Acta Traditional Medicine
Volume 01, Issue 02, 2022, (Page no.13-37)

Journals url:
http://traditionalmedicine.actabotanica.org/

COMPARATIVE EFFECTS OF PREPARED AND VENDED HERBS ON SELECTED DIARRHOEGENIC BACTERIA IN EKITI STATE

OMOYA F. O[1]. AND AJIMOJUOWO F. B[1,2].*

[1]Department of Microbiology, Federal University of Technology, Akure, Ondo State, Nigeria

[2] College of Health Science and Technology, Ijero Ekiti, Ekiti State, Nigeria

Corresponding author: ojobolaoluwatosin01@gmail.com

ABSTRACT

Herbs contamination by some pathogenic bacteria poses a major threat to the usage as a treatment option for diarrheagenic bacteria. This study was carried out to determine the comparative effects of prepared and vended herbs sold within Ado-Ekiti metropolis on selected diarrhogenic bacteria. Herb was prepared in the laboratory under a strict aseptic condition, and a total of 50 vended herbs samples were purchased from vendors across ten different locations within Ado-Ekiti, Ekiti State. All herb samples were analyzed using standard microbiological techniques, total bacteria count, Staphylococcal counts, coliform counts and fungal counts were carried out. Bacteria colonies on the agar plates were identified using morphological and biochemical characterization. The susceptibility testing of the vended and prepared herbs on selected diarrheagenic bacteria; *Shigelladysenteriae*, *Escherichiacoli* and *Salmonellatyphi* were carried out using agar well diffusion method. The haematological and histological effects of the herbs were done using standard haematological and histological procedures respectively. The data generated were analysed using analysis of variance (ANOVA) and the level of significance was set at 0.05. There was a low microbial count in the prepared herb compared to the vended herbs, Staphylococcal counts, coliform counts and fungal counts of the vended herbs ranged from 15.50±0.65 to 52.00±0.36 cfu/100ml, 7.00±0.40 to 22.00±0.13 cfu/100ml, 8.50±0.15 to 36.50±0.20 cfu/100ml and 1.50±0.50 to 8.00±0.40 sfu/100ml respectively while the total bacterial counts of prepared herbs ranged from 1.00±0.01 to 2.00±0.00 cfu/100ml. There was a low microbial count in the herbs vended by those with more than 9 years herb vending experiences compared to others. Nine different bacteria species were isolated from vended herb samples with *Escherichiacoli* and *Staphylococcus aureus* 8(22.2%) being the most frequently occurred bacteria and followed by *Enterobacter aerogenes* and *Pseudomonas aeruginosa* was4(11.11%) occurrence. The laboratory prepared herbs exhibited a better antibacterial activity against the tested diarrheagenic bacteria (*S.dysenteriae* (6.00±0.00 mm), *E.coli* (10.00±0.10 mm) and *S.typhi* (8.00±0.40 mm) compared to the vended herbs. The bacteria isolated from the vended herbs showed higher resistant against augmentin and amoxicillin. The haematological

profile of the rats showed a significant increase in the white blood cell counts. There were also multiple foci of hepatic necrosis with cellular aggregation around the same foci in the liver and diffuse tubular degeneration and necrosis with the tubular epithelium completely eroded in the kidney of the rat. However, occurrence of these bacteria in vended herbs could predispose consumers to the risk of infections with pathogenic and antimicrobial resistant bacteria. Also, the herbs could be a source of toxicity showed in both histological and haematological parameters, which could pose a life-threatening disease to the consumer. Therefore, steps involve in the preparation of the herb must be standardize to prevent the microbial contamination.
Keywords: VENDED HERBS, DIARRHOEGENIC, BACTERIA, EKITI-STATE, ANTIBIOTICS

Introduction

Herbs are plants with savory or aromatic properties that are used for flavoring and garnishing food, medicinal purposes, or for fragrances; excluding vegetables and other plants consumed for macronutrients. Culinary use typically distinguishes herbs from spices. Herbs generally refers to the leafy green, stem and roots or flowering parts of a plant (either fresh or dried) (Dillehay *et al*., 2014).

Herbal medicines (HM) are plant-derived materials or plant extracts which are used as therapeutic substances (Meshack *et al*., 2013) and can also be used as dietary supplements. World Health Organization (WHO) (2011) defined Herbal Medicines Product (HMP) as botanical medicine or phytomedicine. WHO (2008), describes traditional Herbal Medicines Product (HMP) as herbs, herbal materials, herbal preparations, and finished herbal products that contain parts of plants or other plant materials as active ingredients that used for prevention or treatment of different ailments. The usage of HMP in prevention and treatment of human diseases has potentially maintained its popularity worldwide (WHO, 2017).

There are many different methods of preparing medicinal herbs. Although, there are no really any set rules for the preparation of herbs (Abubakar and Haque, 2020). The common methods are: Drying method, Decoction method, Macerations method, Oil Infusions, Teas, Tinctures,andPowder.

The general benefit of herbs include: Strengthen the Immune System, Anti-inflammatory Properties, Reduce Blood Sugar & Cholesterol Levels, Prevent Alzheimer's Disease, Prevent Cancer, Skin care, Hair care and Dental care. Other benefit of herbs include carminative (cure gastric issues), diaphoretic (control water retention), lipolytic (help in weight loss), anti-spasmodic, analgesic (relieve pain), deodorant, aphrodisiac (increase sexual drive), antiseptic, digestive, and stimulant when taken in correct dosage (Allaby, 2012).

In Nigeria, there has been a remarkable increase of public awareness and usage of herbal medicinal products (HMP) in the treatments and/or prevention of diseases accompanied by a lot of advertisement in the newspapers, radios, billboards and television media. The contribution of herbs cannot be neglected in the society since they are easily accessible health care options for people with limited financial resources (WHO, 2007). Modern medicine is well developed in most parts of the world but the problem of antibiotic resistance worldwide poses a major threat to the effective treatment of bacteria infection particularly diarrhogenic bacteria (Iguchi *et al*., 2015). Nevertheless, a large number of people in developing countries especially in rural area, usually perceived herbs as being natural and therefore safe, but they are not necessarily free from microbial and other contaminants which are due to factors such as adulteration, substitution,

cross contamination, lack of standardization, inappropriate use of antimicrobials and poor handling practices and/or poor storages (Jonas, 2015).

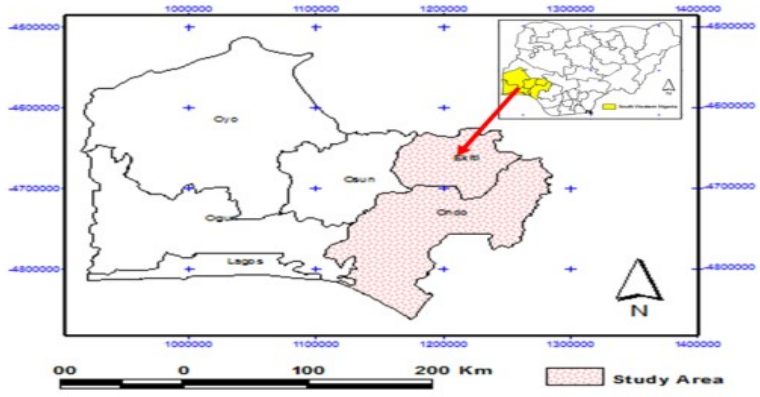
Diarrheagenic bacteria are bacteria that can cause diarrheal diseases (WHO, 2017). Diarrhoea is defined as the passage of three or more loose or liquid stools per day (or more frequent passage than is normal for the individual). Frequent passing of formed stools is not diarrhoea, nor is the passing of loose, "pasty" stools by breastfed babies (WHO, 2017). Diarrhoea is usually a symptom of an infection in the intestinal tract, which can be caused by a variety of bacterial, viral and parasitic organisms. Infection is spread through contaminated food or drinking-water, or from person-to-person as a result of poor hygiene (WHO, 2017). The main bacterial pathogens causing diarrhea are diarrheagenic *Escherichia coli* (DEC) that consists of enteroaggregative *E. coli* (EAEC), enteropathogenic *E. coli* (EPEC), enterotoxigenic *E. coli* (ETEC), enterohemorrhagic *E. coli* EHEC and enteroinvasive *E. coli* (EIEC), *Salmonella*, *Shigella* spp. (S. *dysenteria, S. sonnei, S. flexneri*) *Campylobacter* (*C. coli, C. jejuni*), *Vibrio* (*V. vulnificus*, *V. parahaemolyticus, V. cholerae*), *Yersinia enterocolitica* and *Aeromonas hydrophila* (Sjoling *et al*., 2015).

Despite the fact that many studies conducted have shown beneficial/antibacterial effects of some herbs, there is still little information about vended herbs and laboratory prepared herbs under aseptic condition. It is therefore becomes necessary to compare the effect of prepared and vended herbs on selected diarrhogenic bacteria. This study then aimed at to compare the antibacterial effects of the prepared and vended herbs in Ado-Ekiti on selected diarrhogenic bacteria.

3.0 Methodology

Study Location

This study was conducted at Ado Ekiti and Akure, Nigeria. Ekiti State was created from a portion of Ondo State in 1996 and has Ado-Ekiti as its capital. The state is named for the Ekiti people, a Yoruba subgroup that makes up the bulk of the state's population.



Geographical relationship between study area (Ekiti State).

