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Extraction, Characterization and Application of Natural Dye Extract from Beetroot (*Beta vulgaris*) on Cotton Fabric

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Abstract

Using a Soxhlet system, natural dye was extracted from beetroot (Beta vulgaris) and applied to cotton fabric. With the use of FT-IR, GC-MS, and UV-visible spectroscopy, the extracted dye was analyzed. On the cotton cloth that had been dyed, the properties of scouring, wash fastness, and light fastness were assessed. Using a mordant, the cotton fabric was treated with the natural dye extract. The dye extract has a red colour. According to the FTIR characterization data suggested the presence of N-H, C-H, C=C, C-H, C-N and C-Cl functional groups in the dye extract. Fifty one compounds were isolated and identified by the GC-MS Melezitose, 5-Hydroxymethylfurfural and 4-Mercaptophenol has the higest percentage area (70.19 %) and Cis-10-Nonadecenoic acid, 9-Octadecenoic acid, (E)-, and cis-Vaccenic acid has the lowest percentage area (0.07%). N-H and C=C were the chromophores found in the dye extract.The color fastness to wasshing test revealed that the materials' fastness grade increased to a range of 3, indicating a fair grade of fastness, when the dye was applied using $SnCl_2$ as a mordant.While the color fastness to light showed that cotton fabric was dyed with mordants, the fastness grade increased to a range of 5, which indicates a moderate fading resistance, when the dye was applied to the fabrics using $FeSO_4$ as the mordant. Using $K_2Cr_2O_7$ as the mordant, the fastness grade slightly decreases to a range of 1, indicating very poor grade of fastness. The result of colour

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fastness and $SnCl_2$ light fastness of the dye demonstrated a moderate resistance to fading and a fair color look on cotton fabric. The colour fastness to rubbing showed that 3-4 and 2-3 colour changes were experienced for dry and wet rubbing respectively with mordant. The outcome of colour fastness of the dye showed a very good affinity to remain on the fabric when mordant was applied.

Keywords: Beta Vulgaris, Dye, Extraction, Cotton Fabric

INTRODUCTION

Since prehistoric times, natural dyes have been widely utilized to colour textile materials. 2600 BC is the earliest known written account of the usage of natural dyes, and it was discovered in China (Samanta and Agarwal, 2020). Concerns about environmental sustainability and the demand for healthier, more ethically manufactured products have led to the resurgence of natural dyes, which have their roots in ancient traditions and societies (Hannigan et al., 2022).

Societies, customs, and identities have been woven into textiles for millennia. These colors, which are made from minerals, plants, and even insects, promise a more environmentally responsible future while also providing a link to our historical past (Styles, 2014). As a result of these worries, natural dyes are becoming more and more popular again. Natural dyes offer a more eco-friendly option.

They are produced from biological sources such as roots, leaves, flowers, and barks. Customers that care about the environment will choose them because they are frequently non-toxic, biodegradable, and can be sourced sustainably (Abrahart et al., 2018). Furthermore, the revival of natural dyes protects indigenous wisdom, cultural legacy, and customary craftsmanship that have been woven into cloth dying for ages (Wang, 2019).

In this study, we investigate natural dyes, specifically the extraction, characterisation, and application of beetroot dye on cotton fabrics. By investigating this method, we hope to further the discussion regarding sustainable methods in the textile industry while also bringing attention to the potential of beetroot as a natural dye source. Natural dyes are often more environmentally friendly and have superior biodegradability.

According to Oenal et al., (2018) and Samanta et al. (2020),they are readily available, renewable, non-toxic, and non-allergic to skin. An increasing number of people are becoming interested in using natural colorants for dying textiles

In contrast to synthetic colorants, naturally derived colorants yield unique, calming, and gentle hues; yet, there are a few reasons why scientists and researchers are curious to explore the possibilities of naturally extracted colorants:

Natural colorants are widely used and have a lot of potential. There is experimental evidence that some synthetic dyes are toxic and allergic, while natural colorants are non-toxic and non-allergic. Specialty colors are created by skilled natural dyers using unique techniques, and there is scientific information available about the chemical characterizations of various natural colorants, including their extraction and purification. Finally, there is a knowledge base and database on the application of natural colorants on various textiles (Hudu et al., 2020).



