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Dyeing of wool yarn with natural dyes of *Lactarius deliciosus* and *L. sanguifluus* from Turkey

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ABSTRACT

In recent years, the use of synthetic dyes and pigments in textile finishing companies has been discussed for their harmful effects on human health and environment. For this reason, the use of plant–based dyestuffs from the nature has begun to increase because of the biodegradability, non–toxicity, human health and waste water contamination. Natural dyes are substances synthesized by some plants, animals, lichens and fungi in nature. Fungal species, including lichenized fungi have been used as natural colorants in different parts of the world throughout the history. In this study, natural dyes were extracted from *Lactarius deliciosus* and *Lactarius sanguifluus* collected from Osmaniye province (East Mediterranean region) of Turkey. The adsorption UV–Vis spectra of the mushrooms were measured to examine major colorants, and FTIR analysis of natural dyes obtained from the mushrooms extracts was performed. According to the analyses, major colorants in the edible Lactarius species could be azulene and its derivatives. Wool yarn (for carpet) was dyed with these natural dyes by using different mordants. CIELab (L*, a*, b*, c* and h), color differences (ΔE) and color strength (K/S) values of dyed wool yarns were determined. According to the dyeing results, cream and brown colors were obtained from *L. deliciosus* and *L. sanguifluus* respectively; the use of mordant (ferrous sulfate) increased the color strength of dye goods. Besides, properties of rubbing and washing fastness were investigated, and the results of the dyed yarns were low/moderate. This is the first study on dyeing of wool yarns with natural dyes obtained from *L. deliciosus* and *L. sanguifluus* collected from East Mediterranean (Osmaniye province) of Turkey.

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Natural dyeing, Russulaceae, mushroom, mordanting, color measurement

1. INTRODUCTION

Natural pigments and dyes, isolated from different natural sources, have attracted people's attention since ancient times [1–3]. Fungal species containing special natural dyes are widely used as colorants in Europe, America and North Africa [4,5]. Fungal dyestuffs and pigments can be isolated from a wide variety of fungal species and divided into two sections (carotenoids, bensole derivative (I) and quinones, anthraquinones, azulenes, heterocyclic nitrogen–bearing pigments (II)) [6–8]. These products are among the natural chemicals for use in pharmaceutical, food, cosmetic and textile industries [9].

Many dyes found in fungi, are benzoquinone derivatives, especially terpenylquinone compounds [10]. In earlier studies, red colour from *Echinodontinum tinctorum* were obtained by Indians living in America and some species of the genus *Boletus* were also used for painting of furs [11,12]. In Europe, fungi were used as textile colorants [13]. It was known that some species which belong to the genera *Telephora* sp., *Hydnellum* sp., *Sarcodon* sp. and *Phelledon* sp. containing benzoquinone derivatives were expressed to give blue hues to wool yarns [4,10,13,14]. Further on, some colorant components such as atromentin, polyphoric acid, telephoric acid, grevillin, physcion, emodin, hispidin, anthraquinones, iminoquinone, dermoquinone, dermocybin, pulvinic acid, norbadione A, laetiporic acids were found in *Boletus* sp., *Xerocomus* sp., *Polyporus* sp., *Trametes* sp., *Hydnum* sp., *Suillus* sp., *Dermocybe* sp., *Paxillus atrotomentosus*, *Sarcodon squamosus*, *Hapalopilus nidulans*, *Cortinarius sanguineus*, *Pisolithus arhizus*, *Laetiporus sulphureus* [15–21].

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The genus *Lactarius* belonging to the family Russulaceae (Basidiomycota) has more than one thousand species [44]. *L. deliciosus* and *L. sanguifluus* (kanlıca, çıntar or çam mantarı in Turkish) are often used as food among local people in Turkey.

Previous studies found different pigments such as red colour 7–acetyl–4–methylazulene–1–carbaldehyde, 7–(1,2–dihydroxy–1–methylethyl)–4–methylazulene–1–carbaldehyde, 7–acetyl–4–methylazulene–1–carboxylic acid in *L. deliciosus* [22] and as beige colour (azulene) in *L. deliciosus* [23], 1,3,5,7(11),9–pentaenyl–14–guaianal) in *L. sanguifluus* [24], brilliant blue colour (1–hydroxymethyl–4–methyl–7–(1–methylethenyl) azulene) in *L. indigo* [25–28], pale green colour aminobenzoquinone blennione [29]. Considering related publications, major colorants in the edible Lactarius mushrooms may be azulene and their structural formula are given in Figure 1.

