https://dx.doi.org/10.4314/jpb.v16i2.11
Vol. 16 no. 2, pp. 165-172 (September 2019)
http://ajol.info/index.php/jpb

Journal of
PHARMACY AND BIORESOURCES

Phytochemical and *in vitro* anti-typhoid properties of leaf, stem and root extracts of *Ficus capensis* (Moraceae)

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Received 9th July 2019; Accepted 20th August 2019

Abstract
Ficus capensis plant and its parts have diverse trado-medicinal application in Nigeria folklore. This study was designed to investigate the *in vitro* phytochemical and anti-typhoid potential of the root, stem and leaves of the plant as an attempt to proffer solution to the challenges faced by modern medicine especially in the complete cure of microbial-associated diseases as a result of their abrupt and unpredictable genetic mutations. The powdered leaf, stem and root of the plant were extracted with various solvents viz., n-hexane, chloroform, methanol and water by serial exhaustive extraction with each extract challenged with *Salmonella typhi* a common typhoid disease-causing organism. The leaf extract contains the highest concentration of all the phytochemical studied except for tannin, which was found to be highest in the stem bark. While the root, stem and leaf extracts of *F. capensis* inhibited the growth of *S.* Typhi in a concentration dependent manner comparable to that of the standard drugs, the reconstitution solvent showed no antibacterial activity. The results show that leaf, stem bark and root extracts of *F. capensis* confer anti-typhoid activity against *Salmonella* Typhi.

Keywords: *Ficus capensis*; Phytochemical; *Salmonella typhi*

INTRODUCTION

Medicinal plants over the years have remained relevant in the treatment of diverse ailments plaguing humankind. Research revealed that more than 80% of African population rely one way or the other on traditional medicine for their primary health care needs [1,2] since modern Western medicine is either unavailable or is simply too expensive for the common man to afford. This is so because plants are rich in a wide variety of secondary metabolites such as tannins, terpenoids, alkaloids, flavonoids, among others that have been found, *in vitro*, to have antimicrobial properties [3]

Typhoid fever is an enteric fever caused by *Salmonella* Typhi [4,5]. The disease is transmitted by water, milk, fruits and vegetables contaminated with the bacterium. It is also transmitted by healthy carriers and contaminated food handlers [6]. It has an incubation period of normally two weeks after which the organism becomes disseminated in the body and can be isolated from urine and feces. Clinically, it is characterized by a continual high fever and headache with the low pulse rate, headache,

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ISSN 0189-8442

enlargement of the spleen and mental confusion. Its burden is highest in developing countries, particularly in Africa. The disease remains endemic in many developing countries as a result of the near lack of good sanitary facilities and waste disposal systems. It has an estimated mortality of up to 30% and could result in over 20 million hospital cases annually, with at least 700,000 deaths [7].

Ficus capensis a native of tropical Africa and the Cape Islands belongs to the mulberry family Moraceae. The plant is a deciduous tree with spreading roots and branches and broad green leaves. *F. capensis* produces fleshy fruits all year round in a single or branched raceme along the trunk and the main branches. It is known as "*uwaryara"* in Hausa, "o*poto"* in Yoruba, "*rima bichehi"* in Fulani, "*akokoro"* in Igbo and "*obada"* in Edo [8,9]. Traditionally, decoctions of the plant parts are used in the treatment of dysentery, oedema, epilepsy, chest ailments, leprosy, tuberculosis, anemia, rickets in infants among others [9,10,11,12]. Aside its trado-medicinal applications, *F. capensis* have been reported based on modern scientific investigations to possess anti-sickling [13,14], antibacterial [11], antiabortifacient [15], immune-stimulatory [16], antidiarrhoeal [17], antioxidant [18] and pro-fertility in treating azoospermia [19].

