

British Microbiology Research Journal 16(4): 1-11, 2016, Article no.BMRJ.28042 ISSN: 2231-0886, NLM ID: 101608140



SCIENCEDOMAIN international www.sciencedomain.org

# The Antibacterial Effect of *Carica papaya* L. Extracts and Their Synergistic Effect with Antibiotic and Non-antibiotic Drugs

# E. Femi Francis<sup>1</sup> and Vimala Jose<sup>1\*</sup>

<sup>1</sup>Department of Botany, St. Thomas' College (Autonomous), Thrissur -680 601, Kerala, India.

### Authors' contributions

This work was carried out in collaboration between both authors. Author EFF designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript and managed literature searches. Authors EFF and VJ managed the analyses of the study and literature searches. Both authors read and approved the final manuscript.

#### Article Information

DOI: 10.9734/BMRJ/2016/28042 <u>Editor(s):</u> (1) Raúl Rodríguez-Herrera, Autonomous University of Coahuila, México. <u>Reviewers:</u> (1) Muhammad Yusha'u, Bayero University Kano, Nigeria. (2) Georgios Androutsopoulos, University of Patras, Rio, Greece. (3) Lung-Chien Chen, National Taipei University of Technology, Taiwan. Complete Peer review History: <u>http://www.sciencedomain.org/review-history/15811</u>

**Original Research Article** 

Received 30<sup>th</sup> June 2016 Accepted 5<sup>th</sup> August 2016 Published 16<sup>th</sup> August 2016

# ABSTRACT

**Aims:** Antibacterial activity of *Carica papaya* leaf and seed extracts, their synergism with antibiotic and non-antibiotic drugs and GC-MS analysis of extracts.

**Study Design:** Antibacterial activity was evaluated by Disc and Well diffusion method. Synergism with antibiotic drug, Gentamicin, and non-antibiotic drug, Vitamin C, were done by disc diffusion method. GC-MS analysis carried out in GC-MS equipment (Thermo Scientific Co.).

Place and Duration of Study: Study was conducted in Department of Botany and Department of Chemistry, St. Thomas' College, Thrissur between December 2015 to April 2016.

**Methodology:** We include 3 gram negative (*E. coli, Klebsiella pneumoniae* and *Pseudomonas aeruginosa*) and a gram positive (*Staphylococcus aureus*) bacteria in this study. Antibacterial activity of *Carica papaya* extracts (water, petroleum benzene, chloroform and ethanol extracts) against these bacteria's were studied. Their synergisms with antibiotic as well as non antibiotic drugs were also evaluated. GC-MS analysis of all the extracts were also done.

Results: In the antibacterial activity assessment, all the four extracts of tender leaves were effective

<sup>\*</sup>Corresponding author: E-mail: vimalajoseparaemackel@gmail.com;

Francis and Jose; BMRJ, 16(4): 1-11, 2016; Article no.BMRJ.28042

against *E. coli* than other plant materials. Seed extracts were more effective against *P. aeruginosa* and *S. aureus*. In synergistic analysis, water and ethanol extracts of all the plant materials have an enhanced effect with gentamicin against *E. coli* and *P. aeruginosa*. Yellow leaves extracts along with gentamicin exhibited an inhibition zone which is greater than that of gentamicin alone. Vitamin C gave enhanced activity against all the tested bacteria when combined with papaya extracts. GC-MS analysis proved that more number of bioactive components were present in petroleum benzene extract of tender leaves than all other extracts.

**Conclusion:** The results shows that *Carica papaya* extracts have antibacterial activity and when they were combined with antibiotic and non antibiotic drugs. In GC-MS analysis, tender leaves exhibited more bioactive components.

Keywords: Carica papaya; antibacterial activity; synergism; antibiotic drug; non-antibiotic drug; GC-MS analysis.

#### **1. INTRODUCTION**

Medicines have an important role in our day to day life. Since time immemorial plants have been used for treatment of various ailments. Even today several important drugs used in modern system of medicines are obtained from plants. Use of medicinal plants has figured in several ancient manuscripts like *Rig-Veda*. In *Ayurveda*, definite properties of drugs obtained from plants are used to improve the overall health and wellbeing. Medicinal importance of a plant is due to presence of some alkaloids, glycosides, resins, volatile oils, gums, tannins, etc. These active principles usually remain concentrated in storage organs of plant, *viz*, roots, seeds, bark, leaves etc.

