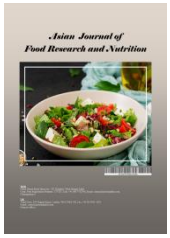

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Antibacterial Effect of Awolowo Weed (*Chromolenaodorata*) Extract on *Salmonella typhii* Isolated from Typhoid Patients

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Authors' contributions

This work was carried out in collaboration among all authors. Author ECF designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Authors NNE and OOJ managed the analyses of the study. Authors MOA and AECE managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

In this study, it was determined what phytochemical components and antibacterial properties *Chromolena dorata* leaves had against Salmonella species that had been isolated from typhoid patients. The antibacterial test of the leaf extracts against Salmonella Typhi was conducted after phytochemical screening. *Chromolena odorata* samples were extracted using methanol, ethanol, and water. The extracts were then subjected to normal phytochemical screening procedures. The

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agar well diffusion technique was used to test the extracts' antibacterial activity against *Salmonella Typhi* at doses of 50, 100, 200, 400, and 500 mg/mL. Results revealed seven secondary plant metabolites, including cardiac glycosides, steroid, alkaloids, flavonoids, terpenoids, tannins, and saponins. When used against *Salmonella typhi*, the ethanol and aqueous extracts shown antibacterial activity with a zone width of inhibition of 14 mm and 15mm respecively at a dosage of 50 mg/mL. The zone width of inhibition for the ethanol extracts was 23 mm at 250 mg/mL and 27 mm at 500 mg/mL, respectively. At 500 mg/mL, three extracts showed the greatest zone width of inhibition, but the ethanol extract and the aqueous extract showed the least inhibition at 100 mg/mL and 50 mg/mL, respectively. The antibacterial activity of the methanol, ethanol, and aqueous extracts against *Salmonella typhi* were statistically significant at (P.05). For methanol, ethanol, aqueous extracts, and amoxicillin (control), the lowest inhibitory concentration of the extracts was 250, 200, and 100 mg/mL, respectively. As a result, *Chromolena odorata* may be used for research purposes, and plant extracts work synergistically with other therapeutic plants.

Keywords: Antibacterial effects; Chromolena odorata leaves extract; Salmonella typhii.

1. INTRODUCTION

The threat of escalating antimicrobial resistance has constrained and decreased the efficacy of the majority of therapeutic antibiotics, making infectious diseases (IDs) one of the ten most lethal diseases in the world [1]. Antibiotic overuse results in higher rates of antimicrobial resistance, which leads to more fatalities from this condition worldwide (Zachariades et al., 2021). A sizable genus of gram-negative bacteria belonging to the Enterobacteriaceae family, Salmonella includes more than 2300 serotypes, is highly adapted to grow in both humans and animals, causes a wide range of diseases, and may also be linked to localised infections and/or bacteremia [2]. Members of the genus Salmonella are responsible for the human intestinal illness known as salmonellosis. Salmonella are pervasive diseases that may spread outside of living hosts and are found in people, cattle, wild animals, reptiles, birds, and insects. One of the most prevalent cases of an intestinal illness that is transferred from animals to people appears to be salmonella infection. Food items including meat, dairy, and eggs as well as direct oral contact between animals and people are both ways that the disease is spread [3]. According to Brooks et al. [4], salmonella contamination of food goods can considerably lower consumer demand and impact producer revenues. Since bacterial resistance to single-target antibiotics develops too quickly to be sustainable and resistance to new drugs frequently arises even before they are commercially available, a multi-target drug approach may be a viable solution to the Anti-Microbial Resistance issues [5].

As they battle against predators like bacteria, fungi, viruses, insects, and herbivores from their germination to maturity stages and go through difficult defence mechanisms for survival, naturally occurring chemicals originating from plants have the capacity to multi-target [6]. Therefore, it is practical and economical to investigate possible antibacterial compounds from plant sources rather than creating new antibiotics. In addition to being costly and ineffective, synthetic medications frequently have problems with adulterations and unwanted effects. Customers are particularly worried about the product's pathogenicity and high fatality rate. Therefore, as technology develops, scientists are pushed to develop fresh concepts for unique and alternative medications to replace the use of microbially resistant ones [7].

