MedCrave Step into the World of Research
International Journal of Complementary & Alternative Medicine

Research Article

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Studies on the synergistic effectiveness of methanolic extract of leaves and roots of *Carica papaya* L. (papaya) against some bacteria pathogens

Abstract

Carica papaya L. (Papaya) is a major food crop with several nutritional values. Different parts of the plant are used for the treatment of several types of diseases among traditional health practitioners in many developing countries including Nigeria. This study investigated the synergistic effectiveness of methanolic leaves and roots extract of papaya. The dried roots and leaves of papaya were extracted using methanol for 48hours and agar well diffusion method was used to test for sensitivity. The mean zone of inhibition by *Staphylococcus aureus*, *Escherichia coli* and *Bacillus subtilis* was 11.33mm, 11.00mm and 9.00mm, respectively for leaves, 10.00mm, 10.33mm and 10.33mm, respectively for roots, and 11.67mm, 12.00mm, 10.00mm, respectively for synergy of roots and leaves at 100% concentration. Significant variation ($P<0.05$) exists at 100% and 90% concentrations of the extracts. The study revealed that the synergy of roots and leaves had slight superior effectiveness against *Staphylococcus aureus*, *Escherichia coli* and *Bacillus subtilis*.

Keywords: antibacterial, *Carica papaya*, human health, medicinal plants, microbial pathogens

Volume 11 Issue 6 - 2018

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Received: May 21, 2018 | **Published:** November 20, 2018

Introduction

Antimicrobial resistance is a major global concern.[1–8] In addition, emerging and reemerging microbial infections are also major concern to public health[7,9–12] especially in tropical nations. These have led to research into alternative means of controlling bacterial diseases. Dey et al.,[8] also reported overuse of antibiotics is contributing to growing number of infections that are becoming resistant due to mutation as a result of increased exposure to medications. Typically, resistance to antimicrobial agents has resulted to treatment failure and increased health care cost and in severe instances leading to morbidity and mortality.[8]

Carica papaya (Paw-paw or papaya) which belong to the Caricaceae family is an important food crop in many tropical countries including Nigeria. Several varieties of papaya exist which are mainly distinguished based on their shape and endocarp colour (red, orange, yellow etc).[13] Papaya is known to contain several nutrients and vitamins.[13] According to Vij & Prashar[14] papaya has antioxidant properties and contain Vitamin B (such as panthothenic acid and folate), vitamin A, C and E, minerals such as magnesium and potassium, fiber and digestive enzyme (papain). Probably due to this it have been widely consumed raw and processed further especially as canned foods such as jam.[13,15]

Papaya has some medicinal values.[16] Papaya is used by folk medicine practitioners as herbal remedy for the prevention and treatment of several diseases. According to Panzarini et al.,[17] the curative potentials of papaya could be due to the presence of various bioactive and phytochemical nutrients. Igwe[18] have revealed the presence of Hexahydro-1-aHnaphtho[1,8a-b]oxiren-2(3H)-one (cyclic ketonic ether), 3,7-dimethyloct-7-en-1-ol (Alkene alcohol), 3-methyl-4-(phenylthio)-2-enyl-2,5- dihydrothiophene-1,1- dioxide (Aromatic thiophene dioxide), Cyclopentaneundecanoic acid, methyl ester (Fatty acid ester), 3,7,11,15-tetramethyl-2- hexadecen-1-ol (Alkene alcohol), 9-octadecenamide (Amide (alkaloid) in leaves of papaya. The authors further reported that the bioactive compound have pharmacological potentials.

According to Jaiswal et al.,[19] Vij & Prashar[14] the leaves, barks, roots, latex, fruit, flowers, and seeds of papaya is used for the treatment of several disease conditions. Papaya also confers several health benefits including improving cardiovascular system, protection against heart diseases, heart attack, stroke and prevention of colon cancer.[14] Based on the traditional claims about therapeutic potentials of the plants, several scientific validations have been made about the plants including its potentials as antimicrobial (viz: antifungal, antibacterial, antiviral), anti-amoebic, anti-carminative, anti-fertility, anti-tumor, anti-malaria, anti-cancer, anti-sickling, anti-helminthes, immune-modulatory, hypoglycemic, wound healing, diuretic, histaminergic, hepatoprotective, immunological, effect on muscle smoothing, hepatotoxicity, anti-inflammatory, anti-hypertensive and hypolipidemic, neuroprotective, abortifacient etc.[14,17,19–21] Several solvents have been used for the extraction of plant parts of papaya. Among the different parts studied, the leaves and root have been extracted using acetone, methanol, water, petroleum benzene, chloroform, ethanol, isopropanol, petroleum ether.[8,18,22–24] Despite the widely reported anti-microbial properties of papaya, the synergistic effectiveness of both leaves and roots are scanty in literature, hence the need for this study.

