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Phytochemical screening and anti-tussive studies of aqueous and alcoholic extracts of *Aneilema aequinoctiale*

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ABSTRACT

This work investigates the phytochemical composition and anti-tussive effect of aqueous and alcoholic extract of *Aneilema aequinoctiale*. The phytochemical screening of the *Aneilema aequinoctiale* showed quantitatively the presence of alkaloids, saponin, steroid, cardiac glycosides, flavonoids and tannins. The possible anti-tussive effects of the ethanolic and aqueous extract of the leaves, root, stem and whole plant of *Aneilema aequinoctitale* on *Streptococcus pyogenes, Klebsiella pheumonia, Cryphtococcus neoformans* and *Streptococcus pneumonia* were determined and compared. The extracts of the whole plant exhibited the widest inhibition zones followed by extracts of the leaves and stem, while the root extracts showed the least inhibition zones. The results are discussed with reference to the nutritional and medicinal values of the plant *Aneilema aequinoctiale.*

Introduction

The use of plant as a source of drug for treating different ailments is as old as mankind with recorded practices dating back to 3000 years. Medicinal plants form the basis of medicinal treatment in many developing countries and the research on this plant's extracts may yield useful information, which could lead to the discovery of potentially important pharmaceutical products. Many plants have been used traditionally for years for healing on trial and error basis and some of them do give positive result but there may not be sufficient scientific data to confirm their efficacy.

The compilation of useful drugs derived from medicinal plants is impressive; these include; heart drugs, analgesics, anesthetics, antibiotics, anti-cancer and anti-parasitic compounds, anti-inflammatory drugs, oral contraceptic hormones, as well as laxative diuretics [1].

A medicinal plant is any plant in which one or more of its organs contains substances that can be used for therapeutic purposes or which are precursors for synthesis of useful drugs [2]. The choice of special plant materials for treatment of ailment by our ancestors, was not based on knowledge of the chemical constituents, but was probably based on their observation of the effect on certain things in the environment. The presence of bioactive agents in various plants has been associated with their potency as drugs. A good example is the steroidal glycosides found in the leaves of Foxglove (*Digitalis purpurea L*) which give the plant its cardiotonic activity [2].

However, pharmaceutically active principles of medicinal plants are localized in various parts of the plant and environmental conditions may influence the quality and quantity of the final product. Hence, pharmacognostic descriptions include instructions on the part of the plant to be used and the preferred geographical regions of the plants. Studies have shown that the extracts from the bark, leaves and roots of the plant locally known as "Dongoyaro" *(Azadirachta indica*) (neem) is used mostly for the treatment of malaria, *Salacia pyriformes* (Mbang-enang in Ibibio) is used in the treatment of malaria and jaundice, while *Vernonia amygdalina* (bitter leaf) is used for skin disease as well as in managing diabetes *milletus*. *Rauvollia vomitoria* is effective in the treatment of hypertension and mental disorders [3]. Eseyin *et al.*, [4] found out that the leaves and stem of *Telfairia occidentalis* (fluted pumpkin) could be used in managing diabetes. *Bryophylium pinatum* known locally as ndodop in Ibibio is used externally and internally for the cure of catarrh, cough and asthma. It is also used for the treatment of epilepsy; this may be attributed to its high saponins content [5]

Other plants of important medicinal use are *Elephantopus scaber* is used against inflammation of uterus and ovaries as well as an anti-tussive concoction. Odoemena *et al.*, [6] reported the anti-bacterial activity of the root extract of *Telfairia occidentalis* (fluted pumpkin) against *Staphylococcus aureus, Streptococcus pyogents, Shigella dysenteriae* and *Klebsiella pneumonia.*

Many species of leguminous plants are known to possess anti-microbial properties for example lathyrus and lotus species are known to inhibit growth of *Chorella vulgaris, E. coli, S. aureus* and *Candida* species [7]. Leaf extract from *Cassia alata* was found to inhibit the growth of dermatophytes cultured in Sabouraud Dextros Agar (SDA) [8].

