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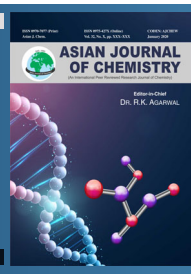

Psuderanthimum reticulatum: A New Green Source for Dyeing of Cotton Fabric

S. Margrat Sheela*, J. Rosaline Vimala, M. Stella Bharathy, A. Agila, S. Sharmila and A. Leela Devaki

Department of Chemistry, Holy Cross College (Affiliated to Bharathidasan University), Tiruchirappalli-620002, India

*Corresponding author: E-mail: msheela86@gmail.com

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In the present work, cotton fabrics were dyed using the acidified aqueous extract of *Psuderanthimum reticulatum* leaves with different mordants such as ferrous sulphate, stannous chloride, copper sulphate, potash alum and ferric alum by premordanting technique and various shades were obtained. The washing, perspiration and light fastness properties of the dyed fabrics were carried out by standard methods, ISO 105-C06 A2S-2010, ISO 105 E04-2013 and ISO 105 B02 Methods 3:2014, respectively. Cotton fabrics dyed with copper sulphate and ferric alum showed excellent to very good properties towards the entire fastness tests studied. The bioactive colourant present in the aqueous extract of *Psuderanthimum reticulatum* responsible for dyeing is anthocyanin pigment and it was identified by chemical test, UV & FTIR spectral and HPLC analysis.

Keywords: *Psuderanthimum reticulatum*, Natural dyeing, Colour fastness, Anthocyanin.

INTRODUCTION

Textile industries use synthetic dyes in larger amount to meet the global need for dyeing of fabrics because they are cost effective, give wide range of bright shades and exhibit improved fastness properties to the dyed materials [1]. Due to the large production and application of synthetic dyes, vast amount of waste and unfixed colorants are released to the environment, which disturbs the eco-balance and cause serious health hazard. At present, natural dyes gained more importance because of its eco-friendly nature. They produce very uncommon, soothing shades to the fabrics therefore the use of non-toxic, non-allergic and eco-friendly natural dyes on textiles has become significant importance due to increasing environmental awareness [2]. The alchemy of colours started from an ancient time. Nature provide wide spectrum of colours. The primary sources of natural dyes are plants, animals and minerals. Natural dyes are classified based on their source, structure and method of application. Dyes are classified into indigoid, anthroquinone, flavones, α-hydroxy napthoquinones, dihydropyrans, carotenoids and anthocyanidins based on its structure [3]. The natural colourant anthocyaninis belong to the flavonoid group of phytoconsituents. Anthocyanin gives red, blue or violet colours to the plants. They are water soluble pigments and it is having high hydrogen bonding capacity. Anthocyanins are substituted glycosides of salts of phenyl-2-benzopyrilium in which the hydroxyl group at 3, 5, 7 positions and phenyl ring has 1-3 hydroxyl or methoxy groups at 3′, 4′, 5′ position. They are more sensitive to pH. The sugar moieties like glucose, rhamnose, xylose, glactose, arabinose and fructose *etc.*, are present in anthocyanins through glycosidic linkage. The sugar molecules are frequently attached to the hydroxyl groups at 3 and 5 positions. The basic structure of anthocyanin is anthocyanidin which contain no sugar molecules. The most common anthocyanidins are cyaniding, delphinidin, pelargonidin, peonidin, malvidin and petunidin [4]. The most used cellulosic fiber in the world is cotton. It exhibits the following properties like good absorbency, colour retention, machine-washable, good strength, easy to handle, *etc.* This make the cotton fiber is suitable for everyone [5]. Most of the natural colourants were mainly obtained from plants. Ghurde *et al.* [6] carried out the natural dyeing of cotton fabrics using the extract of *Ixora coccinea* floral petals under normal conditions and tested the colour fastness of the dyed fabric towards washing properties. The spectroscopic analysis of the dye colour gives the identification of the functional groups which is present in the dye extract. Extraction of dyes from walnut shells using Soxhlet apparatus, application of dye to polyamide fabrics and characterization of the colorant