Papaya is the only species in the genus Carica of the plant family Caricaceae [1]. The papaya is a large, tree-like plant, with a single stem growing from 5 to 10 m tall, with spirally arranged leaves confined to the top of the trunk. The flowers appear on the axils of the leaves, maturing into large spherical or pear shaped fruits - 15-45 cm long and 10-30 cm in diameter. The fruit is a type of berry [2]. They clumped near its top end of the trunk. Inside, the fruit features numerous black pepper-corn like seeds, encased in a mucin coat, at its hollow central cavity. The flesh is orange in color with either yellow or pink hues, soft in consistency and has deliciously sweet, musky taste with rich flavor [3]. Now it is considered as valuable nutraceutical fruit plant. It can be chosen as a source of "Papain" for the development of various industrial and pharmaceutical products for various diseases [4]. Rich loamy soil is most suitable for the growth of papaya. It is usually propagated by seeds obtained from well mature fruits.

So the present study was designed keeping in view the resistance of bacteria to most of the drugs commonly used today. It was aimed to analyze the antibacterial activity and its synergistic effect with antibiotic and nonantibiotic drugs against the most common disease causing bacteria's such as *Escherichia coli, Klebsiella pneumoniae., Pseudomonas aeruginosa* and *Staphylococcus aureus.* 

### 2. METHODOLOGY

#### 2.1 Collection of Plant Materials

Tender leaves, mature green and yellow leaves and mature white seeds of *Carica papaya* L. were collected from different locations in Thrissur. Leaves were dried under shade and seeds under sunlight. Dried leaves and seeds were grinded into fine powder.

#### 2.2 Extraction of Plant Materials

#### 2.2.1 Aqueous extraction

50 gm of finely powdered leaves and seeds were separately weighed out in a weighing balance and tied up in muslin cloth. It was concentrated by boiling in a beaker of distilled water in a hot plate and then concentrated to dryness with a vacuum evaporator. Dilutions of the concentrated samples were prepared in DMSO and it was used as aqueous extract in the antibacterial analysis. The concentration of plant extracts was fixed as 500  $\mu$ g/ml for the purpose of this study.

#### 2.2.2 Organic solvent extraction

25 gm of finely powdered plant parts were taken in Soxhlet apparatus along with 300 ml of organic solvents such as petroleum benzene, chloroform and ethanol based on their polarity. The plant extracts obtained after extraction were concentrated with a vacuum evaporator. The solution was stored in well closed containers under refrigeration conditions and dilutions of the plant extract in DMSO were used for antimicrobial studies. The concentration of plant extracts used for antimicrobial study was 500  $\mu$ g/ml.

#### 2.3 Antibacterial Activity

Bacterial samples were isolated from a local clinic (Poly Clinic Laboratories, Thrissur, Kerala, India). *Staphylococcus aureus* ATCC 6538, *Klebsiella pneumoniae* ATCC 700603, *Pseudomonas aeruginosa* ATCC 15442 and *Escherichia coli* ATCC 2392 were used in the study.

#### 2.3.1 Paper disc diffusion assay

Suspensions of testing microorganisms were spread on medium. The filter paper discs of 5mm diameter was placed on the agar plates which was inoculated with the tested microorganisms and then impregnating with 20  $\mu$ l of plant extract. Respective solvents were used as the negative control and gentamicin disc as the positive control. The plates were subsequently incubated at 37 °C for 24 hours. After incubation the growth inhibition zone were quantified by measuring the diameter of the zone of inhibition in mm [5].

#### 2.3.2 Well diffusion assay

An inoculam suspension was swabbed uniformly to solidified Nutrient agar for bacteria, and the inoculam was allowed to dry for 5 min., holes of 5 mm in diameter were made in the seeded agar using cork borer. Aliquot of 20  $\mu$ l from each plant crude extract was added into each well on the seeded medium and allowed to stand on the bench for 1 hour for proper diffusion and thereafter incubated at 37 °C for 24 hour. Respective solvents were used as the negative control and gentamicin (10  $\mu$ g/ml) as the positive control. The resulting inhibition zones were measured in mm [6].