Euphatorium odoratum (L.) or *Chromolaena odorata* (L.) are both members of the Asteraceae family. The most infamous perennial scrambling weed in the Asteraceae family, *Chromolaena odorata* has shown to be a serious ecological and economic burden in many tropical and subtropical parts of the world [8]. One of the deadliest invasive alien plant species in the humid and semi-humid tropics of the world, the weed is also known as awolowo weed [9]. It typically invades fallows or freshly cleared ground and is frequently shaded out after forest trees and shrubs are completely established. It is a plant of secondary succession.

Awolowo weed is usually a hindrance to the growth of other plants, according to research done by Aliyu [10]. It is a traditional medicinal herb that is well-known for its capacity to treat wounds. Particularly, the various components of this plant have been utilised to treat skin infections, burns, and wounds. It has also been demonstrated to have antioxidant, anti-

hepatotoxic, anti-inflammatory, anticancer, and anti-diabetic effects. Alkaloids, flavonoids, flavanone, essential oils, phenolics, saponins, tannins, and terpenoids are some of its phytochemical constituents. This plant also contains significant amounts of eupolin, chromomoric acid, quercetagetin, and quercetin all of which contribute to its remedial properties [11]. Ash (11%), crude fat (11%), fibre (15%), moisture (15%), crude protein (18%), and carbohydrate (31%) were all present in the dried leaf of C. odorata [12]. Its active phytochemical substances are as follows: flavonoid aglycones (flavanones, flavonols, flavones) including acacetin, chalcones, eupatilin, luteolin, naringenin, kaempferol, quercetin, quercetagetin, and sinensetin, terpenes and terpenoids; essential oils; alkaloids including pyrrolizidine; saponins and tannins; phenolic acids including ferulic acid, protocatechuic acid; phytoprostane compound including chromomoric acid [13].

Basic phytochemical screening is doing straightforward chemical tests to find alkaloids, tannins, saponins, flavonoids, digitalis glycosides, and other compounds in a plant extract. In addition to being costly and ineffective, synthetic medications frequently have problems with adulterations and unwanted effects. Customers are particularly worried about the product's pathogenicity and high fatality rate. Therefore, as technology develops, scientists are pushed to develop fresh concepts for unique and alternative medications to replace the use of microbially resistant ones [7]. The study's importance lies in its ability to provide pertinent advice for the advancement and successful implementation of *Chromolaena odorata*. The success of this effort will encourage the development of several novel medications with different antibacterial efficacies. The purpose of the study is to identify through phytochemical screening the components of *Chromolaena odorata* extract that are active against salmonella spp. that have been isolated from typhoid patients. This requires collecting 500g of the plant, preparing and collecting the extract using ethanol and water, and identifying the antimicrobial properties against salmonella spp isolated from typhoid patient.

2. MATERIALS AND METHODS

2.1 Sample Collection

Samples of *Chromolena odorata* were gathered in Awka, Anambra state. Whereas the fresh leaves were chopped into smaller sizes using a knife and tray after drying for three days at room temperature and without direct sunlight, the sliced leaves were then further ground into powder using an electric grinder. Using a computerised weighing scale, the weight of the powdered sample was calculated. The laboratory and other resources were procured from the Alpha Research Laboratory in Awka, Anambra state, for the actual work.

2.2 Methods of Extraction

Extract of *Chromolena Odorata* in hexaneThe 100g of plant material that was gathered was air dried, ground up, and then macerated with N-hexane. After standing for 48 hours, it was filtered. The filtrate was dried in a rotary evaporator at 55°C while being evaporated under decreased pressure.

2.3 Ethyl acetate Extract of *ChromolenaOdorata*

Chromolena odorata's plant residue (100 g) was dried before being utilised for ethanol extraction. After being re-soaked in 5 L of ethanol for 48 hours, it was filtered. The filtrate was dried in a rotary evaporator at 55°C while being evaporated under decreased pressure.

2.4 Extract of *Chromolena Odorata* in Ethanol

100 g of freshly powdered *Chromolena Odorata* was steeped in 2.5 L of ethanol for 48 hours before being filtered. Additionally, the filtrate was dried with a rotary evaporator at 55°C while being evaporated under decreased pressure.

2.5 Initial Plant Chemical Screening

According to [14], the extracts will go through a preliminary chemical screening to see if any active phytochemical ingredients are present or not.

2.5.1 Analyse for alkaloids

The extracts were filtered after being treated with weak (10%) hydrochloric acid. Multiple alkaloidal reagents were used to treat the filtrates.

The Mayer test Potassium mercuric iodide, Mayer's reagent, was used to treat the extracts. All extracts include alkaloids because of their cream-colored appearance.

