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Antimicrobial Effect of the Leaf Extract of *Psidiumguajava* L. *and Carica Papaya* L.

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Abstract: **Medicinal plants have been used from centuries to treat infectious diseases as an alternative form of health care. Even though pharmacological industries have produced a number of new antibiotics in the last decades, resistance to these antibiotics by microorganisms have increased. The present study evaluated the antimicrobial effect of *Psidiumguajava* L. and *Carica papaya* L. as used in the traditional system of medicine. Their effects were determined using standard method (agar well diffusion). The results obtained showed that both plants had a broad spectrum effect on the test isolates with zones of inhibition ranging from 16.0 to 14.0 mm for *P. guajava* and *C. papaya* at 200 mg/ml respectively for S. *aureus,* 18.4 to 13.0 mm for *E. coli,* 15.0 to 13.5 mm for *S. typhi,* 16.5 to 14.0 mm for *Pseudomonas spp* and 13.0 to 11.8 mm for *C. albican* respectively. *Psidiumguajava* showed more effect compared to *Carica papaya* with MIC at 50 mg/ml and 100 mg/ml respectively. The paper therefore reports the broad spectrum antimicrobial activities of *Psidiumguajava* L. and *Carica papaya* L. leaf extracts on different clinical isolates.**

Keywords: Psidiumguajava L., Carica papaya L., antimicrobial, minimum inhibiting concentration, ethanol extract, aqueous extract.

1. INTRODUCTION

Practitioners of traditional medicine believe that the constituents of plants are unique as they contain both active ingredients and "non-active" components that playa role in enhancing the well-being of their patients. Are kindled interest in the pharmaceutical importance of plants have led to the discovery and adoption of plant extracts which were commonly used in traditional medicine, as alternative source of remedy [1]. It is, however, observed that these practices vary from one country to another. Numerous plants and herbs are used all over Nigeria by traditional medicine practitioners. The use of herbs is the most ancient approach to healing known. The herbal medicines may be in form of powders, liquids, or mixtures, which may be raw, or boiled, ointments, liniments, and incisions [2]. Roots, barks, and leaves of various plants are employed in ethno-medicine. Plant extracts are given singly or as concoctions for various ailments. More than 70% of the people living in Nigeria depend on these various form of concoctions and herbal decoctions for the treatment of some diseases [3].

Psidiumguajava L. (Guava) belongs to the family of Mytaceae and is an evergreen shrub native to tropical America that has naturalized in south East Asia. The Guava leaf extract has been reported to possess a wide spectrum of activities against a variety of human ailment[3, 4, and 5]. Over twenty compounds have been reported to be present in the leaf [6, 7 and 8]. Aqueous leaf extract contains tannin, while ethanolic extract is enriched with anthocyans, alkaloids flavonoids, tannins and steroids/ terpenoids[9].

Carica papaya belongs to the family of Caricaceae, and several species of Caricaceae have been used as remedyagainst a variety of diseases[10]. Papaya offers not only the luscious taste but is a rich source of antioxidantnutrients such as carotenes, vitamin C and flavonoids; the B vitamins, folate and pantothenic acid; and the minerals,potassium and

magnesium; and fibres[11]. Together, these nutrients promote the health of the cardiovascular systemand also provide protection against colon cancer. The fruit is valued for its proteolytic enzymes including papain, which is used like bromelain, a similar enzyme found in pineapple, to treat sports injuries, other causes of trauma, and allergies [12]. Biochemically, its leaves and fruit are complex, producing several proteins and alkaloids with important pharmaceutical and industrial applications[28]. Carapine, an alkaloid present in papaya, can be used as aheart depressant, amoebicide and diuretic. The fruit and juice are consumed for gastrointestinal ailments; a fresh leafpoultice is used to treat sores. The fresh root with sugarcane alcohol can be taken orally or as a massage to soothe rheumatism. A flower decoction is taken orally for coughs, bronchitis, and asthma and chest colds. In some countries, the seeds are used as an abortifacient and vermifuge. The present study was undertaken to evaluate the antimicrobial activities of aqueous and organic extract of *Psidiumquajava* L *and Carica papaya* leaves on clinical isolates.

2. METHODS

A. Collection of Plant Material:

The plants used in this study was collected in the botanical garden of the institution (Abia State Polytechnic, Aba) in a sterile polythene bag, rinsed, dried in an oven at 40^{o}C for five hours. The dried leaves were pulverized using sterile laboratory mortar and pestle to obtain a powder form. These were stored in air tight glass containers from sunlight until required for analysis.

B. Test Isolate:

The test organism used, were all human pathogenic organism from clinical origin. They were obtained from stock cultures in the Department of Microbiology,Abia State Polytechnic, Aba.The organisms were collected, identified and subculture in nutrient agar slants and incubated at 37^{o}C for 24 hours. They were then kept as stock in the refrigerator at 4^{o}C until when required for the analysis.