(Encyclopedia Britannica, 2021)

Study Design

A cross-sectional descriptive study was conducted which involved the purchase of vended herbs for laboratory analyses. In addition, a checklist was used to collect information on herbs handling practices, water used for preparation packaging materials, and state of the product collected from the vendors.

Sample Size

The sample size for this study was calculated by usingthe formula for unknown population, by Kothari (2004).

$$n=Z^2SD^2/e^2,$$

where Z=standard error from the mean,≈1.96 at 95% confidence interval; standard deviation(SD) ≈0.180 or 18.0%; and e=tolerable samplingerror/precision, ≈0.05 at 95% confidence interval.Then, the sample size was calculated as:

$$n= \frac{[(1.96)^2 \times (0.180)^2]}{(0.050)^2} = 50 \text{ samples}$$

Therefore 50 vended herbs samples were collected from vendors across different locations within Ado Ekiti. The samples were transported to the laboratory for analysis. For the Prepared herbs, the raw herbs used for the production of the vended herbs were gotten from the farm and they were prepared using the same method the vendor used for their preparation.

Questionnaire

A structured questionnaire was used to assess possible factors for bacterial contamination in Herbs vended in Ado metropolis.

Collection of Vended herbs

About 5mls of vended herbs sample was purchased into a sterile bottle and was transported to the laboratory as soon as possible (Jonas, 2015).

Sources of the test bacteria

Diarrheagenic bacteria (*Escherichia coli, Shigella dysenteria,* and *Salmonella typhi*) were gotten from the microorganism bank of Ekiti State University Teaching Hospital, Ado Ekiti.

Preparation of Herbs

Laboratory prepared herbs was done under aseptic condition by following the same method used by the vendor. Decoction method of herb preparation was used for preparation of herbs in this study (Abera, 2014). Continuous hot extraction utilizing a predetermined amount of water as a solvent is what this procedure entails. Plant matter that has been dried, ground, and powdered is put into a spotless container. After that, water is added and mixed. Then, to speed up the extraction, heat is used throughout the operation (Ingle *et al*., 2017, and Majekodunmi, 2015). The procedure just takes a few minutes, typically around 15 of them. Typically, the solvent (alcohol) to crude drug ratio is 4:1 or 16:1. It is used to extract plant material that is both heat- and water-soluble (Ingle *et al*., 2017, and Majekodunmi, 2015).

Determination of total plate counts (TPC) and total coliform counts (TCC)

The total plate count was done on nutrient agar while total coliform count was done on MacConkey agar. The membrane filtration technique was used to determine the plate counts. In this technique, 5mls sample is passed through the membrane using a filter funnel and vacuum system. All of the organisms in the sample are concentrated on the membrane's surface. After

that, the membrane is placed in a specific plate with a pad that has been saturated with the right medium and the trapped bacteria. The formation of colonies of organisms on the upper side of the membrane is facilitated by the transfer of nutrients through the filter during incubation (Devika *et al*., 2019). The colonies thus formed can be transferred to confirmation media.

Morphological and Biochemical Characterization of Bacteria

The pure culture of each isolate was examined. Microscopic examination, staining reactions and biochemical tests were carried out on the isolates according to the methods described by Cheesbrough*et al.* (2000). They were Gram staining, Motility test, Catalase test, Oxidase test, Citrate utilization test, Methyl Red Test, Voges Proskauer Test, Indole test, Hydrogen sulphide (H_2S) production, Sugar Fermentation Test.

Determination of the antibacterial activities of the diarrhogenic bacteria

Evaluation of antibacterial activities of the both Prepared and Vended herbs were performed on Mueller Hinton Agar (MHA) using agar disc diffusion method according to CLSI (2012).

Collection of Herb Samples

Herb samples were collected from different vendors (five vendors in a park) three times a week between October, 2019 and December, 2019 from randomly selected 10 motor parks namely; Odo-Ado, Ajilosun, Basiri, Old garage, Okesa, Orita blessing, New grage, Olaniyi, Lagos and Abuja park in Ado Ekiti, Ekiti State, Nigeria.

Antimicrobial susceptibility Profiles of isolated Bacteria

Antibiotic susceptibility profile of bacteria was determined by the disc diffusion method with the use of Mueller-Hinton agar, according to the Bauer-Kirby method (Cheesbrough, 2014). The following clinical antibiotics, with their concentrations given in parentheses were used in the antibiograms as recommended by Committee for Clinical Laboratory Standards (2019), for gram negative bacteria; Tetracycline (30µg), Ofloxacin (30µg), Gentamicin (20µg), Chloramphenicol (30µg), Augmentin (30 µg), Ceftriazone (30 µg), Nitrofuration (300 µg), Cotrimozazole (25 µg), Ciprofloxacin (10 µg) and Amoxicillin (30µg) while gram positive was tested against Cotrimozazole (25 µg), Erythromycin (10µg), Gentamicin (20µg), Augmentin (30 µg),Streptomycin (10 µg), Cloxacilin (5 µg) Tetracycline (30µg) and Chloramphenicol (30µg). Multidrug resistance was defined in this study as resistance to more than one antibiotics tested.

Antibacterial potency of the herb *in vitro* on selected bacterial pathogens

The antibacterial activities of herbs wereaccessed as described by Oluyege and Adelabu (2010), with slight modification. The sterile petri dishes were filled with 25ml of Muller Hinton agar and allowed to solidify. Prior to streaking the plates with bacterial culture (*Shigella dysenteriae, Salmonella typhi, Escherichia coli, Staphylococcus aureus* and *Bacillus cereus*), 5mm diameter wells were punched in the medium using a sterile borer. The bacterial cultures were inoculated by spreading in the petri plates using sterile cotton swabs. Then 0.1ml of the herb sample was directly introduced into the well made on the surface of Muller Hinton agar containing bacterial lawn. The inoculated plates were incubated overnight at 37^{o}Cand the diameter of zone of inhibition was measured in mm.

Source of Experimental Wistar Albino rats

Experimental rat were obtained from animal house, Department of Microbiology, School of Sciences, Federal University of Technology, Akure and kept in a wooded cage for 14 days for acclimatization under standard environmental condition.

Collection of blood sample

This was done according to the method used by Cheesbrough (2014). At the end of the treatment period, rats were anaesthetized using chloroform vapour prior to dissection. Blood samples were

collected by motordiac puncture into two different tubes, one containing anticoagulant [ethylene diamine tetraacetic acid (EDTA)] and the other without anticoagulant. The blood was used for the determination of haematological parameters.

Haematology Analysis

Packed cell volume (PCV) was determined by microhaematocrit technique using capillary tubes. Red blood cell counts (RBC) and white blood cells were determined as described by Cheesbrough (2014). Differential leucocytic counts (DLC) and the haemoglobin (Hb) concentration (determined using cyanomethaemoglobin method). Haematological indices such as the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated from determined values of PCV, RBC and Hb as described by Ochei and Kolhakltar (2000).

Toxicological effects of the Herbs on selected Organs in Albino rat

Cross sections of heart, kidney and liver tissue were prepared and analyzed using conventional techniques described by Cheesbrough (2014). After sacrificing the animals, small pieces of liver were fixed in 10% formalin, dehydrated in ascending grades of alcohol and cleared in xylene. The fixed tissue were embedded in paraffin wax and sectioned into five micrometres thick with the rotary microtome, then stained with haematoxylin and eosin. Then the sections were examined with light microscope and photographed using a microscopic camera.

Results

Socio demographic characteristic of the herb vendors at selected motor park in Ado Ekiti, Ekiti-State.

The socio demographic characteristic of the herb vendors at selected motor park in Ado Ekiti State is shown in Table 1. There were 10 different motor parks selected for this study. The distribution of the parks in relation with the gender showed that all the vendors in the various parks selected were female with overall distribution rate of 50 (100.0%). The age distribution of the vendor showed that Age groups 31-40years had the highest number of vendor with distribution rate of 23 (46.0%). However, the level of education of the vendor showed that vendor with secondary school had the highest distribution of 45 (90.0%) compare with their primary school counterpart with 5 (10.0%) distribution rate.

Comparative Microbial contamination of vended and prepared herbs at different motor parks in Ado-Ekiti, Ekiti State.

The comparative microbial contamination of vended and prepared herbs at different motor parks in Ado Ekiti is shown in Table 2. It was observed that herbs vended at Lagos motor park had the highest mean total viable bacterial count of 52.00± 0.36cfu/100ml, followed by Okesa park, Odo-Ado park, Basiri park, Abuja park, New Garage, Orita blessing, Olaniyi park, Ajilosun park and Old garage with the mean total viable counts of 38.00±0.34cfu/100ml, 37.00±0.21cfu/100ml, 24.50±0.95cfu/100ml, 24.00±0.70cfu/100ml, 24.00±0.11cfu/100ml, 20.50±7.50cfu/100ml, 19.00±0.50cfu/100ml, 15.50±0.65cfu/100ml and 16.50±0.10cfu/100ml respectively.

