Antibacterial activity of the methanolic leaf extract of some medicinal plants used by traditional birth attendants in Sokoto Metropolis

Abstract

Leaves of *Ocimum basilicum, Leptadania hastata* and *Momordica balsamina* are locally used by traditional birth attendants at pre and post-partum. The present study investigates the phytochemical and antimicrobial activity of the leaf extracts against isolates of *L. monocytogenes*. Standard microbiological techniques and polymerase chain reaction were used to isolate and identify the bacteria. Phytochemical analysis revealed the presence of tannins, flavonoids, carbohydrates, alkaloids, terpinoids and glycosides in the studied extracts. MIC of the extract of *M. balsamina* shows a total inhibition of *Listeria monocytogenes* at a concentration of 2,000µg/mL. No isolate was inhibited at concentration lower than 125µg/mL of *M. balsamina* extract. MIC of the extract against *L. hastata* was observed at a concentration of 2,000µg/mL. *O. basilicum* extract inhibited the growth of the isolates at a concentration of 2,000µg/mL. The MBC of the extracts, was between 5,000-0.61µg/mL were used, however, no antibacterial activity was observed on the bacterium with any of the three plants extracts at all concentration levels (0.61-5000πg/mL). The leaf extracts demonstrated significant inhibitory activity but lack bacteriocidal effects on *L. monocytogenes*. It was recommended that further studies on the preparation, effective doses and side effects of these extracts in animal models are warranted.

Keywords: Antibacteria, *Ocimum basilicum, Leptadania hastata* and *Momordica balsamina,* Medicinal plants, Traditional Birth Attendants, Sokoto.

Introduction

For many years, traditional medicines from flowers, bark, leaves and fruits of plants have been utilized for medicinal purposes (Ogbuewu *et al.*, 2011). This has been the practice of several people throughout the beginning of human civilization. This is

because, plants are the richest resource of drugs of traditional modern medicines, nutraceuticals, food supplements, pharmaceutical intermediates and chemical entities for synthetic drugs (Hameed *et al*., 2015). Thus, traditional medicine has existed for ages and has relied largely on experience handed down from one generation to another.

The genus *Ocimum* comprises more than 150 species and is considered as one of the largest genera of the Lamiaceae family (Hameed *et al*., 2015). *Ocimum basilicum* (sweet basil) known in Hausa as *Dodoya*, is an annual herb which grows in several regions all over the world. The plant is widely used in food, oral care products and the essential oil of the plant is also used as perfumery (Chiang, 2005; Ali *et al*., 2015). Koba *et al.* (2009), reported *O. basilicum* and *O. gratissimum* as potential insecticides.

Leptadenia hastata Pers. Family name Asclepiadaceae, commonly known in Hausa as *Yadiya* is also a traditional plant used by indigenous traditional birth attendants at the third trimester of pregnancy. *Momordica balsamina* from the Family Cucurbitaceae (called *Garahuni* in Hausa) is a plant widely used as an anti-inflammatory, treatment of stomach ache and the leaves as breast milk stimulant (Abubakar *et al*., 2007).

The need for new antimicrobial agents is greater than ever because of the emergence of multidrug resistance in common pathogens, the rapid emergence of new infections, and the potential for use of multidrug-resistant agents in bioweapons (Spellberg *et al.*, 2004). Paradoxically, some pharmaceutical companies have indicated that they are curtailing anti-infective research programs. There still exist a vast number of tropical trees with tremendous medicinal potentials but with no empirical proof to support claims of efficacy (Akindahunsi and Salawu, 2005; Kotta *et al.*, 2013).

