Research Article

ANTIBACTERIAL EFFICACY OF COTTON FABRIC DYED WITH NATURAL DYE FROM FRUITS OF MULBERRY VARIETIES COMMONLY GROWN IN TAMIL NADU

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ABSTRACT

Ecofriendly dyeing efficacy of cotton fabric with the pigments of mulberry fruits and its antibacterial functionality were investigated. The pure extract, ethanol and aqueous extracts were tested for their dye ability. The colour values referred in terms of L, a and b indicated purple to red colour of the pigment. The crude extract of fruits shown inhibitory zone against *Bacillus subtilis*, *Staphylococcus aureus* and *Escherichia coli* in well diffusion assay. The cotton fabric dyed with fruit extract had fastness to light, UV and also thermal stability. The results of this research indicated a potential commercial application of the mulberry fruit in textile technology.

INTRODUCTION

Mulberry (*Morus* sp.) is distributed in most part of the world. It is extensively cultivated in eastern, central and southern Asia for silk production. Most of the Indian mulberry varieties of mulberry belongs to *M. indica* (Datta, 2000). *M. indica* fruits are ovoid or cylindrical, the unripe fruits are red in colour and become black on ripening (Ozgen *et al.*, 2009).

The fruit has a pleasant flavour with slight acidic taste and has an attractive red colour (Koyuncu *et al.*, 2004: Ozgen *et al.*, 2009). Mulberry fruit have been reported to exhibit, antioxidant (Kim *et al.*, 1999, 1998; Naderi *et al.*,2004), antimicrobial activity (Khalid *et al.*, 2011) anti pyretic activities (Jawad *et al.*, 1988) and anti-inflammation (Butt *et al.*, 2008). These activities are generated by anthocyanins, which are a group of naturally occurring phenolic compound that are responsible for the colour of mulberries (Liu *et al.*, 2004).

There is an increasing interest in adding value to textiles all over the world for much demand functionality. Among these, development of antibacterial textile finish is highly indispensible. Antimicrobial textiles found to have many medical applications such as wound dressing and product carrier materials (Mahesh *et al.*, 2011; Saranyadevi *et al.*, 2014).

The use of natural product such as natural dye for antimicrobial finishing of textiles is widely practiced. The use of natural dye for antimicrobial finishing of cotton fabrics has been widely reported (Gupta *et al.*, 2004, 2005; Singh *et al.*, 2005). Eco-friendly dyeing of fabric with fruit colourant containing anthocyanins also been successfully taken to commercial application (Wang *et al.*, 2016). The dyeability of anthocyanin in extract of mulberry fruit on cotton fabric was found (Wang *et al.*, 2016).

The present study investigated the effectiveness of dye extract from mulberry fruits of varieties common in Tamil Nadu region and the antimicrobial functionality of dyed cotton fabric.

MATERIALS AND METHODS

The fresh fruits of mulberry (*M. indica*; variety V1) were collected from mulberry farms in Coimbatore district of Tamil Nadu. Dark coloured fruits were selected, the peduncle removed before the fruits were used for extraction process. Woven cotton fabric was purchased from market and used.

Bacterial Cultures

Two gram positive bacteria, *Staphylococcus aureus* (ATCC 6538) and *B. substilis* (ATCC 19659) and one gram

negative bacteria, *Escherichia coli* (ATCC 9863) maintained on agar slants were used for antimicrobial susceptibility testing.

Preparation of Mulberry Fruit Extract

Approximately 500 g of fresh fruits were mashed in an electrical blender. The fresh juice obtained was filtered using a nylon cloth and then centrifuged at 5000 rpm for 15 minutes. The supernatant was collected and kept in the dark at 4^0C until further analysis.

Aqueous fruit extract was prepared by adding 500 ml of water to 100g of fruit sample while grinding. Mulberry juice from the pulp was separated using nylon filter cloth and centrifuged at 4500 rpm for 10 minutes and supernatant was collected.

Figure 1: Inhibition of aqueous extract of mulberry fruit to *Bacillus subtilis*

Ethanolic extract was prepared by adding 95 ml of 95 per cent ethanol and 5ml of HCL instead of water for 100 g fresh mulberry fruit. Extract was centrifuged finally to collect 100 ml of mulberry fruit extract.