Scientists in the world over resolved in proving the efficacy of the plant extracts against diseases acclaimed by the herbalist. In a similar manner *Aneilema aequinoctiale* known as Ekpa-Ekpa Ikpaha by Ibibio people [9]. This research work entails determination of phytochemical properties of *Aneilema aequinoctiale* and anti-tussive effect of its aqueous and alcoholic extracts. Antimicrobial efficacy of the plant extracts will be studied using some cough causing organisms such as: *Klebsiella pneumonia, Streptococcus pyogenes, Streptococcal pheumonia* and *Cryptococcus neoformans.*

Botany of *Aneilema aequinoctiale*

Aneilema aequinoctiale is a rainforest plant that is spread across Africa. It belongs to the plant family of commelinaceae and of genus *Aneilema* R. Br. And the species is *aequinoctiale.* The common English names are "day flower" or "white and

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blue" flower. The Yoruba's refer to it as "goolofun-fun" "godobo odo (meaning 'stream') or "Ito-ipere". In Edo State, it is called "Ohiovbu", while the Igbo's know it as "Oboogu uku" or "Ode oa Ogu Oji". The Ibibio's of Akwa Ibom State know *Aneilema aequinoctiale* as "ekpaekpa ikpaha" which mean "even if all die, it will not die" [9].

Moreso, the commelinaceae are herbs comprising about 50 genera and 700 species that are often somewhat succulent and frequently have cymose inflorescences sometimes subtended by a boat-shaped spathe. The leaves are alternate, simple, parallel-veined and usually with a closed sheating base. The flowers are bisexual and actinomorphic or commonly slightly to strongly zygomorphic. The perianth is in two usually differentiated series. The calyx is unequal, distinct, deliquescent petals. The androecium typically comprises six distinct stamens but commonly three or sometimes more are reduced to staminodes, the gynoecium consist of a single simple pistil of three carpels, a single style and a superior ovary containing three or occasionally by abortion only two locales, each containing one few axile ovules. The fruit is usually a loculicidal capsule or is sometimes indehiscent. *Aneilema aequinoctiale* species has zygomorphic flowers with one petal greatly reduced or obsolete. Among the green sepals, only two stamens appear to be functional, another species like *zebrine pendula*, (wandering jew), has antinomorphic trimerous flower with 6 stamens. The inflorescence is cymose and is subtended by a boat-shaped brat. It is common for filament to be hairy in this family, also in *Dichorisandra thyrsiflora* (blue ginger). This unusual member of the family has petaloid sepals that are white on the front and purple on the back. Also, the anthers of this species open by terminal pores [10]

2.0 Material and methods

2.1 Sample Collection and Treatment

The entire plant comprising, root, steam and leaves of *Aneilema aequinoctiale* was collected from different forest areas in Nsit Ubium and Uyo Local Government Area in Akwa Ibom State, Nigeria. The plant was identified traditionally by the Christ herbalist, and a taxonomist in the Faculty of Pharmacy and Department of Botany respectively. They were treated by the methods recommended by Williamson *et al.,* [11]. The plant was separated from weeds and dirt, washed and rinsed with distilled water. It was sun-dried for 5 days, ground with pestle and mortar into coarse power and packed in an air-tight plastic container for future used.

2.2 Extraction of the Plant Sample

Five hundred grams (500g) of each of the samples was weighed into a conical flask and was extracted with 70% ethanol at room temperature (27^oC) for 72 hours with occasional shaking. Aqueous extract was obtained by extracting the weighed sample with distilled water. Fresh samples (crushed) and uncrushed dried samples were also treated as above. Each extract was filtered using a clean muslin cloth and green coloured filtrates were obtained. The filtrates were concentrated at temperature between $45 - 50^oC$ over water bath until a paste was obtained. The extracts were labeled accordingly, cooled and stored in desiccators for further analysis in the laboratory.

2.3 Phytochemical Analysis of the Extracts

The freshly prepared aqueous and ethanolic extracts of *Anielema aequinactilale* leaf, stem and root were tested for the presence of phytochemical constituents using standard procedures [12].

1) Test of Alkaloids

0.5g of the aqueous and ethanol plant extracts were each measured into three separate test tubes for the test and the colour change for each test was noted.

(a) A few drops of freshly prepared Dragendorff reagent were added to the first test tube. A pink or red precipitate was an indication of a positive test.

(b) A freshly prepared Mayer's reagent was added to the extract in the second test tube. A milky or cream colour was an indication of a positive test.

(c) A few drops of picric reagent were added to the extract in the third (3^{rd}) test tube. A white or yellow precipitate indicated a positive test.

2) Test for Steroids and Terpenes

To 0.05g of each extracts, 3.0 ml chloroform was added and filtered, 10 drops of ethanoic anhydride and 21 drops of concentrated H_2SO_4 were added to the filtrate and the colour change was observed. Pink colour at interphase was taken as positive test for terpenes, bluish green interphase was positive test for steroids.

