Phytochemical, Minerals and Antimicrobial Evaluation of *Trianthema portulacastrum (aizoaceae)* Leaf extract

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ABSTRACT:

The leaf extracts of the plant was analyzed for its phytochemical, mineral and antimicrobial activities using the standard method. The Phytochemical constituent revealed the presence of tannins in all the solvent system used for extraction. Alkaloids and phenol were present in dichloromethane, ethyl acetate, methanol and aqueous extract but absent in ethanol extract. Flavonoids, cardiac glycoside and saponins were present in aqueous and ethanol extract. Terpenoids was absent in aqueous but present in the other solvent used. The proximate composition revealed carbohydrate has the highest percentage content of 21.24%, protein: 21.10%, Fibre: 18.40%, Ash: 17.82% while lipid and moisture content are 11.94% and 9.5% respectively. The mineral composition in the leaf showed the contents (mg/g) as in order of Ca: 7.3303mg/g>K: 2.3297mg/g>Na: 1.3327mg/g>Mg: 0.7427mg/g>Fe: 0.6234mg/g>Mn: 0.2344mg/g>Cu: 0.0555mg/g>Zn: 0.0435mg/g>Cr: 0.0330mg/g>Pb: 0.0198mg/g>Ni: 0.0054>Cd: 0.0022mg/g The PPT/DCM/HP, FILT/DCM/HP, ETOA/HP and MTH/HP extract showed sensitivity in stephlococcus aureus, FILT/DCM/HP and MTH/HP are sensitive in Meth. resist staph aureous and Acinetobacter sp. The zone of inhibition against the test microorganisms occurred in the four (4) extract at 20mm, 20mm, 20mm and 22mm for staphylococcus aureus and the extract, PPT/DCM/HP(24mm), FILT/DCM/HP(20mm) and MTH/HP) in streptococcus pneumonia and PPT/DCM/HP(27mm), FILT/DCM/HP(20mm) and MTH/HP(20mm) for Acinetobacter sp. The results shows that the leaf extract plays significant role in the body metabolic system

Key words: Medicinal Plant, Minerals, Alkaloids, Flavonoids, Antimicrobial**.**

INTRODUCTION

Trianthema portulacastrium is a species of flowering plants in the ice plants family known by the common name desert horse purslane. It is a native to areas of several continents including Africa and North America and belongs to the family of *Aizoaceae*. In Nigeria, in the Northern part, Hausa call it Baba bajibji, in the Southeastern part, Igbo, call it, Ntioke; Ntilimoke and in the Southwestern part, Yoruba, call it Papasan.

The plant which can grow, often considered as a weed, all the aerial parts can be eaten, especially as salad plant. It is widespread as weed and has been ranked the eight most common plants in the world[1]. Purslane has a long history of use for human food, animal feed and medicinal purposes. Apart from its alimentary use, purslane has been traditionally used as medicinal plants. It has anti-inflammatory and analgesic properties (World Agricultural Production WAP[2] and antioxidant

activity. This plant can be used externally for various skin complaints, such as eczema, ulcers and acne and to relieve insect bites. It can also be used for cough, bronchitis and fever[3].The aim of this research work was to evaluate the Phytochemical, Proximate, antioxidant activity and its antimicrobial screening on the leaf extract using dichloromethane, ethyl acetate and methanol.

MATERIALS AND METHODS

Collection of Plant material

The fresh leaf of *Trianthema Portulacastrum* were collected from Sheda, Kwali, F.C.T. Abuja, Nigeria. The Plant leaf was identified by the Taxonomist at the National Institute for Pharmaceutical Research and Development (NIPRD), Idu, Industrial Area, Abuja, Nigeria. The leaf was washed and dried for two weeks, was then powdered using a grinder and stored in a dry container. The herbarium voucher number (*Trianthema Portulacastrum* NIPRD/H/7342)

Preparation of Plant Extract

The n-hexane, dichloromethane, ethyl acetate and methanol leaf extracts of the medicinal plant were obtained successively by macerating the powdered leaves material[4]. 20ml of n-hexane were added to dichloromethane extract and left to precipitate, then filtered. The extracts were concentrated using a rotary evaporator under vacuum with the water bath set at 45^{o} C. Both the precipitate and filtrate were left to dry and weighed

Phytochemical Screening

Standard methods as described by[5,6] were adopted for the phytochemical screening of the extracts of *Trianthema Portulacastrum* leaf.

Test for Alkaloids: 5g of the powdered leaves were placed in a test tube and 20ml methanol poured into the test tube. The mixture was allowed to boil for 2minutes in water bath, cooled and then filtered. Two drops of dragendoff's reagent (Solution of Potassium Bismuth Iodide) was added to 2ml of filtrate; Two drops of wagner's reagent (Solution of Iodine and Potassium Iodide) was added to 5ml of the filtrate; Two drops of Meyers reagent (Potassium mercuric +Iodide solution) was added to another 2ml filtrate and two drops of hager's reagent (saturated solution of Picric acid) was added to a 5ml of filtrate. The alkaloids are precipitated from the above solutions giving characteristic colours-reddish brown, reddish brown, cream and yellow respectively.

