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# ANTIBACTERIAL AND ANTIOXIDANT PROPERTIES OF SILK FABRIC DYED WITH *CICHORIUM INTYBUS* ROOT EXTRACT

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Natural dye, *Cichorium intybus*, Antibacterial, Antioxidant, Mordant, Dyeing, DPPH

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**ABSTRACT:** The present study was conducted to investigate the color strength (K/S), colorfastness, antibacterial and antioxidant properties of silk using fabric *Cichorium intybus* root extract as a natural dye. Comparative results of color fastness (fastness to wash, rub, light and perspiration) and colorimetric properties (CIELab and K/S values) of dyed silk samples were studied to quantify the effect of mordants and mordanting method. Alum and myrobalan were used as mordants. Three methods of mordanting techniques were used (pre, meta and post-mordanting). The samples showed acceptable color strength and colorfastness values. The antibacterial activity of the dyed samples with and without mordants was tested against common pathogens *Escherichia coli* and *Staphylococcus aureus*. The antioxidant property of the dyed fabric was analyzed by DPPH method. *Cichorium intybus* proved to be very effective in inhibiting microbial growth. The results of DPPH assay were positive for all the samples. The dyeing of silk fabric with *Cichorium intybus* root extract provided a multifunctional textile material with both antioxidant and antibacterial properties.

**INTRODUCTION:** Value addition in clothing has changed the global textile scenario [1]. Research has quite convincingly shown that apparel consumers all over the world are demanding functionality in the products [2]. The wellness or health-promoting aspects of textile finishes have become a delightful functional matter in the 21st century. The extracts of natural products and selected essential oils are added to textiles, which not only have healing properties but also keep the wearer fresh and vigorous [3]. The market for cosmetotextiles and medical textiles has greatly expanded in recent years.

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As textile materials have found applications in the cosmetic field, more and more commercial cosmetic textile agents are now available in the market [4]. Plants are the basic source of knowledge of modern medicine. Almost all the parts of the plant, namely leaves, flowers, fruits, roots, stem, and seeds are known to have various medicinal properties including antibacterial and antioxidant properties [5].

*Cichorium intybus* L. (Asteraceae) as an important medicinal herb has been used in folk medicine for liver disorders, gallstones and inflammations of the urinary tract since the 17th century. The plant was used traditionally for the treatment of diarrhea, to strengthen the prostate and other reproductive organs, for the treatment of pulmonary disease and cough, cancer, hangover, for purification of biliary tract, liver complaints, as spasmolytic, to relief of symptoms related to mild digestive disorders (such as feeling of abdominal fullness, flatulence, and

slow digestion) and temporary loss of appetite. It is also reported to be used in the treatment of sore throat, hemorrhoids, tuberculosis, abdominal cramps, melancholy, deafness, rashes and as a laxative for children [6]. Chicory is commercially used as a vegetable, fresh or cooked, while the ground and roasted roots are widely used for blending with coffee powder. It is well-known from the literature that the main active compounds of chicory are: inulin, fructooligosaccharides, caffeic acid derivatives, flavonoids and polyphenols [7].

*Cichorium intybus* L. has been investigated previously to assess the content of phenolic compounds, mainly cinnamic acids and flavonoids [8]. Based on the studies carried out in chicory, worldwide report shows that the roots and leaves of this plant possess strong antibacterial and antioxidant properties [9, 10, 11]. However, no report exists so far on the extraction of natural dye from *Cichorium intybus* L. root and its application on silk fabric. The present study was undertaken to investigate the antibacterial and antioxidant property of the silk fabric dyed with *Cichorium intybus* root extract.

## MATERIALS AND METHODS:

**Materials:** Plain weave silk fabric, having specifications 47 g/m², 130ends/inch and 82 picks/ inch, was used for the study. The roasted chicory root powder was purchased from the market.

**Extraction of Color Component:** The extraction was carried out under an alkaline condition with MLR 1:40 at pH 10 for 90 min at a temperature of 90 °C. The hot solution was then filtered through a Whatman no.1 filter paper to obtain a clear filtrate. The filtrate was evaporated to produce crude which was further dried for 24 h and ground manually using mortar and pestle to form a powder.