were studied by Mirjalili and Karimi [7]. Efficiency of dyeing on wool, cotton and silk fabrics by using natural obtained from kitchen waste of dry skin extract of *Allium cepa* was studied by Vankar *et al.* [8]. The plant extract showed good wash and light fastness for cotton and silk fabrics and gave the hue colours. Uddin [1] has studied the dyeing of silk using mango leaves. The dyed materials were evaluated by measuring the fastness properties and colour yield. Mango leaves have good potentiality for dyeing of silk fabric. Deo and Desai [9] investigated dyeing of cotton and jute fabrics using the aqueous extract of tea by pre-mordant, post-mordant and meta-mordant techniques. The colourant present in tea extract was tannin and the results for wash and light fastness of the dyed fabrics were good to excellent colour and deep shades were obtained. Study on dyeing of cotton fabric using henna leaves extract and effect on UV radiation was done by Iqbal *et al.* [10]. The natural dye contains Lawsone that imparts yellowish green colour. Different mordants were used to improve the colour fastness properties of irradiated dyed cotton fabrics. Iqbal and Liaqat [11] investigated direct dyeing of cotton using natural dye extracted from *Punica granatum* bark in aqueous medium and three mordants were used. The fastness properties such as rubbing, perspiration and light were also assessed. Jahan and Datta [12] have studied the dyeing of cotton and silk fabric using the madder as the natural dye.

In this work, cotton fabrics were dyed with an eco-friendly dye extracted from *Psuderanthimum reticulatum* leaves. The plant belongs to the Acanthaceae family. *Pseuderanthemum reticulatum* is a vertical open shrub; the stems lightly branched unless pinched out at intervals when young. The height of the shrub is 10 to 15 cm long and spread 30-75 cm. The leaves are broadly elliptic and are having deep purple at the back and green in front. During summer, tubular white flowers, spotted rose-red or purple at the bases are produced. They have high pest tolerance, grow at faster rate and don't have any fragrance. They are used as an ornamental plant and its leaves & juice are used to treat headache, fever, back pain and cold [13].

EXPERIMENTAL

Psuderanthimum reticulatum leaves were collected from the campus of St. Joseph's College, Tiruchirappalli, India and the plant was authenticated from the Rapinat Herbarium, Centre for Molecular Systematics, St. Joseph's College (Autonomous), Tiruchirappalli, India. Pure cotton fabric was purchased from the local market for dyeing. 5% (w/v) of analytical grade of ferrous sulphate, stannous chloride, copper sulphate, potash alum and ferric alum were used for mordanting.

Phytochemical tests: Preliminarily the aqueous extract of the fresh leaves of *Pseuderanthemum reticulatum* was subjected to the following photochemical tests to find out the phytoconstituents.

Alkaloids

Mayer's test: Mayer s reagent (2 drops) was added with 1 mL of the leaf extract. A white or cream precipitate shows the presence of alkaloids.

Hager's test: Hager's reagent (saturated aqueous of picric acid, 2 mL) was added with 1 mL of leaf extract. A prominent yellow precipitate shows the presence of alkaloids.

Carbohydrates

Molish's test: An alcoholic solution of α-naphthol (2 drops) was added into the leaf extract. The mixture was shaken well and slowly added 1 mL of conc. sulphuric acid and allow to stand for few minutes. A violet ring shows the presence of carbohydrates.

Benedict's test: Benedict's reagent (0.5 mL) was added into 0.5 mL of leaf extract. The mixture was heated on a water bath. A characteristic coloured precipitate shows the presence of sugar.

Ninhydrin test: Ninhydrin solution (2 drops, 10 mg of ninhydrin solution in 200 mL of acetone) was added into 2 mL of leaf extract. Purple colour shows the presence of amino acids.

Tannins: Few drops of 0.1% ferric chloride was added into the leaf extract. A brownish green or a blue-black colour indicates the positive result of tannins.

Flavonoids: Few drops of leaf extract were added with 5 mL of dilute ammonia solution and a few drops of concentrated H_2SO_4. Yellow coloration disappeared on standing indicate the presence of flavonoids.

Anthocyanin: HCl (2 mL) was added into the leaf extract and the solution was heated for 5 min. The red or pink colour remains stable and addition of 2 mL of NaOH into the leaf extract in another test tube and the formation of green or blue colour indicates the presence of anthocyanin.

