

Elemental Composition and Phytochemical Studies of Aqueous Leaf Extract and Stem Bark Extract of *Crateva adansonii*

Absract

Objective: To investigate the elemental and Phytochemical components of aqueous extracts of the leaves and stem bark of the plant, *Crateva adansonii*. **Methods:** The elemental analysis was done using atomic absorption spectrophotometer (AAS) Model 400. Carbohydrate, anthraquinone, saponins, tannins, alkaloids, steroids, flavonoids, terpenoids and phlobatanins were determined using standard methods. **Results:** The phytochemical studies revealed the presence of saponins and flavanoids in both the leaf and stem bark extracts. Tannins, alkaloids, steroids and terpenoids were present only in the leaf extracts. While carbohydrate was found only in the stem bark extracts. Anthraquinone and phlobatanins were absent in both the leaf and stem bark extract. The anions and cations detected included sodium(Na^+), potassium(K^+), calcium(Ca^{2+}), magnesium(Mg^+), manganese(Mn), copper(Cu^{2+}), zinc(Zn^{2+}) iron(Fe^{2+}), lead(Pb^{2+}), cobalt(Co^{2+}), lithium(Li^+), chloride(Cl^-) and sulphate (SO_4^{2-})ions, suggesting that the plant contains pharmacologically active ingredients as well as metal cations found important in antioxidative stress roles. The results gotten from elemental analysis were subjected to statistical analysis using students'-test statistic at $P<0.05$ level of significance to find out which of the extracts that contained higher concentrations of the above elements (cations and anions). Statistical analysis done showed that sodium, potassium, calcium, magnesium, manganese, iron and phosphorous ions were significantly higher($P<0.05$) in the stem bark extract compared to the leaf extract. Calcium, magnesium and manganese were significantly higher in the leaf extracts than in the stem extracts. **Conclusion:** The results show that *Crateva adansonii* contains relevant elements in electrolyte balance functions which may be exploited in the management of hypertension.

1.0. Introduction

The use of plants in treating diseases has been existing before the inception of civilization. There are strong beliefs in the efficacy of traditional medical systems worldwide [1]. Medicinal plants play a key role in the human health care. [2] A number of plants have been used by traditional medicine practitioners in Orlu, Eastern Nigeria where there are many popular native medicine practitioners or traditional doctors. Such plants include *Sacoglottis gabonensis*, a Nigeria alcoholic beverage additive which contains antioxidant properties against experimental membrane peroxidation [3]. Also, *Crateva adansonii* (three-leaved plant) has been used by Nzeh Native Doctor Nwaele Okereke Okwaraozurumba the grandfather of one of the authors of this article in the management of wounds and sores, hypertension, diabetes, boils and treatment of venereal diseases and to boost female fertility in mothers. The above would suggest that the plant possesses principles that are pharmacologically active against the

above disease conditions. It also suggests possible role for the plant during oxidative stress conditions caused by some of the diseases. *Crateva adansonii* was described by Augustin Pyramus de Candolle in 1824 as a specie in the genus *Crateva* which contains 16 to 25 species and belongs to the family of Capparaceae. The plant (*Crateva adansonii*) is perennial without flowers and seeds but grows vegetative through roots that travel underground and sprouts at certain places. It can also grow by stem cutting. The leaves are used in curing boils, whitlows, and also as fertility drugs and management of rashes.

From time immemorial, man has been in search for plant, animal and other materials that can be used to take care of the pains, deformities, ailments and diseases that inflict some of the unfortunate member of our society [4]. It has been observed that traditional medicine practitioners tend to hide the identities of plants used for different ailments largely for fear of lack of patronage should the sufferer learn to cure himself. Traditional medicine, a major African socio-cultural heritage obviously in existence for several decades was believed to be primitive and wrongly challenged with animosity by foreign religions and conventional or orthodox medical practitioners [5].

There is paucity of information on the medicinal properties of *Crateva adansonii* species being discussed in this paper. This research has, therefore addressed itself to investigate the phytochemical and elemental compositions of crude extracts of the stem bark and leaves of *Crateva adansonii* towards establishing the scientific bases for the long practice of using the plant in the medicinal practices enumerated above.



Fig 1: Photographs of *Crateva adansonii* obtained from Orlu, Imo State, Nigeria

2.0. Materials and methods

2.1. Collection and preparation of samples

The plant was identified by Mr. Ozioko of Botany Department, University of Nigeria, Nsukka.

Samples were prepared according to the standard procedures [1]. Fresh stem and leaves cuttings were brought from Federal low Cost Estate, Orlu, and Imo State, Nigeria and kept in cellophane and transported to University of Maiduguri, Nigeria for onward analysis. The leaves and stem bark were air-dried in the shade and pulverized to obtain a coarse powder using pestle and mortar. They were scrapped and the aqueous extract prepared. 30 g of dry leaves of the plant, *Crateva adansonii* was weighed into one litre of hot distilled water and left to stay overnight. It was filtered and 530 ml of the filtrate used for analysis. 30g stem cuttings of the

plant was weighed into one litre of distilled water and boiled for 1h 30 min. It was cooled, filtered and 550 ml of the filtrate used for analysis.

