

## MULTIPLE DRUG RESISTANCE (MDR) AND EXTENSIVELY DRUG RESISTANCE (XDR) *ESCHERICHIA COLI* ISOLATES FROM DIARRHEIC STOOL AND THEIR SUSCEPTIBILITY PROFILE TO *MORINDA LUCIDA* AQUEOUS LEAF EXTRACT

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### ABSTRACT

**Introduction:** *Escherichia coli* is a Gram negative bacteria. It has been implicated in many cases of resistance recently.

**Aim:** To determine the incidence of MDR and XDR *Escherichia coli* isolates from diarrheic stool and their susceptibility profile to *Morinda lucida* aqueous leaf extract

**Methods:** Diarrheic stool samples were collected from patients at Ahmadu Bello University Teaching Hospital (ABUTH) and Ahmadu Bello University Medical Centre (ABUMC) Zaria, Nigeria. *Escherichia coli* were isolated and identified. The susceptibility of the isolates to the antibiotics were assessed using disk diffusion method. After which multiple drug resistance (MDR) and extensively drug resistance (XDR) *E. coli* were identified. The activity of *Morinda lucida* aqueous leaf extract at different concentrations was tested against the MDR and XDR isolates using agar-well diffusion method.

**Results:** Highest antibacterial efficacy was observed with Ceftriaxone, Cefotaxime, Chloramphenicol, Ciprofloxacin and Gentamicin. While the lowest antibacterial activity was observed with Amoxicillin-Clavulanic acid, Tetracycline, Ampicillin, and Sulphamethoxazole-Trimethoprim. However, for *M. lucida*, the range of average zones of inhibition obtained were 10.00-15.22mm. The higher concentration of *M. lucida* aqueous leaf extract produced better activity than some of the antibiotics used.

**Conclusion:** Antimicrobial resistance is a real challenge to public health. Therefore, measures such as sensitization, surveillance of antimicrobial resistance across different regions and antibiotics policies must be strictly adhered to in curtailing these rising cases of resistance. This study justifies the use of *M. lucida* as an alternative medicine by some members of the populace in the treatment of cases of gut infections.

**Key words:** *Escherichia coli*, multiple drug resistance (MDR), extensively drug resistance, (XDR), *Morinda lucida*.

## INTRODUCTION

Resistance among bacteria now ranks as one of the most significant global health concerns [1], especially since there is a halt over recent years in the development of new antimicrobials [2]. The extensive use of antimicrobials in animals in order to prevent and treat diseases has also contributed to the problem of resistance [3]. Especially, considering the fact that antimicrobials used in animal farming and that of human medicine seems largely similar [4].

The burden of infections caused by these resistant strains of bacteria is enormous. These strains are associated with a higher mortality rate, increased length of hospital stay and increased health costs [5 and 6]. The rapid increase in the spread of these antimicrobial resistant organisms, coupled with the unavailability of effective antimicrobial agents has made the World Health Organization (WHO) to warn against “post-antimicrobial era”, where people die because of common infections and minor injuries [7].

*Escherichia coli* is a Gram negative bacterium from the family Enterobacteriaceae [8]. *E. coli* mainly inhabits the lower intestinal tract of warm-blooded animals, including humans, and is often discharged into the environment through faeces or wastewater effluent [8]. *E. coli* is a common enteric organism and one of the predominant species in most bacterial infections [9].

Most *E. coli* strains live harmlessly in the intestines and rarely cause disease in healthy individuals. Nonetheless, a number of pathogenic strains can cause diarrhea or extra-intestinal diseases both in healthy and immunocompromised individuals. Diarrheal illnesses are a severe public health problem

and a major cause of morbidity and mortality in infants and young children [10]. Low- and middle-income countries in Africa, Asia and Latin America are the most affected regions with diarrheal diseases occurring more often with lethal outcomes mainly due to poor living conditions (inadequate water supplies, poor environmental hygiene and sanitation, and insufficient education) [11]. *E. coli* strains that causes diarrhea have evolved by acquiring, through horizontal gene transfer, a particular set of characteristics that have successfully persisted in the host [12].