In spite of the advances in disease management and medicine, disease-causing microorganisms continually undergo genetic variations which enable them mutate and offer resistance to common antibiotics. The emergence and spread of *Salmonella typhi* resistance to many commonly used antibiotics is now a subject of international concern. This problem is as a result of increase hospitalizations, health costs, and mortality. It is against this background that the search for modern and efficacious antimicrobial agents in disease management and control is unending in line with one of the World Health Organization interventions [20,21]. This study hence was carried out to give scientific credence or otherwise to the claim that *F. capensis* leaf, stem and root have a positive effect against *S.* Typhi as claimed in folklore medicine in comparison with that of established antibiotics *vis-a-vis* ascertaining the plant part with better activity.

EXPERIMENTAL

Plant collection and treatment. The plant's stem bark, root, and leaves were collected from various locations in the Igala speaking areas of the north-central Nigeria. They were authenticated by experts in the Biological Sciences Department of Kogi State University, Anyigba, Kogi State Nigeria. A voucher specimen was deposited at the herbarium of Biological Sciences Department. Soil particles were removed from the plant parts by washing under running tap water. They were shade dried for 2 weeks before being oven dried at 40^{o}C for 24 hours. The dried plant samples were reduced to fine powder with the aid of mortar and pestle. They were weighed into a clean glass container and labeled.

Extraction. Uniform weight of the plant samples was extracted successively with solvents of increasing polarity starting with n-hexane, followed by chloroform, methanol and finally distilled water. 100 g of each of the pulverized plant sample was macerated in 600 ml of n-hexane in a capped vessel. After 24 hours, the macerate was filtered through Whatman No 1 filter paper. The filtrate was concentrated using a rotary evaporator and dried on a water bath to obtain the n-hexane extract (NHE). The residue obtained from the filtration was collected, dried and macerated in 600 ml of chloroform for another 24 hours; the filtrate was concentrated as done for n-hexane to produce the chloroform extract (CFE). The procedure was repeated for methanol and water to obtain the methanol (MEE) and water extracts (WAE) respectively. The extracts were stored in

clean, labeled, capped brown bottles and maintained at 4ºC until ready for use. All extracts were weighed with the percentage yield calculated determined relative to the starting material.

Bacteria culture. Clinical isolate of *Salmonella* Typhi was obtained from hospitalized typhoid fever patients at Maria-Goretti Hospital, Anyigba, Kogi State. The *b*acterial strain was sub cultured on nutrient agar followed by incubation at 37±1°C for 24 hours before being used for the study

Confirmation of test organism. Biochemical analysis methods as described by MacFaddin [22-24] were carried out on the test bacteria isolate for confirmation. The Bergey's Manual of Systematic Bacteriology [25] was used for species authentication.

Standardisation of inoculum for antibacterial screening. The method of Rajarkaruna *et al.* [26] was used for the standardization of the inoculum. Nutrient Broth for the standardization of the inoculum was prepared by adding 2 g of nutrient broth to 40ml of distilled water in a 250 ml conical flask and then digesting it on a hot plate. The clear solution was then poured into clean McCartney bottles and sterilized by autoclaving at 121ºC for 15 minutes. A loop full of the pure isolate of the test microorganism was aseptically suspended into 15 ml of the broth in the McCartney bottle and then labeled. The culture was incubated at 37ºC for 24 hours before use.

Phytochemical Screening. The pulverized plant samples of *F. capensis* were screened for the presence of secondary metabolites. The quantity of each phytoconstituents was determined according to the procedure described by Sofowora [27] and Evans [28].

Determination of inhibitory activities of extracts. The antibacterial activity of the aqueous extract of the samples were determined by the agar well diffusion technique described by Odama *et al.*, [29] as modified by Musa *et al.*, [30]. A suspension of the standardized inoculum (0.2 ml) was seeded into 600 ml of molten Mueller-Hinton agar at a temperature of 40ºC. The seeded agar was poured aseptically into sterile petri dishes and allowed to set at room temperature. The solidified agar was bored with a sterile 8 mm cork borer to create five wells on the agar plate about 10mm deep. The wells were filled with 0.1 ml of 12.5 mg/ml, 25 mg/ml, 50 mg/ml, and 100 mg/ml each of NHE, CFE, MEE and WAE extracts and the fifth well was filled with n-hexane, chloroform, methanol and water respectively to serve as the solvent control. Similarly, three plates, which serve as the positive control, were prepared for the standard antibiotics, gentamycin, amoxicillin and chloramphenicol. The plates were incubated for 24 hours at 37ºC. The resulting zones of inhibitions in mm around each well were then measured using a transparent ruler. The experiment was carried out in triplicate

