Assessment of the Dyeing and Antimicrobial Activity of the Extract of the Root Bark of Cochlospermum Tinctorium Oncotton, Silk and Polyesterfabrics Usingal₂so₄, Cuso₄ and Feso₄ as Mordants

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Abstract: The root bark of Cochlospermumtinctorium (Rawaya) is used in the treatment of various disease in man such as diabetes, yellow and malaria feverin Nigeria but in other parts of the world, is used against intestinal worms, bilharzia and gastro-intestinal disease like ulcer. Cochlospermum tinctorium (Rawaya) is a savannah plant found in farm across Northern Nigeria. In the present study, the dye extracts obtained from the bark of C.tinctorium was used in the dyeing of cotton, silk and polyester fabrics. The fabrics were assessed for antimicrobial and fastness properties. The antimicrobial assessment was performed quantitatively by disc diffusion method, parallel streak method and quantitatively by percentage reduction test against the test organisms Escherichia coli and Staphylococcus aureus. The fabrics were also assessed for fastness properties such as wash fastness and light fastness as per the AATCC standards. The natural extract hasantimicrobial activity with no clear growth of bacteria under them .However ,the dyed fabrics alone showed uninhibited bacterial growth This is in contrast when mordants were applied where reduction in percentage of E.Coli and SAureus is varied with different and amount of mordants. This showed that using mordant had better antibacterial activity because it is well known that metallic salts exhibit toxic effects against pathogens.

Date of Submission: 01-03-2019

Date of acceptance: 18-03-2019

I. Introduction

Nigeria has a rich plant biodiversity and one of the biggest in the world, with so many plant species of the plant kingdom, Nigeria is treasure house of diverse natural product. One of such product is the natural dye and found as a pigment in leaves, fruit, seed and roots. In addition to their yielding characteristics some of them possess medicinal value.

The plants exhibit a wide range of colors and not all the pigment can be used as dyes. Some of the dyes are not water soluble and cannot be absorbed on fibers and leather other, fade when exposed to sun light.

Due to lack of technical knowledge of extraction and dyeing technique, the applications of natural dyes have not been commercially successful. In addition due environmental legislation to convert toxic waste from effluent discharge of textile companies that are toxic to human and animals, the need to shift to the more environment friendly method of dyeing fabrics[Ado etal,2015].

Dyer (1976) indicates that the effectiveness of natural dyes differ with each plant, with distinct differences in the color obtained at different times of the day. Some may require mordant to improve their fastness but others may be used as direct dyes on fiber. Natural dyes also have various exceptions[Lokhande etal,1998;1999]: they are not substantive, with little or no coloring power in themselves except when used in conjunction with mordants although most of them produce very colorful effects so amazing to behold [Kundal,2015]. Natural dyes initially appear vivid but they soon fade; very few of them prove to be colorfast. It is this fastness problem that led to the discovery of mordants ;natural acids and oxides that react both with the dyestuff and the fibers to form an insoluble compound that "fixes" the color firmly in the fibers and prevents the dye from dissolving easily. Some natural dyes also require mordants to make them colorfast.

Natural dyes are known for their use in coloring of food substrate, leather as well as natural protein fibers like wool, silk and cotton as major areas of application since pre-historic times [Baraji etal,2015;Collier etal,2001]. The use of non-allergic, non-toxic and eco-friendly natural dyes on textiles have become a matter of importance due to the increased environmental awareness in order to avoid some hazardous synthetic dyes (Elnagar etal, 2014). However, worldwide the use of natural dyes for the coloration of textiles has mainly been

confined to artisan / craftsman, small scale / cottage level dyers and printers as well as to small scale exporters and importers producers dealing with high-valued eco-friendly textile production and sales [Argawal,2009]. Recently, a number of commercial dyers and small textile export houses have started looking at the possibilities of using natural dyes for regular basic dyeing and printing of textiles to overcome environmental pollution caused by the synthetic dyes. Natural dyes produce very uncommon, soothing and soft shades as compared to synthetic dyes. The present study examines the performance behavior of the root bark of cochlospermum tinctorium in the dyeing of fabrics and its antimicrobial activity on the fiber.