Dragendorff's reagent (potassium bismuth iodide) was used to treat the extracts, and the

formation of a reddish brown precipitate revealed the presence of alkaloid in all of the extracts.

Hager's test: After the extracts were treated with picric acid and the Hager's reagent, a yellow precipitate formed, indicating the presence of alkaloids in all of the extracts.

Wagner's test: After the extracts were treated with the Wagner's reagent (iodine solution), brown precipitate formed, indicating that all of the extracts contained alkaloids.

2.6 Cardiac Glycosides Test

The Keller-Killani test shows that all extracts contain glycosides when a small amount of the extracts are dissolved in glacial acetic acid, a few drops of ferric chloride solution are added, and then concentrated sulfuric acid is added. This procedure results in the formation of a red ring at the intersection of the two liquids.

2.7 Flavonoids Test

Test by Shinoda The extracts were dissolved in alcohol, and after that, a piece of magnesium and concentrated hydrochloric acid were heated and added drop by drop. The presence of flavonoids may be seen by the magenta colour of all extracts.

Test for ferric chloride: A few drops of neutral ferric chloride were added to the extracts. All of the extracts had a blackish red coloration.

2.8 Saponins Test

Foam test: The extracts were shaken vigorously in a graduated cylinder for 15 minutes after being diluted to 20 ml with distilled water. The development of foam in the test tube's upper portion shows that saponins are present in all extracts.

2.8.1Check for steroids in 2.7

Salkowski's response Chloroform and concentrated H2SO4 were each added to 2 ml of extract. a good shake. Acid layer fluoresced greenish yellow whereas the chloroform layer was red.

According to the Liebermann-Burchard test, none of the extracts contained any steroid molecules when they were exposed to concentrated sulphuric acid, a few drops of glacial acetic acid, and acetic anhydride.

2.9 Tannins Test

Lead acetate solution: After the extracts were exposed to a 10% lead acetate solution, a white precipitate formed, indicating that tannins were present in all of the extracts.

2.10 Phenol Testing

Test for ferric chloride: 1ml of the test sample's diluted aqueous solution was mixed with 2 drops of a neutral ferric chloride solution. Phenolic compounds are visible as a greenish purple colour.

2.11 Sample Collection

Patients with typhoid infections were given a single blood culture. Ten blood samples in total were taken. The Brain Heart Infusion Broth Agar (Oxoid, England) was incubated at 37°C for 3 days after receiving 2 ml of venous blood in an aseptic fashion and inoculating it onto 18 ml of the medium (Naidoo, et al., 2011).

2.12 Identification

Before serologic testing was carried out, the isolates were screened using Kliger Iron Agar Agar (Oxoid, England), Simmon Iron Medium Agar Agar (Oxoid, England), Ureabase Agar (Lab M, England), and Simmons citrate agar (Lab M, England) (Cheesbrough, 2020).

2.13 Serological Evaluation

Using the Well colex colour Salmonella test kit, serologic identification of Salmonella species was carried out. A suspension tube containing 200 l of sterile saline was gently emulsified with one or two probable Salmonella colonies from the culture plate. After shaking ferociously for a short period of time, re-suspended latex reagents 1 and 2 were put into separate circles on a flat reaction card while holding the container vertically. In two of the reaction circles containing latex reagents 1 and 2, respectively, around 40 l of the bacterial suspension was added and stirred. Without removing the card rotator, the card was placed on a suitable flat bed rotator and ran at 150 rpm for 2 minutes. Then, it was switched off and checked for agglutination. controls that are positive using the reagents for positive controls (green, blue, and and red control) were carried out along side with the latex reagent 1 and 2 respectively without the inoculums. Results were interpreted according to the manufacturer guidelines for usage of the kit (Cheesbrough, 2020).

2.14 Disc Preparation

Using a perforator, a disc with a 6mm diameter was cut from whatman filter paper. The discs were sterilised in a hot air oven for two hours at 1600 C using a bijou bottle. According to (Cheesbrough, 2020), it was taken out and allowed to cool before being used once more.

2.15 Making an Antibiotic Disc

Amoxillin was individually dissolved in 2 ml of water at a dose of 250 mg each.To ensure that the medicine would adhere to the paper discs, 1 ml of the various stock solutions was diluted in 1 ml of water before being put to 10 of them in a glass petri dish. This drying process took three hours at 400°C in the oven (Cheesbrough, 2020).