RESULTS AND DISCUSSION

The percentage yield of the extracts is shown in table 1. The use of water as the solvent of choice in Igala folk medicine for the extraction of this plant is validated by the high yield of the aqueous extracts obtained from the plant except for methanol, which has better extraction yield from the stem bark of the plant. This shows *F. capensis* sequester more readily into aqueous extracts.

Phytochemical screening. The quantitative phytochemical screening of *F. capensis* showed the presence of important secondary metabolites which is presented in Table 2. The presence of these phytochemicals is responsible for the diverse trado-medicinal application of the plant. Quantitatively, phenol has highest concentration while phytate and oxalate were present in the least concentration in all the extract examined. Although phytate and oxalate are regarded as

antinutritional factors, their concentration below the critical level shows that they will not inhibit the bioavailability of the nutrients inherent in the plant. The leaf was reported to contain the highest concentration of all phytochemicals screened for. The presence of cardiac glycoside, saponin, alkaloid, flavonoid and tannin is in agreement with the findings of Ehimwenma and Osarieme [31].
Flavonoid is one of the most diverse and widespread group of natural compounds. They possess broad-spectrum chemical and biological activities such as anti-allergenic, anti-viral, anti-inflammatory, anti-cancer and vasodilating actions [32-36]. The presence of saponins in all extract of *F. capensis* justifies its use locally to stop bleeding and in treating wounds [12]. Saponins also possess hemolytic, cholesterol binding and antimicrobial activity [32,37,38]. Tannins, having astringent properties, hasten the healing of wounds and inflamed mucous membranes [39]. The high saponin content in the leaf when compared to the root and stem bark corroborated the findings of Solomon-Wisdom *et al.,* [40] as well as contradict the findings of Oyeleke *et al.* [11] which also reported the presence of all the phytochemicals identified except saponins.

Antibacterial screening. The result of the antimicrobial screening when the NHE, CFE, MEE and WAE when challenged with *S.* Typhi as well as the response of test isolate to some standard antibiotics (positive control) and the reconstitution solvent (negative control) are shown in Figures 1 and 2 respectively. The result showed that the extracting solvents of n-hexane, chloroform, methanol and water had no effect on the test organism (Figure 2) as shown by its zero millimetre zone of inhibition. This shows that the solvent of reconstitution of the extracts had no effect on the antibacterial activity of the extracts. This agrees with the findings of Dickson *et al*., [41] and Kumuda *et al*., [42]. The standard antibiotic susceptibility to the test organism is in the order gentamycin > chloramphenicol > amoxicillin. This is in agreement with the report of Dickson *et al*., [41] Akinyemi *et al.* [43], Ugboko and De [44] and Matthew *et al.,* [45] who all reported the resistance of *S.* Typhi to chloramphenicol in Nigeria. This confirms the gradual cessation of the prescription of chloramphenicol in the treatment of typhoid fever. The root, stem and leaf extracts of *F. capensis* inhibited the growth of *S.* Typhi in a concentration dependent manner except the root extract of the n-hexane extract with values comparable to standard antibiotics. This result in agreement to the finding of Dickson *et al*., [41] but disagrees with the finding of Oyeleke *et al.,* [11] who reported that the methanol leaf and stem bark extracts of *F. capensis* produced no activity against *S. typhi*. The difference in our finding with that of Oyeleke *et al.* [11] could be due to differences in the *S. typhi* strain used or perhaps the source of the plant sample since several studies have shown that geographical location, age of plant at harvest, season of harvest, postharvest storage, solvent of extraction and method of extraction influence the secondary metabolites present in a plant. This by extension will influence their pharmacological activity [46-53].