Okesa park had the highest *Staphylococcus* count of 22.50±0.13cfu/100ml, followed by new garage with mean staphylococcal count of 16.50±0.56 cfu/100ml, while Lagos park, Ajilosun park, Odo-Ado park, Old garage park and Olaniyi park had the mean Staphylococcal counts of 15.00±0.30cfu/100ml, 14.00±0.30cfu/100ml, 12.50±0.15cfu/100ml, 9.50±0.25cfu/100ml, 8.00±0.15cfu/100ml and 7.00±0.40cfu/100ml respectively. No *Staphylococcus* species was isolated in Abuja Park and Orita blessing park. The mean coliform count showed that Okesa Park had the highest mean coliform count of 36.50±0.20cfu/100ml, while motor parks Odo-Ado,

Ajilosun, Basiri, Old garage, Orita blessing, new garage, Olaniyi parks, and Lagos parks had the mean total coliform count of 16.50±0.45cfu/100ml, 11.00±0.40cfu/100ml, 8.50±0.50cfu/100ml, 13.00±0.00cfu/100ml, 16.50±0.50cfu/100ml, 8.50±0.15cfu/100ml, 18.50±0.95cfu/100ml respectively. No coliform was isolated in herbs vended at Abuja parks. The total fungal count of the herbs showed that Lagos parks had the highest mean fungal count of 10.50±0.35sfu/100ml, while parks Odo-Ado, Ajilosun, Basiri, Old garage, Okesa, Orita blessing, New garage, Olaniyi parks and Abuja parks had the mean fungal count of 3.00±0.10sfu/100ml, 4.00±1.00sfu/100ml, 8.00±0.20sfu/100ml, 8.00±0.40sfu/100ml, 7.50±0.50sfu/100ml, 8.00±0.14sfu/100ml, 3.50±0.15sfu/100ml, 3.00±0.00sfu/100ml, 1.50±0.50sfu/100ml respectively. However, the prepared herb had the mean total viable bacterial count of 2.00±0.00cfu/100 ml and mean total fungal count of 1.0±0.01sfu/100 ml. No staphylococcal and coliform counts were seen in the prepared herbs.

Effect of vendors age, level of education, household number, year of job experience, tribe on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State

Tables 3, 4, 5, 6 and 7 shows the effects of vendors' age, level of education, household number, year of job experience, tribe on microbiological quality of the herb vendors at selected motor parks within Ado Ekiti. It revealed that Age groups 32-40 years had the highest mean total viable bacterial count of 30.60±0.68cfu/100 ml.The vendor with secondary school education had the highest mean total viable bacterial count, mean staphylococcal count and the mean total fungal counts of 28.56±0.38sfu/100ml, 10.94±1.99sfu/100ml and 5.77±0.97sfu/100ml respectively. The mean total coliform count of vendor with primary education is higher than the secondary with mean count of 14.00±3.00cfu/100ml to 12.61±0.57cfu/100ml in secondary school educated. Vendors with house household size 3,4,5 and 6 had the mean total viable bacterial count of 4.00±0.00cfu/100ml, 31.20±0.63cfu/100ml,18.33±0.18cfu/100ml and 28.67±0.12cfu/100ml respectively. The vendor with job experience of 9yrs and above had the least mean count for total viable bacterial, total staphylococcal count, total coliform count and total fungal count of 16.60±0.37sfu/100ml, 3.00±0.00sfu/100ml, 5.00±0.00sfu/100ml and 1.00±0.00sfu/100ml respectively, followed by those with 5 to 8yrs experience with mean total viable bacterial count, total staphylococcal count, total coliform count and total fungal count of 24.00±0.00cfu/100ml, 9.40±0.23cfu/100ml, 9.60±0.25cfu/100ml and 4.20±0.10cfu/100ml respectively. Yoruba tribes had the highest means of total bacterial count, total staphylococcal count and total fungal count of 27.89±0.53cfu/100ml, 11.00±1.89cfu/100ml and 5.84±0.92sfu/100ml compared into their Hausa counterpart with mean of total bacterial count, total staphylococcal count and total fungal count of 13.00±0.00cfu/100ml, 0.00±0.00cfu/100ml and 3.00±0.39sfu/100ml.

Comparative Antibacterial potency of prepared and vended herbs at different motor parks in Ado-Ekiti on selected diarrheagenic bacteria

The vended herbs sold in Odo Ado, New garage, Lagos park, Abuja park had mean sensitivity to*Shigella dysenteriae* of 0.50± 0.05mm, 5.00 ±0.00mm, 8.50±0.70mm, 2.50±0.20mm respectively. *Escherichia coli* sensitivity with the mean sensitivity of 4.50±0.15mm, 1.01±0.00mm, 1.50±0.20mm from the vended herbs sold in Old garage, Orita blessing, New garage, Lagos park and Abuja park respectively. Herbs from Ajilosun, Basiri, Odo-Ado, Okesa and Olaniyi motor parks were not sensitive to the *E. coli*. However, *Salmonella typhi* was sensitive to herbs from Odo-Ado, Basiri , Old garage, Orita blessing, Olaniyi park, Lagos park with mean sensitivity of 2.50±0.15mm, 2.50±0.50mm, 0.50±0.10mm, 2.50±0.50mm,

7.0±0.00mm, 4.50±0.50mm respectively, other herbs from parks like Ajilosun, Okesa, New garage and Abuja parks were not sensitive to *Salmonella typhi*.

The prepared herb shows the mean sensitivity of 6.00±0.00mm, 10.00±0.10mm and 8.00±0.40mm to *Shigella dysenteriae, Escherichia coli and Salmonella typhi* respectively. The Ciprofloxacin antibiotics was use as a positive antibiotic control showed a high mean sensitivity of 21.04±.77mm, 26.46±0.41mm and 24.03±0.11mm to *Shigella dysenteriae, Escherichia coli and Salmonella typhi* respectively.

Effect of vendors' year of job experience, level of education, age, tribe on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria

Table 9, 10, 11 and 12 shows the effect of vendors' year of job experience, level of education, age, tribe on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria. The herbs prepared by the vendor with the job experience of 5 to 8 years had the mean sensitivity of about 3.60±0.56mm,0.80±0.04mm and 2.40±0.15mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively, while herb produced by those vendors with job experience of less than 1year had the mean sensitivity of 0.25±0.02mm, 1.25±0.75mm and 2.00±0.91mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively. Also, herbs produced by the vendor with 9 years and above experience had the mean sensitivity of 0.00±0.00mm, 1.09±0.04mm and 7.00±0.01mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively.

The herbs produced by the vendor with primary education had the mean sensitivity of 4.00±0.04mm, 2.00±0.00mm, and 4.00±0.01mm to *Shigella dysentariae, Escherichia coli* and *Salmonella typhi* respectively while herbs produced by the vendor with secondary education had the mean sensitivity of 1.39±0.6mm, 0.78±0.03mm and 2.37±0.56mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively.

The herbs produced by vendor within age group 11-20 years inhibited *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* with mean sensitivity of 0.00±0.00mm, 2.00±0.00mm and 3.00±0.00mm respectively. Also herbs produced by vendors with age group 21-30 years had the mean sensitivity of 0.85±0.18mm,0.42±0.04mm,1.71±0.56mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively. Herbs produced by vendor within age group 31-40 were sensitivity to *Shigella dysventeriae, E. coli* and *Samonella typhi* with mean sensitivity of 1.90±0.32mm, 1.10±0.64mm, 1.90±0.03mm respectively, while vendor within age group 41-50 years had the mean sensitivity of 4.00±0.00mm, 1.00±0.00mm and 2.50±0.50mm to *Shigella dysventeriae, Escherichia coli* and *Salmonella typhi* respectively.

The herbs produced by Yoruba tribe had the mean sensitivity of 1.74±3.02mm, 0.84±0.64mm and 1.89±2.42mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively, while those herbs produced by Hausa vendor had the mean sensitivity of 0.00±0.00mm, 10.00±0.10mm and 3.00±0.01mm to *Shigella dysenteriae, Escherichia coli* and *Salmonella typhi* respectively.

Occurrence of bacterial in herbs vended at different motor parks in Ado-Ekiti

Table 13 showed the occurrence of bacterial in herbs vended at different motor parks in Ado-Ekiti. A total of nine different bacteria species were isolated from various vended herbs within Ado Ekiti, with *Escherichiacoli* and *Staphylococcusaureus* 8(22.22%) each having the highest occurrence across the locations, followed by *Enterobacter aerogenes* and *Pseudomonasaeruginosa* having the occurrence of 4(11.11%) each. *Klebsiellapneumoniae*, *Shigella dysentarie* and *Salmonella typhi* had the isolation rate of 3(8.33%) each. The least

isolated bacteria were *Proteus mirabillis* and *Citrobacter freundii* with the isolation rate of 2(5.56%) each. All 9 bacteria species were isolated from herbs vended at Odo-Ado Park except *Shigella dysenteriae*, *Pseudomonas aeruginosa, and Proteus mirabilis.* The only four bacteria species isolated from herbs vended at Ajilosun Park were *Escherichia coli, Shigella dysenteriae, Pseudomonas aeruginosa* and *Staphylococcus aureus.* Bacteria species isolated from herbs vended at Bashiri Park include *Escherichia coli, Citrobacter freundii, and Staphylococcus aureus.* The bacteria isolated from Old garage Park included *Escherichia coli, Enterobacter aerogenes, and Staphylococcus aureus.* Herbs vended at Okesa showed bacteria contamination with *Escherichia coli, Shigella dysenteriae, Salmonella typhi* and *Staphylococcus aureus.* Herbs vended at Orita blessing showed bacteria contamination with *Klebsiella pneumoniae, Enterobacter aerogenes, Pseudomonas aeruginosa* and *Proteus mirabilis.* The two bacteria species that were isolated from herbs vended at New garage Park are *Escherichia coli,* and *Staphylococcus aureus.* Also, the bacteria species isolated from herbs vended at Olaniyi Park are *Escherichia coli, Klebsiella pneumoniae*, *Salmonella typhi*, *Pseudomonas aeruginosa,* and *Staphylococcus aureus.* The bacteria species isolated from herbs vended at Lagos Park are *Escherichia coli, Enterobacter aerogenes*, and*Staphylococcus aureus.* The two isolated bacteria species from herbs vended at Abuja are *Pseudomonas aeruginosa*, and*Staphylococcus aureus.*