2.16 Making Discs for Ethanolic Extract

1 gramme of an ethanol-based leaf extract was combined with 2 ml of water. In a test tube, the mixture was correctly prepared, and 1ml of it was then added to a glass petri dish that had 10 paper discs. To enable the plant extract to adhere to the paper disc for future usage, the disc was dried in the oven (Cheesbrough, 2020).

Testing for Susceptibility Using Plant Extract SDA and nutrient agar plates were infected with the appropriate test organisms using a syringe and needle, and then each test organism was disseminated using a glass spreader. Each test organism's plates in aqueous and ethanol extract were done in triplicate. In an incubator, the plates were allowed to dry for 15 minutes. Using flamed but cooled forceps, the previously indicated dry water and ethanol discs were placed onto the surface of the infected agar plates. They weren't too close together so that the ensuing zone of inhibition wouldn't overlap. To see the zone of growth inhibition caused by the extract, the plates were incubated at 37°C for 18–24 hours (Cheesbrough, 2020).

2.17 Antibiotic Susceptibility Test

All isolates underwent an antibiotic sensitivity test utilising Kirby Bauer's paper disc diffusion methods utilised amoxicillin after the agar plate inoculation of test organisms. The plates were given 15 minutes in an incubator to dry. Using sterile forceps, the antibiotic discs were applied to the agar. To avoid the zones of inhibition on each disc overlapping, they were separated from one another. To detect the antibiotics' zone of inhibition, the plate containing the antibiotic disc was then incubated at 37°C for two hours (Cheesbrough, 2020).

3. RESULTS

3.1 Phytochemical Screening of Extracts

The result of the phytochemical screening revealed the following metabolites as present in the methanol, ethanol and aqueous extracts.The phytochemical studies of the ethanol extract revealed the presence of four (4) out of the seven (7) phytochemicals with presence of alkaloids, saponins, phenol, tannins, compounds while flavonoids, steroid and glycoside was absent. The ethyl acetate extract showed the presence of five phytochemicals (glycoside, steroid, tannin, saponin and phenol) while alkaloid and flavonoid were absent. Finally the result also showed that methanol extract reveled the presence of five (5) out of the seven (7) phytochemicals with presence of glycosides, saponins, phenol, tannins, steroid compounds while flavonoids, and alkaloids, were absent.

Table 1. Qualitative phytochemical composition of different extracts of *C. pentandra*

Metabolites	Methanol	Ethanol	Aqueous
Saponins	+	+	+
Tannins	+	+	+
Reducing sugars	+	-	-
Carbonyl or aldehyde	+	+	-
Phlobatannins	+	-	+
Steroids	-	-	-
Flavonoids	+	-	+
Terpenoids (Salkowski method)	+	+	+
Alkaloids	+	+	+
Cardiac glycoside	+	+	-
Anthroquinones	+	+	-

Key: +++ = Present in high concentration; ++ = Present in moderate concentration; + = Slightly or sparingly present; - = Absent.

3.2 Isolation of *Salmonella typhii*

The growth pattern of the blood Brain Heart Infusion Broth Agar (Oxoid,England), which was incubated at 37^{o}C for 3days are shown below.

Table 2. Bacteria growth pattern and bacteria count of isolate

Sample	Nature of growth	Total bacterial Count (Cfu/ml)
Sample 1	Moderate	0.91 x10^4
Sample 2	Moderate	0.75x10^4
Sample 3	Heavy	3.25 x10^4
Sample 4	Scanty	1.65 x10^4
Sample 5	Moderate	3.25 x10^4
Sample 6	Moderate	0.96 x10^4
Sample 7	Moderate	1.27 x10^4
Sample 8	Moderate	0.91 x10^4
Sample 9	Scanty	0.75x10^4
Sample 10	Moderate	1.27 x10^4

