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Macronutrients, Phytochemicals, Mineral Elements, Functional Groups and Health Risk Assessment of *Chrysophyllum albidum* Fruit Obtained in Zaria, Nigeria

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ABSTRACT

Background: This study assessed macronutrients, phytochemicals, mineral elements, functional groups and health risk of *Chrysophyllum albidum* fruit obtained in Zaria. macronutrients (%) were 20.82 ± 0.31, 4.76 ± 0.03, 8.42 ± 0.13, 4.27 ± 0.11, 5.98 ± 0.12 and 55.27 ± 0.52 for moisture, ash, crude fibre, crude fat, crude protein and carbohydrates respectively.

Research Methods: Phytochemical analyses of fruit extracts revealed the presence of alkaloids, tannins, steroids and saponins.

Research Result: The concentrations (mg/kg) of mineral elements were 23.00, 17.40, 8.00, 4.50, and 3.90 for Cu, Mn, Fe, Ni and Pb respectively. Metal concentrations were higher than the Permissible Limits except for Ni (4.50 mg/kg) which was lower than the 10.00 mg/kg acceptable limit by FAO/WHO. MPI revealed strong overall metal concentration (8.7203) in the fruit, the HQ for Fe (0.0737) and Mn (0.8021) was < 1, however the HI (12.3358) was >>1. Hence, there could be health risk concerning the metals.

Conclusion: FTIR analysis revealed the presence of hydroxyl (OH) group of alcohols and phenols, alkanes (C-H), alkynes, C=C stretch of a ketones, C–O stretch of esters and carboxylic group and C-F stretch of alkyl group.

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1. INTRODUCTION

Chrysophyllum albidum (C. albidum) is common fruits consumed in Nigeria and other parts of Africa. It is known as African star apple or just star apple. It belongs to the family of sapotaceae and the class ericales. In Nigeria, it is commonly known as "Agbalumo" (Yoruba), "Udara" (Igbo/Ibibio) and "Agwaluma" (Hausa) (Ibrahim et al., 2021; Esiere et al., 2023). Reports from literature have shown that African star apple have essential nutrients and health-promoting phytochemicals (Ibrahim et al., 2021). Apart from the phytochemicals, some essential/ non-essential metals are also found in the fruits. Heavy metals are not only contained in fruits like the *C. albidum* but are also found in processed foods and fresh water sources (Nwokem et al., 2023; Okon et al., 2023a; Anweting et al., 2024). Heavy metals in fruits could be linked to assimilation from the soil and this can adversely affects plants (Ebong et al., 2022; Okon et al., 2023b). From studies, the fruit has been found to be a good source of vitamins and flavours to diet (Umelo, 1997; Adisa, 2012). *C. albidum* has been noted to be of great medicinal value (Onyekwelu and Stimm, 2011; Agustin, 2018; Esiere et al., 2023).

In terms of environmental services, it is a good species for carbon sequestration; the trees are used as windbreakers, shade and as ornamental plant. It thrives on all kinds of soil and plays vital role in reclamation of wasteland (Esiere et al., 2023). Many people in Zaria consume this fruit (*C. albidum*), hence, the need to know the macronutrients compositions, phytochemicals, mineral elements present and health risk associated with its consumption. This study therefore aimed at assessing the proximate compositions, phytochemicals, mineral elements, functional groups and health risk concern of *C. albidum* (African star apple) fruit obtained in Zaria.

Figure 1. Different part of *Chrysophyllum Albidum* (African star apple) - leaves, fruits and seeds (source: *Ibrahim et al*., 2021).

2. METHODS

Chemical reagents used were of analytical grade and were obtained from Sigma-Aldrich Chemicals (Steinheim Germany). Deionized water was used throughout the experiment. Fruit samples of *Chrysophyllum albidum* were obtained from Sabon Gari Central Market, Zaria. They were stored in a new clean polyethene bag and taken to laboratory for analysis.

