Available online at www.elixirpublishers.com (Elixir International Journal)

Applied Chemistry

Elixir Appl. Chem. 113 (2017) 49299-49303

ISSN: 2229-712X

The Physiochemical Investigation of the Aqueous Extract of the Leaves of Four Nigerian Medicinal Plants. (*Eremomasta polysperma, Ocimum gratissimum, Carica papaya and Starchytarpheta cayenneisis*).

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ARTICLE INFO

Article history:

Received: 07 November 2017;
Received in revised form:
9 December 2017;
Accepted: 19 December 2017;

Keywords

Physiochemical,
Aqueous Extract,
Medicinal Plants,
Eremomastaxpolysperma,
Ocimum gratisssimum,
Caricapapaya,
Starchytarpheta cayennensis.

ABSTRACT

The physiochemical properties of the leaf extract of four Nigeria medicinal plants were investigated in order to ascertain their level of safety for human consumption. The results obtained from the analysis show a range of temperature 25.50 + 0.003, 26.00+0.0020C, as against 30.000C obtained for the water used for the extraction, pH range 5.50+0.003 - 8.0+0.002. All the other extracts showed greenish coloration except *Eremomastex polysperman* extract that appears bluish purple. Electrical conductivity 24.00+0.012-145.20+0.053uscm-1. Suspended solid ranged between 1.60+0.001-3.60+0.008, dissolved solid 12.00+0.003-98.20+0.005, titratable alkalinity and acidity ranged 25.34+0.003-500.00+0.0012 and 40.44+0.005-900.00+0.128mg/l respectively. Biochemical oxygen demand BOD and dissolved oxygen DO values ranged 118.00+0.008-360.00+0.025 and 1.26+0.007-1.50+0.008 respectively. Total hardness ranged from 31.61+0.012 to 48.00+0.015. Finally sulphate, nitrate and chlorides values ranged within 2.10+0.003-24.20+0.004, 2.10+0.005-4.32+0.005 and 30.74+0.025-309.41+0.043 (mg/l) respectively. The extraction of these leaves in water has strongly altered the entire physiochemical parameter of drinkable water. These variations do not show any definite correlation. However when compared with the WHO permissible level despite the variations over 85% of the values were within the permissible limits. Thus these extract may not pose any danger with respect to physicochemical properties except for acidity. Thus one may apply caution when consuming these extract.

Introduction

In recent years, there has been on gradual revival of interest in the use of medicinal plants in developing countries because herbal medicines have been reported safe and without any adverse side effect especially when compared with synthetic drugs (Eiocan and Bara, 2007; Iwu *et al.*, 1999). Thousands of rural communities still depend mainly on folklore medicine to cure diseases in developing countries because they are cheap due to their proximity, reliability and age long practice. They are used by about 80% of the marginal communities around the world (Ibironke and Olusola, 2013).

Eremomastax polysperma belong to the acanthacaae family. It is commonly found in the tropic and the sub tropical forest (Pandey, 1996). It is known as pindula in swahili and edemididuot in Ibibio (Ajibesin *et al.*, 2008; Essiet, 2010). The part of the plant commonly used for medicinal purposes is the leaf. Petiole anatomy of the leaves show a succulent leaf with a purple under side maturity (Essiet, 2010). Oral administration of *E. polysperma* leaf mixed with egg helps in the treatment of anemia and internal heat (Etukudo, 2003). It is also used to treat pen figures in children (Bassey, and Effiong, 2011). The leaf extract is used for the treatment of male infertility among Ifa Nkari people of Akwa Ibom State (Erhabor *et al.*, 2013). The people of Cameroon employ the plant variously for malaria, kidney pain, nerves pain etc (Dibong *et al.*, 2011).

Ocimum gratisssimum commonly known as scent leaf is widely distributed in the tropics of Africa and Asia. It belongs to the family labiates and is the most abundant of the genes *Ocimum*. In the southern part of Nigeria, the plant is called "effinrin-nia" by the Yoruba speaking tribe, "Ahuji" in Igbo while in the northern part of Nigeria, the Hausas call it "Daidoya" (Effraim *et al.*, 2003). In the South-South particularly Akwa Ibom State, the leaf of this plant is popularly used for the treatment of malaria, skin infection, cough, diarrhea, fever and diaphoresis and also the water extract for enema.

