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Preliminary Efficacy Assessment of Some Selected Indigenous Plant Species of Adamawa State, Nigeria on *Salmonella typhi*

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Authors' contributions

This research was carried out in full support of the authors. Author MA designed the entire study and protocols, helped in the collection and preparations of plant materials. Author MYT managed the literature search, interpreted the results, carried out statistical analyses and prepared the first draft of the manuscript. Author LTT proof read and corrected the draft. All authors read and approved the final manuscript.

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ABSTRACT

Aims: To screened for phytochemicals and determine the in-vitro efficacy of the plants extracts on *Salmonella typhi*.
Study Design: Phytochemicals and in-vitro efficacy assessment of seven medicinal plants in comparison to the standard antibiotics.
Place and Duration of the Study: Department of Biological Science Technology, Federal Polytechnic Mubi, Adamawa State, between October to December, 2017.
Methodology: isolation and identification of the test organism; preparation of plants extracts; phytochemical analyses of the plants parts on aqueous extracts; *in vitro* susceptibility test (agar well diffusion assay),
Results: Of the thirteen (13) phytochemicals screened from seven (7) medicinal plants, none of the plants possessed all the bioactive components. However, the least possessing six (6)

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phytochemicals (*Acacia* sp) while the highest possessed nine components (*Moringa oleifera* and *Carica papaya*). Tannins and steroids were present in all the plant leaves while free anthraquinones were lacking in those leaves. The efficacy of the medicinal plants on the test organism was carried out by agar well diffusion method. At 50 mg/ml concentration, aqueous extracts of the plants leaves showed no inhibitory effect on *S. typhi*. However, at 100 mg/ml concentration, growth of *S. typhi* was inhibited variably with the highest zone (14 mm) produced by *M. oleifera* and *Psidium guajava* extracts. For acetone extracts, variable zones of inhibition were produced by the leaves of all the plants at both 50 and 100 mg/ml concentration with the highest zone (25 mm) produced by *C. papaya* at 100 mg/ml. Statistically, the zones of inhibition produced by acetone extracts at 100 mg/ml concentration was significantly higher than those produced by acetone extracts at 50 mg/ml (p=0.000) and aqueous extracts at 100 mg/ml (p=0.009). However, there was no statistical difference between zones of inhibition produced by aqueous extracts at 100 mg/ml and acetone extracts at 50 mg/ml on *S. typhi* (p=0.197). The result further showed that *S. typhi* was highly susceptible to ciprofloxacin at both 50 and 100 mg/ml with a zone of inhibition greater than those of the aqueous and acetone extracts. Synergy among the seven plants leaves was higher at 100 mg/ml concentration for both aqueous and acetone extracts. However, the acetone extracts of the combined plants leaves produced highest zones of inhibition on *S. typhi* than combined aqueous extracts at both concentrations. Interestingly, the synergy of the aqueous plants extracts at 50 mg/ml produced a relative antibacterial effect on *S. typhi* (10 mm) when compared to the individual plants which demonstrated the non-inhibitory effect.
Conclusion: These results upheld the traditional ideology of using these plants singly or combine in the management of typhoid fever.

Keywords: Efficacy; Salmonella; phytochemicals; extracts.

1. INTRODUCTION

One of the laudable breakthrough which Science and Technology have brought to humans since early 1940s was the discovery of antibiotics. However, this giant stride was short-lived with the incursion of antibiotic resistance among bacterial pathogens. Unfortunately today, the rate at which pathogenic microorganisms are gaining resistance to highly acclaimed and accepted antimicrobials considering the relative absence or few flow of new antimicrobials to the markets, the number of drug options leaves us perilously close to none or only a single effective agent for some life-threatening infections [1]. If this trend is allowed to continue unchecked, sooner than expected, the resurgence of 'pre-antibiotic era' will set in and therapy for common bacterial infections will be unreached [2]. For these reasons, the quest for novel antimicrobials was shifted from synthetic drugs to natural products, especially from plants.