Steroids: The leaf extract was added with 2 mL of acetic anhydride and 2 mL of H_2SO_4. The colour changed from violet to blue or green indicates the presence of steroids.

Terpenoids: Leaf extract (5 mL) was added with 2 mL of chloroform and 3 mL of conc. H_2SO_4. Reddish brown colour interface indicates the presence of terpenoids.

Phytosterols (Libermann-Burchard's test): The leaf extract was added with 2 mL of acetic anhydride and 2 drops of conc. H_2SO_4. A colour change shows the presence of phytosterols.

Phenolic compounds (ferric chloride test): Neutral $FeCl_3$ solution (5%, 5 mL) was added into the leaf extract. A dark green colour confirmed the presence of phenolic compounds.

Glycosides: HCl (1 mL) is added to 0.5 mL of the leaf extract and boilied for 30 min and 2 mL of Fehling's solution was added and again heated for 5 min. Formation of brick red precipitate shows the presence of glycosides [14].

Steps involved in dyeing

Extraction of dye: Fresh leaves of *P. reticulatum* (20 g) were cut into small pieces and allowed to boil for 30 min in a 100 mL of distilled water with 2 mL of concentrated HCl, then the aqueous extract of the plant material was cooled and filtered through Whatmann No.1 filter paper and stored in a brown bottle and refrigerated for further use. The pH of the extract was 1.1.

Scouring: In scouring natural impurities such as wax, oil, *etc*. in the fabric surface were removed and making the fabric more hydrophilic and suitable for dyeing. Scouring of cotton

fabric was done by treating them with 4% sodium carbonate solution by keeping the material to liquor ratio of 1:40 and allowed to boil for 30 min. The scoured materials were thoroughly washed with tap water and dried at room temperature. These materials should be soaked for in clean water 30 min before mordanting [15].

Mordanting: The scoured cotton fabrics were treated with different mordants such as 5% ferrous sulphate, 5% stannous chloride, 5% copper sulphate, 5% potash alum and 5% ferric alum, respectively. The wetted sample were entered into the mordant solution and allowed to boil for 30 min keeping the material to liquor ratio of 1:40. The mordanted cotton materials should be used immediately for dyeing because mordands are sensitive to the light.

Dyeing: The cotton fabrics mordanted with different mordants were dyed with the aqueous dye extract of *Psuderanthimum reticulatum* by keeping the material to liquor ratio as 1:40 and boiled for 30 min. After the dyeing process, the dyed materials were left in the dye bath overnight. The dyed cloths were taken out on the next day and it was washed well with the tap water, squeezed out and dried in the open air.

Determination of colour fastness of dyed fabrics: The term colour fastness used in the dyeing of textile materials which denotes the material colour's resistance to fading. The external conditions such as washing, light, rubbing, human perspiration, *etc*, which affect the colour of the textiles. Therefore, the dyed fabrics must be tested for their colour fastness to washing, rubbing, light, *etc*. Colour fastness studies of dyed fabrics were carried out according to the norms of ISO. In the present work, Wash fastness test was carried out as per ISO 105-C06 A2S-2010. Colour fastness to perspiration was carried out by ISO 105 E04-2013 method and light fastness using ISO 105 B02 Method 3:2014 test. Using standard grey scales, the test fabrics were assessed against washing and perspiration fastness. Light fastness was assessed on 4 point scale (blue wool standard) [2].

Identification of colorant

Test for anthocyanin: The aqueous extract of *Psuderanthimum reticulatum* was subjected to the chemical test to confirm the presence of the natural pigment anthocyanin. The presence of anthocyanins has been identified by adding 2 mL of the aqueous plant extract with 2 mL of 2 N HCl. The appearance of a pink-red colour that turns green on the addition of NaOH indicates the presence anthocyanin [16].