Plate I: Beetroot (Beta vulgaris)

MATERIALS AND METHODS

Sample collection and preparation

Beetroot plants (Beta vulgaris) are cultivated in Plateau State, Jos Nigeria. The product was bought from Terminus Market, Jos and cotton fabrics were bought from wukari main market, Taraba state. The was washed thoroughly with distilled water, peeled and cut it into small pieces, shade-dried and pulverized using mortar and pestle, which was ready for extraction.

Soxhlet extraction

An analytical weighing balance was used to measure the sample, which weighed two hundred grams and was placed within the thimble, and a round-bottom flask that was fitted

inside the thimble for extraction received 200 mL of 100% methanol. The heating mantle was turned on, and sample underwent an extraction process lasting six hours at a temperature of 64.7 °C, or the boiling point of methanol. Excess solvent was removed using a rotary evaporator, leaving the dye in a dry form. For the extracted dye samples, the colour and percentage yield were ascertained, Nwonye & Priscilla 2017.

UV/Visible Spectroscopy

A quartz cell with a 1-cm route was filled with the dye extract, and the UV-Vis spectrophotometer was used to measure the results. Using a scan from 200 to 800 nm, the sample's distinctive absorption spectra were produced (Grifoni *et at.,* 2013).

GC/MS-Gas Chromatography mass spectrophotometer

The extracted dye was analyzed using a Shimadzu GC-2010 linked to an MS QP-2010. The Restec Rtx-5MS column, which had an ID thickness of 0.25µm and measured 60 mm by 0.25 mm, was packed with 95% dimethyl polysiloxane. A 1 µl injection volume and a 1 ml/min flow rate of helium gas were employed as the carrier gas.

FTIR spectroscopy analysis

The dye extract was measured with FTIR spectrophotometer using a Shimadzu IR prestige 21 spectrometers in the wavelength range of 400-4000 cm^{-1} Margaris, (2014).

Preparation of dye bath

The beetroot (Bete vulgaris) dye extract is soluble in methanol but insoluble in water. As a result, 1 g of the dye extract, measured into a 250 ml beaker, was dissolved in 5 ml of methanol and diluted to 55 ml in order to prepare the dye bath. Renu & Sangita, (2017). The dye extract, methanol, and water have a 1:5:50 ratio.

Mordanting method (pre-mordanting)

The mordanting will be carried out before dyeing of the cotton fabric. 3% concentration of mordant will be prepared which will be used to carry out the mordanting. The fabric will be immersed into 50 ml of the mordant solution for about 30 min at 60°C, after which the fabric will be taken out and squeezed by hand and then immersed in the dye bath. (Khin *et al.,* 2017).

Dyeing the cotton fabrics

One gram of the scoured and bleached fibers was added to the dye bath solutions previously mentioned. The materials were left in the dye-bath until they had absorbed the color after it had been gently heated. At 100 °C, the dyeing process started and continued for an hour. Following the conclusion of the dyeing procedure, the materials were taken out of the dye solution and given ten minutes to air oxidize before being rinsed with cold water to get rid of any loose dye particles that had stuck to the surface of the dyed materials. The fastness characteristics of the dyed materials were then examined after they had air dried, Cardon & Jansen (2012).

Colour Fastness Test

Wash fastness of dyed samples

The dyed specimens of dimension 5 cm x 4 cm were placed between two pieces of undyed white fabrics of the same dimension. Three pieces attached to each were held together by stitching round the edges to make a composite specimen. The composite specimen was agitated with 10 steel balls in a solution made-up of 5 g/l soap and 2 g/l soda ash with liquor ratio 1:50 as stipulated by ISO 3 standards. The washing was carried out at 60 °C ± 2 °C for 30 minutes in a launder-o-meter. The composite specimen was then rinsed, separated and dried. The change in colour of the test samples and the staining of the adjacent undyed white fabric was assessed with references to the grey scale (ISO 9001 2000 group). Onyesm, (2017).

