

## Exploring the Color and Biofunctional Potential of Six Natural Textile Dyes: Eucalyptus, Weld, Madder, Annatto, Indigo, and Woad

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### ABSTRACT

Concerns over the environmental and health impacts of synthetic dyes have intensified interest in natural dyes as sustainable and eco-friendly alternatives. Despite this potential, natural dyes have traditionally produced limited color ranges and exhibited poor fastness, and research exploring their broader applications remains scarce. Beyond coloration, natural dyes can impart additional functional properties to textiles, including antibacterial and UV-protective effects. This study presents a systematic review of 38 publications examining six plant-derived dyes: eucalyptus (*Eucalyptus globulus* Labill.), weld (*Reseda luteola* L.), madder (*Rubia tinctorum* L.), annatto (*Bixa orellana* L.), true indigo (*Indigofera tinctoria* L.), and woad (*Isatis tinctoria* L.). These dyes were selected following an initial assessment of plant-based dyes with primary color potential, considering both their chromatic and biofunctional properties. The review evaluates how dyeing parameters and the use of auxiliary agents influence color outcomes and functional attributes. Findings include a summary of chromatic possibilities across different materials and processing conditions. Integrating natural dyes with environmentally friendly auxiliary products emerges as a promising strategy to expand the color palette while maintaining sustainability. Further studies are needed to optimize natural dyeing techniques and advance the development of eco-conscious textile coloration.

**Keywords:** Natural dyes, Medicinal properties, Functional textiles, Color, Textile design, Sustainability

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### Introduction

Textile industry pollution represents a significant global challenge [1], contributing to the release of hazardous chemicals into the environment, which can disrupt ecosystems and pose risks to human health [2]. Despite growing environmental concerns, synthetic materials and chemicals continue to dominate textile manufacturing due to their efficiency, durability, and ease of use, especially in dyeing operations [3]. In some cases, heavy metals and chemical auxiliaries—including acids, promoters, alkalis, oils, and softeners—are employed to enhance color strength (K/S) and improve color fastness [4].

Color strength reflects the extent to which a dyed material absorbs light relative to an undyed or untreated sample. It can be quantified using the Kubelka-Munk equation, which establishes a linear relationship between reflectance and the concentration of dye on a fiber. In this equation, R represents the percentage reflectance of a light source at a given wavelength on an opaque material layer, K denotes the absorption coefficient, and S the scattering coefficient [5]. Higher K/S values indicate greater dye uptake by the fabric, resulting in more vivid and intense coloration [6].

$$\frac{K}{S} = \frac{(1 - R)^2}{2R} \quad (1)$$

Color fastness refers to a material's ability to maintain its original color over time and resist the transfer of its dyes to neighboring materials. Various factors, including light exposure, washing, friction, and perspiration, can impact textile coloration, affecting both quality and appearance [7]. Assessment of color fastness can be performed qualitatively through visual inspection or quantitatively using precise color measurement techniques. Changes in color are evaluated by comparing samples before and after exposure to specific conditions, while color transfer, expressed as staining, is assessed by examining the color change on adjacent fabrics [8, 9].

Several standardized methods exist to evaluate color fastness under different conditions and for diverse applications. The primary organizations responsible for developing these standards include the International Standards Organization (ISO), the American Association of Textile Chemists and Colorists (AATCC), and the European Committee for Standardization (CEN). Examples of these standards are ISO 105, AATCC 16, and EN ISO 105, which employ different rating scales. The gray scale, ranging from 1 (very poor) to 5 (excellent), is commonly used for general fastness evaluation. The blue scale, applied to assess light fastness, ranges from 1 (very poor) to 8 (excellent). Additionally, the CIE Lab color system quantifies color differences by measuring reflectance values of test and standard samples at multiple wavelengths using a spectrophotometer. The resulting color difference is calculated using the CIE Lab formula:

$$\Delta E_{ab}^* = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2} \quad (2)$$

The  $\Delta$  symbol means "difference in",  $L^*$  value indicates lightness (vertical axis on the color space diagram with values from 0 to 100, black to white),  $a^*$  value indicates red-green component of a color (+ $a^*$  being red and - $a^*$  being green) and  $b^*$  value indicates yellow-blue axis (+ $b^*$  being yellow and - $b^*$  being blue) [5, 10].

In addition to chemicals used to enhance K/S values and color fastness, dyed textiles often undergo additional finishing treatments, such as waterproofing, abrasion resistance, and antimicrobial protection, which typically involve synthetic chemical agents [4]. Prolonged human exposure to synthetic dyes and certain auxiliary chemicals can lead to chronic health issues, including dermatitis, respiratory disorders, and carcinogenic effects [11–13]. Contact with these substances begins early in life and continues over the lifespan, with the skin serving as the primary route of absorption [14]. Regions of the body with higher perspiration and friction are particularly susceptible, increasing the risk of dermatological problems [11, 12].

Growing demand for sustainable practices and a focus on human well-being have prompted the textile industry to adopt eco-friendly dyeing techniques [15]. Consequently, natural products such as plant extracts are increasingly used as safer alternatives to synthetic dyes, offering reduced environmental impact and lower health risks [3, 16–18]. Plant-based dyes have a long history of use, providing not only coloration but also potential health benefits [19, 20]. Just as the skin absorbs harmful chemicals, it can also take up beneficial bioactive compounds present in natural dyes.

Among the key functional properties relevant to textiles and wearer well-being are antimicrobial activity and ultraviolet (UV) protection [21]. Antimicrobial finishes are critical for controlling microbial growth, reducing odor, and preventing damage to the textile. Factors such as fiber properties, substrate structure, chemical treatments, and environmental conditions like temperature and humidity facilitate microbial colonization [3, 19]. Close contact with the human body further supports the adherence, transfer, and proliferation of microorganisms, which can cause odors, stains, discoloration, loss of material properties, and skin infections. Antimicrobial treatments help mitigate these issues [22].

Protection against UV radiation is another important factor for human health, as excessive UV exposure can damage the skin [18]. Although synthetic agents like phenyl salicylates, benzophenones, and certain acids provide UV protection, they are environmentally hazardous [3]. Interestingly, some natural dyes, including madder and indigo, have demonstrated inherent UV-blocking capabilities [18, 23, 24].

Aesthetic qualities, particularly color, remain central to the textile industry, highlighting the importance of both dye selection and dyeing processes [25]. While color occurs naturally in textiles, it is also introduced through dyes and pigments. The primary difference between these two types of colorants lies in solubility: pigments are insoluble compounds that remain chemically and physically unaffected by the substrate, whereas dyes are typically soluble in water (or otherwise treated to eliminate crystalline structures) and interact with the substrate during application [8]. Selection of dye type depends on the fiber being treated.