Materials and methods

Samples procurement, preparations and extraction

Leaves and roots of papaya used for the study were obtained in a private residence in Yenagoa metropolis, Bayelsa state, Nigeria. The fresh roots and leaves were washed with water. The plants parts was shade dried before maceration using pestle and mortar. The plants parts were further blended to fine powder. Then after, 40g of the samples

were soaked in 100ml of methanol for 48hours.[11] Furthermore, 20g of each of the powered plant parts (roots and leaves) were mixed together making the mixture 40g, and it was soaked in methanol for 48hours as well.[10,11] The soaked plant parts was filtered using muslin cloth, and the filtrate was re-filtered using Whatman filter paper.[10,11] The solvent were allowed to evaporate in a water bath.

Dilution of the extracts

Extract dilution method previously described by Kigigha et al.,[4] and applied by Izah et al.,[9,10] Izah & Aseibai,[11] Kigigha et al.,[12] was adopted for this study. The extract was considered as 100% concentration which was then diluted into 95%, 90% and 85% using sterile water.

Source of microbes

Staphylococcs aureus, Escherichia coli and *Bacillus subtilis* used were obtained from Medical Microbiology units, Federal Medical Centre, Yenagoa, Bayelsa state, Nigeria. The purity and characteristics of the bacteria isolates was determined following the scheme provided by Cheesbrough.[25] Each isolates under investigation were inoculated into prepared sterile peptone water and incubated for 24hours prior to us.[10,11]

Antimicrobial screening of the extract

The sensitivity screening was carried out using Agar well diffusion method that was previously described by Lino & Deogracious[26] cited in Doherty et al.,[27] with slight modification by Agu &Thomas,[28] Kigigha et al.,[3,4] Epidi et al.,[1,2] Izah et al.,[9,10] Izah & Aseibai.[11] Nutrient agar prepared according to the manufacturers' instruction was dispensed onto sterile Petri dish and it allowed to solidify in triplicate. Furthermore, 300µL peptone water containing 24hours incubated isolates was spread over the surface of the agar. 3 wells of 6mm in diameter were made in each agar plate. Then 300µL of the extracts were dispensed into the agar wells and it was incubated for 24hours. The resultant zones of inhibition were measured.

Statistical analysis

Descriptive statistics (Mean±standard error) was carried out, and One-way analysis of variance was used to establish significance level at P=0.05. Duncan multiple range test statistics was used to discern the difference between means.

Results and discussion

The zone of inhibition of methanolic leaves and roots extract of *Carica papaya* at different concentrations is presented in Table 1. The mean zone of inhibition by *Staphylococcus aureus, Escherichia coli* and *Bacillus subtilis* was 11.33mm, 11.00mm and 9.00mm, respectively for leaves, 10.00mm, 10.33mm and 10.33mm, respectively for root, and 11.67mm, 12.00mm, 10.00mm, respectively for synergy of roots and leaves at 100% concentration. The mean zone of inhibition by *Staphylococcus aureus, Escherichia coli* and *Bacillus subtilis* was 8.33mm, 8.33mm and 7.00mm, respectively for leaves, 7.33mm, 8.33mm and 7.67mm, respectively for root, and 9.00mm, 9.00mm, 7.67mm, respectively for synergy of roots and leaves at 95% concentration of the extracts. The mean zone of inhibition by *Staphylococcus aureus, Escherichia coli* and *Bacillus subtilis* was 4.67mm, 5.00mm and 0.00mm, respectively for leaves, 0.00mm, 7.00mm and 0.00mm, respectively for roots, and 4.67mm, 5.00mm and 0.00mm, respectively for synergy of leaves and root. Basically, there was significant variation at 100% concentration of extracts (P=0.016) and 90% concentration of extracts (P=0.021). Furthermore no significant difference (P=0.122) exist at 95% concentration of the extracts. Typically as the dilution increased the inhibition of the extract decreased. This trend have been reported by Izah et al.,[9,10] Izah & Aseibai,[11] Kigigha et al.[4,12] Typically, studies have shown that papaya have antibacterial potentials.[8,18,22–24] The antibacterial potentials could be due to the phytochemical and bioactive constituents.[1–7,9–11] Saeed et al.,[20] reported that papaya contain several phytochemicals, including polysaccharides, vitamins, minerals, enzymes, proteins, glycosides, saponins, flavonoids, and phytosterols. Doughari et al.,[23] showed that methanolic extract of root of papaya have revealed the presence of saponins, alkaloids, tannins and phenol, while hot water and ethanol extract of same plant roots contain glycosides. Igwe[18] reported that alkaloid confers antibacterial activity. Igwe,[18] Francis & Jose[24] reported that papaya have bioactive constituents with pharmacological properties. The bioactive constituents are influenced by the extraction solvents.[24]