Light fastness

Strips of the fabric and the blue wool standards were cut and mounted on a cardboard paper and half portions of the specimens were covered to obstruct the source of light from getting to that portion. The specimens were exposed due south at an angle of 45^0C sloping from the horizontal to natural day light for 72 daylight hours. The specimens were then removed after 72 daylight hours and the extents of their fading were assessed in comparison with the blue wool standards.

Rubbing fastness properties

First, the dyed fabric (4x2 cm) was cut, and then the white, undyed cloth (4x2 cm) was cut for both wet and dry rubbing. For ten minutes, the surface of the dyed fabric was rubbed with the white, undyed fabric during the dry rubbing process. The undyed white fabric was

first submerged in wet water and rubbed against the dyed fabric for ten minutes in order to perform wet rubbing Geetam *et al.,* (2017).

RESULTS AND DISCUSSION

Extraction of Beetroot plant (Beta vulgaris)

The dye from beetroot was extracted using the Soxhlet extraction method. Plate 2 displays the dye that was produced from beetroot. As can be seen from Table 1, the outcome of the beetroot dye extraction process required 200 g of the sample. Following the extraction, the sample's weight dropped to 124.64 g, and 25.36 g of dye were extracted. Due to Betalain the dye extract has a red colour and a percentage yield of 16.91% (Nwonye & Priscilla 2017., Aimable *et al.,* 2024).



Plate 1, Slide Beetroot Plate 2 Soxhlet apparatus Plate 3 Dye extracted from Beta vulgaaris

Table 1. Extraction of dye from *Beta vulgaris* (Beetroot)

UV-VIS Analysis for beta vulgaris content

A UV-VIS analysis was performed to assess the content of betalain in the dye extract. Figure 1 depicts the spectrum obtained after scanning the dye at wavelengths ranging from 200 nm to 800 nm. The highest absorbance was used to calculate the betalain content. The study's UV-VIS findings showed that a peak was present at 620 nm, indicating the presence of betacyanin in the dye extract, which is responsible for its red colour with maximum absorbance of 1.35_{Abs} as illustrated in Figure 1. This suggests that the chromophores C=C and N=H may be present. Shivani & Gunjan (2017).

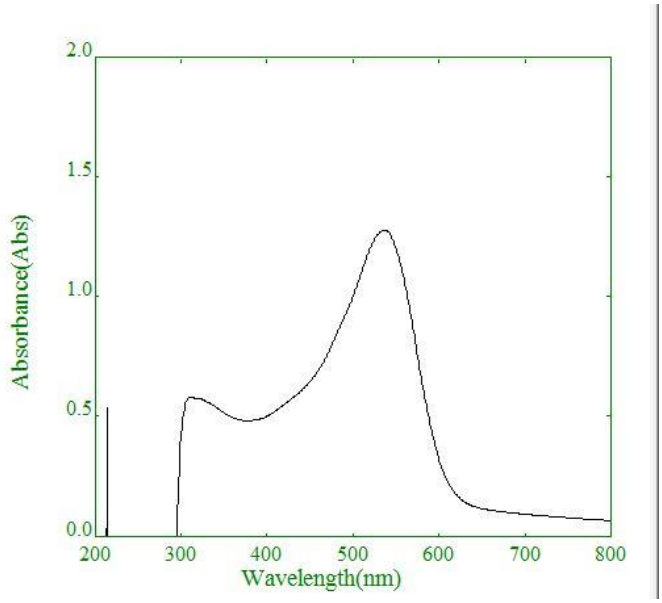


Fig.1 UV- Visible spectra of beetroot dye extract scanned in the range of 200cm-1 –

Sample	Samples before extraction (g)	Samples after extraction (g)	Amount of dyes extracted(g)	% Yield	Colour of the dye extracted
Beetroot	200	156.23g	38.64	19.32	Red

800cm-1 for the wavelength and 0.0 to 2.0 for the Absorbance of beetroot dye

GC-MS analysis of Dye extract from beetroot (*Beta vulgaris)*

The dye extracts obtained were subjected to GC-MS analysis for the determination of various volatile and semi volatile compounds in beetroot of *Beta vulgaris* the GC-MS examination of the dye extract from beetroot of *Beta vulgaris* led to the identification of 50 compound. The retention time (RT), % of peak area, molecular formula, molecular weight and the characterized compounds are listed in Table 2. The compounds identified from the dye extract are, Melezitose, 5-Hydroxymethylfurfural, 4-Mercaptophenol, had the highest

percentage area of (70.19%) and Cis-10-Nonadecenoic acid, 9-Octadecenoic acid, (E)-, and cis-Vaccenic acid has the lowest percentage area (0.07%). It is important to note that just because two or more compounds have similar retention times does not imply that they are the same. Furthermore, the interaction of different substances might enhance or decrease the time they remain inside the column. (Yakubu *et al.,* 2023).

