

## Characterisation of Colourant Extracted from Riped Cashew Fruits

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### ABSTRACT

*This study reports the extraction of colourant from ripe cashew fruits (*Anacardium occidentale*) using ethanol, methanol and water as solvents. The research is also to determine the constituents of the ripe cashew fruits, as well as the functional groups of the constituents. Distilled water was found to be the most suitable solvent for the extraction of the dye from the cashew fruits. Characterization of *Anacardium occidentale* was done using infrared and ultraviolet spectrophotometry. The result reveals the type of chromophores in the active dye compound. The observed absorbance revealed the highest values at the 533.0nm for methanol, 740.0nm for ethanol and 731.5nm for distilled water. The greater volume of compounds present in the different extracts from methanol, ethanol and distilled water is derivatives of benzene and naphthalene. However, derivatives of ketone was also observed from the ethanol and methanol extract. The maximum wave length 533nm, 740.00nm and 731.50nm reveal the chromophoric groups necessary for the formation of colours {mixtures of yellowish and reddish brown}. From the spectroscopic results of the extract derivatives of ketone, benzenes and naphthalenes were evident. Other groups present include C-H, C=O, C=C, N-H, O-H, C=C, =C-O-C, C=N, and CH<sub>3</sub>. It is therefore concluded from the results that anthraquinone, naphthaquinone and indigoid dyes could be present in the extract of the cashew fruit.*

### INTRODUCTION

Although dyeing of textile fabrics with dyes obtained from various natural resources has been extensively investigated, little information is available on the identification and characterization of the natural dyes<sup>1</sup>. Natural dyes, being plant metabolites, are present only in small amounts in dye-bearing materials along with large quantities of other nondye materials. The dye content may vary according to the age, part of the plant, and agroclimatic conditions<sup>2</sup>,

and it is important to know the dye content in order to get reproducible shades. While procuring the dye materials, pricing should match the dye content and when powdered dye materials or extracts are used, these should be authentic. Thus determination of dye content as well as characterization of dye material is important in the case of natural dyes. Absorption spectroscopy is very successfully used for measuring<sup>3</sup> the dye content of synthetic dyes but has limited applicability for natural dyes as these dyes are usually not a single chemical entity but a mixture of closely related compounds and in many cases there are no clearly defined

absorption maxima<sup>4</sup>. A literature survey shows that the earliest attempts to characterize the natural dyes were made in the context of identifying the dyes present on historical textiles kept in museums or those found in archaeological excavations. Different techniques including high performance liquid chromatography (HPLC), thin-layer chromatography (TLC), high-performance thin-layer chromatography (HPTLC), UV visible, and mass spectroscopy have been employed for this purpose<sup>5,6</sup>.

Characterization is a process of identifying various chemical bonds that are present in dye (colourant) e.g. auxochromes and chromophores in the plants extract using IR, UV, liquid chromatography, spectrophotometry. For the fact that plants extracts usually occur as a combination of various types of bioactive compounds or photochemical with different polarities, their separation still remains a big challenge in the process of identification and characterization of bioactive compounds<sup>7</sup>. It is a common practice in isolation of these bioactive compounds that a number of different separation techniques such as TLC, column chromatography, flash chromatography, sephadex chromatography and HPLC should be used to obtain pure compounds. The pure compounds are then used for the determination of structure and colorimetric activity. Besides that, non-chromatographic techniques such as immunoassay, uses monoclonal antibodies (MABS), photochemical screening assay, fourier – transform infrared spectroscopy (FTIR), can also be used to obtain and facilitate the identification of these compounds.

Several studies in the last ten to twenty years ago have characterized and promoted the use of natural dyes from different plant species<sup>8,9</sup>

partly because of recent scientific development in instrumental method of colour measurements, analysis and structure determination<sup>10</sup>. Consequently, many potential dye-yielding plants and major dye components have been identified through colorimetric and spectroscopic investigations<sup>11</sup>. Based on instrumental approach, several dye yielding constituents exhibiting different colours, have been demonstrated in plants<sup>12</sup>. The identification of natural dyes found in plants and how to extracts dyes from them have been a major interest. The manufacture of synthetic dyes has been found to be energy intensive with adverse impact on environment, in addition to its pollution, serious health hazards like allergy, have been associated with the synthetic dyes.