II. Materials Method

General Description

Scientific Name: Cochlospermumplanchonii

Family: Cochlospermaceae

Cochlospermumtinctorium is a bushy plant attaining about 50cm in height. It has widespread occurrence in the savannah and shrub land throughout the drier areas of West Africa region the common vernacular names in Nigeria include Rawaya,Kyamba (Hausa), Obazi (Igbo) and Sewutu (Yoruba).

Botanical Description

Shrubwith woody subterranean rootstock from which, in the rainy season, annual, leafy shoots up to 2.5 m tall are produced. Leaves alternate, palmate (3-)5-lobed; stipules sub late; petiole up to 10 cm long; blade in outline, base cord ate to cuneate, lobes oblong, basally connate for half to two-thirds of their length, apex rounded, margin entire, rarely dented, upper surface dark green and almost glabrous, lower surface paler and soft-hairy. Inflorescence terminal, with 3-7 fascicled branches, rarely lax; bracts triangular. Flowersbisexual, actinomorphic, 5-merous; sepals unequal, elliptical-oblong to broadly ovate, the outer 2 shorter than the inner 3,usually velvety; petals obviate, shallowly emarginated, golden yellow; stamens numerous; ovary superior, globes, style elongate, linear. Fruit 3-5-valved capsule, ovoid, or pyriform. **Seeds** reniform, black, with loosely attached, long, white hairs. *Cochlospermum planchonii*flowers towards the end of the rainy season. Fruits are produced 1-2 months after flowering.

III. Dye Extraction

Aqueous Extraction

To prepare the natural color on the trial plant, the extraction technique of korankye et al, 2015 was adoptedIn the aqueous technique,Cochlospermumtinctorium (Rawaya) (10g) was grinded to powder and put in a beaker to which water (300ml) was added, kept undisturbed for 2, 3 and 5 days to get maximum extracting of the colorant.. Note the beakers were covered with a kitchen foil and left in a cool place. The extracts were used in the dyeing experiment and filtered, diluted and examined under UV to obtain the maximum wave length of absorption. It is the extract with highest o maximum wave length value that was used in the dyeing experiment[Shahin etal2014].

Ethanolic Extraction

To prepared ethanolic extract, ethanol (100ml) was added to the root of Cochlospermumtinctorium (Rawaya) (20g) in a sealed conical flask and kept at room temperature for 48 hours. The solution was filtered to obtain a clear filtrate to remove ethanol. The filtrate was condensed through a rotary vacuum evaporator at 60° C for 30mins. The ethanol free filtrate was used for the preliminary assessment of antimicrobial activity. Preliminary Assessment of Antimicrobial Activity of Root Bark ofCochlospermumTinctorium (Rawaya) Extract The antimicrobial activity of the crude extract of Root of CochlospermumTinctorium (Rawaya) was assessed preliminary by disc diffusion method. Filter paper disc (diameter 2 ml)was prepared by treating with both aqueous and ethanolic extract (100ml) and allowed to air dry. The discs were placed in intimate contact with bacteriostatic agar, which has been previously inoculated with the test organisms. Two test organisms namely Staphylococcus Aureus and Escherichia coli were used in the study. The plates were incubated at 37° C for 18-24 hours and the presence of a clear zone of inhibition indicates the antimicrobial effectiveness of the extracts.

Preparation of Fabrics

The starch content in fabrics can be removed by scouring the fabric with pre weighted soda ash (0.5g) and washed in a solution container (equivalent of 2% of weight of fabric) and detergent at 5.5% wt. of fabric at 1:4 material to liquor ration added to the water and boiled at 80°C and stirred well. The stirring was continuous for 2-3 minute at 85°C. The scoured material is washed with tap water and dried at room temperature.

Bleaching

Colored material or threads containing coloring matter must be removed from substrate. Bleaching is to discolor the organic material giving the color (destroying the chromospheres). This was achieved by treating the scoured cloth with ascorbic acid (4g) in NaOCl (3ml) in water (200ml) at 90°C for 10 minutes. The fabric was washed with distilled water and dried for dyeing or mordanting.