Test for Flavonoids: 10ml of ethyl acetate was added to about 0.2g of the powdered plant material and heated on a water bath for 3minutes. The mixture was cooled, filtered and 4ml of filtrate is shaken with 1ml of dilute ammonia solution. The layers are allowed to separate and the yellow colour in the ammonical layer indicates the presence of flavonoids.

Test for Terpenoids: 5ml of plant extract were mixed in 2ml of chloroform and 3ml concentrated sulphuric acid carefully added to form a layer. A

reddish brown colour interface was formed to show positive results for terpenoids.

Test for Saponin: 2g of the powdered sample was boiled in 20ml of distilled water in water bath and filtered.10ml of the filtrate was mixed with 5ml of distilled water and shaken vigorously for a stable persistent froth. The froth was mixed with three drops of Olive oil and shaken vigorously, then observed for the formation of emulsion.

Test for Steroids: A 9ml portion of ethanol was added to 1g of the powdered leaves. This was refluxed for a few minutes and filtered. The filtrate is concentrated to 2.5ml on a boiling water bath and 5ml of hot water was added. The mixture is allowed to stand for 1hour and the waxy matter filtered off. The filtrate extracted with 2.5ml of chloroform using a separating funnel. 0.5ml of the chloroform extract in a test tube. 1ml of concentrated sulphuric acid was added to form a lower layer. A reddish-brown interface shows the presence of steroids.

Test for Tannins: 0.5g of the powdered leaves sample was boiled in 20ml of water in a test tube and then filtered. A few drop of 0.1% ferric chloride was added and observed for brownish green or blue- black colouration.

Test for cardiac glycosides (Keller –killani Test): 5ml of the extracts was treated 2ml of glacial acetic acid containing a drop of ferric chloride solution. This was underplayed with 1ml concentrated sulphuric acid. A brown ring of the interface is a deoxysugar characteristic of cardenolides. A violet ring appears below the brown ring, while in the acetic acid layer, a greenish ring may form just gradually throughout thin layer.

Proximate Analysis

The proximate analysis was carried out in triplicate with the result being presented as mean values. The analysis was carried out by adopting standard method as described[7].

Determination of Minerals Content

The mineral content of the sample were analysed by AOAC method[7,8] using Atomic Absorption Spectrophotometer .But Na and K were determined by flame photometer. The digested samples in Conc.HNO_3 were made up to 100ml and stored in polypropylene container. Atomic Absorption Spectrophotometer was employed to determine the elements.

Anti-microbial Screening

The antimicrobial screening of the dichloromethane, ethyl acetate and methanol of Horse Purslane leaves were carried out by well diffusion method[9]. The plates were prepared by pouring sterile Muller Hinton agar into sterile petric dishes that were autoclaved. Sterilized cotton swabs were dipped in the bacterial culture in nutrient broth and swabbed on the agar plate.

Sizes were cut with proper gap in the medium and the extracts were added into it. The plates were allowed to stand for one hour, to allow pre-

diffusion of the extracts into the medium[10]. The plates were incubated at 37°C for 24hours.The standard drug used was Sparfloxacin, Ciprofloxacin and Streptomycin. At the end of the incubation period, inhibition zone were measured in millimeter. The studies were carried out in triplicates.

RESULTS AND DISCUSSION

The Phytochemical screening as shown in Table-1, revealed the presence of tannins in all the leaf extracts. Alkaloids and phenols were absent in ethanol extract but present in dichloromethane, ethyl acetate, methanol and aqueous extract. Flavonoids, cardiac glycoside and saponins were present in aqueous and ethanol extracts but absent in dichloromethane, ethyl acetate and methanol. Terpenoids was absent in aqueous extract but present in the other solvents used.

The plant leaf sample was investigated for its proximate composition as shown in Table-2. The result revealed that carbohydrate has the highest percentage (%) content of 21.24%, the protein contain 21.10%, which signified that the leaf can be use as a good source of carbohydrate and protein, fibre 18.40%, ash contain 17.82%, lipids and moisture revealed 11.94% and 9.50% respectively.

Table 3 revealed that the highest quantity of calcium Ca (7.3303mg/g) was found in the leaf of *Trianthema Portulacastrum* followed by potassium K (2.3297mg/g). The presence of minerals such as calcium Ca and potassium K which help to promote adequate removal of the anti-nutritional factor by formation of complex formation, complex formation between calcium and oxalates makes more calcium unavailable. The leaf also revealed that Sodium Na and Magnesium Mg have the highest contribution. The mineral composition in the leaf investigated showed the contents (mg/g) as in the order of Ca>K>Na>Mg>Fe>Mn>Cu>Zn>Cr>Pb>Ni>Cd . This macro element plays an important and vital role in our body system, potassium K and sodium Na regulate the osmotic and acid base balance of the body fluid system, and they are the major cations of intracellular fluid. Sodium Na shows a special role in nervous transmission and is considered an active transport system of sugars and amino acids. Potassium K plays an active and important role in ionic basis of muscle excitability and also as cofactor for several reactions in carbohydrate metabolism.