This study is aimed at determination of the chemical constituents of colourant from ripe cashew fruits through characterization of the colourant extract which can lead to formulation of its structural formula

## **MATERIALS AND METHODS**

### ***Collection of raw Material***

The fruit of cashew (*Anacardium occidentale*) were collected in April when the fruits were predominantly available. Due to the fact that there was abundant sample of the cashew fruits (*Anacardium occidentale*), sample was collected when it was ripe and immediately processed.

### ***Identification the Material/Sample***

The material, cashew fruits (*Anacardium occidentale*), that was used for the study was identified by using the standard botanical identifying features (SBIF).

### **Extraction of Colourant**

The samples were reduced—by cutting the cashew fruits into smaller size and putting them in an electric blender which grinds the cashew fruits until it becomes fluidy. It was poured out from the blender into a bowl, sieved in order to extract the liquid (colourant extracts). The colourant extract was divided according to the various solvents: water, methanol and ethanol.

**Extraction:** 100g of cashew fruits (*Anacardium occidentale*) was weighed using an electronic weighing balance. The weighed samples were—put into the empty beaker of 100ml capacity. 500ml of distilled water was added into the beaker containing the colourants from the cashew fruits. It was stirred continuously for one hour, after which the colourant extract solution (mixture) was poured through fine sieve mesh (to remove unwanted shaft), which was done at room temperature. The filtrate was kept for further analysis. Analysis (characterization) was carried out within 24 hrs after completion of

the extraction process. The same procedure was used for the other solvents.

### **Characterization of Dye**

Ultraviolet visible absorption (UV/VIS) and infrared spectroscopes are proven valuable tools for the characterization and identification of compounds, or functional group chemical bonds present in an unknown mixture of plant extracts<sup>12</sup>. They are used as qualitative tool to identify and characterize molecular species or some of their properties like molecular structure and characteristic absorption spectra that at least constitute their characteristic finger prints<sup>13</sup>. The infra-red spectral assignment of unsaturated bond of unknown samples determines the chromophore in the active compound<sup>14</sup>. The uv/vis is recorded by irradiating with uv/vis light that is continuously varied in wave length. When the wave length corresponds to the homo and lumo orbital of a conjugated system, some of the radiation is absorbed by the samples. Same procedure was used for the extracts from the other solvents

**Table1: Summary of Extraction Protocol**

S/NO	Solvent used	material(g)	beaker(g)	solvent(ml)	extracts(ml)	Temp. (°C)	Time (hrs)
1	Methanol	100	340.4	500	501	27	24
2.	Ethanol	100	278.0	500	501	27	24
3.	Distilled water	100	282.2	500	503	27	24

## RESULTS AND DISCUSSION

**Table2-Infra-Red (IR) Spectra of extract Using Distilled Water**

Peak	Position	Height	Intensive rating	Functional group
1	452.32	17.596	Strong	C-X (X is bromide iodide)
2	1042.56	51.188	Medium-strong	=C-O-C
3.	1464.02	50.628	Strong	C-H bend
4.	1641.48	29.828	Strong	C=O stretch
5.	2101.51	51.852	Medium weak	C=C
6.	2364.81	53.591	Medium	C=N
7.	3455.59	17.733	Weak-medium	N-H stretch
8.	4041.97	49.924	Medium	).H stretch

**Table3-Infra-Red (IR) Spectra of extracts Using Methanol**

Peak	Position	Height	Intensive rating	Functional group
1	441.71	17.818	Strong	C-X (X is bromide iodide)
2	1032.92	66.651	Medium-strong	=C-O- stretch
3.	1254.74	70.618	Medium-strong	=C-O- C
4.	1437.98	69.825	Medium	CH <sub>3</sub> bend
5.	1645.33	64.449	Weak-Strong	C=N
6.	2115.02	71.309	Medium	C=C
7.	2944.44	65.737	Weak	C-H
8.	3404.42	50.715	Medium	O-H
9	3874.16	68.781	Medium	O-H

