

Chemical composition and medicinal potentials of the bark of *Erythrophleum ivorense* A. Chev

R. O. Ogboru*, L. O. Akideno and E. A. Owoeye

Forestry Research Institute of Nigeria, Moist Forest Zonal Research Station, P. M. B 2444, Benin-city, Edo- State, Nigeria.

*Corresponding author. Email: rachov44@gmail.com, rachov44@yahoo.com

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ABSTRACT: Determination of the chemical composition of the bark of *Erythrophleum ivorense* A. chev showed some proximate compounds such carbohydrates, protein, lipids and vitamin C, and not absent in essential food fibre. Essential minerals of sodium, phosphorus, iron, copper and zinc in contents levels needed for a healthy life, except sodium, with a very high content level hence should be diluted properly when administered as a laxative. Some phytochemicals such as phenols, steroids, tannins, flavanoids, alkaloids, saponin and cardiac glycosides were present which gives credence to the medicinal benefits of the bark of the wood despite its wide timber used for construction and toxic nature.

Key words: *Erythrophleum ivorense* bark, phytochemical screening, proximate content, mineral elements, total antioxidant capacity.

INTRODUCTION

The role of forest in our atmosphere cannot be over emphasized. The necessity to study forest plant species according to WHO (2001) was based on extensive use of plant portions in folk medicine and offered a basis for the daily living of the people. A large number of African indigenous trees species have been recognized to have health defensive properties and uses (Okeno et al., 2003; Einosho and Ayorinde, 2008). *Erythrophleum ivorense* belongs to the family Caesalpiniaceae (Leguminosae-Caesalpinioideae) and is found majorly in the Gambia to the Central African Republic and Gabon. The timber of *Erythrophleum ivorense* in Nigeria, is marketed as 'Iyin' in Edo, 'erun' in Yoruba and 'ihi' in Igbo (Aigbokhan, 2014). It is called 'Ordeal tree', 'missanda', 'alui', 'bolondo', 'Sasswood tree' (English). Lim du Gabon, Tali (France) and Mancone (Italian) around parts of Africa where it is found (Burkill, 1985). Some species of plant in the *Erythrophleum* genus are also identified to be venomous and toxic to some livestock. The bark of *Erythrophleum ivorense* traded as 'sassy-bark', 'man cona bark', 'casca bark' or 'ecorce de tali' and has several medicinal uses (ITTO, 2004). Bark extracts of *Erythrophleum ivorense* are taken orally in Sierra Leone as an emetic and purgative and is applied externally to relieve pain (Richter

and Dallwitz, 2000). According to Betti (2004), it is taken orally as a laxative and outwardly to relieve pains. In Ivory-Coast, extract from young branches of crushed *Erythrophleum ivorense* is rubbed on skin to treat chicken pox (PROTA1, 2008). The bark and occasionally the seeds are widely used as hunting and ordeal poison (Barkarr and Janos, 1996). In Liberia and Gabon, the bark of *Erythrophleum ivorense* is preferred to that of *Erythrophleum suaveolens* (Bosch et al., 1996). *Erythrophleum ivorense* occurs scattered in evergreen primary and secondary forest and moist semi-deciduous forest (Diabate et al. 2005). It has been classified as a non-pioneer light demander. Seedlings are often found in smaller forest gaps. *Erythrophleum ivorense* can be propagated in the nursery as its seeds take three weeks to germinate. The bark is also used as fish poison in Sierra Leone (PROTA, 2008). The wood is quite hard and hefty and suitable for woodwork, flooring, rail way sleepers, harbor and dock work, turnery, bridges and constructions. It is also used for boat building and wheel hubs. It makes excellent charcoal and good firewood. In Sierra-Leone and Ivory Coast the bark is used for tanning (PROTA, 2008). A bark decoction added to fermenting palm wine would make it a more potent drink (Voorhoeve, 1979). In trade