Carica- papaya is one of the medicinal plants that have been used worldwide as a remedy, food, cosmetic and widely cultivated around the world it is one of the most popular and economically important plants in the world as its fruit is a common delicacy. (Rivera *et al.*, 2010). *C. papaya* leaf extract has potential anti-sickling (inhibition of sickle cell formation) properties. (Imago *et al.*, 2009). People in rural have used *papaya* leaf as alternative to treat dengue and dengue hemorrhagic fever. They believe that, this plant leaves can increases the platelet level of dengue patient shortly after receiving juice, boiled or raw of the leaves (Chavan, *et al.*, 2005).

Starchytarpheta cayennenisis is a species of flowering plant which belong to the family verbenaceae. It is commonly known as seemai nayruvi. It grows along roadsides, on disturbed sites, grass fields, brushwood, young forest, water

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sides and moreover cultivated as a hedge plant (Backer *et al.,* 1965). This plant has some used in traditional medicine, several Americans recognize extract of the plant as treatment to ease the symptoms of malaria. The boiled juice or a tea made from the leaves or the whole plant is taken to relieve fever and other symptoms. It is also used for dysentery pain and liver disorders. (Frelich *et al.*, 2008). A tea of the leave is also taken to control diabetes. Laboratory test shows that the plant has anti-inflammatory properties (Schapoval *et al.,* 1998).

Among the uses of these plants it is generally known that various forms of the water extract of almost all are used to treat one form of ailment or the other. Thus the need to examine the effect of these leave extract on the quality of water becomes almost necessary and will be investigated in this research.

Materials and Method

Sample Collection and Preparation

The four plant samples were obtained from different locations of Akwa Ibom State. *E. polysperma* from Nkari in Ini Local Government Area *O. gratissimum* from Abiakpo Ntak Inyang in Ikot Ekpene L.G.A, *C. papaya* from Abak Town in Abak L.GA. and *S. cayannensis* from Afaha Eket in Eket L.G.A. all in Akwa Ibom State, South-South of Nigeria. The samples were all separately packed in different dirt free polythene bags and transferred to the laboratory where the plant were washed, cut into small sizes and transferred separately into air tight extraction tanks and soaked with 1000ml of distilled water for 24 hours. After which the sample was filtered using Whatman No. 1 filter paper and a funnel. The filtrate was then stored and used for all analysis.

Laboratory Analysis: Standards methods according to Ademoroti, (1996) WHO (1995, 2008), AOAC (1984, 2004) were used for this study.

Temperature: The extracts and water temperatures were taken insitu by immersing the bulb containing the mercury of the mercury-in-glass thermometer below the surface immediately after preparation and values recorded.

- **pH**:The pH of all the extract and the water were obtained using electronic pH meter: model 3320 JENINAY after standardization. The pH meter probe was inserted into the liquid containers and the values obtained from the read out meter were recorded.
- **Appearance**: Since there is no standard method or standard terminology recommended (Ademoroti, 1960). The appearance of the extract was recorded as visually observed by the researcher.
- **Suspended or filterable, dissolved or nonfilterable and total solids**: Were all determined as described by Admoroti (1996).
- **Electrical Conductivity**: was determined as described by Ademoroti (1996). The conductivity cell was rinsed with three tube of standard KCl solution and the resistance of the fourth was measured and value recorded as R (KCl). Then the cell was rinsed with one tube of the extract solution and the resistance of the second tube measured. In this manner all the four extract were measured and also the water used for the extract. The following calculation were done.

First calculation of the cell constant K (cm^{-1})

$K = R\ (KCl) \times Ct\ (cm^{-1})$

Where R (KCl) is the resistance of standard KCl solution

Ct=Conductivity of the standard KCl solution of 25^0C.

$=0.0014135cm^{-1}$ or $1413\mu Scm^{-1}$

$$Cs = \frac{K}{R_S} = \frac{R\ (KCl) \times Ct\ (cm^{-1})}{R_S}\ Scm^{-1}$$

Where Rs =Measured resistance of samples (extracts or water).