*E. coli* are often used as an indicator of fecal contamination of drinking water, and ingestion of water contaminated with *E. coli* can lead to serious complications including diarrhea, enteritis, and mortality in some cases [13, 14, 15].

Antimicrobial-resistant (AMR) bacteria are widespread in environmental waters, and *E. coli* is of great concern as a reservoir for AMR, due to its numerous natures in such environments with a high risk of transfer from the environment to humans [16, 17 & 18]. The quality of drinking water has an important role in human infection and disease. The dissemination of the resistance gene represents a serious public health threat, which causes dangerous limitation of treatment option, and significant mortality rate [19, 20].

The use of medicinal plants has always been part of human culture and is wide spread in Africa [21]. Brimstone tree (*Morinda lucida*) is a medium size tree with short crooked branches, white flowers and shining foliage. Although it is very bitter, yet the whole plants, leaves, stem bark and roots are known to have medicinal properties [22]. The many benefits derived from *M. lucida* is owed to the high contents of vitamins A, K and E, alkaloids and other phytochemicals which are

powerful antioxidant bioactive components like flavonoids which are effective as free radical scavengers, have anti-allergic, anti-inflammatory, anti-viral, anti-proliferative and anti-carcinogenic properties [22]. *M. lucida* is readily available throughout the year in southwestern Nigeria [22]. The decoction is reported to have anti-diarrheal properties and intestinal worm infestation [23, 24 and 22].

Our study therefore determined the susceptibility pattern of *E. coli* to some commonly used antibiotics and aqueous leaf extract of *M. lucida*.

## MATERIALS AND METHODS

### Study Area

The study area was Zaria, Kaduna State, Nigeria. However, the research was conducted in the Department of Pharmaceutical Microbiology, Faculty of Pharmaceutical Sciences, Ahmadu Bello University (A.B.U.), Zaria, Nigeria.

### Collection of samples

Diarrheic Stool samples were collected from Medical Microbiology Laboratory at Ahmadu Bello University Teaching Hospital (ABUTH) and Ahmadu Bello University Medical Centre (ABUMC), Zaria. About 5-10g of stool was scooped into well labeled sterile wide mouth universal sample bottle using sterile plastic spoon. After collection, the samples were transported immediately on ice packs to the department of Pharmaceutical Microbiology laboratory, ABU Zaria for analysis.

### Ethical Consideration

Ethical approval was obtained from the Ahmadu Bello University ethics committee

with the ethical approval reference number ABUTHZ/HREC/F19/2018.

### Microbiological Analysis of Stool Samples

A loopful of liquid stool or fecal suspension was enriched in selenite F broth for 18-24 hours at 37°C and then sub-cultured on MacConkey agar and Nutrient agar. The plates were incubated at 37°C for 24 hours. Further tests were carried out on the resultant isolates. Of which the presumptive colonies (lactose fermenters and non-lactose fermenting organisms on MacConkey agar) were sub-cultured onto selective media Eosin methylene blue (EMB) to get pure cultures which will be stored on Nutrient agar slants, until required. These isolates were preserved by refrigeration for further bacterial identification. All media preparations were carried out according to the manufacturer's instructions and sterilized at 121°C for 15 minutes.

### Identification of the isolates

Gram staining, colony morphology on MacConkey agar, Nutrient Agar, Eosin methylene blue agar and biochemical characterization was used to identify the organisms [25; 26 and 27].

### Biochemical characterization of isolates

All isolates were biochemically characterized using the Microbact and Microgen™ GnA+B - ID System (for some) for Enterobacteriaceae (Microbact 24E Oxoid, UK and Microgen Bioproduct, Surrey, UK). Biochemical tests were performed strictly following the manufacturer's instructions. After which pure cultures of *E. coli* were preserved on nutrient agar slants in Refrigerator.