Historically, humans have sourced animals and plants to develop dyes and pigments capable of coloring various materials [26, 27]. However, natural textile colorants face limitations that impede their widespread use, including

high production costs, variability in raw material quality, complex and time-intensive extraction processes, poor fastness, and a lack of standardized procedures for color production, such as dyeing parameters and mordanting techniques [28]. Unlocking the full potential of natural dyes is essential to expand the available range of colors. Special attention should be given to natural dyes that can function as primary colors, enabling the formulation of diverse secondary shades [24].

#### *Primary colors and color mixing in textiles*

Primary colors are fundamental hues that cannot be created by combining other colors, yet they serve as building blocks for generating the full spectrum of additional colors [8, 29]. Color mixing can occur in two main systems: additive (light-based) and subtractive (pigment-based). Additive mixing involves light, where red, green, and blue combine to produce white light. In subtractive mixing, colors are created by pigments, with magenta, cyan, and yellow as primary colors [10, 29]. Since magenta and cyan are rare in nature, traditional color theory for natural dyes adopted red, yellow, and blue as the primary colors for subtractive mixing before synthetic dyes were developed [30]. In this system, mixing pigments typically results in darker, less luminous shades [8, 10, 29].

In textile applications, combining primary colors is a standard approach to achieve a broad range of shades. Using primary colors from natural dyes offers the possibility of expanding the palette of naturally dyed textiles [24]. However, achieving consistent and strong coloration with natural dyes is challenging [31]. Dye uptake depends on factors such as the chemical structure and molecular weight of the dye molecules, dye bath pH and temperature, fiber type and structure, and the presence and type of mordants [32]. Substantivity—or the affinity between the dye and the fiber—is often low for natural dyes, leading to weak fastness because many dyes lack reactive functional groups [28]. Mordants are traditionally used to improve dye fixation, yet many are toxic and pose environmental and health risks due to high metal concentrations [33, 34]. The choice of mordants and dyeing methods significantly influences both color strength (K/S values) and fastness properties [35].

Vat dyes, such as true indigo (*Indigofera tinctoria*) and woad (*Isatis tinctoria*), offer an alternative approach by not requiring mordants. These dyes are insoluble in water but can be chemically reduced to a soluble form, which penetrates fibers and then oxidizes back to its insoluble state, eliminating the need for additional mordants [7, 36]. Based on an initial review of plant-based dyes producing primary colors—yellow, red, and blue—six dyes were selected for further study due to their color properties and potential medicinal benefits: eucalyptus (*Eucalyptus globulus* Labill.) and weld (*Reseda luteola* L.) for yellow, madder (*Rubia tinctorum* L.) and annatto (*Bixa orellana* L.) for red, and true indigo and woad for blue. True indigo and woad are separate species, with “indigo” here referring specifically to *Indigofera tinctoria*.

This study systematically examines existing research on the medicinal and chromatic characteristics of textiles dyed with these six natural dyes. It investigates how various dyeing parameters—including method, concentration, temperature, duration, fiber type, and mordanting—affect both color quality and functional properties. The literature reveals a lack of recent research on advancements in this field, highlighting the need for an integrative overview that considers both coloristic and medicinal aspects. The findings provide a foundation for future experimental studies.

## **Materials and Methods**

A systematic review was performed following Torracco’s approach [37], focusing on six natural textile dyes: eucalyptus, weld, madder, annatto, indigo, and woad. The review addressed five aspects: (i) primary chromatic and medicinal properties; (ii) dyeing techniques and mordant usage; (iii) treatments and auxiliary substances; (iv) color strength; and (v) fastness characteristics.

The literature selection was carried out in three stages. Initially, two Scopus database searches were conducted. Next, publications from two specialized research centers were reviewed. After screening for relevance, 38 studies were included in the final analysis (**Figure 1**).

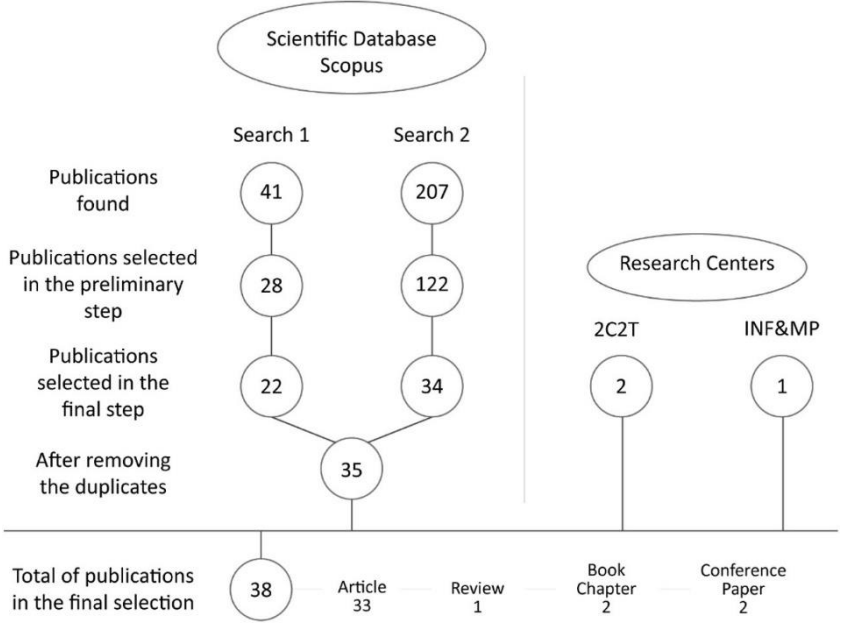


Figure 1. Literature search workflow

Data for this study were collected through a structured search in the Scopus database, as well as publications from two specialized research centers: the Centre for Textile Science and Technology (2C2T, Portugal) and the Institute of Natural Fibers and Medicinal Plants (INF&MP, Poland). The database search used a combination of keywords targeting six plant-based dyes—eucalyptus, weld, madder, annatto, indigo, and woad—and their application in textiles, considering coloristic and functional properties. Keywords included plant names (scientific and common), textile-related terms (textile\*, fashion), color descriptors (color\*, colour\*, dye\*), and functional/medicinal properties (medicinal, antimicrobial, antibacterial, anti-inflammatory, anti-UV, antioxidant). Only peer-reviewed research articles, review papers, book chapters, and conference proceedings were considered. The search was limited to publications between 2015 and 2023, and the initial step screened titles, abstracts, and keywords to remove irrelevant studies. From this, 41 publications were initially retained. A more detailed review led to the pre-selection of 28 studies, which were further filtered based on full-text availability, English language, specific focus on the dye species, and relevance to textile dyeing methods. This process resulted in 22 studies being included from the Scopus search.