2.2. Phytochemical screening

This was carried out according to standard procedures [6]. Terpenoids and steroids were determined by adding 4 mg of the extract to 0.5 ml of acetic anhydride and 0.5 ml of chloroform. Then, concentrated solution of tetraoxosulphate(VI) acid (H_2SO_4) was added slowly. Red violet colour was observed for terpenoids and green colour for steroids. Presence of alkaloids was determined by stirring 2ml each of both leaf and stem extracts with 5 ml of 10 % aqueous hydrochloric acid (HCl) in a steam bath for 20 min. This was cooled and filtered. Two drops of Mayer's reagents was added to 1ml of the filtrate and observed for turbidity or presence of reddish brown precipitate. Presence of tannins was determined by adding 2 ml each of leaf and stem extracts in different test-tubes. This was followed by the addition of one drop of ferric chloride reagent to each test tube and observed for blue or green colour. Two mls of both leaf and stem extracts were boiled separately with 10 % aqueous HCl and observed for formation of red precipitate which confirms presence of phlobatannins. 5 ml of diluted ammonia solution was added to a portion of aqueous filtrate of each stems and leaf extract. This was followed by the addition of 5ml concentrated tetraoxosulphate (vi) acid (H_2SO_4) and observed for yellow colouration which confirms the presence of flavonoids. Presence of steroids was determined by adding 2 ml of acetic anhydride to 2ml of each sample with 2 ml of concentrated H_2SO_4 and observed for colour change from violet to blue or green. Each of 5ml of both the leaf and stem bark extracts was added to 4 drops of α -naphthol and mixed. This was followed by the addition of 4mls of concentrated tetraoxosulphate(vi) acid (H_2SO_4) down the side of the test tube. A purple colour was observed in the solution for stem extract which is an indication of the presence of carbohydrate in the stem extracts.

2.3. Quantitative test for elemental composition

The elemental composition was determined using atomic absorption spectrophotometer (model 400). The samples were subjected to high-energy thermal environment in order to produce excited-state atoms. This environment can be provided by a flame or, more recently, plasma. Since the excited state is unstable, the atoms spontaneously return to the ground state and emit light. The emission spectrum of an element consists of a collection of emission wavelengths called emission lines because of the discrete nature of the emitted wavelength. The intensity at an emission line will increase as the number of excited atoms of the element increases. The ground state atom absorbs light energy of a specific wavelength as it enters the excited state. As the number of atoms in the light path increases, the amount of light absorbed also increases. A quantitative determination of the concentration of the trace elements was made by measuring the amount of light absorbed.

2.4. Statistical method

The statistical method used is students'-test statistic. This statistical method was applied in order to find out if the values gotten for the elemental composition in the plants extracts are significantly different from one another. This was done at $P<0.05$ level of significance.

3.0. Results

Tables 1 -3 below show the results of the phytochemical screening and elemental analysis carried out on the aqueous extract of both the leaf and stem extracts of *Crateva adansonii* plant. Carbohydrate was present in the stem bark extract only, while saponins and flavanoids were present in both leaf and stem bark extracts. Tannins, alkaloids, steroids and terpenoids were present in leaf extract only. Anthraquinone and phlobatannins were absent in both extracts (Table 1).

Table1: Phytochemical screening of *Crateva adansonii*

Test	Leaf Extract	Stem Bark
Carbohydrate	-	+
Anthraquinone	-	-
Saponins	+	+
Tannins	+	-
Alkaloids	+	-
Steroids	+	-
Flavanoids	+	+
Terpenoids	+	-
Phlobatanins	-	-

Key:+=positive

--=negative

Sodium (Na^+),potassium(K^+),cupper(2^+),zinc(Zn^{2+}) and cobolt(Co^{2+})and phosphorous(P) ions were significantly lower in concentration($P<0.05$) in leaf extract than in steam bark extract while concentration of calcium(Ca^{2+}),magnesium(Mg^+), manganese(Mn) and iron(Fe^{2+}) ions were higher in leaf than steam bark extracts. Lead(Pb^{2+}) ion was not detected in both extracts. Lithium (Li) ion was also not detected in steam bark extract (table 2).

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Table 2: Cationic composition of *Crateva adansonii* leaf and stem bark extract

Cation	Leaf Extract (mg/l)	Steam	Bark
(mg/l)			
Na ⁺	1.38±0.00 ^a		
221.00±1.00 ^b			
K ⁺	175.00 ^c ±1.00		
930.00±1.5 ^d			
Ca ²⁺	327.10±0.10 ^e		
141.60±0.20 ^f			
M ^{g+}	98.16±0.20 ^g		
48.88±0.01 ^h			
Mn	2.95±0.10 ^z		
0.91±0.02 ^y			
Cu ²⁺	0.002±0.00 [*]		
0.08±0.01 [*]			
Zn ²⁺	4.71±0.10 ^k		
6.32±0.02 ^k			
Fe ²⁺	0.53±0.02 ^m		
0.02 ⁿ ±0.02			
Pb ²⁺	0.00		0.00
Co ²⁺	0.05±0.01 ^l		
0.06±0.01 ^l			

151	Li ⁺	0.02±0.00*	0.00*
152	P	41.60±1.00 ^s	
153		85.70±0.10 ^p	

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155 Results are mean± SD of triplicate determination. Values with different superscripts in a row are
156 significant($P<0.05$).