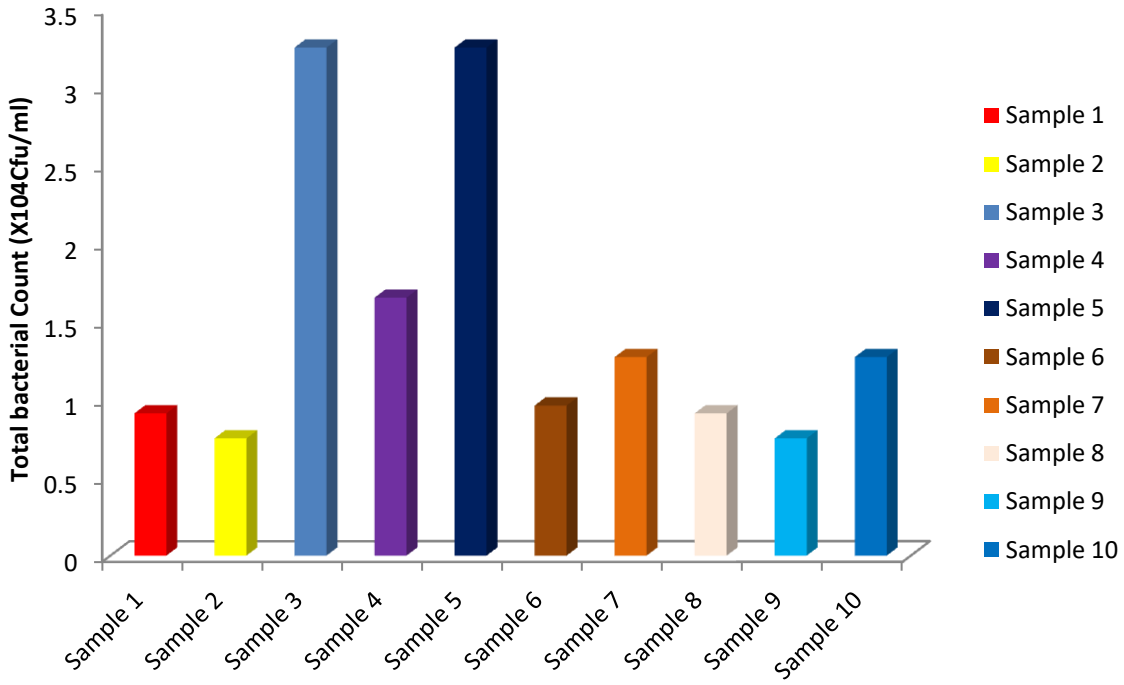

Fig. 1. Bacteria growth pattern and bacteria count of isolate

Table 3. Morphological Characteristics

Isolate	Colour	Form	Elevation	Cell arrangement	Gram reaction	Suspected organism
A	Grennish	Irregular	Flat	Small rods	Positive	*Citrobacterspp*
B	Yellow	Circular	Flat	Large rods	Negative	*Proteus spp*
C	Cream	Circular	Flat	Short rods	Negative	*Klebsiellaspp*
D	pink	Irregular	flat	Short rods	Negative	*Esherichia Coli*
E	Cream	Irregular	Flat	Coccus	Positive	*Shigellaspp*
F	Greenish	Circular	Flat	Large rods	Negative	*salmonella sp*

Table 4. Biochemical characteristics

Isolate	Motility	Coagulase	Catalase	Probable organism
A	-VE	-VE	+VE	*Citrobacterspp*
B	+VE	-VE	+VE	*Proteus spp*
C	-VE	-VE	-VE	*Klebsiellaspp*
D	-VE	+VE	-VE	*salmonella sp*
E	+VE	-VE	-VE	*Shigellaspp*
F	+VE	-VE	-VE	*Salmonella spp*

3.3 Antibacterial Activity

The zone of inhibition for the various extracts at different concentrations are shown in Table 4. The highest zone of inhibition of the extracts against *Salmonella* Typhi (*S*. Typhi) were 26 mm, 27 mm and 27 mm at 500 mg/mL for methanol, ethanol and aqueous extracts respectively and 29 mm for the control (amoxicillin). This was followed by the zone of inhibition of 24 mm, 23 mm and 25 mm at 250 mg/mL for methanol, ethanol and aqueous extracts respectively and 25 mm for the control at 250 mg/mL.

3.4 Minimum Inhibitory Concentration (MIC)

The results of the MIC of the different extracts against *Salmonella.typhi* L for methanol, ethanol, aqueous extracts and amoxicillin (control) respectively.

