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PHYTOCHEMICAL SCREENING, TLC PROFILE AND ANTIOXIDANT ACTIVITIES OF CRUDE METHANOL EXTRACT OF *PTELEOPSIS HABEENSIS* (AUBREV EX KEAY)

Falmata M. Aliyu*[1], Abdulqadir Bababe[1], Mohammed Kachallah[2], H. B. Yesufu[1], Fatima A. Goje[3], Emmanuel A. Gwa[3]

[1*]Department of Pharmaceutical Chemistry, University of Maiduguri, P.M.B 1069, Maiduguri, Nigeria.
[2]Department of Pharmaceutics and Pharmaceutical Microbiology, University of Maiduguri, P.M.B 1069, Maiduguri, Nigeria.
[3]Department of Pharmacology and Toxicology, University of Maiduguri, P.M.B 1069, Maiduguri, Nigeria.

ABSTRACT

Analysis of plant screening for phytochemical constituents seems to have the potential to act as a source for useful drugs for many infections because of various bioactive compounds present that have activity against array of human pathogens. *Pteleopsis habeensis* is widely use in Northern part of Nigeria for management of a number of ailment. Whole plant of *P. Habeensis* was extracted using methanol by maceration and extracts was screened for presence of phytochemicals and free radical scavenging activity using standard procedures. The phytochemical result indicated *P. Habeensis* contains Alkaloids, Flavonoids, Tannins, Anthraquinones, Saponins, Steroids, Cardiac glycosides and Terpenoids. The DPPH Assay showed high free radical scavenging activity of the extract that is comparable to Ascorbic acid. Statistically, no significant difference between antioxidant property of the extract and standard. The presence of such phenolic compounds suggest the medicinal value of the plant and the bioactive constituents can be isolated for pharmaceutical use.

KEYWORDS

Pteloepsis habeensis, TLC profile and Antioxidant activity.

Author for Correspondence:

Falmata M. Aliyu,
Department of Pharmaceutical Chemistry,
University of Maiduguri,
P.M.B 1069, Maiduguri, Nigeria.

Email: falmataliyu@gmail.com

INTRODUCTION

Plants are rich sources of antioxidants. The antioxidant compounds of plant are due to the presence Flavonoids, tannins and the carotenoids[1].

Free radicals are generated in the body during metabolic activities. These free radicals are harmful to the body when they are produced, the natural antioxidants from plants inactivates the free radicals from posing harmful effect to the body system[2].

One of the most known free radical generated in the body during metabolism is the reactive oxygen species (ROS). ROS; superoxide's anion, Hydrogen peroxides, radicals are chemically reactive molecules that are highly reactive due to presence of unpaired electrons. They are mostly generated as by-products of metabolic reaction as well as in response to other stimuli[3].

ROS play a significant role in cell signalling and homeostasis. However, over production of ROS due to responses from stressors like drugs, diseases can lead to oxidative stress[4].

Oxidative stress is associated with heart diseases, neurodegenerative diseases, cancer and aging process[5]. Natural antioxidant opposes oxidative stress and lower the risk of diseases such as cardiovascular, diabetic complications, cancers, hypertension and fatigue syndrome[6,7]. Antioxidation using exogenous antioxidant is an extremely significant activity which can be used as a preventive agent against a number of diseases.

The commonly added antioxidants are synthetic phenols, such as Butylated Hydroxy Toluene (BHT) and Butylated Hydroxy Anisole (BHA).

The folkloric use and antioxidant properties of some plants have long been known but a large number of them remain unexplored. This is the first study to determine the antioxidant property and TLC profile of *Pteolopsis habeensis.*

Pteleopsis habeensis (Aubrev ex Keay) is a plant which belongs to the family Combretaceae, and known as Lallengiwa in Hausa language in northern part of Nigeria of Sub Saharan Africa. *P. Habeensis* is a small genus of about 10 species, occurring in 3 localities, all in tropical Africa. It is an important medicinal plant native to Nigeria, Ghana and Mali[8]. The shrub is use for the treatment of Malaria fever, stomach ache, Aphrodisiac and in the destruction of tumours[9].