C. Extraction of Plant Material:

The ethanol and aqueous extract of both plant samples was carried out according to [29]. 10g of the powdered leaves was weight out and dissolved in 100ml of both solvent in a sterile beaker and allowed to stand for 24hours. The mixture was filtered using What Man No.1 filter paper and the extract was evaporated to dryness at 45^{o}C. The residues obtained were reconstituted in 100% ethanol and water at stock concentration, stored in the refrigerator at 42^{o}C until used.

D. Sensitivity Test:

The disc diffusion technique was used [13]. The circular discs were put in the different reconstituted extract. The discs were allowed to absorb the extract which becomes condition. The test organism were cultured by evenly spreading into sterile agar media on separate Petri dishes then the disc haven allowed to air dried were placed on the inoculated plates at appropriate distance from one another. The plates were allowed to stand on the table for some minute and incubated at 37^{o}C for 24hrs for bacterial and at room temperature for 2-5 days for fungi after which clear zones (zones of inhibition) were measured using a transparent rule. Three measurements were taken for each sample and a mean value was recorded.

E. Determination of Minimum Inhibitory Concentration:

The minimum inhibitory concentration was determined for both plants using agar dilution technique. Nutrient agar plates containing varying concentration of 100mg/mL – 250mg/ mL of both plants extract (aqueous and ethanol) were prepared and inoculated with the test isolate in separate Petri dish. The plates were incubated; the lowest concentration of the various extract that completely prevents the growth of the organism was recorded as the minimum inhibitory concentration.

3. RESULT

Table 1 shows the comparative zones of inhibitions for the ethanol extract of *P.quajava*and *C. papaya* leaves extract. The result showed that *P. quajava*had more effect on the test isolates compared to *C. papaya* with *E. coli* being the most susceptible isolates (18.4mm) for *P. quajava* and *C. albican*as the least (13.0 and 11.8mmrespectively) for both plants. *S. aureus*was the most susceptible isolate for *C. papaya* (14.0mm) at 200mg/ml. however, the test control drug (Gentamycine) showed more effect than the plant extract with *Pseudomonas* spp. being the most susceptible.

Table 2 showed the comparative effect of the aqueous extract of both plants on the clinical isolates. *E. coli* was the most susceptible isolate (14.6mm) for *P. quajava* and *S. aureus*(12.3mm) for *C. papaya*while *C. albican*was the least susceptible isolate (11.8 and 9.5mm respectively) for both plant at 200mg/ml.

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TABLE 1: MEAN DIAMETER OF ZONES OF INHIBITIONS FOR THE EFFECT OF THE ETHANOL EXTRACTS *OF P. GUAJAVA* AND *C. PAPAYA* LEAF EXTRACTS.

Organism	Concentration(mg/ml)	*P. quajava*	*C. papaya*	Gentamicine	Nystatin
S.aureus	50	8.0	NIL		
	100	10.0	8.1		
	150	14.1	11.2	24.5	NA
	200	16.0	14.0		
E. coli	50	11.2	NIL		
	100	14.5	7.0		
	150	15.0	11.0	22.0	NA
	200	18.4	13.0		
S. typhi	50	7.5	NIL		
	100	9.6	6.2		
	150	13.5	10.0	20.7	NA
	200	15.0	13.5		
*Pseudomonas*spp	50	8.6	NIL		
	100	11.9	6.3		
	150	14.4	10.2	25.2	NA
	200	16.5	14.0		
C. albican	50	6.9	NIL		
	100	9.3	NIL		
	150	11.2	7.9	NA	23.0
	200	13.0	11.8		

TABLE 2: MEAN DIAMETER OF ZONES OF INHIBITIONS FOR THE EFFECT OF THE AQUEOUS EXTRACT OF *P. GUAJAVA* AND *C. PAPAYA* LEAF EXTRACTS.

Organism	Concentration(mg/ml)	*P. quajava*	*C. papaya*	Gentamicine	Nystatin
S.aureus	50	NIL	NIL		
	100	8.1	NIL		
	150	11.2	9.0	24.4	NA
	200	14.0	12.3		
E. coli	50	NIL	NIL		
	100	8.1	NIL		
	150	11.2	8.6	22.0	NA
	200	14.6	11.2		
S. typhi	50	NIL	NIL		
	100	NIL	NIL	20.7	NA
	150	9.2	7.2		
	200	12.8	10.2		
*Pseudomonas*spp	50	NIL	NIL		
	100	NIL	NIL		
	150	9.4	7.1	25.2	NA
	200	13.3	11.3		
C. albican	50	NIL	NIL		
	100	NIL	NIL	NA	23.0
	150	7.9	6.8		
	200	11.8	9.5		

NB: NA= Not applicable

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TABLE 5: MINIMUM INHIBITORY CONCENTRATION OF THE ETHANOL EXTRACT OF *P. GUAJAVA* AND *C. PAPAYA* LEAF EXTRACTS.

