**Extraction and Application of Marigold Dyestuff with \((Ocimum tenuiflorum)\) Oil on Cotton and Polyester Activity against Antibacterial**

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**Abstract:** A lot of attention has been given by the researchers to seek cheap and natural sources of colorant materials and strategies to improve their fastness against the light, crocking, and washing of textile materials. Marigold flowers were taken to extract dyestuff and applied on the surface of cotton and polyester fabrics to study the colorfastness in dry and wet conditions. Oil was also extracted from leaves of *Ocimum tenuiflorum* (tulsi) and added in the prepared dye bath to induce antimicrobial properties in the dyed fabrics. Colorfastness of fabrics was evaluated by following AATCC 8-2007 test method. Antimicrobial activity was determined by following recommendations given by AATCC 147-16 test procedure. It was concluded that the scale of colorfastness ranged between 4 and 5 for polyester and cotton fabrics respectively that depict excellent fastness properties. *Ocimum tenuiflorum* oil helped to reduce the growth of bacteria on treated fabrics. It was found that extracted oil provided satisfactory protection against *Staphylococcus aureus* and *Escherichia coli* on both types of fabrics.

**Keywords:** Antimicrobial, *Ocimum tenuiflorum*, Marigold, Dyestuff, Cotton, Polyester

**1. INTRODUCTION**

Technology is advancing at a phenomenal pace. Every industry is looking for innovative materials and techniques to satisfy its customers. The use of a natural substance such as plants, flowers, herbs, insects, and rocks are used to extract dyes and oils for finishing treatments on various types of fabrics. Most of these substances have medicinal properties and are extensively used in the field of textiles as antimicrobial or antibacterial agents while preparing spinning, dyeing, or finishing solutions [1]. The concept of green consumerism has opened new avenues in this field of research. It is mainly due to the reason that natural dyes do not create harm to the wearer as well as do not damage the environment at any stage of production and application. These substances are regarded as environment-friendly [2]. The use of natural products for dyeing and finishing purposes in the textile industry has been diminishing due to a lack of useful techniques to extract dye and quick application procedures for durable effects. So, that’s why nowadays manufacturers are willing to adapt synthetic dyes which are a great source of creating air and water pollution. These dyes are notorious for damaging public health. Moreover, they also create waste disposal problems. [3]. It should be the responsibility of textile manufacturers and technologists to use eco-friendly substances and to adapt eco-friendly procedures to satisfy their consumers [4].

Microorganisms are present almost everywhere and can multiply very quickly. They may cause discoloration, create a smell, develop stains, break yarns, and lose tear and tensile strength in the textile materials [5]. Most synthetic substances are used as antibacterial agents in textile industries that present certain environmental and health hazards. So, a lot of attention has been given in inducing antimicrobial properties in fabrics by using natural...
Natural herbs are used to produce many natural antibacterials as compared to synthetic products. Most of the plants have antibacterial characteristics due to the presence of certain compounds such as tannins, phenols, lactones, saponins, and terpenoids [7]. One of the most important reasons for using plants as an antibacterial source is the elimination of side effects even in case of prolonged usage [8]. Similarly, most of the synthetic substances and dyes are toxic, hazardous, and carcinogenic. They also cause the environment to pollute [9].

*Ocimum tenuiflorum* is a natural herb (Fig 1) that belongs to labiates family containing 70% eugenol, 20% methyl eugenol, and a lesser amount of other ingredients such as β-caryophyllene and oleanolic acid [10]. It has been observed that during olden days, tulsi leaves were used as antibacterial agents in the field of textiles and also used in pharmaceutical products [11]. There are more than forty compounds in tulsi. It has volatile oil in the form of rosmarinic acid which serves as an antioxidant. It has medicinal characteristics due to the presence of eugenol. Tulsi seeds contain fatty acids and sitosterol that helps to produce antibodies to fight against certain diseases [12]. Tulsi leaves are used to extract oil by grinding them into fine particles and adding in the spinning, dyeing, or finishing solution. It helps to restrict the growth of bacteria [13].

Natural extracts in the form of oils and dyestuffs are currently used in the textile industry due to their inherent characteristics of killing bacteria. They are non-toxic and non-allergic. They help to save our environment [14]. Therefore a lot of attention has been given to study the effect of various natural herbs and plants on the efficiency of fabrics.

*Ocimum tenuiflorum* leaves are grown in Pakistan as a perennial shrub with approximately 6-9 inches height of the plant and are commonly known as tulsi. The leaves are somewhat round at the edges and are 2.5 inches in length upon maturity. The leaves have the aroma and are mostly used in medicines [15]. Pakistan is also well known for the production and cultivation of flowers such as Marigold, Red rose, Chrysanthemum, Lilies, Jasmine, and Gladiolus. The climate and land both are favorable in Pakistan for their growth but due to the lack of available resources, the production is only limited to small scale and cater to the needs of local consumers [16].

This research was aimed to study the effect of tulsi leaves in terms of providing antibacterial properties to the naturally dyed fabrics with marigold flower.