### 2.4 Synergism of Plant Extracts along with Antibiotic and Non-antibiotic Drugs

The bacterium was inoculated on the surface of nutrient agar. Subsequently, the antibiotic disc

(diameter=5 mm) was placed on the surface of each inoculated plate and then added 20  $\mu$ l of *Carica papaya* extract to identify synergism effect between the papaya extracts and antibiotics. To identify synergism between the papaya extracts and Non-antibiotics, 10  $\mu$ l of Vitamin C and 10  $\mu$ l of papaya extracts were mixed and put together on a filter paper disc which was left for one hour to dry. The plates were incubated at 37 °C for 24 h. The diameters of clearing zones were measured.

### 2.5 GC-MS Analysis

2 µl of the petroleum benzene extracts of Carica papaya was employed for GC- MS analysis. The phytochemical investigation of petroleum benzene extracts was performed on a GC-MS equipment (Thermo Scientific Co.) Thermo GC-TRACE ultra ver.: 5.0, Thermo MS DSQ II. Experimental conditions of GC-MS system were as follows: TR 5-MS capillary standard non-polar column, dimension: 30 Mts, ID: 0.25 mm, Film thickness: 0.25 µm. Flow rate of mobile phase (carrier gas: He) was set at 1.0 ml/min. In the gas chromatography part, temperature programme (oven temperature) was 40 °C raised to 250 °C at 5°C/min and injection volume was 1 µl. Samples dissolved in chloroform were run fully at a range of 50-650 m/z and the results were compared by using Spectral library search programme. Interpretation on mass spectrum of GC-MS was done using the database of Thermo Scientific GC MS, Department of Chemistry, St. Thomas' College, Thrissur. The mass spectrum of the unknown component was compared with the spectrum of the known components stored in the GC - MS Library.

### 3. RESULTS AND DISCUSSION

### **3.1 Antibacterial Activity Assessment**

The water extract of *Carica papaya* tender leaves have shown an inhibitory zone of  $15\pm1.247$  mm in disc diffusion method and petroleum benzene extract of tender leaves has highest inhibition zone ( $9.5\pm0.236$  mm) in well diffusion method in *E. coli* (Table 1). Chloroform extract of seeds of papaya shows an inhibition zone of  $7.33\pm0.272$ mm in disc diffusion method and chloroform extract of green leaves has maximum inhibition zone of  $22.66\pm1.905$  mm in well diffusion method against *K. pneumoniae* (Table 2). Petroleum benzene extract of seed has maximum inhibition zone in disc diffusion method (15.0 mm) and in well diffusion method water extract of seed shows maximum diameter in inhibition zone (8.66  $\pm$  0.272 mm) against *P. aeruginosa* (Table 3). The inhibition zone formed by ethanol extract of green leaves is maximum (10 $\pm$ 0.707 mm) against *S. aureus* in disc diffusion method. But in well diffusion method chloroform extract of seed has maximum inhibition zone (13.33  $\pm$  1.963 mm) (Table 4).

#### 3.2 Synergism of Plant Extract with Antibiotic Drug Gentamicin

The synergistic effect of Carica papaya extracts along with gentamicin shows that there is enhanced effect when water and ethanol extracts of all the plant materials such as tender leaves. green leaves, yellow leaves and seeds combined with gentamicin against E. coli (Table 5). Water and petroleum benzene extract of tender leaves of papaya along with gentamicin show enhanced effect against Klebsiella pneumoniae. Chloroform and ethanol extracts of tender leaves have enhanced effect on the bacteria when compared to the zone formed by the extracts alone but they have a zone which is lesser than that of Gentamicin alone (Table 6). The tender leaves extract of Carica papaya exhibits an enhanced effect on P. aeruginosa along with Gentamicin. But its inhibition zone is reduced than that of gentamicin alone (22.33  $\pm$  0.272 mm). Green leaves extracts do not have any effect on P. aeruginosa, but when they combines with gentamicin inhibition zone is formed which is smaller than that of inhibition zone formed by gentamicin alone (Table 7). The water extract of tender leaves, all the extracts of green leaves and yellow leaves has much greater effect than that of gentamicin alone  $(19.66 \pm 0.720 \text{ mm})$ against S. aureus (Table 8).