3) Test for Saponins

(a) Frothing Test: 0.5g of each extract was shaken vigorously with distilled water in a test tube. Frothing which persisted on warming was taken as preliminary evidence for the presence of saponins.

(b) Fehlings solution was added to 0.5g of each extract and warmed. The presence of brick red precipitate confirmed the presence of saponins.

(c) 1.0 ml Na_2CO_3 was added to 0.5g of each extract and fehlings solution was also added and warmed, brown precipitate confirmed the presence of saponins.

4) Test for Cardiac Glycosides

(a) Lieberman's test was carried out by dissolving 0.5g of plant extract in 2mls of ethanoic anhydride and cooled in ice. Sulphuric acid was then carefully added. A colour change from violet to blue, then to green indicated the presence of a steroidal nucleus i.e aglycone portion of the cardiac glycoside.

(b) Salkowski test was carried out by dissolving 0.5g of the plant extracts in 2mls of chloroform, sulphuric acid was carefully added to form a lower layer. A reddish-brown colour at the interface indicated a positive test.

5) Test for Tannins

(a) The plant extract of weight 0.5g was taken in 100 ml beaker, was stirred. 10.0 ml of distilled water and 2mls of bromine water was added and stirred. For a positive test, tannin decolorized bromine water.

(b) The plant extract of weight 0.5g was dissolved in distilled water and ferric chloride reagent was added to the filtrate. A blue-black, green or blue green precipitate was taken as evidence for the presence of tannins.

6) Test for Carbohydrate

The plant extract of weight 0.2g was carefully mixed with 4 drops of Molisch's reagent and $1cm^3$ of conc. H_2SO_4 was added. A purple colour indicated a positive test for carbohydrates.

7) Test for Flavonoids

(a) Shinoda's test was carried out as follows: The plant extract was dissolved in concentrated hydrochloric acid. Few pieces of magnesium metal were added to 5mls of the extract. The formation of orange, red, crimson or magenta was taken as a positive test, for the presence of flavonoid.

(b) Ammonia test was carried out by weighing 0.2g of plant extract into a test tube.

Table 1: Summary of Phytochemical Test Result

Test	Observation	
	Ethanolic extract	Aqueous extract
1 Alkaloid Dragendorff test	+++	+
(i) Mayer's test	-	-
(ii) Picric acid test	-	-
2 Steriods	+++	++
3 Saponins		
(a) frothing test	++	+
(b) fehling's solution test	+	+
(c) Na_2CO_3 and fehling solution test	+++	+
4. Cardiac Glycosides		
(a) Lieberman's test	-	++
(b) Saikowski test	-	++
5. Tannins		
(a) 5% ferric chloride test	++	-
(b) Bromine Water test	+	-
6. Carbohydrates	+++	++
7. Flavonoids		
(a) Shinoda's test	+	+
(b) Ammonia test	+	+

KEY: +++ = High, ++ = Moderately, + = Trace, - = Absent or neglibible

Table 2: Antimicrobial Screening of *Aneilema aequinoctiale* Extract on *Streptococcus pyogenes*

	Conc.	Zones of Inhibition in mm			
Extract	(mg/ml)	Leaves	Stem	Root	Whole plant
	100	2.10±0.04	2.00±0.35	1.90±0.01	9.00±0.14
Ethanolic extract	90	1.80±0.07	1.56±0.25	1.50±0.31	6.90±0.10
	80	1.70±0.43	1.60±0.18	1.40±0.17	6.70±0.10
	70	1.50±0.06	1.30±0.14	1.20±0.14	5.00±0.07
	60	1.40±0.01	1.20±0.20	1.10±0.13	4.70±0.04
	50	1.30±0.02	1.10±0.10	1.00±0.07	4.40±0.14
	40	-	-	-	-
	30	-	-	-	-
	20	-	-	-	-
	10	-	-	-	-
Aqueous extract	100	1.85±0.08	1.70±0.10	1.60±0.02	8.20±0.14
	90	1.80±0.14	1.50±0.30	1.40±0.07	6.90±0.14
	80	1.80±0.13	1.45±0.15	1.30±0.06	7.60±0.10
	70	1.18±0.24	1.02±0.10	1.00±0.19	5.20±0.28
	60	1.00±0.12	0.80±0.20	0.60±0.20	4.40±0.20
	50	0.85±0.30	0.60±0.20	0.50±0.04	4.00±0.28
	40	0.50±0.19	0.30±0.04	0.20±0.10	2.00±0.07
	30	0.50±0.40	0.30±0.07	0.20±0.30	3.00±0.07
	20	0.50±0.30	0.30±0.14	0.20±0.02	2.00±0.07
	10	0.50±0.10	0.30±0.03	0.20±0.01	2.00±0.01