Table 1: Percentage yield of *F. capensis* roots, stem bark and leaves extracts

Plant part	Percentage yield			
	n-Hexane	Chloroform	Methanol	Water
Leaves	0.759	1.526	3.965	11.548
Stem bark	0.951	0.156	1.245	1.015
Roots	0.764	0.459	2.033	4.862

Table 2: Phytochemical composition of *F. capensis* roots, stem bark and leaves

Phytochemical	Concentration (mg/g)		
	Roots	Stem Bark	Leaves
Cardiac Glycoside	0.53	0.30	0.35
Flavonoid	1.89	0.53	4.20
Alkaloid	0.80	1.80	7.00
Saponin	0.60	0.18	0.80
Phenol	110.21	204.39	494.95
Tannin	3.19	2.64	2.57
Phytate	0.16	0.14	0.30
Oxalate	0.02	0.03	0.05

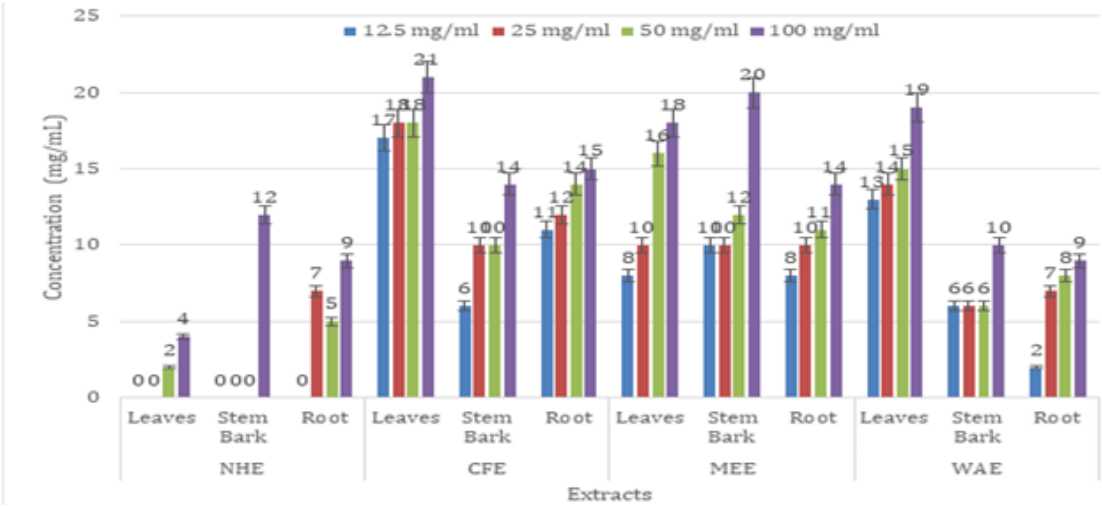

Figure 1: Zone of Inhibition for the extracts of *F. capensis* against *S. typhi* at different concentrations (NHE = N – Hexane Extract, CFE = Chloroform Extract, MEE = Methanol Extract, WAE = Water Extract)

