrophilicity and increase flexibility of fabrics [131]. Chemical treatment, which can cause undesirable weight and strength losses as well as irretrievable yellowing in the example of PA and PAN fabrics, is challenging to control [132]. The method also harms the environment because it uses a lot of energy and chemicals. Utilizing enzymes to adapt the surface of synthetic fabrics is a recently discovered substitute [133]. Substantial depilling, effective desizing, elevated hydrophilicity and reactivity with cationic dyes, and enhanced oily stain release were all outcomes of the enzymatic treatment [108].

5.8 Catalase

The enzymes that accelerate the breakdown of hydrogen peroxide into water and oxygen are referred to as catalases, also known as hydroperoxidases. After desizing and scouring but before dyeing, H₂O₂ is used in bleaching in the textile industry [134]. Historically, hydrogen peroxide was destroyed with a reducing agent and was rinsed with water. Conventionally, sodium bisulfate, which requires high temperatures and thorough rinsing, is used to eliminate hydrogen peroxide after bleaching [120]. Catalases can be used to break down extra hydrogen peroxide at low temperatures, this makes the procedure more affordable and environmentally friendly. They are made by a range of microorganisms, comprising bacteria and fungus, and many of them perform best at temperatures around 20°C and a pH of 07 [72]. The synthesis of microbial CATs will only be economically viable when using recombinant strains and low-cost technologies, or for CATs with particular qualities like thermostability or action at alkaline or acidic pH [135]. CATs from animal resources (bovine liver) are often inexpensive. The use of immobilized enzymes could lower the rate of enzyme for the breakdown of hydrogen peroxide in bleaching wastes, permitting not only the retrieval of the enzyme but also the reutilization of preserved decolorizing over-flows for coloring [136].

6. Future prospects

In practically every stage of manufacturing textile fibers, enzymes can be employed to create ecologically acceptable alternatives to chemical processes. Amylases for desizing, cellulases, laccases for denim finishing, and proteases included in commercial products are just a few examples of existing successful commercial uses. Commercial enzyme-based techniques for the bio-modification of artificial and natural fibers must first undergo additional study before they can be put into use. The quest for novel enzyme-producing microorganisms and enzymes derived from microorganisms is an important area of research. Future textile processing still has a lot of room for novel and enhanced enzyme uses.

7. Conclusion

Enzymes are the greatest substitute for the best textile processing they not only help the environment but also save a great deal of money by consuming less energy and water, which lowers the production cost. It appears that all processes will be able to be carried out utilizing enzymes in the future as the employment of diverse

enzymes is still in its infancy, but their inventive uses are growing and expanding quickly into every aspect of textile production. Companies that produce enzymes are always working to make their products better for a wider variety of usage scenarios. The biggest barrier to employing enzymes is their high price. The textile industry was identified as a market area with great potential for implementing biotech but limited biotech awareness at the moment. Through the adoption and implementation of the enzymatic process, high value-added textile products with top quality may be produced while consuming less power, water, and other resources as well as enforcing to assure economic and environmental improvements along with sustainable development and social responsibility. In the area of treating textiles, enzymes are becoming much more prevalent and they can be used much more extensively in the textile industry if their expense can be controlled.

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