Antibiotic susceptibility Patterns of bacterial isolated from herbs vended at different motor parks in Ado-Ekiti

Table 14 showed the antibiotic susceptibility patterns of bacterial isolated from herbs vended at different motor parks in Ado-Ekiti. Two (25.0%) *Escherichia coli, Klebsisella pneumonia* 3(100.0%), *Shigella dysentariae* 1(33.33%), *Salmonella typhi* 1(33.33%), *Pseudomonas aeruginosa* 1(25.0%) and 4(50.0%) *Staphylococcus aureus* were resistant to augmentin. One 1(12.50%) *Escherichia coli*, 1(33.33%) *Klebsiella pneumonia*, 1(33.33%) *Shigella dysentariae*, 1(33.33%) *Salmonella typhi* and 1(50.00%) *Citrobacter freundii*. The bacteria specie that was resistant to Ofloxacin was *Klebsiella pneumoniae.Escherichia coli* 2(25.00%), 1(33.33%), 1(33.33%) *Klebsiella pneumoniae*, 1(33.33%) *Shigella dysentariae* and 1(33.33%) *Salmonella typhi* were resistant to amoxicillin. *Escherichia coli* 2(25.00%) and *Shigella dysentariae* 1(33.33%) were resistant to Gentamicin. Escherichia coli 1(12.50%) was resistant to sparfloxacin. All bacteria isolate from the herbs vended in Ado were sensitive to ciprofloxacin and perfloxacin

Effects of herbs vended at different location in Ado motor parks on hematological parameters of albino rat

The Effects of herbs vended at different location in Ado motor parks on hematological parameters of albino rat is showed in Table 15. The packed cell volume of the rats feed with herbs vended from the 10 different location within Ado Ekiti metropolis showed that the PCV of rat were within the range of 33.00±0.52% to 40.00±0.74% which fed below the PCV of the control rat (40.75±0.39%). The haemaoglobin concentrations of the rat fed with the herbs from various Parks were lower (<12.11±0.02g/l) than the control rats except for those fed with herbs from Old garage (12.40±0.16g/l) was higher than that of the control rat (12.11±0.02g/l). The red blood cell counts of the rat were within the range of 2.00±0.00x10^{12}g/l to 2.50±0.04 x10^{12}g/l which were within that of the control rat (2.48±0.01 x10^{12}g/l) except in Ajilosun with red blood cell of 2.50±0.04which was a little bit higher than was obtainable in the control rat. The white blood cell counts of the rats were lower than that of the control which was within the range of 1.20±0.29x10^{6}/mm^{3} to 4.50±0.27 x10^{6}/mm^{3} compare to the control (4.72±0.51 x10^{6}/mm^{3}). The platelet count of the rats showed that there was an appreciable increase in the platelet count of

some rats fed with vended herbs from Odo Ado, Basiri, Orita blessing and Abuja park compare to the control rate ($136.00\pm0.00x10^9$/l).

Photomicrograph of the histology of the Liver of rat administered with herbs

Plates 1 and 2 revealed the photomicrograph of the histology of liver of rats administered with herbs vended at New garage and Lagos park in Ado Ekiti respectively. Although the photomicrograph revealed Liver section with some areas of normal architecture, there are multiple foci of hepatic necrosis with cellular aggregation around the same foci among the rat feed at the herbs vended at new garage in Ado Ekiti while there was no visible lesions seen. This suggested that there was no visible histological damage in the Liver of rats administered with herbs vended at Lagos park in Ado Ekiti. Plate 3 revealed the photomicrograph of the histology of liver of control rat with no lesion.

Also, Plate 4 and 5 revealed the photomicrograph of the histology of kidney of rats administered with herbs vended at New garage and Lagos park in Ado Ekiti. There was slightly normal Kidney architecture, but there is a severe diffuse tubular degeneration and necrosis with the tubular epithelium completely eroded in the Kidney of rats administered with herbs vended at New garage in Ado Ekiti. However, the Photomicrograph of the histology of the kidney of rats administered with herbs vended at Lagos Park in Ado Ekiti showed that there was no visible lesions seen. Plate 6 revealed the photomicrograph of the histology of kidney of control rat with no lesion.

Table 1: Socio-Demographic characteristics of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State

Socio-demographic characteristics		Names of motor parks										Total (%)
		Odo-Ado	Ajilosun	Basiri	Old garage	Okesa	Orita Blessing	New garage	Olaniyi park	Lagos park	Abuja park	
Gender	Female	5	5	5	5	5	5	5	5	5	5	50(100.00)
	Male	0	0	0	0	0	0	0	0	0	0	0(0.00)
Age (year)	11 – 20	0	0	0	0	0	3	0	0	0	0	3(6.00)
	21 – 30	5	0	5	0	0	2	3	0	0	4	19(38.00)
	31 – 40	0	5	0	5	2	0	2	5	3	1	23(46.00)
	41 – 50	0	0	0	0	3	0	0	0	2	0	5(10.00)
Level of education	Primary	0	0	0	0	0	3	0	0	2	0	5(10.00)
	Secondary	5	5	5	5	5	2	5	5	3	5	45(90.00)
Tribe	Yoruba	5	5	5	5	5	2	5	5	5	5	47(94.00)
	Igbo	0	0	0	0	0	0	0	0	0	0	0(0.00)
	Hausa	0	0	0	0	0	3	0	0	0	0	3(6.00)
Religion	Christianity	5	2	5	1	5	3	4	0	5	0	30(60.00)
	Muslim	0	3	0	4	0	2	1	5	0	5	20(40.00)
Household size	3	0	1	0	0	0	0	0	1	1	0	3(6.00)
	4	2	1	2	4	5	3	3	2	2	1	25(50.00)
	5	0	1	1	0	0	2	0	1	1	1	7(14.00)

	6	3	2	2	1	0	0	2	1	1	3	15(30.00)
Marital status	Single	4	0	0	0	0	4	1	0	1	0	10(20.00)
	Married	1	5	5	5	5	1	4	5	2	5	38(76.00)
	Divorced	0	0	0	0	0	0	0	0	2	0	2(4.00)

Table 2: Comparative Microbial contamination of vended and prepared herbs at different motor parks in Ado-Ekiti, Ekiti state.

Motor Park locations	Total viable bacterial counts (cfu/100ml)	Total Staphylococcal counts (cfu/100ml)	Total coliform counts (cfu/100ml)	Total fungal counts (sfu/100ml)
Odo-Ado	37.00 ± 0.21^{b}	9.50 ± 0.25^{b}	16.50 ± 0.45^{b}	3.00 ± 0.10^{a}
Ajilosun	15.50 ± 0.65^{b}	14.00 ± 0.30^{b}	11.00 ± 0.40^{b}	4.00 ± 1.00^{a}
Basiri	24.50 ± 0.95^{b}	12.50 ± 0.15^{b}	8.50 ± 0.50^{b}	8.00 ± 0.20^{b}
Old garage	16.50 ± 0.10^{b}	8.00 ± 0.15^{b}	13.00 ± 0.00^{b}	8.00 ± 0.40^{b}
Okesa	38.00 ± 0.34^{b}	22.00 ± 0.13^{b}	36.50 ± 0.20^{b}	7.50 ± 0.50^{b}
Orita Blessing	20.50 ± 7.50^{b}	0.00 ± 0.00^{a}	16.50 ± 0.50^{b}	8.00 ± 0.14^{b}
New garage	24.50 ± 0.11^{b}	16.50 ± 0.56^{b}	16.50 ± 0.55^{b}	3.50 ± 0.15^{a}
Olaniyi park	19.00 ± 0.50^{b}	7.00 ± 0.40^{b}	8.50 ± 0.15^{b}	3.00 ± 0.00^{a}
Lagos park	52.00 ± 0.36^{c}	15.00 ± 0.30^{b}	18.50 ± 0.95^{b}	10.50 ± 0.35^{b}
Abuja park	24.00 ± 0.70^{b}	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}	1.50 ± 0.50^{a}
Prepared	2.00 ± 0.00^{a}	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}	1.00 ± 0.01^{a}

Values are presented as mean±SE, values in the same column carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 3: Effect of vendors age on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State

Microbial load	Age of vendors (year)				Prepared herb
	11 to 20	21 to 30	31 to 40	41 to 50	
Total viable bacterial counts (cfu/100 ml)	13.00±0.00^{b}	29.14±0.82^{c}	30.60±0.68^{c}	10.00±0.06^{b}	2.00±0.00^{a}
Total Staphylococcal counts (cfu/100ml)	0.00±0.00^{a}	9.14±0.78^{b}	12.40±0.07^{b}	10.50±1.50^{b}	0.00±0.00^{a}
Total coliform counts (cfu/100ml)	17.00±0.00^{b}	11.57±0.37^{b}	13.40±0.67^{b}	11.50±0.50^{b}	0.00±0.00^{a}
Total fungal counts (sfu/100ml)	3.00±0.00^{a}	5.57±0.66^{a}	5.80±1.37^{a}	7.00±0.00^{a}	1.00±0.01^{a}