The PPT/DCM/HP, FILT/DCM/HP, ETOA/HP and MTH/HP extract in Table-4, showed sensitivity in *Staphylococcus. aureus* except MTH/HP in *Escherichia Coli* and FILT/DCM/HP resistant to *Klebsiella Pneumoniae* microorganism. FILT/DCM/HP and MTH/HP are sensitive in *Meth. resist staph. aureus* and *Acinetobacter. Sp;* PPT/DCM/HP and ETOA/HP are resistant to *Meth. Resist staph. aureus* and *Acinetobacter. Sp.* The zone of inhibition of the extract against the test micro-organism in Table-5 investigated occurred in the four (4) extract for *staphylococcus*

microorganism and for the extract of PPT/DCM/HP, FILT/DCM/HP and MTH/HP as shown in *Streptococcus. Pneumoniae* and *Acinetobacter*. *Sp.* The control against the test organism in Table-6 and Table-7 revealed that *Acinetobacter*. *Sp.* was sensitive against the drugs used. Similarly, the zone of inhibition against test micro-organism occurred at 27mm for Sparfloxacin, at 31mm for Ciprofloxacin 31mm and *at* 29mm for *Streptomycin* respectively.

Tables-1: Phytochemical Screening

No.	Parameters	Dichloromethane	Ethanol	Methanol	Aqueous	Ethanol
1	Alkaloid	+	+	+	+	-
2	Flavonoid	-	-	-	+	+
3	Terpenoid	+	+	+	-	+
4	Phenol	+	+	+	+	-
5	Saponin	-	-	-	+	+
6	Tannin	+	+	+	+	+
7	Cardiac glycoside	-	-	-	+	+

Table-2: Proximate Analysis

Parameters	Moisture	Lipid	Ash	Fibre	Protein	Carbohydrate
Percentages %	9.50	11.94	17.82	18.40	21.10	21.24

Table-3: Mineral composition

No.	Parameters	Conc. mg/g
1	Iron Fe	0.6234
2	Copper Cu	0.0555
3	Potassium K	2.3297
4	Magnesium Mg	0.7427
5	Chromium Cr	0.0330
6	Lead Pb	0.0198
7	Sodium Na	1.3327
8	Manganese Mn	0.2344
9	Cadmium Cd	0.0022
10	Zinc Zn	0.0435
11	Nickel Ni	0.0054
12	Calcium Ca	7.3303

Table-4: The Antimicrobial Activity

Test Organism.	PPT/DCM/HP	FILT/DCM/HP	ETOA/HP	MTH/HP
Methicillin resist staph aureus	R	S	R	S
Vancomycin resist enterococci	R	R	S	R
Staphylococcus aureus	S	S	S	S
Escherichia coli	S	S	S	R
Acinetobacter sp.	R	S	R	S
Klebsiella pneumoniae	S	R	S	S
Salmonella sp	S	R	R	S
Heamophilus influenza	S	R	S	R
Streptococcus pneumonia	R	S	R	S

Key; S=Sensitive; R= Resistance

Table-5: Zone of Inhibition of the extract against the test microorganism.

Test Organism	PPT/DCM/HP	FILT/DCM/HP	ETOA/HP	MTH/HP
Methicillin resist staph aureus	0	21	0	20
Vancomycin resist enterococci	0	0	21	0
Staphylococcus aureus	20	20	20	22
Escherichia coli	20	19	21	0
Acinetobacter sp.	27	20	0	23
Klebsiella pneumonia	0	0	0	23
Salmonella sp	0	0	0	20
Heamophilus influenza	0	0	22	0
Streptococcus pneumonia	24	20	0	21

Table-6: The Control against the microorganisms

Organism	Sparfloxacin	Ciprofloxacin	Streptomycin
Methicillin resist staph aureus	S	R	R
Vancomycin resist enterococci	R	S	R
Staphylococcus aureus	S	R	S
Escherichia coli	R	S	R
Acinetobacter sp	S	S	S
Klebsiella pneumonia	S	R	S
Salmonella sp	R	S	R
Heamophilus influenza	S	R	R
Streptococcus pneumonia	S	R	R

Key; S= Sensitive; R= Resistance

Table-7: Zone of Inhibition of Drugs against the test microorganism

Test Organism	Sparfloxacin	Ciprofloxacin	Streptomycin
Methicillin resist staph aureus	30	0	0
Vancomycin resist enterococci	0	29	0
Staphylococcus aureus	32	0	30
Escherichia coli	0	37	0
Acinetobacter sp	27	31	29
Klebsiella pneumonia	30	0	31
Salmonella sp	0	39	0
Heamophilus influenza	28	0	0
Streptococcus pneumonia	25	0	0

Abbreviations:

PPT= Precipitate; FILT=Filtrate; ETOA= Ethyl acetate; MTH= Methanol and HP= *Trianthema Portulacastrum*

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