**Table 4: Infrared (IR) Spectra of Extract Using Ethanol Extract**

Peak	Position	Height	Intensity rating	Functional group
1	434	14.734	Strong	C-x (x is bromine)
2	880.53	34.039	Strong	C-H
3	1056.06	46.801	Medium-strong	C-O stretch
4	1412.9	48.933	Medium	-CH <sub>3</sub>
5	1653.05	48.667	Strong	C=O
6	1924.06	55.57	Medium-strong	X=C=Y
7	2142.99	55.303	Medium – strong	C-C
8	2373.49	55.672	Very weak-weak	C=C stretch
9	2529.73	51.925	Strong	O-H
10	2904.89	43.875	Strong	O-H
11	2968.95	43.016	Strong	C-H stretch
12	3380.76	39.182	Weak-medium	N-H symmetric
13	4034.25	59.925	Medium	O-H
14	4251.25	66.667	Medium	O-H
15	4339.02	59.831	Medium	O-H

**Table 5: UV/VIS Spectrophotometry Results for Distilled Water Extract**

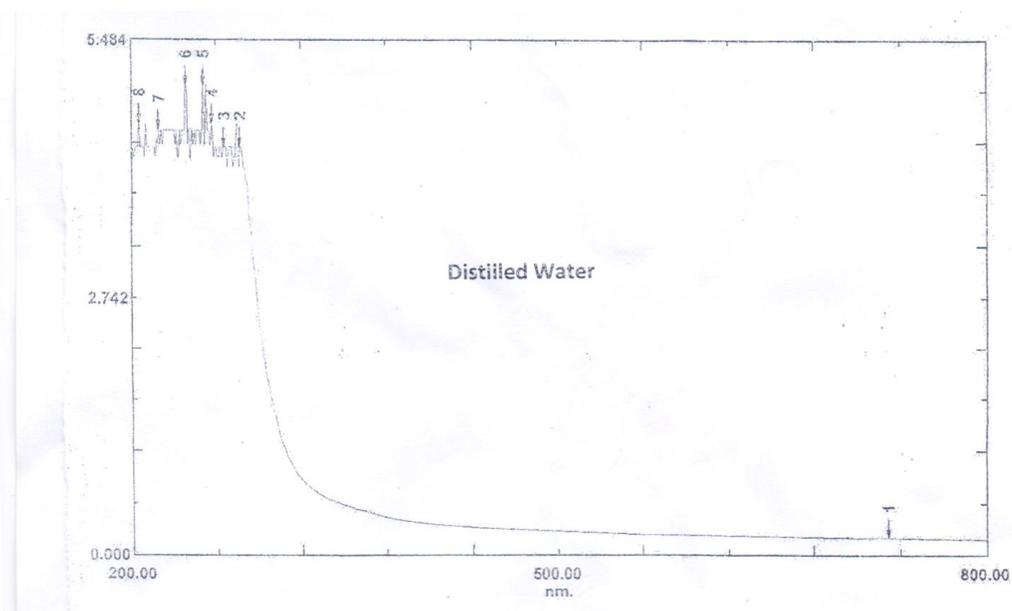
S/NO	Wave length (nm)	Valley absorption	Group Present
1	731.50	0.174	
2	277.00	4.340	Naphthalene
3	265.00	4.340	Naphthalene
4	257.00	4.579	Naphthalene
5	250.50	5.000	Benzene
6	238.00	5.000	Benzene
7	219.00	4.516	Benzene
8	205.00	4.579	Benzene