Mordanting

Mordanting of fabrics is performed before or after the dyeing. In general $k_2Cr_2O_7$, $Al_2(SO_4)_3$, $CuSO_4$ or $FeSO_4$ 2% -6% is dissolved in distilled water (100ml). The scoured cloth is dipped into the mordant solution and the temperature raised to dye bath 80°C for 30 minutes .The cloth is then cooled, washed and air dried. In dyeing of fabric with mordant.

Dyeing

The fabric cloth and thread samples are dyed with extracted color. The dye extract prepared by adding of natural dye extract (10ml) in the deepest shade in water (ML)(1:20) (200lm) and the temperature is raised gradually to 80oC and maintained with continuous stirring for 45minutes. The fabrics are removed from the dye bath and washed.

Post mordanting

In the post mordanting process the fabrics cloth and thread sample are dyed with natural plant extracted using M:L ratio 1:20. The dye bath is heated to 80°C and temperature maintained for 45 minutes with continuous stirring. Then the fabrics are removed washed and dried. Wet sample of fabric are dipped into the mordant solution and brought to a temperature of 80°C for 30 minutes. Then cooled for 15 minutes, washed and dried with water and air dried

Effect of Dye without Mordant

The effect of dye without mordanting the fabric was also studied. Then the fabric was treated withtopol(Color fixative) and dried in sunlight. Different cloth and thread were experimented for cotton dyeing. Cloth, polyester, cotton thread and silk thread were used in the experiments to observe the strength of dye. Similarly the effect of various mordants on color of dye extracted from the root of Cochlospermumtinctorium (Rawaya)were also studied on cloth and thread found best in the above experiment. This was achieved by incorporating different mordant like $Al_2(SO_4)_3$, CuSO₄ or FeSO₄separately, each at a concentration of 3% of the dye extract (5ml).Cloth and thread pieces were individually soaked with the mixture of extract-mordant solution.

Antimicrobial Activity Assessment of Dyed Cotton Fabric Qualitative Assessment by Agar Diffusion Method (SN 95920-1992)

Fabric dyed with Root of Cochlospermumtinctorium (Rawaya) and un dyed cotton material samples were placed in close contact with bacteriostatic agar which has been previously inoculated of test organism inpetridish. Two test organisms namely *staphylococcus aureus* and *Escherichiacoli* were used for the study. The plates were incubated at 37^oC for 18-24 hours. After incubation a clear area of uninterrupted growth underneath and alongside test material is measured aszone of inhibition in mm.

Qualitative Assessment in Parallel Streak Method (AATCC 147-2004)

Sterile bacteruiostasis agar was dispensed in Petridis 24 hours broth culture of the test organism (SAureus and E.coli).using 2mm inoculation loop, 1 loop full of culture was loaded and transfer to the surface of the agar plate by making 7.5cm long parallel streaks 1cm apart in the center of the plate without refilling the loop. The test specimens are gently pressed transversely, across the 5 inoculums of streaks to ensure intimate contact with the agar surface. The plates are incubated at 37° C for 18-42 hours. After incubation a streak of interrupted growth underneath and along the side of the test material is measured zone of inhibition in mm.

Quantitative Assessment by Percentage Reduction Test (AATCC100-2004)

Specimens of the test material were shaken in a known concentration of bacterial suspension and the reduction in bacterial activity in standard time was measured. The efficiency of the antimicrobial treatment is determine by comparing the reduction in bacterial concentration of treated sample with that of control sample expressed as a percentage reduction in standard time

Percentage reduction = $A-B/B \times 100$

A and B = concentration of surviving cell (CFU/ML) for the flasks containing control (blank cotton fabric) and test sample (natural dyed treat fabric) after 18 hours contact time.