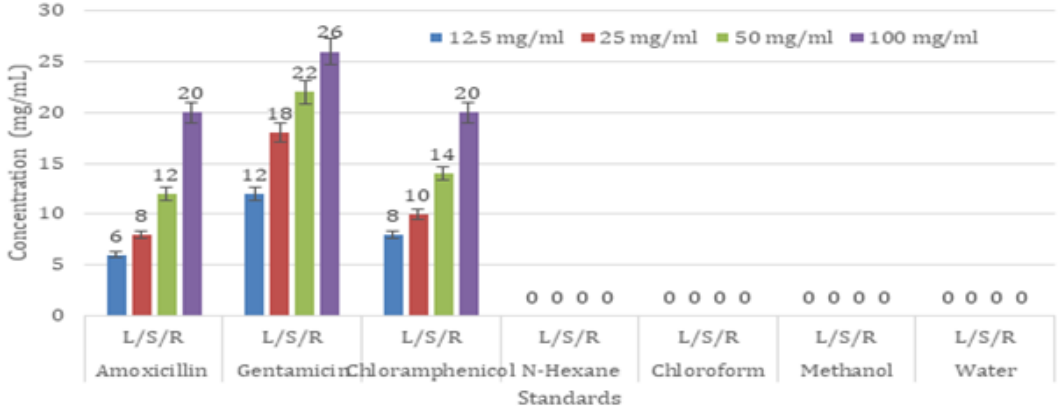

Figure 2: Zone of Inhibition for the positive (standard antibiotics) and negative (solvent) controls against *S. typhi* at different concentrations (L/S/R = Leaf, Stem and Root Extract)

Several phytochemicals like tannins, alkaloids, saponins, flavonoids, and phenols have been known to possess antibacterial properties including *Salmonella* Typhi as reported by Kennedy and Wightman [54], Choudhury *et al*., [55] and Ogbiko *et al*. [3]. They also reported that the presence of more group of phytochemical diversity gives synergic effects in many biological applications. The disparity between the activities of the extracts and the standard antimicrobial drugs may be due to the mixtures of bioactive compounds present in

the extracts compared to the pure compound contained in the standard antibiotics 3,56]. These phytochemicals are all present in *F. capensis* extracts hence the extract is effective against *Salmonella* Typhi and can be used for suppression of typhoid fever.

Conclusion. Considerable antimicrobial activities of the different plant extracts were recorded in this work and these would be as a result of the presence of the bioactive compounds in *Ficus capensis*. This study has confirmed and justified the use of *F. capensis* as an herbal preparation for the treatment of typhoid fever. This research is a promising start for future research on the isolation and characterization of potent anti-typhoid agent from the root, stem and leaf of *F. capensis*.

REFERENCES

1. Abushama MF, Hilmi YI, Abdalgadir HM, Fadul E, Khalid HE. Lethality and antioxidant activity of some Sudanese medicinal plants fixed oils. *European. J. Med. Plants*. 2014; 4(5): 563-570.

2. Vasanthi P, Ganapathy M, Evanjelene VK, Ayyavuv N, Angamuthu J. Phytochemical screening and antioxidant activity of extracts of the leaf and bark of *Albizzia lebbeck* (Benth). *Academia Journal of Medicinal Plants.* 2014; 2(2): 26-31.

3. Ogbiko C, Eboka CJ, Tambuwal AD. Extraction and Fractionation of Whole Black Seed Plantain (*Plantago rugelii* Decne) for *in-vitro* Antioxidant, Antibacterial and Phytochemical Screening. *J. Appl. Sci. Environ. Manage*. 2018; 22(5): 613– 618.

4. Parry CM, Hien TT, Dougan G, White NJ, Farrar JJ. Typhoid fever. N Engl *J Med*. 2012; 347(22): 1770-1782.

5. Parry CM. Untreatable infections? The challenge of the 21st century. *South-east Asian J Trop Med Public Health*. 1998; 29(2): 416-24.

6. Birgitta J, Yvonne A, Carl E. Effect of regulation and education on reptile associated salmonellosis. *Emerging Infectious Diseases*. 2005; 9(5): 585 – 591.

7. Edelman R, Levine MM. Summary of an international workshop on typhoid fever. *Rev Infect Dis*. 1986; 8(3): 329-349.

8. Gills LS. Ethnomedical Uses of Plants in Nigeria. Uniben Press, Edo State, Nigeria. 1992; 121 p.

9. Ayinde BA, Owolabi OJ. Effect of the aqueous extract of *Ficus capensis* Thunb. (Moraceae) leaf on gastrointestinal motility. *Journal of Pharmacognosy and Phytotherapy*. 2009; 1(3): 31-35.

10. Ahmadu AA, Zezi AU, Yaro AH. Anti-diarrheal activity of the leaf extracts of *Daniella oliveri* Hutch and Dalz (Fabaceae) and *Ficus sycomorus* Miq (Moraceae). *African Journal of Traditional, Complementary, and Alternative Medicine*. 2007; 4 (4): 524-528.