Dissolved Oxygen (DO): DO was determined via the method of AOAC (2004) using Winkler's method where the burette readings were taken at the meniscus and recorded in mg/l as against the equation given by Boyd and Lichtkoppler (1979). For dissolved oxygen concentration in sampled water.

$$\text{Conc. of DO (mg/L)} = \frac{V_{(D)} \times N_{(D)} \times 8 \times 1000}{\text{Volume of Sample}}$$

Where (d) = volume of sodium thiosulphate used in titration and N (D) = Normality (0.025N) of sodium thiosulphate

Biochemical Oxygen Demand (BOD): This was determined as in DO except that the remaining samples were kept in an incubator in the dark at 20^0C for 5 days before the DO test was repeated.

Acidity and alkalinity were determined by methods of WHO (1995) and AOAC (2004).

Acidity: 50ml of each sample was measured into 250ml conical flasks. Three (3) drops of phenolphthalein indicator were added in each case and the solution titrated with $C0_2$ free NaOH solution (0.02m) until appearance of a faint pink colour (pH 8.3) was observed indicating the end point of the titration.

Calculation: $\text{Acidity (Mg/l } CaCO_3) = \frac{V \times M\ \ 100,000}{\text{Volume of Sample}}$

Where V = Vol of NaOH; M = molarity of NaOH. Molecular weight of $CaCO_3$ = 100g = 100,000mg.

Alkalinity: To $50cm^3$ of the samples 3 drops of phenolphthalein was added and solution titrated with 0.02m HCl until the colour disappears, and the phenolphthalein alkalinity was calculated as follows:

Alkalinity: $\text{(mg/l) as } CaCO_3 = \frac{V_P \times M \times 100,000}{\text{Vol of sample}}$

Where M=Molarity of acidVp= Volume of acid

Water Hardness: was determined by titration method using ethylene-diamin-tetraethanoic acid commonly called EDTA and the indicator solution of eriochrome Black T. 25ml of the sample was diluted to about 50ml with distilled water in an erlenmeyear flask. 1ml buffer solution was added and then titrated with EDTA until the final end point is obtained.

Calculation: Hardness (EDTA) mg/l as $CaCO_3$

$$= \frac{V \times A \times 1,000}{\text{Vol of sample}}$$

Where V = volume of titrant A = mg $CaCO_3$ equivalent to 1ml EDTA.

Nitrate was determined as described by AOAC (2004).

Nitrate (NO_3): 10ml of the different samples were transferred into different 25ml beakers and 2ml brucine reagent added, 10ml concentrated H_2SO_4 was then added rapidly and mixed for about 30 seconds then allowed to stand for 5mins. The flask was allowed to set in cold water for another 5 minutes and made up to mark with distilled water. The absorbance was read using UV/Vis spectrophotometer at 470nm.

Chloride: was determined according to method of WHO (1995). Three drops of 5% K_2CrO_4 indicator were added to 25ml of each sample and titrated with standard 0.02M $AgNO_3$ solution until the colour charged to brick red.

Results and Discussion

The physiochemical properties of the leave extract of four Nigerian medicinal plants along with the values of the water from which the extracts were prepared, WHO (2001) and NSDWQ (2007) permissible limits are presented in table 1.

Table 1: Results of the Physiolchemical Properties of the Aqueous extract of four Medicinal Leave.

