Trad. Med. J., January-April 2023
Vol. 28(1), p 1-9
ISSN-p : 1410-5918 ISSN-e : 2406-9086

Submitted : 27-10-2022
Revised : 12-12-2022
Accepted : 24-01-2023

# Phytochemical Constituents, Nutritional and Antibacterial Potentials of Selected Medicinal Plants

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## ABSTRACT

Alternate medicine practices using natural plant extract may be of great importance in combating public health challenges. Medicinal plants are the bases for the development of modern drugs; accepted to treat infectious diseases globally. This study investigated the phytochemical constituents, proximate composition, and antibacterial activity of *Allium sativum* (garlic)*, Chromolaena odorata* (Siam weed), and *Talinum triangulare* (Waterleaf) due to their reported medicinal properties. The phytochemical constituents and proximate composition were analysed using standard protocols. Agar diffusion technique was used to evaluate the antibacterial activities of the plants. All three plants included innumerable quantities of phytochemicals such as alkaloids, tannins, phlobatannins, saponins, flavonoids, anthraquinones, steroids, phenol, and cardiac glycoside. Proximate analysis indicated that the moisture (9.88%), fiber (4.57%), crude fat (2.89%), crude protein (3.67%), ash (3.78%), and carbohydrate (75.21%) present in *Allium sativum*; moisture (9.26%), fiber (15.28%), crude fat (3.56%), crude protein (18.86%), ash (11.76%), and carbohydrate (41.28%) present in *Chromolaena odorata*; moisture (10.37%), fiber (16.43%), crude fat (2.23%), crude protein (11.88%), ash (13.29%), and carbohydrate (45.80%) present in *Talinum triangulare*. The crude extract of *Chromolaena odorata* had the highest zone of inhibition against *Pseudomonas aeruginosa* (12.5mm). These bioactive substances, such as phenol, alkaloids, flavonoids, saponins, and tannins, augment the therapeutic characteristics of these plants, making them a common component of traditional remedies. They may serve as primes to drug discovery and be integrated into the health care delivery system. The investigations proved these plants' medicinal potential and further demonstrated *Chromolaena odorata* and *Talinum triangulare*'s nutritional potential. They are good sources of income for individuals and bioprospecting.

**Keywords:** *Antibacterial; Clarias gariepinus; Medicinal plants; Phytochemicals; Pseudomonas aeruginosa*

## INTRODUCTION

Ever since ancient times, medicinal plants have been utilized to treat a variety of human and animal illnesses. All civilizations are reported to have traditions of using herbs to promote healing. Synthetic drugs have negative side effects, based on this and many other reports from researchers worldwide, herbal medication is increasingly gaining popularity day by day (Rout *et al*., 2012). Because they are extensively available, many therapeutic herbs have gained appeal. The invention of modern pharmaceuticals initiated with plants, and there has been a shift in public opinion toward wider acceptance of herbal treatments to treat infectious diseases globally (Ates *et al*., 2003), this is perhaps due to the over-prescription and misuse of synthetic antibiotics (Tiamiyu and Soladoye, 2015) which eventually result in the development of antibiotic resistance. An estimated 50% of the medications used in contemporary Western medicine have been modeled after plant materials (Robert *et al*., 2011). Different chemical components found in medicinal plants have specific physiological effects on both human and animal bodies. These bioactive compounds include sugars, proteins, alkaloids, phenols, tannins, flavonoids, saponins, and steroids. (Kumar *et al*., 2012). Various substances are present in the leaves, stems, roots, and bark of these plants and are produced by primary or secondary metabolism. (Vijayalakshmi and Ravindhran, 2012). The structure of these secondary metabolites varies greatly; many of them are aromatic compounds, the majority of which are phenols or their oxygen-substituted derivatives. The antioxidant qualities of medicinal plants that are high in phenolic compounds have been extensively researched. The primary source of natural antioxidants is planted phenolic

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compounds such as flavonoids, phenolic acids, and tocopherol **(**Prasad *et al*., 2012**).**