The United States Food and Drug Administration (FDA) approval of new antibacterial agents decreased by 56% over the past 20 years (1983–1987 vs. 1998–2002) (Shlaes

and Moellering, 2002). Projecting future development, new antibacterial agents constitute 6 of 506 drugs disclosed in the developmental programs of the largest pharmaceutical and biotechnology companies. Despite the critical need for new antimicrobial agents, the development of these agents is declining. Solutions encouraging and facilitating the development of new antimicrobial agents are needed (Spellberg *et al.*, 2004). The purpose of this research is to determine the antibacterial activity of the leaf extracts of (*Ocimum basilicum*, *Leptadania hastata* and *Momordica balsamina*) against isolates of *L. monocytogenes* isolated from vegetables.

Materials and Methods

Study Area

Sokoto is the capital of Sokoto State, located in the North Western part of Nigeria. With a land area of approximately 56,000 square kilometers, it is located between longitudes 11°30" to 13°50" East and latitude 4° to 6° North (Anon, 2001). The state is bordered in the North by Niger Republic, Zamfara State to the East and Kebbi State to the South and West (Anon, 2001). Sokoto is located in the Sudan Savannah vegetation belt with sandy soil and a humidity of below 40% year-round except during the rainy season when it rises to 60% (Ileoje, 1971). The two dominant seasons are the wet (June-October) and dry (November-May) seasons. The former begins in June and lasts up to October, while the latter begins in November and last up to May (Mahmuda *et al*., 2020). Sokoto state is second in the nation's livestock population with estimates of about 3 million cattle, 4 million sheep, 4.5 million goats and 3 million poultry (Buhari, 2008; SSIPC, 2008).

Collection and Identification of Plant Materials

The plant materials (leaves) were collected from farmlands and local markets within Sokoto metropolis, Nigeria. The plants were collected into clean poly bag and transported to Herbarium of Botany Unit, Department of Biological Sciences, Usmanu Danfodiyo University Sokoto, Nigeria for identification and issuance of a voucher number.

Isolation of Listeria/Test Isolates

Vegetables which include onions, cabbages, tomatoes and lettuce were obtained from five different selling points at Kasuwar daji, Sokoto. Each specimen was put in separate plastic bag and transported to the Microbiology Department laboratory UDUS for analysis.

Morphological and Biochemical Characteristics

Listeria spp cultures were enriched and subcultured according to the procedure previously described by Navas *et al*., (2007). *Listeria monocytogenes* was confirmed by morphology and gram staining was performed in accordance to the procedure as documented by Koneman, (2005). Active motility of the bacteria was determined using the method and characteristics described elsewhere. Some biochemical tests (Catalase and Oxidase) were perfomed using the procedure described by (Cheesborough, 2010).

Molecular Identification of *Listeria* species

DNA extraction was done as previously documented (Wu *et al.*, 2018; Hwang *et al.*, 2014).

Amplification of 16S rRNA region of the bacterial genome

Sixteen *S* (16S rRNA) region with the following primer sequence was used 27F **(**5'-AGAGTTTGATCMTGGCTCAG-3') 907R (5'-CCGTCAATTCMTTTRAGTTT-3') as described by Jiang *et al*. (2006) A 25 µl reaction mix containing the following component: Qiagen, toptag PCR master mix 2.5 µl 0.2 µl, of each forward and reverse primer, 17.1 µl of nuclease free-water, 5 µl of DNA, template in a 0.2 microtube (PCR tube) the tubes were capped and transferred into applied Biosystem 7700 thermocycler with the following cycling conditions: denaturation at 94°C for 30 s, annealing at 55°C for 30 s, and extension at 72°C for 1 min with a final extension at 72°C for 10 min for 35 cycles. The DNA bands were then viewed in a 1% Agarose gel (Murakami *et al.*, 1991) by illumination with UV light and images recorded by photography (Biorad imager).

Phytochemical analysis

The extract was used for screening of phytochemicals such as proteins, flavonoids, glycosides, carbohydrates, terpinoids and tannins. Preliminary phytochemcial screening was conducted on the plant extract, according to the standard procedures described previously (Okwu, 2005; Ayoola *et al*., 2008; Santhi *et al*., 2011).