Digestion of sample was carried out in line with standard method as described by American Public Health Association (APHA, 1999). Proximate analysis for moisture content, fat content, ash content and crude fibre were determined in accordance with the official methods of the Association of Analytical Chemists (AOAC, 2004). Nitrogen was determined by the Kjeldahl method and the percentage of nitrogen was converted to crude protein by multiplying by 6.25. Carbohydrate content was determined by the difference (Ekwumemgbo et al., 2015; Ibrahim et al., 2021). For digestion of sample for determination of crude protein, Some 2.0 g

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of the sample was weighed into Kjeldahl flask. 5.0 g anhydrous sodium sulphate was added and then 5 ml of concentrated H2SO4 was added. The digestion flask was placed in the digestion rack and heated gently to avoid frothing for 2 hours until a clear bluish solution was obtained. The digest was allowed to cool to room temperature and was transferred into a volumetric flask (250 ml) and made up to the mark with distilled water.

Mineral elements (Metals) were determined, Metals determined include copper (Cu), iron (Fe), manganese (Mn), nickel (Ni) and lead (Pb). Metal contents of the digests were determined using Microwave Plasma Atomic Emission Spectrophotometer (MPAES, model 4200, USA) in the Multi-User Science Research Laboratory of Chemistry Department, Ahmadu Bello University, Zaria. Calibration curves were prepared separately for each of the metals by running different concentrations of standard solutions. The instrument was set to zero by running the respective reagent blanks and average values of three replicates analyses were taken for each determination (Okon et al., 2022).

Fruit extract was prepared for phytochemical screening/FT-IR analyses, to achieve this, fresh fruits of *C. Albidum* were washed thoroughly with tap water to remove dust particles and other dirt and then rinsed properly with distilled water. The fruits were opened up by which the pulp (flesh) was collected. The fruit pulp were air-dried for 10 days and then ground to a fine powder with a Thomas-Wiley laboratory mill (Model 4) and stored in a clean polythene bag for further use (Ekwumemgbo et al., 2023). 25 g of fine powder was boiled in 200 mL of deionized water for 5 min and the extract was then filtered using Whatman filter paper No 1. The aqueous plant extract was collected in an amber-coloured bottle and stored at 4 oC for further use. Other extract were done using acetone and methanol.

For determination of phytochemicals present, crude extract of the fruit was screened, phytochemicals screened include alkaloids, tannins, flavonoids, steroids and saponins in accordance with standard methods by Harborne (1998), Adesanya et al. (2022) and Datti et al. (2020) as described in Table 1.

Table 1. Phytochemical Screening

Phytochemicals	Test	Observation
Alkaloids	1.0 cm^3 of the extract + 3 drops of Dragendoff's reagent. (Dragendoff's test) (Adesanya *et al.*, 2022)	Orange-red precipitate with turbidity indicates present of alkaloid.
Tannins	2.0 cm^3 of the extract diluted with distilled water + 3 drops 5% ferric chloride ($FeCl_3$) solution. (Ferric chloride test) (Adesanya *et al.*, 2022)	Deep blue colour indicates the presence of tannins.
Flavonoids	2.0 cm^3 of the extract + 4 drops of 1% NaOH (Datti *et al.*, 2020)	Appearance of yellow colour indicates the presence of flavonoids
Steroids	2.0 cm^3 of the extract + 5 cm^3 of acetic Chloroform followed by 5 cm^3 cm^3 of conc. H_2SO_4 added by the sides of the test tube (Salkowski Tests) (Datti *et al.*, 2020)	Formation of a red colour on standing shows the presence of steroids.
Saponins	2.0 cm^3 extract + 5.0 cm^3 distilled water and Shaken well for 5 min. (Froth test) (Datti *et al.*, 2020)	Formation of honeycomb froth for about 15 min. indicate and confirms the presence of saponin

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Human health risk assessment for heavy metals was carried out using the following indices: Metal Pollution Index (MPI), Estimated Daily Intake (EDI), Hazard Quotient (HQ) otherwise known as Health Risk Index (HRI) and Hazard Index (HI) in accordance with Mawari *et al*., (2022).

Metal Pollution Index (MPI) gives the overall studied metals (Pb, Cd, As, and Hg) concentration in fruit studied. It was gotten by calculating the geometrical mean of concentrations of the metals as presented in Equation (1) (Sharma *et al*., 2016; Jolly *et al*., 2019).

$$\text{MPI (mg/kg)} = (C_1 \times C_2 \times \ldots C_n)^{1/n} \quad (1)$$

Where C_n = concentration of metal in nth sample.