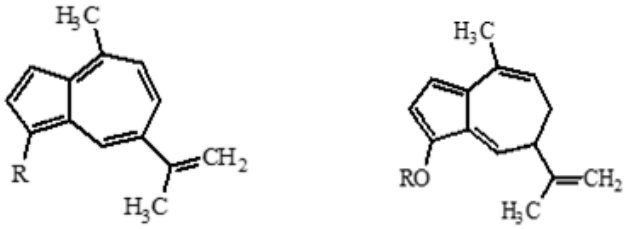

Figure 1. Chemical structure of azulene (lactarazulene (R =CH3), dihydroazulen–1–ol (R =H))

Mycologists have so far identified about 120.000 fungal species [30], but this number increases day by day with the detection of new species [31-36]. However, the number of fungal species in the world is estimated to be 2.2-3.8 million [37]. When examined ecologically, mushrooms serve as an interesting source of raw materials and have faster growth potential and high yields in a short time than higher plants [38]. Thus, mushrooms are more suitable and economical for new technologies such as biotechnological products [39-40].

Natural dyes are mostly eco-friendly, biodegradable, less toxic, and less allergenic as compared to synthetic dyes. In spite of many advantages of natural dyes, their short range of shades, non-availability of standard shade cards and low fastness results have been improved by using metallic mordants. Natural dyes are mostly non-substantive and must be applied on textiles by the help of mordants, usually a metallic salt, having an affinity for both the coloring matter and the fiber. After combining with dye in the fiber, these metallic mordants turn into an insoluble precipitate or lake; hence, both the dye and mordant get fixed to become wash fast to a reasonable level [41,42].

Recently, eco–friendly natural dyes have attracted the attention of many researchers and the use of these dyes is increasing day by day in many products such as textiles, cosmetics and food [43–49]. However, uses of fungal species for dyeing textiles have not been investigated enough until today. In the present study, natural dyes obtained from *L. deliciosus* and *L. sanguifluus* were studied on dyeing of wool yarns.

2. EXPERIMENTAL

2.1. Material

2.1.1. Plant material (Mushroom samples)

L. deliciosus and *L. sanguifluus* (voucher no: FBozok222 and FBozok223, respectively) were collected from Osmaniye province (37°01'15" N, 36°13'53" E, 571 m) of Turkey (Figure 2).

Figure 2. *L. deliciosus* (left) and *L. sanguifluus* (right)

2.1.2. Studies on wool

Straygarn woolen yarns, which is %100 pure (Yarn count: Nm 4/2, Twist per meter: 256 t/m), were supplied from Karatepe Kilim Cooperative in Osmaniye, Turkey. Wool used to dye was obtained from sheep which are native to the Eastern Mediterranean region. The hank samples (each 3 g) were produced using hank winder for dyeing.

2.1.3. Chemicals

The metallic salts employed were alum ($KAl(SO_4)_2.12H_2O$) and ferrous sulphate heptahydrate (Fe_2SO_4, $7H_2O$) for pre-mordanting. Acetic acid ($C_2H_4O_2$, glacial) was used to adjust the pH. Mushroom samples were extracted in ethanol. Nonionic detergent (Setalan HE, Setas–Chem.) was utilized to wash after dyeing. All chemicals were of laboratory grade.

2.2. Methods

2.2.1. Natural dye extraction

Fresh mushroom samples were dried in dehydrator for 48 h at 50 °C and pulverized by using a blender (Waring Blender, HGB2WTS3). 50 g powder from both mushroom samples were extracted in 500 ml of ethanol during 24 h at 50 °C and filtered by Whatman filter paper. Ethanol was then removed by using a rotary evaporator at 78 °C. Natural colorants obtained from both mushroom samples were preserved in a refrigerator (+4 °C) until used.