Testing for Antibacterial Activity

The leaf extracts were tested for their antibacterial activities in vitro against strains of Listeria isolated above on Mueller Hinton agar (MHA) using a modification of the Kirby Bauer disk diffusion method. Chloramphenicol (30µg) was used as standard drug (as reference) in bactericidal analysis (Ogidi *et al*., 2015).\

Minimum Inhibitory Concentration (MIC)

The estimation of MIC of the crude extracts were carried out using the method of Akinpelu and Kolawole (2004). The MIC was taken as the lowest concentration that prevented the growth of the bacteria.

Minimum Bactericidal Concentration (MBC)

The MBC of the plant extracts were determined by a modification of the method of Spencer and Spencer (2004). The MBC was taken as the concentration of the extract that did not show any growth on a new set of agar plates.

Statistical Analysis

SPSS version 25.0 was used for statistical analysis, data generated was presented in the form of tables and graphs where appropriate. All measured parameters were calculated and expressed as Mean ±Standard Deviation or percentages. For all numerical values, homogeneity of variances was tested using the Bartlett's test. Probability of 0.05 ($p < 0.05$) was used as the criterion of significance.

Results

The study determines the antimicrobial activity of *M. balsamina, O. Basilicum* and *L. hastata* leaves extracts using nine (9) isolates of *L. monocytogenes* isolated from vegetables in Sokoto metropolis. The source and biochemical reaction of each isolate included in the study are depicted in Table 1. Three of the isolates (O4, O9 and O14) were recovered from onion samples, two from (L5 and L13) lettuce, two (T9 and T13) from tomatoes and another two isolates (C2 and C3) from cabbages. The corresponding morphologic characteristics (growth on Listeria agar, Gram stain and motility) and

some biochemical reactions (catalase and oxidase) for each of the isolates are shown in the same table. Based on the molecular identification, a 956 bp gene from the 18S ribosomal subunit was amplified (Figure 1).

The phytochemical analysis revealed the presence of tannins, saponins, flavonoids, carbohydrates, alkaloids, phenols, terpinoids and glycosides in all the studied extracts. Cholesterol, was found to be present in all the extracts except in *L. hastata*. However, phlabotannins tested positive only for the *O. basilicum* leaf extract (Table 2). Antibiogram patterns of *L. monocytogenes* isolated from vegetables against some selected antibiotics (Table 3).

The MIC of the nine isolates to the extract of *M. balsamina* shows a 100% inhibition of all the nine tested *L. monocytogenes* isolates at a concentration of 2,000 µg/mL. At 2,000 µg/mL 8 (88.9%) out of the 9 isolates were inhibited. This number was reduced to 4 (44.4%) isolates inhibited at 500 µg/mL, a number that was further reduced to only 2(22.2%) and 1(11.1%) isolate when the concentration was reduced to 500 and 125 µg/mL of the extract. No isolate was inhibited at concentration lower than 125 µg/mL of *M. balsamina* extract (Table 4).

Table 1: Morphological and Biochemical identification of *L. monocytogenes* from vegetables

S/N	Isolate	Gram Rxn	Shape	Motility	Catalase	Oxidase	Suspected Organism
1	O4	+	R	Motile	+	-	Listeria *sp*
2	O9	+	R	Motile	+	-	Listeria *sp*
3	C3	+	R	Motile	+	-	Listeria *sp*
4	L5	+	R	Motile	+	-	Listeria *sp*
5	T13	+	R	Motile	+	-	Listeria *sp*
6	L13	+	R	Motile	+	-	Listeria *sp*
7	C2	+	R	Motile	+	-	Listeria *sp*
8	O14	+	R	Motile	+	-	Listeria *sp*
9	T9	+	R	Motile	+	-	Listeria *sp*