Values are presented as mean±SE, values in the same row motorrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 4: Effect of vendors level of education on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State

Microbial load	Level of Education		Prepared herb
	Primary	Secondary	
Total viable bacterial counts (cfu/100 ml)	14.50±1.50^{b}	28.56±0.38^{c}	2.00±0.00^{a}
Total Staphylococcal counts (cfu/100ml)	6.00±0.60^{b}	10.94±1.99^{b}	0.00±0.00^{a}
Total coliform counts (cfu/100ml)	14.00±3.00^{b}	12.61±0.57^{b}	0.00±0.00^{a}
Total fungal counts (sfu/100ml)	5.00±0.20^{b}	5.77±0.97^{b}	1.00±0.01^{a}

Values are presented as mean±SE, values in the same row carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 5: Effect of vendors household number on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State

Microbial load	Household size				Prepared herb
	3	4	5	6	
Total viable bacterial counts (cfu/100 ml)	4.00±0.00^{a}	31.20±0.63^{c}	18.33±0.18^{b}	28.67±0.12^{c}	2.00±0.00^{a}
Total Staphylococcal counts	9.0±0.33^{b}	10.40±0.50^{b}	9.67±0.25^{b}	11.17±0.18^{b}	0.00±0.00^{a}

(cfu/100ml)					
Total coliform counts (cfu/100ml)	12.00±0.00^{c}	13.70±0.44^{c}	6.33±0.32^{b}	14.50±0.12^{c}	0.00±0.00^{a}
Total fungal counts (sfu/100ml)	7.00±0.13^{b}	5.20±0.14^{b}	4.67±0.17^{b}	6.83±0.88^{b}	1.00±0.01^{a}

Values are presented as mean±SE, values in the same row carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 6: Effect of vendors' year of job experience on Microbiological quality of herbs vended at different motor parks inAdo-Ekiti, Ekiti State

Microbial load	Year of Job Experience (year)				Prepared herb
	≤ 1	2 to 4	5 to 8	9 and above	
Total viable bacterial counts (cfu/100 ml)	28.50±0.10^{c}	32.20±0.87^{c}	24.00±0.00^{c}	16.60±0.37^{b}	2.00±0.00^{a}
Total Staphylococcal counts (cfu/100ml)	6.50±0.24^{b}	13.30±0.32^{c}	9.40±0.23^{c}	3.00±0.00^{b}	0.00±0.00^{a}
Total coliform counts (cfu/100ml)	16.75±0.65^{d}	13.50±0.37^{d}	9.60±0.25^{c}	5.00±0.00^{b}	0.00±0.00^{a}
Total fungal counts (sfu/100ml)	5.25±0.22^{b}	7.10±0.13^{b}	4.20±0.10^{b}	1.00±0.00^{a}	1.00±0.01^{a}

Values are presented as mean±SE, values in the same row carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 7: Effect of vendors tribe on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State

Microbial load	Tribes		Prepared herb
	Yoruba	Hausa	
Total viable bacterial counts (cfu/100 ml)	27.89±0.53^{c}	13.00±0.00^{b}	2.00±0.00^{a}
Total Staphylococcal counts (cfu/100ml)	11.00±1.89^{b}	0.00±0.00^{a}	0.00±0.00^{a}
Total coliform counts (cfu/100ml)	12.52±0.24^{b}	17.00±0.07^{b}	0.00±0.00^{a}
Total fungal counts (sfu/100ml)	5.84±0.92^{b}	3.00±0.39^{b}	1.00±0.01^{a}

Values are presented as mean±SE, values in the same row carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 8: Comparative Antibacterial potency of prepared and vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria

Motor park locations	*Shigella dysenteriae*	*Escherichia coli*	*Salmonella typhi*
Odo-Ado	0.50±0.05[b]	0.00±0.00[a]	2.50±0.15[b]
Ajilosun	0.00±0.00[a]	0.00±0.00[a]	0.00±0.00[a]
Basiri	0.00±0.00[a]	0.00±0.00[a]	2.50±0.50[b]
Old garage	0.00±0.00[a]	4.50±0.15[c]	0.50±0.10[b]
Okesa	0.00±0.00[a]	0.00±0.00[a]	0.00±0.00[a]
Orita Blessing	0.00±0.00[a]	1.01±0.00[b]	2.50±0.50[b]
New garage	5.00±0.00[c]	1.00±0.00[b]	0.00±0.00[a]
Olaniyi park	0.00±0.00[a]	0.00±0.00[a]	7.00±0.00[c]
Lagos park	8.50±0.70[c]	1.00±0.00[b]	4.50±0.50[b]
Abuja park	2.50±0.20[b]	1.50±0.20[b]	0.00±0.00[a]
Prepared	6.00±0.00[c]	10.00±0.10[d]	8.00±0.40[c]
Ciprofloxacin	21.04±0.77[d]	26.46±0.41[e]	24.03±0.11[d]

Values are presented as mean±SE, values in the same column carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 9: Effect of vendors' year of job experience on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria

Year of Job Experience (year)	*Shigella dysenteriae*	*Escherichia coli*	*Salmonella typhi*
Less the 1 year	0.25±0.02[b]	1.25±0.75[a]	2.00±0.91[a]
2 to 4	1.40±0.97[b]	0.90±0.06[a]	1.20±0.46[a]
5 to 8	3.60±0.56[b]	0.80±0.04[a]	2.40±0.15[a]
9 and above	0.00±0.00[a]	1.09±0.04[a]	7.00±0.01[b]
Prepared	6.00±0.00[c]	10.00±0.10[b]	8.00±0.40[b]
Ciprofloxacin	21.04±0.77[d]	26.46±0.41[c]	24.03±0.11[c]

Values are presented as mean±SE, values in the same column carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 10: Effect of vendors' level of education on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria

Level of Education	*Shigella dysenteriae*	*Escherichia coli*	*Salmonella typhi*
Primary	4.00±0.04[b]	2.00±0.00[a]	4.00±0.01[a]
Secondary	1.39±0.63[a]	0.78±0.03[a]	2.37±0.56[a]
Prepared	6.00±0.00[c]	10.00±0.10[b]	8.00±0.40[b]
Ciprofloxacin	21.04±0.77[d]	26.46±0.41[c]	24.03±0.11[c]

Values are presented as mean±SE, values in the same column carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 11: Effect of vendors' age on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria

Microbial load	Age of vendors (year)				Prepared herb
	11 to 20	21 to 30	31 to 40	41 to 50	
Shigella dysenteriae	0.00±0.00[a]	0.85±0.18[ab]	1.90±0.32[b]	4.00±0.00[c]	6.00±0.00[c]
Escherichia coli	2.00±0.00[a]	0.42±0.04[a]	1.10±0.64[a]	1.00±0.00[a]	10.00±0.10[b]
Salmonella typhi	3.00±0.00[a]	1.71±0.56[a]	1.90±0.03[a]	2.50±0.05[a]	8.00±0.40[b]

Values are presented as mean±SE, values in the same row carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 12: Effect of vendors' tribe on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria

Tribes	*Shigella dysenteriae*	*Escherichia coli*	*Salmonella typhi*
Yoruba	1.74±3.02[b]	0.84±0.64[a]	1.89±2.42[a]
Hausa	0.00±0.00[a]	2.00±0.06[b]	3.00±0.01[b]
Prepared	6.00±0.00[c]	10.00±0.10[c]	8.00±0.40[c]
Ciprofloxacin	21.04±0.77[d]	26.46±0.41[d]	24.03±0.11[d]

Values are presented as mean±SE, values in the same column carrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

Table 13: Occurrence of bacterial in herbs vended at different motor parks in Ado-Ekiti

Bacterial isolates	Location of motor parks											Total (%)
	Odo-Ado	Ajilosun	Basiri	Old garage	Okesa	Orita Blessing	New garage	Olaniyi park	Lagos park	Abuja park	Prepared	
Escherichia coli	+	+	+	+	+	-	+	+	+	-	-	8(22.22)
Klebsiella pneumoniae	+	-	-	-	-	+	-	+	-	-	-	3(8.33)
Shigella dysenteriae	-	+	-	-	+	-	-	-	-	+	-	3(8.33)
Salmonella typhi	+	-	-	-	+	-	-	+	-	-	-	3(8.33)
Enterobacter aerogenes	+	-	-	+	-	+	-	-	+	-	-	4(11.11)
Pseudomonas aeruginosa	-	+	-	-	-	+	-	+	-	+	-	4(11.11)
Proteus mirabilis	-	-	-	-	-	+	-	-	-	-	-	1(2.78)
Citrobacter freundii	+	-	+	-	-	-	-	-	-	-	-	2(5.56)
Staphylococcus aureus	+	+	+	+	+	-	+	+	+	-	-	8(22.22)
Total	6	4	3	3	4	4	2	5	3	2	-	36

Key: + = present, - = absent

Table 14: Antibiotic susceptibility Patterns of bacterial isolated from herbs vended at different motor parks in Ado-Ekiti