Paper chromatography: In partition chromatography, the samples are distributed in two liquid phases. Acidified methanolic extract (1 part of conc. HCl and 99 part of methanol) of *Psuderanthimum reticulatum* was used in paper chromatography to confirm the presence of anthocyanin pigment. In Paper chromatography technique, the methanolic extact of the sample was spotted on the chromatography paper. For the development of chromatogram butanol:acetic acid:water system was (5:4:1) used [17]. Retention time (R_f) of the coloured spot was calculated by the following formula:

$$R_f = \frac{\text{Distance travelled by the sample}}{\text{Distance travelled by the solvent}}$$

UV and FTIR spectrum: The UV-visible absorption spectrum of the *Psuderanthimum reticulatum* extract was recorded between 200 and 800 nm using double-beam UV-visible spectrophotometer (Model-HITACHJ-U2910). The FTIR spectrum of the extract was also taken using FTIR spectrometer (Shimadzu) and characteristic peaks were detected.

HPLC analysis: Analytical HPLC analysis of *Psuderanthimum reticulatum* was carried out using the acidified methanolic extract [18]. High performance liquid chromatography (HPLC) is the one of the chromatography techniques, which is used to identify, quantify and purify the multi-component present in the mixture. Mostly, reverse HPLC mode utilization is preferred due to its best separation (stationary phase-non polar; mobile phase-polar). This technique is used for the quantification and characterization of secondary metabolites in the plant extract. In the present work, the extract was analyzed using Waters HPLC equipped with a reverse-phase C18 column (4.5 × 250 mm) along with 2545 quaternary gradient module and 2998 photodiode array detector (Vienna, Austria). Before injection the sample was filtered through syringe filter (PVDF filter membrane with polypropylene housing pore size 0.2 µm, Whatman, GE Healthcare, UK) and the mobile phase was water:acetonitrile (50:50). The flow rate for the elutaion was 1 mL min^{-1}. 20 µL of sample was injected and the detection was carried out at 520 nm.

RESULTS AND DISCUSSION

Qualitative analysis of *Pseuderanthemum reticultum* leaf extract: The preliminary phytochemical screening of *Pseuderanthemum reticultum* revealed the presence of anthocyanin, terpenoids, tannis, carbohydrates and alkaloids. The results of the qualitative analysis of the plant extract are shown in Table-1.

TABLE-1
SCREENING OF THE AQUEOUS
EXTRACT OF *Pseuderanthemum reticultum*

Phytochemicals	Observed
Alkaloids	+
Carbohydrates	+
Amino acids	–
Tannins	+
Flavonoids	+
Anthocyanin	+
Steroids	–
Terpenoids	+
Phytosterols	–
Phenolic compounds	+
Glycosides	+

+ Indicates the positive result (presence); – indicates the negative result (absence)

Principle of dyeing: Mordants play an important role in imparting colours to the fabric. They stabilize the colour of the anthocyanins pigments, which are having free hydroxyl group and are capable of metal chelation. Most of the natural dyes have no substativity on cellulose or other textile fibres without the use of mardant. The mordant create an affinity

between the fibre and the pigmant molecules. The metalic salts form metal complexes with the fibres and the pigmens. After mordanting the metal salts anchored to the fibres and act as the bridging link between the pigment molecules and the fibres in formation of insloulbe complexes. The dyeing was carried out under acid condition, since the anthocyanin pigments are more stable at low pH. Most of the results from the literature showed that the highest pigment retention observed at lower pH < 3.0. Under this acidic condition, the anthocyanins primarily exist in the form of a red flavylium cation. The cotton fabrics dyed with the aqueous extract of *Psuderanthimum reticulatum* using different mordants such as ferrous sulphate, stannous chloride, copper sulphate, potash alum and ferric alum gave different shades and the Fig. 1 showed the effect of dyeing on cotton fabrics after mordanting [19].

Fastness properties of dyed fabrics

Washing fastness: Wash fastness is referred to the properties to stain the colour on the surface of textile materials during washing. Wash fastness is influenced by the rate of diffusion of dye present inside the fiber. In wash fastness test, the dyed cotton fabric was in contact with different adjacent fabrics and is agitated mechanically under described conditions of time and temperature in a soap solution. After rinsing and drying, the change in the colour of the dyed fabric and the staining of the adjacent fabrics were assessed under grey scales [20]. The results of the fastness properties of the cotton fabrics dyed with the aqueous extract of *Psuderanthimum reticulatum* with different mordants were assessed by ISO 105-C06 A2S-2010 method. The overall result of the colour fastness to washing of the dyed cotton fabrics on staining was very good to excellent. Copper sulphate mordanted fabric showed excellent wash fastness towards colour change. The mordants ferrous sulphate and ferric alum exhibited well to better fastness whereas stannous and potash alum showed poor washing fastness.