Table 1 Zone of inhibition (mm) of methanolic leaves and roots extract of Carica papaya

Plant parts	Isolates	100%	95%	90%	85%
	Staphylococcus aureus	11.33±0.33^{bc}	8.33±0.67^{ab}	4.67±2.233^{ab}	0.00±0.00
Leaves	*Escherichia coli*	11.00±0.00^{bc}	8.33±0.67^{ab}	5.00±2.52^{ab}	0.00±0.00
	Bacillus subtilis	9.00±0.00^{a}	7.00±0.00^{a}	0.00±0.00^{a}	0.00±0.00
	Staphylococcus aureus	10.00±0.58^{ab}	7.33±0.33^{ab}	0.00±0.00^{a}	0.00±0.00
Roots	*Escherichia coli*	10.33±0.33^{abc}	8.33±0.33^{ab}	7.00±0.00^{b}	0.00±0.00
	Bacillus subtilis	10.33±0.88^{abc}	7.67±0.67^{ab}	0.00±0.00^{a}	0.00±0.00
	Staphylococcus aureus	11.67±0.88^{bc}	9.00±0.58^{b}	4.67±2.33^{ab}	0.00±0.00
Synergy of leaves and roots	*Escherichia coli*	12.00±0.00^{c}	9.00±0.58^{b}	5.00±2.52^{ab}	0.00±0.00
	Bacillus subtilis	10.00±0.58^{ab}	7.67±0.33^{ab}	0.00±0.00^{a}	0.00±0.00

Data were expressed as mean ± standard error; Different letters along the column indicate significant variation (P<0.05) according to Duncan statistics

Citation: Izah SC, Uhunmwangho EJ, Dunga KE. Studies on the synergistic effectiveness of methanolic extract of leaves and roots of *Carica papaya* L. (papaya) against some *bacteria* pathogens. *Int J Complement Alt Med.* 2018;11(6):473–476. DOI: 10.15406/ijcam.2018.11.00429

The synergy had higher effect for *Staphylococcus aureus* and *Escherichia coli.* While *Bacillus subtilis* were in the order; roots> synergy> leaves. The values in this study had some similarity with previous works; Igwe[18] reported zone of inhibition of leaves isopropanol extracts of papaya as 14.33mm, 11.39mm, 12.98mm and 12.37mm for *Staphylococcus aureus*, *Streptococcus faecalis*, *Escherichia coli* and *Proteus mirabilis* respectively at 100% concentration. Dey et al.,[8] reported zone of inhibition of ethanolic root extracts of papaya as 19.83mm, 15.5mm, 9.83mm, 14.5mm and 10.5mm for *Staphylococcus aureus*, *Escherichia coli, Proteus mirabilis*, *Klebsilla pneumoniae* and *Staphylococcus epidermidis* respectively at 100mg/ml concentration. Doughari et al.,[23] also reported that methanolic root extract of papaya is potent against *Staphylococcus aureus, Streptococcus pyogenes, Streptococcus pneumonia, Bacillus cereus, Escherichia coli, Pseudomonas aeruginosa, Proteus mirabilis, Salmonella typhi* and *Shigella flexneri*. The authors further reported viability of solvent in the order; methanol> acetone> hot water.

