

International Journal of Advanced Research in Biological Sciences

www.ijarbs.com



Research Article

Phytochemical analysis and *in-vitro* Antimicrobial activity of Aqueous and Solvent extracts of *Carica papaya* against clinical Pathogens

R. Sumathi* and M. Gowthami

PG Dept. of Microbiology, Kanchi Shri Krishna College of Arts and Science, Kilambi, Tamil Nadu, India - 631 551.

Corresponding author e-mail: renusumathi@rediffmail.com

Abstract

The plant materials such as leaves, stem and root of disease free *Carica papaya* were collected from Kaveripakkam, Vellore district, Tamilnadu. The dried powdered plant material is subjected to solvent extraction using the solvents cold water, hot water and ethanol. Antimicrobial assay of plant extract against clinical isolates by AWD assay. Only the leaf extracts showed inhibitory effect against *Candida albicans*, whereas stem and root extracts were ineffective. Among the leaf, stem and root extracts, the leaf extract is found to exhibit more antimicrobial activity than the stem and root.

Keywords: *Carica papaya*; solvents; Antimicrobial assay; *Candida albicans*.

Introduction

A wide range of medicinal plant parts is used for extract as raw drugs and they possess varied medicinal properties. The different parts used include root, stem, flower, fruit, twigs exudates and modified plant organs. While some of these raw drugs are collected in smaller quantities by the local communities and folk healers for local used, many other raw drugs are collected in larger quantities and traded in the market as the raw material for many herbal industries.

Medicinal plants are of great importance to the health of individuals. The value of these plants lies in some chemical substances that produce a definite physiological action on the human body (Hill, 1952). The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids, and phenolic compounds (Mbojikwe,

2004; Hill, 1952). Many of these indigenous medicinal plants are used as spices and food plants. *Carica papaya* belongs to the family *Caricaceae* that has been used to treat various ailments.

Considering the vast potentiality of plants as sources for antimicrobial drugs with reference to antibacterial and antifungal agents, a systematic investigation was undertaken to screen the local flora for antibacterial and antifungal activity of *Carica papaya*.

Materials and Methods

Collection of plant materials

The plant *Carioca papaya* was used for this project work and it was identified as papaya leaf, stem, and

root. These were then collected in a sterile polytene bag, rinsed, dried and made into a powdery form before uses.

Preparation of Plant Extract (Omojosola and Awe, 2004)

The *Carica papaya* (dry, green leaves, stem, root) was separately extracted with cold aqueous, hot water and cold ethanol. These were carried out by suspending 5 grams of the finely ground leaves, stem, root in 50ml of distilled water or 70% ethanol. The hot water extraction was done at 80°C in a water bath for ½ hours. The ethanolic extraction was done at 28±1°C for 72 hours. The extracts were then decanted and filtered through a Whatman filter paper. The filtered extract was then sterilized using a membrane filter and evaporated after used. A known quantity of leaf powder (5gm) was taken in each of the 250ml beaker and added with 100 ml of cold water, hot water and ethanol. The preparation was kept at room temperature for 48 hours and rapidly stirred using separate glass rod every 4 hours. After 48 hours, the each leaf, stem, root extract was filtered through Whatmann No. 1 filter paper to exclude the leaf, stem and root powder. The leaf, stem, and root extract was taken in separate beaker and kept in a water bath at 40-50°C until the solvent gets evaporated. A greasy final material was obtained from the leaf, stem and root extract. Each extract was transferred to sterile screw capped bottles, labeled and stored under refrigerated condition till use.

Experimental microorganism

Gram negative strains is *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi* and Gram positive strain is *Staphylococcus aureus* were used for the study. Fungal strain is *Candida albicans* also tested for antibacterial assay.

Antimicrobial assay of extracts (Jasso de Rodríguez et al., 2005)

The agar well method of the agar diffusion technique was used to determine the antibacterial activity of the plant extracts. Preparation of filter paper disc impregnated with *Carioca papaya* extracts. Filter paper disc of 6 mm diameter were

cut using a punching machine in Whatmann No. 1 filter paper. The discs were sterilized by dry heat sterilization. 20µl of each leaf, stem, root extracts were added to the separate discs. The dried extract impregnated discs were used for testing antimicrobial activity against clinical isolates by disc diffusion method.