KEY: isolates from this point onwards were identified by an annotation that includes a single letter and a number; the letter represents the first alphabet in name of the vegetable from which it was isolated while the number is a serial number that identified a vegetable from a collection. Thus;
O4 = Onion 4th isolate; O9 = Onion 9th isolate; C3 = Cabbage 3rd isolate; L5 = Lettuce 5th isolate; T13 = Tomato 13th isolate; L13 = Lettuce 13th isolate; C2 = Cabbage 2nd isolate; O14 = Onion 14th isolate; T9 = Tomato 9th isolate; R=Rod;

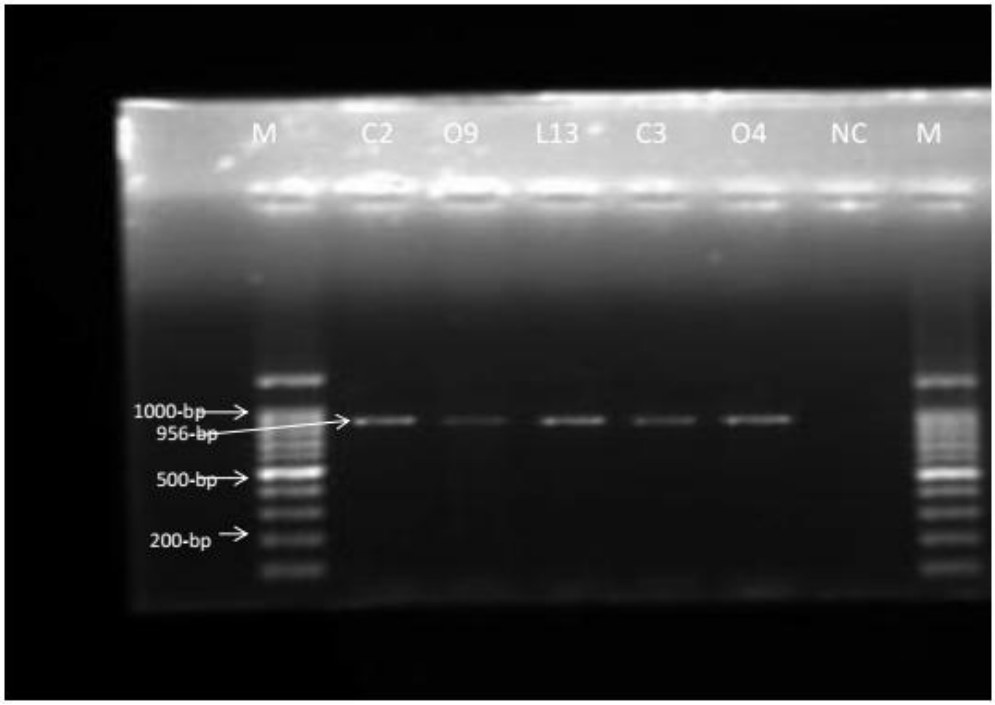

Figure 1: Agarose gel electrophoresis pattern of 16S ribosomal subunits specific for *Listeria monocytogenes*. M=DNA Ladder; C2, O9, L13, C3, O4 =samples; NC= Negative control.

Table 2: Preliminary screening for Phytochemicals composition of some plants used by traditional birth attendants

Phytochemicals	***Mormodica balsamina***	***Ocimum basilicum***	***Leptadania hastata***
Tannins	+	+	+
Phlabotannins	-	+	-
Saponins	+	+	+
Flavonoids	+	+	+
Cardiac glycosides	+	+	+
Carbohydrates	+	+	+
Cholesterol	+	+	-
Terpinoids	+	+	+
Alkaloids	+	+	+
Phenols	+	+	+
Glycosides	+	+	+

Table 3: Antibiogram patterns of *L. monocytogenes* isolated from vegetables against some selected antibiotics