**Selection of Mordant:** Considering the eco-friendliness and cost-effectiveness, the natural mordant myrobalan [12] and metallic mordant alum [13] were chosen for the dyeing process.

**Dyeing and Mordanting of silk fabric:** The dyeing was carried out at 90 ºC with the dye bath pH 5 containing 4 % of dye in crude, based on the weight of fabric (owf) at MLR 1:40 in a temperature controlled open bath dyeing machine for 60 min. The fabric samples were pre, meta and post mordanted using 4% (owf) solutions of each of potassium aluminium sulfate and myrobalan separately with MLR 1:40 for 30 min at 60 ºC. The fabrics were subsequently rinsed and dried. The dyed samples were subsequently washed in 1 GPL non-ionic detergent solution at 60 ºC for 15 min and dried at room temperature.

**Color Value Measurement:** Color was evaluated using K/S and CIELAB values with illuminant D65/10º observer on Gretag Macbeth Color Eye 7000 A Spectrophotometer. Four measurements were made for each sample, and the variation in percentage reflectance values over a range of 350–750 nm was recorded. The K/S values were assessed using the Kubelka-Munk equation.

$$K / S = (1-R)^2 / 2R$$

Where, R is the observed reflectance, K is the absorption coefficient, and S is the light scattering coefficient [14, 15, 16].

**Fastness Tests:** Color fastness properties of the samples were assessed using ISO standards. Colorfastness to washing of dyed fabric was determined as per ISO 105-C06 test method. Colorfastness to crocking/ rubbing was assessed as per ISO105-X12 test method. Colorfastness to light was determined as per AATCC 16.3:2012 method and Colorfastness to perspiration were assessed as per ISO 105-E04 1994 method [15, 16, 17, 18].

**Determination of Antibacterial Activity of Dyed Fabrics:** The antibacterial activity was assessed by qualitative testing of fabric using Antibacterial Activity Assessment of Textile Materials: Parallel Streak Method (AATCC Test Method 147-2004) [19]. *Escherichia coli* (*E. coli*), a gram-negative bacterium and *Staphylococcus aureus* (*S. aureus*), a pathogenic gram-positive bacterium were selected for the study [20, 21] as they are recommended in most test methods [22, 23]. Briefly, the test strains were inoculated in sterile nutrient broth and were allowed to grow for 18 h at 37 ºC. Sterile nutrient agar plates were prepared for each test strain and one loop (4 mm in size) full of overnight grown bacterial culture was inoculated on the surface of nutrient agar plate by making five parallel inoculum streaks in such a way that it should cover the central area of the Petri dish without refilling the loop of the same inoculum.

Then *Cichorium intybus* treated fabrics were placed transversely across five inoculum streaks made earlier on the plate. The fabrics were pressed gently using a sterile spatula (flame sterilization) to ensure that *Cichorium intybus* treated fabrics have intimate contact with nutrient agar surface. Simultaneously, the plate containing untreated fabrics was also maintained as control, and the plates were incubated for 24 h at 37 ºC. At the end of the incubation, a clear area showing no growth along the sides of *Couroupita guianensis* treated fabrics indicates the antibacterial activity, and then ZOI was calculated. The average width of the inhibition zone on either side of the *Cichorium intybus* treated fabrics along the streak was calculated using the following equation,

$$W = (T-D) / 2$$

Where, W = Width of clear zone of inhibition (mm); T = Total diameter of test specimen and the zone of clearance (mm); D = Diameter of the test specimen (mm).