The Liliaceae family's *Allium sativum*, also known as Garlic, is legendary for both its culinary and therapeutic benefits. Garlic has proven antioxidant, antiviral, antibacterial, and antifungal properties in addition to its reputation as a healthful meal (Fratianni *et al*., 2016). Additionally, it has been shown to have anti-cancer and anti-atherosclerotic effects (Bongiorno *et al.,* 2008). *Chromolaena odorata* R. M. King and H. Rob known as Siam weed is an Asteraceae plant. In tropical Africa, Siam weed has developed a reputation as a remedy for a number of illnesses, including fever, diarrhea, toothaches, and malaria (Olajide *et al*., 2000). The plant decoction is used therapeutically as a treatment for coughs and colds or in baths to cure skin conditions. It has also been widely used as an efficient treatment for diarrhea, malaria fever, toothaches, diabetes, skin conditions, dysentery, and colitis (Akinmoladun *et al*., 2007). Waterleaf (*Talinum triangulare*), is a member of the Portulacaceae plant family. Crude protein, total lipids, essential oils, cardiac glycosides, flavonoids, and polyphenols are all abundant in the *Talinum triangulare* (Ogunnowo, & Alao-Sanni, 2010). Preliminary investigations on the phytochemistry of *T. triangulare* showed the presence of omega-3 fatty acids as well as high quantities of nutrients necessary for growth and development, including minerals (such as calcium, potassium, and magnesium), soluble fibers (such as pectin), and vitamins (such as C, tocopherols, and beta-carotene) (Airaodion *et al*., 2019). The medicinal therapy of cardiovascular disorders including stroke and obesity has been linked to waterleaf (Pavithra *et al*., 2017), and traditionally it is used as a softener of other vegetal species.

The economic situation in Nigeria is depressed just as it has been observed in many developing countries and this has reduced the purchasing power of the majority, It is now clear that local medicinal plants will become more and more important to the food, nutrition, and health security of rural residents, urban slum dwellers, and their domestic animals. Apart from these studies, works relating to the evaluation of phytochemicals of medicinal plants such as *Allium sativum, Chromolaena odorata* and *Talinum triangulare* which bioactive compounds can be useful in nutritional and therapy of humans and animals are meager in our country. Given these facts, the present work was embarked on to evaluate the status of different bioactive compounds in the dried powder of *Allium sativum* bulb and the leaves of *Chromolaena odorata* and *Talinum triangulare* to ascertain their nutritive and therapeutic potentials.

## MATERIALS AND METHODS

### *Plant Collection and Plant Powder Production*

Healthy, new foliage of Siam weed and water leaf were garnered from the wild, and bulbs of garlic were sourced from native markets in the Southwestern part of Nigeria. The plants were documented by the technical staff of the Department of the Botany University of Ibadan. The voucher example was placed with the herbarium of the University of Ibadan for future reference. The foliage was rinsed thoroughly with clean water. The foliage was mechanically milled into powder after being air dried for a few weeks in the shade at 25 ±2 °C on a side bench in the lab. Garden-fresh garlic rhizomes were peeled, sliced into smaller pieces, and then oven-dried at 70° C until a constant weight was obtained. Using an electric food processor, the dried garlic was ground into a powder. For later use, the plant powder was stored in sealed plastic containers with the proper labels.

### *Phytochemical Analysis*

Each powdered plant component's extract was utilized to conduct phytochemical analyses and determine its contents, following standard methods as outlined by (Sofowora, 1993), and the outcomes are shown in Table 1.

### *Solvent extraction of sample*:

About 20 g powder sample of each medicinal plant was extracted with methanol for 16 hours in the Soxhlet Unit. The extract was then dried on the hot plate at 30°– 40°C till the solvent got evaporated and the dried powder was stored in the fridge at four-degree centigrade for further analysis.

### *Test for Taninns*:

1 ml of plant extract was boiled in 20 ml of water, filtered, and then reconstituted. The filtrate was mixed with 0.1% ferric chloride. Observed blue-black or green coloring proved tannin was present.