From time immemorial, plants have traditionally provided succor for numerous illness and are a source of hope for novel antimicrobials. This is because plants seem to possess numerous prospects when compared to synthetic drugs. These prospects include; better patient tolerance and fewer side effects when they are used for therapeutic purposes, relatively inexpensive and readily available and affordable, widely and highly accepted due to a long history of use, etc [3]. Medicinal plants are reservoirs of biologically active ingredients such as phytochemicals. Phytochemicals have been reported widely to possessed antimicrobial activities. The plants leaves under study are often used traditionally especially in Adamawa State Nigeria in the treatment and management of typhoid fever mostly in the form of concoction. Typhoid is caused primarily by *Salmonella typhi* and less commonly by the paratyphi sub species.

Salmonella is a Gram-negative, motile bacteria belonging to the family Enterobacteriaceae. These are found almost everywhere being an enteric pathogen. Cases of typhoid fever are increasing in an alarming rate especially in tropical countries like Nigeria. The pathogens typically gain entry into water systems through faecal contamination and man is infected after ingestion of such contaminated water.

Various plant extracts and phytochemicals have been widely investigated as possible potentials for the development of new and novel antimicrobials effective against disease-causing organisms and which could also help to manage the problem of multidrug-resistant organisms.

Therefore, this study reports the phytochemical nature of medicinal plants which are often used traditionally to manage typhoid fever in Adamawa State and the *in vitro* efficacy of the plants extracts on typhoid causing organism.

2. MATERIALS AND METHODS

2.1 Study Area

Mubi metropolis is a geo-political area comprising of two local government areas; Mubi North and Mubi South. The metropolis is located between latitudes 10° 05' and 10° 30'N of the equator and between longitude 13° 12' and 13° 19'E of the Greenwich meridian. The two Local government areas occupy a land area of 192,307 Km^2 and support a total population 260,009 people (National Population Census 2006). The area shares boundary with Maiha L.G.A in the South, Hong L.G.A in the West, Michika L.G.A and Cameroon Republic in the East. The major ethnic groups in Mubi includes; Fali, Gude, Kilba, Higgi, Margi and Nzanyi [4].

2.2 Collection of Plant Materials

The leaves of all the plants (*Mangifera indica*, *Psidium guajava*, *Carica papaya*, *Acacia* sp, *Musa paradisiaca, Moringa oleifera* and *Citrus sinensis*) were collected at the end of the rainy season (4^{th} - 8^{th} October, 2017) around Dipalma area of Mubi metropolis. All the plants parts were identified and authenticated in the Botany unit of Department of Biological Science Technology, Federal Polytechnic Mubi, Adamawa State.

2.3 Extraction Procedure

The leaves of all the plants were properly washed, air-dried at room temperature and ground to fine powder. 50 g of each of the powdered plant part was soaked in 200 ml distilled water and was allowed to stand for 48 hrs at room temperature after thorough vortexing. Each mixture was filtered using whatman no.1 filter paper. The filterate were concentrated in vacuo using rotary evaporator. Similar procedure was followed to obtain acetone extracts of the plants parts using acetone as extracting solvents. The aqueous and acetone dried extracts of all the plants part were properly labelled and stored in sample bottles at 4°C prior to use [5].

2.4 Qualitative Phytochemical Screening

Qualitative phytochemical screening for the presence of thirteen (13) phytochemical compounds were carried out on the aqueous extracts of all the plants parts using standard procedure as described by Sofowora [6] and Egwaikhide et al. [7].

2.4.1 Test for terpenoid

Five (5) ml of each extract was mixed in 2 ml of chloroform. 3 ml of concentrated H_2SO_4 was then added to form a layer. Formation of Reddish-brown precipitate coloration at the interface indicates the presence of terpenoids.

2.4.2 Test for free anthraquinones

Five (5) ml of chloroform was added to 0.5 g of the powdered dry plant parts of each specimen. The resulting mixture was shaken for 5 minutes after which it was filtered. The filtrate was then shaken with equal volume of 10% ammonia solution. The presence of a bright pink colour in the aqueous layer indicates the presence of free anthraquinones.

2.4.3 Test for cardiac glycosides

Five (5) ml of each extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was underplayed with 1 ml of concentrated sulphuric acid. A brown ring at the interface indicates the deoxysugar characteristics of cardenolides. A violet ring may appear below the ring while in the acetic acid layer, a greenish ring may be formed.