statistics, *Erythrophleum ivorense* is regarded as the fourth most important timber of Cameroon and was imported to China in 2005 (PROTA, 2008). It is an evergreen tree that can grow up to 40 metres tall, bole cylindrical but sometimes fluted at base with or without buttresses. Its leaves are alternate and flowers are bisexual. The toxicological nature of the specie has been reported by Amoah et al. (2014). They worked on methanol extract on the bark and were administered to rats which caused an increase inflammatory in the liver and kidney of the rats. The phytochemical screening of the plant also revealed the presence of alkaloids, tannins, flavonoids, steroids, cardiac glycosides and terpenoids but they were not quantified. Also Sima et al. (2016) worked on phytochemical screening identifying the phytochemicals, antioxidant and antimicrobial properties of the bark of *Erthrophleum ivorense* A. Chev. Hence, this research work quantified the exact amount of the phytochemicals present contained in the bark *Erythrophleum ivorense* and the proximate analysis to show if it is fit for consumption. This study therefore attempts to identify the chemical compositions contained in the bark of *Erythrophleum ivorense* through proximate analysis, phytochemical screening, mineral analysis and total antioxidant capacity.

MATERIALS AND METHODS

Collection of plant samples

Bark samples of *Erythrophleum ivorense* A. Chev were obtained from the Federal Department of Forestry, Benin City, Edo State, Nigeria in October, 2016. It was authenticated by Mr Emmanuel Isebemhe, the taxonomist of the moist Forest Research station. The Bark sample was air-dried for one month at the research station, Benin City and pulverized to powder and sent to the Delta State University Chemistry Laboratory in Abraka for the chemical analysis.

Preparation of aqueous extract of plant samples

The aqueous extract of bark of *Erythrophleum ivorense* sample was primed by soaking 10 g of powdered samples in 200 ml of distilled water for 12 hours. The extracts were tried for carotenoids, terpenoids, alkaloids, tannins, flavanoids, saponin, cardiac glycoside, anthraquinone, phenols, polyphenols and steroids

Phytochemical analysis

Biochemical tests were conducted on the aqueous extract of the bark sample by using standard methods adopted by Sofowora (1993), Harbone (1973), Oyoyede (2005), Ogboru et al. (2015) and Owoeye et al. (2016).

Qualitative analysis on phytochemical constituents

Test for tannins

0.5 g of powdered sample was boiled in 20 ml of distilled water in a test tube and then filtered. The filtration method used here is the normal method, which includes a conical flask and filter paper. 0.1% of FeCl_2 was added to the filtrate and observed for brownish green or a blue black coloration, which shows the presence of tannins.

Test for saponins

2 g of powdered bark sample was boiled together with 20 ml of distilled water in a water bath and filtered. 10 ml of the filtrate was mixed with 5 ml of distilled water in a test tube and shaken vigorously to obtain a stable persistent froth. The frothing was then mixed with 3 drops of olive oil and observed for the formation of emulsion, which indicates the presence of saponins.

Test for flavonoids

A few drops of 1% NH_3 solution was added to the aqueous extract of the bark sample in a test tube. A yellow coloration was seen indicating flavonoids compounds was present.

Test for cardiac glycosides

1 ml of concentrated H_2SO_4 was prepared in a test tube. 5 ml of aqueous extract from the bark sample was mixed with 2 ml of glacial $\text{CH}_3\text{CO}_2\text{H}$ containing 1 drop of FeCl_3 . The above mixture was carefully added to the 1 ml of concentrated H_2SO_4 so that the concentrated H_2SO_4 was underneath the mixture. A brown ring appeared, indicating the presence of the cardiac constituent.

Test for phenols

To 3 ml of extract, 3 ml of lead acetate solution was added. The occurrence of white precipitates indicates the presence of phenols.

Test for alkaloids

To 3 ml of filtrate, 1 ml of Hagers reagent (Saturated picric acid solution) was added. The appearance of yellow precipitate shows alkaloids present.

Test for steroids

2 ml of acetic anhydride was added to 0.5 g extract with 2 ml of sulphuric acid and observes for the colour change

from violet to blue or green in samples indicating the presence of steroids.