Results are mean of triplicate determinations ± SD

Table 3: Antimicrobial Screening of *Aneilema aequinoctiale* Extract on *Klebsiella pneumonia*

Extract	Conc. (mg/ml)	Zones of Inhibition in mm			
		Leaves	Stem	Root	Whole plant
Ethanolic extract	100	4.70±0.07	3.20±0.11	2.10±0.03	13.00±0.20
	90	4.60±0.04	3.00±0.20	1.60±0.07	11.20±0.07
	80	4.30±0.02	2.80±0.01	1.40±0.12	9.50±0.28
	70	4.20±0.30	2.78±0.07	1.30±0.04	10.40±0.21
	60	3.90±0.40	2.56±0.12	1.20±0.04	9.70±0.07
	50	3.80±0.28	2.50±0.04	1.20±0.11	8.50±0.01
	40	3.50±0.24	2.20±0.02	1.10±0.01	9.80±0.01
	30	3.30±0.21	2.10±0.01	1.00±0.20	7.40±0.03
	20	3.10±0.04	2.00±0.12	1.00±0.01	7.10±0.14
	10	0.60±0.02	0.30±0.11	0.10±0.10	2.00±0.11
Aqueous extract	100	3.70±0.01	1.40±0.07	0.20±0.01	7.30±0.01
	90	2.20±0.14	1.30±0.06	0.60±0.02	6.10±0.01
	80	2.00±0.07	1.20±0.07	0.60±0.11	5.80±0.28
	70	2.00±0.05	1.10±0.04	1.50±0.14	5.60±0.14
	60	1.70±0.04	1.00±0.12	0.40±0.01	4.20±0.07
	50	1.60±0.17	0.80±0.13	0.60±0.02	4.00±0.14
	40	1.70±0.01	0.70±0.11	0.50±0.14	3.90±0.01
	30	1.30±0.01	0.80±0.19	0.40±0.10	3.50±0.26
	20	1.20±0.02	0.70±0.11	0.40±0.2	3.40±0.13
	10	-	-	-	-

Results are mean of triplicate determinations ± SD

Table 4 Antimicrobial Screening of *Aneilema aequinoctiale* Extract on *streptococcus pneumonia*

Organism	Ethanolic Extract Conc. In mg/ml	Zones of Inhibition in mm			
		Leaves	Stem	Root	Whole plant
Ethanolic extract	100	7.90±0.02	5.20±0.02	2.60±0.09	18.70±0.07
	90	7.70±0.01	5.00±0.07	2.50±0.01	17.20±0.07
	80	7.30±0.04	4.70±0.11	2.30±0.01	16.30±0.14
	70	7.00±0.11	4.60±0.01	2.30±0.02	15.90±0.21
	60	6.10±0.20	4.59±0.01	2.20±0.04	15.00±0.35
	50	5.40±0.20	3.60±0.02	1.80±0.30	12.80±0.07
	40	5.30±0.21	3.52±0.05	1.70±0.01	12.60±0.01
	30	4.90±0.01	3.20±0.06	1.60±0.02	10.70±0.07
	20	4.40±0.28	2.80±0.04	1.40±0.11	10.60±0.21
	10	4.30±0.03	2.70±0.07	1.30±0.35	9.30±0.01
Aqueous extract	100	5.40±0.02	3.50±0.01	1.70±0.06	31.60±0.14
	90	4.70±0.01	3.10±0.01	1.50±0.12	11.30±0.01
	80	4.65±0.01	3.00±0.22	1.60±0.10	11.10±0.07
	70	4.50±0.17	3.00±0.03	1.50±0.01	11.00±0.35
	60	4.60±0.16	2.90±0.04	1.40±0.02	11.90±0.21
	50	4.30±0.04	2.80±0.21	1.30±0.10	10.40±0.07
	40	2.20±0.03	2.60±0.21	1.20±0.07	10.00±0.35
	30	3.70±0.03	2.40±0.01	1.10±0.06	8.20±0.02
	20	2.90±0.07	2.20±0.08	0.90±0.40	8.00±0.04
	10	2.80±0.06	2.10±0.21	0.80±0.07	7.80±0.01

Results are mean of triplicate determinations ± SD

Table 5: Antimicrobial Screening of *Aneilema aequinoctiale* Extract on *Cryptococcus neoformans*