Antibiogram of carica papaya extracts

The sterilized Muller Hinton Agar medium was poured into a sterile Petri plate. After solidification, a lawn culture of the organism was made and it is allowed to dry for 5 minutes. The filter paper discs impregnated with *Carioca papaya* ethanolic leaf, stem, root extracts were placed on to the surface of the medium 3mm apart and gently pressed in order to adhere the discs. After incubation the zone of inhibition around the disc were measured.

Results

Phytochemical screening

The qualitative analysis of the phytochemical constituents of the crude extracts of the *Carica papaya*. The analysis gave positive results for alkaloids, saponins, tannins and terpenoids for crude aqueous extracts of leaves, stem and root. On the other hand, the ethanolic extracts show positive result for alkaloids, flavonoids, tannins, terpenoids and reducing sugars. This observation indicates that the difference in activity could be due to the differences in the phytochemical composition of the extracts (Arunkumar and Muthuselvan, 2009; Yebpella et al., 2011).

Antimicrobial assay

The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* leaf against *Staphylococcus aureus* was shown in Table-1. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* stem against *Staphylococcus aureus* was shown in Table-2. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* root against *Staphylococcus aureus* was shown in

Table.1 Antibacterial activity of *C. papaya* leaf extract against *S.aureus*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	16
Hot aqueous	9	16
Ethanol	12	16

Table.2 Antibacterial activity of *C. papaya* stem extract against *S.aureus*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	8	16
Hot aqueous	11	16
Ethanol	12	16

Table .3 Antibacterial activity of *C. papaya* root extract against *S.aureus*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	16
Hot aqueous	5	16
Ethanol	12	16

Table . 4 Antibacterial activity of *C. papaya* leaf extract against *E.coli*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	12	14
Hot aqueous	11	14
Ethanol	15	14

Table.3 The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* leaf against *Escherichia coli* was shown in Table. 5 The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya*

Table .5 Antibacterial activity of *C. papaya* stem extract against *E.coli*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	8	16
Hot aqueous	11	16
Ethanol	12	16

Table.6 Antibacterial activity of *C. papaya* root extract against *E.coli*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	16
Hot aqueous	-	16
Ethanol	10	16

Table .7 Antibacterial activity of *C. papaya* leaf extract against *S.typhi*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	7	16
Hot aqueous	10	16
Ethanol	14	16

Table.8 Antibacterial activity of *C. papaya* stem extract against *S.typhi*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	16
Hot aqueous	9	16
Ethanol	10	16

stem against *Escherichia coli* was shown in Table.5.

The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* root against *Escherichia coli* was shown in Table. 6. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* leaf against *Salmonella typhi* was shown in Table 7.

Table .9 Antibacterial activity of *C. papaya* root extract against *S.typhi*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	13	16
Hot aqueous	6	16
Ethanol	10	16

Table .10 Antibacterial activity of *C. papaya* leaf extract against *P.aeruginosa*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	13
Hot aqueous	12	13
Ethanol	16	13

Table. 11 Antibacterial activity of *C. papaya* stem extract against *P.aeruginosa*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	13
Hot aqueous	12	13
Ethanol	16	13

Table. 12 Antibacterial activity of *C. papaya* root extract against *P.aeruginosa*

Extract	Zone of inhibition in mm	Standard antibiotics (Tetracycline)
Aqueous	-	13
Hot aqueous	12	13
Ethanol	16	13

Table . 13 Antifungal assay of *Carica papaya* against *Candida albicans*

Extract	Zone of inhibition in mm			Standard antifungal agent (fluconazole)
	Leaf	Stem	Root	18
Aqueous	10	-	-	
Hot aqueous	8	-	-	
Ethanol	12	-	-	

The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* stem against *Salmonella typhi* was shown in Table 8. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* root against *Salmonella typhi* was shown in Table.9. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* leaf against *Pseudomonas aeruginosa* was shown in Table.10.

The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* stem against *Salmonella typhi* was shown in Table 8. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* root against *Salmonella typhi* was shown in Table.9. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* leaf against *Pseudomonas aeruginosa* was shown in Table.10. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* stem against *Pseudomonas aeruginosa* was shown in Table 11. The antibacterial activity of cold aqueous, hot aqueous and ethanolic extract of *Carica papaya* root against *Pseudomonas aeruginosa* was shown in Table 12.