**Table 6: UV/VIS Spectrophotometry Results for Methanol Extract**

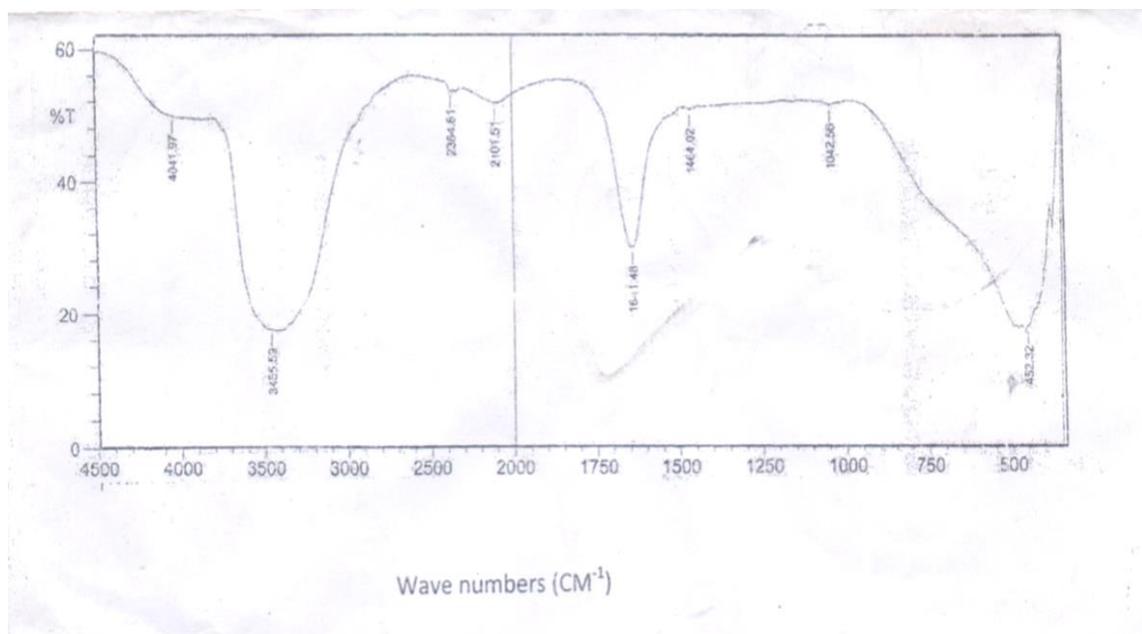
S/NO	Wave length (nm)	Valley absorption	Group present
1	533.00	0.098	Ketone
2	266.00	3.084	Naphthalene
3	259.50	3.061	Naphthalene-
4	225.00	4.215	Benzene
5	218.00	4.366	Benzene