2.2.2. Spectrophotometric analysis

To examine absorbance properties of *L. deliciosus* and *L. sanguifluus* crude extracts, the adsorption UV–Vis spectra were measured and the obtained spectra in full UV–Vis range are presented in Figure 3. The absorption spectra of natural dyes from *L. deliciosus* and *L. sanguifluus* in the present study found to be between 250 and 400 nm for azulene and its derivatives as in the study of Matênovâ et al. (2014) [50]. It is thought that major colorants in the edible Lactarius species could be azulene and its derivatives.

2.2.3. FTIR analysis

An FTIR (Fouirer Transform Infrared Spectroscopy, PerkinElmer Spectrum 65) that have Universal ATR (attenuated total reflectance) sampling accessory was used and IR spectra were obtained in the 600–4000 cm^{-1} (Figure 4). Measurements from each sample were done 5 times in the room temperature (25 ^{o}C). It was known that the absorbance of azulene was about 760 cm^{-1} in FTIR [45].

2.3. Mordanting and Dyeing

Pre–mordanting method was applied to study and the wetted wool yarns were mordanted by using 3 % owf of ferrous sulfate and potassium aluminum sulfate mordants at 80 °C and liquor ratio 1:50 for 45 minutes. After mordanting, each sample was not rinsed but only squeezed.

The wool yarns used in the production of carpets and rugs were dyed with natural dyes extracted in the laboratory. Different dyeing concentrations (1, 2 and 3 g/l) and mordant materials (Ferrous Sulphate and Potassium Aluminum Sulfate) were used for dyeing wool yarns. 150 ml of original dye solution was used for each a hank sample at liquor ratio 1:50. Acetic acid was used to adjust the pH of the dyebath to 5. Dyeing was done at 80 °C for 60 min. The dyed wool yarns were then washed with 2 g/l nonionic (Setalan HE, Setas–Chem.) detergent for 30 min at 80 °C, rinsed with tap water and dried at room temperature in shade. Laboratory type dyeing machine was used in all dyeing processes.

2.4. Color Measurement

Swatch cards were made by using yarn sample winding machine for color measurement (spectrophotometer). A spectrophotometer (Minolta CM 3600D) coupled to a PC was used to measure the color coordinates of dyed yarn between 400–700 nm under D65/10° illuminant. K/S (Color Strength) values were obtained from Kubelka Munk Equation (1);

$$K/S = (1–R)^2/2R \quad (1)$$

where K is a constant associated with the light absorption of the fabric, predominantly determined by dyestuff, S is the constant of light scattering of the fabric, determined only by the textile material, and R is the reflection of the dyed fabric measured at the wavelength of maximum light absorption. The CIE L*a*b* formula is used to assess small color differences and recommended for use by DIN 6174. The color difference is determined using a color difference formula from the colorimetric measures L*, a*, b* which result from the CIE tristimulus values X, Y, Z. In the CIE L*a*b* uniform color space, the coordinates are:

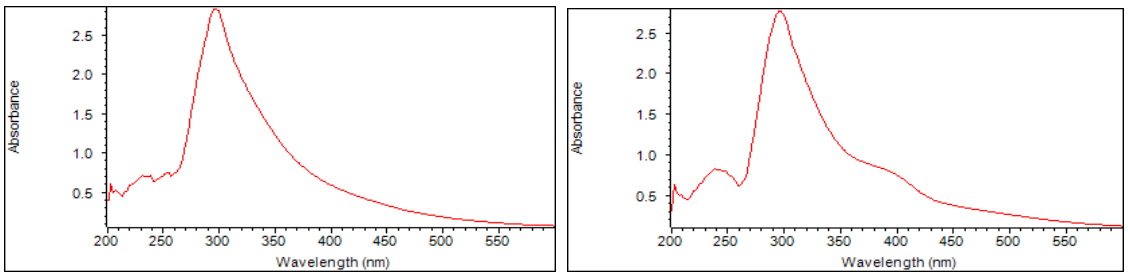

Figure 3. The absorption UV–Vis spectra of *L. deliciosus* (left) and *L. sanguifluus* (right)