Wash Fastness (AATCC-110-2006)

Fabric sample of size 6 x2Cm was taken and staple along with fresh piece on the technical face distilled water (150ml) and Detergent (0.0225g) are added to each beaker containing steel ball in metal container and then closed and rotated for 2minutes at 140° C.The sample is than added and rotated for 5 minutes. The sample is rinsed with water 3times and then excess water is removed. The sample is dried at 40° C.For one hour and evaluated.

Light Fastness (AATCC Test Method 16-1993)

Each treated and untreated fabric sample is exposed to sun light for 10 hour. After that the change in color of the test fabric sample is evaluated with the geometric grey scale and the numerical ratings are assigned for both treated and untreated fabric.

Acidic and Alkaline Perspiration (IS: 971-1956)

The acidic test liquor is prepared by dissolving NaCl (2.65g) and urea (0.75g/liter) and pH adjusted to 5.6 with acetic acid. The alkaline test liquor is prepared by dissolving NaCl (3g/liter) and pH adjusted to 7.5 with NaHCO₃. The composite specimen is then wetted thoroughly in acidic test liquor using liquor to specimen ratio 50:1 (mg) and allowed to retain in the liquor for 30 minute at room temperature. The specimen must be uniformly saturated. The liquor was poured 0ff and the specimen is placed between two glass plates under a face of 4.5kg. The plate is placed in a hot oven for 4 hour at 37°C. at the end this period the specimen is removed and test piece are separated, dried at 60°C. The alkaline procedure is repeated as above with composite. The change in color of test piece (alkaline and acidic) and degree of staining is evaluated with the geometric gray scale and numerical rating.

IV. Results And Discussion

Assessment Of Dye Up Take By Cotton And Polyester Fabrics, Cotton And Silk Threads

The application of dye in textile industry has been for the purpose of dyeings yarn ,woven cloth, cloth warp earlier e.t.c. thus aspect of dyeing is not only in demand in textile industry but also in leather, food and pharmaceuticals. The rich bioderversity of Nigeria should provide the raw materials for subtutinable linkage to be devided between collection and usage. The present study investigates the extraction of dye from the root of cochlospermum tinctorium and its application in the dyeing of cotton and polyester fabrics, cotton and silk threads in the presence of $(Al_2SO_4)_3$, CuSO₄ and FeSO₄ as mordants. The lack of any literature report on the use of cochlospermum tinctorium and its application in the dyeing of fabric with mordant or its chemical composition further necessatated the work.

S/N	Sample	Color obtained with	Color obtained with	Color obtained with	Color obtained with
		treatment of dye without	treatment of mordant FeSO4	treatment of mordant	treatment of mordant
		mordant		CuSO ₄	$(Al_2SO_4)_3$
1	Cotton fabric	yellow Brown	Dark Ash	Brownish brown	Pink
2	Polyester	very light brown	Light Ash	Brown green	Pale Pink

 Table 1 Effect of different mordant with dye extract from cochlospermumtinctorium on cotton and polyester fabrics

 Table 2 Effect of different mordant with dye extract from cochlospermumtinctorium on cotton thread and silk

thread

S/N	Sample	Color obtained with	Color obtained with	Color obtained with	Color obtained with
		treatment of dye without	treatment of mordant	treatment of mordant	treatment of mordant
		Mordant	FeSO ₄	CuSO ₄	$(Al_2SO_4)_3$
1	Cotton thread	Orange	Dark Ash	Dark green	Pink
2	Silk thread	Shiny orange	shiny Ash	Shiny green	shiny Pink

When the cotton, polyester ,cotton and silk threads were treated with the extract of cochlospermum tinctorium, the colours obtained are given in Tables1 and 2. The colour shade was yellow brown for cotton and light brown for polyester . The coloration of the cotton thread was orange while silk thread appeared as shiny orange. The choice of extract for coloration of fabrics was made using extract with maximum absorption. The dyebath were prepared from those extract for the dyeing and post mordanting process. When the effect of mordants on the dyeing of fabrics was investigated using $(Al_2SO_4)_3$, $CuSO_4$ and $FeSO_4$ the results obtained are given in Tables1 and 2... In all the dyeing process post mordanting procedure was examined because the dyestuff is bound stronger than that of the pre mordanting exercise where it is weakly held. The purposeof mordanting is to bind the dyestuff on the fiber to form a complex with the resultant increase in dye uptake. In thus way the metal ions act as electron acceptors from the electron donating group of dye to form