To ensure comprehensive coverage, a second search was conducted with a minor adjustment to the Boolean operators, which yielded 207 articles, including 36 duplicates from the previous search. After screening and applying inclusion criteria, 34 studies were selected. Merging results from both searches, and adding one missing paper, resulted in 35 studies.

Publications from 2C2T and INF&MP were also assessed using similar criteria. Of six preliminary papers, three met the requirements for inclusion. Combining the database and center-derived studies, a total of 38 publications were included in the review. Selected studies were organized by title, authorship, publication type, source, and year. A narrative synthesis was performed, summarizing key findings per dye in **Table 1**.

### Results and Discussion

This review analyzed six plant-based dyes—eucalyptus, weld, madder, annatto, woad, and indigo—focusing on their applications in textile coloration and associated functional properties. Each dye was assessed for its frequency of citation in the literature, medicinal benefits, color fastness performance, K/S values, textile substrates used, auxiliary agents, and mordanting methods. The results indicate that these dyes differ in both their coloristic and functional potential, with some offering additional bioactive properties such as antimicrobial or UV protection. **Table 1** synthesizes the main findings, highlighting differences in dyeing performance, treatment requirements, and possible health-related functionalities of fabrics treated with these natural dyes.

Table 1. Main findings of the research.

Natural Dye / Plant Source	Reference(s)	Year	Textile Substrate	Main Colors Obtained	Key Functional Properties Reported	Color Fastness Highlights	Other Notable Findings
Eucalyptus (Eucalyptus globulus Labill.)	Das & Das [19]	2022	Cotton	Yellow–brown shades	↑ K/S with concentration & alum mordant; strong antibacterial (especially vs Gram-positive)	Good wash/light fastness; activity ↓ after 20 washes	Stronger effect on Gram-positive bacteria
	Endris & Govindan [38, 39]	2021-22	Cotton	Yellow	Excellent K/S; essential oil post-treatment ↑ antibacterial activity	Very good light, wash, rub fastness	Essential oil finishing markedly improves antibacterial performance
	Yilmaz [40]	2021	Wool	Golden yellow → brown (mordant-dependent)	Antibacterial vs S. aureus (non-mordanted) & vs E. coli (with mordants); highest K/S with CuSO <sub>4</sub>	Good light & wash fastness	Wide shade range with different mordants
	Maqbool <i>et al.</i> [41]	2019	Cotton	Not specified	Excellent UV protection, good insect repellency, antibacterial activity	Good overall fastness, moderate K/S	Multi-functional performance among seven plant dyes
	Silva <i>et al.</i> [42]	2018	Cotton	Not specified	Good antimicrobial & anti-UV; chitosan pre-mordanting ↑ dye uptake & fastness	Improved fastness (eliminates electrolytes)	Sustainable chitosan mordanting process

			<b>Madder</b> ( <i>Rubia tinctorum</i> L.)						<b>Weld</b> ( <i>Reseda luteola</i> L.)						
Fernandes <i>et al.</i> [49]	2022	Cotton	Sadeghi-Kiakhani <i>et al.</i> [46-48]	2021-22	Wool / Cotton	Elmaaty <i>et al.</i> [13]	2023	Wool	Sadeghi-Kiakhani <i>et al.</i> [45]	2018	Cotton	Dumitrescu <i>et al.</i> [44]	Schmidt-Przewozna & Zajaczek [43]	2022	Linen
Yellow → purple (pH-dependent)			Red-orange			Reddish-brown → bright orange			Yellow			Yellow	Lemon yellow → olive/brown		
Cationization ↑ dye uptake; plasma + metallic mordants ↑ fastness & UV protection			Dendrimer, AgNPs, SA-AgNPs pre-treatments → dramatic ↑ K/S, antioxidant & antimicrobial			↑ K/S with bio- & chemical mordants; good-excellent antibacterial activity			Chitosan-cyanuric chloride pretreatment gave higher K/S & microbial reduction than alum			Excellent UV protection, moderate antibacterial activity	Antimicrobial, excellent UV protection, positive skin hydration		
Improved with simultaneous metallic mordanting			Very good fastness			Very good to excellent fastness			Good overall fastness			Poor wash/acid perspiration; moderate light	Not detailed		
pH strongly influences final color			50 % less dye & time needed with advanced pre-treatments			Wide shade palette with different bio-mordants			Biomordant hybrid superior to alum			Mimosa + alum mordanting system	Meta-mordanting dramatically changes hue		

True Indigo ( <i>Indigofera tinctoria</i> L.)	Annatto ( <i>Bixa orellana</i> L.)			Alves <i>et al.</i> [50]		
	Chattopadhyay <i>et al.</i> [62]	Moses & Venkataraman [59–61]	Jajpura <i>et al.</i> [58]	Multiple others [22, 51–57]	2022	Cotton
	2015	2016-17	2019	2016-21		
	Jute	Cotton	Various	Wool, silk, cotton		
	Light yellow → dark red	Orange-yellow	Yellow to red	Red–orange–brown	Orange–red	
	Very good UV protection & antimicrobial activity with bio/inorganic mordants	↑ antibacterial & UV protection after alkali/enzyme pre-treatment	Carotenoid-based; good biological activity on natural fibers	Consistent antibacterial, antioxidant, anti-wrinkle & excellent UV protection	Quebracho & laccase systems ↑ K/S, wash/light fastness & UPF	
	Moderate light, very good rub & wash	Excellent light, satisfactory wash, poor rub	Variable	Generally good–excellent wash & light fastness	Higher with enzyme + metal combinations	
	Myrobalan/pomegranate + metal mordants give best results	Pre-treatment critical for performance	Potential in textile & leather sectors	Silver nanoparticles & dendrimers markedly enhance performance	Bio-mordants competitive with synthetic ones	
True Indigo ( <i>Indigofera tinctoria</i> L.)	Reningyas <i>et al.</i> [63]					
	2021					
	Cotton					
	Blue					
	Nano-chitosan + ZnO coating ↑ light fastness & UV protection					
	Improved light fastness					
	ZnO nanoparticles act as UV blocker					