Isolate	DA	RD	AMC	F	TEC	CIP	TE	FOX	QD	C	AMP	SXT	CN
T9	I	S	S	S	S	S	S	S	S	S	S	R	S
O14	S	S	S	S	S	S	S	S	S	S	S	S	S
L5	S	S	S	S	S	S	S	S	S	S	S	S	S
L13	R	S	S	R	R	S	S	R	R	S	R	S	S
O9	S	S	R	R	S	I	S	R	S	S	R	S	R
C2	R	S	S	R	S	S	S	S	S	S	R	R	S
O4	R	S	R	R	S	I	R	R	S	I	R	S	S
C3	S	S	S	I	S	S	S	R	S	I	R	R	S
T13	S	S	S	S	S	S	S	S	S	S	S	S	S

KEY: DA Clindamycin; RD Rifampin; AMC Augmentin; F Nitrofurantoin; TEC Teicoplanin; CIP Ciprofloxacin; TE Tetracycline; FOX Cefoxitin; QD Quinopuristin/Dalfostrin;C Chloramphenicol; AMP Ampicillin; SXT Septrin; CN Gentamycin; R resistant; S sensitive.

Table 2: Minimum Inhibitory Concentration of *L. monocytogenes* against the leaf extracts of *Momordica balsamina*

Extract Conc. (µg/mL)	Isolates								
	O4	O9	C3	L5	T13	L13	C2	O14	T9
$MB_{2,000}$	-	-	-	-	-	-	-	-	-
$MB_{1,000}$	+	-	-	-	-	-	-	-	-
MB_{500}	+	+	+	-	-	-	-	+	+
MB_{250}	+	+	+	-	-	+	+	+	+
MB_{125}	+	+	+	+	-	+	+	+	+
$MB_{62.5}$	+	+	+	+	+	+	+	+	+
$MB_{31.25}$	+	+	+	+	+	+	+	+	+
$MB_{15.63}$	+	+	+	+	+	+	+	+	+
$MB_{7.81}$	+	+	+	+	+	+	+	+	+
$MB_{3.91}$	+	+	+	+	+	+	+	+	+
$MB_{1.95}$	+	+	+	+	+	+	+	+	+
$MB_{0.98}$	+	+	+	+	+	+	+	+	+

Key:

MB = *Momordica balsamina,* O4 = Onion 4,O9 = Onion 9, C3 = Cabbage 3, L5 = Lettuce 5, T13 = Tomato 13, L13 = Lettuce 13, C2 = Cabbage 2, O14 = Onion 14 and T9 = Tomato 9

Table 5 depicts the MIC of the test isolates when exposed to a doubling serial dilution of *L. hastata*. Inhibition of visible bacterial growth with this extract was observed in only 3(33.3%) of the 9 isolates at a concentration of 2,000 µg/mL. Only 1(11.1%) of the isolates was inhibited at 1,000 µg/mL concentration, all the lower concentrations showed no inhibitory activity on any of the isolates tested.

When the extract of *O. basilicum* was used to test the MIC of the same 9 isolates in a serial doubling dilution protocol ranging from 2,000-0.98 µg/mL only 1(11.1%) isolate,

T13, was inhibited at a concentration of 2,000 µg/mL. All other concentrations showed no inhibitory effect on any of the isolates (Table 6).

Owing to the high MIC observed in the extracts, particularly with that of *O. Basilicum*, the concentration of the extracts used was increased from 2,000 to 5,000 µg/mL when determining the Minimum Bactericidal Concentration (MBC) of *O. basilicum* extract under investigation. no bacteriocidal activity was observed on any of the 9 isolates with any of the 3 extracts (Table 7).

Table 3: Minimum Inhibitory Concentration of *L. monocytogenes* against the leaf extracts of *L. hastata*