Perspiration fastness: The ability of not to fade and not to stain when dyed fabric is perspired is called perspiration fastness. In the perspiration fastness test, artificial perspiration with different acid and alkaline is used to imitate the sweat situation when human wear textiles. During the test, the dyed sample stitched with different adjacent fabric and soaked in artificial perspiration and the excess liquid was removed then the sample was dried under specific temperature, pressure and time. Finally the discoloration of the original dyed fabric and staining on adjacent sample were evaluated using gray cards [21]. ISO 105 E04-2013 test was carried out for perspiration fastness. Evaluation of fastness to perspiration showed good to excellent result towards staining on different fabrics in both acidic and alkaline medium. Colour change due to perspiration on cotton fabrics dyed with ferric alum, copper sulphate and ferrous sulphate showed very good result in both the medium.

Light fastness: Light fastness is the resistance to fading of dyed textile when exposed to day light. The high energy UV rays accelerate the fading of dye. The resistance of a dye to photochemical and chemical attack is an inherent property of the dye chromophore. The substituent present in the pigments play an important role in determining the light fastness properties. In the analysis of light fastness, the dyed sample is exposed to light for certain time (24 to 72 h) and the change in colour is compared with the original unexposed sample [15].

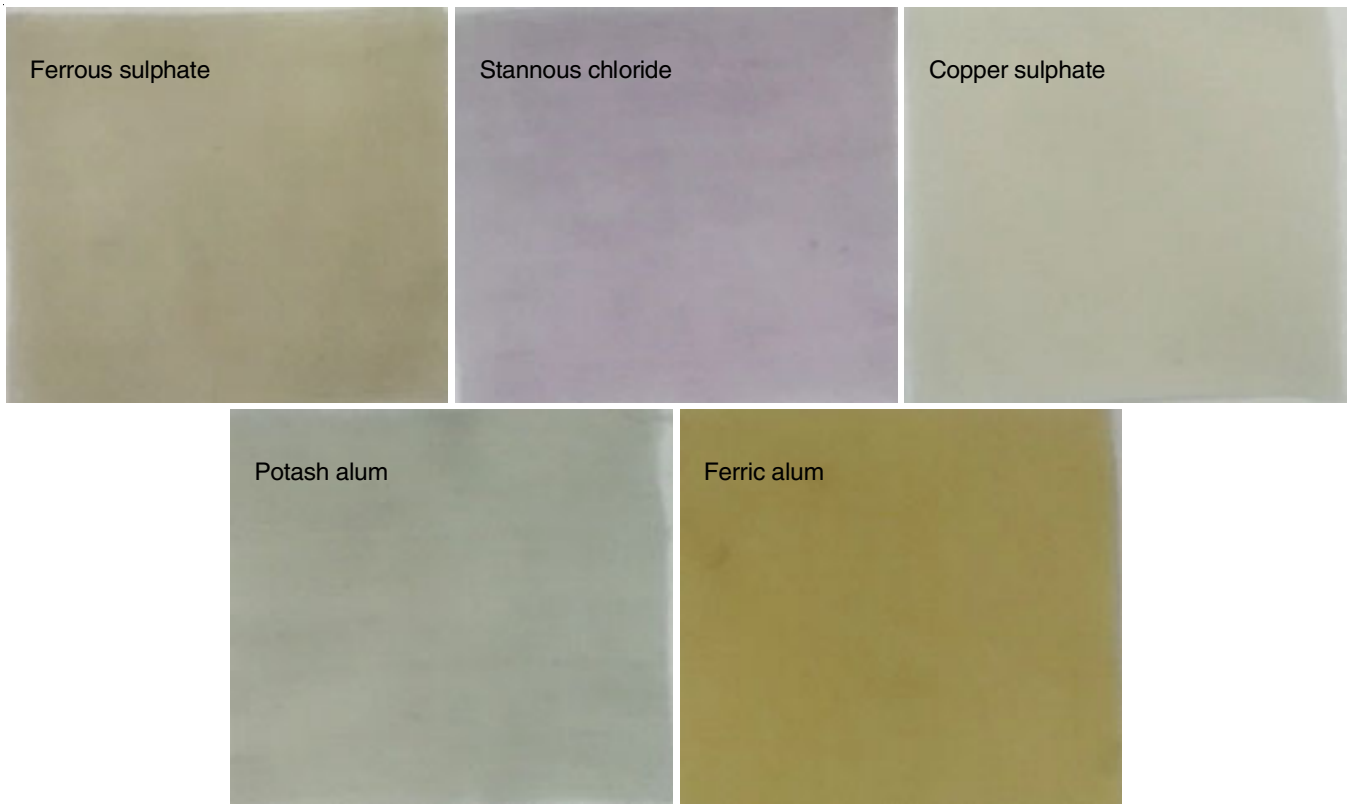

Fig. 1. Cotton fabrics dyed with the aqueous extract of *Psuderanthimum reticulatum*

Light fastness of the dyed fabric was assessed on 4-point scale (ISO 105 B02 Method 3:2014). Ferric alum, copper sulphate and ferrous sulphate mordanted cotton fabrics showed excellent light fastness properties whereas cotton fabric mordanted with stannous chloride and potash alum showed poor and little bit improved grade towards light fastness properties, respectively [22] (Table-2).