**Table 7 : UV/VIS Spectrophotometry Results for Ethanol Extract**

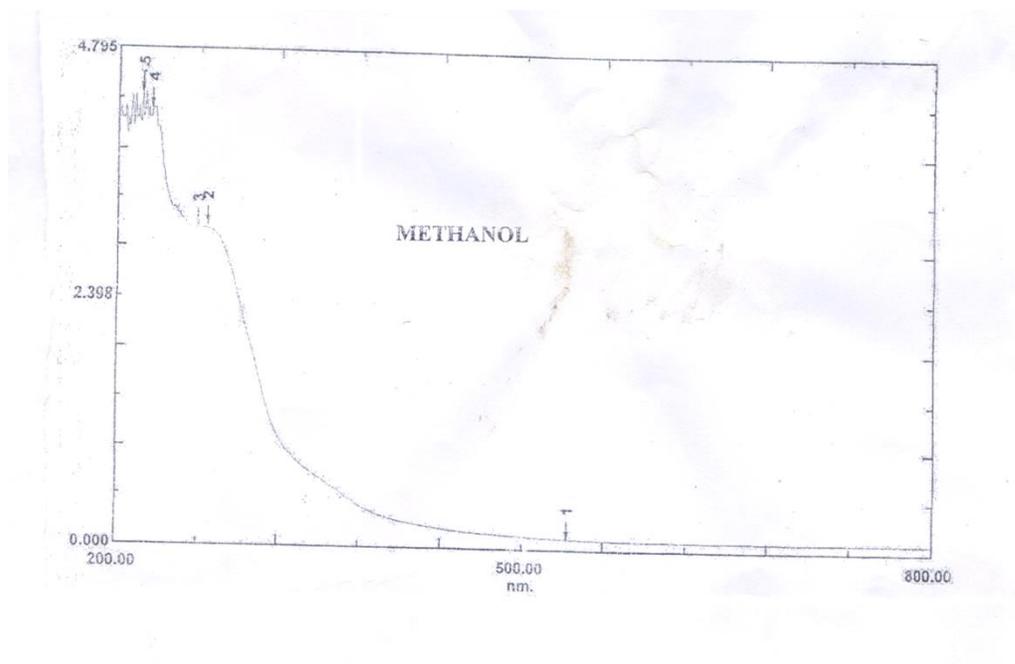
S/NO	Wave length (nm)	Valley absorption	Group present
1	740.00	0.067	-
2	305.00	4.199	Aldehyde
3	294.00	4.100	Aldehyde
4	286.00	4.187	Naphthalene
5	280.00	4.178	Naphthalene
6	269.50	4.198	Naphthalene
7	264.00	4.197	Naphthalene
8	255.00	4.304	Benzene
9	247.50	4.330	Benzene
10	240.00	4.287	Benzene
11	230.00	4.269	Benzene
12	223.00	4.269	Benzene
13	216.00	4,244	Benzene
14	207.00	4.120	Benzene



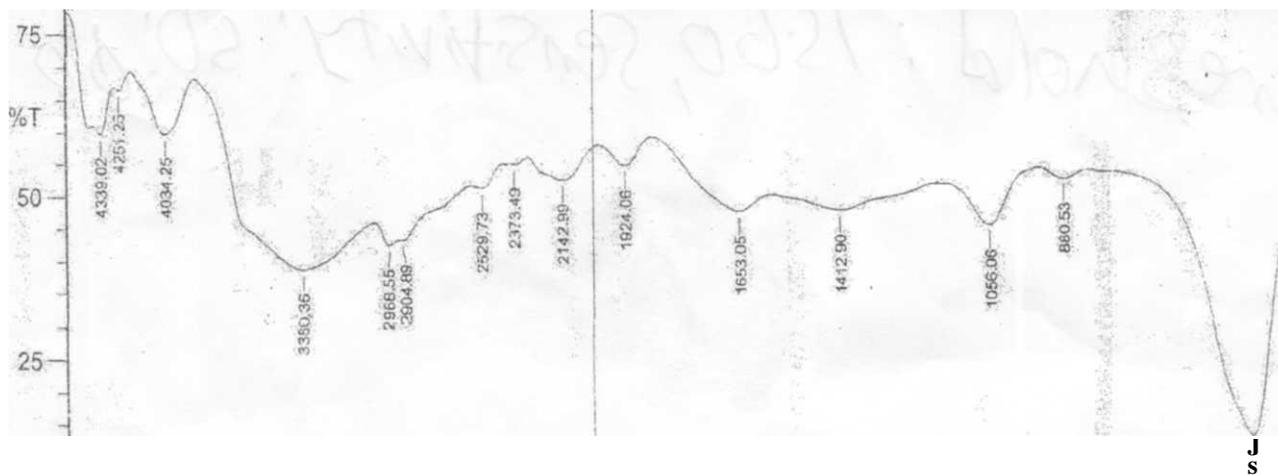
**Fig 1: Peak Finding Results for Distilled Water Extract**



**Fig. 2: Peak Finding Result for Extract Using Methanol**



**Fig 3: Absorption Spectra per Wavelength Range Methanol Extract**



**Fig. 4: Peak Finding Result for Ethanol Extract**

**DISCUSSIONS**

Distilled water, methanol and ethanol were the three solvents used for the extraction in equal

volume of 500ml. The extraction result reveals that distilled water has the highest yield of extract (503ml), it is followed by methanol and ethanol extracts with a yield of (501ml) for both solvent. The difference in volume of yield extracts might be based on the volatility of ethanol and methanol in comparison with distilled water at the temperature the extraction was carried out.