Organism	Plant / MIC (mg/mL)	
	P. quajava	*C. papaya*
S. aureus	50	100
E. coli	50	100
S. typhi	50	150
P. aeroginosa	50	100
C. albican	50	150

TABLE 6: MINIMUM INHIBITORY CONCENTRATION OF THE AQUEOUS EXTRACT OF *P. QUAJAVA* AND *C.PAPAYA* LEAF EXTRACTS.

Organism	Plant/ MIC (mg/mL)	
	P. quajava	*C. papaya*
S. aureus	100	150
E. coli	100	150
S. typhi	150	150
P. aeroginosa	150	150
C. albican	150	150

4. DISCUSSION

The use of medicinal plants to treat and manage various forms of diseases and dysfunctions is becoming increasingly popular and has received wide acceptance [14]. Nigeria, an important nation of biodiversity, is enriched with herbal resources. Reports on the effects of these medicinal plants on animal and human health are diverse. Although these effects are largely attributed to the active components of these plant materials [15, 16], yet informationon the chemical composition of many of these plant materials are still scarce [17]. The chemical evaluation ofmedicinal plants and their isolates have transformed traditional medicine from an almost invisible trade into a modern industrial enterprise, capable of making significant contribution to both health care delivery and economic growth of most developing countries [4, 18]. Moreover, the World Health Organization (W.H.O.) had recognized traditional herbal medicine as a building block of primary health care[19].

In the present study, the result for the effect of ethanol and aqueous extract of *Psidiumguajava* and *Carica papaya* showed that the plants were effective against the test isolate at different concentrations showing varying zones of inhibitions. This finding can be attested to other works by different authors like[30]who reported the effect of *P. guajava* on multidrug resistance Staphylococcus, Amit and Shweta (2012), reported the effect of the plant (*P. guajava*) *on Pseudomonas* spp, *Staphylococcus* and *E. coli.* Okunola et al (2012) reported the effect of *C. papaya* on similar isolates including *klebsiella pneumonia*, *Enterococcus*, and *Proteusspp*. However, *P. guajava* showed more effect than *C. papaya*. The present result indicated that the strong anti microbial activity exhibited by the leaf extracts of *P. guajava* was possibly due to protein degrading activity of the extract. Tannin, known to be present in the aqueous and ethanol extract, reportedly have protein-binding activities and can interfere with many substances [18].

The results of this study demonstrated that the organic extracts were more effective than aqueous extracts. This may be due to the better solubility of the active components in the organic solvent. The ethanol extracts demonstrated a higher activity than the aqueous extracts in both leaf samples. The better efficacy of the ethanol extract as against the aqueous extract maybe because different solvents have different polarities, hence different degrees of solubility for the various phyto-constituents[30]. Based on the limited effect of activity of the other extracts compared with the ethanol extracts, it suggests that the active component ismore soluble in ethanol than in the other solvents.

Comparing the sensitivity of the microbial strains to both the plant extracts and to synthetic antibiotics, the result showed that the plant extracts compete favourably with the drugs and can be used as an alternative to the antibiotics as the zones on inhibition shown were very comparable and the extracts have lesser side effects which are often associated with the use of antibiotics [20, 21]. Also the issue of resistance to these extracts cannot arise as is found with antibiotics [22]. The results

obtained support the fact that further work needs to be done to determine and identify, purify and quantify the antibacterial compound within these plants and also to determine their full spectrum of efficacy.

The MIC result showed that increasing concentration has an increasing efficiency in inhibiting the organisms used. Since the MIC values indicated the definite nature of the antimicrobial activities of this plant, the inhibition zones values, only, indicated extent of effectiveness of the extract with increasing concentration.

Although the mechanism of action of this extract is not understood. It has been proposed that its action against the bacteria and fungi may be due to the inhibition of cell wall formation in the cell resulting in a leakage of cytoplasmic constituents by the bioactive components of the extract [23, 24]. While phytochemical compounds such as tannin coagulate the wall proteins, saponins facilitated the entry of toxic material or leakage of vital constituents from the cell [25]. Flavonoids inhibit the activity of enzymes by forming complexes with bacterial cell walls, extracellular and soluble proteins, more lipophilic flavonoids disrupt cell wall integrity [26] or microbial membranes [27] at low concentrations.

In conclusion, plant-based antimicrobials have enormous therapeutic and preferential potential. They can serve thedesired purpose with lesser side effects that are often associated with synthetic antimicrobials. The antimicrobial activity of P. *guajava and C. papaya* leaves was demonstrated in this study. Demonstration of antimicrobial activity against the test isolates is an indication that there is possibility of sourcing alternative antibiotic substances in these plants for the development of newer antibacterial agents.

The results obtained in this study contribute to the scientific validation for the use of these medicinal plants in traditional medicine and serve as a guide for selection of plants with antimicrobial activity for further phyto-chemical work on isolation and identification of the active compounds.

5. CONCLUSION

The result of the present study indicates that both plant extracts showed antibacterial and antifungal activities against test organisms with *Psidiumguajava* being more effective than *Carica papaya*. The paper therefore suggests further research into investigating other biological and phytochemical constituents of both plants for scientific uses in relevant fields of study.

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