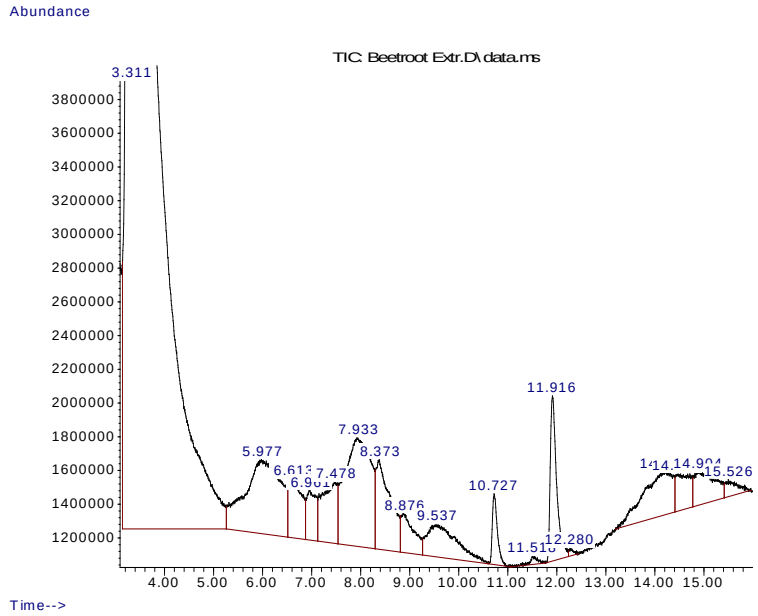


Fig. 2 GCMS spectra of beetroot dye extract scanned in abundance against time for the amount of substances in beetroot dye.

Table 2 GC-MS Interpretation

Peak	**Retention time**	**% of peaks area**	**Compound identified**	**Nature of compound**	**Molecular formular**	**Molecular weight**
1	3.310	70.19	Melezitose	Organic compound	$C_{18}H_{32}O_{16}$	504.4
		70.19	5-Hydroxymethylfurfural	Organic compounds	$C_6H_6O_3$	126.11
		70.19	4-Mercaptophenol	Organic compounds	C_6H_6OS	126.17624
2	5.976	5.44	Cyclohexanone, 4-ethyl-	Organic compounds	$C_8H_{14}O$	126.1962
		5.44	2-Piperidinone, N-[4-bromo-n-butyl]-	Organic compounds	$C_9H_{16}BrNO$	234.13
		5.44	d-Gluco-heptulosan	Keto sugar	$C_7H_{14}O_7$	210.18
3	6.612	1.55	Benzenemethanol, 3-fluoro-	Aromatic compound	C_7H_7FO	126.13
		1.55	Pyrazole-5-carboxylic acid, 3-methyl-	Hetrocyclic organic	$C_4H_6N_2O$	82.10