Physiochemical parameters	***E. polyspermea***	***O. gratissimm* Extract**	***C. papaya Extract***	***S. cayemensis* Extract**	**Distilled Water**	**WHO 2001**	**Nsdwa (2007)**
Temperature ^{0}C	25.50±0.013	26.00±0.002	26.00±0.002	25.50±0.003	30.00	40.00	Ambient
pH	8.0±0.002	5.0±0.003	5.0±0.003	5.50±0.002	6.50	65-8.5	65-8.5
Appearance	Bluish purple	Greenish	Greenish	Greenish	Clear	-	-
Electrical conductivity (μSCM^{-1})	14±0.011	85.63±0.011	24.00±0.012	145.20±0.003	2.00	1000	1000
Suspended solid (mg/l)	3.60±0.008	2.80±0.004	1.60±0.001	1,80±0.001	-	<10	-
Dissolved solid (mg/l)	12.00±0.003	20.60+0.003	20.40±0.011	98.20±0.005	0.12	-	500.00
Total solid (mg/l)	15.60±0.001	23.40±0.21	22.00±0.007	100.20±0.009	0.12	<1,510	-
Titretable Alkalinity $CaCO_3$ (mg/L)	65.31±0.008	500.00±0.012	25.34±0.003	90.00±0.011	5.00	120	-
Biochemical oxgen demand (BOD)	360±0.025	118.00±0.008	308.00±0.013	150.00±0.043	10.00	0.2-5.0	-
Dissolved oxygen (DO)	1.85±0.007	1.52±0.001	2.50±0.008	1.26±0.0.007	2.20	7.5	-
Total Hardness	42.30±0.013	36.610±1.012	31.44±0.012	48.00±0.015	0.81	10-5.0	-
Sulphate(SO_4^{2-})	24.20±0.004	2.100±.0.003	18.00±0.011	13.95±0.005	3.56	42.00-45.00	-
Nitrate (No^-_3)	2.81±0.002	432±0.005	2.10±0.005	4.31±0.006	1.66	10.00	-
Titreable acidity $CaCO_3$	40.44±0.005	-	110.01±0.014	900.00±0128	-	-	0.2
Chloride content (mg/l)	188.00±0.125	34.67±0.031	30.74±0.025	309.41±0.043	21.65	250	-

Means of triplicate determination + standard deviation

Discussion

The results as presented in table 1 shows diverse variation in the physiochemical properties of the leaf extract of these medicinal plants the results were. Compared with the world health organization standards for drinking water and the value obtained for the distilled water from which the extract were prepared, since these extract are usually consumed orally as tea or juice and the following observation may be highlighted.

The temperature of the original water was reduced from 30°C to 25.5°C by *E. polysperm* and *S. cayennensis*, to 26°C by *O. gratissimum*, and C. papaya. Thus showing a general reduction in temperature, this may likely be the reason behind the constant use of these plants by locals for the treatment of internal heat. However since these values are within the recommended or permissible limit of temperature (40°C) by WHO (2001) for drinking water. The plants may not pose any danger to its consumer in term of temperature.

The pH values obtained for the leaves showed that only *E. polysperma* extract indicated a value (8.0) higher than the lower limit of the permissible level though still within the range of 6.8-8.5. While all other showed pH values lower than the lower limit. They also show lower level than the distilled water from which the extracts were prepared. This is an indication of increasing acidity of the water by *O. gratissium, C. papaya and S. cayennensis*, while the pH of *E. polysperma* increased from 6.5 to 8 indicating an increase in the basicity of the water solution. A pH of 7 in water solution is considered neutral and counting above 7 each number is ten times more basic than the previous while each number below is ten times more acidic than the previous (Adekunle *et al.*, 2004). Though the pH has no effect on human health, but its indirect action on physiological processes cannot be over looked (NSDWQ, 2007). According to James (2004) increased in acidity correlate increase in protein concentration.

The total dissolved solids (TDS) of all the extracts were higher than that for the water from which they were prepared. This shows that particles of the plants have dissolved into the liquid. This is basically why all the liquid change from clear colourless water to greenish due to green particles from the plant leaves. However the level reported for all the leaves are for less than the permissible limit of 1500mg/l by WHO, 500mg/l by NAFDAC (2001) and NSDWQ (2007). The TDS is the term used to describe the organic salt and small amount of organic matter present in water solution.

Total suspended solid is lower in all the mixture than the dissolved solid. The principal constituents are usually calcium, magnesium, sodium, potassium, carbonate, hydrogen carbonate, chloride, sulphate and nitrate anion (WHO, 1996). The presence of this TDS in water may be inacceptable because of its flat lipid taste that it results into. (Bruvold and Ongerth, 1969). Also suspended solid were generally higher in all the extract that the water but lower than the dissolved solids in all the samples. The values are within the permissible limit of 10 by WHO, (2001). Thus these extract may not pose any danger. As most of its solids are already dissolved into the liquid and only a few are suspended as colloidal particle.

