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Research paper

Phytochemical Composition of Ethyl Acetate, Dichloromethane, n-Hexane and Methanol Leaf Fractions of Gongronema Latifolium

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Abstract

The phytochemicals present in ethyl acetate, dichloromethane, n-hexane, and methanol leaf fractions of Gongronema latifolium were identified using a phytochemical profiling method. The crude extract of Gongronema latifollium (990g) was dissolved in distilled water and partitioned successively and exhaustively with methanol, n-hexane, dichloromethane, and ethyl acetate using a separating funnel. Alkaloids were present in high concentration (+++) in all four leaf extracts. Flavonoids and terpenoids were observed to be present in high concentration (+++) except in n-hexane and ethyl acetate leaf extracts, respectively, which are moderately high concentration (++). Cardiac glycoside was moderately high (++) in all four leaf extracts. From the results of this study, it can be suggested that the fractions of Gongronema latifolium contain significant phytochemicals in very high and moderately high concentrations, which may significantly alleviate some health challenges.

Keywords: *Phytochemicals; Concentration; Alkaloid; Flavonoid; Antioxidant.*

1. Introduction

1.1. Background of the study

The variety in the bio-system of plants in the tropical rainforests is embedded with countless quantities of natural plant products, which are of high nutritional and medicinal benefits. One such natural plant is Gongronema latifolium Benth, which belongs to the family of Asclepiadaceae, formerly known as Marsdenia latifolia (Okafor, 1975). Gongronema latifolium Benth (Asclepiadaceae), which is known as a climbing perennial and edible herbaceous plant of non-woody shrub, is commonly grown in Nigeria. It is generally and commonly called Amaranth globe and locally called urasi by Ibibios, Efiks and Quas, utazi by the Igbos and arokeke by the Yorubas (Agbo et al., 2005). The plant is usually found in the tropical rainforest, secondary forests, deciduous forests, and in mangroves. The wide distribution of G. latifolium also extends to Senegal and Ghana, where it is referred to as "Server gasule" and "Akan-Asante aborode" respectively (Hutchinson, 1973). It is a very nutritious and edible plant which contains a sharp and slightly sweet taste when eaten fresh. The leaves of G. latifolium are rich in proteins, fats, minerals, vitamins, and many amino acids, which are important and contribute collectively due to its high nutritional value (Eleyinmi, 2007). "Utazi" is commonly utilized as a vegetable in salad preparations and soup or as a spice in dried form, as powder (Dalziel et al.,1937; Okafor, 1975; Morebise et al., 2002).
Medicinally, the sliced plant is added to water or boiled with lime juice for at least three days to produce liquor; it can work as a laxative against intestinal worms or for colic and stomach pain (Okafor, 1975; Onike, 2010). The leafy part of the vegetable can be planted from its seed or by planting the cut softwood or root. In Eastern Nigeria, the leaves are used to prepare soup for mothers during the lactating period to reduce post-partum contraction and stimulate appetite. A study has revealed from an animal study of the ethanolic extract of G. latifolium on male Wistar rats, which exhibited an increase in follicle-stimulating hormone, serum concentration of testosterone and luteinizing hormone at doses of 100 mg/kg and 200 mg/kg body weight, even decreased in the levels of progesterone.
Gongronema latifolium contains several active chemicals of which some have been elucidated, thereby having better medicinal qualities; therefore, this present work identifies the phytochemicals present in ethyl acetate, dichloromethane, n-hexane, and methanol leaf fractions of Gongronema latifolium.

2. Methodology

2.1. Materials

2.1.1. Chemicals and equipment

Chemicals and reagents of analytical grade were used for this study. The equipment and glass wares used in this research include cages, centrifuge (Microfield 80-2), refrigerator, syringe and needle, plain tubes, EDTA tubes, dissecting set, hand gloves, spectrophotometer (721 UV-5500 (PC), Xian Toption Instrument Company, China) light microscope (BH2, Olympus Corporation, Japan) automated micropipette, test tubes, micro plate reader and water bath.