				compound		
		1.55	Cyclohexanone, 4-ethyl-	Aromatic compound	$C_8H_{18}O$	126.20
4	6.961	0.97	5-Acetoxymethyl-2-furaldehyde	Organic compounds	$C_8H_8O_4$	168.15
		0.97	4-Hepten-3-one, 5-methyl-, (E)-	Organic compounds	$C_8H_{14}O$	126.20
		0.97	4-Hepten-3-one, 5-methyl-, (Z)-	Organic compounds	$C_8H_{14}O$	126.20
5	7.476	1.85	2-Piperidinone, N-[4-bromo-n-butyl]-	Organic compounds	$C_9H_{16}BrNO$	234.13
		1.85	Beta.-D-Glucopyranose, 4-O-, Beta -D-galactopyranosyl-	Aldehyde compound	$C_{12}H_{22}O_{11}$	342.3
		1.85	Bicyclo[3.1.1]heptan-3-one, 6,6-di-methyl-2-(2-methylpropyl)-	Bicyclic ring compound	$C_{13}H_{22}O$	194.31
6	7.933	5.74	Cyclohexan-1-ol-2-amine, O,N,N-tri acetyl-	Aromatic compound	$C_{22}H_{23}NO_4$	365.42
		5.74	Beta.-D-Glucopyranose, 4-O-.beta-D-galactopyranosyl-	Aldehyde compound	$C_{12}H_{22}O_{11}$	342.3
		5.74	Beta.-D-Glucopyranose, 4-O-.beta-D-galactopyranosyl-	Aldehyde compound	$C1_2H_{22}O_{11}$	342.3
7	8.374	2.69	11-Octadecenoic acid, methyl ester	Fatty acid compound	$C_{19}H_{36}O$	296.4879
		2.69	Methyl 11-oxo-9-undecenoate	Ketone compound	$C_{12}H_{20}O_3$	212.28
		2.69	cis-13-Octadecenoic acid, methyl ester	Fatty acid compound	$C_{19}H_{36}O_2$	296.4879
8	8.877	1.05	Cyclohexan-1-ol-2-amine, O,N,N-tri acetyl-	Aromatic compound	$C_6H_{10}O$	98.14
		1.05	beta.-D-Glucopyranose, 4-O-.beta -D-galactopyranosyl-	Aldehyde compound	$C_6H1_2O_6$	180.16
		1.05	5,6,6-Trimethyl-5-(3-oxobut-1-enyl)-1-oxaspiro[2.5]octan-4-one	Organic compound	$C_{13}H_{16}O_4$	236.26
9	9.536	1.99	2-Methyl-Z,Z-3,13-octadecadienol	Organic compound	$C_{19}H_{36}O$	280.49
		1.99	2-Dodecen-1-yl(-)succinic anhydrid	Organic compound	$C_{16}H_{26}O_3$	266.38
		1.99	13-Octadecenal, (Z)-	Fatty aldehyde	$C_{18}H_{34}O$	266.5
10	10.726	0.74	Diisooctyl adipate	Organic compound	$C_{22}H_{42}O_4$	370.574
		0.74	Hexanedioic acid, bis(2-ethylhexyl) ester	Ester compound	$C_{22}H_{42}O_4$	370.6
		0.74	Hexanedioic acid, mono(2-ethylhexy-l)ester	Ester compound	$C_{22}H_{42}O_4$	370.6
11	11.521	0.13	2-Dodecen-1-yl(-)succinic anhydrid	Organic compound	$C_{16}H_{26}O_3$	266.3758