Estimated Daily Intake (EDI) gives an estimate of the daily concentration of the studied metals ingested into human body (Bayo *et al*., 2021). It was calculated as given in Equation (2).

$$\text{EDI} = \frac{C_{\text{metal}} \times D_{\text{fruit intake}}}{\text{BWaverage}} \quad (2)$$

Where: EDI = Estimated Daily Intake; C_{metal} = concentration of the metal in the fruit samples (mg/kg); $D_{fruit\ intake}$ = Daily intake of the fruit, a minimum of 400 g (0.4 kg) per day in Nigeria (Amao, 2018). BWaverage = average body weight of a Nigerian = 62 kg (Bayo *et al*., 2021).

Hazard Quotient (HQ) is an indication of potential hazards to human health when foods contaminated with these metals are consumed (Mawari *et al*. 2022). it was calculated as expressed in Equation (3).

$$\text{HQ} = \frac{\text{EDI}}{\text{RfD}} \quad (3)$$

Where: HQ= Hazard quotient; EDI = Estimated daily intake; RfD = Oral reference dose. The oral reference dose (RfD) of Cu, Fe, Mn, Ni and Pb are 0.04, 0.7, 0.14, 0.02 and 0.004 mg/kg/day respectively (Wadood *et al*., 2021; Bayo *et al*., 2021).

Hazard Index (HI) gives the overall toxic risk or adverse health effects of the metals considered (Onyele and Anyanwu, 2018). It is calculated as expressed in Equation (4).

$$HI = \sum_{i=1}^{n} (\text{HQ})i \quad (4)$$

Where: HI is the hazard index and n is the total number of metals studied, HQ = Hazard Quotient. When HI ≤ 1 and HQ ≤ 1, there would be no obvious health risk (FAO/WHO, 2010; Ekeanyanwu *et al*., 2015).

3. RESULTS AND DISCUSSION

3.1. Proximate Composition of *Chrysophyllum albidum* Fruits

The results of proximate composition of the fruits are presented in Figure 1 for moisture, ash, crude fibre, crude fat (Lipid), crude protein and carbohydrate. The proximate compositions (%) are 20.82 ± 0.31, 4.76 ± 0.03, 8.42 ± 0.13, 4.27 ± 0.11, 5.98 ± 0.12 and 55.27 ± 0.52 for moisture, ash, crude fibre, crude fat (Lipid), crude protein and carbohydrate respectively. The result followed the order: carbohydrates > moisture > crude fibre > Crude

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Protein > Ash > Crude fat (Lipid). Moisture content was found to be 20.82 ± 0.31%, This value is higher than the 7.18%, 7.76% and 17.03% earlier reported by Bolanle et al. (2017), Ibrahim et al. (2017) and Ibrahim et al. (2021) respectively. The higher moisture content in this study could be attributed to level of freshness and succulence of the fruit at time of analysis. Moisture content is an index of water activity; it is used as measure of the stability and susceptibility to microbial contamination (UmiKalsum, 2014).

Ash content of the *C. albidum* fruit obtain in this study was 4.70 ± 0.03 %, which is higher than the 2.95 % earlier reported, by Bolanle et al. (2017). However, It is lower than the 6% and 8% reported by Karaye et al. (2020) and Wakawa and Chiroma (2020) respectively for Balanites aegyptiaca (Desert date), suggesting that the ash content in *Chrysophyllum albidum* could be lower than that of Balanites aegyptiaca. Ash content is a measure of the total amount of minerals present, which is a measure of the amount of specific inorganic components such as iron, sodium, potassium and zinc. Determination of ash and mineral contents of foods is important for nutritional labelling and quality assessment (McClements and Decker 2009).

Crude fibre was found to be 8.42 ± 0.13%; this value is higher than the 6.6% and 5.67% earlier observed by Ibrahim et al. (2021) and Bolanle et al. (2017) respectively. However, the value obtained in this work (8.42 ± 0.13%) is lower than the early report (9.2%) by Ibrahim et al. (2017). Consumption of fruit with adequate crude fibre can prevent colon cancer and digestive disorder (Gabriel et al., 2018). The crude fat (4.27 ± 0.11%) observed in this study is lower than the 13.25 % and 6.97% earlier reported by Ibrahim et al. (2021) and Ibrahim et al. (2017) respectively. Nevertheless, it was higher than the 3.67% reported by Bolanle et al. (2017).