<b>Woad</b> ( <i>Isatis tinctoria</i> L.)	Bektaş <i>et al.</i> [36]	2016	Cotton & wool yarn	Dark blue (wool) / light blue (cotton)	Antigenotoxic effect; detoxifies sodium dithionite	Not detailed	Environmentally safer reduction process
	Basak <i>et al.</i> [3]	2018	Cotton/jute	Blue	Good fastness properties	Good overall fastness	Traditional blue dye with reliable performance
	Moses & Venkataraman [59–61]	2016-17	Cotton	Blue	Highest antibacterial activity among tested dyes; excellent UV protection	Excellent light/wash, poor rub	Best antimicrobial performer in the series
	Barani [64]	2020	Wool	Deep blue → brown-yellow (with AgNPs)	AgNPs (pre- or simultaneous) ↑ K/S dramatically	Not detailed	Silver treatment changes hue and depth
	Tambi <i>et al.</i> [24]	2021	Polyester	Blue, gray, green (combinations)	Good K/S (>1.7), antibacterial & UV protection, low antioxidant	Not detailed	Mixing with pomegranate/kunkum creates new shades

The analysis revealed that madder was the most frequently reported natural dye, appearing in 16 of the reviewed studies, whereas weld and woad were the least cited. All studies employed standardized methods to assess color performance and related properties. Among the investigated dyes, the most commonly reported bioactive properties were antibacterial activity and protection against ultraviolet (UV) radiation; however, woad lacked documented UV-protective effects. The antibacterial activity was generally more pronounced against gram-positive bacteria compared to gram-negative.<sup>1</sup>

<sup>1</sup> Gram-positive and gram-negative bacteria are two groups of bacteria with different cell wall structure and health implications. Gram-positive bacteria, such as *Staphylococcus* species, have a thick cell wall and can cause infections like skin infections. Gram-negative bacteria, such as *Escherichia coli*, have a thin cell wall and can cause infections like pneumonia and urinary tract infections.



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Several dyes also exhibited additional functional benefits, including anti-inflammatory, antioxidant, and anticancer effects. Beyond medicinal properties, some dyes demonstrated other advantageous characteristics, such as insect repellency, pleasant aroma, and anti-wrinkle effects.

Regarding color performance, all dyes showed satisfactory color fastness, though only a single study reported the fastness of woad. Cotton was the predominant textile substrate used for dyeing, followed by wool. Pre-mordanting emerged as the most common treatment, yielding the best color results across most dyes. Alum and chitosan were the most frequently employed mordants.

Alum, typically a hydrated double sulfate of aluminum combined with potassium, ammonium, or sodium, was found to be safe and effective. Potassium alum ( $\text{KAl}(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$ ) was the specific form used in the studies reviewed. Chitosan, a biopolymer derived from the deacetylation of chitin found in crustacean shells and certain fungal cell walls, has significant potential as a biodegradable, renewable, and non-toxic alternative to synthetic polymers. Its production can utilize seafood industry waste, offering a sustainable replacement for several harsh chemicals in textile and chemical processes. Given its environmentally friendly properties, chitosan is expected to see increased adoption in future textile applications.

#### *Eucalyptus dye*

Eucalyptus has long been utilized for its essential oils across diverse fields, including medicine, perfumery, and textile coloration [65]. The plant contains high levels of lignin, which provide resistance against microbial and enzymatic degradation, as well as numerous bioactive properties such as anti-inflammatory, antibacterial, antiseptic, anticancer, and astringent effects [40, 65]. While most research has concentrated on using eucalyptus leaf essential oils, recent studies highlight its potential as a natural textile dye.

Seven publications, including one review, were analyzed for this dye. All studies applied eucalyptus leaves or extracts to dye cotton or wool fabrics. Reported colors ranged from yellow to brown, depending on factors such as dye concentration, dyeing technique, and choice of mordant. For example, Yılmaz [40] observed that alum-mordanted wool fabrics yielded a golden yellow, tin chloride produced bright yellow, iron sulfate resulted in beige, and copper sulfate produced brown. Among these, the copper sulfate treatment showed the highest K/S values, while alum yielded the lowest, indicating a strong correlation between mordant type and both color intensity and absorption.

Other studies noted that increasing dye concentration generally enhanced K/S values at constant temperature and time [19, 38]. Endris and Govindan [39] reported that raising temperature in cotton dyeing did not affect K/S, suggesting lower temperatures may be more efficient. In contrast, Silva *et al.* [43] found that dye uptake on chitosan pre-mordanted knitted cotton increased with temperature, enhancing K/S. These discrepancies likely arise from differing dyeing methods: the padding technique, in which pressure drives the dye into fibers, versus the exhaustion method, which relies on diffusion and benefits from higher temperatures. Additional factors, such as fiber characteristics and mordant selection, also influenced outcomes.

Pre-mordanting and meta-mordanting with alum or aluminum sulfate were the most common approaches to improve K/S and fastness properties [19, 38–40]. Fastness tests for light, washing, and rubbing demonstrated satisfactory results, attributed to tannins in eucalyptus extracts, which function both as mordants and dyes [42]. Use of alternative or bio-mordants, such as chitosan, often improved fastness by one grade on gray scale evaluations. Chitosan forms crosslinks with fibers, generating positive sites that bind to the negatively charged dye molecules, enhancing color retention [42]. For instance, pre-mordanting with chitosan improved wash and rub fastness (dry and wet) from 3 to 4–5 on gray scale ratings. Comparisons of dyed fabrics with and without aluminum sulfate showed improvements in wash fastness from 1–2 to 3–4, dry rub from 4 to 4–5, wet rub from 2–3 to 3–4, and light fastness from 3 to 4 [38]. Yılmaz [40] reported that wool fabrics dyed without mordants or with alum, tin chloride, or copper sulfate achieved maximum fastness (value of 5), except for iron sulfate, which slightly reduced wash fastness to 4–5, likely due to insoluble complex formation.

All studies reported antibacterial activity for eucalyptus-dyed textiles, particularly against gram-positive bacteria. Das and Das [19] demonstrated that higher dye concentrations produced larger inhibition zones, although antibacterial activity decreased after 20 wash cycles, retaining 56–88% of the original effect. Mordant selection also influenced antibacterial performance. Yılmaz [40] observed 99.99% efficacy against *Staphylococcus aureus* regardless of mordant use, but *Escherichia coli* inhibition was only evident with alum, tin chloride, copper, or iron sulfate. Endris and Govindan [38] showed partial inhibition of *S. aureus* on aluminum sulfate-mordanted cotton but no effect against *E. coli*; however, treating fabrics with eucalyptus leaf essential oil fully eliminated both bacteria even after five washes, with partial retention after ten washes. Similar trends were reported by Maqbool *et al.* [41] and Silva *et al.* [42], confirming stronger activity against *S. aureus* and improved efficacy with chitosan pre-treatment.