### 3.3 Synergism of Plant Extracts with Nonantibiotic Drug Vitamin C

Ethanol extract of tender leaves and petroleum benzene and chloroform extract of green leaves have an enhanced effect on *E. coli* when combined with Vitamin C (Table 9). Chloroform and ethanol extract of tender leaves; petroleum benzene extract of green leaves and chloroform extract of yellow leaves and seeds along with Vitamin C have an enhanced effect on *Klebsiella pneumoniae* (Table 10). Ethanol extract of tender leaves and green leaves and chloroform extract of green leaves and seeds have an enhanced effect when combined with Vitamin C (Table 11). Chloroform and ethanol extracts of tender leaves along with Vitamin C produces an inhibition zone having diameter of 6.0 mm in *S. aureus* (Table 12).

## 3.4 GC-MS Analysis

GC MS analysis of petroleum benzene extract of tender leaves shows a chromatogram having 15 peaks (Fig. 1-A). The chemical constituents comprises 9-octadecenoicacid(z)-tetradecylester; Decvl oleate: Cyclobuta(a)dibenzo(c.f)cyclo heptadiene,7-oxo-; Naphthalene-1,2,3,4-Benzene-1, tetrahydro-1-phenyl-; 1'-(1,2cyclobutanediyl)bis-,trans-; Benzene-1,1'-(1,1,2,2-tetramethyl-1,2-ethanediyl)bis-; Caffeine; decanoicacid, ethylester; Hexa 2.3-Dihydroxypropylelaidate; Hexadecanoic acid,1-(hydroxylmethyl)-1,2-ethanediylester; Linoleicacid ethylester; 9,12,15-octadecatrienoic acid, 2,3-dihydroxypropylester,(z,z,z); N.N'-Bis(carbobenzyloxy)-lysine methyl(ester); Octadecane, 3-ethyl-5-(2-ethylbutyl)and Oleicacid, eicosyl ester.

GC MS analysis of petroleum benzene extract of green leaves produce a chromatogram (Fig. 1-B) having peaks of 10 compounds. They includes Ethanol,2-(9-octadecenyloxy)-,(z); n-Hexadecanoic acid; 3-oxo-5-phenylpentanoicacid, ethylester; 5,9-undecadien-2-one,6,10-dimethyl-,(E)-; 4-Hydroxy- $\beta$ -ionone; Octahydrobenzo[b]pyran,4a-acetoxy-5,5,8a-trimethyl-; 13,heptadecyn-1-ol; Hexadecanoic acid, ethyl ester and 9-octadecenoicacid(z)-2-hydroxy-1-(hydroxymethyl) ethyl ester.

12 peaks are seen in the chromatogram of yellow leaves. The major components present in the yellow leaves are 5,9-undecadien-2-one,6,10dimethyl-(E)-; Ingol 12-acetate; 2,7-Diphenyl -1,6-dioxopyridazino[4,5:2',3']pyrrolo[4',5'd]pyridazine; Ethyl iso-allocholate; (5.9-Dimethyl-1-(3-phenyl-oxiran-2-yl)-deca-4,8-dienylidene-1-(2-phenyl-aziridine-1-yl)-amine;13-Heptadecyn-1ol;1-Heptatriacotanol;Ethanol,2-(9octadecenyloxy)-,(z)-; 9,19-cyclolanost-24-en-3ol,acetate,(3); 1H-cyclopropa(3,4) benz(1,2e)azulene-5,7b,9,9a-tetrol,1a,1b,4,4a,5,7a,8,9octahydro-3-(hydroxylmethyl)-1,1,6,8-tetrame thyl-5,9,9a-triacetate,  $[1aR-(1a\alpha, 1b\beta, 4a\beta, 5\beta, 7b\alpha,$ 8α.9β.9aα)]and Cholest-22-ene-21-ol.3.5dehydro-6-methoxy-, pivalate.