Extract Conc. (µg/mL)	Isolates								
	O4	O9	C3	L5	T13	L13	C2	O14	T9
$LH_{2,000}$	+	-	-	+	-	+	+	+	+
$LH_{1,000}$	+	+	+	+	-	+	+	+	+
LH_{500}	+	+	+	+	+	+	+	+	+
LH_{250}	+	+	+	+	+	+	+	+	+
LH_{125}	+	+	+	+	+	+	+	+	+
$LH_{62.5}$	+	+	+	+	+	+	+	+	+
$LH_{31.25}$	+	+	+	+	+	+	+	+	+
$LH_{15.63}$	+	+	+	+	+	+	+	+	+
$LH_{7.81}$	+	+	+	+	+	+	+	+	+
$LH_{3.91}$	+	+	+	+	+	+	+	+	+
$LH_{1.95}$	+	+	+	+	+	+	+	+	+
$LH_{0.98}$	+	+	+	+	+	+	+	+	+

Key:

LH = *Leptadania hastate*, O4 = Onion 4, O9 = Onion 9, C3 = Cabbage 3, L5 = Lettuce 5, T13 = Tomato 13, L13 = Lettuce 13, C2 = Cabbage 2, O14 = Onion 14 and T9 = Tomato 9

Table 4: Minimum Inhibitory Concentration of *L. monocytogenes* against the leaf extracts of *O. basilicum* used in the study

Extract Conc. (µg/mL)	Isolates								
	O4	O9	C3	L5	T13	L13	C2	O14	T9
$OB_{2,000}$	+	+	+	+	-	+	+	+	+
$OB_{1,000}$	+	+	+	+	+	+	+	+	+
OB_{500}	+	+	+	+	+	+	+	+	+
OB_{250}	+	+	+	+	+	+	+	+	+
OB_{125}	+	+	+	+	+	+	+	+	+
$OB_{62.5}$	+	+	+	+	+	+	+	+	+
$OB_{31.25}$	+	+	+	+	+	+	+	+	+
$OB_{15.63}$	+	+	+	+	+	+	+	+	+
$OB_{7.81}$	+	+	+	+	+	+	+	+	+
$OB_{3.91}$	+	+	+	+	+	+	+	+	+
$OB_{1.95}$	+	+	+	+	+	+	+	+	+
$OB_{0.98}$	+	+	+	+	+	+	+	+	+

Key:

OB = *Ocimum basilicum*, O4 = Onion 4, O9 = Onion 9, C3 = Cabbage 3, L5 = Lettuce 5, T13 = Tomato 13, L13 = Lettuce 13, C2 = Cabbage 2, O14 = Onion 14 and T9 = Tomato 9

Table 5: MBC of *L. monocytogenes* against the leaf extracts of *M. balsamina, O. basilicum* and *L. hastata*

Extract Conc. (µg/mL)			Isolates								
			O4	O9	C3	L5	T13	L13	C2	O14	T9
$MB_{5,000}$	$OB_{5,000}$	$LH_{5,000}$	+	+	+	+	+	+	+	+	+
$MB_{2,500}$	$OB_{2,500}$	LH_{2500}	+	+	+	+	+	+	+	+	+
$MB_{1,250}$	$OB_{1,250}$	$LH_{1,250}$	+	+	+	+	+	+	+	+	+
MB_{625}	OB_{625}	LH_{625}	+	+	+	+	+	+	+	+	+
$MB_{312.5}$	$OB_{312.5}$	$LH_{312.5}$	+	+	+	+	+	+	+	+	+
$MB_{156.25}$	$OB_{156.25}$	$LH_{156.25}$	+	+	+	+	+	+	+	+	+
$MB_{78.13}$	$OB_{78.13}$	$LH_{78.13}$	+	+	+	+	+	+	+	+	+
$MB_{39.06}$	$OB_{39.06}$	$LH_{39.06}$	+	+	+	+	+	+	+	+	+
$MB_{19.53}$	$OB_{19.53}$	$LH_{19.53}$	+	+	+	+	+	+	+	+	+
$MB_{9.77}$	$OB_{9.77}$	$LH_{9.77}$	+	+	+	+	+	+	+	+	+
$MB_{4.88}$	$OB_{4.88}$	$LH_{4.88}$	+	+	+	+	+	+	+	+	+
$MB_{2.44}$	$OB_{2.44}$	$LH_{2.44}$	+	+	+	+	+	+	+	+	+
$MB_{1.22}$	$OB_{1.22}$	$LH_{1.22}$	+	+	+	+	+	+	+	+	+
$MB_{0.61}$	$OB_{0.61}$	$LH_{0.61}$	+	+	+	+	+	+	+	+	+