11. Oyeleke SB, Dauda BEN, Boye OA. Antibacterial activity of *Ficus capensis*. *African Journal of Biotechnology*. 2013; 7(10): 1414-1417.

12. Adebayo-Tayo BC, Odeniyi AO. Phytochemical screening and microbial inhibitory activities of *Ficus capensis; Afr. J.f Biomed. Res.* 2012; 15: 35-40.

13. Umeokoli BO, Onyegbule FA, Gugu TH, Igboeme SO. Evaluation of the erythropoietic and anti-sickling properties of *Ficus capensis* leaf extract in the treatment of anaemia. *Planta Medica*. 2013; 79(13): 79 – 89.

14. Mpiana PT, Mudogo V, Tshibangu DST, Kitwa EK, Kanangila AB, Lumbu JBS, Ngbolua KN, Atibu EK, Kakule MK. Antisickling activity of anthocyanins from *Bombax pentadrum*, *Ficus capensis*, *Zizyphus mucronata*: Photo degradation effect. *J. Ethnopharmacol*. 2008; 120: 413-418.

15. Owolabi OJ, Nworgu ZA, Falodun A, Ayinde BA, Nwako CN. Evaluation of tocolytic activity of ethanol extract of the Stem bark of *Ficus capensis* Thunb. (Moraceae). *Acta Poloniae Pharmaceutica ñ Drug Research*. 2009; 66(3): 293–296.

16. Daikwo OA, Tende JA, Okey SM, Eze ED, Isa AS. The effect of aqueous extract of leaf of *Ficus capensis* Thunb (Moraceae) on *in vivo* Leukocyte Mobilization in Wistar Rats. *British Journal of Pharmacology and Toxicology*. 2012; 3(3): 110-114.

17. Owolabi OJ. Active ileum relaxant fractions from the leaves of *Ficus capensis* Thunb (Moraceae). *Nigerian Journal of Pharmaceutical Sciences*. 2013; 12(1): 1-7.

18. Ramde-Tiendrebeogo A, Tibiri A, Hilou A, Lomp M, Millogo-Kone H, Nacoulma OG, Guissou IP. Antioxidative and antibacterial activities of phenolic compounds from Ficus sur Forssk. and *Ficus sycomorus* L. (Moraceae): Potential for sickle cell disease treatment in Burkina Faso. *Int. J. Biol. Chem. Sci*. 2012; 6(1): 328-336.

19. Gelfand M, Mavi S, Drummond RB, Ndemera B. The traditional medicinal practitioner in Zimbabwe. Mambo Press, Gweru, Zimbabwe. 1985: 411 p.

20. Butler T, Linh NN, Arnold K, Pollack M. Chloramphenicol resistant typhoid fever in Vietnam associated with R factor. *Lancet.* 1973*;* 2(836): 983-985.

21. Chen S, Zhao S, White DG, Schroeder CM, Lu R, Yang H. Characterization of multiple-antimicrobial-resistant salmonella serovars isolated from retail meats. *Appl Environ Microbiol*. 2004; 70(1): 1-7.

22. MacFaddin JF. Biochemical Tests for Identification of Medical Bacteria. 3rd Edition. ASM Press, Washington, DC. 2000; 87 p.

23. Fobres BD, Sham A. Weissfield. Diagnostic Microbiology. 11th Edition, Andrew Allen Press, Canada, 2002; 56-59 p.

24. Leboffe MJ, Pierce BE. Microbiology Laboratory Theory and Application. 1st Edition, Morton Publishers, USA. 2002; 76-79 p.

25. Sneath PHA, Mair NS, Sharpe ME, Holt JG. Bergey's Manual of Systematic Bacteriology. Vol. 2, Williams and Wilkins, Philadephia. 1986; 98 p.

26. Rajarkaruna N, Harris CS, Towers GHN. Antimicrobial activities of plants collected in Sri Lanka. *Journal of Pharmaceutical Biology*. 2002; 40(3): 235-224.