### Determination of Antibiotic Susceptibility Pattern of the Isolates

Isolates were tested for susceptibility to the following 10 antibiotics, commonly used in humans for the treatment of Gram-negative pathogens; Gentamicin (10 $\mu$ g), Ampicillin (10 $\mu$ g), Ceftriaxone (30 $\mu$ g), Trimethoprim/Sulphamethoxazole (1:19) (25 $\mu$ g), Ciprofloxacin (5 $\mu$ g), Nalidixic acid (30 $\mu$ g), Tetracycline (30 $\mu$ g), Cefotaxime (30 $\mu$ g), Chloramphenicol (30 $\mu$ g) and Amoxicillin/clavulanic acid (20:10). All antibiotic discs were sourced from Oxoid, UK.

Antimicrobial susceptibility testing was performed for all the identified *Escherichia coli* isolates using the disc diffusion method and results interpreted using Clinical and Laboratory Standards Institute (CLSI) [28].

The surface of Mueller Hinton agar was inoculated with 0.1ml of standardized test organism (using micropipette) and evenly spread using sterile swab stick.

The antibiotic discs were then placed gently on the surface of the Mueller Hinton agar plate using disc dispenser (Oxoid, UK) and incubated at 37°C for 24 hours.

### Determination of MDR, XDR and MAR Index of the Isolates

MDR was determined by those organisms that were resistant to one or more of the agents in three or more classes of antimicrobials; While XDR were those that were resistant to all antibiotics tested except two or less [29].

MAR Index was calculated as the ratio between the number of antibiotics that an isolate is resistant to and the total number of antibiotics the organisms were exposed to. MAR Index greater than 0.2 means that the

high-risk source of contamination is where antibiotics are frequently used [30; 31].

### Collection and Identification of plant materials

*Morinda lucida* leaves were obtained in Ibadan and identified at the Herbarium Section, Department of Botany, Faculty of Life Science, ABU, Zaria.

### Preparation of plant materials

The *Morinda lucida* leaves specimen freshly obtained were washed with running tap water and shade-dried at room temperature. The dried plant samples were then grinded into coarse powdered form [32]. The coarse powdered samples were stored in an air-tight container within a desiccator until required for use [33; 34].

### Preparation of crude extracts

The extraction and screening were carried out using the method described by [35 and 32]. Five hundred grams (500 g) of the dried powdered samples of the leaves of *M. lucida* were extracted with 2.5 L of water for 72 hours. Extracts were filtered using muslin cloth and solvents removed through evaporation using water bath at 40°C. The dried extracts were finally transferred into sterile sample bottles for storage at refrigerated temperature until when required for use [34].

### Preparation of Different Concentrations of Plant Extract

One gram (1g) of aqueous extract was added to 5ml of sterile distilled water to give a concentration of 200 mg/ml. Other concentrations of 100, 50, 25, 12.5 and 6.25 mg/ml were prepared by double dilution method as described by Iqbal and Arina [36].

### Determination of Antibacterial Activity of the Extracts with the

Antibacterial susceptibility was tested using the modified Kirby-Bauer disc diffusion method as used by Adeshina *et al.* [37]. The surface of Mueller Hinton agar was flooded with 1 ml of 0.5 McFarland standardized inoculum and allowed to cover the entire surface, after which the excess was discarded into a container of disinfectant. The Mueller Hinton agar surface was allowed to dry, after which wells were bored on the surface of the inoculated agar plates using sterile stainless Cork borer (4mm). The bottom of the wells were sealed with one drop of molten Muller Hinton agar.

Different concentrations of *M. lucida* leaves were prepared. After which, 0.2 ml of the different concentrations of the extract was transferred into the well using micro pipette. The wells were sufficiently spaced to prevent

the resulting zones of inhibition from overlapping. Pre-incubation diffusion time (45 minutes to 1 hour) was allowed, after which the petri-dishes were incubated at 37°C for 24 hrs.

The zones of inhibition surrounding the wells and discs were measured in millimeters using a meter rule [28]. Clear inhibition zones around the wells indicated the presence of antimicrobial activity. The resulting zones of inhibition was recorded as mean  $\pm$  standard error of mean.