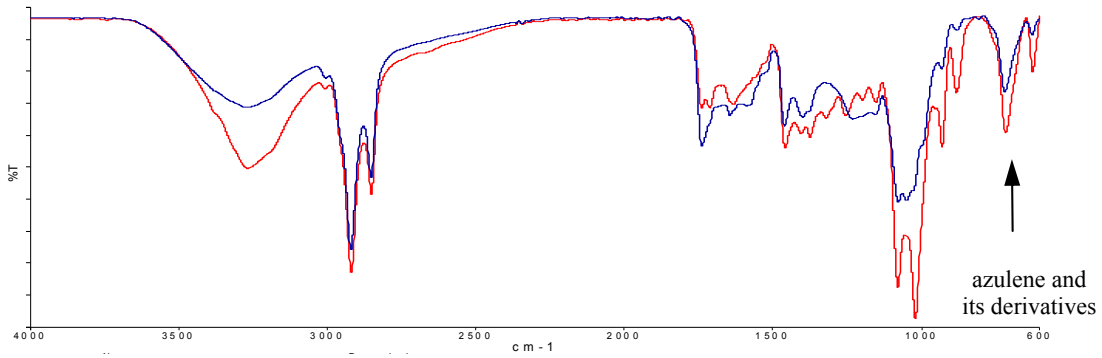

Figure 4. FTIR analysis of natural dyes obtained from *L. deliciosus* (blue) and *L. sanguifluus* (red) extracts

L* - the lightness coordinate.

a* - the red/green coordinate, with +a* indicating red, and -a* indicating green.

b* - the yellow/blue coordinate, with +b* indicating yellow, and -b* indicating blue

According to DIN 6174, the ΔE*ab color difference is calculated as Equation (2) [51,52]**:**

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

$\Delta L^* = L_i^* - L_o^*$ (light difference)

$\Delta a^* = a_i^* - a_o^*$ (red-green difference)

$\Delta b^* = b_i^* - b_o^*$ (yellow-blue difference)

ΔE^*_{ab} : total color difference

i: Sample o:Reference

2.5. Fastness Properties

The washing and rubbing (crock) fastnesses of the naturally dyed yarn samples were determined according to TS EN ISO 105-C06 A1S (steel balls are not used) and TS EN ISO 105–X12 standards after conditioning in standard atmospheric conditions (20ºC±2 temperature and 65 ± 4 % RH) for 24 hours. The wash fastness of the dyed samples was assessed both in terms of alteration of shades and degree of staining on white cotton adjacent fabric. Dry and wet rub fastness of dyed wool yarn samples was tested using the Crockmeter. Washing and dry cleaning fastness machine was used to measured wash fastness values of dyed wool yarn samples. The results were evaluated with the gray scale values (rating 1-5, 1= poor, 5=excellent) for washing fastness and rubbing fastness.

3. RESULTS AND DISCUSSION

As a result of color measurements, brown shades of color were obtained by using two various mordants at different concentrations (1, 2 and 3 g/l) for dyeing with *Lactarius deliciosus* and *Lactarius sanguifluus*. Color measurements on spectrophotometer are given in Table 1–4. Cream–its shades and brown–its shades in wools dyed with extracts of *L. deliciosus* and *L. sanguifluus* were obtained for dyeing without mordant (Table 1 and 2). In the use of ferrous sulphate mordant ($FeSO_4$), darker colors (dark brown) in *L. deliciosus* and brown in *L. sanguifluus* were obtained with high K/S values (Figure 5), while light brown and its shades with *L. deliciosus* extract and cream and its shades with *L. sanguifluus* extract were obtained with aluminum potassium sulphate ($KAl(SO4)_2.12H_2O$) respectively. Lightness (L*) values decreased while red/green (a*) values in wool dyed with *L. deliciosus* extract increased with the increase in concentration and the use of mordant.