Bacterial isolates (n=Number of isolates)	Antibiotics (% resistant)									
	AU	S	CPX	OFX	PEF	AMX	CN	SP	CH	SHT
Escherichia coli (n = 8)	2(25.00)	1(12.50)	0(0.00)	0(0.00)	0(0.00)	2(25.00)	2(25.00)	1(12.50)	4(50.00)	0(0.00)
Klebsiella pneumoniae (n = 3)	3(100.00)	1(33.33)	0(0.00)	1(33.33)	0(0.00)	1(33.33)	0(0.00)	0(0.00)	1(33.33)	0(0.00)
Shigella dysenteriae (n = 3)	1(33.33)	1(33.33)	0(00.00)	0(00.00)	0(00.00)	1(33.33)	1(33.33)	0(00.00)	1(33.33)	1(33.33)
Salmonella typhi (n = 3)	1(33.33)	1(33.33)	0(0.00)	0(0.00)	0(0.00)	1(33.33)	0(0.00)	0(0.00)	1(33.33)	1(33.33)
Enterobacter aerogenes (n = 4)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	2(50.00)	0(0.00)
Pseudomonas aeruginosa (n = 4)	1(25.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(25.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)
Proteus mirabilis (n = 1)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(100.00)	0(0.00)
Citrobacter freundii (n = 2)	0(0.00)	1(50.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	1(50.00)
Staphylococcus aureus (n = 8)	4(50.00)	0(0.00)	0(0.00)	0(0.00)	0(0.00)	7(87.50)	0(0.00)	0(0.00)	0(0.00)	0(0.00)

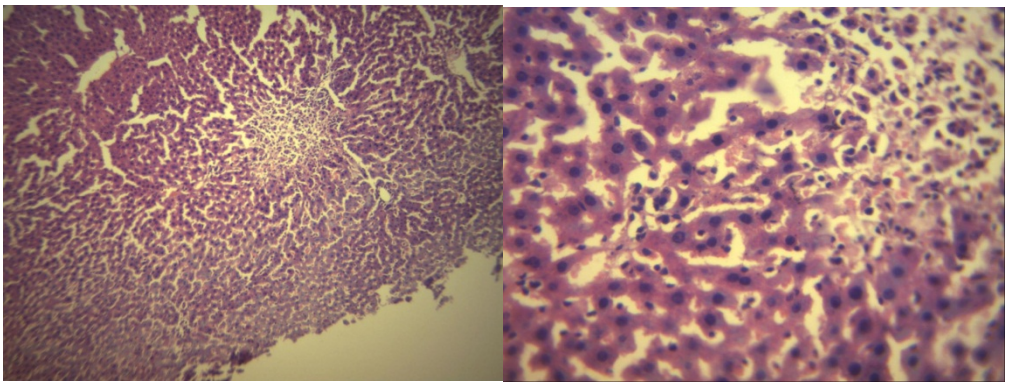
Key: AU=Augumentin, S = , CPX =Ciprofloxacin , OFX =Oxfloxacin , PEF =Pefloxacin , AMX =Amoxylin , CN = , SP = Sparfloxacin , CH = , SHT

Table 15: Effects of herbs vended at different location in Ado motor parks on hematological parameters of albino rat

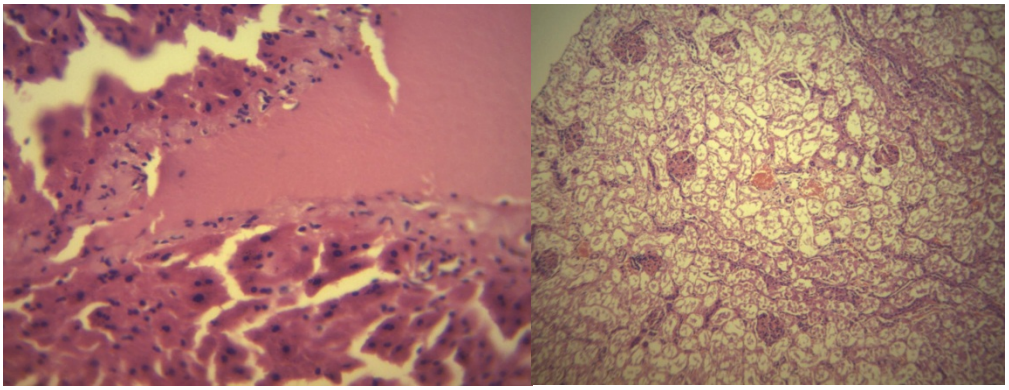
Herbs sources	PCV (%)	HB (g/l)	RBC 10^{12}g/l	WBC 10^{6}/mm^{3}	MCV (fl)	MCH (pg)	PLT 10^{9}/l	MCHC (g/l)
Odo Ado	38.00±1.02^{ab}	12.10±0.39^{a}	2.08±0.09^{a}	2.20±0.31^{c}	6.60±0.24^{a}	1.40±0.38^{a}	143.00±0.00^{c}	34.00±0.00^{b}
Ajilosun	40.00±0.74^{b}	12.00±0.53^{a}	2.50±0.04^{a}	4.40±0.26^{d}	7.60±0.18^{a}	1.10±0.00^{a}	133.00±0.00^{b}	30.60±0.02^{a}
Basiri	37.00±1.23^{a}	12.00±0.28^{a}	2.00±0.03^{a}	1.80±0.74^{b}	7.80±0.31^{a}	1.50±0.00^{a}	138.00±0.00^{c}	33.90±0.18^{b}
Old garage	40.00±0.33^{b}	12.40±0.16^{a}	2.00±0.02^{a}	1.20±0.56^{a}	8.50±0.11^{b}	1.30±0.13^{a}	120.00±0.00^{a}	28.00±1.32^{a}
Okesa	39.00±0.41^{b}	12.00±0.77^{a}	2.30±0.02^{a}	4.40±0.22^{d}	6.50±0.00^{a}	1.30±0.03^{a}	118.00±2.73^{a}	32.10±0.05^{b}
Orita blessing	36.00±0.31^{a}	11.00±0.62^{a}	2.20±0.11^{a}	3.20±0.21^{c}	7.30±0.06^{a}	1.20±0.00^{a}	138.00±1.84^{c}	32.20±0.11^{b}
New garage	33.00±0.52^{a}	10.30±0.19^{a}	2.10±0.18^{a}	1.20±0.29^{a}	6.80±0.03^{a}	1.10±0.02^{a}	123.00±0.00^{a}	32.10±0.31^{b}
Olaniyi park	36.00±0.07^{a}	11.00±1.36^{a}	2.20±0.07^{a}	1.84±0.18^{b}	7.00±0.00^{a}	1.10±0.01^{a}	130.00±0.21^{b}	32.20±0.51^{b}
Lagos park	38.00±0.36^{ab}	12.00±0.92^{a}	2.00±0.00^{a}	4.50±0.27^{d}	8.00±0.00^{ab}	1.50±0.02^{a}	122.00±0.14^{a}	32.00±0.00^{b}
Abuja park	36.00±0.16^{a}	11.00±0.33^{a}	2.00±0.00^{a}	4.20±0.22^{d}	7.50±0.31^{a}	1.30±0.01^{a}	138.00±0.00^{c}	32.20±0.05^{b}
Control	40.75±0.39^{b}	12.11±0.02^{a}	2.48±0.01^{a}	4.72±0.51^{d}	8.11±0.03^{ab}	1.13±0.02^{a}	136.00±0.00^{b}	33.63±0.14^{b}

Values are presented as mean±SE, values in the same column motorrying same superscript are not different significantly according to new Duncan's Multiple Range test at $p<0.05$

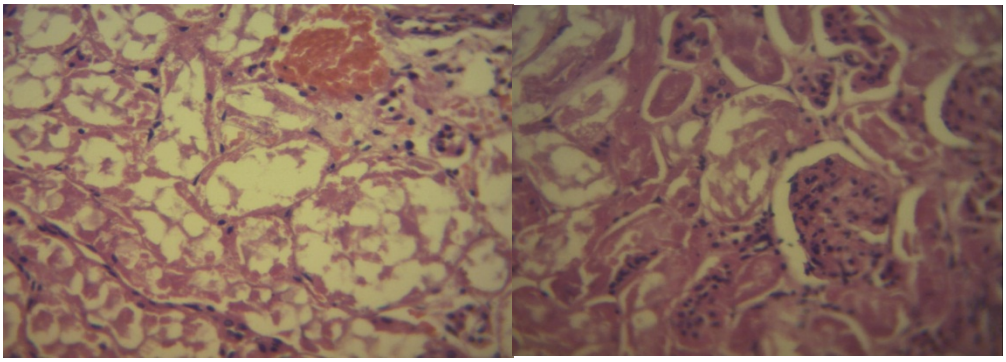
Keys: PCV = Packed cell Volume, HB = Haemoglobin Concentration, WBC = White Blood Cell , RBC = Red Blood Cell, MCH = Mean Cell Haemoglobin, PLT = Platelet, MCV = Mean Cell Volume, MCHC = Mean Cell Haemoglobin Concentration



(A) (B)



(C) (D)



(E) (F)

A-Plate 1: Photomicrograph of the histology of the Liver of rat administered with herbs vended at New garage in Ado Ekiti
B- Plate 2: Photomicrograph of the histology of the Liver of rats administered with herbs vended at Lagos park in Ado Ekiti
C-Plate 3: Photomicrogragh of the Histology of the Liver of control rat

D-Plate 4: Photomicrograph of the histology of the Kidney of rats administered with herbs vended at New garage in Ado Ekiti
E-Plate 5: Photomicrograph of the histology of the kidney of rats administered with herbs vended at Lagos park in Ado Ekiti
F-Plate 6: Photomicrograph of the histology of the kidney of control rat

DISCUSSION

The Socio-Demographic characteristics of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State showed that all the vendors were female 50 (100.0%). This might be because herbs preparation requires a lot of house hold dedication, although male subjects might have involved in one process or the other but female get more involved in the preparation and marketing. Also, in this study location, it is not common to see a male subject hawking because it is assumed that kind of job is for the female subjects. This is supported by Adele (2000) who reported thatin Nigeria, there has been a remarkable overwhelming increase of public awareness and usage of herbal medicinal products (HMP) in the treatments and/or prevention of diseases accompanied by a lot of advertisement from the female subjects.
There were more of the female subjects within age group 21-30yrs and 31-40yrs with occurrence rate of 19(38.0%) and 23(46.0%) respectively compared to other age groups. This might be because these age groups are active and full of energies used for day-to-day activities. Since street vending requires a lot of energy for the marketing and selling of the herb products to their prospective customers. This is in agreement with the study conducted by Abera (2014). Also, most of the female vendors were married with occurrence rate of 38(76.0%). This could be because most of the vendors were within the ripe marriage age.