157 The concentration of the anions, sulphate and chloride anions(SO_4^{2-} and Cl^-) were significantly
158 higher in leaf extract ,85.77mg/l ±0.02 and 1935 mg/l±2.00 than in stem extracts,51.18±0.02
159 and 673 mg/l±1.00 ($p<0.05$) (Table 3).

160 Table 3: Anionic composition of *Crateva adansonii*

161	Samples	Leaf Extract	Steam Extract
162	SO₄²⁻(mg/l)	85.77±0.02 ^a	1935.00± 2.00
163	^b		
164	Cl⁻(mg/l)	51.18±0.02 ^c	673.00±1.00 ^d

165 Results are mean± SD of triplicate determination. Values with different superscripts in a row are
166 significant($p<0.05$).

167 4.0. Discussion

168 The phytochemical analysis done in this work showed that the plant, *Crateva adansonii* contains
169 saponins and flavanoids in both leaf and stem bark extracts. These constituents were known to
170 be bioactive. [6], [7] The presence of saponins in both leaf extracts and stem bark of the plant,
171 *Crateva adansonii* suggests that the extracts from *Crateva adansonii* may be used in the
172 treatment of hormone dependent disorders, including cancer and postmenopausal symptoms.
173 Presence of saponins has been known to protect against attack by potential pathogen[7] and
174 this may account for the antifungal, antimicrobial, anti-yeast and anti-inflammatory activities of
175 the plant. However, tannins, alkaloids, steroids and terpenoids were found present in the leaf
176 extracts but absent in the stem bark of *Crateva adansonii*. This suggests that the leaf extracts of
177 the plant may be used in the treatment of gastrointestinal disorders, diarrhea and malaria.[9]
178 The presence of steroids also suggests that the plant may be a source of vitamin D. [8]
179 Carbohydrate was absent in leaf extracts but present in stem extract. This again suggests that
180 the stem bark extracts have nutritional values as they might be sources of carbohydrates.
181 Similar work had been reported by William. [9]

Furthermore, the elemental analysis done in this work shows that *Crateva adansonii* plant contains significant amount of Na^+ , K^+ , Ca^+ , Mg^+ , Mn , Zn^{2+} and P in both leaf and stem bark extracts. Thus, suggesting that these extracts may be good sources of these ions. From the results analyzed, sodium, potassium, zinc and phosphorous were significantly higher in concentration in stem extracts than in leaf extracts (table 2). Na , K and Ca show the role of the extract as a source of important cations of extracellular and intracellular fluids and hence electrolyte balance functions. The presence of Fe , Cu , Zn , Mn , all redox active cations suggest antioxidant properties and cytoprotection while absence of Pb and negligible presence of Co showed that the environment is not polluted with these heavy metals. The higher concentrations of Na^+ , K^+ , Zn^{2+} and P ions found in stem bark extract than the leaf extracts suggest that the stem bark of *Crateva adansonii* may be a better source of these ions than leaf extracts. This agrees with the similar work of William. [9] Cu , Fe , Co and Li ions occurred in trace amount. This could equally mean that the plant (*Crateva adansonii*) may not be good sources of Cu , Fe , Co and Li ions. The concentration of Fe^{2+} was found to be significantly higher ($p < 0.05$) in leaf than in stem bark of *Crateva adansonii* which suggests that leaf extracts may be a better source of Fe^{2+} than stem bark of *Crateva adansonii*. The absence of Cd and Pb which are heavy metals, in both extracts might be an indication that the area from which the plant was obtained were not polluted with these metals. Cadmium (Cd) and Lead (Pb) had been reported by Ofoegbu, [10] Akubugwo, [11] to be among the common heavy metals found in polluted areas. Chloride ions (Cl^-) and sulphate ions (SO_4^{2-}) were highly concentrated in both extracts but the concentration of Cl^- and SO_4^{2-} were higher in leaf than in stem bark extract. This shows that the extracts might be good sources of these ions but could be better when using leaf extract. The presence of SO_4^{2-} shows the plant can supply substrates for detoxification of autooxidisable drugs while Cl^- plays role in electrolyte balance roles and regulations.

4.1. Conclusion

This current study therefore provides some scientific justification for the utilization of the plant, *Crateva adansonii* by herbalists and the Nigerian populace to treat boils, withlow, skin rashes, fungal diseases, diarrhoea, malaria and infertility. The plant can therefore be said to be potentially medicinal. Further studies should be directed towards the toxicological assessment of the plant extracts.

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