2.4.4 Test for phlobatannins

Deposition of a red precipitate when an aqueous extract of each plant sample was boiled with 1% aqueous hydrochloric acid was taken as evidence for the presence of phlobatannins.

2.4.5 Test for polyphenols

Two grams (2 g) of powdered sample was boiled with distilled water for 30 min, and then 1 ml of 5% ferric chloride (Iron III chloride) and 1 ml of 1% potassium ferric cyanides was added to the solution. It was filtered and observed for the formation of blue-green colour.

2.4.6 Test for tannins

To 3 ml of extract, 3 ml of 5% w/v ferric chloride solution was added. A bluish-black or brownish-green precipitate indicates the presence of tannins.

2.4.7 Test for saponins

One gram (1 g) of each powdered dried stain was separately boiled with 10 ml of distilled water in a bottle bath for 10 min. The mixture was filtered while hot and allowed to cool. 2.5 ml of filtrate was diluted to 10 ml distilled water and shaken vigorously for 2 min (frothing indicated the presence of saponins in the filtrate).

2.4.8 Test for combined anthraquinones

One gram (1 g) of powdered sample of each specimen was boiled with 2 ml of 10% hydrochloric acid for 5 minutes. The mixture was filtered while hot and the filtrate was allowed to cool. The cooled filtrate was partitioned against equal volume of chloroform and the chloroform layer was transferred into a clean dry test tube using a clean "pipette". An equal volume of 10% ammonia solution was added into the chloroform layer, shaken and allows to separate. The separated aqueous layer was observed for any colour change. Delicate rose pink colour shows the presence of combine anthraquinones.

2.4.9 Test for alkaloids

A small portion of crude extract was dissolved in 5 ml of 1% hydrochloric acid, filtered and tested with Dragendorff's reagent and Mayer's reagent separately. Any precipitate or turbidity with the reagent suggests the presence of alkaloids.

2.4.10 Test for steroids

A small portion of the extract was dissolved in 1 ml of chloroform and filtered. To the filtrate on ice, 1 ml of acetic acid was added and then a few drops of concentrated sulphuric acid were run down the side of the test tube. The appearance of blue, bluish-green or a rapid change from pink to blue colours indicates the presence of steroids.

2.4.11 Test for flavonoids

One gram (1 g) of the powdered dried leaves of each specimen was boiled with 10 ml of distilled water for 5 minutes and filtered while hot. Few drops of 20% Sodium hydroxide solution were added to 1ml of the cooled filtrate. A change to yellow colour which on addition of acid changed to colourless solution depicted the presence of flavonoids.

2.4.12 Test for carotenoids

One gram (1 g) of each specimen sample was extracted with 10 ml of chloroform in a test tube with vigorous shaking. The resulting mixture was filtered and 85% sulphuric acid was added. A blue colour at the interface shows the presence of carotenoids.

2.4.13 Test for reducing compound

To about 1 g of each sample in the test tube, 10 ml distilled water was added and the mixture boil for 5 minutes. The mixture was filtered while hot and the cooled filtrate makes alkaline to litmus paper with 20% sodium hydroxide solution. The resulting solution was boiled with an equal volume of Benedict qualitative solution in a water bath. The formation of a brick-red precipitate depicts the presence of reducing compound.

2.5 Test Organisms

The test isolates used for this study was *Salmonella typhi* isolated from stool samples collected from Mubi General Hospital, Adamawa State.

2.6 Culture and Identification of the Test Organism

A loopful of the stool sample was introduced into prepared selenite F broth in a test tube and incubated for 24 hrs at 35-37°C. After incubation, a loopful of the broth was streak on Salmonella-Shigella agar (SSA) and replicated on Deoxycholate citrate agar (DCA). The plates were incubated for 24 hrs at 37°C. Pure colonies were identified based on their morphological, cultural and biochemical properties on Kligler iron agar. Colonies that are creamy, colourless with dark centres in both media were taken as *Salmonella typhi* [8].