### *Assessment for Phlobatannins:*

Aqueous hydrochloric acid (1%) was used to boil 2 ml of plant extract. The existence of phlobatannins was confirmed by the deposition of a red precipitate.

***Test for Saponin*:**

5 ml of the extract was boiled in a water bath with 20 ml of distilled water before being filtered. Filtrate (10 ml) and distilled water (5 ml) were combined, and after a vigorous shake, a stable persistent foam was allowed to form. Then, the foam was vigorously shaken while three drops of olive oil were added. The development of an emulsion revealed the existence of saponin.

***Anthraquinone testing***

In 5 ml of concentrated sulfuric acid, 2.5 g of extract was dissolved, then filtered. To dissolve the filtrate, 2.5 ml of chloroform was used. 0.5 ml of 10% diluted ammonia was then pipetted into the chloroform layer in a tube. Anthraquinones are evident when pink, red, or violet color develops.

***Test of Flavonoids***:

The plant extract (5ml) was added to 1% Aluminum chloride solution (3ml). The existence of flavonoids was established by observing a yellow coloration. Diluted ammonia solution (5ml) was added to the already prepared mixture and then the addition of concentrated $H_2SO_4$, the mixture was allowed to stand. The disappearance of yellow coloration confirmed the presence of flavonoids.

***Analyze for steroids***

The plant extract (2ml) was added to acetic anhydride (2ml) and then $H_2SO_4$ (2ml) was added. The occurrence of steroids was established as the color changed from violet to green or blue.

***Phenol:***

A 30 ml test tube was filled with 5 ml of the extract, followed by 10 ml of distilled water. In addition, 30 minutes were given for 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol to react. The emergence of a bluish-green hue was interpreted as a sign that phenol was present.

***Test for Terpenoids (Salkowski test):***

The plant extract (5 ml) was mixed with chloroform (2 ml) and then concentrated $H_2SO_4$ (3 ml) was added carefully to form a layer. A reddish-brow coloration at the interface indicated the presence of terpenoids.

***The Keller-Killani test (for cardiac glycosides and cardenolides):***

Concentrated H2SO4 (1 ml) was added after the plant extract (5 ml) had been combined with glacial acetic acid and one drop of ferric chloride solution. If a brown ring—a cardenolide typical of deoxysugar—formed at the contact, the presence of cardenolides would have been established. The emergence of a violet-green ring proved that glycoside was present.

***Analyze for alkaloids:***

In a steam bath, the plant extract (1ml) was mixed with 5ml of 1% aqueous HCL before being filtered while still hot. The residue was mixed with distilled water. A few drops of Mayer's reagent (potassium mercuric iodide solution), Dragendorff's reagent (potassium bismuth iodide solution), or Wagner's reagent were added to one milliliter of the filtrate (solution of iodine in Potassium iodide). Using Mayer's reagent, cream color production demonstrated the presence of alkaloids. Dragendorff's reagent or Wagner's reagent-induced reddish-brown precipitate further demonstrated the presence of alkaloids.

**Proximate Composition:**

According to the official procedures of analysis established by the organization of official analytical chemists, plant extracts were assessed for their immediate composition. (AOAC, 2005). All investigations were carried out in duplicate. The outcomes of the proximate composition forprotein, ash, and carbohydrate were presented in figure 1 while moisture content, fiber, and fat were presented in figure 2.

**Evaluation of antibacterial activity of the plants**

***Preparation of crude extracts***

To extract 50ml of the undiluted crude juice, fresh leaves of Siam weed, Waterleaf, and Garlic were each macerated separately in a mortar and pestle. This juice was later filtered with a membrane filter of pore size 0.45ul to obtain a sterile juice. The obtained crude extracts were poured into a sterile bottle at concentrations of 100%, and two folds dilution was used to serially diluted extract further into 50% and 25% concentrations. They were stored in air-tight bottles and labeled appropriately.