Test for terpenes

0.5 g of plant extract in a test tube was added with 2 ml of chloroform and 5 ml of concentrated sulphuric acid carefully. A reddish brown interface to show positive result for terpenes.

Test for anthraquinones

0.5 g of plant extract is shaken with 10 ml of benzene and filtered. 5 ml of 10% ammonia is added to the filtrate. The mixture is shaken and the presence of pink, red or violet colour indicates the presence of anthraquinones.

Quantitative analysis on phytochemical constituents

Phenols

The quantity of phenols was determined using the spectrophotometer method. The fat free plant sample was boiled with 50 ml of Ether ($\text{CH}_3\text{CH}_2)_2\text{O}$) for 15 mins. 5 ml of the boiled sample was then pipette into 50 ml flask, and 10 ml of distilled water was added. After the addition of distilled water, 2 ml of NH_4OH solution and 5 ml of concentrated $\text{CH}_3(\text{CH}_2)\text{CH}_2\text{OH}$ was added to the mixture. The sample was made up to the mark and left for 30 mins to react for colour development and measured at 505 nm wavelength using a spectrophotometer.

Cardiac glycosides

The Baljets reagent method was used (95 ml of 1% picric acid + 5ml of 10% NaOH). 1 g of powdered sample was soaked overnight with 70% ethanol and filtered. The extract was purified by lead acetate and Na_2HPO_4 and 1 ml of freshly prepared Baljets reagent was added. The solution was put in the curvette and read at 495 nm in the UV spectrophotometer.

Alkaloids

5 g of plant sample was prepared in a beaker and 200 ml of 10% $\text{CH}_3\text{CO}_2\text{H}$ in $\text{C}_2\text{H}_5\text{OH}$ was added to the powdered plant sample. The mixture was covered and allowed to stand for four hours. The mixture was then filtered and the extract was allowed to become concentrated in a water bath until it reaches $\frac{1}{4}$ of the original volume. Concentrated NH_4OH is added until the precipitation is complete. The whole solution is allowed to settle and the precipitate was collected and washed with dilute NH_4OH and then filtered. The residue is alkaloid which is then dried and weighed.

Tannins

Quantity of tannins was determined by using the spectrophotometer method. 0.5g of plant sample is weighed into a 50 ml plastic bottle. 50 ml of distilled water is added and stirred for 1hour. The sample is filtered into a 50 ml volumetric flask and made up to mark. 5ml of the filtered sample is then pipette out into test tube and mixed with 2 ml of 0.1M FeCl_3 in 0.1M HCl and 0.008M $\text{K}_4\text{Fe}(\text{CN})_6 \cdot 3\text{H}_2\text{O}$. The absorbance is measured with a spectrophotometer at 395nm wavelength within 10mins.

Saponins

20 g of powdered bark of plant sample was put in a conical flask and 100 ml of 20% $\text{C}_2\text{H}_5\text{OH}$ was added to the plant sample. The sample was heated over a hot bath for 4 hours with continuous stirring at about 55°C . The mixture was then filtered and the residue re-extracted with another 200 ml of 20% $\text{C}_2\text{H}_5\text{OH}$. The combined extracts were reduced to 40 ml over a water bath at about 90°C . The concentrated solution was then transferred into a 250 ml separator funnel and 20 ml of $(\text{CH}_3\text{CH}_2)_2\text{O}$ was added to the extract and shaken vigorously. The aqueous layer was recovered while the $(\text{CH}_3\text{CH}_2)_2\text{O}$ layer was discarded and the purification process was repeated twice. 60 ml of $n\text{-C}_4\text{H}_9\text{OH}$ was added and the combined $n\text{-C}_4\text{H}_9\text{OH}$ extracts was washed twice with 10 ml of 5% NaCl . The remaining solution was then heated in a water bath and allowed to evaporate. After the evaporation, the samples were dried in the oven to a constant weight.

Flavonoids

10 g of the plant sample was repeatedly extracted with 100 ml of 80% aqueous methanol at room temperature. The entire solution was then filtered through filter paper and the filtrate was transferred into a water bath and the solution was evaporated into dryness. The sample was then weighed until a constant weight was obtained.