**Determination of Antioxidant Activity (DPPH Assay):** Radical scavenging activity of the dyed cotton fabric was determined by measuring the decrease in absorbance of 2, 2-Diphenyl-1-picrylhydrazyl radical (DPPH$^{\bullet}$) at 517 nm [24]. The DPPH assay is a widely used method to evaluate the ability of antioxidants [25] to scavenge free radicals which are known to be a major factor in biological damages caused by oxidative stress. This assay is known to give reliable information concerning the antioxidant ability of the tested compounds [26, 27, 28]. This method is based on the ability of DPPH radical to react with hydrogen donor species such as phenolics and flavonoids present in the extracted material. Upon receiving a proton from the donor species, it loses its color and becomes yellow.

As the concentration of phenolic compounds increases, their DPPH radical scavenging activity also increases [29]. The fabric samples were kept immersed in DMSO for 1 h at MLR 1:10. The DMSO solution was then filtered and collected from each fabric sample. The solution was evaporated to dryness, and the crude collected was weighed and dissolved in DMSO (10mg/ml). This was considered a pure sample extract for testing the antioxidant property. 0.3mM solution of DPPH was prepared in 100% methanol. To 1 ml of this solution, three different concentrations 100 µl, 300 µl, and 500 µl of sample extract and standard solution (Ascorbic acid) were added separately. The final volume was made up to 4ml by adding 100% methanol to each sample mixture and also for a standard solution (Ascorbic acid). The same reaction mixture without the extracted sample but with an equivalent amount of standard phosphate buffer was taken as control. All the sample mixtures and control were shaken thoroughly and kept in the dark at room temperature for 30 min. The absorbance of the reaction mixtures was measured at 517 nm. The radical scavenging activities were expressed as a percentage of inhibition and calculated according to the following equation.

$$\% \text{ DPPH inhibition} = [(AC-AS)/AC] \times 100$$

Where, AC = absorbance of control and AS = absorbance of test sample [30, 31].

## RESULTS AND DISCUSSION:

**Color Characteristics:** K/S values of the samples indicate that the concentration and kind of mordant and mordanting methods significantly affect the lightness, shade and color yield [25]. Results concerning color depth (K/S values) of silk fabric dyed with fruit extract of *Cichorium intybus* obtained with and without the use of mordants are given in **Table 1**. From the results, it is clear that both mordants and mordanting methods influence the color strength values.

All the mordanted samples show higher dye uptake than that of the unmordanted (control) sample. Maximum color strength of K/S (15.14) was obtained with sample pre-mordanted with myrobalan, whereas the unmordanted (control) sample showed the lowest K/S value of 7.36. The process of dying with different mordants gives a shade change from light to golden brown. Myrobalan mordanted samples showed higher color strength valued compared to alum mordanted samples. Varied hues of color are obtained from pre, meta, and post-mordanting with alum and nyrobalan.

**Fastness Properties:** The evaluation of color fastness to washing, rubbing, light and perspiration using alum: pre, simultaneous and post-mordanting

method and myrobalan: pre, simultaneous and post-mordanting method is presented in **Table 2**. All the alum mordanted samples including control sample which were subjected to washing fastness showed fair color fastness to color change and poor to fair color fastness to staining. The myrobalan mordanted samples have shown good to fairly good color fastness to color change and color staining. The dry and wet rubbing fastness grades for control and alum mordanted samples were 2-3 (poor to fair) and grades for myrobalan mordanted samples were in between 4 and 3-4. The light fastness of control and alum mordanted samples was very poor to poor, whereas the myrobalan mordanted samples showed fairly good to fair fastness. In case of perspiration fastness, the grades for color change in both acidic and alkaline media for control and all alum mordanted samples were fair to fairly good (3 to 3-4), and grades for color staining were 2-3 (poor to fair). All the myrobalan mordanted samples showed better color fastness properties compared to control and alum mordanted samples.

