WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.074
Volume 8, Issue 3, 147-155. Research Article ISSN 2277– 7105

DETERMINING THE ANTIMICROBIAL ACTIVITY OF THE EXTRACTS OF GINGER AND TURMERIC INCLUDING ITS AQUEOUS, ETHANOLIC AND n-HEXANE EXTRACTS AND DETERMINING ITS LEVEL OF EFFICACY COMPARED TO A STANDARD ANTIMICROBIAL DRUG

***[1]Ike C. O., [2]Orjioke N. M., [1]Okafor G. U., [1]Udeozo I. P., [3]Ugwu M. C., [1]Agu F. C. and [1]Chukwu S. C.**

[1]Department of Industrial Chemistry, Enugu State University of Science and Technology, Enugu State, Nigeria.

[2]Department of Science Laboratory and Technology, University of Nigeria Nsukka, Enugu State, Nigeria.

[3]Department of Medical Laboratory Science, College of Health Sciences, Nnamdi Azikiwe University, PMB 5001Nnewi, Anambra State, Nigeria.

Article Received on
31 Dec. 2018,

Revised on 21 Jan. 2019,
Accepted on 10 Feb. 2019

DOI: 10.20959/wjpr20193-13968

***Corresponding Author**
Ike C. O.
Department of Industrial Chemistry, Enugu State University of Science and Technology, Enugu State, Nigeria.

ABSTRACT

Ginger and Turmeric are not just used for culinary but exhibits good therapeutic qualities used for the prevention and treatment of various ailments. Chemical analysis has proved that each consists of over 400 different phytochemicals. This work investigates the antimicrobial activities of the aqueous and the inorganic extracts of ginger and turmeric with gram +ve (S. aureus) and gram – ve (E. coli and S. typhi) strains using Ciprofloxacin as the standard antibiotic. Disc diffusion and serial dilution were the methods used in carrying out the tests. The zone of inhibition which determines if the bacteria are resistant, intermediate or susceptible to the extracts and minimum inhibitory concentration which determines the lowest value at which the extracts are efficient. The zone of inhibition ranged from 9-13 mm for the ginger extracts and 11-16.2 mm for the turmeric extracts while the MIC values ranged 0.05-.06 mg/ml for the ginger extract and 0.1-1.0 mg/ml for the turmeric extract. In conclusion, the test bacteria showed susceptibility to all extracts, however, the bacteria showed overall higher susceptibility to the ginger extracts.

KEYWORDS: Aqueous Extract, Ginger, Tumeric, n-Hexane, Ethanol, antimicrobial agent.

INTRODUCTION

Humans developed antimicrobials to destroy disease-causing microbes. The most commonly known antimicrobials are antibiotics, which target bacteria (Giedraitienė et al., 2011). Other forms of antimicrobials are antivirals, antifungals, and antiparasitics (Okonko, et al., 2008). Antimicrobial drugs have caused a dramatic change not only in the treatment of infectious diseases but in the fate of mankind (Saga et al., 2009). Antimicrobial chemotherapy made remarkable advances, resulting in the overly optimistic view that infectious diseases would be conquered in the near future (Pathak et al., 2018). However, in reality, emerging and re-emerging infectious diseases have left us facing a counter-charge from infections (Iroha et al., 2013). Infections with drug-resistant organisms have become a grave problem in clinical practice that is difficult to solve. (Saga, 2009).

Infections caused by antimicrobial-resistant micro-organisms are difficult to treat, resulting in poor outcomes for patients and economic impacts through increased healthcare costs and lost productivity (Regea, 2018).The incidence of AMR is rapidly increasing in frequency and geographical spread. A recent high profile report estimates that, by 2050, 10 million people will die every year due to AMR unless a global response to the problem of AMR is mounted (Nikhil et al, 2017).