UV protection was also noted, often correlated with darker shades and higher K/S values. Maqbool [41] reported that some cotton fabrics with low K/S still achieved excellent Ultraviolet Protection Factor (UPF) ratings, highlighting the influence of dye chemistry. Silva *et al.* [42] found that increasing dye concentration and pre-mordanting with chitosan enhanced UPF values, likely due to greater absorption of flavonoids and other UV-absorbing compounds.

Beyond medicinal properties, eucalyptus dye demonstrated insect-repellent effects and a pleasant fragrance, as reported in four of the seven studies [19, 38, 39, 41].

#### *Weld dye*

Weld plants have been recognized for both their health benefits and their ability to produce vibrant yellow hues in textiles [26]. Research on weld as a natural dye highlights its good washing and light fastness, alongside antimicrobial activity and potential skin benefits, such as enhanced hydration and UV protection [26, 66].

Only three publications were included for weld dye. Two studies focused on cotton fabrics, while one explored linen. The resulting colors ranged from yellow to brown [43–45]. Different mordants were applied to achieve various shades: Schmidt-Przewozna and Zajacek [45] reported that pre-mordanting with alum or gall oak (*Quercus infectoria* G. Olivier) and meta-mordanting with specific chemicals produced lemon yellow (alum), bright yellow (sodium carbonate), beige (citric acid), olive green (copper sulfate), and brown (iron sulfate). It should be noted that certain mordants, particularly copper and iron salts, pose toxicity concerns.

Mordants were commonly employed to enhance color intensity, often in combination with bio-mordants [43, 44]. Chitosan–Cyanuric Chloride hybrid (Ch–Cy) treatments were also used; Sadeghi-Kiakhani *et al.* [45] observed that Ch–Cy increased K/S values more effectively than aluminum sulfate pre-mordanting, suggesting deeper dye penetration without the need for metal-based mordants.

Weld dyeing generally improved color fastness, especially with pre-mordanting using alum and gall oak, which increased resistance to washing and light (from 4–5) without affecting rubbing, perspiration, or hot-pressing fastness, which remained high (value 4) [43]. However, some studies reported very poor fastness to washing and acid perspiration (values 1–2) when using mimosa and alum combined. Increasing mordant concentration (e.g., 8% mimosa + 15% alum) enhanced fastness to alkaline perspiration (3–4) and wet rubbing (3–4), while dry rub fastness remained high (4–5), and light fastness was moderate (3) [44].

Medicinally, weld demonstrated strong anti-UV properties, influenced by the type of mordant. Linen fabrics pretreated with alum and gall oak, followed by weld dyeing with soda, reached UPF 25, whereas copper and iron mordants achieved UPF 50 [43, 44]. Antibacterial activity was consistently reported, especially against gram-positive bacteria, and remained effective after multiple washes (10 cycles), with reductions of 91–99% for *E. coli* and *S. aureus* [45]. The Ch–Cy treatment further improved antimicrobial activity. Some studies also highlighted weld's anti-wrinkle effects in textiles.

#### *Madder dye*

Madder was the most frequently cited natural dye in the selected literature, appearing in 16 studies, including one review and one book chapter. Most studies applied madder extracts to cotton or wool fabrics, with some extending to silk and polyester. Madder also demonstrated compatibility with other natural dyes and various mordants, allowing a wide spectrum of color variations [66, 67]. However, most studies focused only on combinations with different mordants.

The resulting colors ranged from red to pink, orange, and yellow, depending on mordant selection. Aluminum sulfate yielded reddish-brown, *Punica granatum* produced deep reddish-orange, *Clonorchis sinensis* produced

brown, and *Rhus coriaria* bright orange [13]. Alum generally produced lighter shades, while combinations with mimosa tannin allowed red-orange to orange variations depending on concentration ratios [55]. Fernandes *et al.* [49] reported that solution pH influenced final fabric color, with higher pH shifting the hue from yellow to purple. Novel eco-friendly treatments were also examined to enhance K/S, fastness, and medicinal properties. Bio-nano mordants altered wool hues from pink to red, and microbial transglutaminase (m-TGase) combined with bentonite increased madder absorption and dyeability [52]. Pretreatments such as cationization, plasma treatment, and silver nanoparticle (AgNP) incorporation were also explored. Cationization introduced positive charges on cotton fibers, enhancing dye affinity and allowing salt-free dyeing [68]. Plasma treatment had minimal effect on saturation, whereas chitosan or AgNPs improved both K/S and color durability, reducing chemical, water, and energy consumption. Wool yarns pretreated with sodium alginate-silver nanoparticles (SA-AgNPs) exhibited significant increases in K/S values [47]. However, the potential environmental risks associated with silver ion release remain a consideration [69].

The review indicated that alum and aluminum sulfate were the most commonly applied mordants, often used alone or combined with treatments such as chitosan or its modified forms. Cai *et al.* [51] reported that alum mordanting increased the K/S value of madder-dyed cotton by approximately 85 % compared to direct dyeing. Sadeghi-Kiakhani *et al.* [46] demonstrated that a chitosan-poly(amidoamine) (chitosan-PAMAM) hybrid enhanced dye uptake by nearly 10 % during the exhaustion dyeing of natural dyes, outperforming unmodified chitosan. Similarly, chitosan-polypropylene imine (CS-PPI) dendrimer treatment improved K/S values, even surpassing those obtained with aluminum sulfate pre-mordanting [48].

Pre-mordanting with alum and chitosan, particularly when combined with other treatments, generally resulted in higher K/S and improved color fastness [45, 46, 53]. In contrast, the sole application of other bio-mordants, including mimosa, quebracho, and laccase, often produced unsatisfactory fastness results [50]. Exceptions were noted for tannin-rich mordants such as pomegranate, *Rhus coriaria*, and *Clonorchis sinensis*, which enhanced dye absorption due to their interaction with alizarin, the principal madder pigment [13].

Both dyeing duration and bath temperature influenced K/S values. Sadeghi-Kiakhani *et al.* [47] observed a slight increase in K/S when dyeing was extended from 30 to 60 minutes, after which values plateaued. Higher temperatures did not consistently improve dye uptake; for instance, dyeing at 70 °C yielded better absorption than at 90 °C, suggesting potential energy savings in wool dyeing.