		0.13	cis-13-Octadecenoic acid	Fatty acid	$C_{18}H_{34}O_2$	282.5
		0.13	trans-13-Octadecenoic acid	Fatty acid	$C_{18}H_{34}O_2$	282.5
12	11.916	2.15	Di(E)-but-2-enyl phthalate	Organic compound	$C_{24}H_{38}O_4$	390.56
		2.15	Bis(2-ethylhexyl) phthalate	Organic compound	$\mathbf{C_{24}H_{38}O_4}$	390.564
		2.15	Phthalic acid, di(2-propylpentyl)ester	Organic compound	$C_{24}H_{38}O_4$	390.56
13	12.282	0.07	cis-10-Nonadecenoic acid	Fatty acid	$C1_9H_{36}O_3$	296.5
		0.07	9-Octadecenoic acid, (E)-	Fatty acid	$C_{18}H_{34}O_2$	282.5
		0.07	cis-Vaccenic acid	Fatty acid	$C_{18}H_{34}O_2$	282.5
14	14.216	2.64	Oleic Acid	Fatty acid	$C_{18}H_{34}O_2$	282.5
		2.64	1,2-Benzisothiazole, 3-(hexahydro-1H-azepin-1-yl)-, 1,1-dioxide	Fatty acid	$C_{13}H_{16}N_2O_2S$	264.34
		2.64	cis-13-Eicosenoic acid	Fatty acid	$C_{20}H_{38}O_2$	310.5
15	14.462	1.05	1,2-Benzisothiazole, 3-(hexahydro-1H-azepin-1-yl)-, 1,1-dioxide	Fatty acid	$C_{13}H_{16}N_2O_2S$	264.34
		1.05	cis-13-Eicosenoic acid	Fatty acid	$C_{20}H_{38}O_2$	310.5
		1.05	Fumaric acid, diundecyl ester	Organic compound	$C_{14}H_{48}O_4$	424.7
16	14.903	1.37	Oleic Acid	Fatty acid	$C_{18}H_{34}O_2$	282.5
		1.37	1,2-Benzisothiazole, 3-(hexahydro-1H-azepin-1-yl)-, 1,1-dioxide	Organic compound	$C_{13}H_{16}N_2O_2S$	264.34
		1.37	Oxacyclotetradecane-2,11-dione, 13-methyl-	Cyclic organic compound	$C_{14}H_{32}O_3$	240.34
17	15.526	0.39	Oleic Acid	Fatty acid	$C_{18}H_{34}O_2$	282.5
		0.39	cis-9-Hexadecenoic acid	Fatty acid	$C_{16}H_{30}O_2$	254.41
		0.39	8-Hexadecenal, 14-methyl-, (Z)-	Fatty acid	$C_{17}H_{32}O$	252.4

FTIR Analysis of dye extract from beetroot (*Beta vulgaris)*

The dye extract underwent Fourier Transform Infrared Spectroscopy to identify its functional groups. The scan was conducted in the 4000-500 cm^{-1} range, as illustrated in table 3. The resulting spectra showed a medium band at 3335.31 cm^{-1}, suggesting the existence of amine groups in the extract based on the N-H Stretching vibration. The peak observed at 2850.36 cm-1 was attributed to the stretching of C-H of the alkanes. The peak observed at 1615.50 cm-1 in the extract was assigned to the C=C stretching of the alkenes. Additionally, the peak at 14459.26 cm-1 was attributed to the C=C stretching of the aromatic compound present in the extract. The presence of C=C stretching of the aromatic. The peak observed at 1212.89 in the extract was assigned to the C=H wagging

stretching in the alkyl halides. The peak observed at 1032.58 cm-1 was attributed to the C=N of the aliphatic amines, as well as the C=Cl stretching, were responsible for the peak observed at 778.21 cm-1 hidden in the fingerprint. (Margaris, 2014., Aimable *et al.,* 2024).

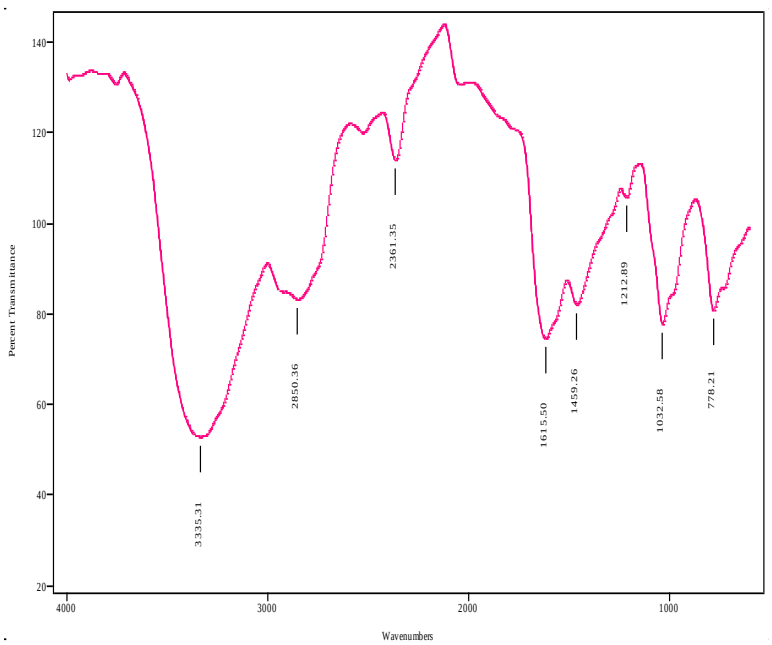


Fig 3 FTIR spectra of beetroot dye extract scanned in the range of 4000cm-1 – 500cm-1for the functional group of beetroot dye