27. Sofowora A. Medicinal Plants and Traditional Medicine in Africa, Spectrum Books Limited, Ibadan. 1993; 199-204 p.

28. Evans WC. Trease and Evans' Pharmacognosy, 15th ed., Churchill Livingstone Harcourt publishers Limited, London. 2002; 204-393 p.

29. Odama IE, Shok M, Olurinola PF. *Ceiba petandra* (Silk cotton tree), Microbiological evaluation of its use in traditional wound treatment In: The State of Medicinal Plant Research in Nigeria (Sofowora, A. Ed.) Ile-Ife Univ. Press Ile-Ife, 1986; 253-259 p.

30. Musa AD, Nwodo OFC, Ojogbane EB. Phytochemical, Antibacterial and Toxicity Studies of The Aqueous Extract of *Eucalyptus camaldulensis* Dehnh. *Asian Journal of Plant Science and Research*. 2011; 1(3): 1-10.

31. Ehimwenma SO, Osarieme TO. *In vitro* antioxidant activity and phytochemical screening of methanol extracts of *Ficus capensis* and *Dacryodes edulis* leaves. *Journal of Pharmacy and Bioresources*. 2014; 11(2): 66-75.

32. Okwu DE. Phytochemicals and vitamin content of indigenous spices of South Eastern Nigeria. *Journal of Sustainable Agriculture and Environment*. 2004; 6: 30 – 37.

33. Nagavani V, Madhavi YD, Bhaskar RP, Koteswara R, Raghava RT. Free radical scavenging activity and qualitative analysis of polyphenols by RP-HPLC in the flowers of *Couroupita guianensis*. *Electronic Journal of Environmental, Agricultural and Food Chemistry*. 2010; 9(9): 1471-1484.

34. Parajuli S, Pun TN, Parajuli, N, Jamakattel-Pandit, N. Antioxidant activity, total phenol and flavonoid contents in some selected medicinal plants of Nepal. *Journal of Health and Allied Sciences*. 2012; 2(1): 27-31.

35. Hamzah RU, Egwim EC, Kabiru AY, Muazu MB. Phytochemical and *in vitro* antioxidant properties of the methanolic extract of fruits of *Blighia sapida, Vitellaria paradoxa* and *Vitex doniana*. *Oxidant, Antioxidant and Medical Sciences.* 2013; 2(3): 215-221.

36 Hamzah RU, Jigam AA, Makun HA, Egwim EC. Phytochemical screening and *in vitro* antioxidant activity of methanolic extract of selected Nigerian vegetables. *Asian Journal of Basic and Applied Sciences*. 2014; 1(1): 1-14.

37. Sodipo OA, Akiniyi JA, Ogunbamosu JU. Studies on certain characteristics of extracts of bark of *Pansinytalia macruceras*. *Global Journal of Pure and Applied Sciences*. 2000; 6: 83-87.

38. Chinedu SN, Olasumbo AC, Eboji OK, Emiloju OC, Arinola OK, Dania DI. Proximate and phytochemical analyses of *Solanum aethiopicum* L. and *Solanum macrocarpum* L. fruits. *Research Journal of Chemical Sciences*. 2011; 1(3): 63-71.

39. Okwu DE, Josiah C. Evaluation of the chemical composition of two Nigerian medicinal *plants. African Journal of Biotechnology*. 2006; 5(4): 357-361.

40. Solomon-Wisdom GO, Shittu GA, Agboola YA. Antimicrobial and Phytochemical Screening Activities of *Ficus Sur* (Forssk). *New York Science Journal*. 2011; 4(1): 15-18.

41. Dickson AM, Gloria OS, Amanabo M, Okwesili, FCN. Evaluation of *in vitro* anti-salmonelal effect of combined crude aqueous extracts of *Ficus capensis* and *Nauclea latifolia* used for the treatment of typhoid fever. *Biokemistri*. 2017; 29(1): 19–23.