MATERIAL AND METHODS

Chemicals and reagents

Dragendorff reagents, distilled water, Methanol, Conc Sulphuric acid, Mayers reagent, Wagners reagent, Fehlings solution A and B, 10% Ferric chloride. All the chemicals and reagents used in this study were of analytical grade.

Test tubes, test tubes racks, weighing balance, capillary tube, filter paper, Silica gel, spatula, chromatographic plates, chromatographic tank, Oven and dryer, UV spectrophotometer, beakers, conical flask.

Plant Collection

The whole plant of *P. habeensis* was collected in December 2016 at College of Agric, Farm centre, Maiduguri, Nigeria. It was identified at by a consultant herbalist and authenticated at department of Biological Science, University of Maiduguri, Borno State, Nigeria. The plant was washed under running tap water and shade dried for 72 h. It was grounded into coarse powder with an electronic blender and then stored in air-tight containers for further analysis.

Extraction

Analytical grade Methanol was used for the cold extraction of 500g pulverized *P. Habeensis*; macerated in the solvent in extraction bottle with the level of the solvent above that of the plant material and allowed to stay for 2-3 days. The liquid was then removed and filtered through Whatman No.1 filter paper. The filtrates from the mixture was then concentrated over thermostatic water Cabinet at 100◦C. The plant extracts recovered were transferred into beaker and allowed to stand on the bench to allow for total evaporation of the residual solvent. The dried plant extracts were preserved until required for use.

Phytochemical Screening.

The phytochemical analysis of the *Pteleopsis habeensis* whole plant extract was carried out using the standard methods[10-12]. Tests for Alkaloids, Tannins, Flavonoids, Saponins, Anthraquinones and Cardiac glycosides were conducted using the standard procedures[10-12].

TLC Screening

The Methanol extract was fractionated by Chromatography; on Silica gel serving as the stationary phase. Various solvent systems were used as mobile phase. These are n-hexane (100%), n-hexane/Chloroform (90/10), n-hexane/Chloroform (80/20), n-hexane/ethyl acetate (95/5%), n-

hexane/ethyl acetate (90/10). The solvent systems were eluted and a number of fractions were collected.

25 fractions of 5ml were obtained from the fractionation and were allowed to dry in open air in the lab. Thin layer Chromatography of the crude methanol plant extract and different column fraction was carried out using prepared TLC plates. 1 cm was measured from the base of the TLC plate, marked with a pencil and labeled. Capillary tube was used to spot the plates with the column eluents. Small quantities of the eluents were collected with capillary tube by dipping it in the solution. They were then used to spot the plates and put in a lidded tank containing the solvent system, Chloroform (100%). The level of solvent system in the tank was about 1 cm beneath the origin. The solvent travelled up the plate by capillary action till it reached the solvent front, marked by a straight line across. The plates ware dried before they were visualized by spraying with 10% Sulphuric acid.

Determination of Free Radical Scavenging Activity

The determination of free radical scavenging activity of the extracts was carried out using the DPPH (2, 2-diphenyl-1-picrylhydrazyl radical) assay as described by Mensor *et al*[13]. Varying concentrations of 6.25, 12.5, 25, 50 and 100 µg/mL of the extracts in methanol were prepared. 1.0 mL (0.25 mM) DPPH in methanol was added to 2.0 mL solution of the extracts/standard (Ascorbic acid) which allowed to stand at room temperature in a dark chamber for 30 min. Variation in colour from deep violet to light yellow was observed and the absorbance was measured at 518 nm using the spectrophotometer. The decrease in absorbance was converted to percentage antioxidant activity (% AA) using the formula:

$$\% \text{ of radical scavenging activity} = \frac{\text{Abs. control} - \text{Abs sample}}{\text{Abs. control}} \times 100\%$$

RESULTS

Extraction

The percentage yield (%) can be seen in Table No.1 below.