2.2. Methods

2.2.1. Collection and identification of plant sample

Fresh Gongronema latifolium leaves were obtained at Ikot Ambang market, Ibiono Ibom Local Government Area, Akwa Ibom State, Nigeria. The plant sample was identified by taxonomist Prof. Margaret I. Essien of the Department of Botany and Ecological Studies, University of Uyo, Akwa Ibom State, Nigeria. It was given the voucher number UUPH 1(b) and was deposited at the herbarium of the Department of Pharmacognosy and Natural Medicine, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria.

2.2.2. Preparation and extraction of plant material

Fresh leaves of Gongronema latifolium were washed, cut into smaller pieces, and dried in the shade for two weeks. The leaves were further pulverized to powder using an electric grinder. The powdered leaf material (5 kg) was gradually macerated for 72 hours in 5litres of 70% ethanol, with intermittent stirring. Thereafter, the suspensions were filtered with cheesecloth and then subsequently with Whatman No. 1 filter paper to obtain the filtrate of the plant sample. The filtrates were dried in a Water Bath at 40 ºC to yield what is referred to as the extracts. It was weighed, and the weights used in estimation of the yield.

2.2.3. Partitioning of crude extract

The crude extract of Gongronema latifollium (990g) was dissolved in distilled water and partitioned successively and exhaustively with methanol, n-hexane, dichloromethane and ethyl acetate using a separating funnel (Pyrex, England). The respective liquid fractions were obtained in each case. The fractions were then concentrated at 40 ºC to dryness using a rotary evaporator. The dried fractions were weighed and preserved in a refrigerator at 4 ºC until subsequently used for the study. The extraction and partitioning procedures are highlighted schematically in Figure 3.1 below.

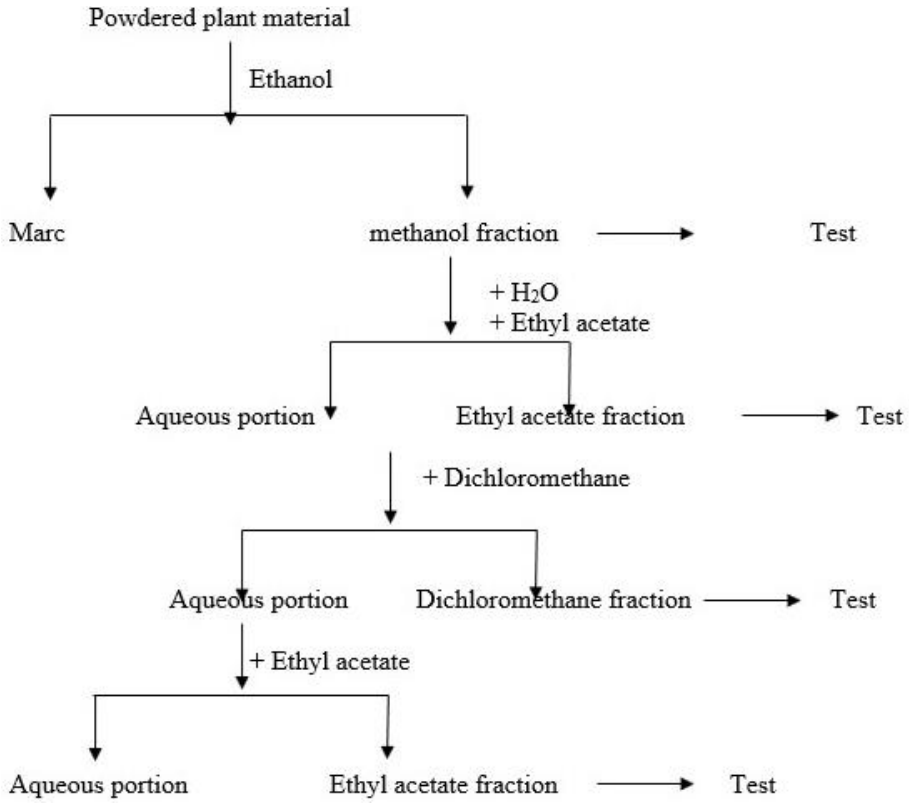

Fig. 1: Partitioning of Crude Extract.