***Test bacterial isolates***

*Pseudomonas aeruginosa* was isolated from a tissue sample that was harvested from the gill of a diseased fish obtained from a local fish pond in Olodo town of Oyo State, Nigeria. The organism was grown overnight at $37^0$C in selective media, Pseudomonas Selective agar (Cetrimide agar), and the organism was further identified and interpreted according to (Austin and Austin, 2007).

**Table I. Phytochemical analysis of the *Allium sativum, Chromolaena odorata, and Talinum triangulare***

| Phytochemicals | *A. sativum* | *C. odorata* | *T. triangulare* |
|---|---|---|---|
| Alkaloids | vvv | vvv | vvv |
| Tannin | Vvv | vvv | vv |
| Phlobatannin | v | vv | v |
| Saponin | vv | vvv | vvv |
| Flavonoids | v | v | V |
| Anthraquinones | v | Nil | v |
| Steroids | v | v | v |
| Terpenes | v | v | V |
| Cardenolides | Nil | Nil | Nil |
| Phenol | vvv | vvv | vvv |
| Cardiac glycoside | v | vv | vv |

Observation Remarks: vvv = Considerable Quantity Present, v v = Modest Quantity Present; v = Trace Quantity Present; Nil = Absolutely Absent

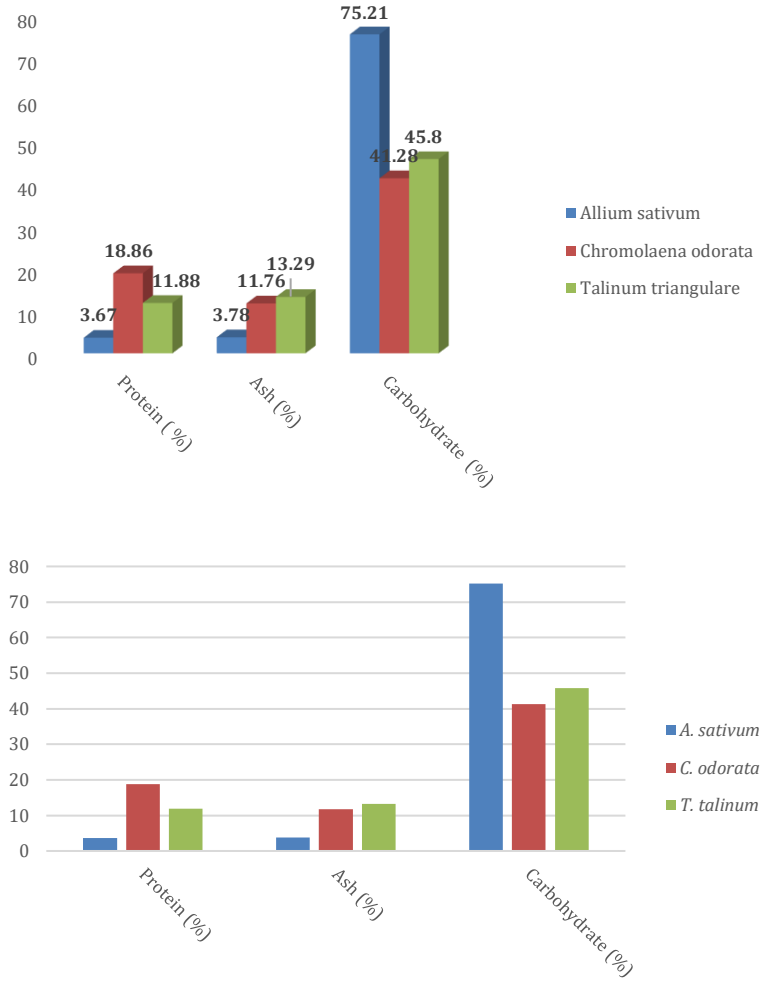


**Figure 1. Proximate analysis (Protein, Ash, Carbohydrate) of *Allium sativum, Chromolaena odorata* and *Talinum triangulare***