Madder dyeing on polyester without mordants achieved moderate-to-good light fastness (3–4) and good-to-excellent washing and rubbing fastness (4–5) [56]. Elmaaty *et al.* [13] found no significant difference in fastness between wool dyed without mordants and those treated with bio-mordants or aluminum sulfate, with values ranging from 4 to 5 after five washes. Wool fabrics pretreated with bio-nano mordants displayed improved washing fastness (2–3) compared to untreated samples [52]. Schmidt-Przewozna *et al.* [54] reported that combining alum with bio-mordants, such as Chebulic Myrobalan (*Terminalia chebula* Retz) or gall oak, significantly enhanced wash fastness: on linen/elastane fabrics, ratings increased from 3–4 to 5; on linen, from 2–3 to 5; and on organic cotton, from 4–4.5 to 5. Similarly, wool pretreated with chitosan achieved excellent washing and rubbing fastness (4–5 and 5, respectively) [53]. Both Ch–Cy pre-treatment and aluminum sulfate pre-mordanting improved fastness across washing, light, and rubbing tests, likely due to amino groups on chitosan forming stronger bonds with dye molecules [45]. SA-AgNPs treatment further enhanced K/S and washing fastness from 4 to 4–5 [57]. Conversely, mimosa alone or combined with alum yielded low fastness to washing and light (1 and 3), likely due to weaker binding with the dye.

Regarding medicinal properties, 13 studies examined antibacterial effects, generally stronger against gram-positive bacteria. Elmaaty *et al.* [13] found bio-mordanted fabrics showed significant inhibition zones (15–25 mm) against gram-negative bacteria but minimal activity (3 mm) against gram-positive strains. This activity is attributed to alizarin release, which disrupts bacterial cell walls. Agnhage *et al.* [56] reported polyester dyed with madder had 86 % antibacterial activity against both bacterial types, compared to <17 % in undyed fabric.

Pre-mordanting with alum improved activity against *S. aureus*, but not *E. coli*. Combining alum with 50 % gall oak increased inhibition to >99 % for both bacteria, maintained after five washes [22]. Untreated wool yarns showed weak antimicrobial activity (20 %), which increased to 74 % with SA-AgNPs and 80–85 % after subsequent madder dyeing. These treatments provided durable antibacterial properties against *E. coli* and *S. aureus*, remaining above 70 % even after 10 washes [47].

#### Madder dye

Pour *et al.* [52] reported that pre-mordanting wool with microbial transglutaminase (m-TGase) and bentonite enhanced the antibacterial performance of madder-dyed fabrics. The antibacterial efficacy of these bio-mordants likely stems from their ability to improve dye fixation within fibers and reinforce dye activity.

In cotton textiles, madder dyeing substantially increased antibacterial properties. Cross-linking the dyed fabric with succinic acid and citric acid (SUA + CA) further enhanced antibacterial activity by over 60 % compared to untreated cotton. This improvement is attributed to the anthraquinone structure of madder, which can interact with bacterial proteins and disrupt their function, affecting both Gram-positive and Gram-negative bacteria. Fabrics with cross-linked madder dye exhibited the highest antibacterial performance, approximately 70 %, due to higher color yield [51].

Other studies also demonstrated strong antibacterial activity in non-mordanted madder-dyed fabrics, with bacterial reductions of 95.3 % for *S. aureus* and 96.2 % for *E. coli*. The linkage of chromophores to glycosides in madder likely contributes to this effect. Pre-mordanting with chitosan and increasing dye concentration further enhanced antibacterial activity, achieving bacterial reductions of 99.2 % (*S. aureus*) and 99.1 % (*E. coli*), with higher concentrations resulting in greater inhibition [53]. Sadeghi-Kiakhani *et al.* [45] observed that chitosan alone produced bacterial reductions below 90 %, whereas Ch–Cy treatment achieved 99.96 % and 98.61 % reductions for *E. coli* and *S. aureus*, respectively, which decreased to 91.14 % and 85.61 % after ten washes.

Madder-dyed fabrics also demonstrated significant UV protection. Dumitrescu *et al.* [55] found that cotton samples pre-mordanted with mimosa or mimosa-alum combinations achieved excellent UV protection (UPF 50+). Polyester fabrics dyed with 3 % and 5 % WOF of madder increased UPF values from 65 (undyed) to 106 and 112, respectively [56].

Antioxidant activity was noted in several studies [47, 49, 54]. Schmidt-Przewozna *et al.* [54] reported that madder-dyed fabrics pre-mordanted with Chebulic Myrobalan, gall oak, oak bark, or alum positively influenced skin health, including reducing skin aging, improving blood vessel tone, enhancing hydration, and minimizing skin lesions. Additionally, madder dye demonstrated anti-wrinkle properties in two studies [45, 51].

#### *Annatto dye*

Annatto has long been utilized in traditional medicine, with reported antibacterial, analgesic, healing, antioxidant, and astringent properties, making it suitable for treating skin conditions [70, 71]. Seven publications were selected, including a review and two book chapters, all employing annatto extracts to dye cotton or jute fabrics. Dyeing produced colors ranging from yellow to red to orange. Mordants were essential for influencing final fabric color [3].

Pre-treatments included sodium hydroxide, morpholine, and cellulase enzyme in three studies [59–61], while one study employed bio-mordants (myrobalan, pomegranate) in combination with inorganic mordants (ferrous sulfate or alum) [62]. The latter approach achieved superior color fastness and K/S values compared to the other treatments. Double-mordanting (bio-mordant followed by metallic mordant) improved color uniformity and consistency. K/S values increased with bio-mordant + iron sulfate but decreased with bio-mordant + alum, likely due to color interactions.

Chattopadhyay *et al.* [62] noted that wash fastness of jute fabrics dyed with annatto varied between 1 and 2 across pH ranges but improved to 3–3.5 when pre-mordanted with myrobalan + iron/alum or pomegranate + iron/alum. Light and rubbing fastness also improved, though to a lesser extent. In cotton fabrics, pre-treatments such as sodium hydroxide, morpholine, or cellulase showed minimal impact on wash fastness [61].

Annatto-dyed fabrics consistently exhibited good UV protection and antimicrobial activity against Gram-positive bacteria. Moses and Venkataraman [61] reported UPF values of 34 for untreated cotton, improving to 37–40 after pre-treatment. Jute fabrics pre-mordanted with double-mordant treatments reached UPF values of 35–38, showing a notable enhancement over non-mordanted samples [62].

In addition to antibacterial and UV protection properties, annatto-dyed fabrics displayed anti-odor and stain-resistant characteristics, which were consistently positive across multiple studies [59–61].