Color characteristics are highly affected by the type of mordant used [42]. In Figure 5, it is observed that while the K/S values of dyed samples with Alum and unmordanted increased depending on concentration, the K/S values of dyed samples with ferrous sulfate did not change much for both mushrooms extracts. In another result, *L. sanguifluus* K / S values are lower than other *L. deliciosus* for dyeing with mordant. While it was found that K/S values increased in the order of ferrous sulfate > alum >unmordanted for *L. deliciosus* extract, and they increased in the order of ferrous sulfate > unmordanted > alum for *L. sanguifluus* extract. It is remarkable that the woolen yarns dyeing with *L. deliciosus* extract using mordant have higher K/S values. This situation is thought to be related to the chemical structure of two different fungi belonging to the genus Lactarius. In addition, the high K / S values obtained from pre-mordanting with ferrous sulphate can be explained by the fact that azulene acts as a ligand for low-value metals such as ferrous and can especially form stronger chemical bonds in organometallic chemistry.

Table 1. Colour measurements of wool dyed with extracts of *L. deliciosus* without and with mordanting

Dyeing		Color measurements							
Mordant	Con. (g/l)	L*	a*	b*	C*	h	K/S		Color
Without	1	60.83	05.81	22.86	23.59	75.74	03.87	Cream	
	2	55.50	07.33	23.07	24.20	72.37	05.32	Ivory	
	3	53.28	07.96	22.94	24.28	70.85	06.06	Grayish Brown	
Potash alum	1	58.24	08.12	25.51	26.77	72.35	05.18	Fawn (Fulvous)	
	2	52.50	08.76	25.24	26.71	70.86	07.48	Light Brown	
	3	46.70	09.21	23.72	25.44	68.78	09.92	Brown	
Ferrous Sulphate	1	46.83	15.19	31.44	34.91	64.21	16.68	Bombay Brown	
	2	41.80	09.97	23.29	25.34	66.82	16.62	Dark Brown	
	3	41.36	09.56	22.18	24.15	66.69	16.14	Dark Brown	

Con.: Concentrations

Table 2. Colour measurements of wool dyed with extracts of *L. sanguifluus* without and with mordanting

Dyeing		Color measurements							
Mordant	Con. (g/l)	L*	a*	b*	C*	h	K/S		Color
Without	1	62.58	6.59	18.73	19.85	70.60	04.19	Grayish Brown	
	2	54.88	8.88	21.66	23.41	67.70	07.36	Brown	
	3	47.13	9.13	18.43	20.57	63.65	10.44	Dark Brown	
Potash alum	1	63.96	5.36	19.74	20.45	74.80	03.64	Cream	
	2	59.82	7.07	20.60	21.78	71.04	05.27	Cream	
	3	56.48	7.45	20.59	21.90	70.12	06.54	Fawn (Fulvous)	
Ferrous Sulphate	1	49.04	8.68	23.37	24.93	69.62	09.82	Fawn (Fulvous)	
	2	48.92	9.64	24.17	26.03	68.25	10.98	Brown	
	3	48.83	9.70	24.47	26.32	68.37	11.07	Brown	

Con.: Concentrations

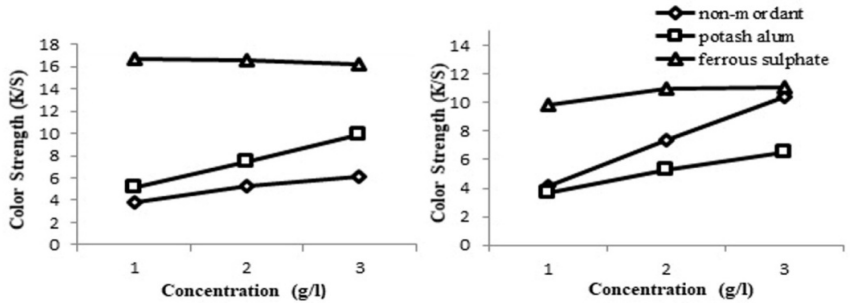

Figure 5. The effect of different mordants on color strength of *L. deliciosus* (left) and *L. sanguifluus* (right) extracts

ΔL*, Δa*, Δb* values and the total color difference (ΔE) can help to explain how the mordant changes color. The samples of dyeing without mordanting were taken as a reference for comparison. Total color difference (ΔE) values are given for three different concentrations in Table 3 and 4. Table 3 and 4 indicate that there are significant color differences between the results of dyeing without and with mordanting. As shown in Table 3, dyeing samples with extract of *L. deliciosus* are dyed darker, redder and more yellow for most concentrations. While dyeing samples with extract of *L. sanguifluus* are dyed darker, redder and more yellow for ferrous sulphate mordant, samples of *L. sanguifluus* are dyed lighter, greener and more yellow for potash alum mordant for all concentrations in Table 4. The red/green (a*), yellow/blue (b*) and croma (C*) values increased depending on dyeing concentration for wool dyed with *L. sanguifluus* extract, while lightness (L*) values decreased.