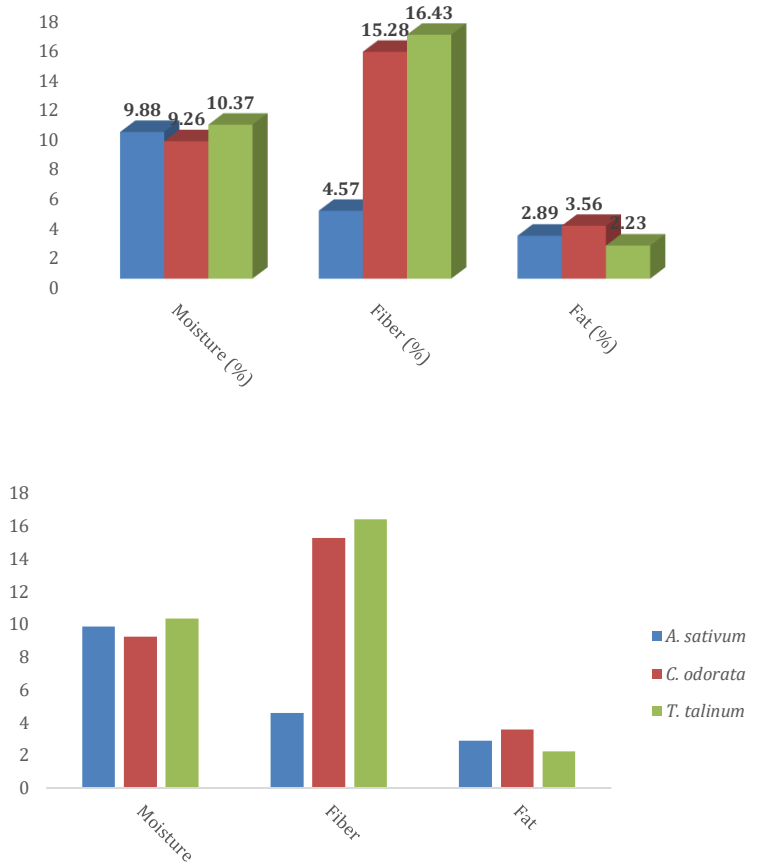


**Figure 2. Proximate analysis (Moisture, Fiber, Fat) of *Allium sativum, Chromolaena odorata,* and *Talinum triangulare***

***Antibacterial Activity***

The Agar diffusion technique as modified by (Osadebe and Ukwueze, 2004) was accepted to evaluate the antimicrobial activity of the plants. Broth cultures of the *P. aeruginosa* containing 1.4 x$10^6$ bacteria/ml (previously determined) were placed into a germ-free Petri dish and 15 ml melted Mueller Hilton Agar was added. The content was meticulously mixed and permitted to harden. Four hovels were prepared in each plate (5.00 mm width) by means of a disinfected cork-borer and an equivalent cork-borer leaf extract was moved into the holes by means of a Pasteur's pipette. An equal volume of distilled water was used as the negative control. The plates were permitted to settle for 60 minutes for the pre-diffusion of the extracts to happen and were incubated at 37°C for 24 h. At the completion of the incubation period, the antimicrobial action was evaluated by calculating the width of the zone of inhibition (ZOI) displayed by the extract (Table II).

**Statistical analysis**

For the descriptive analysis, the SPSS statistical software version 23 was employed.

## RESULTS AND DISCUSSION

The normal metabolic process in plants results in the formation of bioactive compounds often referred to as phytochemicals (Okigbo *et al*., 2009). Alkaloids, flavonoids, coumarins, glycosides, gums, polysaccharides, phenols, tannins, terpenes, and terpenoids are only a few of the classes of these phytochemicals that have been identified (Okwu and Josiah, 2006). According to (Okigbo *et al*., 2009), the beneficial effects of many medicinal plants are a result of synergistic or additive action of these phytochemicals compounds acting at single or multiple target sites of the host associated with a physiological process. This is in contrast to synthetic pharmaceuticals or drugs whose actions are based upon a single chemical. The results obtained in this trial showed