### Statistical Analysis

Data obtained in this study were expressed as Mean  $\pm$  Standard Error of Mean (SEM) using SPSS version 20. Differences were considered significant if  $P < 0.05$ .

## RESULTS

Plate I show the pure colonies of *E. coli* on Eosine Methylene Blue (EMB) agar with clear greenish metallic sheen. A total of 258 samples were collected and a total of 237 bacteria were isolated. Figure 1 shows the prevalence of *Escherichia coli* in comparison to other bacteria isolated. With *E. coli* having a prevalence rate of 24%.



Plate I: Pure Colonies of *E. coli* on EMB Agar

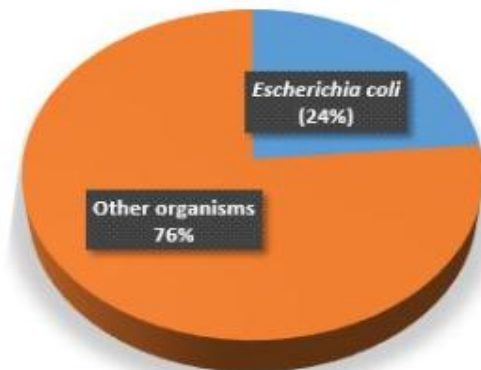


Figure 1: Prevalence of *Escherichia coli*

The isolates were most resistant to Tetracycline, Sulphamethoxazole-Trimethoprim and Amoxicillin-clavulanic acid, followed by Ampicillin, Nalidixic acid, Ciprofloxacin, Cefotaxime but significantly susceptible to Ceftriaxone, Chloramphenicol and Gentamicin (Table 1).

**Table 1: Antibiotic Susceptibility Test of *Escherichia coli* from Diarrhoeic Patients**

S/N	Isolate code	CN	AMP	CRO	SXT	CIP	NAL	TE	CTX	C	AMC	NART
1	T614	S	S	S	S	S	S	S	S	S	S	0
2	T610	S	S	S	S	S	S	R	S	S	S	1
3	T611	S	R	S	S	I	S	R	S	S	I	2
4	S1051	S	R	S	S	S	S	S	S	S	S	1
5	S1044	S	S	S	S	S	R	S	S	S	I	1
6	S1089	S	R	S	R	S	S	R	S	S	R	4
7	S1022	S	S	S	R	S	S	R	S	S	R	3
8	T603	S	S	S	R	S	S	S	S	S	I	1
9	T609	S	S	S	S	S	S	S	S	S	I	0
10	S1003	S	S	S	S	S	R	R	S	R	S	3
11	S1129	S	S	S	S	S	S	R	S	R	R	3
12	T156	R	R	S	R	R	R	R	S	S	R	7
13	T172	S	S	S	S	S	S	S	I	S	R	1
14	T986	S	S	S	S	S	S	S	S	S	I	0
15	T2467	S	R	S	R	S	S	R	S	R	S	4
16	S1087	S	S	S	R	S	S	R	S	S	I	2
17	T0230	S	S	S	S	I	S	R	S	S	S	1
18	S1266	S	R	R	R	R	R	R	R	S	R	8
19	T6000	S	R	S	R	S	S	R	S	I	R	4
20	T0020	S	S	S	S	S	S	S	S	S	R	1
21	T630	S	R	S	R	R	R	R	S	I	R	6
22	S1179	S	R	R	R	R	R	R	R	S	I	7
23	T5726	R	R	S	R	R	S	I	S	S	R	5
24	T211	S	S	S	S	S	S	S	S	S	I	0
25	S1001	S	S	S	R	S	R	R	S	S	S	3
26	T1871	S	S	S	R	S	S	I	S	S	R	2
27	T613	S	S	S	S	S	S	S	S	S	I	0
28	T1628	S	S	S	S	S	S	S	R	S	S	1
29	T602	S	R	S	S	S	S	I	I	S	S	1
30	T197	S	S	S	S	S	S	R	S	S	R	2
31	T187	S	R	S	R	S	R	S	S	S	R	4
32	T2561	S	R	S	R	R	R	R	S	S	R	6
33	T214	S	R	R	R	R	R	R	R	S	R	8
34	T183	S	R	R	R	S	S	R	R	S	R	6
35	S1182	S	R	R	R	R	R	R	R	R	R	9
36	T620	S	S	S	S	S	S	R	S	S	S	1
37	S1090	S	R	S	S	S	S	R	S	S	R	3
38	T2648	S	R	R	S	R	R	S	R	S	R	6
39	T229	S	R	S	R	R	R	I	S	R	R	6
40	S1434	S	S	S	S	S	S	R	S	S	S	1