Table 5: Zone of inhibition of the different concentrations of extracts against *S. Typhi* (mm)

Extract	Concentration (mg/mL)			
	100.00 mg/ml	200.00 mg/ml	250.00 mg/ml	500.00 mg/ml
Methanol	10.0±1.0^{a}	15.0±2.0^{a}	24.0±1.0^{a}	24.0±2.0^{b}
Ethanol	14.0±1.0^{a}	17.0±1.5^{a}	23.0±1.0^{b}	24.0±1.0^{a}
Aqueous	15.0±2.0^{b}	21.0±1.0^{a}	25.0±1.5^{a}	27.0±1.0^{a}
Amoxicillin	20.0±1.5^{a}	23.0±1.0^{a}	25.0±1.0^{b}	28.0±2.0^{b}

Values are expressed as mean±SD. Values with the same alphabets are significantly different (P≤.05)

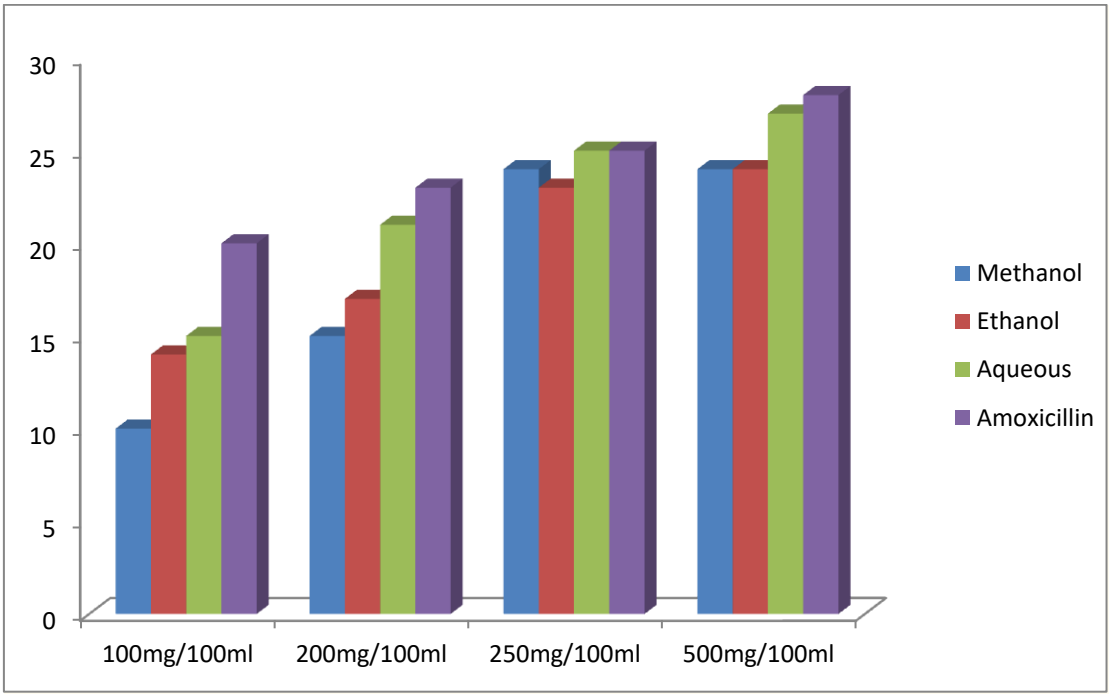

Fig. 2. Zone of inhibition of the different concentrations of extracts against *S. typhi*

Table 6. Minimum inhibitory concentration (MIC)

Microorganisms	Ethyl acetate	Ethanol extract	Methanol extract	Amoxicillin
S. typhi (mg/mL)	200.00	200.00	250.00	100.00

Table 7. Qualitative phytochemical

Metabolites	Methanol	Ethanol	Aqueous
Saponins	+	+	+
Tannins	+	+	+
Reducing sugars	+	-	-
Phlobatannins	+	-	+
Steroids	-	-	-
Flavonoids	+	-	+
Terpenoids	+	+	+
Alkaloids	+	+	+
Cardiac glycoside	+	+	-
Anthroquinones	+	+	-

Key: +++= Present in high concentration; ++ = Present in moderate concentration; + = Slightly or sparingly present; - = Absent

Table 8. Quantitative phytochemical composition

Phytochemicals	Mean value(mg/100g)
Alkaloid	8.8200
Flavonoid	1.0567
Phenol	100.5267
Saponin	34.1667
Tannin	18.3667
Steroid	.1300
Terpenoid	3.3000

**Values are mean scores ± Standard deviation of three (3) replicates*

4. DISCUSSION

Chromolaena odorata (Awolowo) leaves underwent phytochemical screening, and the results are shown in Table 1. Based on the presence of phenols, glycosides, steroids, tannin, and saponins in all of the leaves, the sample was found to be rich in phytonutrients. According to Echo and Ikenebomeh (2018), phytochemicals are bioactive, non-nutritive, naturally occurring plant components found in fruits, vegetables, and spices. Alkaloids, saponins, phenol, tannins, and compounds were all present in the ethanol extract according to the phytochemical analyses, however flavonoids, steroids, and glycosides were not present. Five phytochemicals (glycoside, steroid, tannin, saponin, and phenol) were present in the ethyl acetate extract while alkaloid and flavonoid were not. Finally, the results showed that five of the seven phytochemicals—glycosides, saponins, phenol, tannins, and steroid compounds—were present in the methanol extract, whereas flavonoids and alkaloids were not.