Ginger as a natural antibiotic is the earliest known medicinal plant. It has been shown to be effective in treating diseases in humans, poultry and aquaculture owing to its antimicrobial, antioxidant, growth promotion and immune stimulant properties. An optimized dose of ginger is recommended in diets. Ginger has been used as a spice for over 2000 years (Shubha, 2015). It is also called “The Great Medicament” in Ayurvedic medicines and is generally considered as a safe herbal medicine (Yuan et al., 2016). Ginger contains natural organic materials beneficial to health and enhances resistance to infectious diseases by increasing non-specific and specific immune mechanisms (Shagufta, 2010).The rhizome of ginger has been shown to be effective in the control of a range of illnesses including diarrhea, stomach ache, asthma etc. (Yassan *et al.*, 2016).

Turmeric is an ancient spice derived from the rhizomes of curcuma longa, which is a member of the ginger family (Anusha et al., 2016). According to the Indians, turmeric is known as the golden spice. This rhizome is also used in India for medicine purposes; traditional medicine

as a household remedy for various diseases including anorexia, cough, diabetic wounds, hepatic disorders etc (Yadav et al., 2017). In addition to its use as spice, turmeric and its components especially curcumin and essential oils so wide spectrum of biological actions (Rathaur et al., 2012). Turmeric contains proteins (6.3%), fat (5.1%), minerals (3.5%), carbohydrate (69.4%) and moisture (23.1%). Curcumin is one of the curcuminoids and is a bioactive compound found in turmeric(Faten et al., 2016). It is commonly referred to as holy powder and is a natural antioxidant. Turmeric is widely applied in culinary, dye, as indicator for chemical analysis and traditional uses (Bagchi Anamika, 2012).

This study is geared at determining the antimicrobial activity of the extracts of ginger and turmeric including its aqueous, ethanolic and n-hexane extracts and hence determine its level of efficacy compared to a standard antimicrobial drug (ciprofloxacin) which is itself in conformity with standards set by the National Agency for Food and Drug Administration and Control (NAFDAC) and the World Health Organization (WHO, 2001).

MATERIALS AND METHODS

Preparation of Extracts

Three types of extracts; aqueous, ethanol and n-hexane extracts each from ginger and turmeric were prepared separately. The fresh ginger and turmeric rhizomes were washed, peeled, sliced and oven dried at low heat to remove all moisture. After drying, the ginger and turmeric rhizomes were ground to fine powder separately using an electric blender.

Soxhlet Extraction

This process was used for both the ethanolic and n-hexane extracts of ginger and turmeric and is named as Ginger ethanol extract and Turmeric ethanol extract, Ginger n-hexane extract and Turmeric n-hexane extract.

The ground plant material was placed in the extraction chamber which was suspended above the flask containing ethanol and n-hexane solvents respectively and below a condenser.

The flask was heated and the solvent evaporated and moved into the condenser where it was converted into a liquid that trickled into the extraction chamber containing the plant material.

The extraction chamber was designed so that when the solvent surrounding the sample exceeded a certain level, it overflowed and trickled back down into the boiling flask. At the end of the extraction process, the flask containing the ethanol and n-hexane extracts was

removed and each concentrated on a water bath at 78^{o}C and then left to dry at room temperature.

Aqueous Extraction

The ground plant samples of ginger and turmeric were extracted with distilled water by maceration. 30g of the samples was soaked with 200ml of distilled water for 72h at room temperature. After that, the resulting extracts was filtered using filter paper (Whatman no.) The filtrates obtained were concentrated by heating over water bath. The extracts were labeled Ginger Aqueous Extract and Turmeric Aqueous Extract respectively.

Preparation of Test Bacteria

Three different bacterial strains were used. Two species of gram negative bacteria, E. coli and S. typhi and one gram positive specie, S. aureus were obtained from stock cultures of the department of Applied Microbiology, Esut, Nigeria. The bacteria were sub cultured on appropriate agar then incubated at 37^{0}c overnight. They were then standardized by matching to the 0.5 McFarland turbidity standards using sterile saline to produce approximately 1.5x10^{8} colony forming units (cfu) per ml.

Antimicrobial Assay

As described in the NCCLS manual (CLSI, 2015) the surface of the Mueller Hinton agar plate sub cultured was divided into quadrants by drawing horizontal and vertical lines across the middle point. The plates are inoculated by streaking evenly the suspension of the test organism (E. coli) using sterile cotton swabs. The plates were kept aside and left to air dry.