Extract	Conc. (mg/ml)	Zones of Inhibition in mm			
		Leaves	Stem	Root	Whole plant
Ethanolic extract	100	6.40±0.30	4.10±0.41	2.00±0.41	15.50±0.10
	90	6.35±0.17	4.00±0.01	1.60±0.21	13.99±0.50
	80	5.80±0.05	3.90±0.02	1.50±0.33	13.20±0.03
	70	5.70±0.25	3.80±0.30	1.50±0.01	13.00±0.02
	60	5.60±0.25	3.40±0.10	1.20±0.21	12.20±0.01
	50	5.55±0.07	3.30±0.20	1.20±0.28	12.10±0.01
	40	5.40±0.40	3.30±0.30	1.10±0.08	11.0±0.14
	30	5.40±0.02	3.20±0.41	1.10±0.07	11.70±0.01
	20	4.20±0.01	3.00±0.11	1.00±0.01	9.20±0.02
	10	4.10±0.01	3.00±0.01	1.00±0.01	10.10±0.14
Aqueous extract	100	3.60±0.30	2.00±0.20	1.70±0.06	9.60±0.04
	90	4.40±0.14	1.90±0.14	1.00±0.02	8.30±0.01
	80	2.90±0.11	1.90±0.20	0.90±0.20	7.70±0.01
	70	2.70±0.20	1.70±0.07	0.80±0.01	7.20±0.04
	60	2.60±0.40	1.60±0.01	0.80±0.10	7.00±0.35
	50	2.50±0.01	1.60±0.21	0.80±0.10	5.90±0.04
	40	2.30±0.01	1.40±0.01	0.70±0.13	6.40±0.28
	30	2.30±0.03	1.30±0.31	0.70±0.12	6.30±0.01
	20	2.00±0.40	1.30±0.30	0.60±0.40	5.90±0.45
	10	-	-	-	-

Results are mean of triplicate determinations ± SD

5mls of ethyl acetate was added and heated. It was cooled and filtered; 4.0ml of the filtrate was shaken with 1.0ml of dilute ammonia solution. The colour changed was observed.

2.4 Anti-microbial Evaluation of *Anielema aequinoctiale* Extract

Nutrient agar of 2.8g was weighed and dissolved in 100ml of distilled water and allowed to stand for 20 minutes, then autoclaved at 121^{o}C for 15 minutes at 10 psi. On cooling to 45^{o}C; the medium was dispensed into petri-dishes. All the plates were allowed to set on the bench. The test organisms *Kliebsiella pneumonia, Streptococcus pyogenes, Cryptococcus neofromans,* and *Streptococcus pneumonia* were inoculated into the nutrient agar plates, and labeled appropriately. Sterile No. 1 Whatman filter paper disc of 0.5mm in diameter were impregnated separately with the ethanol extract and aqueous extracts of *Aneilema aequinoctiale.* Extracts of variable conc. ranging from 10% to 100% were tested. The disc was allowed to absorb the plant extract, and were aseptically removed and placed on the surface of the inoculated plates in such a way that the test organisms were exposed to the extracts of different concentrations. The plates were labeled accordingly and incubated at 37^{o}C for 24 hours using Gallen Kamp inculator.

The zones of inhibition induced by the plant extract of different concentrations were measured in mm. An inhibition zone diameter of 10mm and above indicated the susceptibility of the organism to the antimicrobial agent [12].

4.0 Result and discussion

4.1 Phytochemical Screening Test Results

Table 1 present the result of the photochemical screening of the alcoholic and aqueous extract of *Aneilema aequinoctiale.* The result revealed the presence of some classes of chemical compounds in the plant.

Alkaloids

Dragendorf test showed that alkaloids were highly present in the ethanolic extract while it occurs in trace amount in the aqueous extract, whereas Mayer's test and picric acid test in both ethanolic and aqueous extract did not show the presence of alkaloids. It may be elaborated as metabolic end product, as reservoir for protein synthesis or protective devices to discourage animal or microbial attack [13]. This probably explains why the plant could be used as an anti-tussive agent.

Steroids

Steroid level in the ethanolic extract is high while it is moderately present in aqueous extract. This may be as a result of the less polar organic nature of ethanol when compared with water and as such, favours extraction of steroids more than water. A steroid is a lipid characterized by the presence of a hydrogenated cyclopentanophenanthrene ring system. Steroids are mainly made in living tissues from the triterpenoid lanosterol.

Saponins

The amount of saponins found in the plant extracts was high. The ethanolic extract showed high content of saponins while the aqueous extract showed moderate content of saponins.