Percentage of crude protein content was 5.98 ± 0.12. Crude protein represents the amount of amino acids in foods; proteins are good source of essential amino acids, such as lysine, tryptophan, methionine, leucine, isoleucine and valine, which are essential to human health (Nelson and Cox 2005). Crude protein content (5.98 ± 0.12%) obtained is higher than the 3.85% earlier reported by Ibrahim et al. (2021), but lower than the 16.40% reported for B. aegyptiaca by Wakawa and Chiroma (2020) and the 14.87 % and 15.75 % reported for S.agilo and S.aubergine fruits respectively by Edem et al. (2009). These suggest that *C. albidum* fruit could be low in protein content.

Carbohydrates exhibited highest concentration (55.2 ± 0.527%), this shows that the fruit is a source of energy (UmiKalsum, 2014). However, the value (55.27%) is lower than the 66.45% and 75.00% repoerted by Ibrahim et al. (2017) and Bolanle et al. (2017). This could be attributed to more hydration from moisture content of 20.82% obtained in this work, as the moisture content obtained by Ibrahim et al. (2017) and Bolanle et al. (2017) were 7.76% and 7.18% respectively.

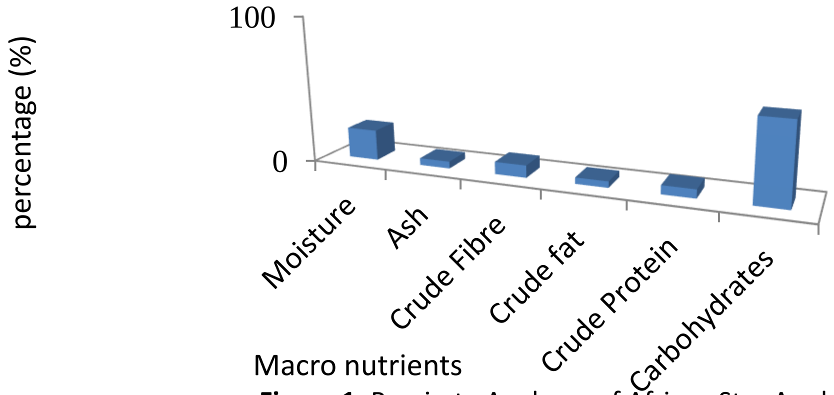

Figure 1. Proximte Analyses of African Star Apple

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3.2. Elemental Analyses

The metal elements determined were Cu, Fe, Mn, Ni and Pd, the results are presented in Figure 2. Cu exhibited the highest concentration (23 mg/kg). The high concentration of Cu could be linked to high soil availability of Cu and bioaccumulation. Copper is required for the growth of plants and animals, but at high concentration, it can affects plants and may cause kidney damage to human (Bayo et al., 2021). The concentration of 23 mg/kg obtained exceeds the (FAO/WHO) acceptable limit of 2.0 mg/kg for Cu. The 23 mg/kg obtained is higher than the 3.275 mg/kg reported by Ibrahim et al. (2021), this could emanate from heavy metal level at point source.

The concentration obtained for Fe was 8.00 mg/kg; this has exceeded the FAO/WHO (2010) acceptable limit of 1.00 mg/kg (Bayo et al., 2021). The result obtained is lower than the 40.875 mg/kg reported by Ibrahim et al. (2021) but higher than the 2.36 mg/kg reported by Asare et al. (2015). Fe plays a substantial role in the formation of hemoglobin and transfer of oxygen in the human body (Vallee and Auld 1990), however, excess Fe can affect early growth in human (Thankachan and Kurpad, 2018).

For Mn, the level obtained was 17.40 mg/kg, which is higher than the FAO/WHO (2010) acceptable limit of 5.00 mg/kg (Bayo et al., 2021). The 17.40 mg/kg obtained is far higher than the 0.58 mg/kg and 0.45 mg/kg earlier reported by Asare et al. (2015) and Ibrahim et al. (2017) respectively. Though Mn is an essential element, excessive accumulation in the body is harmful (Sharma et al., 2006). The value observed for Ni was 4.50 mg/kg which is considerably lower than the permissible limit of 10.00 mg/kg set by joint FAO/WHO Codex (2001). A value of 3.90 mg/kg was obtained for Pb, which is noticeably higher than the 0.50 mg/kg acceptable limit (Bayo et al., 2021). The value (3.90 mg/kg) is higher than 0.9 mg/kg earlier reported by Musa et al. (2014); this could have been caused by higher contamination resulting from more accumulation in soil over time and consequently transferred to plants.