Paper disks (6mm) were then impregnated with the extracts labeled Ginger ethanol extract, Turmeric ethanol extract, Ginger aqueous extract, Turmeric aqueous extract Ginger n-hexane Extract and Turmeric n-hexane Extract and kept aside. Ciprofloxacin (10µg) disks were used as standard.

To each quadrant of the agar plate, inoculate a paper disk using forceps sterilized before and after use by dipping in ethanol and passing through a Bunsen burner. Plates used were incubated at 37^{o}C for 24hrs. After incubation period, the plates were examined and the diameter of each zone was measured for inhibition zone around the disks for each extract and recorded. The procedure was repeated for the two other test bacteria.

Determination of Minimum Inhibitory Concentration

Ginger and turmeric extracts with different solvents were examined for their Minimum Inhibitory Concentrations using method described by Iram Gull et al (2012). The extracts were diluted ranging from 100mg/ml to 0.01mg/ml and studied for their effects against the various bacterial strains. Sterile discs were dipped in different dilutions of the aqueous, ethanolic and n-hexane extracts of ginger and turmeric and placed over agar plates seeded with uniform concentration of each bacterial culture separately. The zone of inhibition in each case was measured as the diameter of the clearing zones and recorded.

RESULTS

The results obtained in this study are summarized below. Table 1 shows the antibacterial activity of ginger extracts measured as diameter (mm) of zone of inhibition. The zone of inhibition for the standard is high when compared with the aqueous, ethanolic and n-hexane extracts. Also, the zone of inhibition is high for the ethanolic extract when compared with aqueous and n-hexane extract in *E. coli*. The ethanolic extract showed a high zone of inhibition when compared with aqueous and n-hexane extract in *S. aureus*. The n-hexane extract showed a high zone of inhibition when compared with the aqueous and ethanolic extract while the aqueous and ethanolic extract had the same zone of inhibition in *S. typhi*.

Furthermore, the aqueous extract had a high zone of inhibition in *S. typhi* compared to *E. coli* and *S. aureus*. Also, the ethanolic extract had high zone of inhibition in *E. coli* compared to *S. aureus* and *S.* typhi. The n-hexane extract had high zone of inhibition in *S.* typhi compared to *E. coli* and *S. aureus* in the study.

Table 1: Antibacterial activity of Ginger extracts measured as diameter (mm) of zone of inhibition.

Bacteria strain	Aqueous extract	Ethanol extract	n-Hexane extract	Standard
E. coli (-ve)	10.30	13.00	9.30	19.00
S. aureus (+ve)	9.00	9.30	8.20	22.00
S. typhi (-ve)	11.00	11.00	13.00	20.00

Table 2 shows the antibacterial activity of turmeric extracts measured as diameter (mm) of zone of inhibition. The zone of inhibition for the standard is high when compared with the aqueous, ethanolic and n-hexane extracts. Also, the zone of inhibition is high for the aqueous extract when compared with ethanolic and n-hexane extract in *E. coli*. The ethanolic extract showed a high zone of inhibition when compared with aqueous and n-hexane extract in *S.*

aureus. The aqueous extract showed a high zone of inhibition when compared with the ethanolic and n-hexan extract while the ethanolic and n-hexane extract had the same zone of inhibition in *S. typhi*.

Furthermore, the aqueous extract had a high zone of inhibition in *S. aureus* compared to *E. coli* and *S. typhi*. Also, the ethanolic extract had high zone of inhibition in *S. aureus* compared to *E. coli* and *S.* typhi. The n-hexane extract had high zone of inhibition in *S. aureus* compared to *E. coli* and *S.* typhi in the study.

Table 2: Antibacterial activity of Turmeric extracts measured as diameter (mm) of zone of inhibition.