**Table II. Plant leaf crude extract's antibacterial properties**

| Plants Tested | Concentrations | Diameter of zone of inhibition (mm) | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | Mean | SD | CV | Min | Max | Median | Sum |
| ***Allium*** | 100% | 12.00 | ±1.41 | 23.57% | 10.00 | 16.00 | 11.00 | 48 |
| ***Sativum*** | 50% | 11.50 | ±1.71 | 29.70% | 8.00 | 16.00 | 11.00 | 46 |
| | 25% | – | – | – | – | – | – | – |
| ***Chromolaena*** | 100% | 12.50 | ±1.26 | 20.13% | 10.00 | 16.00 | 12.00 | 50 |
| ***odorata*** | 50% | 9.50 | ±0.96 | 20.16% | 8.00 | 12.00 | 9.00 | 38 |
| | 25% | 6.00 | ±0.82 | 27.22% | 4.00 | 8.00 | 6.00 | 24 |
| ***Talinum*** | 100% | – | – | – | – | – | – | – |
| ***triangulare*** | 50% | – | – | – | – | – | – | – |
| | 25% | – | – | – | – | – | – | – |
| **Distilled water** | | – | – | – | – | – | – | – |

SD= Standard Deviation, CV= Coefficient of Variations,– = No inhibition, mm= millimetres

the presence of alkaloids, tannins, phlobatannins, saponins, flavonoids, anthraquinones, steroids, terpenes, phenol, and cardiac glycoside in varied proportions and the absence of cardenolides as they were presented in Table I. The result showed that alkaloids and phenol are present in substantial quantities in all tested medicinal plants. Tannin is present in substantial quantity in *Allium sativum* and *Chromolaena odorata* while saponins are also present in substantial quantity in *Chromolaena odorata* and *Talinum triangulare* and moderately present in *Allium sativum.* A moderate quantity of tannins is found in *Talinum triangulare* and a moderate quantity of cardiac glycoside is equally found in *Chromolaena odorata* and *Talinum triangulare* however a trace quantity is found in *Allium sativum*. Phlobatannins are moderately present in *Chromolaena odorata* and their trace quantity is present in both *Allium sativum* and *Talinum triangulare*. Flavoid, steroid, and terpenes are all present in trace quantity in all the tested plants, and also trace quantity of anthraquinones is present in *Allium sativum* and *Talinum triangulare* but absent completely in *Chromolaena odorata*. It was pointed out by Heldt, (2005) that most of the phytochemicals present in plants are made through biosynthesis in the metabolic pathways. These bioactive are responsible for the therapeutic potential of these medicinal plants. However, variability in the phytochemicals content of the medicinal plants has been observed (Borokini and Omotayo, 2012), and this could be due to variation in the qualitative and quantity of bioactive compounds present in the plants as it was influenced by soil quality, cultivation period and season of collection of the plant materials. In this present study, the bioactive compounds which were identified were similar to what was obtained in the previous phytochemical screening of qualitative organic analysis of aqueous, ethanolic, chloroform, and petroleum extract of Garlic*,* Siam weed*,* and Waterleaf in the previous studies however the quantity varied. Plants' active components, which are being researched to use as pharmacological instruments to promote health and wellness, are thought to have therapeutic potential (Liu *et al*, 2006). According to Okwu, (2004) and Rios and Recio, (2005), West Africa's plants are so diverse that there is scarcely any sickness that both animals and humans cannot be treated by these plants. Many of these indigenous medicinal plants are utilized as spices and food plants, and they are occasionally added to dishes for medical purposes. Therefore, plants, especially the higher ones have been described as the sleeping giants for drug development (Edeoga, *et al*., 2005). Phenolics are common secondary metabolites in plants that have a variety of medicinal applications, including antioxidant, antimutagenic, anticarcinogenic, free radical scavenging, and lowering cardiovascular problems (Yen *et al*., 1993). The hydroxyl group is primarily responsible for the phenolics' capacity to scavenge free radicals. All of the examined plants have detectable amounts of phenol, making them potential antioxidants. It is well-recognized that free radicals play a significant part in a wide range of clinical symptoms. We are shielded from a number of diseases by antioxidants, which combat free radicals. They either work by removing reactive oxygen species from the environment or by defending the antioxidant defense mechanisms (Umamaheswari and Chatterjee, 2008). The importance of phenol as an analgesic, antipyretic, and anti-inflammatory phytochemical had also been documented (Michael, 2008). Reports had