2.7 Determination of Antibacterial Activity

The dried aqueous and acetone extracts of all the plants parts were reconstituted with distil water and glycerol respectively to obtain a final concentration of 50 and 100 mg/ml for this test. The susceptibility test was done using agar well diffusion method. 0.1 ml aliquot of the test organism suspension (equivalent to 0.5 McFarland standards) was transferred onto dried agar plates in duplicate and was spread evenly with a sterile bent glass rod. After drying, four (4)

wells were bored (using 6 mm diameter cork borer) into the dried nutrient agar plates and 0.5 ml each of the extracts was aseptically introduced into the wells. Glycerol was introduced into another well on a separate plate as control. The plates were then incubated at 37°C for 24 h after which zones of inhibition were measured in millimetres (mm) and recorded appropriately [5]. For the purpose of comparison, standard antibiotic (Ciprofloxacin) was also tested on the test organism at 50 and 100 mg/ml.

2.8 Statistical Analysis

Duncan multiple range test of one way ANOVA was use to determine significance difference between diameters of zones of inhibition produced by aqueous and acetone extracts of all the plants on *S. typhi* at 50 and 100 mg/ml concentrations. Statistical difference was taken when p≤0.05.

3. RESULTS

The result in Table 1 shows the phytochemical components of seven (7) medicinal plants leaves using their aqueous extracts. Out of the thirteen (13) phytochemicals screened, none of the plant possessed all the bioactive components. However, the least possessed six (6) phytochemicals (*Acacia* sp) while the highest possessed nine components (*M. oleifera* and *C. papaya*). None of the plant possessed free anthraquinones. Moreso, all the plants lack combined anthraquinones and carotenoids except *P. guajava* and *M. indica* respectively. Contrariwise, tannins and steroids were present in all the plant leaves. The result also showed that the presence of other phychemicals in the plants leaves was variable.

The result in Table 2 shows the effect of aqueous and acetone extracts of all the plants leaves on *S. typhi*. The result revealed that zones of inhibition produced against *S. typhi* by the extracts of all the plants increases with increase in concentration of the plants extracts. At 50mg/ml concentration, aqueous extracts of the plants leaves showed no inhibitory effect on *S. typhi*. However, at 100 mg/ml concentration, growth of *S. typhi* was inhibited variably with the highest zone (14 mm) produced by *M. oleifera* and *P. guajava* extracts.

For acetone extracts, the zones of inhibition produced by the leaves of all the plants were variable at both 50 and 100 mg/ml concentration with the highest zone (25 mm) produced by *C. papaya* at 100 mg/ml. However, acetone extracts of *C. papaya* and *C. sinensis* leaves had no inhibitory effect on *S. typhi* at 50mg/ml concentration.

Statistically, the zones of inhibition produced by acetone extracts at 100 mg/ml concentration was significantly higher than those produced by acetone extracts at 50 mg/ml (p=0.000) and aqueous extracts at 100mg/ml concentration (p=0.009). However, there was no statistical difference between zones of inhibition produced by aqueous extracts at 100 mg/ml and acetone extracts at 50 mg/ml on *S. typhi* (p=0.197).

Table 1. Phytochemical components of the medicinal plants

Phytochemicals	Tested plant species						
	MO	CP	CS	MP	MI	PG	AS
Terpenoids	+	+	+	-	+	+	+
Free anthraquinones	-	-	-	-	-	-	-
Cardiac glycosides	+	+	-	+	-	+	+
Phlobatannins	+	+	+	+	-	-	-
Polyphenols	+	+	+	+	-	-	+
Tannins	+	+	+	+	+	+	+
Saponins	-	+	+	+	+	+	-
Combined anthraquinones	-	-	-	-	-	+	-
Alkaloids	+	+	+	-	+	+	-
Steroids	+	+	+	+	+	+	+
Flavonoids	+	+	+	+	+	-	+
Carotenoids	-	-	-	-	+	-	-
Reducing compounds	+	-	-	-	+	+	-

Legend: += presence, -= absence, MO= Moringa oleifera, CP= Carica papaya, CS= Citrus sinensis, MP= Musa paradisiaca, MI= Mangifera indica, PG= Psidium guajava, AS= Acacia sp