Typically, the variation in bioactive constituent of the essential oil and phytochemical compound suggests that there is no single mechanism of action at cellular level.[18] The authors further reported that irreversible destruction of bacteria cells, that induce material losses (cytoplasmic), leakage of ions, loss of energy substrate, leading to lysis of bacteria (cytolysis) and death is a possible action. Several factors affect the sensitivity of extracts. Some of them including nature of medium used which may affect the diffuse ability of the agent, environmental condition of such as pH of the medium and incubation temperature,[23] solvent used for extraction[1–4,24] age of the plant,[9–11,24] metabolism, physiology, nutrition and biochemistry of the organisms,[1–4,6,7, 9–12] concentration of the extracts and well as the approach used for the sensitivity testing. The effectiveness of the extracts against *Escherichia coli* (gram negative) and *Bacillus subtilis* and *Staphylococcus aureus* (gram positive organism) suggests that the extracts of the papaya roots and leaves can be used for the development of broad spectrum antibiotics. This finding correlates with previous works by Doughari et al.,[23] Dey et al.,[8] Igwe.[18]

Conclusion

The challenges of microbial resistances and emergence of new microbial infections have intensified the quest for new source of antimicrobials. Within the past few decades, research unto plant for treatment of several diseases condition and control of parasites, intermediate host, vector causing/transmitting diseases have increased. This study investigated the effectiveness of roots and leaves extracts of papaya against some common bacteria pathogens. The study found that at certain concentration of the methanolic extract of the plant there was inhibition. Furthermore, the synergy showed apparent increase in inhibition zones for both gram positive and gram negative bacteria under study compare to individual plant parts.

Acknowledgements

None.

Conflict of interest

Authors declare that there is no conflict of interest.

References

1. Epidi JO, Izah SC, Ohimain EI. Antibacterial and synergistic efficacy of extracts of Alstonia boonei tissues. *British Journal of Applied Science.* 2016;1(1):21–26.
2. Epidi JO, Izah SC, Ohimain EI, et al. Antibacterial and synergistic potency of tissues of *Vitex grandifolia. Biotechnological Research*. 2016;2(2):69–76.
3. Kigigha LT, Biye SE, Izah SC. Phytochemical and antibacterial activities of *Musanga cecropioides* tissues against *Escherichia coli*, *Pseudomonas aeruginosa Staphylococcus aureus, Proteus* and *Bacillus* species. *International Journal of Applied Research and Technology.* 2016;5(1):100–107.
4. Kigigha LT, Izah SC, Ehizibue M. Activities of *Aframomum melegueta* Seed against *Escherichia coli*, *S. aureus* and *Bacillus* species. *Point Journal of Botany and Microbiology Research.* 2015;1(2):23–29.
5. Kigigha LT, Kalunta CG. Antimicrobial efficacy of leaf extracts of Piper nigrum against *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans*. *J Basic Pharm Toxicol*. 2017;1(2):32–36.
6. Kalunta CG. Antimicrobial effect of different seed extracts of Piper nigrum against *Escherichia coli*, *Staphylococcus aureus* and *Candida albican*. *Biotechnol Res*. 2017;3(3):71–76.
7. Izah SC, Uhunmwangho EJ, Eledo BO. Medicinal potentials of *Buchholzia coriacea* (wonderful kola). *Medicinal Plant Research*. 2018;8(5):27–43.
8. Dey A, Stanley S. Preliminary studies on combinational antibacterial effects of root extracts of Withania somnifera and Carica papaya. *World Journal of Pharmaceutical Research*. 2017;6(4):1431–1443.
9. Izah SC, Uhunmwangho EJ, Etim NG. Antibacterial and synergistic potency of methanolic leaf extracts of *Vernonia amygdalina* L. and *Ocimum gratissimum* L. *Journal of Basic Pharmacology and Toxicology*. 2018;2(1):8–12.
10. Izah SC, Zige DV, Alagoa KJ, et al. Antibacterial Efficacy of Aqueous Extract of *Myristica fragrans* (Common Nutmeg). *EC Pharmacology and Toxicology*. 2018;6(4):291–295.
11. Izah SC, Aseibai ER. Antibacterial and Synergistic activities of methanolic leaf extract of Lemon grass (*Cymbopogon citratus*) and rhizome of Ginger (*Zingiber officinale*) against *Escherichia coli*, *Staphylococcus aureus* and *Bacillus species*. *Acta Scientific Microbiology.* 2018;1(6):26–30.
12. Kigigha LT, Selekere RE, Izah SC. Antibacterial and synergistic efficacy of acetone extracts of Garcinia kola (Bitter kola) and Buchholzia coriacea (Wonderful kola). *Journal of Basic Pharmacology and Toxicology*. 2018;2(1):13–17.
13. Izah SC, Aseiba ER, Orutugu LA. Microbial quality of polythene packaged sliced fruits sold in major markets of Yenagoa Metropolis, Nigeria. *Point Journal of Botany and Microbiology Research*. 2015;1(3):30–36.
14. Vij T, Prashar Y. A review on medicinal properties of *Carica papaya* Linn. *Asian Pacific Journal of Tropical Disease*. 2015;5(1):1–6.
15. Adedeji TO, Oluwalana IB. Physico-chemical, sensory and microbial analysis of wine produced from watermelon (citrulluslanatus and pawpaw (*Carica papaya*) blend. *Food Science and Quality Management*. 2013;19:41–50.
16. Chukwuka KS, Okonko IO, Adekunle AA. Microbial Ecology of Organisms Causing Pawpaw (*Carica papaya L.*) Fruit Decay in Oyo State, Nigeria. *American-Eurasian Journal of Toxicological Sciences*. 2010;2(1):43–50.