41	T2507	S	S	S	S	S	S	S	S	S	I	0
42	T2693 <sub>a</sub>	S	R	S	R	S	I	S	S	S	R	3
43	T2589	S	S	S	R	S	R	R	S	S	R	4
44	T228	S	R	R	R	R	R	R	R	S	R	8
45	T2475	S	R	S	R	S	R	S	S	S	R	4
46	T2693 <sub>b</sub>	S	S	S	R	S	S	R	S	S	I	2
47	T5696	S	R	S	R	S	S	R	S	S	R	4
48	S1476	S	S	S	S	S	S	S	S	S	S	0
49	T2610	S	S	S	S	S	S	S	S	S	I	0
50	S1445	S	S	S	S	S	S	S	S	S	I	0
51	T2587	S	S	S	S	S	S	S	S	S	I	0
52	T0636	S	S	I	R	S	S	R	S	S	R	3
53	T597	S	S	S	R	S	S	I	S	S	S	1
54	T151 <sub>b</sub>	S	R	R	R	R	I	R	R	S	R	7
55	T2458	S	R	R	R	R	R	R	R	S	R	8
56	S1310 <sub>a</sub>	S	R	R	R	R	R	R	R	S	R	8
NRPA		2	26	10	30	14	18	31	11	5	29	

Key: S = Sensitive, I=Intermediate, R = Resistant, CN=Gentamicin, AMP = Ampicillin, CRO = Ceftriaxone, SXT = Sulphamethoxazole-Trimethoprim, CIP = Ciprofloxacin, NAL = Nalidixic acid, TE = Tetracycline, CTX = Cefotaxime, C = Chloramphenicol and AMC = Amoxicillin-clavulanic acid, NRPA = Number of *E. coli* resistant to each antibiotics

The results of antibiotics susceptibility test shows that the average zones of inhibition (mm) within the area studied to the antibiotics tested were Gentamicin (21.75), Ampicillin (14.80), Ceftriaxone (28.98), Sulphamethoxazole-Trimethoprim (15.39),

Ciprofloxacin (23.59), Nalidixic acid (18.71), Tetracycline (14.25), Cefotaxime (28.34), Chloramphenicol (24.16) and Amoxicillin-Clavulanic acid (13.55) (Table 2).

**Table 2: Average Zone of Inhibition (mm) using Standard Antibiotics**

Organism	CN	AMP	CRO	SXT	CIP	NAL	TE	CTX	C	AMC
	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D	Mean ± S.D
<i>Escherichia coli</i>	21.75 ± 3.96 <sup>bc</sup>	14.80 ± 7.98 <sup>ab</sup>	28.98 ± 8.86 <sup>ab</sup>	15.39 ± 10.4 <sup>a</sup>	23.59 ± 10.4 <sup>ab</sup>	18.71 ± 8.62 <sup>abc</sup>	14.25 ± 7.73 <sup>a</sup>	28.34 ± 8.88 <sup>ab</sup>	24.16 ± 6.75 <sup>a</sup>	13.55 ± 5.08 <sup>a</sup>

Key: S.D = Standard deviation, CN = Gentamicin, AMP = Ampicillin, CRO = Ceftriaxone, SXT = Sulphamethoxazole-Trimethoprim, CIP = Ciprofloxacin, NAL = Nalidixic acid, TE = Tetracycline, CTX = Cefotaxime, C = Chloramphenicol and AMC = Amoxicillin-clavulanic acid.