Table 2. Diameter of zone of inhibition on *Salmonella typhi*

Plant part	Zone of inhibition (MM)			
	Aqueous extracts		Acetone extracts	
	50 mg/ml	100 mg/ml	50 mg/ml	100 mg/ml
Moringa oleifera	-	14	11.0	18
Mangifera indica	-	11	10.0	16
Psidium guajava	-	14	8.0	10
Carica papaya	-	-	-	25
Acacia spp	-	13	10.0	20
Musa paradisiaca	-	11	10.0	20
Citrus sinensis	-	10	-	16
C7	10	15	13	21

Legend: C7= combination of 7 herbs (serial no 1-7). -= no zone of inhibition

Synergistically, the antibacterial effect of the seven plants leaves extract was higher at 100mg/ml concentration for both aqueous and acetone extracts. However, the acetone extracts of the combined plants leaves produced the highest zones of inhibition on *S. typhi* than combined aqueous extracts at both concentrations. Interestingly, synergy of the aqueous plants extracts at 50 mg/ml produced relative antibacterial effect on *S. typhi* (10 mm) when compared to the individual plants which demonstrated non-inhibitory effect.

The results further showed that *Salmonella typhi* was highly susceptible to ciprofloxacin at both 50 and 100 mg/ml with diameter of zone of inhibition greater than those of the aqueous and acetone extracts (Fig. 2).

4. DISCUSSION

Phytochemicals are the secondary metabolites usually found in plants parts and are known to carry out specific functional roles. For example, phytochemicals help to defend plant against intrusion by microorganisms, insects and herbivores. In the ancient times, before the advent of antibiotics, man used phytochemicals in the form of plants as the only and sole weapon to fight against infectious diseases. In recent times they are still indispensable as they constitute the central components of today's pharmaceuticals. With the incidence of multidrug-resistant pathogens and the menace they constitute to therapy, researchers are focusing on plants derivatives as possible solutions [2,9]. Phytochemicals can have complementary and overlapping mechanisms of action in the body which includes hormone metabolism, modulation of detoxification enzymes, stimulation of the immune system and antioxidant effects. Water was used for extraction to subject the plants parts to the methods often used traditionally, while acetone was used for extraction because it is believed that organic solvents possessed the potentials to extract potent bioactive ingredients with antibacterial activity than water.

The Phytochemical screening in this study was carried out only on aqueous extracts of all the plants leaf.

In this study, tannin was detected in the aqueous extract of all the plant leaves. In contrast to the findings of this study, previous studies showed that *C. papaya* [10] and *M. oleifera* [11] lack tannins. In line with this study however, several other studies detected tannins in *M. oleifera* [12,13,14], *P. guajava* [15,16,17], *M. indica* [15,18], *C. papaya* [15,19], *C. sinensis* [20] and *Acacia* spp [21]. More so, steroids was detected in all the plant species as shown in our study which is in agreement with the report of Yahaya et al. [19], Ekwenye and Edeha [20] and Krishna et al. [18] on *C. papaya, C. sinensis* and *M. paradisiaca* aqueous extract respectively. On the contrary, previous reports were unable to detect steroids in *M. indica* [22,23], *P. guajava* [16] and *M. oleifera* [12,24]. Terpenoid in our study was not detected only in *M. paradisiaca* and this contradicts previous findings on the same plant part [15,18]. Contrary to our findings, a study was unable to detect terpenoid in *C. papaya* [15]. Also, the findings of our study showed that flavonoid was not detected only in *P. guajava*. On the contrary, several studies report the presence of flavonoid on the same plant part [15,16,17,25]. More so, some studies reported the absence of flavonoid in aqueous extract of *C. papaya* [10] and *Acacia* sp [21] which was contrary to the findings of our study. The findings that combine anthraquinones was only detected in *P. guajava* as shown in our study agrees with the report of