Source: Field Data (2024).

2.2.4. Phytochemical analysis

Aliquots of the ethanolic extracts were subjected to phytochemical investigation, using the method described by Trease and Evans 2001. The tests carried out were to confirm the presence or absence of alkaloids, saponins, flavonoids, tannins, glycosides, steroids, carbohydrates, Protein, Anthraquinones, Terpenoids, and cardiac glycosides.

3. Results and Discussion

3.1. Results

3.1.1. Phytochemical screening of Gongronema latifolium methanol fraction

The result of the phytochemical screening of the methanolic fraction of G. latifolium is presented in Table 1. A total of eleven (11) phytochemical principles were screened for in the methanol leaf fraction of G. latifolium. Alkaloids, flavonoids, steroids, saponins, and terpenoids were present in higher concentrations, while protein, carbohydrate, and cardiac glycoside were present in moderately high concentrations. Similarly, tannins and anthraquinones were present in small concentrations. However, glycosides were completely absent in the methanol fraction of Gongronema latifolium.

Table 1: Phytochemical Analysis Result of Methanolic Fractions of G. Latifolium Leaves

S/No	Chemical constituents	Gongronema latifolium
I	Alkaloids	+++
II	Flavonoids	+++
III	Saponins	+++
IV	Glycosides	-
V	Carbohydrates	++
VI	Protein	++
VII	Steroids	+++
VIII	Anthraquinones	+
IX	Tannins	+
X	Terpenoids	+++
XI	Cardiac glycosides	++

Source: Field Data (2024).

− not present;
+ present in small concentration;
++ present in moderately high concentration;
+++ present in high concentration;

3.1.2. Phytochemical screening of Gongronema latifolium ethyl acetate fraction

The result of the phytochemical screening of the ethyl acetate fraction of G. latifolium is presented in Table 2. A total of eleven (11) phytochemical principles were screened for in the ethyl acetate leaf fraction of G. latifolium. Flavonoids, steroids, and alkaloids were present in higher concentrations, while saponin, carbohydrate, protein, anthraquinones, terpenoids, and cardiac glycoside were present in moderately high concentrations. Tannins were present in small concentrations; however, glycosides were completely absent in the ethyl acetate fraction of Gongronema latifolium.

Table 2: Phytochemical Analysis Result of Ethyl Acetate Fractions of G. Latifolium Leaves

S/No	Chemical constituents	Gongronema latifolium
I	Alkaloids	+++
II	Flavonoids	+++
III	Saponins	++
IV	Glycosides	-
V	Carbohydrates	++
VI	Protein	++
VII	Steroids	+++
VIII	Anthraquinones	++
IX	Tannins	+
X	Terpenoids	++
XI	Cardiac glycosides	++

Source: Field Data (2024).

− not present;
+ present in small concentration;
++ present in moderately high concentration;
+++ present in high concentration;

3.1.3. Phytochemical screening of Gongronema latifolium dichloromethane fraction

The result of the phytochemical screening of the dichloromethane fraction of G. latifolium is presented in Table 3. A total of eleven (11) phytochemicals were screened in the dichloromethane leaf fraction of G. latifolium. Flavonoids, terpenoids, and alkaloids were present in higher concentrations in the fraction, while saponin, carbohydrate, protein, steroids, and cardiac glycoside were present in moderately high concentrations. Anthraquinones and tannins were present in small concentrations; however, glycosides were completely absent in the dichloromethane fraction of Gongronema latifolium.