Identification of anthocyanin pigment

Paper chromatography: The R_f value of the coloured pigment of the methanolic extract of *Pseuderanthemum reticulatum* was calculated from the paper chromatogram using butanol:acetic acid:water (5:1:4) solvent system was found to be 0.46 and value indicates the presence of anthocyanin pigments [23].

UV and FTIR studies: The absorption peak observed at 521 nm in the double beam UV-visible spectrophotometer confirmed the presence of anthocyanin pigment present in the aqueous extract of *pseuderanthemum reticulatum.*

The IR spectrum of the extract was recorded using FTIR spectrometer is shown in Fig. 2. The leaf extract exhibited strong absorption band belonging to the saturated hydrocarbon groups (2951.09 cm^{-1} peak corresponds to methyl group, while 2839.22 cm^{-1} peak is due to methylene group). Absorption peak at 1646.28 cm^{-1} corresponds to carbonyl group. Several ther absorption bands were identified including those within the range of 1750-800 cm^{-1}, which are due to C=C-C aromatic ring. The IR region at 1542-965 cm^{-1} is usually referred to as the finger print region and various IR bands, including those corresponding to the vibrations of C-O, C-C and C-H bonds, occur in this region. These provide important information regarding organic compounds such as sugars, phenolic group. The deformation of C-C bonds in the phenolic group absorbed in the region at 1500-1400 cm^{-1} [24].

HPLC analysis: Anthocyanins present in the methanolic extract of *Pseuderanthemum reticulatum* was further confirmed by HPLC technique. The retention time observed in the chromatogram (Fig. 3) and its corresponding absorbance at 541.1, 548.4, 533.8 nm from the UV-visible spectrum index plot also confirmed the presence of anthocyanins, which are responsible for dyeing [25].