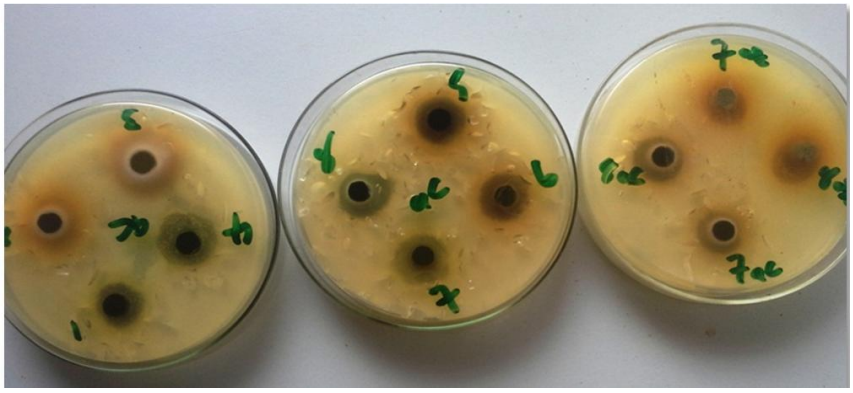

Fig. 1. Plates showing diameter of zones of inhibition of the medicinal plants on *S. typhi*
Legend: ac= acetone extract, 1= M. oleifera, 2= M. indica, 3= P. guajava, 4= C. papaya, 5= Acacia spp, 6= M. paradisiaca, 7= C. sinensis, 8= plant part not included in this study. 7ac= combination of seven plants (acetone extract), 7aq= combination of seven plants (aqueous extract), 8ac and 8aq (not included in this study)

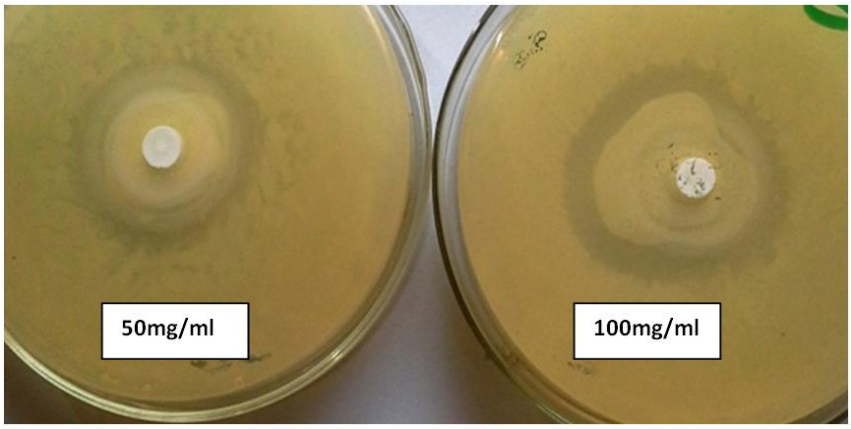

Fig. 2. Plates showing diameter of zones of inhibition of ciprofloxacin against *S. typhi*

Ali et al. [16]. Also, free anthraquinone was not detected in aqueous extracts of all the plant spp used in this study. This finding agrees with previous report on *M. indica* [22] and *M. oleifera* [12,24]. The finding that carotenoid was detected only in *M. indica* as shown in the present study was contrary to previous report [22] which showed that carotenoid was absent in the same plant leaf.

Furthermore, the findings of our study showed that saponin was not detected in *Acacia* spp and *M. indica*. This observation concurred with the reports of Okoro et al. [21] and Okoko et al. [22] on the same plants. In contrast to our observation, the absent of saponins in *C. papaya* [10] and *M. paradisiaca* [18] was reported in earlier studies. Our study also revealed the absent of alkaloid in *Acacia* spp and *M. paradisiaca* which was contrary to the report of Okoro et al. [21] and Bisht et al. [15] on the same plants. However, in agreement with our findings, the report of Krishna et al. [18] revealed that alkaloid was not detected in the aqueous extract of *M. paradisiaca* leaf. Cardiac glycoside in our study was not detected in *C. sinensis* and *M. indica*. This observation was contrary to earlier report on *M. indica* [22,23] but concur with another report on *C. sinensis* [20]. In contrast to our findings also, previous studies report the absence of cardiac glycosides in *Acacia* sp [21] and *C. papaya* [10]. Also, polyphenols in our study was not detected in *M. indica* and *P. guajava*. This observation was not in agreement with previous report on the same plants [16,22,23].

Plants are valuable sources of biological active principles which are stored in its various parts. Their production and release into solution are triggered by external challenges. Consequently, variation in the phytochemicals as reported in

this study might be due to physiological age of the plant, variation in soil types or soil nutrients, geographical location on which the plant is growing, percentage humidity of the harvested material, situation and time of harvest, method of extraction and extracting solvent [5].