To all the isolates tested, the high the concentrations in mg/ml of the *Morinda lucida* aqueous extract the higher the

susceptibility of the isolates which reflects on their zone of inhibition.

**Table 3: Antibacterial Susceptibility Result using *Morinda lucida* Aqueous Extract (Concentrations in mg/ml)**

S/N	Isolate code	200	100	50	25	12.5	6.25	C	Remark for isolates as per antibiotics used
1	S1089	12	11	10	10	10	10	10	MDR
2	S1022	14	13	12	10	10	10	10	MDR
3	S1003	17	15	13	11	10	10	10	MDR
4	S1129	15	13	12	10	10	10	10	MDR
5	T156	15	13	12	11	10	10	10	XDR
6	T2467	12	11	10	10	10	10	10	MDR
7	S1266	15	13	11	10	10	10	10	XDR
8	T6000	15	13	11	10	10	10	10	MDR
9	T630	13	11	10	10	10	10	10	MDR
10	S1179	17	13	12	11	10	10	10	MDR
11	T5726	13	12	11	10	10	10	10	MDR
12	S1001	16	14	13	11	10	10	10	MDR
13	T187	16	13	12	10	10	10	10	MDR
14	T2561	14	12	10	10	10	10	10	MDR
15	T214	13	11	10	10	10	10	10	XDR
16	T183	16	14	12	11	10	10	10	MDR
17	S1182	14	13	11	10	10	10	10	XDR
18	T2648	19	16	13	12	11	10	10	MDR
19	T229	15	13	11	10	10	10	10	MDR
20	T2589	16	13	12	11	10	10	10	MDR
21	T228	13	11	10	10	10	10	10	XDR
22	T2475	16	14	12	11	10	10	10	MDR
23	T5696	21	17	15	13	10	10	10	MDR
24	T0636	15	13	12	11	10	10	10	MDR
25	T151 <sub>b</sub>	14	12	11	10	10	10	10	MDR
26	T2458	17	14	13	12	10	10	10	XDR
27	S1310 <sub>a</sub>	18	15	13	11	10	10	10	XDR

**Keys:** MDR=Multiple Drug Resistance, XDR=Extensively Drug Resistance, C=Control.

The zones of inhibition obtained with the aqueous leaf extracts of *M. lucida* range from 10.00 – 15.22 (Table 4).

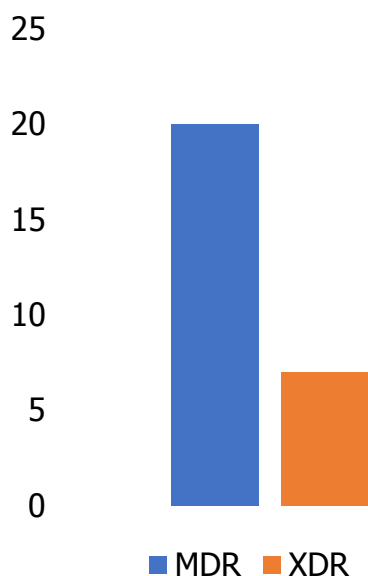
**Table 4: Average Zone of Inhibition (mm) using the Morinda Aqueous Extract**

Organism	Mean $\pm$ S.D in mg/ml						
	200	100	50	25	12.5	6.25	C
<i>Escherichia coli</i>	15.22 $\pm$ 2.10 <sup>a</sup>	13.07 $\pm$ 1.52 <sup>a</sup>	11.63 $\pm$ 1.25 <sup>a</sup>	10.59 $\pm$ 0.80 <sup>a</sup>	10.04 $\pm$ 0.19 <sup>a</sup>	10.00 $\pm$ 0.00 <sup>a</sup>	10.00 $\pm$ 0.00 <sup>a</sup>

Key: S.D=Standard deviation, C=Control.

Twenty 20 (35.7%) of the *E. coli* isolates were multidrug resistance while 7 (12.5%) were extensively drug resistant (Fig. 2.0).