been made on flavonoids to be antidiarrhoeal (Schnier *et al.,* 2005), antibacterial (Galeotti *et al*., 2008), and antimicrobial (Cushnie and Lamb, 2005). This could explain the rationale behind the use of plants like *Chromolaena odorata*, which has been used to treat a variety of ailments, including gonorrhea, chicken pox, measles, sore throats, coughs, piles, and toothaches (Bamisaye *et al*., 2014). Saponins had also been implicated as antimicrobial and antifungal (Forester and Hartmut, 2006) and therefore these plants could be useful for the management of fungi diseases such as ringworm, guinea worm, and skin diseases. Additionally, it improves nutrient absorption and aids with digestion (Forester and Hartmut, 2006). Similarly, tannins have been revealed to have antiviral (Lü *et al*., 2004), antibacterial (Akiyama *et al* 2001), and antiparasitic (Kolodziej and Kiderien, 2005) activities. Therefore, it is not surprising that Siam weed is used traditionally in Nigeria to cure skin conditions as well as various illnesses caused by viruses and bacteria. Additionally, tannins have been linked to a faster rate of blood coagulation and a reduction in blood pressure. This might be the basis of its efficiency as a blood clotting agent and as a therapeutic plant for controlling hypertension. The presence of saponins and tannins in Siam weed could be responsible for its hemostatic activity, thus supporting its traditional use for wound healing. Our results equally revealed the presence of high-quality crude protein present in Siam weed. This indicates the nutritional protein supplementation potential of this plant and this could be useful in large animal feed such as cattle. The attention of the Federal Government of Nigeria is drawn to this revelation to put an end to the farmers' and cattle rearers' constant clashes. Siam weed grows as an invasive plant in many parts of Nigeria and is therefore available throughout the year. A number of therapeutic uses such as anti-viral, anti-bacterial, anti-fungal, and antioxidant abilities had been attributed to *Allium sativum,* and more so it is commonly used today in many cultures as a seasoning or spice. The result also revealed that *T. triangulare* leaves contain a substantial amount of bioactive compounds.

The result of the study showed that *Chromolaena odorata* had the highest antibacterial activity at 100% crude extract i.e undiluted crude extract with a diameter of 12.5 mm zone of inhibition followed closely *by Allium sativum* with a diameter of 12.0 mm. The two folds dilutions of extract obtained from the two plants still showed antibacterial activity though lesser than the undiluted crude extract of both plants. These findings correlate with the observation of previous workers that the plants such as *Chromolaena odorata* contain antimicrobial substances (Douye *et al.*,2013).

## CONCLUSION

According to the current investigation, the plants under study contained components that are significant in the pharmaceutical industry. The detected phytochemicals' bioactivity has been supported by a large body of evidence from earlier investigations. Numerous studies have shown that the presence of these phytochemicals gives the plants under study physiological and therapeutic qualities that can be used to treat a variety of diseases. This is further validation of the facts concerning these plants' medicinal and nutritional capabilities that have already been mentioned. As a result, these plants' extracts may be a reliable source of therapeutic medicines. They can be effective income generators for people, and more research is encouraged to clarify the potential mechanism of action of these bioactive chemicals. It is advised that additional work be done to isolate, purify, and define the active ingredients responsible for the activity of these plants to further investigate and study this observation.

## ACKNOWLEDGMENTS

We would like to express our deepest thanks to all staff of the Food and Meat Hygiene Laboratory of the Department of Veterinary Public Health and Preventive Medicine of the University of Ibadan, Faculty of Veterinary Medicine, University of Ibadan, Ibadan, Nigeria for their cooperation and technical assistance.

## CONFLICT OF INTEREST

Authors declare that there is no conflict of interest to reveal.

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