2. MATERIALS AND METHODS

This study was experimental. The selected fabrics (100% cotton and 100% polyester) were obtained from Nishat Mills Pvt Limited at Lahore and these materials were evaluated for their construction parameters (Table 1). The selected materials were in grey condition. So, the fabrics were scoured by boiling them in a 10% solution of Sodium Hydroxide for 15 minutes to remove any dust particles, starch, or other foreign matter. Then washed and rinsed under running cold water. The samples of fabrics were mordanted with 15 g / liter (2%) of copper sulphate for 1 hour at a temperature of 75-80 °C. After mordanting, the specimens were squeezed out to remove the excessive liquor and finally immersed in a prepared dyestuff [17].

Marigold flowers were collected from the local market to prepare the dyestuff. The flowers were dried under the sunlight to remove the moisture content so that they can be stored without delay for
Table 1. Construction parameters of collected samples

<table>
<thead>
<tr>
<th>Sample</th>
<th>Fiber content</th>
<th>Fabric mass (GSM)</th>
<th>Thread count (warp/weft)</th>
<th>Yarn linear density (warp) (tex system)</th>
<th>Yarn linear density (weft) (tex system)</th>
<th>Weave</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>Cotton (100%)</td>
<td>155</td>
<td>105/90 = 195</td>
<td>12.451</td>
<td>11.584</td>
<td>Plain</td>
</tr>
<tr>
<td>BB</td>
<td>Polyester(100%)</td>
<td>150</td>
<td>90/95 = 185</td>
<td>15.675</td>
<td>14.768</td>
<td>Plain</td>
</tr>
</tbody>
</table>

Colorfastness is the characteristic of fabric to resist change of applied color on any external substance especially white colored fabric. The fabrics were assessed for their colorfastness by following AATCC Test 8-2007 test method [21] (Fig 2). Conditioning was performed by following ASTM D 1776 procedure [22] for 4 hours in a standard testing atmosphere 70 ± 2F and 65 ± 2% r.h. Two sets of specimens 50mm x 130mm were cut from each category for wet and dry conditions respectively. White cloth was used as a standard against dyed specimens for rubbing.

Crockmeter was used to evaluate the transfer of color from a dyed fabric to the standard one. For wet conditions, the specimen was wetted with distilled water. Grayscale ranging from 5 to 1 was taken to give ranks to the tested samples. 5 stands for excellent where no color was lost and 1 stands for poor dye absorption where complete color was lost. Other ranks lie between these two ranks.

The dyed fabrics were evaluated for their antimicrobial activity by following the Qualitative method AATCC 147 [23]. Specimens with a dimension of 25 x 50 mm were taken out from each sample. Petri dishes were sterilized at 120°C for 15 minutes. Sterilized nutrient agar (15mm) was then applied in each petri dish. It was turned into the gel before inoculation. Broth culture in distilled water was used as inoculum for 24 hours. With the aid of the inoculum loop, two microbes named *Staphylococcus aureus* (Fig 3) and *Escherichia coli* (Fig 4) were streaked out by drawing 5 horizontal lines. Each specimen was carefully pressed against these parallel lines with a sterilized spatula. The Petri dishes were incubated at 37°C for 24 hours. The plates were assessed for their microbial activity along with the inoculum streaks by measuring the zone of inhibition in millimeters around each specimen [6].
3. RESULTS AND DISCUSSIONS

Dyestuff obtained from marigold flower by boiling dry petals presented good results on various types of fabrics. Different types of flowers and herbs are an excellent source of extracting the dye and applied on the surface of fabrics [24]. It was mainly due to the use of natural plants and herbs which have low molecular weight than chemical or synthetic dyes. It helps the dye to adhere strongly to the surface of textile material [25]. It is observed from Table 2 that the colorfastness of all treated fabrics ranges from excellent to very good. Cotton fabric ‘AA’ showed better results in both dry and wet conditions as compared to ‘BB’. Dye penetration is largely dependent on the surface area between solid and solvent. It was observed that due to the increase in total surface area by increasing solid particles, the total mass transfer of respective dye to a solvent was also accelerated [26]. Colorfastness should be checked both in dry and wet conditions.

Table 2. Crocking of samples in dry and wet conditions

<table>
<thead>
<tr>
<th>Sample</th>
<th>Crocking (Dry condition)</th>
<th>Crocking (Wet condition)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>BB</td>
<td>4</td>
<td>4.5</td>
</tr>
</tbody>
</table>

To improve the color fastness of dyed fabrics, certain mordants act as fixing agents that can be used. Mordants increase the capacity of dye particles to strongly attach to the fiber and make it colorfast for a longer period [27]. The obtained results are due to the reason that the dye strongly adheres to the surface of the fabric easily [28-30]. Silk samples mordanted with copper sulphate and ferrous sulphate present excellent colorfastness properties [31]. Results are also similar to the findings of another study conducted by Jha et al., [32] that copper sulphate and ferrous sulphate used in marigold dye stuff showed good to excellent ratings with a range of 4 to 5. The measurement of colorfastness is determined by the staining of samples against standard fabric [33]. It was found that copper sulphate used as a mordant in cotton dyeing showed better color fastness against rubbing, washing, and light [34]. It was due to the presence of metal ions that serve as electron acceptors for electron donors to form coordination bonds in the presence of dye molecules. Mordants such as Copper sulphate and Ferrous sulphate present high dye take-up by bridging up the gap between dye and fiber structure. The dye extracted with marigold flower along with the mordant insolubilize applied dye thus making it more colorfast [35].