Key:

MB = *Momordica balsamina*, OB = *Ocimum basilicum*, LH = *Leptadania hastate*, O4 = Onion 4, O9 = Onion 9, C3 = Cabbage 3, L5 = Lettuce 5, T13 = Tomato 13, L13 = Lettuce 13, C2 = Cabbage 2, O14 = Onion 14 and T9 = Tomato 9

Discussion

In this study, isolates of *L. monocytogens* were recovered from 60 samples of vegetables comprising of onions, lettuce, cabbage and tomatoes. This represents an isolation rate of 15.0%. In an earlier study by Weiss and Seeliger (1975), in Germany, the researchers isolated 154 strains of *L. monocytogenes* from soil and plants. Nearly 10% of the corn plants and 13% of the grain plants in the research were positive for *L. monocytogenes***.** However, a more recent report showed that 25.58% vegetable samples were positive for *Listeria* spp. and only one sample (carrot) was positive for *L. monocytogenes* out of 43 samples in total collected from field and greenhouse (Kljujev *et al*., 2018) is present in many animals, including humans, so it is not too surprising that the organism can also be isolated from faeces of these animals, on the land they occupy, in sewage, in soils to which sewage is applied and on plants, including those producing vegetables, which grow in these soils.

The antimicrobial activities of the common herbs/plants (*Ocimum basilicum, Leptadania hastata* and *Momordica balsamina*) was thus investigated. The findings of this study on the phytochemical screening of *Ocimum basilicum* agreed with those reported by several workers. For instance, Sekar *et al*. (2009) reported that the leaves of *Ocimum basilicum* are rich in tannins, flavonoids, cholesterol, terpernoids, glycosides, cardiac glycosides and phlobatannins which were also similar with those found in this research. In fact, the results showed that in all the three samples studied, the qualitative phytochemical analysis of the crude extract appears to be marginally different with one or two bioactive compounds absent and/or present with a varied concentration of tannins, saponins, flavonoids, carbohydrates, alkaloids, phenols, terpinoids, glycosides, cardiac glycosides, cholesterols and phlabotannins. Some of these phytochemicals may be believed to be responsible for high therapeutic potency (including antibacterial, antitoxic etc.).

Moreover, Tannis were reported to possess physiological astringent properties, which hasten wound healing and ameliorate inflamed mucus membrane (Sirinthipaporn and Jiraungkoorskul, 2017). They also have haemostatic properties (de Jesus *et al*., 2012). The presence of saponins in the plant could be responsible for the traditional use in relaxation of muscles and in treatment of wound as practiced generally in Northern Nigeria (Sasidharan *et al.,* 2010). Alkaloids were also reported to have analgesic, anti- inflammatory and adaptogenic activities which help to alleviate pains, develop resistance against diseases and endurance against stress (Gupta and Birdi, 2017). The leaf extracts of *Ocimum basilicum* are rich in various phytochemicals and nowadays these compounds are used tremendously to synthesize drugs for pharmacological actions and are substantial for different foodstuffs (Falowo *et al.,* 2019). And it may be the reason why the traditional birth attendants use the leaves of the plant during the third trimester of pregnancy.

It is also reported that the fruits are common ingredients in Indo Pakistan pickles and are often used in curries and meat dishes. Tender shoots are usually consumed with Okra soup by the Kanuris of Borno State where the plant is locally known as dagdawu. The leaves and fruits were observed to have hypoglycemic effects in rats (Akinniyi *et al.,* 1999; Karumi and Bobbi, 1999). The leaves were found to be highly hemolytic and hepatotoxic in Wistar rats. Furthermore, the fruits were observed to be toxic to various organs and tissues of rats in very high dose as reported by Shettima *et al. (*2001).