In a previous study, anthraquinone dyestuff (60g anthraquinone powder from 10 kg fresh mushroom) was obtained in fruiting bodies of *Cortinarius sanguineus* and *C. semisanguineus* collected from Kuopio and Tuusula region of Finland, respectively [53] and natural and synthetic fibres were dyed with pure emodin and dermocybin found as major compounds in both *Cortinarius* species, as well as tap water extract. It was found that natural anthraquinones which produced light and dark colors such as yellow, orange and brown, could be suitable in dyeing and printing different textile materials. Maldonado and Ibarra [23] stated that *L. deliciosus,* which contained lactarazulene, lactaroviolin, has blue, reddish crystal, violet pigments and that extract of this mushroom painted the yarns in beige color by using different mordants (iron sulfate, tin chloride, potassium dichromate, alum and copper sulfate mordants).

K/S values increase, and L values decrease with the use of ferrous sulfate mordant. In case of ferrous sulphate mordant, brownish black shades are observed due to reacted with oxygen in air and light. The ferrous and ferric forms are found together on the fiber and their spectra overlap that alter λ_{max} and the dyed fibers appear darker [54]. In other words, iron sulfate can form stable complex structures with both wool fibers and natural dyestuff such as "dihydroazulen–1–ol (R=H).

The wool fibre morphology, chemical constituents and its complex histological structure play a major role in dyeing. Wool mainly consists of amino acid units, which is composed of free amino and carboxyl groups. During the dyeing process, auxochrome groups of dyestuff bind with amino groups of the fibers via hydrogen bonding. Azulene is an organic compound. The organic compounds of the mushrooms (Figure 1) show dyeing properties for wool fibers due to their chromogen group such as benzene ring. Therefore, it is expected that chemical interactions between the dyestuffs in the mushrooms and the yarns occurred between $-R=CH_3$ or $-R=H$ groups of the dye molecule and functional groups of the fabrics via H-bonding. When the different dyes are combined with the mordant to form dyed-mordant complexes, different shades are than attained under different concentrations, and fastness such as washing and rubbing of many natural dyes could be improved by treatment with definite metal ions [55].

Rubbing and washing fastness values of dyed wool yarns with and without mordant are given in Table 5. Rubbing fastness is found to be in range of 2-4/5. All mordanted wool yarns and dry rub fastness are found to have better fastness values than non–mordanted and wet rub fastness, respectively. When comparing rubbing fastness, similar values were observed for *L. deliciosus* and *L. sangiufluus*. Washing fastness is also found to be in range of 2-4/5. The obtained staining washing fastness is good and the same as each other. But the changing washing fastness is bad/moderate. In addition, it can be said that the use of mordant changes slightly the washing fastness values for *L. sangiufluus* and *L. deliciosus*.

In fact, the metal complexes were easily formed with the fibers, and this could create affinity between natural dye and wool fiber [56]. Low washing and wet rubbing fastness values might to be due to the increased size of dye molecules when mordant molecules bind to the fiber and because water molecules removed some water–soluble dyes by the action of rubbing and washing conditions [57, 58].