Table 3 FT-IR Interpretation

S/N PRESENT	PEAK AREAS (nm)	POSSIBLE FUNCTIONAL GROUP
1.	3335.31	N-H (Amines)
2.	2850.36	C-H (Alkanes)
3.	2361.36	----------
4.	1615.50	C=C (Alkenes)
5.	1459.26	C=C (Alkenes)
6.	1212.89	C-H (Alkyl halides)
7.	1032.58	C-N (Aliphatic amines)
8.	778.21	C-Cl (Alkyl halides)

Wash Fastness properties of dye extract from Beetroot (beta vulgaris) on cotton fabric.

The results of the wash fastness properties of dye extract from Beetroot (*beta vulgaris*) on cotton fabric were presented in Table 3 The result showed that when the dye was applied

to the fabric using $SnCl_2$, as mordant the fastness grade slightly increases to a range of 3. This indicates fair grade of fastness. When the dye was applied to the fabrics using $FeSO_4$, as mordant the fastness grade slightly decreases to a range of 2. This indicates poor grade of fastness. When the dye was applied to the fabrics using $K_2Cr_2O_7$ as mordant the fastness grade slightly decreases to a range of 1, This indicates very poor grade of fastness.

Table 4 Wash Fastness Properties of dye extract from beetroot on cotton fabric

Fabric type	**Mordants**	**Wash fastness**
Cotton fabric A	$K_2Cr_2O_7$	1
Cotton fabric B	$FeSO_4$	2
Cotton fabric C	$SnCl_2$	3

Wash Fastness: 1 – very poor; 2 – poor; 3 – fair; 4 – good; 5 – excellent

Light Fastness of dye extract from Beetroot (*beta vulgaris*) on cotton fabrics

Table 5 shows the light fastness results of the dye extract from Beetroot (*beta vulgaris*) on Cotton fabric. The result show that when the cotton fabric was dyed with mordants which, the fastness grade increases to a range of 5 which indicates a moderate fading resistance.

Table 5 Light Fastness Properties of dye extract from beetroot on cotton fabric

Fabric type	Mordants	Light fastness
Cotton fabric A	$K_2Cr_2O_7$	1
Cotton fabric B	$FeSO_4$	3
Cotton fabric C	$SnCl_2$	5

1 – very poor; 2 – poor; 3 – fair; 4 – moderate; 5 – good; 6 – very good; 7 – excellent; 8 – outstanding.

Rubbing fastness properties of dyed fabric

The results of rubbing fastness qualities of cotton dyed samples using Beta vulgaris dye extract are shown in Table 6. It was discovered that dyeing the cloth with a mordant (both dry and wet) resulted in a rubbing fastness grade of 3-4, which indicates fair to good for dry rubbing, and a rubbing fastness grade of 2-3, which indicates poor to fair for wet rubbing. (Samanta and Agarwal, 2020., Yakubu *et al.,* 2023)

Table 6. Rubbing fastness properties of dyed fabric

Fabric Type	Dry rubbing	Wet rubbing
Dyed fabric	3-4	2-3

Rubbing fastness; 1- very poor; 2- poor: 3- fair; 4- good; 5-excellent

CONCLUSION

Natural colourants were successfully extracted from beetroot plant using soxhlet techniques. The dye extracts were applied on cotton fabrics with mordants. The results obtained in this study suggest that the natural dye extracted from beetroot plant possess intrinsic affinity for natural (cotton) fabric. However, the addition of mordants improved the fastness performance of the dye extract on the fabrics. Stannous chloride was preferred as a potential mordant because of its ability to produce better dye uptake on the fabrics.

Characterization of the dye extract revealed the presence of N-H (Amines), C-H (Alkanes), C=C (Alkenes), C-H (alkyl halides), C-N (Aliphatic amines) and C-Cl (alkyl halides) are functional groups in the dye components. The chromophores presence in the dye extract are C=C and N-H.

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