The presence of tannins, as shown by the results, points to this plant's potential to be a significant anti-diarrheal and anti-haemorrhagic agent (Asquith and Butler, 1986). Because of their connection to substances employed as sex hormones, steroidal chemicals are significant in medicines (Akinmoladun et al., 2017). The tannins found in fertility tree leaves may have contributed to their bitter flavour and have been shown to speed up the recovery of wounds and irritated mucous membranes (Wise et al., 2017). According to several reports, cardiac glycosides are useful for treating heart disorders and are known to reduce blood pressure. The findings of this study thus imply that spices are a great source of bioactive chemicals that may have significant socioeconomic significance (Henneberg and Stohmann, 2014).

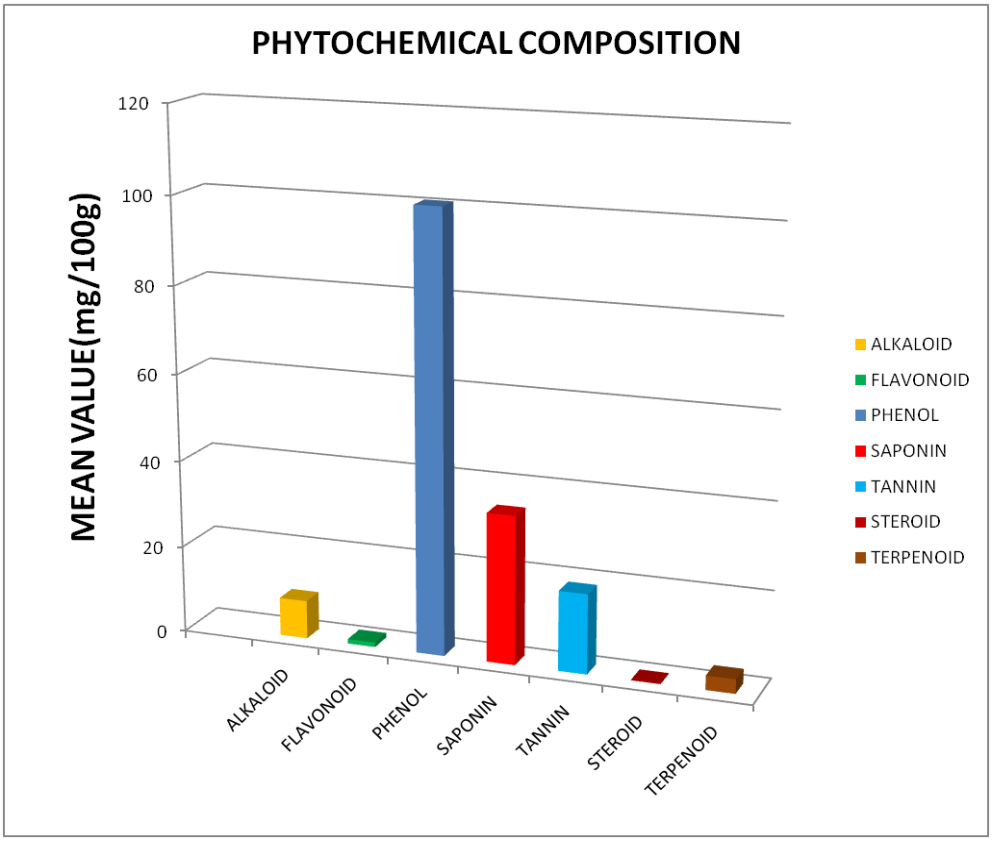

Fig. 3. Mean values of Phytochemical Composition

Alkaloids are substances that benefit plants by acting as a deterrent to pests and predators. These most likely gives these category of drugs their antibacterial properties. Many medical plants that contain alkaloids are thought to have been employed by early humans as painkillers, recreational stimulants, or in religious rites to induce trances that allowed for communion with deities or ancestors.