**TABLE 1: K/S AND COLOR CO-ORDINATES OF SILK FABRIC DYED WITH *CICHORIUM INTYBUS* ROOT EXTRACT**

| Sample | Mordanting Method | K/S | L* | a* | b* | c | H |
|---|---|---|---|---|---|---|---|
| Control | - | 7.36 | 62.996 | 10.865 | 38.101 | 39.62 | 74.083 |
| Alum | Pre | 10.64 | 56.653 | 11.922 | 37.845 | 39.678 | 72.515 |
| Mordanted | Meta | 8.28 | 62.744 | 10.405 | 38.754 | 40.126 | 74.971 |
| | Post | 10.38 | 55.066 | 11.644 | 35.519 | 37.379 | 71.85 |
| Myrobalan | Pre | 15.14 | 54.753 | 8.087 | 33.672 | 34.629 | 76.496 |
| Mordanted | Meta | 13.72 | 58.339 | 7.343 | 34.283 | 35.061 | 77.911 |
| | Post | 14.64 | 56.181 | 10.121 | 35.677 | 37.085 | 74.163 |

**TABLE 2: FASTNESS PROPERTIES OF SILK FABRIC DYED WITH *CICHORIUM INTYBUS* ROOT EXTRACT**

| Sample | Mordanting Method | Washing fastness grades | | Rubbing fastness grades | | Light fastness grades | Perspiration fastness grades | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | Acidic | | Alkaline | |
| | | CC | CS | Dry | Wet | CC | CC | CS | CC | CS |
| Control | | 3 | 2-3 | 3 | 3 | 1-2 | 3 | 2-3 | 3 | 2 |
| Alum | Pre | 3 | 2-3 | 2-3 | 2-3 | 1-2 | 3-4 | 2-3 | 3-4 | 2-3 |
| Mordanted | Meta | 3 | 2-3 | 2-3 | 2-3 | 1 | 3 | 2-3 | 3 | 2-3 |
| | Post | 3 | 3 | 2-3 | 2-3 | 1 | 3 | 2-3 | 3 | 2-3 |
| Myrobalan | Pre | 4 | 4 | 3-4 | 3 | 3 | 4 | 3-4 | 4 | 3-4 |
| Mordanted | Meta | 4 | 3-4 | 3-4 | 3-4 | 3-4 | 4 | 3-4 | 4 | 3 |
| | Post | 4 | 4 | 4 | 3-4 | 3 | 4 | 4 | 4 | 3 |

**Antibacterial Properties:** The antibacterial property is qualitatively evaluated by AATCC Test Method 147-2004 against gram-negative (*E. coli*) and gram-positive (*S. aureus*) bacterium. The results **Table 3** show that all the treated fabrics, including unmordanted sample, have very good antibacterial property to gram-negative and gram-positive bacterium, *E. coli* and *S. aureus*. In all the cases, there is a good zone of inhibition ranging from 1.4 mm to 3.7 mm against *E. coli* and 1.2 mm to 2.65 mm against *S. aureus*. The mordanted samples showed an increased zone of inhibition compared to unmordanted sample (dyed without mordant). From the results shown in **Table 3**, it is clear that all the myrobalan mordanted samples showed a higher zone of inhibition compared to alum mordanted samples.

**TABLE 3: ANTIMICROBIAL ACTIVITY OF SILK FABRIC DYED WITH *CICHORIUM INTYBUS* ROOT EXTRACT**

| Sample | Mordanting Method | Zone of inhibition, mm | |
|---|---|---|---|
| | | *E. coli* | *S. aureus* |
| Positive Control | - | 4.1 | 4.9 |
| Negative Control | - | Nil | Nil |
| Unmordanted | - | 1.4 | 1.2 |
| Mordanted | | | |
| Alum | Pre-mord | 2.7 | 2.2 |
| | Meta-mord | 2.9 | 1.7 |
| | Post-mord | 2.7 | 1.6 |
| Myrobalan | Pre-mord | 3.7 | 2.65 |
| | Meta-mord | 3.4 | 2.4 |
| | Post-mord | 3.7 | 2.7 |

**Antioxidant Properties:** DPPH radical scavenging activity of Ascorbic acid, control and pre, post and simultaneous mordanted samples with alum and myrobalan are shown in **Fig. 1** and **2**. The activity was increased by increasing the concentration of the sample extract (100 µl, 300 µl, and 500 µl). DPPH antioxidant assay is based on the ability of 2,2-Diphenyl-1-picrylhydrazyl radical (DPPH), a stable free radical, to decolorize in the presence of antioxidants. The DPPH radical contains an odd electron, which is responsible for the absorbance at 517 nm and also for a visible deep purple color. When DPPH accepts an electron donated by an antioxidant compound, the DPPH is decolorized, which can be quantitatively measured from the changes in absorbance. Ascorbic acid was used as Standard. All the samples dyed with *Cichorium intybus* root extract showed excellent increased % inhibition with the increase in the concentration of extract proving it to be a potential antioxidant.