Saponins are generally known for their antibacterial and antifungal properties (Odebiyi and Sofowora, 1978) and are partly responsible for the therapeutic value of the plant in control of cough.

Cardiac Glycosides

Cardiac glycosides were absence in the ethanolic extracts while aqueous extracts showed moderate cardiac glycosides. The detection of cardiac glycosides in the plants revealed that it could be used in the production of antibiotic drugs. It can be used as anti-tumour agent; it helps in strengthening the contraction of heart muscles. It can be used for treatment of asthma [14].

Tannins

The level of tannin in ethanolic extract was moderate but negligible in the aqueous extract. The presence of tannins is of interest as they are known to posses anti-fungicidal properties and confer protection against parasitic infection [15].

Flavonoids

The quantity of flavonoids found in both ethanolic and aqueous plant extract was at trace level. Flavonoids are compounds distributed in nature as pigment in flowers and fruits. Some medicinal uses of flavonoids include hypercholestrolamia, hyperglyceamia, antioxidant, anti-cancer, anti-inflammatory, weight loss, blood cleanser [16].

4.2 Anti-Microbial Screening

Table 2 - 5 present the result of anti-microbial potency of the whole plant and different parts of *Aneilema aequinoctiale* the above results showed that ethanolic extracts of *Aneilema aequinictiale* exhibited a higher antimicrobial potency against the four test organisms than the aqueous extracts. However, the result also shows that the concentration of the plant extracts influence the diameter of the zone of inhibition. It follows therefore that the higher the concentration of the extract the more efficacious is the antimicrobial activity of the plant (*Aneilema aequinoctiale*) extract.

Different parts of the plants exhibited different levels of antimicrobial activity. The whole plant recorded the largest zone of inhibition. When compared with the leaf, stem, and the roots of the plant (i.e. whole plant >leaves > stem > root). Also the ethanolic extract showed the largest inhibition zone of (18.7±0.07 – 9.30±0.10) mm with *Streptococcus pneumonia*, with *Cryptococcus neoformane* (15.50±0. 10 – 10.10±0.14) mm, with *Klebisella pneumonia* (13.00±0.20 – 2.00± 0.11) mm, and with *Streptococcus pyogenes* (9.00.± 0.14 – 0.00) mm, respectively at 100mg/ml of extract. The least inhibition zones were observed in aqueous extract tested against *Klebsiella pneumonia* (7.30±0.01 – 0.00mm), *Streptococcus pyogenes* (8.20 ±0.14 –2.00±0. 14–2.00±0.01mm), *Crypotococcus neoformans* (9.60 ± 0.04 – 0.00mm) and *streptococcus pneumonia* (13.60 ± 0.14 – 7.80 ± 0.01mm).

t-test was used for the comparison of ethanolic and Aqueous analysis of *Aneilema aequinoctiale* extract on Organism. When t_{cal} is greater than $t_{critical}$, (2.976), it means that the result varies significantly. The result obtained vary significantly at 95% confidence interval, 4 degree of freedom, except the result obtained for the extract on *Streptococcus pyogenes*, at 80g/ml for leaves extract and 60g/ml for stem extract.

Conclusion

The phytochemical composition analysis of *Aneilema aequinoctiale* extract has shown that alkaloids, steroids, saponins and tannins is present in a high amount in ethanolic extract and moderate in aqueous extract while cardiac glycosides appears moderate in aqueous extract and negligible in ethanolic extract, this revealed its major contribution in the anti-tussive effect of the plant and other medicinal characteristics.

The antimicrobial screening showed that the plant extracts had inhibitory effect on the microorganisms used, (that is *Streptococcus pyogenes, Klebsiella pneumonia, Streptococcus pneumonia* and *Cryptococcus neoformans*) thus *Aneilema aequinoctiale* may be efficacious in the alleviation and treatment of cough. The findings revealed that the concentration of the plant extract is very vital in inhibiting the activity of the cough causing organisms.

Anti-tussive effect of its aqueous and alcoholic extracts was aimed at verifying or ascertaining the traditional belief about the plant in the treatment of cough. The results obtained from this research showed that the plant (*Aneilema aequinoctiale*) is rich in bioactive compounds such as flavonoids, cardiac glycosides, terpenes, saponins and tannins, and could be a good source of raw materials for pharmaceutical industry.

Finally, the results obtained in this work have justified to some extent the use of *Aneilema aequinoctiale* by the local people and herbalist for the treatment of cough.

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