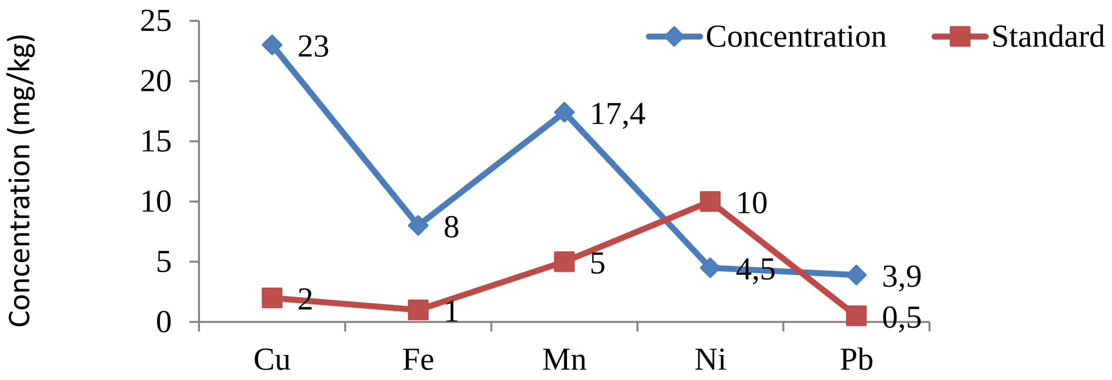

Figure 2. Elemental Analysis of the Fruits with Standard Values

3.3. Phytochemical Analyses

The result of preliminary phytochemical analyses is presented in Table 2, the phytochemicals screened were alkaloids, tannins, flavonoids, steroids and saponins. Flavonoids were not detected in all the extracts. This is in contrast with Ibrahim et al. (2017) and Bolanle et al. (2017). Alkaloids exhibited strongest presence in distilled water extract while that of tannins was in methanol extract. Steroids showed same strength of presence in extracts from all solvents (distilled water, acetone and methanol). Saponins were not detected in distilled water extract but were evenly detected in acetone and methanol

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extracts. The presence of phytochemicals in plants makes the plants to possess antibacterial potentials against human pathogens and hence, finds application in treatment of illness (Nwokonkwo, 2014) and as reducing and stabilizing agent in green synthesis of metal nanoparticles (Ekwumemgbo et al. 2023; Kpega et al. 2023).

Table 2. Phytochemical screening of *C. albidum* Fruits using Distilled Water, Acetone and Methanol as Solvents

Phytochemicals	Distilled Water	Acetone	Methanol
Alkaloids	++	+	+
Tannins	+	+	++
Flavonoids	-	-	-
Steroids	+	+	+
Saponins	-	+	+

keys: + presence, ++ strong presence, - absence

3.4. Health Risk Assessment

Health Risk Assessment is presented in Table 3. From the analyses, Metal Pollution Index (MPI) showed a strong concentration of 8.7203 mg/kg and this could cause health risk. Cu had the highest Estimated Daily Intake (EDI) of 0.1484 mg/kg leading to Hazard Quotient (HQ) of 3.7100 which is higher than 1, hence, Cu may pose a health risk. Mn with EDI of 0.1123 mg/kg followed this though with HQ of only 0.8021. Despite Mn having higher concentration (17.40 mg/kg) in the fruit compared to Pd (3.90 mg/kg) and Ni (4, 50 mg/kg), its HQ (0.8021) is lower than that of Pb (6.3000) and Ni (1.4500). This is due to its higher reference oral dose (RfD) of 0.14 (mg/kg/day) compared to that of Pb (0.004 mg/kg/day) and Ni (0.02 mg/kg/day). Same can be said for Fe with fruit concentration of 8.00 mg/kg but with lowest HQ of 0.0737 compared to Pb and Ni.

The HQ for Mn and Fe were less than 1, revealing that more of the fruit can be consumed without serious health risk from Mn and Fe (Bayo et al., 2021). The value for Hazard Index (HI) was 12.3358, this value is an indication that the consumption of *C. albidum* in Zaria may result to public health concern as it is far above 1 (Jolly et al., 2019).