coordination bond with hydroxyl group of cellulose as well as the dye. The color obtained after mordanting depends on the nature of metal salt for complex formation and in thiswork the coloration was as follows: The cotton fabric changed to Ash in the presence of $FeSO_4$; brownish green (CuSO₄)and finally to a pink colored material with (Al₂SO₄)₃. The polyester was light to pale colored materials were obtained with $FeSO_4$, CuSO₄ and (Al₂SO₄)₃. The cotton thread changed fro brown to Dark ash, Dark green to pink while silk thread change from shiny, light green to shiny pink.

S/N	Sample	Wash fastness	Change in color	Assessment
1	A1	4	Very slight alteration	Very good
2	A2	2	Distinct alteration	Fair
3	A3	4	Very slight alteration	Very good
4	A4	1	Great alteration	Poor
5	B1	3	Appreciable alteration	Good
6	B2	4	Very slight alteration	Very good
7	B3	1	Great alteration	Poor
8	B4	1	Great alteration	Poor
9	C1	3	Appreciable alteration	Good
10	C2	2	Distinct alteration	Fair
11	C3	1	Great alteration	Poor
12	C4	1	Great alteration	Р
13	D1	4	Very slight alteration	Very good
14	D2	4	Very slight alteration	Very good
15	D3	1	Great alteration	Poor
16	D4	4	Very slight alteration	Very good

Table 4 Wash Fastness of theMordanted and Unmordanted fabric

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S/N	Sample	Light fastness	Assessment
1	A1	1	Poor fastness
2	A2	3	Moderate fastness
3	A3	3	Moderate fastness
4	A4	3	Moderate fastness
5	B1	1	Poor fastness
6	B2	2	Slight fastness
7	B3	2	Slight fastness
8	B4	3; C	Moderate fastness

Key: A1 Cotton Dyed with Extract ;A2 CottonDyed with FeSO4;A3 Cotton Dyed with $AL_2(SO4)_3$; A4 Cotton Dyed with $CuSO_4$; B1 Polyester Dyed with Extract; B2 Polyester Dyed with $FeSO_4$; B3 Polyester Dyed with $AL_2(SO4)_3$; B4 Polyester Dyed with $CuSO_4$; C1 Silk Thread Dyed with Extract ;C2 Silk Thread Dyed with $FeSO_4$; C3 Silk Thread Dyed with AL_2SO_4 ; C4 Silk Thread Dyed with $CuSO_4$; D1 Cotton Thread Dyed with $FeSO_4$; D2 Cotton Thread Dyed with $FeSO_4$; D3 Cotton Thread Dyed with Al_2SO_4 ; D4 Cotton Thread Dyed with $CuSO_4$ with $CuSO_4$

Fastness Characteristics

It is reported in the literature (Dyer, 1976) that the effectiveness of natural dyes differ with each plant, with distinct differences in the color. It is also widely understand that natural dyes are not substantive (Rosenberg,) with little or no coloring power and have poor to moderate light and wash fastness characteristics compared to synthetic dyes which are good, and can be better when used in conjunction with mordant. Poor light fastness to moderate was observed for cotton fabric with the extract and mordant. In general the variation is more pronounced in the case of polyester (Table 5). Poor fastness characteristics is attributed to photo oxidation of the dye chrome molecule and can be improved by application of cationic mordants that can bound the dye molecule on the fabric. The purpose of mordanting is therefore to increase the uptake of a dye through formation of a complex between the metal, dye and functional group of the fabrics. Cotton, polyester, cotton thread and silk thread have hydroxyl, amide or carbonyl groups on the fabric which can be occupied by the metal ions as electron acceptor from the fiber functional group. The interaction increases the uptake and better coloration and fastness.Where light fastness is slight or poor (Table 5) indicated that complexation does not much help which is a common feature of natural dyes. When the wash fastness of the mordanted and un mordanted fabric was investigated in (Table 4) was obtained and the ratings were good to fair for samples (A2, B2, C2 and D2), appreciable to great alteration for samples (A4, B1, B3, B4, C1, C3 and D3) and very good wash fastness characteristics for samples (A1, A3, B2, D1, D2, and D4) treated with extract and FeSO₄, CuSO₄ and (Al₂SO₄)₃ as mordants. Whencolor fastness to perspiration was investigated, the results obtained are given in Table 6. The response to perspiration was good for acid and alkaline environment moderate for polyester fabrics.