The antagonistic effect of the plants extracts on *S. typhi* was demonstrated at 50 and 100 mg/ml concentration for aqueous and acetone extracts of the plants leaves. Aqueous extracts of all the plants leaves at 50 mg/ml concentration had no inhibitory effect on *S. typhi*. This is in agreement with previous reports on the plants under study [13,21,22,26,27]. However, contrary to our findings, the aqueous extracts of *C. papaya* [19] and *P. guajava* [16] were reported to have inhibitory effect on *S. typhi* at 50 mg/ml concentration. More so, the activity of acetone extracts at 50mg/ml demonstrated significant antibacterial effect on *S. typhi* except for *C. papaya* and *C. sinensis* on which the organism was resistant. This is quite similar to earlier report on *M. oleifera* [24] and *M. indica* [22,28].

Lack of inhibitory effect on *S. typhi* by aqueous extract of *C. papaya* leaf at 100 mg/ml concentration as shown in our study concurred with earlier reports [10,26], at the same time went contrary to the studies of Yahaya et al. [19] which in their studies revealed that aqueous extract of *C. papaya* leaf had antibacterial effect on the organism under study. It was also observed in our study that at 100mg/ml concentration, aqueous extracts of all the plants except *C. papaya* had antibacterial activity against *S. typhi*. This observation concurred with the reports of earlier studies on *M. oleifera* [11] and *P. guajava* [16,27]. On the contrary, several authors have reported non-inhibitory effect of aqueous extract of *M. oleifera* leaf [13,24], *M. indica* leaf [29] and *Acacia* sp leaf [21] on *S. typhi* at 100 mg/ml concentration.

The acetone extracts of all the plants part at 100 mg/ml concentration exhibit remarkable and significant antibacterial activity against *S. typhi*. This observation was reported previously on *M. oleifera* [24] and *M. indica* [22,28].

The impressive and significant antibacterial activity demonstrated on *S. typhi* by acetone extracts at 100 mg/ml concentration and lack of significant difference in zone of inhibition between acetone extracts at 50 mg/ml and aqueous extracts at 100 mg/ml concentration point to the superiority of acetone (organic solvent) over water in extracting ingredients and biological active components with antibacterial activities. To buttress our findings, previous study has shown that type of solvents and method of preparation of the extracts greatly affects antimicrobial activity of plants [30]. Moreover, previous studies reported that the antibacterial activity of plants extracts is largely dependent on its molecular weight and diffusion rates through agar rather than its concentration [31]. The lower the molecular weight of plant active components the higher the rate of diffusion through agar and consequently the higher the efficacy on bacterial isolates. Therefore, the variation in zones of inhibitions as shown in our study could be attributed to any of the outlined factors.

Furthermore, it was observed that the diameter of the zones of inhibition of the aqueous and acetone extracts of all the plants leaves increases as their concentrations increases. This is in agreement with previous reports [3,19,28]. The findings of the study also showed that synergy among the plants (combination of all the plants leaves) have improved the efficacy of the plants extracts on *S. typhi* especially the aqueous extracts as shown by their diameter of zones of inhibition. This finding upheld the traditional philosophy on the use of these plants in synergy in treating and managing typhoid fever especially in Adamawa State.

The diameter of zone of inhibition produced by ciprofloxacin on *S. typhi* was higher than those produced by acetone and aqueous extracts of all the medicinal plants at the same concentrations. This could partly be attributed to the fact that the standard antibiotic is a synthetic drug and in a purified state, while our extracts are in crude forms, full of impurities and other substances which may hamper the diffusion and biological activity of the active ingredients. Moreover, the concentration of the active ingredients in the crude extracts might be limited. These and other factors may have contributed to lower zones of inhibition demonstrated by our crude extracts in comparison to those produced by ciprofloxacin.

5. CONCLUSION

All the plants used in our study are phytotherapeutic plants often used in folk medicine because it is believed to possess biologically active ingredients that help in the treatment and management of various illnesses. Its inhibitory effects on *Salmonella* singly or in combination provides basis for its

recommendation for its use in the treatment of gastroenteritis, typhoid fever and other related diseases.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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