The infrared spectral assignment of unsaturated bonds of the extract is presented in Table 2, 3, and 4 for distilled water, methanol and ethanol extracts respectively. The results reveal the type of chromophores in the active dye compound. The critical conjugated compounds mostly present are C-H, C=O, C=C, N-H, O-H, C=C, =C-O-C, C=N, CH<sub>3</sub>. The chromophoric compounds absorb light in the visible region.

The spectral result of the three extracts (ethanol, methanol and distilled water), shows the absorption peak for each of the solvent. The distilled water extract shows a medium absorption peak at (4041.97) which reveals the presence of OH-stretching, the methanol extracts shows a medium absorption peak at (3874.16) and the ethanol extracts shows absorption peak at (4339.02) respectively.

The absorption spectral of the different extracts methanol, ethanol and distilled water; across the ultraviolet and visible wave lengths measured are as shown in table 5,6

and 7 respectively. The observed absorbance reveal the highest values at the 533.00nm for methanol 740.00nm for ethanol and 731.50nm

for distilled water. This indicates that the greater volume of compounds present in the different extracts from methanol, ethanol and distilled water is derivative of benzene and derivative of naphthalene. However, derivative of ketone was also observed from the ethanol and methanol extract. The maximum wave length 533nm, 740.00nm and 731.50nm reveal the chromophoric groups necessary for the formation of colours reddish brown.

Figure1 reveals that peak eight has the highest absorbance of 4.579 with wave length of 205.00 indicating the presence of derivative of benzene. Figure 2 indicates that peak nine (9) has the highest absorbance of 4.330 with wave length 247.50 showing the presence of derivative of benzene also. Lastly, figure3 shows that peak five has the highest absorbance of 4.366 with wave length of 218.00 showing the presence of derivative of benzene.

### *Class of dye extract*

Considering the chemical groupings present in the colour extracts (dyes) as revealed by infrared and ultra violet spectroscopy particularly the chromophoric groups, the dye extract could have the following structure.

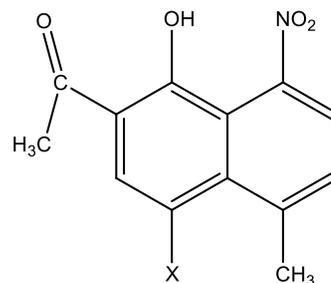


Figure 1 which is distilled water extract, reveals that peak eight has the highest absorbance of 4.579 with wave length of 205.00 the compound present is a derivative of benzene. Figure 2 in which ethanol was used as solvent indicates that peak 9 has the highest absorbance of 4.330 with wave length 247.50, the compound present is a derivative of benzene. Lastly, for methanol solvent, figure iii it shows that peak 5 has the highest absorbance of 4.366 with wave length of 281.00, the compound present is a derivative of benzene.

## CONCLUSIONS

The study carried out on characterization of extracts of *Anacardium occidentale* (cashew fruit) reveals the active dye compound present in the cashew fruit. This study also indicates that this dye compound can be selectively extracted with solvents such as distilled water, ethanol and methanol. Distilled water was found to be the most suitable solvent for the extraction of dye compound from cashew fruits (*Anacardium occidentale*). This study also reveals the presence of C-H, C=O, O-H, C=C, C=N, CH<sub>3</sub>, C≡C and the presence of aromatic compounds such as a derivative of benzene, ketones and naphthalene from ultraviolet spectra.

Lastly, this study suggests a possible presence of three types of dye compounds in the *Anacardium occidentale* extract, based on the data obtained, these are: indigoid dyes, anthraquinone and naphthaquinone, Based on the finding from this study, the following recommendation is made.

Another research should be carried out on *Anacardium occidentale* fruit extracts to confirm the presence of the highlighted chromophores and auxochromes and an attempt should be made to synthesize the dye with the above chromophores, auxochromes and structure.

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