S/N	Color fastness to perspiration	Color fastness rat	ing of sample dyed with	Assessment
		extract		
		Alkali	Acid	
1	Cotton fabric	4	4	Good
2	Polyester	3	3	Moderate

Table 6 Assessment Of perspiration

Preliminary Assessment of Antimicrobial activity

When the preliminary assessment of antimicrobial activity 0f the extract of Cochlospermumtinctorium was investigated in aqueous and ethanolic solution, the result in Table 3 was obtained and the highest zone of inhibition was withethanolic solution for both bacteria and low in aqueous solution. In the overall, the response to treatment was highest with *S.aureus* in both solution.

Table 5 Prenninary Assessment of Antimicrobial Activity of Natural Dye				
S/No	Extract of Root of Cochlospermumtinctorium (Rawaya)	Antimicrobial activity		
		(zone of inhibition in mm)		
		E.Coli	S.aureus	
1	Aqueous	6	7	
2	Ethanolic	10	13	

Table 3 Preliminary Assessment of Antimicrobial Activity of Natural Dye

Qualitative Assessment by Agar Diffusion Method (SN 95920-1992)

The result of agar diffusion method against the test organisms *S.aureus* and *E.coli* indicated that there was no zone of inhibition but displayed clear growth of bacteria for both treated fabric and untreated fabric. Investigated antimicrobial activity showed that untreated and treated cotton fabric against *E.coli* and *S.aureus* does not inhibit microbial activity.

Qualitative Assessment in Parallel Streak Method (AATCC 147-2004)

For qualitative assessment of microbial activity, cotton fabric treated with extract only and fabric treated with extract and mordant were subjected to parallel streak test. Untreated cotton fabric showed clear growth of bacteria under them with no zone of inhibition, indicating that the cotton fabric itself does not inhibit microbial activity. However Treatment with extract and mordant inhibit microbial growth.

Conclusion

Natural dyes have now entered a new era where they are required and used for many intrinsic value. Local plant are one sources of natural colorant grouped under plant sources. In this worh the c colouration of cotton and polyester fabrics and cotton and silk threads have improved in the presence of mordants but more noticeable with $FeSO_4$, Al_2SO_4 which is deeper in shade or pink colored. This suggests that $FeSO_4$ has greater coordination power and therefore high dye uptake. The fastness characteristic is fair to moderate but the best performance is dying of cotton with ferrous sulphate The greater the coordination of metal ion on the fabric, the better the wash fastness and thus behavior has not been reported in the literature using the extract of cochlospermum tinctorium. A full ingestigation of the aqueous and ethanolic root extract was conducted on bacterial specimen E.coli and S.aureus with the fabric. The fabrics dyed with root of cochlospermum tinctorium that was treated with mordant displayed clear inhibition of growth of bacteria which suggest that the extract of root material can significally facilitate the production of quality fabrics that are safe to used[Mohammed etal;2013].

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IOSR Journal of Applied Chemistry (IOSR-JAC) is UGC approved Journal with Sl. No. 4031, Journal no. 44190.

Ibrahim Sada. " Assessment of the Dyeing and Antimicrobial Activity of the Extract of the Root Bark of Cochlospermum Tinctorium Oncotton, Silk and Polyesterfabrics Usingal2so4, Cuso4 and Feso4 as Mordants." IOSR Journal of Applied Chemistry (IOSR-JAC) 12.3 (2019): 29-35.