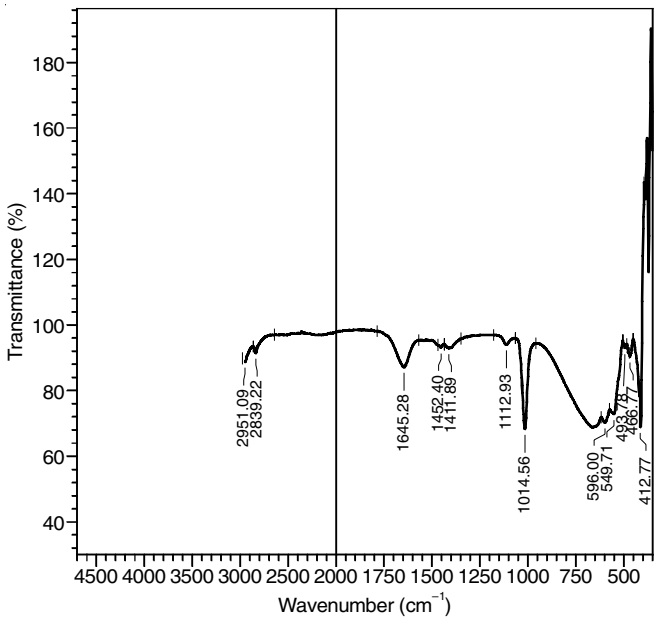

Fig. 2. FTIR spectrum of *pseuderanthemum reticulatum* plant extract

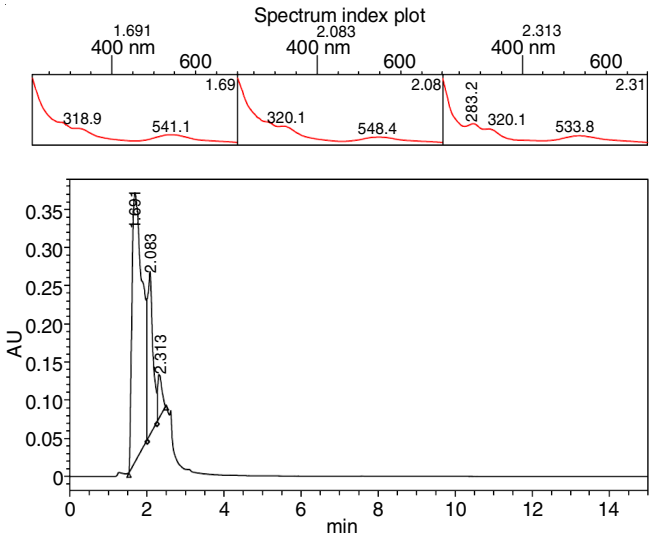

Fig. 3. HPLC chromatogram of *Psuderanthimum reticulatum* leaves extract

TABLE-2
COLOUR FASTNESS DATA ON DIFFERENT CONDITIONS

Mordants	Change in shade		Staining at different fabrics					
	Washing†	Light‡	Cotton		Polyester		Acrylic	
Ferrous sulphate	2-3	4	4-5		4-5		4-5	
Stannous chloride	1	1	4-5		4-5		4-5	
Copper sulphate	4	4	4-5		4-5		4-5	
Potash alum	1	2-3	4-5		4-5		4-5	
Ferric alum	3	4	4-5		4-5		4-5	
	Colour fastness to perspiration							
	Acidic*	Basic**	Acidic*	Basic**	Acidic*	Basic**	Acidic*	Basic**
Ferrous Sulphate	3-4	3-4	4-5	4-5	4-5	4-5	4-5	4-5
Stannous chloride	3	1	4-5	4-5	4-5	4-5	4-5	4-5
Copper sulphate	4	4	4-5	4-5	4-5	4-5	4-5	4-5
Potash alum	1	1	4-5	4-5	4-5	4-5	4-5	4-5
Ferric alum	4	4	4-5	4-5	4-5	4-5	4-5	4-5

†For Washing [ISO 105-C06 A2S-2010]; ‡For Light [ISO 105 B02 Method 3:2014].
*For acidic perspiration: [ISO 105 E04-2013]; **For basic perspiration: [ISO 105 E04-2013].

Conclusion

In the present work, cotton fabrics were dyed with the aqueous extract of *Psuderanthimum reticulatum* leaves with different mordents such as ferrous sulphate, stannous chloride, copper sulphate, potash alum and ferric alum. Different shades were produced after dyeing by premordanting technique. The standard (ISO) colour fastness tests were carried out and the results revealed that the cotton fabrics dyed with copper sulphate and ferric alum showed excellent to very good fastness properties towards washing, perspiration and light respectively. The ferrous sulphate mordanted fabric exhibited very good to good fatness properties in the entire fastness test. The principle of dyeing depends on the formation of stable complex between the natural pigment of the plant extracts and the metal of the mordant. Mordants form insoluble compounds of the dye within the fiber. Dyeing of cotton fabrics using *Psuderanthimum reticulatum* plant extract is an eco-friendly, safe and cost effective process. The natural pigment present in the extract which is responsible for the dyeing was anthocyanin and it was identified and confirmed by chemical test, paper chromatography, UV, FTIR and HPLC techniques. The present work gives more importance towards the utilization of eco-friendly dye for dyeing of fabrics in the textile industry using natural sources. It has been found that there is a lot of scope to use *Psuderanthimum reticulatum* as a potential dye in the textile industry.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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