Proximate composition determinations

The proximate composition of the bark of *Erythrophloeum ivorense* A.Chev was determined using the methods of the AOAC (1990). These analysis included carbohydrate, crude protein, moisture content, ash, crude fibre, fat and vitamin C.

Antioxidant analysis

Total Antioxidant capacity was determined by the Phospho-molybdenum method according to the procedure

Table 1. Phytochemistry.

Phytochemicals	Qualitative Remarks	Quantitative remarks(mg/kg)
Tannin	+	31.17±0.04
Flavanoid	+	33.76±0.25
Alkaloid	+	58.89±0.20
Saponin	+	41.63±0.11
Cardiac glycoside	+	37.45±0.16
Anthraquinon	-	-
Phenolic	+	10.08±0.38
Terpenes	-	-
Carotenoid	-	-
Steroids	+	06.03±0.02

(+) Present (-) Absent.

described by Prieto et al. (1991). This was also adopted by Vernon et al. (1999), Zongo et al. (2010) and Ogboru et al. (2016). Total Phenols were determined using the method described by Obadoni and Ochuko (2001). For the extraction of phenolic content, the fat free sample was boiled with 50 ml of ether for 15 mins. 5 ml of the extract was pipette into a 50 ml volumetric flask and then 10 ml of distilled water was added. The samples were made up to the mark and left to react for 30 mins for colour development. The absorbance at 505 nm was read at a UV-spectrophotometer.

Mineral element analysis

Milled samples (5 g) were burnt to ash in a furnace at 550°C for 24 hours. The resulting ash was cooled in a desiccator and weighed. 2 ml of Concentrated HCl was added to melt the ash and a few drops of HNO₃ were added (AOAC, 1990). The solution was placed in a steaming water bath and evaporated almost to dryness. The contents were transferred to 100 ml volumetric flask and diluted to volume with distilled water and appropriate dilutions were made for each element before analysis.

Determination of Sodium (Na) and Phosphorus (P) by flame photometer Principle

The flame photometer emits radiant energy when atoms of an element arrival to their ground state after their excitation by the high temperature of the flame. The degree of emission is related to the concentration of the element in the solution. Na and K analysis of the sample were done by the method of flame photometry.

Determination of Iron (Fe), Zinc (Zn) and Copper (Cu)

The same wet sample solutions of the bark of *Erythrophleum ivorense* was analyzed for the determination of Iron content, Zinc and Copper using the Atomic

absorption spectrophotometer with Model Agilent technologies 55A. Different electrode lamps were used for each mineral. The equipment was run for standard solutions of each mineral before and during determination to check that it is working properly. The dilution factor was 100.

RESULTS AND DISCUSSION

Phytochemistry

The phytochemical screening of the bark of *Erythrophleum ivorense* which was sourced from Federal Department of Forestry, Benin City, revealed that alkaloids (53.89 ± 0.20 mg/kg), Phenolic compounds (10.08 ± 0.38mg/kg), Flavanoids (33.76 ± 0.25 mg/kg), Tannins (31.17 ± 0.04 mg/kg), Saponins (41.63 ± 0.11 mg/kg), Cardiac glycosides (37.45 ± 0.16 mg/kg) and steroids (06.03 ± 0.02 mg/kg) were significantly present in the sample (Table 1).

Erythrophleum alkaloids have similar pharmacological activities as digitoxine and ouabain which have shown to have anti-hypersensitive effects, antiarrhythmic effect, antimalarial activity and anticancer actions (PROTA, 2008).

Anthraquinones and steroids constituents promote the plant in the treatment and therapeutic applications as arrow poisons or cardiac drugs as laxatives. The presence of anthraquinones was reported to have anti-oxidant, antimicrobial, anti-viral, anti-malaria and anti-tumor activities. The presence of alkaloids also indicates that the bark of *Erythrophleum ivorense* can be use as muscle relaxant in clinics. The presence of flavonoids in a plant indicates its anti-allergic, anti-inflammatory, anti-cancer, anti-oxidant and hypo-lipidemic effects (Skene and Sutton, 2006). Flavanoids have also been reported to be potent antioxidant and free radicals scavengers capable of protecting cell membranes from damage (Noda et al., 2000)

Tannins are used in the dyestuff productions as caustics for cationic dyes (tannin dyes) and also inks (iron gallate

Table 2. Proximate Analysis.