17. Panzarini E, Dwikat M, Mariano S, et al. Administration Dependent Antioxidant Effect of *Carica papaya* Seeds Water Extract. *Evidence-Based Complementary and Alternative Medicine.* 2014;2014:281508.

18. Igwe OU. Chemical constituents of the leaf essential oil of *Carica papaya* from South East Nigeria and its antimicrobial activity. *International Journal of Research In Pharmacy And Chemistry*. 2015;5(1):77–83.

19. Jaiswal P, Kumar P, Singh VK, et al. Carica papaya Linn: a potential source for various health problems. *Journal of Pharmacy Research*. 2010;3(5):998–1003.

20. Saeed F, Arshad MU, Pasha I, et al. Nutritional and Phyto-Therapeutic Potential of Papaya (*Carica Papaya Linn*.): An Overview. *International Journal of Food Properties*. 2014;17(7):1637–1653.

21. Krishna KL, Paridhavi M, Patel JA. Review on nutritional, medicinal and pharmacological properties of Papaya (*Carica papaya* Linn.). *Natural Product Radiance*. 2008;7(4):364–373.

22. Rubaka C, Ndakidemi P, Malebo HM, et al. Individual and Combined Antibacterial Activity of Crude Extracts from Medicinal Plants *Carissa spinarum* Linn and *Carica papaya* Linn. *European Journal of Medicinal Plants*. 2014;4(12):1513–1523.

23. Doughari JH, Elmahmood AM, Manzara S. Studies on the antibacterial activity of root extracts of *Carica papaya L*. *African Journal of Microbiology Research.* 2007:37–41.

24. Francis EF and Jose V. The Antibacterial Effect of *Carica papaya* L. Extracts and Their Synergistic Effect with Antibiotic and Non-antibiotic Drugs. *British Microbiology Research Journal*. 2016;16(4):1–11.

25. Cheesbrough M. *District Laboratory Practice in Tropical Countries.* Low price Edition part 2. England: Cambridge press; 2004.

26. Lino A, Deogracious O. The invitro antibacterial activity of *Annonna senegalensis, Sacuridecae longipendiculata* and *Steganotaema araliacea.* Uganda medicinal plants. *Journal of African Health Science.* 2006;6(1):31–35.

27. Doherty VF, Olaniran OO, Kanife UC. Antimicrobial activities of Aframomum Melegueta (Alligator pepper). *International Journal of Biology*. 2010;2(2):126–131.

28. Agu GC, Thomas BT. Antibacterial Activities of Ethanol and Aqueous Extracts of Five Nigerian Medicinal Plants on Some Wound Pathogens. *Nature and Science.* 2012;10(2):78–84.

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