The total alkalinity for all the extract are far higher than that of the water (5.00mg/l) but the values obtained for *E. polysperma*, *C. papaya* and *S. cayenensis* are within permissible level of 120 (WHO, 2001), but *O. gratissium* showed a very high level of 500mg/l which is far above the permissible limit. The first three plants mentioned here may not be dangerous in terms of alkalinity. Alkalinity is composed primarily of carbonate and bicarbonate and act as stabilizer for pH (Chavan *et al.*, 2005). While the continuous consumption of *O. gratissium* may not be healthy though may likely help to control stomach acidity to some extent. This has to be proven by nutritionist and health researchers.

Electrical conductivity is generally high for all the extract than for the water used, though the order of its increase is *C. papaya* (24.00). < *E. polysperma* (33.14) <*O. gratissimum* (55.63), < *S. cayennensis* (145.20) while the WHO (2001) permissible limit for electrical conductivity is 1000mg/l. These leaves extract are not dangerous with respect to conductivity. The highest value of conductivity 145.20μscm^{-1} recorded for *S. cayennensis* is also marked with highest level of chloride. Thus the *S. cayennensis* extract is a stronger electrolyte than the other extracts. Nevertheless the Cl^- content of all the plant extract are higher than that of the water from which they were prepared chloride is found in form of sodium or potassium in living cells constituting the most abundant electrolyte in most plant and animal cells.

According to Eneobong, (2002) and Lee, (1984), the function of chloride in living cell are in the maintenance of the electrolytic balance of the body fluid. The Cl^- content of *E. polysperma* (188m/l), *C. papaya* (30.74mg/l), *O. gratissimum* (34.67mg/l) are within the WHO permissible level of 250mg/l while the value 309.41mg/l obtained for *S. cayennensis* is above the permissible limit and should be administer with caution.

The dissolved oxygen obtained for the leaves were 1.85, 1.52, 2.50 and 1.26mg/l for *E. polysperma*, *O. gratissimum*, *C. papaya and S. cayannensis* respectively.

The value obtained for the water used was 2.20 while the permissible limit is 7.5. However, all the value are yet within the permissible level. Dissolved oxygen is essential to all forms of aquatic life including the organism that break down pollutants (Terezinha *et al.*, 2016). Its presence in the extract therefore may be useful to its consumers.

The level of BOD in the extracts were 360.00, 118.00, 308.00 and 150.00 for *polysperma, gratissimum, papaya and cayennessis* respectively against 10.00 recorded for the water from which the extract were prepared. Meanwhile the WHO permissible level is 0.20-5.00. Therefore all the extracts are highly BOD contaminated.

Total hardness was observed to have risen in all the extracts compared to the level in the water and all values are for above the permissible levels. These extract therefore can be regarded as hard water. This is an evidence of rise in calcium, magnesium contents of the water, showing some good levels of calcium in the plant. Since they are not within tolerant levels, the extracts are calcium contaminated and too hard for drinking purposes.

The extracts contain sulphates that are higher 24.20mg/l, 18.00mg/l and 13.93mg/l for *polysperma*, *papaya* and *cayennensis* than 3.56 obtained for the water used while 2.10mg/l recorded for *gratissimum* is lower than the value 3.56 for water. However all the values are below the lower limit of the permissible range of 42.00-45.00 by WHO (2001). Therefore the extract are SO_4^{2-} deficient and may not be healthy for drinking purpose.

Finally the values recorded for nitrates in all the extracts, 2.81,4.32, 2.1 and 4.31 for *polysperma*, *gratissimum*, *papaya* and *cayennensis* respectively are all within the permissible limit of 10.00 though they are all higher than the value 1.66 recorded for the water used. These extract therefore may not pose any danger in terms of nitrate to its users. Nitrates and phosphates are two of the major nutrients regards as major chemical factors that control phytoplankton growth productivity and abundance in water.

Conclusion

It is evidence from this research that these plants have altered to a great level the different physiochemical parameters of water by either increasing or reducing the levels as the case may be. Their characteristics must be very different as revealed by the diverse ways the levels of these parameters vary in the extract. There is no definite correlation in the order of increase or decrease among the levels of the different parameter there is therefore no doubt that they will also function differently serving different purposes. Therefore to be able to effective include these plant in any nutritional or herbal use, comprehensive nutritional and medical investigation need be thoroughly embarked upon, so that it users be appropriately informed and given correct prescription. This will help control problems of self-medication and addictions that may be fatal.

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