Bacteria strain	Aqueous extract	Ethanol extract	n-Hexane extract	Standard
E. coli (-ve)	14.20	13.20	12.20	19.00
S. aureus (+ve)	15.00	16.20	14.00	22.00
S. typhi (-ve)	12.00	11.00	11.00	20.00

The results in table 3 and 4 have shown that the MIC values of turmeric and ginger against bacterial strains ranged from 0.05 mg/ml to 0.1 mg/ml. The data in table 3 shows that the ethanol and n-hexane extract of ginger had lower MIC values in comparison to the aqueous extract tested against the bacterial strains. In the case of the extracts, the lowest MIC value for *E. coli* 0.08 mg/ml, S.aureus 0.5 mg/ml and S. typhi 0.09 mg/ml was observed with the ginger extract. The data in table 4 indicates the lowest values of turmeric extracts for E. coli, S.aureus and S. typhi were 0.1 mg/ml, 0.2 mg/ml and 0.2 mg/ml.

Table 3: Minimum Inhibitory Concentration of Ginger in mg/ml against test bacteria.

Bacteria strain	Aqueous extract	Ethanol extract	n-Hexane extract	Standard
E. coli (-ve)	0.1	0.05	0.08	0.00018
S. aureus (+ve)	0.6	0.5	0.5	0.0005
S. typhi (-ve)	0.2	0.1	0.09	0.00026

Table 4: Minimum Inhibitory Concentration of Turmeric in mg/ml against test bacteria.

Bacteria strain	Aqueous extract	Ethanol extract	n-Hexane extract	Standard
E. coli (-ve)	0.1	0.3	0.2	0.00018
S. aureus (+ve)	0.2	0.8	1.0	0.0005
S. typhi (-ve)	0.2	0.9	1.0	0.00026

DISCUSSION

The antimicrobial effect of ginger and turmeric was evaluated by disc diffusion method. The results indicated that the different extracts of the spices have broad spectrum antimicrobial activity with variable degrees of sensitivity of the test bacteria toward the extracts.

The data showed in table 1 and 2 reveals that all the turmeric extracts exhibited higher inhibition zone on all test bacteria except on the ethanolic or n-hexane extracts of ginger which showed equal or higher degree of inhibition. These findings are in accordance with Mohammad *et al*, (2010). Similarly, the essential oil extracted from Zingiber officinale exhibited activity against food-borne pathogenic fungal and bacterial species (Singh et al., 2008). Though the exact antibacterial mechanism of action of ginger and turmeric is not yet clear, there are hypothesis which involves hydrophobic and hydrogen bonding of phenolic compounds to membrane proteins, followed by partition in lipid bilayer, membrane disruption, destruction of electrons transport systems, cell wall perturbation (Souza et al., 2005).

The minimum inhibitory concentration was determined by making dilutions of the different extracts of ginger and turmeric using method described by Iram Gull et al., (2012). It was interesting to note that, both gram +ve and gram –ve bacteria were sensitive to all the extracts of ginger and turmeric but gram -ve bacteria showed overall sensitive than the gram +ve bacteria. This result is in accordance with results seen in Chandarana et al., (2005) and Onyeagba et *al.*, (2004).

CONCLUSION

The results of this present study have provided justification for the therapeutic potential of medicinal plants. The practice of using this plants as supplementary or alternative medicine in developing countries like Nigeria will not only reduce the clinical burden of drug resistance development but also side effects and cost of treatment with allopathic medicine. However, further clinical evaluation of medicinal plants in vivo experiments is required to be carried out for enablement of low cost treatment with little or no side effects and for the prevention of recurring infections.

ACKNOWLEDGEMENTS

Our immense gratitude goes all those who contributed to the success of this research.