#### *True indigo dye*

Indigo has been used since ancient times to produce blue dyes, which are typically mordant-free and possess a range of bioactive properties, including anti-UV, anticancer, cytotoxic, antiseptic, and wound-healing effects. Additionally, indigo can support the treatment of infections as well as dermatological and respiratory conditions [72, 73].

Nine studies were included in this review, one of which was a book chapter. Cotton was the most commonly dyed material using indigo leaves or extracts, followed by polyester, silk, silk-polyester blends, and lyocell. Indigo dyeing produced shades from blue to gray. Tambi *et al.* [24] reported that combining indigo with other natural dyes, such as pomegranate and kumkum, can generate secondary hues. For instance, mixing indigo and kumkum at a 50:50 ratio yielded green, while a 75:25 ratio of indigo to pomegranate produced gray.

Shade and brightness were also influenced by post-treatments. Barani [64] found that indigo-dyed wool without silver treatment appeared blue, whereas silver pre-treatment or simultaneous treatment produced brown-yellowish or darker blue shades with differing chromaticity. This suggests that silver ions interact with indigo and alter its optical properties.

Although natural dyes can be challenging to extract and standardize, blending primary dyes or using different mordants provides a practical way to achieve a variety of colors while simplifying industrial applications [28]. Tambi *et al.* [24] observed that in binary dye mixtures, the component present in higher proportion predominantly determined the resulting shade, while variations in concentration affected hue and lightness.

Several pre-treatments were applied to enhance K/S values and color fastness, including AgNPs, chitosan, ZnONPs, sodium hydroxide, morpholine, cellulase enzyme, and hydrochloric acid. Generally, good K/S values were achieved. For instance, indigo-dyed wool without silver treatment exhibited lower K/S across visible wavelengths compared to silver-treated samples [64]. Silk and silk-polyester-lyocell blends pre-treated with hydrochloric acid showed strong light and wash fastness, though rub fastness remained low [74]. The typically high wash fastness of indigo-dyed fabrics is attributed to the insoluble nature of indigo, which forms covalent bonds with cellulose fibers, stabilizing the color. Simultaneous silver treatment improved light fastness (6–7 to 7–8 on the blue scale) while maintaining wash fastness (4–5 on the gray scale), using sodium dithionite as a reducing agent [64].

Sodium hydroxide treatment of cotton enhanced wash fastness (4–5) compared to morpholine, cellulase, or untreated fabrics (3–4 to 4) and also improved light and rub fastness, indicating greater durability [61]. Reningtyas *et al.* [63] reported that nanochitosan and ZnONPs applied sequentially as anti-UV agents increased ZnONP uptake by cotton up to fourfold, improving light fastness. Sodium dithionite and sodium carbonate were employed as reducing agents in these dyeing processes.

High-temperature, high-pressure (HTHP) dyeing of polyester achieved satisfactory fastness: light (5–6), wash (4), and rub (3–4), highlighting the role of temperature in polyester dyeing [24]. However, indigo-dyed fabrics generally display low rub fastness, as reported by Jeyaraj *et al.* [74], where wash and light fastness were good (4), but rub fastness remained between 1–2 (dry) and 2 (wet).

Regarding medicinal properties, six studies reported antibacterial activity and four studies indicated anti-UV effects. Quantitative antibacterial testing showed a bacterial reduction greater than 83 % [24]. In qualitative assessments, the bacterial inhibition zone (BIZ) for dyed chitosan-treated cotton ranged from 26–40 mm, while for silk and its blends, BIZ ranged from 36–42 mm [60, 74, 75]. Antibacterial activity was generally stronger against *S. aureus* than *E. coli*.

Moses and Venkataraman [61] found that untreated cotton dyed with indigo provided moderate UV protection (UPF 33). Pre-treatments with sodium hydroxide, morpholine, or cellulase only slightly improved UPF (36–38). In polyester, indigo dyeing significantly increased UPF from 42.97 (undyed) to 198.40, as indigo molecules penetrate fibers and block UV radiation [24].

Indigo-dyed fabrics also exhibited anti-odor and stain-resistant properties, confirmed in three of seven publications [59–61]. However, antioxidant activity of indigo-dyed polyester was notably lower compared to other dyed fabrics [24].

#### *Woad dye*

Three studies were included in this review, one of which was a book chapter. Various fibers were tested with woad, with cotton being the most frequently used, followed by wool, silk, and jute. According to the literature, woad dye produces shades ranging from blue to gray and can be combined with other natural dyes to create additional colors. For example, combining woad with weld and gall oak yielded a green color [21]. In this process, silk fabrics were first mordanted with alum, then dyed separately with weld and gall oak, and also in combination at different ratios to achieve green. Color outcomes were also influenced by the type of fiber; woad-dyed wool yarns tended to be darker blue than cotton yarns [36]. Basak *et al.* [3] reported that woad provides consistent blue



coloration on both cotton and jute with satisfactory fastness, although none of the selected studies conducted formal color fastness testing.

Sodium dithionite was highlighted as the preferred reducing agent in both traditional and industrial woad dyeing [36]. However, this chemical and its derivatives are significant environmental pollutants and pose health risks, including genotoxicity in human lymphocytes. Interestingly, woad was found to counteract these genotoxic effects due to its antigenotoxic properties.

The antibacterial potential of woad was assessed only in a study evaluating green fabrics created by overlapping different natural dyes with an alum mordant [21]. The results indicated that darker fabrics exhibited higher bacterial reduction, with 91–99 % inhibition of *S. aureus* in samples dyed with woad, gall oak, and weld combined. Conversely, samples dyed solely with woad and weld showed no bacterial reduction, suggesting that the antibacterial effect was attributable to gall oak rather than woad.

### Final discussion

Various mordants and treatments have been employed in textile dyeing to enhance color properties and achieve diverse shades, as summarized in **Table 2**. Eucalyptus and weld dyes can produce yellow tones with or without mordants such as alum or aluminum sulfate. When eucalyptus is mordanted with copper sulfate, the resulting color shifts to brown, whereas weld under the same treatment produces olive-green. Both madder and annatto dyes yield orange and red tones in unmordanted fabrics, but madder can be combined with auxiliary agents and treatments to generate additional colors such as pink, gray, brown, beige, and purple. Indigo-based dyes primarily produce blue and gray shades, but when combined with other natural dyes like pomegranate, kumkum, or gall oak, green hues can be obtained.