The growth of microbes on fabrics has diverted the attention of textile technologies towards the development of antimicrobial treatments. Recently a
lot of damage occurred during the storage of textiles due to microbial attack. The use of antimicrobial finishes saves money by providing durable effects [1]. *Ocimum tenuiflorum* was best to serve as a dyestuff for wool and wool related fabrics. The prepared dyestuff was adhered to the surface of the fabric and dyed well [36]. Natural dyes extracted from certain plants and herbs act as a defensive mechanism against many bacteria, microbes, or other pathogens. Tulsi oil used in dyestuff restricted the growth of micro-organisms due to the slow release of many active agents from the substrate and assist in damaging the cell membrane of microbes. As Tulsi belongs to the family of the natural herb, it does not create any harm to the wearer thus providing him safety [37]. The ingredients found in tulsi such as caryophyllene, phytol, and germacrene are proved to have antimicrobial properties used for many purposes [38].

It can be observed from Table 3 that the zone of inhibition of positive gram bacteria for dyed samples ranges from 55mm and 45mm whereas 47mm and 39mm for negative gram bacteria in AA and BB respectively. It shows that treated fabric restricts the growth of bacteria. On the other hand, undyed samples showed a zone of inhibition ranges from 3mm and 5mm. It shows the attraction of bacteria towards their surface. One of the possible reasons for the obtained results may be that Tulsi leaves have antimicrobial properties and provide protection when treated with cotton fabrics. Tulsi oil extracted from its leaves help in preventing the growth of Gram-positive and Gram-negative bacteria [39]. It is observed that fine particles of *Ocimum tenuiflorum* (Tulsi) were locked in between the yarn structure and presented antibacterial properties. Tulsi extract has filled the gaps between yarns and helped to create a smooth surface in the form of lamination. Tulsi compounds protect more than 70% against antimicrobial activity on cotton and cotton blended fabrics. The finish made with tulsi leaves served excellent protection against the growth of bacteria on various types of polymers [40]. In another study, the extract of *L. fischeri* was used to determine the antioxidant capacity of its compounds. It was found that the presence of polyphenols with hydroxyl head groups served as efficient antioxidants [41].

Oil extracted from tulsi possess higher antibacterial characteristics against *Staphylococcus aureus* and *Escherichia coli*. The inhibition behavior of selected oils against various tested bacteria was due to the presence of certain compounds such as eugenol, camphor, thymol, carvacrol, and cinnamaldehyde [42].

The pharmacological behavior of many herbs provides a path to the researchers in the synthesis and extraction of compounds for antibacterial properties in many fields. It was observed that extracted compounds from Levofloxacin had good antimicrobial properties against two selected microbes such as *Candida albicans* and *Bacillus subtilis* [43].

Antibacterial agents assist in killing microorganisms present on and around the fabric surface. It is usually observed that the larger zone of inhibition gives better results against the growth of bacteria and vice versa [44, 45]. The antimicrobial activity of tulsi is due to the presence of phenol, quinine, and tannin. These ingredients help in

<table>
<thead>
<tr>
<th>Sample</th>
<th>Bacterial species</th>
<th>Zone of inhibition (mm)</th>
<th>Growth observation</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>Dyed sample</td>
<td>Undyed sample</td>
</tr>
<tr>
<td><strong>AA</strong></td>
<td><em>Staphylococcus aureus</em></td>
<td>55</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td><em>Escherichia coli</em></td>
<td>47</td>
<td>3</td>
</tr>
<tr>
<td><strong>BB</strong></td>
<td><em>Staphylococcus aureus</em></td>
<td>45</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td><em>Escherichia coli</em></td>
<td>39</td>
<td>3</td>
</tr>
</tbody>
</table>
durability and longer life of dyed fabrics [46]. It has been investigated that eugenol restricts the growth of microbes and bacteria more than 70% in most of the studies [47]. The antibacterial agents present in tulsi leaves permeate through the cell membrane of microbes and kills them. These leaves also assist in protecting our environment [48].

4. CONCLUSIONS

It was concluded from the present work that Marigold flowers are suitable to be used a dye on cotton and polyester fabrics as they have good fastness properties. Ocimum tenuiflorum oil is quite a good source of developing antimicrobial properties in fabrics during dyeing or finishing stages. The extraction of the application process of dyeing fabrics was eco-friendly. It is the need of an hour to create awareness among all stakeholders to adopt such methodology which is safe for both human health and our environment too.

5. REFERENCES


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