Measurement of the *M. indica* Fruit Extract Colour

Colour values of the mulberry fruit extract were measured using Hunter Colour Meter. Based on the spectra, the apparatus calculated and retuned the colour parameters. The colour coordinates of the uniform colour space CIE-LAB, L^*, a^*, b^* were determined by its reflectance (L^*) and chromaticity (a^* and b^*) values. The L^* value indicated brightness ranging from black (L^*=0) to white (L^*=100). The a^* value range from -60 (green) to 60 (red) and b^* value range from -60 (blue) 60 (yellow).

Stability of Mulberry Fruit Extract Dyed Cotton Fabric to Ultra Violet Radiation, Light and Heat of Sun

The cotton fabric was soaked in different extracts of mulberry fruit for 1h and used for the study. Hand oven cotton fabric was used in the study. The fabric dyed with different extracts of fruit was kept under UV rays for 30 min and the colour retention was calculated in comparison with fresh dyed fabric. The stability to sun light and heat was examined by exposing the aqueous extract dyed fabric to direct sun light for continuous 8 h of sunshine. At the end of exposure the colour retention was calculated in comparison with fresh dyed fabric (Fig. 2a-b).

Figure 2(a): Dyed cotton fabric with aqueous fruit extract

Figure 2(b): Dyed cotton fabric with pure fruit extract

Preliminary Assessment of Antibacterial Activity of Mulberry Fruit Extract

The antimicrobial activity of the Mulberry fruit extract was assessed by well diffusion assay (Perez *et al*., 1990). *In vitro* antibacterial activity of pure, aqueous and ethanolic fruit extract were tested against *S. aureus, B. subtilis and E. coli*. All the glasswares and media used for the assay was sterilized in autoclave at 121 C and 15 lb pressure for 15 minutes. The sterile media was poured into sterile petriplates and allowed to solidify at room temperature. Using sterile glass spreader 1000 µl of bacterial suspension was spread uniformly on the respective solidified medium. Wells were bored in the medium using cork borer. 100 µl fruit extract was transferred into the well and incubated for

24 hrs at 37℃. After 24 h, the diameter of resulting zone of inhibition was measured.

Antimicrobial Activity of Mulberry Fruit Pigment Dyed Cotton Fabric

Cotton fabric was cut into 1cm^2 pieces and soaked in various extracts of mulberry fruit for one hour. *S. aureus, B. subtilis and E. coli* were used as test organisms. The fabric dyed with aqueous extract of mulberry fruit was placed on the centre of the respective agar plates. Undyed fabric served as control. Zone of inhibition was measured at the end of 48 h of incubation.

Figure 3(a): Dyed fabric after exposure to UV rays

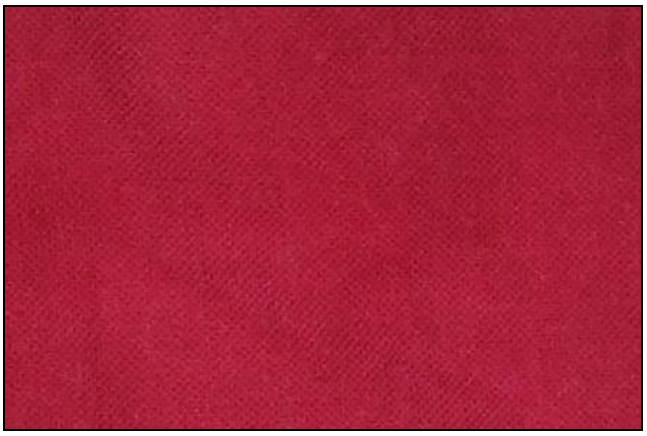

Figure 3(b): Dyed fabric after exposure to sunlight for 2 days (8h/day of sunshine)

RESULTS AND DISCUSSION

Fruit Colour of Mulberry Fruit Extracts

Colour values of different mulberry fruit extracts determined using Hunter colour meter are given in Table 1. values of pure mulberry fruit extract are higher than aqueous and ethanolic extract. In pure extract a* value is higher when compared to L* and b* value indicating the purple to red colour of the mulberry fruit juice. However, ethanol is not a good extractant of colour of mulberry fruit based on the colour parameters of fruit extracts.

Stability of Mulberry Fruit Pigment Dyed Cotton Fabric to UV Rays, Light and Heat of Sun

Stability of mulberry fruit pigment dyed cotton fabric to UV rays, light and heat of sun was exhibited in figures 3a-b. Exposure of aqueous extract dyed fabric to UV for 30 minutes resulted in minimal colour degradation. In sunlight exposed fabric cloth until 2days (8h/day of sunshine) no colour fading occured but after 3days (8hr/days of sunshine) colour fading and degradation were seen.