Antifungal assay

The antifungal assay of *Carica papaya* leaf, stem and root extracts against *Candida albicans* were shown in Table 13.

Discussion

Carica papaya plants produce natural compounds (annonaceous acetogenins) in leaf bark and twig tissues that possess both highly anti-tumour and pesticidal properties. It was suggested that a potentially lucrative industry based simply on production of plant biomass could develop for production of anti-cancer drugs, pending Food and Drug Agency approval, and natural (botanical) pesticides (Ayoola and Adeyeye, 2010). The high level of natural self-defence compounds in the tree makes it highly resistant to insect and disease infestation (Joel L. Mattsson, 2007).

Papaya contains many biologically active compounds. Two important compounds are chymopapain and papain, which are supposed to aid digestion. It has been used for treating digestive problems and intestinal worms. The softening qualities of papain have been taken advantage of, in the treatment warts, corns, Sinuses, and Chronic forms of scaly eczema, cutaneous tubercles, and other hardness of the skin, produce by irritation. Papain also is used to treat arthritis (Abhishek Mathu et al., 2011). The aqueous and ethanolic extracts investigated revealed the presence of alkaloids, flavonoids, tannins, cardiac glycosides, and reducing sugars, however, all the phytoconstituents were more in the alcoholic extraction than the aqueous as indicated by the intensity of the different confirmatory colours, In the present study, the extracts of *Carica papaya* were prepared using solvents like cold aqueous, hot aqueous and ethanol by solvent extraction method. Antibacterial and antifungal activities were tested against both Gram positive and Gram negative bacteria and medically important fungi *Candida albicans* by agar well diffusion method. In the antibacterial activity assay, the zone of inhibition of both Gram positive and gram negative was more in ethanolic extract than cold aqueous and hot aqueous extracts. In case of antifungal activity, the zone of inhibition was observed only in leaf extracts and no activity was seen in aqueous extracts. Thus, *Carica papaya* could become promising natural antimicrobial agents with potential applications in pharmaceutical industry for controlling the pathogenic bacteria. However, if plant extracts are

to be used for medicinal purposes, issues of safety and toxicity will always need to be considered.

References

- Abhishek Mathur, Stish K, Verrma, Santosh K, Singh and GBKS Prasad VK. Investigation of the antimicrobial, antioxidant and anti-inflammatory activity of compound isolated from *Murraya koenigii*, IJABT. 2011; 2(1).
- Arunkumar S and Muthuselvan M. Analysis of phytochemical constituents and antimicrobial activities of *Aloe vera* L. against clinical pathogens. World J Agric Sci. 2009;5:572-576
- Ayoola, P.B., and A. Adeyeye. phytochemical and nutrient evaluation of *Carica papaya* (pawpaw) leaves. IJRRAS 5 (3) December 2010
- Hill, R., 1952. The Elastic Behaviour of a Crystalline Aggregate. Proc. Phys. Soc. A 65 349. doi:10.1088/0370-1298/65/5/307
- Jasso de Rodríguez D, Hernández-Castillo D, Rodríguez-García R, Angulo-Sánchez JL. Antifungal activity in vitro of *Aloe vera* pulp and liquid fraction against plant pathogenic fungi. Ind Crops Prod. 2005;21:81-87. doi: 10.1016/j.indcrop.2004.01.002.
- Joel L. Mattsson. Mixtures in the real world: The importance of plant self-defense toxicants, mycotoxins, and the human diet. Toxicology and Applied Pharmacology 223 (2007) 125-132
- Mbojikwe . L.O., 2004. The Identification and Structural Characterisation of the Active Acaricidal Principle in the Aqueous Stem Bark Extract of *Adenium obesum* . NVRI SEMINAR SERIES pp.7-9.
- Omojola, P. F. and Awe, S. 2004. The Antibacterial activity of the leaf extract of *Anacardium occidentale* and *Gossypium hirsutum* against some selected microorganisms. Bioscience Research Communication 16 (1).
- Yebpella GG, Hammuel C, Adeyemi HMM, Magomya AM, Agbaji AS and Shallangwa GA. Phytochemical screening and a comparative study of antibacterial activity of *Aloe vera* green rind, gel and leaf pulp extract s. Int Res J Microbiol. 2011;2:382-386.