REFERENCES

1. Anusha, A., Kumar, R and Shaik, R. Phytochemical Evaluation Curcuma Longa and Curcumin; *International Journal Of Pharmaceutical And Chemical Sciences*, 2016; 5(4): 5005–2277.
2. Bagchi Anamika: Extraction of Curcumin. *Journal of Environmental Science, Toxicology and Food Technology*, 2012; 1(3): 01-16.
3. Chandarana, H., Baluja, S and Chanda, S.V: Comaparision of Antibacterial Activities of Selected Spices of Zingiberene Family and Some Synthetic Compounds. *Turk Journal of Biological sciences*, 2005; *29: 83-97.*
4. De-Souza, E.L., Stamford, L.M., Lima, E.D., Trajano, V.N and Barbosa J.M. Antimicrobial effectiveness of spices: an approach for use in food conservation systems. *Brazillian Archives of Biology and Technology, 2005; 48(4): 549-558.*
5. Faten, F.A. Hammam, O., El-ahwany, E., El-wakil, E., Kandil, S., Taleb, H., El-sayed, M, Hassenein, H. Bioactive chemical constituents of Curcuma longa L. rhizomes extract inhibit the growth of human hepatoma cell line, *Acta Pharm*, 2016; 66: 387–398.
6. Giedraitienė, A., Vitkauskienė, A., Naginienė, R., Pavilonis, A. Antibiotic Resistance Mechanisms of Clinically Important Bacteria. *Medicina (Kaunas)*, 2011; 47(3): 137-46.
7. Gull, I., Saeed, M., Shauhat, H., Aslam, M., Samra, Q., Athar, M. Inhibitory Effect Of Allium Sativum And Zingibere Officinale Extracts On Clinically Important Drug Resistant Pathogenic Bacteria. *Annals of Clinical Microbiology and Antimicrobials,* 2012; *11: 8.*
8. Iroha, I. R, Nwakeze, A.E., Afiukwa F. N., UduIbiam, E. O., Nwuzo, A.C., Oji A.E and Ngwu T. N; *International Research Journal of Basic and Clinical Studies*, 2013; 1(4): 46-52.
9. Nikhil, S., Sangeeta, G. and Vaibhav, R. Comparative Antimicrobial Study of Ethanolic Extract of Leaf and Rhizome of Curcuma longa Linn. *Pharmacogn J., 2017; 9(2): 208-2.*
10. Onyeagba, R.A., Ugbogu, O.C., Okeke, C.U and Irokasi O: Studies on the antimicrobial effects of garlic (Allium sativum Linn), ginger (Zingiber officinale Roscoe) and lime (Citrus aurantifolia Linn). *African Journal of Biotechnology*, 2004; *3: 552-554.*
11. Okonko, I.O., Fajobi, E.A., Ogunnusi, T.A., Ogunjobi, A.A. and Obiogbolu, C.H.(2008): Antimicrobial chemotherapy and Sustainable Development: The past, The Current Trend, and the future, *African Journal of Biomedical Research*, 2008; 11: 235–250.

12. Pathak, M., Kumar, A., Singh, U.K. Phytochemical Screening & Antibacterial activity of Scindapsus officinalis fruit extract against Human Pathogens: An Ethanomedicinal plant; *Journal of Pharmacy and Biological Sciences*, 2018; 3(4): 41-48.
13. Regea, G. Review on Antibiotics Volume 13, Issue 5, PP 36-40 Resistance and its Economic Impacts. *Journal of Pharmacology & Clinical Research*, 2018; 2473-5574.
14. Saga, T., Yamaguchi, K. History of Antimicrobial Agents and Resistant Bacteria. *Japan Medical Association — Journal*, 2009; 52(2): 103–108.
15. Shagufta, N., Safia, J., Saiqa, I., Farkhanda, M., Farah, A. and Aamir, A.L. Antibacterial Activity of Curcuma Longa Varieties against Different Strains of Bacteria, *Pak. Journal of Botany, 2010; 42(1): 455-462.*
16. Shubha, R.S. Medicinal uses of ginger (Zingiber officinale Roscoe) improves growth and enhances immunity in aquaculture, *International Journal of Chemical Society*, 2015; 3(2): 83-87.
17. WHO. Legal Status of Traditional Medicine and Complementary/ Alternative medicine: A world wide review. WHO Publishing 1, 2001.
18. Yadav, P.R. and Tarun, G. Versatility of turmeric: A review the golden spice of life; *Journal of Pharmacognosy and Phytochemistry*, 2017; 6(1): 41-46.
19. Yassen, D., Ibrahim, E. Antibacteerial Activity of Crude Extracts of Ginger (Zingiber Officinale Roscoe) On Escheria Coli and Staphylococcus Aureus: A Study in Vitro. *Indo American Journal of Pharmaceutical Research, 6(06).*
20. Yuan, H., Ma, Q., Ye, L. and Piao, G. The Traditional Medicine and Modern Medicine from Natural Products. *Molecules*, 2016.