Thirty-four (34) (60.7%) of the isolates had their MAR Index greater than or equal to 0.2 ( $\geq 0.2$ ) while 22 (39.3%) had their MAR Index less than 0.2 (Table 5)



**Figure 2: Multidrug Resistant (MDR) and Extended Drug Resistant (XDR) *Escherichia coli***

**Table 5: Multiple Antibiotic Resistance Index of the Isolates**

S/N	MAR Index	Frequency	Percentage MARI	
1	0.0	10	17.9	} 39.3
2	0.1	12	21.4	
3	0.2	05	8.9	} 60.7
4	0.3	07	12.5	
5	0.4	07	12.5	
6	0.5	01	1.8	
7	0.6	05	8.9	
8	0.7	03	5.4	
9	0.8	05	8.9	
10	0.9	01	1.8	
<b>Total</b>		<b>56</b>	<b>100</b>	

## DISCUSSION

The prevalence of *E. coli* varies from one region to another. The prevalence rate of *E. coli* in this study was found to be 24%. A contrasting prevalence was obtained by Cho *et al.* [38]; they observed from their studies 99.3% (455/458) of the total samples to be positive for *E. coli*. Ekenyia *et al.* [39] observed 45.52% of *E. coli* in their studies. The differences in the prevalence rate could be as a result of differences in location, sample source and size. These could influence the distribution, and ultimately the recovery rate of *E. coli* from the samples.

High level of resistance was observed with Amoxicillin-Clavulanic acid, Tetracycline, Ampicillin, and Sulphamethoxazole-Trimethoprim which could be due to antibiotics misuse. However, the low level of resistance to Ceftriaxone, Cefotaxime, Chloramphenicol, Ciprofloxacin and Gentamicin could be due to the cost of these antibiotics and method of administration. Other studies showed similar results to the ones obtained in this study; for instance, in a study by Mahmoud *et al.* [40], *E. coli* isolates showed a high rate of tetracycline resistance (97.7%), followed by gentamicin (57.7%), ciprofloxacin (46.6%), ceftriaxone (35.5%), and chloramphenicol (31.1%). In contrast to the result in this study, Mapanguy *et al.* [41] reported low resistance to Amoxicillin-Clavulanic acid (1%) and resistance to Ceftazidime (65%); Manishimwe *et al.* [42] reported 56.8% resistance to Ceftriaxone and 54.5% resistance by *E. coli* to Ciprofloxacin. Differences could be as a result of location and rate of antibiotics misuse.

Resistance obtained against some of the antibiotics could be as a result of abuse of antibiotics which includes self-medication, purchase of over-the-counter drugs and

antibiotics sold by the road side, which was indicated by 34 isolates (60.7%) having MAR Index of  $\geq 0.2$ . Multiple Antibiotics Resistance Index  $\geq 0.2$  originates from a high-risk source of contamination where several antibiotics are frequently used [30; 31].

The higher concentrations of *M. lucida* aqueous leaf extract produced better activity than the lower concentrations on the MDR and XDR isolates. High level of resistance was obtained against the lower concentrations of the extract. Other studies however were in contrast to this. Studies by Fakoya *et al.* [43] showed that *M. lucida* extracts were more active against all the tested bacteria than the standard antibiotics, chloramphenicol and ciprofloxacin even at the same concentrations of 5, 10 and 20 mg/ml. The results of their study showed that the extracts of *M. lucida* has the potentials of inhibiting the growth of *E. coli* and *Salmonella* species.

## CONCLUSION

High prevalence of multiple drug resistance *E. coli* was observed in this study. This has a very significant implications on human health. This could lead to prolong hospital stay (as samples were obtained from hospitals), treatment failure and mortality. So, measures such as sensitization, surveillance of antimicrobial resistance and antibiotics policies must be strictly adhered to in curtailing this rising case of antibiotic resistance.

*M. lucida* aqueous leaf extract produced some level of activity against some of the MDR *E. coli* isolates tested. This justifies the use of this plant as an alternative medicine in the treatment of some diarrheal illnesses.

**Authors Contribution:**

GOA, JAO and AMM supervised the Research, MMB and IJC did the research and assisted by SMS and VAO. All authors read through the manuscript and consented to its publication.

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