Also, there were more of the vendors with secondary education compared to the primary education. This could be because preparation of herbs does not require formal education. This is in agreement with Tapsell *et al.,* (2006), who reported that there are many different methods of preparing medicinal herbs and there are no really any set rules for the preparation of herbs.

About 47(94.0%) of the vendor were Yoruba. This might be because the study was conducted in the southwest region which is majorly Yoruba speaking part of Nigeria. It is agreement with Abera (2014) who reported that in Nigeria, “Agunmu” is locally produced by traditional people of Yoruba land. It is the most common and easiest way of preparing herbal materials.

The comparative microbial contamination of vended and prepared fever herbs at different motor parks in Ado-Ekiti, Ekiti State. The total bacterial count in Ajilosun was significantly lower in the herbs vended at Ajilosun compared to those vended at Lagos Park. The low bacterial count recorded for the laboratory prepared herb compared to those vended at the motor parks could be due to the fact that the herb was prepared under septic condition. This suggested that herbs sold in those motor parks were not prepared under hygienic condition and as such can be a vehicle of transmission of microbial pathogen and can lead to disease condition. This is in agreement with WHO (2014) that reported that herbs is a known vehicle that serves as means of transmission of food-borne pathogens to humans. Herbs could be contamination by microorganisms because of the unhygienic handling practices and low level of safety knowledge of the Herbs vendors. This could predispose patients and other consumers of the products to the risk of infections with pathogenic and antimicrobial resistant bacteria (Jonas, 2015).

Finding from the present study showed that the total coliform count was significantly higher in the herbs vended at most of the motor Parks compared to those prepared in the laboratory and those vended at Abuja Park. The term "total coliforms" represents a group of non-spore forming, gram-negative, facultatively anaerobic, bacillus bacteria that strongly ferment lactose to acid and gas following 42–48 h incubation at 35±2 °C (Halkman, 2014). Coliforms, which are commonly found in the intestines of man and other warm-blooded animals (Halkman, 2014) include some strains of *Escherichia* spp, *Klebsiella* spp, and *Citrobacter* spp. among others. These three organisms mentioned were among those that were isolated from vended herbs at the motor parks with *E. coli* isolated from almost all herbs vended at the different motor parks except herbs gotten from Orita-blessing and Abuja parks. These signify potential contamination by fecal matter and its associated human pathogens, threatening public health (Walusansa *et al*., 2022). In the present study the most isolated bacteria, *Escherichia coli*, is believed to be one of commonest fora that inhabit the intestinal tract of vertebrates (Walusansa *et al*., 2022). Therefore, the presence of this coliform is widely used as a reliable basis to infer the degree of fecal contamination (Walusansa *et al*., 2022). Actually, no coliform organism is acceptable in any product intended for human consumption (Martin *et al*., 2016).

The effect of vendors' level of education on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State showed that herbs vendors with secondary education had highest microbial load compare to those with primary education. This could suggest that the level of education does not have effect on the preparation of good quality herbs. Although, the reason for the increase microbial load among those with secondary education is not too clear. Since those with secondary education should have better understanding about the need to practice hygiene than their counterpart (Timmis *et al*., 2019).

The effect of vendors household number on microbiological quality of the herb vendors at selected motor parks in Ado-Ekiti, Ekiti State showed that those vendor with 3 and 5 households had a reduce microbial count compare with those with 4 and 6 house household. This shows that the number of household may not have effect on the quality of the herbs vended in the Ado metropolis. Since there are no significant differences in term of the counts in relation to the number of households. This disagreed with the finding of Omoya and Kola-Oladejo, (2021) who reported that high number of vendors' household size, herbs packaged with plastics and nylon contributed significantly to high coliform counts.

The effect of vendors' year of job experience on microbiological quality of herbs vended at different motor parks in Ado-Ekiti, Ekiti State showed that there is reduction in the total bacteria count, Staphylococcus count, coliform count and fungal count as the year of experience increases. This suggested that those that have preparing or vending these herbs for more than 9 years had developed aseptic skill overtime and this has contributed to the reduction in the bacteria counts. This is supported by Kitula (2007) who reported that experience of the vendor also minimizes microbial contamination during production.

The antibacterial potency of the vended herbs and the laboratory prepared herb on the isolated diarrheagenic bacteria isolated in this study which included *Shigella dysenteriae*, *Escherichia coli* and *Salmonella typhi* was compared to ciprofloxacin, a broad-spectrum antibiotic. The potency of ciprofloxacin against diarrheagenic bacteria is well established in studies such as that of Tribble, (2017). As expected, Ciprofloxacin has a wider mean diameter zone of inhibition around the isolated bacteria compared to that observed from the herbs. It is also important to note

that the laboratory prepared herbs had more potency against the isolated bacteria compared to the herbs vended at the different motor parks. The only exception to this was the fact that herbs vended at Lagos park had higher antibacterial potency against *Shigella dysenteriae* compared to the laboratory prepared herbs. Among the herbs vended at the different motor parks, those vended at Olaniyi park had more potency against *Salmonella typhi* while those vended at old garage motor park was more potent against *Escherichia coli*.

The effect of vendors' tribe on Antibacterial potency of vended herbs at different motor parks in Ado-Ekiti, Ekiti State on selected diarrheagenic bacteria showed that herbs prepared by Yoruba tribe had a wider zone of inhibition against *Shigella dysenteriae*, while for antibacterial potency against *Escherichiacoli* and *Salmonellatyphi*, those herbs prepared by the Hausa tribe expressed wider zone of inhibition compared to herbs from the Yoruba tribe. However, herbs prepared in the laboratory had significantly wider zone of inhibition against all the bacteria compared to herbs vended at the motor parks.

The occurrence of bacterial in herbs vended at different motor parks in Ado-Ekiti showed that there were seven (7) different bacteria isolates belonging to seven (7) different bacteria genera isolated from this study; *Escherichia coli* and *Staphylococcus aureus* had the highest occurrence, followed by *Enterobacter aerogenes, Pseudomonas aeruginosa* while *Klebsiella pneumoniae, Shigella dysenteriae,* and *Salmonella typhi*. *Citrobacter freundii* and *Proteus mirabilis* had the lowest occurence. This is supported by Oluyege and Adelabu (2010) who reported that pathogenic microorganisms commonly isolated from herbs pose a serious threat to human health. Some of these pathogens include *Escherichia coli, Staphylococcus aureus, Salmonella spp., Listeria monocytogens, Bacillus* spp*., Mycobacterium* spp*., Campylobacter* spp., *Clostridium* spp., *Pseudomonas aeruginosa* and *Proteus* spp.

Higher occurrence of *S. aureus* in vended herbs could pose a greater health risk to the consumers. This bacterium species is normal skin flora, they live as a commensal organism on the in the nose and throat of humans and animals (Varshney *et al*., 2018), it is an important cause of food poisoning following ingestion of preformed heat-resistant toxins. It has been demonstrated that ingestion of Staphylococcus enterotoxins (SEs) within food cause food poisoning, which is characterized by severe vomiting and diarrhea. Those symptoms occur within hours after eating of SE contaminated food (Marrack and Kappler, 2014).

The antibiotic susceptibility patterns of bacterial isolated from herbs vended at different motor parks in Ado-Ekiti showed the antibiotic resistance of the isolated bacteria against the 10 tested varied across the isolated bacteria. It is noteworthy that none of the bacteria was resistant against ciprofloxacin (CPX) and perfloxacin (PEF). There was a high resistance of *Staphylococcus aureus* against Amoxicillin (AMX) and Augmentin (AU). This is in agreement with WHO (2014) who reported that apart from herbs being potential carrier of pathogens can cause serious health risk to consumers due to their antimicrobial resistance. Report on global surveillance of antimicrobial resistance (AR) revealed that AR is no longer a prediction for the future; it is happening right now, across the world, and is putting at risk the ability to treat common infections in the community and hospitals (WHO, 2014).

The haematological parameters of the rats feed with the herbs in various locations in Ado metropolis. Although, the packed cell volume, haemoglobin concentration and the red blood cell count of albino rats feed with herbs were similar to that of control. Nevertheless, there was a

significantly increased in the white blood cell count of most of the rats feed with herbs in some locations. There are multiple foci of hepatic necrosis with cellular aggregation around the same foci in the liver and there is a severe diffuse tubular degeneration and necrosis with the tubular epithelium completely eroded in the Kidney of rats administered with herbs vended at New garage in Ado Ekiti and some other locations. This suggested that the bacteria could have contaminated the herbs, the toxic effects of the chemical constituents of plants used in the preparation of the vended herbs could have contributed to the results obtained in this study. This is supported by Fennell *et al*. (2014) who reported that plant could be a source of toxins produced in the herbs and could therefore be a source of the toxicity showed the histological and hematological parameters.