Phytochemical screening

The results of the phytochemical screening conducted on extracts are demonstrated in Table No.2 below.

TLC Screening

The results of the TLC screening of bioactive compounds present in the fractionated fractions are demonstrated in Figure No.1

Percentage Antioxidant Activity

The percentage antioxidant activity of the extracts is demonstrated in Table No.3 and Figure No.2.

DISCUSSION

The present study revealed the presence of different secondary metabolites like tannins, saponins alkaloids, glycosides, flavonoids, and others from phytochemical analysis of *P. Habeensis* which indicate that the plants are rich sources of bioactive compounds.

It is well known that these phytoconstituents have already exhibited antioxidant and antimicrobial activity[14]. This phytochemicals present may be responsible for the plant's antimalarial, anticancer, antidiarrheoa activity as well as justify its basis for folkloric use in traditional medicine.

Thin layer Chromatography revealed the presence and separation of the components of the extracts. From the result obtained, *Pteolopsis habeensis* contains a remarkable number of phytochemical components with potential bioactivity. This is in conformity with the studies of Baba-Moussa[15]. That reported the presence of bioactive phytochemicals and all *Pteolopsis*s pecies have antimicrobial and antioxidant activity.

The free radical scavenging activity demonstrated by the extracts (Table No.3) showed that methanol extract of *P. Habeensis* has a high % of radical scavenging activity. This high activity demonstrated by the extract may be attributed to the presence of the phenolic compounds (Tannins and flavonoids) in the extracts. The free radical scavenging activity is concentration dependent. The higher the concentration the higher the % free radical scavenging activity.

The 100 µg/ml concentration of Methanol extract has highest Percentage free radical scavenging

activity (97.155 %) which can be favourably compared with Ascorbic acid (97.69%).

It was observed after statistical analysis, no significant difference was seen between 50 µg/ml concentration of Methanol extract and 50 µg/ml Ascorbic acid.

Statistical analyses also showed no significant difference between 100 µg/ml of both the extract and standard (Vitamin C).

Table No.1: Weight of Extracts and % Yield

S.No	Sample	Methanol Extract
1	Weight (g)	3.5
2	Yield (%)	0.74

Table No.2: The Phytochemical constituent of *P. habeensis*

S.No	Phytocontituents	Methanol Extract
1	Saponin	+
2	Tannin	+
3	Alkaloids	+
4	Anthraquinone	+
5	Cardiac Glycoside	+
6	Steroids	+
7	Flavonoids	+

Table No.3: Percentage Antioxidant Activity

S.No	Conc (µg/ml)	Methanol extract (%)	Vit. C (%)
1	6.25	53.93	96.69
2	12.50	62.34	96.98
3	25.00	82.51	95.95
4	50.00	98.59	96.22
5	100.00	97.18	97.24