Shade variations are influenced not only by mordants and treatments but also by several other factors. The reviewed studies indicate that chromatic properties are affected by: i) the dyeing method employed, ii) dye and mordant concentrations, iii) dyeing temperature and duration, and iv) the type of fiber. These factors collectively determine the final color and fastness properties of natural dyeing.

**Table 2.** Color palette obtained from selected natural dyes on textile substrates as a function of auxiliary products and mordanting/post-treating agents (Compiled and systematized from recent literature)

Auxiliary product / Treatment	Eucalyptus	Weld	Madder	Annatto	True Indigo	Woad
<b>No mordant / untreated</b>	Yellowish-brown [40, 58]	Light yellow [43] / Beige [45]	Red [3] / Yellow–orange [56] / Pink [45, 49, 52] / Brown tones [13]	Orange-yellow [3, 76] / Yellow to red [58]	Blue [3, 24, 59–61, 64] / Gray [24]	Blue [3, 36] / Dark & light blue [36]
<b>Alum</b>	Golden yellow [40] / Darker shades [19]	Yellow [43]	Orange [51] / Pink [49, 50] / Light red [22]	–	–	–
<b>Aluminium sulfate</b>	Yellow [38, 39]	Light yellow [45]	Reddish-brown [13] / Red [45]	–	–	–
<b>Tin chloride</b>	Bright yellow [40]	–	–	–	–	–
<b>Iron sulfate</b>	Beige [40]	Brown [43]	Gray [49, 50]	–	–	–
<b>Copper sulfate</b>	Brown [40]	Olive green [43]	–	–	–	–
<b>Sodium carbonate</b>	–	Bright lemony	–	–	–	–

		yellow [43]				
<b>Citric acid</b>	–	Light creamy [43]	–	–	–	–
<b>Mimosa</b>	–	Yellow [44]	Red [55]	–	–	–
<b>Pomegranate</b>	–	–	Reddish orange [13]	–	Gray / olive green [24]	–
<b>Kumkum</b>	–	–	–	–	Green / bright blue [24]	–
<b>Gall oak</b>	–	–	Dark red [22]	–	–	Green [21]
<b>Gall oak + alum</b>	–	Creamy yellow [43]	–	–	–	–
<b>Quebracho</b>	–	–	Beige [50]	–	–	–
<b>Laccase</b>	–	–	Beige [50]	–	–	–
<b>Silver nanoparticles (AgNPs)</b>	–	–	–	–	Blue → dark blue [64]	–
<b>Chitosan– cyanuric chloride (Ch–Cy)</b>	–	Yellow [45]	Red [45]	–	–	–
<b>Plasma treatment</b>	–	–	Beige / pink [49]	–	–	–
<b>Cationization</b>	–	–	Pink → purple [49]	–	–	–
<b>Citric acid + dicarboxylic acids</b>	–	–	Orangish-brown [51]	–	–	–
<b>Alum + mimosa</b>	–	Yellow → dark yellow [44]	Reddish-orange / orange [55]	–	–	–
<b>Quebracho + alum</b>	–	–	Pink [50]	–	–	–
<b>Quebracho + iron sulfate</b>	–	–	Gray [50]	–	–	–
<b>Laccase + alum</b>	–	–	Pink [50]	–	–	–
<b>Laccase + iron sulfate</b>	–	–	Dark gray [50]	–	–	–
<b>Myrobalan / pomegranate + iron sulfate</b>	–	–	–	Dark red [62]	–	–
<b>Myrobalan / pomegranate + potash alum</b>	–	–	–	Light yellowish-red [62]	–	–

The exhaustion method emerged as the predominant technique for dyeing all the selected natural dyes. Although this method is more time-consuming than alternatives, it is favored because it allows for better control over key parameters such as temperature, dye concentration, processing time, and the amount of auxiliary substances used, which contributes to more consistent results.

All the natural dyes demonstrated antibacterial properties, although these effects tended to diminish over time and with repeated washing. The use of mordants and additional treatments, including plasma processing or essential oil applications, can help maintain and even enhance antibacterial activity. Many mordants themselves possess



medicinal properties, so their use not only improves dye fixation on fibers but also reinforces the intrinsic antibacterial effects of the dyes, resulting in textiles with stronger and more durable antimicrobial performance. In the fashion industry, the drive for novelty and change underscores the need for a diverse color palette. While some colors remain consistently popular, designers require access to a broad spectrum of shades to meet evolving trends. This review analyzed the potential of natural dyes to generate a wide range of colors. A systematic table of the studied dyes was created to provide a clearer view of the chromatic possibilities of each dye, which are often difficult to discern from individual publications. However, each natural dye exhibits unique characteristics that interact differently with various fibers, and the interplay of multiple variables—including dyeing methods, dye and mordant concentrations, temperature, and fiber type—makes standardization challenging. Consequently, optimizing dyeing processes is essential to reproduce consistent shades across multiple dye baths. Currently, much of the available information remains experimental and somewhat vague. At the same time, there is increasing interest in environmentally friendly alternatives to synthetic dyes. Natural dyes offer designers the opportunity to expand the chromatic possibilities in fashion without being restricted to a limited palette.

## Conclusion

Color plays a central role in the fashion and textile industries and strongly influences consumer purchasing decisions. Growing awareness of the environmental and health impacts of synthetic dyes is expected to drive increasing demand for natural alternatives.

Although natural dyes can be challenging to extract and often yield limited color ranges, strategies exist to overcome these limitations. One promising approach involves creating diverse shades from primary natural dyes in combination with various mordants. This strategy could simplify the dyeing process by focusing on only three primary dyes while allowing a broader color spectrum to be achieved.

This study demonstrated that the chromatic properties and antibacterial performance of six natural dyes—eucalyptus (*Eucalyptus globulus* Labill.), weld (*Reseda luteola* L.), madder (*Rubia tinctorum* L.), annatto (*Bixa orellana* L.), true indigo (*Indigofera tinctoria* L.), and woad (*Isatis tinctoria* L.)—are influenced by multiple factors. Mordants and additional treatments can improve colorfastness and antibacterial properties, positioning natural dyes as a sustainable and eco-friendly alternative to synthetic colorants.

The medicinal attributes of natural dyes, particularly antibacterial action and UV protection, are increasingly relevant in textile applications. Preserving these properties during dyeing highlights the importance of optimizing processes. However, the current literature shows gaps, especially regarding weld and woad dyes, indicating a need for interdisciplinary research exploring the synergy between color, medicinal benefits, and textile aesthetics. By addressing these gaps and refining dyeing techniques, natural dyes can become a viable, health-conscious option for the textile industry, supporting sustainability and consumer well-being.

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