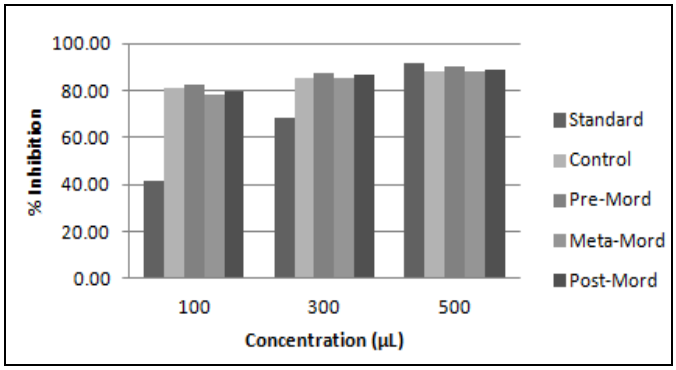


**FIG. 1: DPPH ASSAY OF ALUM MORDANTED SILK SAMPLES DYED WITH *CICHORIUM INTYBUS* ROOT EXTRACT**

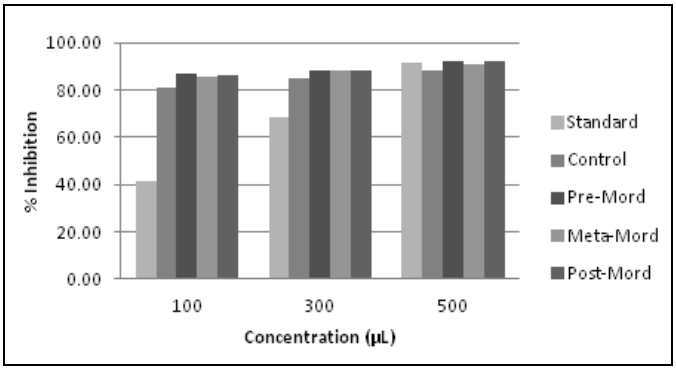


**FIG. 2: DPPH ASSAY OF MYROBALAN MORDANTED SILK SAMPLES DYED WITH *CICHORIUM INTYBUS* ROOT EXTRACT**

The control sample dyed without mordant showed % inhibition of 88.14% at 500 µl concentration. Among all the mordant-dyed samples, silk pre-mordanted with myrobalan showed the highest % inhibition (92.35%) followed by myrobalan post-mordanted with % inhibition of 92.06% and myrobalan meta-mordanted with % inhibition 91.04%. The samples mordanted with myrobalan showed higher antioxidant activity compared to alum mordanted samples. However, all the samples including the control sample (dyed without mordant) showed positive results for DPPH assay.

**CONCLUSION:** Silk fabric was treated with natural dye extracted from *Cichorium intybus* roots to produce a multifunctional textile material with antibacterial and antioxidant properties. From the study, it is revealed that mordants and mordanting methods have a synergistic effect on color strength, colorfastness, anti-bacterial and antioxidant properties. Mordant treatment not only improved the color strength and colorfastness properties of the sample but also added to its antibacterial and antioxidant properties. The dyed samples showed good inhibition to microbial growth. The antioxidant activity of the samples was excellent and found to be increased with the increase in the concentration of the dye extract. The treatment of silk fabric with *Cichorium intybus* root extract provided a textile material with both antibacterial and antioxidant properties and acceptable color strength and color fastness properties.

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**CONFLICT OF INTEREST:** Nil

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