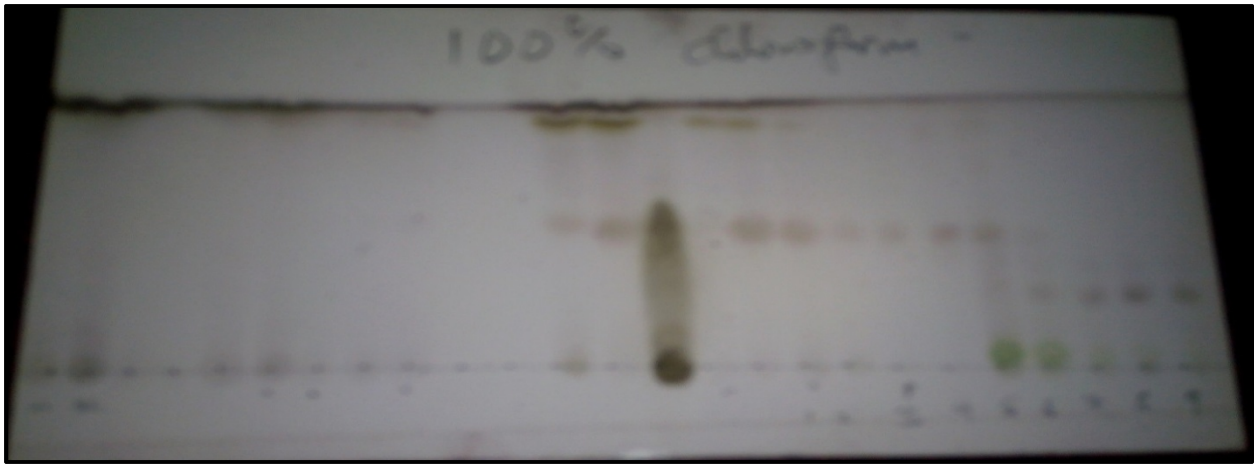

Figure No.1: TLC screening of bioactive compounds present in the fractionated fractions of *P. habeensis* with 100% Chloroform as developing system

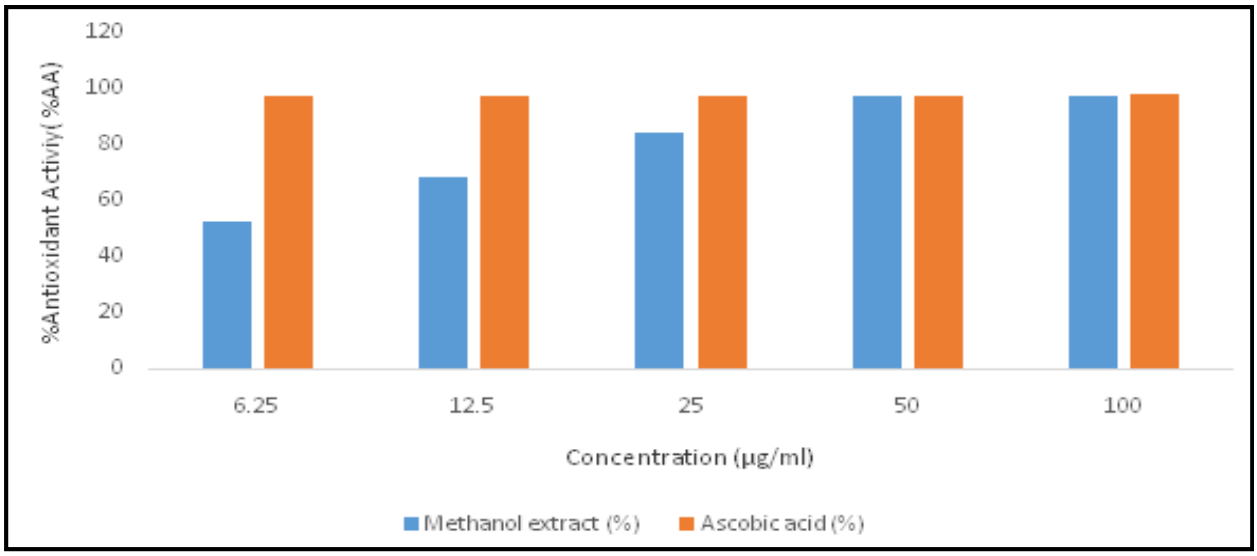

Figure No.2: Percentage Antioxidant Activity of *P. habeensis* extract

CONCLUSION

From the result, it can be concluded that *P. Habeensis* contains bioactive compounds and exhibited profound antioxidant activity that is comparable to Ascorbic acid.

Further studies is required to isolate the principal constituents responsible for the observed antioxidant activity. This may be attributed to the presence of phenolic compounds and other compounds in the extract.

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CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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