Parameters	Mg/kg
Carbohydrates	57.61±0.08
Protein	201.47±0.02
Moisture content	4.38±0.16
Ash	63.11±0.38
Fibre	130.84±0.04
Fat	0.02±1.16
Vitamin C	8.06±0.20

Table 3. Mineral Analysis.

Mineral elements	(ppm)
Zinc	21.66±0.02
Copper	3.77±0.05
Sodium	214.53±0.04
Iron	42.24±0.14
Phosphorus	2.143±0.01

Table 4. Antioxidant Analysis.

Parameters	Values
Total Phenolic (GAE100-1)	10.08±0.03
Vitamin C (mg/kg)	8.06±0.09
Total Antioxidant Capacity mg/Vce	63.38±0.10

ink). In the food industries tannins are used to clarify wines, beer and fruit juices. It can also be used as coagulants in rubber production (Gyamfi and Aniya, 2002). Lately, tannins have enticed scientific attention, especially due to amplified incidence of deadly illnesses such as various cancer and AIDS. Tannin rich medicinal plants are used to heal a lot of illnesses; such as leucorrhoea, rhinorrhea and diarrhea. The presence of a few phytochemicals gives belief to the medicinal benefits that the bark of this plant encompasses.

Proximate analysis

Proximate study revealed that the bark contains little nutrients and minerals. Macro nutrient like protein and Carbohydrates were determined with value of 201.47 ± 0.02 and 57.61 ± 0.08 mg/kg respectively. Other macro nutrients were also relatively high in content level. Crude Lipid had a small content level of 0.02 ± 0.16 mg/kg. Proximate analysis for crude Fibre, Vitamin C, Ash, and Moisture also revealed their contents levels to be 130.84 ± 0.04, 8.06 ± 0.02, 52.37 ± 0.38 and 4.38 ± 0.16 mg/kg respectively (Table 2).

Mineral analysis

The existence of essential minerals and their contents levels analyzed showed that *Erythrophleum ivorense* contains sodium (Na; 214.53 ± 0.04) mg/kg, phosphorus (P; 2.143 ± 0.01) mg/kg, Iron (Fe; 42.24 ± 0.02) mg/kg, Zinc (Zn; 21.66 ± 0.02) mg/kg and Copper (Cu; 3.77 ± 0.05) mg/kg respectively (Table 3). The values ranges are non-lethal, therapeutic and idea for the maintenance of good health except for sodium that was on the very high side.

Total antioxidant analysis

Erythrophleum ivorense bark was found to be very opulent in anti-oxidants and phenols with total anti-oxidant capacity of 63.38.02 ± 0.12 mgVce and phenol total content of 10.08 ± 0.03 GAE100⁻¹ (Table 4). Total Anti-oxidant Capacity in *Erythrophleum ivorense* was moderate. The vital role of anti-oxidants is to neutralise the effect of free radicals which damage health cells. It also strengthens the immune system and prevent diseases (e.g. cardiovascular disease and cancer), thus imparting good health and longevity. Phenols help in providing the body defence against oxidative stress or damage arising from oxidising agents and free radicals particularly when the internal enzymatic mechanisms fail or become inadequate. Besides, they are known for their excellent ability to prevent fatty acids from oxidative decay.

Conclusion

It can be inferred from the phytochemical analysis that the bark of *Erythrophleum ivorense* can be said to have some medicinal potentials due to some phytochemicals present. But because high doses of the bark extract is an enormously strong, rapid acting cardiac poison, in warm blooded animals causing shortness of breath, seizures and cardiac arrest in a few minutes. It is advised that the bark be administered with caution despite it alkaloid content and high sodium level.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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