In this study, the total phenolic, total flavonoid and proanthocyanidin contents were in the ranges of 17-38, 10-16 and 4-10 mg/g respectively depending on the extraction solvent. In their study, they reported that the acetone extract had highest content of total phenol (35.77 mg/g) than the methanol extract and aqueous extract. The flavonoids content of methanol fraction (15.85 mg/g) is higher than that of acetone and water extracts. The methanol extract

(9.69 mg/g) had highest content of proanthocyanidins compared to water and acetone. In fact, *L. hasata* has been repository of bioactive components and thus found numerous applications traditionally by Africans.

The minimum inhibitory concentration (MIC) of the crude leaf extracts of *Ocimum basilicum, Leptadania hastata and Momordica balsamina* on strains of *Listeria monocytogenes* that showed the highest MIC value to *M. balsamina* is O4 with inhibition of bacterial growth being seen to begin at concentrations starting at 2,000 μgmL^{-1} of the crude extract. The lowest MIC value for the same extract was observed in the isolate labelled with inhibition seen in extraction concentration as low as 156.25 μgmL^{-1}. The findings of this results were not surprising because Aliero and Wara (2009), investigated the effect of *L. hastata* leaf extracts on *Bacillus metagarium, Staphylococcus aureus, Escherichia coli, Salmonella paratyphi* and *Pseudomonas aeruginosa*. Aqueous extract markedly inhibited the growth of *S. paratyphi* and *E. coli* at 30 mg/mL^{-1} and *P. aeruginosa* at 60 mg/mL^{-1}. However, the activity exhibited by the methanol extract was generally low and acetone extract did not show any activity against the tested organisms (Aliero and Wara, 2009).

Moreover, *M. balsamina* exhibited significant antibacterial inhibition activity against pathogenic bacteria as reported by Jigam *et al*. (2004). The extract of *M. balsamina* significantly inhibited *Bacillus subtilis, Staphylococcus aureus, Klebsiella pneumoniae, Escherchia coli, Salmonella typhi, Proteus vulgaris* and *Pseudomonas auruginosa* to varying levels of inhibition (Jigam, 2004; Otimenyin *et al*., 2008). Methanolic extract of *M. balsamina* leaves and stem was also reported to show significant inhibitory property against *B. subtilis, E. coli, P. auruginosa,* and *P. mirabilis* (Otimenyin *et al.,* 2008) and whole plant was very effective against *S. aureus, E. coli, P. auruginosa,* and *Salmonella typhi*. The aqueous and ethanolic extract of *M. balsamina* was active against *Salmonella typhi* as put forward by Akinyemi *et al.* (2005).

Effectively, *M. balsamina* exhibited an overwhelming property against micro-organisms (Bacteria) which may inform us why traditional birth attendants used it during third trimester pregnancy period and can as well be used as an antimicrobial agent against several diseases. The minimum bactericidal concentration of the extracts on different strains of *Listeria monocytogenes* was conducted and presented. The results demonstrated a lack of a bactericidal effect of the extracts up to a concentration of 5,000 µg/mL^{-1}.

Conclusion

All the plants used in this study have shown antilisterial properties against the *Listeria monocytogenes* isolated from vegetables in Sokoto. This was observed from their significant inhibitory activities against the organism. The MBC results revealed that all the extracts have no bacteriocidal effects even at increased concentration to 5000 µg/mL.

Recommendations

Antimicrobial assay of leaf extracts of the plants (*O. basilicum, M. balsamina* and *L. hastata*) should be conducted against other bacterial pathogens associated with pregnancy and childbirth to enable an in-depth analysis of their antimicrobial properties.

NOTE:

The study highlights the efficacy of "traditional medicine" which is an ancient tradition, used in some parts of India. This ancient concept should be carefully evaluated in the light of modern medical science and can be utilized partially if found suitable.

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