4. CONCLUSION

Due to decreasing fossil resources in the world, increasing sensitivity to ecological and human health, natural dyes have become increasingly popular. Natural pigments and dyes which extracted from different natural resources such as plant, animal and fungal species since ancient times are used in pharmaceutical, food, cosmetic and textile dyeing industries. In this study, natural dyes from *L. deliciosus* and *L. sanguifluus* collected from Osmaniye province of Turkey were extracted and wool dyeing properties of these natural dyes were investigated. It has been indicated that cream and brown colors can obtained from natural dyestuff extracted from two different fungal species (*L. deliciosus* and *L. sanguifluus*). According to the results of fastness tests, dry rubbing fastness values were high, but wet rubbing and washing (changing) fastness values were low. As a result, these colors can be used easily in textile field and especially in natural dyeing for traditional textile products such as carpets and rugs which are not washed often. It is a fact that commercially natural dyes cannot meet the needs of textile industry in the world and the natural dyes obtained from plants have still some drawbacks. Thus, the papers published associated with the use of bio-colorants show that the poor reproducibility, time-consuming extraction methods, insufficient degree of fixation and low color fastness properties are the main problems ahead the natural dyers to use this products in textile finishing industry. However, considering factors such as health and environment, it has been revealed that natural dyes can be an alternative to synthetic dyes. The study on the mushroom may be beneficial in terms of raising awareness. In particular, babies and elderly people should prefer textile apparel dyed with natural dyes for the sake of health. In addition, this study revealed that some mushrooms that are usually consumed as food and grow in many areas of Turkey can be used for natural dyeing. Fungi that are not used in nature are important for sustainability, because they can be easily used as a dyestuff in homes and cooperatives. In further studies, it is necessary to identify each natural dyestuff in different fungal species and to investigate their staining properties on wool yarns.

Acknowledgement

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Table 3. Comparison of difference color measurement values for natural dyeing with *L. deliciosus* extracts

Reference	Con. (g/l)	Samples Pre-mordanting	ΔL*	Δa*	Δb*	ΔE	Color comparison
Dyeing without mordanting	1	Potash alum	–2.59	2.31	2.65	04.36	Sample is darker, redder, more yellow
		Ferrous sulp.	–13.99	9.38	8.58	18.90	Sample is darker, redder, more yellow
	2	Potash alum	–3.00	1.43	2.17	03.97	Sample is darker, redder, more yellow
		Ferrous sulp.	–13.70	2.64	0.23	13.96	Sample is darker, redder, more yellow
	3	Potash alum	–6.58	1.24	0.78	06.74	Sample is darker, redder, more yellow
		Ferrous sulp.	–11.92	1.59	-0.76	12.05	Sample is darker, redder, more blue

Con.: Concentrations

Table 4. Comparing of color difference measurement values for natural dyeing with *L. sanguifluus* extracts

Reference	Con. (g/l)	Samples Pre-mordanting	ΔL*	Δa*	Δb*	ΔE	Color comparison
Dyeing without mordanting	1	Potash alum	1.38	–1.23	1.01	02.11	Sample is lighter, greener, more yellow
		Ferrous sulp.	–13.54	2.09	4.64	14.47	Sample is darker, redder, more yellow
	2	Potash alum	4.94	–1.81	–1.07	5.37	Sample is lighter, greener, more blue
		Ferrous sulp.	–5.96	0.76	2.51	6.51	Sample is darker redder, more yellow
	3	Potash alum	9.35	–1.68	2.16	9.75	Sample is lighter, greener, more yellow
		Ferrous sulp.	1.70	0.57	6.04	6.30	Sample is darker, redder, more yellow

Con.: Concentrations

Table 5. Rub and wash fastness properties of dyed samples

Mordant	Con. (g/l)	*Lactarius deliciosus*				*Lactarius sanguifluus*			
		Rub Fastness		Washing Fastness		Rub Fastness		Washing Fastness	
		(dry)	(wet)	Changing	Staining	(dry)	(wet)	Changing	Staining
Without Mordant	1	4	2/3	3	4/5	4	3	3	4/5
	2	4	2/3	3	4/5	3	2/3	3	4/5
	3	3/4	2	3	4/5	3	2/3	3	4/5
Potash alum	1	4/5	4	2	4/5	4/5	4/5	3	4/5
	2	4/5	3/4	2	4/5	4/5	4/5	3	4/5
	3	4/5	3/4	1/2	4/5	4	4	3/4	4/5
Ferrous sulphate	1	4/5	4/5	2/3	4/5	4/5	4/5	3	4/5
	2	4/5	4	2/3	4/5	4/5	4	3	4/5
	3	4/5	4	3	4/5	4/5	3/4	3/4	4/5

Con.: Concentrations

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