Method	Inhibition zone (mm)*											
		Dis	c diffusion me	ethod	Well diffusion method							
Solvent	Water	Petroleum	Chloroform	Ethanol	Gentamicin	Water	Petroleum	Chloroform	Ethanol	Gentamicin		
		benzene					benzene					
Plant extract												
Tender leaves	15.0±1.247	6.0±0	6.0±0	6.66±0.544	19.0±3.742		9.5±0.236	11.0 <u>+</u> 2.160	10.66±0.98	28.33±2.178		
Green leaves				8.0 <u>+</u> 0.707	18.0 <u>+</u> 0.943			10.66±098	11.33±0.72	22.0 <u>+</u> 0.471		
Yellow leaves					20.0±0			11.0±0.943	10.0 <u>+</u> 0.471	21.66±0.272		
Seed				6.33 <u>+</u> 0.272	16.0 <u>+</u> 0.816			12.66±1.186	15.33±1.089	19.0 <u>+</u> 0.943		
Negative								7.0 <u>+</u> 0	6.0 <u>±</u> 0			
Control												

### Table 1. Antibacterial activity against E. coli

\* Mean±SE

## Table 2. Antibacterial activity against Klebsiella pneumonia

Method	Inhibition zone (mm)*											
		Dis	c diffusion m	ethod		Well diffusion method						
Solvent	Water	Petroleum	Chloroform	Ethanol	Gentamicin	Water	Petroleum	Chloroform	Ethanol	Gentamicin		
		benzene					benzene					
Plant extract												
Tender leaves	6.0±0		6.0±0	6.5±0.354	14.66 <u>+</u> 0.683			9.0 <u>+</u> 0.471	9.66 <u>+</u> 0.981	20.66 <u>+</u> 0.544		
Green leaves				9.0±0	14.33 <u>+</u> 0.72			22.66±1.905	12.66 <u>+</u> 0.544	18.0 <u>+</u> 0.943		
Yellow leaves	7.0±0		6.0±0	8.33 <u>+</u> 0.544	12.66 <u>+</u> 0.272			13.33 <u>+</u> 2.596	10.33 <u>+</u> 1.186	21.33 <u>+</u> 0.544		
Seed	7.33±0.272		7.33 <u>+</u> 0.272		13.0 <u>+</u> 0			12.5 <u>+</u> 1.768	14.33 <u>+</u> 2.177	20.0 <u>+</u> 0.471		
Negative				6.0±0				7.0±0	9.0±0			
control												

\* Mean±SE

Method	Inhibition zone (mm)*											
		Dise	c diffusion m	ethod		Well diffusion method						
Solvent	Water	Petroleum	Chloroform	Ethanol	Gentamicin	Water	Petroleum	Chloroform	Ethanol	Gentamicin		
		benzene					benzene					
Plant extract												
Tender leaves			6.0 <u>±</u> 0	9.0±0.471	19.0 <u>+</u> 0.471			8.66±0.981	9.0 <u>+</u> 0.816	29.0 <u>+</u> 0.471		
Green leaves					22.0±0.943			7.0±0	9.33±0.981	28.66±.544		
Yellow leaves					23.0±0.471			8.33±0.544	10.0±0.471	29.33±0.272		
Seed	7.66±0.544	15.0 <u>+</u> 0		7.33±0.667	20.66±0.272	8.66±0.272		14.0 <u>+</u> 2.449	11.66±1.44	29.0 <u>+</u> 0.471		
Negative								8.0 <u>+</u> 0	7.0±0			
Control												

## Table 3. Antibacterial activity against P. aeruginosa

\* Mean±SE

Table 4. Antibacterial a	ctivity against	S. aureus
--------------------------	-----------------	-----------