Table 3. Health Risk Analyses

Metal	Concentration (mg/kg)	R_fD (mg/kg/day)	EDI	HQ
Cu	23.00	0.04	0.1484	3.7100
Fe	8.00	0.7	0.0516	0.0737
Mn	17.40	0.14	0.1123	0.8021
Ni	4.50	0.02	0.0290	1.4500
Pd	3.90	0.004	0.0252	6.3000
MPI = 8.7203; HI = 12.3358				

3.5. FT-IR Analysis of the Fruit Extract

The Fourier transform infrared spectroscopy (FT-IR) spectra of the fruit extract are shown in Figure 3. FT-IR analysis was carried out using FT-IR spectrophotometer, Model Carry 630 (Agilent Technologies, USA). The analysis revealed the presence of various characteristic functional groups in the fruit extract. The presence of hydroxyl compound was revealed due to the absorption band at 3280.1 cm–1 as this band correspond to O-H stretching of alcohols. The band at 2922.2 cm–1 indicates C-H stretching of alkanes as it falls within 2990 – 2850 cm–1, 2143.4 cm–1 corresponds to C≡C stretching of alkynes as it falls

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within 2250 – 2100 cm–1, 1729 cm–1 band corresponds to C=O stretch of aldehydes as it falls within 1740 – 1720 cm–1 (Mohrig et al., 2006). The band at 1617 cm–1 reveals the C=C stretch of a ketone, 1364.2 cm–1 corresponds to the O-H bending of phenol.The band at1244.90 cm–1 falls within 1300 – 1000 corresponding to C–O stretching of esters and carboxylic functional groups (Jayachandran et al., 2021). 1028 cm–1 presents C-F stretch of alkyl and aryl halides as it is within 1000 – 1400 cm–1.

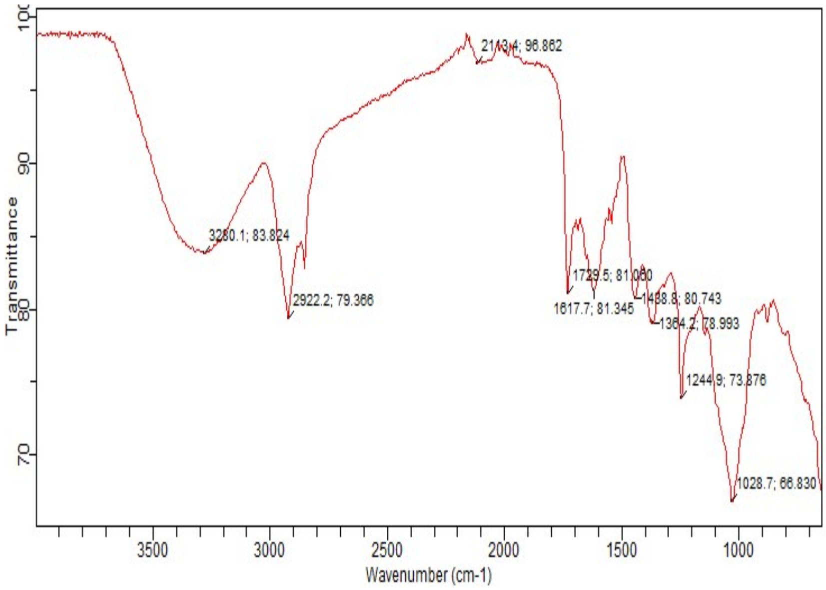

Figure 3. FT-IR Spectra of African Star Apple Fruit Extract

4. CONCLUSION

From the results obtained, *Chrysophyllum albidum* contains some macronutrients (moisture, ash, crude fibre, crude fat, crude protein and carbohydrate). Carbohydrate was highest followed by moisture and was in the order- carbohydrates >> moisture >> crude fibre > crude protein > ash > crude fat. Phytochemical analysis of fruit extracts from the different solvent revealed the presence of alkaloids, tannins, steroids and saponins. Flavonoids were not detected in any of the extracts, saponins were detected only in acetone, and methanol extracts. Mineral elements determined were in the order Cu > Mn > Fe > Ni > Pb. The Metal Pollution Index (MPI) calculated revealed strong overall metal concentration in the fruit, the Hazard Quotient (HQ) for Fe and Mn was less than 1, however the overall health risk index (HI) for all the metals was far above1. Hence, there could be public health risk concerning the metals in the fruit. FTIR spectroscopy revealed the presence of hydroxyl (O-H) group of alcohols and phenols, alkanes (C-H), alkynes, C=C stretch of a ketones, C–O stretching of esters and carboxylic groups and C-F stretch of alkyl group.

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