Table 3: Phytochemical Analysis Result of DCM Fractions of G. Latifolium Leaves

S/No	Chemical constituents	Gongronema latifolium
I	Alkaloids	+++
II	Flavonoids	+++
III	Saponins	++
IV	Glycosides	-
V	Carbohydrates	++
VI	Protein	++
VII	Steroids	++
VIII	Anthraquinones	+
IX	Tannins	+
X	Terpenoids	+++
XI	Cardiac glycosides	++

Source: Field Data (2024)

− not present;
+ present in small concentration;
++ present in moderately high concentration;
+++ present in high concentration;

3.1.4. Phytochemical screening of Gongronema latifolium n-hexane fraction

The result of the phytochemical screening of the n-hexane fraction of G. latifolium is presented in Table 4. A total of eleven (11) phytochemical principles were screened for in the n-hexane leaf fraction of Gongronema latifolium. Terpenoids and alkaloids were present in higher concentrations in the fraction, while flavonoids, saponin, carbohydrate, protein, steroids, anthraquinones, and cardiac glycoside were present in moderately high concentrations. Tannins and carbohydrates were present in small concentrations; however, glycosides were completely absent in the dichloromethane fraction of Gongronema latifolium.

Table 4: Phytochemical Analysis Result of N-Hexane Fractions of G. Latifolium Leaves

S/No	Chemical constituents	Gongronema latifolium
I	Alkaloids	+++
II	Flavonoids	++
III	Saponins	++
IV	Glycosides	-
V	Carbohydrates	+
VI	Protein	++
VII	Steroids	++
VIII	Anthraquinones	++
IX	Tannins	+
X	Terpenoids	+++
XI	Cardiac glycosides	++

Source: Field Data (2024)

− not present;
+ present in small concentration;
++ present in moderately high concentration;
+++ present in high concentration;

3.2. Discussion

The plant kingdom has become a very useful source for man in his desire and search for beneficial products for nutritional and medicinal purposes (Edagha and Aquaisua 2017; Chaachouay and Zidane, 2024). The medicinal value of plants lies in their bioactive phytochemical constituents that produce definite physiological action in the human body (Agidew, 2022). The therapeutic potentials of plants have been linked with their various bioactive properties (Agidew, 2022; Dubale et al., 2023). Studies have shown that medicinal plants can enhance biological activities due to the presence of phytonutrients, which serve as herbal therapies (Dasofunjo et al., 2020).

The Phytochemical Analysis of the plant fractions revealed the presence of tannins, flavonoids, saponins, terpenoids, Cardiac glycosides, and alkaloids, which are suggested to exhibit pharmacological activities. Studies have shown that G. latifolium has a high content of different types of alkaloids, flavonoids, phenol, lignan, terpenes, sterol, saponin, carotenoids (Imoh and Uchegbu 2015, Balogun et al., 2016), which can be of pharmacological importance.

Studies have shown that Flavonoids are known to inhibit the formation of plaques in arteries and so prevent atherosclerosis, hypertension, and other cardiovascular diseases (Panche et al., 2016, Wang et al., 2020). They also contain important antioxidants that help mop up reactive oxygen species known to be responsible for conditions that cause cancers, diabetes, inflammatory and neurodegenerative diseases (Lukong et al., 2018)

Therefore, these phytochemical substances are recognized to play essential roles in medicinal plants' bioactivity, and they may be a source of their therapeutic benefits (Balogun et al 2016).

4. Conclusion

Evidence from this study has revealed that Gongronema latifolium leaves have a good number of phytochemical constituents that qualify it as a medicinal plant. The four different fractions, ethyl acetate, dichloromethane, n-hexane and methanol of gongronema latifolium leaf extract contain moderately high and very high phytochemicals, which can be used in the treatment of different illnesses by practitioners of traditional medicine. However, further studies are needed to ascertain their biological and pharmacological activity.

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