Method		Inhibition zone (MM)*											
		Disc	diffusion me	thod		Well diffusion method							
Solvent	Water	Petroleum	Chloroform	Ethanol	Gentamicin	Water	Petroleum	Chloroform	Ethanol	Gentamicin			
		benzene					benzene						
Plant extract	_												
Tender leaves				$6.66 \pm 0.272$	$23.33 \pm 1.361$			9.0 <u>+</u> 0.816	12.0±1.972	30.0±0			
Green leaves				10.0±0.707	19.0 <u>+</u> 0			8.0 <u>+</u> 0.707	8.0 <u>+</u> 0	26.0±0.471			
Yellow leaves			7.0±0	$7.66 \pm 0.544$	19.66±0.72			11.0±1.699	12.0 <u>+</u> 2.824	29.66±0.272			
Seed	7.0 <u>+</u> 0	8.0 <u>+</u> 0.707	6.5±0.354	$7.66 \pm 0.544$	$20.33 \pm 0.544$			20.33±1.963	13.66±0.272	28.0±0			
Negative control								7.0±0	6.0±0				

\* Mean±SE

Plant extracts	Gentami-cin	Tender	Tender leaves*		leaves*	Yellow	leaves*	Seeds *	
	alone*	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen
Water		15.33±1.44	21.0±0.471	13.33±0.72	18.66±1.905	12.66±0.72	17.0±0.816	13.33±0.72	19.0±1.247
Petroleum benzene			19.0±0.471		19.0±1.247		19.66±1.186		13.66±0.544
Chloroform	19.0 <u>+</u> 3.742	6.0±0	19.66±0.272		17.33±1.186		18.33±0.544		18.0±0.471
Ethanol		7.0 <u>±</u> 0	18.33 <u>+</u> 0.272	6.0 <u>±</u> 0	16.0 <u>+</u> 1.247	6.0 <u>±</u> 0	15.33 <u>+</u> 2.126	6.33 <u>+</u> 0.272	17.33 <u>+</u> 1.186

### Table 5. Synergism with gentamicin against E. coli

#### \* Mean±SE

### Table 6. Synergism with gentamicin against Klebsiella pneumonia

Gentami-cin	Tender leaves*		Green leaves*		Ye	llow leaves*	Seeds *		
alone*	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen	
	6.0±0	15.66±0.720		14.0 <u>+</u> 0.471		13.33 <u>+</u> 0.272	6.0±0	12.33 <u>+</u> 0.272	
		15.33 <u>+</u> 0.981		15.66±0.981		15.66 <u>+</u> 0.544		6.66±0.331	
14.66±0.683		13.33±0.272		10.33±0.272		16.33±0.720		14.33±0.272	
	6.0 <u>±</u> 0	14.0 <u>+</u> 0.471	6.33 <u>+</u> 0.272	15.66±0.544		17.0 <u>+</u> 0.471	6.0±0	13.0±0.943	
	Gentami-cin alone* 14.66±0.683	Gentami-cin  Tend    alone*  Ext    6.0±0     14.66±0.683     6.0±0	Gentami-cin alone*  Tender leaves*    6.0±0  15.66±0.720     15.33±0.981    14.66±0.683     6.0±0  14.0±0.471	Gentami-cin alone*  Tender leaves*  Green    6.0±0  15.66±0.720     14.66±0.683   13.33±0.272    6.0±0  14.0±0.471  6.33±0.272	Gentami-cin alone*  Tender leaves*  Green leaves*    alone*  Ext  Ext+Gen  Ext+Gen    6.0±0  15.66±0.720   14.0±0.471     15.33±0.981   15.66±0.981    14.66±0.683   13.33±0.272   10.33±0.272    6.0±0  14.0±0.471  6.33±0.272  15.66±0.544	Gentami-cin alone*  Tender leaves*  Green leaves*  Ye    alone*  Ext  Ext+Gen  Ext  Ext+Gen  Ext    6.0±0  15.66±0.720   14.0±0.471     14.66±0.683   13.33±0.272   10.33±0.272     6.0±0  14.0±0.471  6.33±0.272  15.66±0.544	Gentami-cin alone*Tender leaves*Green leaves*Yellow leaves*alone*ExtExt+GenExtExt+GenExt+Gen $6.0\pm0$ $15.66\pm0.720$ $14.0\pm0.471$ $13.33\pm0.272$ $$ $15.33\pm0.981$ $15.66\pm0.981$ $15.66\pm0.544$ $14.66\pm0.683$ $13.33\pm0.272$ $10.33\pm0.272$ $$ $16.33\pm0.720$ $6.0\pm0$ $14.0\pm0.471$ $6.33\pm0.272$ $15.66\pm0.544$ $17.0\pm0.471$		