According to theory, saponins inhibit the development and survival of cancer cells by interacting with their cholesterol-rich membranes. Red blood cells can be precipitated and coagulated by saponins. According to Iwu [11], some qualities of saponins include the ability to create foams in aqueous solutions, hemolytic activity, cholesterol-binding abilities, and bitterness.

The majority of biological actions relating to human cell division and proliferation caused by medicinal plant saponins also have an anti-inflammatory impact (Okwu, 2011).

The antibacterial activity of the three extracts of *Chromolaena odorata* (Awolowo) leaves revealed that the ZDI of the control [(20.01.5) mm] is considerably greater (P 0.05) than the extracts' ZDI [(10.01.5) mm] at low doses (10-200 mg/mL). The ZDI of the control did not differ from those of the extracts at greater doses, though (P0.05). An organism is only deemed sensitive to a chemical agent, according to Baker and Silverton (2016), when the ZDI is either greater than or not less than 3 mm below the control, or when it is equal to the control. For methanol, ethanol, and aqueous extracts, respectively, the greatest ZDI of the extracts against S. Typhi are 26 mm, 27 mm, and 27 mm at 500 mg/mL, while the control (amoxicillin) had a ZDI of 29 mm. Following this, the ZDI of 25 mm for the control and 24 mm, 23 mm, and 25 mm at 250 mg/mL for the methanol, ethanol, and aqueous extracts, respectively. These data are deemed sensitive in this work because they are beyond the Clinical Laboratory Standard Institute's reference range (Chakraborty *et al*., 2011).

The antibacterial properties of *Chromolaena odorata* against various well-known diseases were emphasised in this study. Drug resistance is an issue that has rendered certain antibiotics

useless (Ghani and Uheshaja, 2020). Therefore, employing herbs as raw materials to promote health should be re-thought. In Nigeria, *Chromolaena odorata* is a well-known invasive weed that easily spreads and takes up residence in any open area. The ability of *Chromolaena odorata* to demonstrate antimicrobial activities in the current research work points to a potential alternative use of the weed as raw materials for the production of medicine that can be used in diseases caused by *Staphylococcus spp*., *Escherichia coli*, and *Candida albicans* (Paterson, & Zachariades, 2013).

This study demonstrates the nutritional value of awolowo leaves and the importance of using them for overall wellness. Awolowo leaves contain a variety of compounds that can scavenge free radicals, including vitamins, alkaloids, tannins, terpenoids, phenolic acids, flavonoids, and other metabolites that are essentially abundant in antioxidant activity. The fact that these plant metabolites may be genetically altered to improve their output is highly intriguing. This suggests that, in this case, purchasing synthetic medications from over-the-counter drug stores may not be essential (Okon and Amalu, 2013). To tackle the issue of *S. typhi* multi-drug resistance, it is possible to use the antibacterial characteristics of *Chromolaena Odorata* (Awolowo) Leaves extract to create new antibiotics or improve those that already exist but are quickly gaining resistance.

5. CONCLUSION

According to the findings of the phytochemical investigation, the plant's active ingredients, which are responsible for the therapeutic properties of the plant, include alkaloids, saponins, tannins, anthraquinones, flavonoids, phenols, terpenoids, and glycosides. Against *S. typhi*, the extracts showed antibacterial action. The utility of the *Chromolaena odorata* (Awolowo) Leaves in the treatment of illnesses including typhoid fever, diarrhoea, and dysentery was confirmed by the fact that each of the extracts tested exhibited a sizable zone of inhibition against the organisms tested. The indigenous use of *Chromolaena odorata* (Awolowo) Leaves as a medicinal plant in the treatment of typhoid fever has a scientific foundation thanks to the demonstration of antibacterial activity by *Chromolaena odorata* (Awolowo) Leaves extracts against *S. typhi*. The fact that the extracts' antibacterial activity increased following their purification with polar solvents is proof that the plant Awolowo bioactive components are a viable source for an effective, affordable, and widely available antityphoid medication. Therefore, more investigation is required to better define, identify, and characterise bioactive substances and their impact on *S. typhi*, including toxicological investigations. It is advised that more research be done or that toxicity tests be conducted on the bioactive components. There aren't many studies on the Awolowo leaf's capacity to cure wounds; additional study on the seed's characterisation is required. The bacteriological analysis of its herbal extract also requires more investigation.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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