The photomicrograph of the organs (Kidney and Liver) at New garage and Lagos park in Ado Ekiti showed that there is a few renal tubules appear degenerated with the tubules being necrotic and slightly eroded in kidneys of the rats fed with the herbs while the liver showed no visible lesion. This slight degenerative effect seen on the kidney could be due to the chemical constituents of plants used in preparation of herbs which are potentially toxic (Fennell *et al.,* 2014). Contamination of plant used for the herbs preparation by pesticides and other chemicals used in agricultural practices could be source of toxicity (Sharma *et al.,* 2017).

Conclusion

Findings from the present study showed that the total coliform count was significantly higher in the herbs vended at most of the motor parks compared to those prepared in the laboratory and those vended at Abuja Park. It is important to note that the laboratory prepared herbs had more potency against the isolated bacteria compared to the herbs vended at the different motor parks. The only exception to this was the fact that herbs vended at Lagos park had higher antibacterial potency against *Shigella dysenteriae* compared to the laboratory prepared herbs. The significant socio-demographic characteristics of the vendors associated with the prevalence of bacterial contamination include their level of education and year of job experience. Based on deductions from this study there is need to practice personal hygiene by the vendors as some of the bacteria isolated from this study indicate faecal contamination of herbal medicine product.

Recommendation

Herbs is a known vehicle that serves as means of transmission of food-borne pathogens to humans. Pathogenic microorganisms commonly isolated from herbs pose a serious threat to human health. Therefore, the steps involve in the preparation of herbs must be standardized to prevent the survival of microorganisms. I therefore recommend that every locally vended herbs must be quality controlled and monitored by a government agency. As observed from the present study, there is need to increase awareness on the bacterial contamination of vended herbs as the level of literacy was seen to be significantly associated with the prevalence of diarrheagenic bacteria in the present study.

REFERENCES

Abubakar, A. R., and Haque, M. (2020). Preparation of Medicinal Plants: Basic Extraction and Fractionation Procedures for Experimental Purposes. *Journal of pharmacy & bioallied sciences*, *12*(1), 1–10.https://doi.org/10.4103/jpbs.JPBS_175_19.

Adele G Dawson (2000). Herbs, Partners in Life: Healing, Gardening and Cooking with Wild Plants. Bear & Co. pp. 5–6.

Encyclopedia Britannica(2021) Ado-Ekiti | Location, Facts, & History" (https://www.britannica.com/place/Ado-Ekiti) .. Retrieved 2021-06-25.

Allaby, Michael (2012). A Dictionary of Plant Sciences.Oxford University Press.ISBN 9780191079030

Cheesbrough M (2014)District Laboratory Practice in Tropical Countries. 2nd Ed. Cambridge University Press, United Kingdom, 480.

Committee for Clinical Laboratory Standards, (2019).Performance Standards for Antimicrobial *Susceptibility Testing; Twenty-Seventh Informational Supplement*. CLSI document M100-S27 Wayne, PA: ISBN 1-56238-898-3. 34(1): 50-98

Devika M., Bhuvaneshwari G. (2019). Comparative Study of Membrane Filtration and Spread Plate Technique for Dialysis Water Analysis.*Saudi Journal of Pathology Microbiology*4(9): 649-653

Dillehay T, Rossen J, Ugent D, Karathanasis A, Vásquez V, Netherly P (2010). "Early Holocene coca chewing in northern Peru".*Antiquity*.84 (326): 939–953

Fennel, C.W., Light, M.E., Sparg, S.G., Stafford, G.I. and Van Staden, J. (2004). Assessing African medicinal plants for efficacy and safety: agricultural and storage practices. *Journal of Ethno-pharmacology* 95: 113-121.

Halkman, H. B., and Halkman, A. K(2014).Coliform Bacteria.Sci. Direct.Published online.

Iguchi, H.; Yurimoto, H.; Sakai, Y. (2015)."Interactions of methylotrophs with plants and other heterotrophic bacteria".*Microorganisms*.3 (2): 137–151

Jonas Daniel Kira (2015) Prevalence and antimicrobial susceptibility of bacteria isolated from herbal medicines vended in morogoro municipality, Tanzania.A dissertation submitted in partial fulfillment of the requirements for the degree of master of science in public health and food safety of sokoine university of agriculture. Morogoro, Tanzania. Pp. 1-65.

Kitula, R.A. (2007). Use of medicinal plants for human health in Udzungwa Mountains Forests: A case study of New DabagaUlongambi Forest Reserves, Tanzania. *Journal of Ethno-biology and Ethno-medicine*3:7.

Kothari C. R. (2004). Research methodology: Methods and techniques. New AgeInternational.

Kumar, Manish; Singh Tomar, Rajesh; Lade, Harshad; Paul, Diby (2016). "Methylotrophic bacteria in sustainable agriculture". *World Journal of Microbiology and Biotechnology*.32 (7): 120.

Majekodunmi S.O.. (2015) Review of extraction of medicinal plants for pharmaceutical research. Merit research journals. 3:521–7.

Marrack, P. and Kappler, J. (2014).The Staphylococcal Enterotoxins and Their Relatives. Science, 248: 705–711.

Martin, N. H., Trmčić, A., Hsieh, T. H., Boor, K. J., & Wiedmann, M. (2016). The Evolving Role of Coliforms As Indicators of Unhygienic Processing Conditions in Dairy Foods. *Frontiers in microbiology*, *7*, 1549. https://doi.org/10.3389/fmicb.2016.01549

Meshack, O.O., Hezekiah, K.C., Grace, N.T., Godfrey, O.O. and George, O.O. (2013). Microbial Quality of Unregulated Herbal Medicinal Products in Kenya; *African Journal of Pharmacology and Therapeutics*2(3):70-75.

Ochei, J. and Kolkahtar, A. (2000). Bacteriology: Medical Laboratory Science, theory and practice. In:Bulakh P.M. and Deshmukh S. (eds). Tata McGraw-Hill publishing company limited New Delhi. . 525-752. ISBN-13:978-0-07-463223-9.

Oluyege, J.O. and Adelabu, D.M. (2010). Microbial contamination of some hawked herbal products in Ado-Ekiti, Nigeria. *Continental Journal of Microbiology*4:8-14.

Omoya, F.O., and Kola-Oladejo M.O. (2021) Microbial Contamination of Vended Fever Herbs in different Motor Parks in Akure, South-West Nigeria.Global Research Journal of Public Health and Epidemiology. Vol. 9(12)

Sharma, A.; del Carmen Flores-Vallejo, R.; Cardoso-Taketa, A.; Villarreal, M.L. (2017) Antibacterial activities of medicinal plants used in Mexican traditional medicine. Journal of Ethnopharmacology 208, 264–329.

Sjöling Å, Sadeghipoorjahromi L, Novak D (2015) Detection of major diarrheagenic bacterial pathogens by multiplex PCR panels. Tobias. *Microbiology Resource.*172:34-40

Tapsell L. C., Hemphill I, Cobiac L., Sullivan D. R., Fenech M., Patch C. S., Roodenrys S, Keogh J. B., Clifton P. M., Williams P. G., Fazio V. A., Inge K. E. (2006). "Health benefits of herbs and spices: The past, the present, the future". *Medical Journal of Australia.*185 (4): S1–S24.

Timmis, K., Cavicchioli, R., Garcia, J. L., Nogales, B., Chavarría, M., Stein, L., … Harper, L. (2019). *The urgent need for microbiology literacy in society. Environmental Microbiology, 21(5), 1513–1528.* doi:10.1111/1462-2920.14611

Tribble D. R. (2017).Antibiotic Therapy for Acute Watery Diarrhea and Dysentery. *Military medicine*, *182*(S2), 17–25. https://doi.org/10.7205/MILMED-D-17-00068

Varshney, A. K.; Mediavilla, J. R.; Robiou, N.; Guh, A.; Wang, X.; Gialanella, P.; Levi, M. H.; Kreiswirth, B. N. and Fries, B. C. (2018) Diverse Enterotoxin Gene Profiles Among Clonal Complexes of Staphylococcus aureus Isolates From the Bronx, New York. Applied Environmental Microbiology, 75: 6839–6849

Walusansa A., Asiimwe S., Kafeero H. M., Stanley I. J., Ssenku JE, Nakavuma J.L., *et al*. Prevalence and dynamics of clinically signifcant bacterial contaminants in herbal medicines sold in East Africa from 2000 to 2020: a systematic review and meta-analysis. Trop Med Health. 2021;49(1):1–14. https://doi.org/10.1186/s41182-020-00295-8

WHO (2007, Guideline for assessing quality of herbal medicine with Reference to contaminants and residues. World Health Organization, Geneva.

WHO (2011). Food safety and food borne illness; Fact sheet No 237; [http://www.who.int/mediacentre/factsheets/fs237/en] site visited on 10/06/ 2014.

WHO (2013). Diarrheal diseases, fact sheet Number 330, viewed May 2013, [http://www.who.int/mediacentre/factsheets/fs330/en/index.html] site visited on 12/06/2014.

WHO (2017) Fact sheet about diarrhea.World Health Organization, Geneva www.WHO.int

World Health Organization (WHO)(2017). Prioritization of pathogens to guide discovery, research, and development of new antibiotics for drug-resistant bacterial infections, including tuberculosis.World Health Organization.