\* Mean±SE

### Table 7. Synergism with gentamicin against P. aeruginosa

Plant extracts	Antibiotic	Tender leaves*		Green leaves*		Yello	Yellow leaves*		Seeds *	
	alone*	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen	
Water			19.0 <u>+</u> 0		20.66±0.544	7.5 <u>+</u> 1.061	19.0 <u>+</u> 0.471		17.66 <u>+</u> 1.089	
Petroleum benzene		6.0 <u>±</u> 0	19.0 <u>+</u> 0.471		21.66±0.272		20.66±0.544		13.0 <u>+</u> 0.816	
Chloroform	22.33 <u>+</u> 0.272	6.66±0.272	20.0±0.471		21.66±0.272		16.66±2.596		15.33 <u>+</u> 2.228	
Ethanol		$6.33 \pm 0.272$	20.0±0.943		22.0±0.816	6.0±0	19.0 <u>+</u> 0.816	7.0 <u>+</u> 0	17.66±1.186	

\* Mean±SE

Plant extracts	Antibiotic	Tender leaves*		Green	leaves*	Yellow	leaves*	Seeds *	
	alone*	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen	Ext	Ext+Gen
Water		12.66±0.72	20.66±0.544	15.0±1.247	22.33±1.361	15.66 <u>+</u> 0.272	22.33±1.186	17.0 <u>+</u> 5.312	14.0 <u>+</u> 0.816
Petroleum benzene			19.33 <u>+</u> 0.544		23.33±1.361		22.0 <u>+</u> 0.943	6.5±0.354	15.0 <u>+</u> 2.357
Chloroform	19.66±0.72	6.5±0.354	18.66±1.186		22.66±3.662		23.33±2.177		15.66±1.905
Ethanol			18.0 <u>+</u> 0.943	7.33±0.720	23.33±1.785	6.5 <u>+</u> 0.354	23.66±1.515	6.0 <u>±</u> 0	14.0 <u>+</u> 0.471
Ethanol			18.0 <u>±</u> 0.943	7.33 <u>+</u> 0.720	$23.33 \pm 1.785$	6.5 <u>±</u> 0.354	23.66 <u>+</u> 1.515	6.0 <u>±</u> 0	14.0 <u>±</u> 0

### Table 8. Synergism with gentamicin against S. aureus

#### \* Mean±SE

## Table 9. Synergism with vitamin C against E. coli

Plant extracts	Vitamin C	Te	Tender leaves*		en leaves*	Yel	low leaves*	Se	eds*
	alone	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC
Water				7.0±0	6.0±0			6.0±0	7.0 <u>±</u> 0
Petroleum benzene		6.0±0			6.0±0				6.0±0
Chloroform		6.0±0	6.0±0		6.0±0				6.0±0
Ethanol		6.0±0	6.33±0.272	7.0±0.471	6.0 <u>±</u> 0	$6.0\pm0$	6.5±0.354	6.33±0.272	6.33±0.272

\* Mean±SE

### Table 10. Synergism with vitamin C against Klebsiella pneumonia

Plant Extracts	Vitamin C	Ten	der leaves*	Gree	n leaves*	Ye	llow leaves*	Se	eds*			
	alone	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC			
Water								6.5 <u>+</u> 0.354	6.0 <u>+</u> 0			
Petroleum benzene		$6.0\pm0$	6.0 <u>±</u> 0		6.0 <u>+</u> 0							
Chloroform			6.0±0			8.0±0			6.33±0.272			
Ethanol		$6.0\pm0$	6.33±0.272	6.33±0.372	6.0±0	$6.0 \pm 0$	6.5±0.354	$7.66 \pm 0.544$	6.0±0			
	* Mean±SE											

Plant extracts	Vitamin C	Tende	r leaves*	r leaves* Green leaves		Yellov	w leaves*	Se	eds*
	alone	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC
Water									
Petroleum benzene		6.0±0	6.0±0						
Chloroform		6.33±0.272	6.0±0		6.5±0.289				6.0±0
Ethanol		6.66 <u>+</u> 0.474	7.33 <u>+</u> 0.469	7.0 <u>+</u> 0.471	8.0 <u>+</u> 0	7.5 <u>+</u> 0.354	6.66 <u>+</u> 0.272	6.66 <u>+</u> 0.544	6.0 <u>±</u> 0