Table 1: Colour values of mulberry fruit extract measured in Hunter colour meter

Value	Pure extract	Aqueous extract	Ethanolic extract
L	17.66	1.68	0.067
a	19.46	4.62	0.097
b	-9.87	-0.55	-0.016

L^* = black (L^*=0) to white (L^*=100)

a^* = 60 (Green) to + 60 (Red)

b^* = -60 (Blue) to + 60(Yellow)

Antimicrobial Activity of Mulberry Fruit Extracts and Dyed Fabric

Antimicrobial activity of different mulberry fruit extract was measured in cm^2 of zone of inhibition against *B. subtilis, S. aureus and E. coli* (Table. 2) (Fig. 1.) Pure mulberry fruit extract exhibited zone of inhibition of 2.63 to 2.98 cm^2 against *B. subtilis, S. aureus and E. coli* respectively. The activity of aqueous extract was comparable to pure extract (Table 3). There is more than 90 % retention of antimicrobial activity of dyed cotton fabric when calculated in comparison to inhibition zones formed in respective fruit extracts.

The dying ability and the antibacterial potential of fruit of mulberry variety, V1, widely cultivated in Tamil Nadu was examined in the present study. The colour values of whole fruit, aqueous & ethanolic fruit did not show significant variation. The values indicated the presence of higher amount of red colour pigments. The extraction of anthocyanin based pigments is normally accomplished using water and ethanol based solutions (Lee and Wrolstad, 2006). Anthocyanin content of mulberries ranges from 0.16 to 0.18 mg C3G/g dry weight as reported (Gozlekci *et al.,* 2015). The fruit extract exhibited inhibitory activities against *Bacillus subtilis, Staphylococcus aureus* and *Escherichia coli.* Effect against the tested bacteria correlated with the purple colour intensity of the fruit extract. The antimicrobial activity exhibited here, is similar to the findings reported by (Budiman *et al.,* 2017) with *Morus nigra* fresh juice. Chen *et al.,* (2018) had observed

broad spectrum of antibacterial activity in pigmented black mulberry (*Morus nigra*) but not in non-black mulberries.

Dyeing of fabric with natural dye extract from plant and microbes is an environment eco-friendly approach in the fabric industry. Natural dyes with antimicrobial activity is known to have medical applications. The cotton fabric dyed using mulberry fruit extract considerably retained antibacterial properties (Wang *et al.,* 2016). In this context, the antimicrobial properties exhibited by mulberry fruit extract dyed cotton fabric will be of more value to health conscious users. In our study, the use of chemical fixation is avoided and high concentration and duration was provided while dyeing. The dyed cotton fabric exhibited the deep purple colour of the fruit pigment, and this aborigine colour is rare to achieve with natural dyes.

Table 2: Antibacterial activity of mulberry fruit extract as determined by well diffusion assay

Sl. No.	Bacterial culture	Diameter of inhibition zone in cm^2		
		Pure extract	Aqueous extract	Ethanolic extract
1	*Bacillus subtilis*	2.98	2.14	1.28
2	*Staphylococcus aureus*	2.68	2.37	1.29
3	*Escherichia coli*	2.63	2.11	1.29

Table 3: Retention of inhibitory activity of aqueous extract dyed cotton fabric after exposure to UV light and sun

Sl. No.	Bacterial culture	% retention of inhibitory activity	
		UV light exposure	Exposure to sun
1	*Bacillus subtilis*	91.27	86.34
2	*Staphylococcus aureus*	91.31	87.28
3	*Escherichia coli*	91.07	86.33

The degradation of anthocyanin by UV, light and heat has been reported (Aramwit *et al.,* 2010); the longer the period of exposure, the lower both the anthocyanin content and antimicrobial activity (Qin *et al.,* 2010). Since mulberry fruit colour is primarily generated by anthocyanins, the thermal and light stability of the dyed fabric was tested. The UV and sunlight treated dyed fabric retained nearly 90 per cent of colour and antibacterial activity in our study. This difference in stability and activity among the samples may be due to anthocyanin concentration of the extract.

CONCLUSION

Fruit extract of *Morus indica* (var. V1) contains significant antibacterial activity which may be related to the anthocyanin content of this purple coloured fruit. Fruit pigment intensity is much correlated with antibacterial activity. The findings of present study promises the use of mulberry fruit extract as natural antimicrobial agent and its extent use in biomedical applications. Relative stability of antibacterial activity of dyed fabric is a promising finding with functional application in textile industry.

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