42. Kumuda KV, Shashidhara S, Rajasekharan PE, Ravish BS. Study on In vitro antityphoid activity of various root extracts of *Decalepis hamiltonii* (Wight and Arn.). *International Journal of Pharmaceutical & Biological Archives*. 2011; 2(1): 546-548.

43. Akinyemi KO, Coker AO, Olukoya DK, Oyefolu AO, Amorighoye EP, Omonighehin EO. Prevalence

of multi-drug resistant *Salmonella typhi* among clinically diagnosed typhoid fever patients in Lagos, Nigeria. *Zeitschrift für Naturforschung C, A. Journal of Biosciences*. 2000; 55(5-6): 489-493.

44. Ugboko H, De N. Mechanisms of Antibiotic resistance in *Salmonella typhi. International Journal of Current Microbiology and Applied Sciences*. 2014; 3(12): 461-476.

45. Matthew B, Ugboko H, De N. Prevalence of Multidrug Resistant *Salmonella enterica* Serovar Typhi in Kaduna Metropolis, Kaduna, Nigeria. *International Journal of Current Microbiology and Applied Sciences*. 2015; 4(9): 323-335.

46. Calixto JB. Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines (phytotherapeutic agents). *Brazilian Journal of Medical and Biological Research*. 2000; 33: 179-189.

47. Fennell CW, Light ME, Sparg SG, Stafford GI, Van Staden J. Assessing African medicinal plants for efficacy and safety: agricultural and storage practices. *Journal of Ethnopharmacology*. 2004; 95(2-3): 113–121.

48. Ughachukwu PO, Ezenyeaku CCT, Ezeagwuna DA, Anahalu IC. Evaluation of antibacterial activities of *Euphorbia heterophylla. IOSR Journal of Dental and Medical Sciences*. 2014; 13(11): 69-75.

49. Ullah N, Khurram M, Amin MU, Khan TA, Khayyam SU, Khan FA, Najeeb U, Ullah S. Impact of geographical locations on *Mentha spicata* antibacterial activities. *Journal of Medicinal Plants Research*. 2012; 6(7): 1201-1206.

50. Ndhlala AR, Amoo SO, Ncube B, Moyo M, Nair JJ, Van Stadan J. Antibacterial, Antifungal, and Antiviral Activities of African Medicinal Plants. In: Medicinal Plant Research in Africa, Pharmacology and Chemistry. Eds Kuete,V. Elsevier, London. 2013; 621-651 p.

51. Ughachukwu PO, Ezenyeaku CCT, Ezeagwuna DA, Anahalu IC. Evaluation of anti-bacterial properties of ethanol extract of *Ficus exasperata* leaf. *African Journal of Biotechnology*. 2012; 11(16): 3874-3876.

52. Makanjuola SO, Oluwadun A, Thomas BT, Kolawole RM. Influence of Geographical Locations on the Antibacterial Activities of *Aloe vera* Accessions. *Applied Journal of Hygiene*. 2015; 4(2): 25-28.

53. Yadav M, Yadav A, Kumar S, Yadav JP. Spatial and seasonal influences on culturable endophytic mycobiota associated with different tissues of *Eugenia jambolana* Lam. and their antibacterial activity against MDR strains. *BMC Microbiology*. 2016; 16: 44 - 52.

54. Kennedy DO, Wightman EL. Herbal extracts and phytochemicals: Plant secondary metabolites and enhancement of human brain function. *Advances in Nutrition*. 2011; 2: 32-50.

55. Choudhury S, Sharan L, Sinha MP. Phytochemical and antimicrobial standardization of the methanolic leaf extracts of *Murraya koenigii* Linn. *Archives Des Sciences*. 2013; 66(3): 67-80.

56. Gatsing D, Nkeugoauapi CFN, Nkah BFN, Kuiate JR, Tchouanguep FM. *Chenopodium ambrosioides* Linn., an indigenous medicinal plant. *Afr. J. Pharm. and Pharmacol*. 2010; 8(8): 231-234.