## Table 11. Synergism with vitamin C against P. aeruginosa

\* Mean±SE

### Table 12. Synergism with vitamin C against S. aureus

Plant extracts	Vitamin C alone	Tender leaves*		Green leaves*		Yellow leaves*		Seeds*	
		Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC	Ext	Ext+VitC
Water									
Petroleum benzene									
Chloroform			6.0±0						
Ethanol		6.0±0	$6.0\pm0$	7.0±0.471				7.0±0	6.0±0

In all the tables values are expressed as Arithmetic Mean  $\overline{(x)} \pm S.E$  of three replicates

The GC MS analysis of seed extract produces a chromatogram of 5 peaks (Fig. 1-c). They include (isothiocyanatosmethyl)-, Benzene, Hexadecanoic acid, methyl ester; n-Hexadecanoic 9-octadecenoicacid(z)acid; methyl ester and 6-octadecenoic acid. The major component is the 6-octadecenoic acid which is followed by the n-Hexadecanoic acid and Benzene, (isothiocyanatomethyl).

Ethyl iso-allocholate present in yellow leaf extract is a sterol with antimicrobial, diuretic, antiinflammatory, antiasthmatic, anti tumor, cancer preventive and chemopreventive activities. Ingol 12-acetate is a diterpene with antileukemic properties. 5,9-undecadien-2-one,6,10-dimethyl ester is a proven anticancer agent, used as an antidote, it decreases endothelial platelet adhesion and epinephrine production [7].



#### A. Chromatogram of tender leaves

B. Chromatogram of green leaves





Fig. 1. Chromatograms of petroleum benzene extracts

#### 4. CONCLUSION

The aqueous and organic solvents extracts of leaves and seed of Carica papaya exhibits antibacterial activity against the most common disease causing bacteria's such as E. coli, K. pneumoniae., P. aeruginosa and S. aureus. They also found to have synergistic effect with an antibiotic drug Gentamicin and a non-antibiotic drug Vitamin C. The presence of various bioactive compounds justifies the use of the whole plant for various ailments by traditional practitioners. It could be concluded that Carica papaya L. plant is of phytopharmaceutical importance. Studies with these compounds may yield nature friendly strong anti-microbial agents of agricultural importance and anti-cancer drugs.

#### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

### REFERENCES

1. Aravind G, Debjit Bhowmik, Duraivel S, Harish G. Traditional and medicinal uses of *Carica papaya*. Journal of Medicinal Plants Studies. 2013;1(1):7-15.

- Heywood VH, Brummitt RK, Culham A, Seberg O. Flowering plant families of the world. Firefly Books. 2007;88. ISBN 9781554072064
- Available:<u>http://www.nutrition-and-you.com/</u> Papaya fruit nutrition facts and health benefits.
- Krishna KL, Paridhavi M, Patel Jagruti A. Review on nutritional, medicinal and pharmacological properties of papaya (*Carica papaya* Linn.). Indian Journal of Natural Products and Resources. 2008; 7(4):364-373.
- Kumara M, Agarwala R, Deyb K, Raib V, Johnsonc B. Antimicrobial activity of aqueous extract of *Terminalia chebula* Retz. on gram positive and gram negative microorganisms. International Journal of Current Pharmaceutical Research. 2009;1(1):56-60.
- Obeidat M, Shatnawi M, Al-alawi M, Al-Zubi E, Al-Dmoor H, Al-Qudah M, El-Qudah J, Otri I. Antimicrobial activity of crude extracts of some plant leaves. Research Journal of Microbiology. 2012;7:59-67.
- 7. Edward P, Claus E, Varro LR. Pharmacognosy. 6<sup>th</sup> ed. LEA and Febiger. 1987;184-187.

© 2016 Francis and Jose; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

> Peer-review history: The peer review history for this paper can be accessed here: http://sciencedomain.org/review-history/15811