



Different Areas of *Carica papaya* Linn Display Antifungal and Antimicrobial Activity

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The plants of *Carica papaya* are utilised for nutritional and therapeutic purposes. The phytochemical examination of the leaf extract of *Carica papaya* reveals all phytochemical elements, including saponin, flavonoid, glycoside, alkaloid, carbohydrates, tannins, and steroids. A systematic phytochemical analysis was used to check all extracts for the presence of phytochemical components. Free radical scavenging; wound healing; neuroprotection; diuretic; abortifacient; antifertility; anti-sickling; neuroprotective; diuretic; abortifacient; Antimicrobial properties of papaya seed extracts have been shown to be effective against *B. cereus*, *S. faecalis*, *S. aureus*, and a variety of bacterial pathogens. Anti-inflammatory properties of an ethanolic extract of *Carica papaya* L. leaf were investigated in rats with paw oedema. In order to create the extract, many solvents were used such as alcohol, methanol, and ethanol as well as aqueous extracts such as acetone and chloroform. The research indicates that the latex of papaya fruits exhibits substantial efficacy against *Candida albicans*. The well diffusion approach inhibits *S. aureus* more effectively than *E. coli* when using methanol extract. It has antifungal, antibacterial, anti-inflammatory, antioxidant, anti-diabetic, wound-healing, and analgesic properties, among others. According to the findings of this investigation, when given in doses of 50, 75, and 100 mg/ml, *Carica papaya* leaf extract had more antifungal activity against *Colletotrichum gloeosporioides* than *Fusarium* spp., *Rhizopus stolonifer*, and antibacterial activity than aqueous papaya leaf extract.

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1. INTRODUCTION

Carica papaya, often known as papaya, papayer, tinti, and pepol, is a member of the Caricaceae family. *Carica papaya* is a fast-growing tree that may reach a height of 7-8 metres and has a trunk diameter of 20 centimetres, hollow leaf scars, spongy-fibrous tissue, and an extensive root system. It also produces copious amounts of latex throughout the entire plant [1].

A year-round fruit that is readily available and packed with nutrients is the papaya. An abundance of antioxidants may be found in fruits and vegetables, including beta-carotene, vitamin C, vitamin B complex, flavonoids, folate, and pantothenic acids [2,3]. Meat is tenderised with papain and particular enzymes found in papaya fruits and latex. The papain enzyme is isolated as dried and refined latex from green grapes. Papain is supposed to aid in the treatment of skin discolorations, diphtheria, and even cancer [4].

Papaya's nutritional and culinary value is well-known across the world. The papaya plant's leaves, fruit, seeds, latex, and roots all contain bioactive substances. There are several plant compounds that have analgesic and antibacterial properties as well as cardiogenic and digestive, emmenagogue, hypotensive, and laxative properties [5]. Dengue patients were treated with a palm oil solution made from papaya leaves from the *Carica* genus. Scientific evidence supports the use of *Carica papaya* leaf extract in the treatment of dengue fever [6]. *Candida albicans* has been demonstrated to be inhibited by papaya seed extract and oil [7].

In order to analyse the biological activity of *Carica papaya* fruits, shoots and leaves and seeds and roots and latex, several investigations have been carried out [8]. Jam, desserts, and pulp from *Carica papaya* have more nutritional content than the leaves and seeds, which are used to create tea and flour.

The pulp contains three substantial sources of vitamin A, C, and E antioxidants [9]. One of the fastest growing trees in the world, the *Carica papaya* may reach heights of 10 to 12 feet. It is comparable to pepsin in that it also contains a soft yellow resin, fat, albuminoid sugar, and pectin in green fruits. Glucosides carposide and carpine are found in the leaves [10]. Spiral-leaved leaves adorn the top of the trunk of the

papaya tree, which grows to enormous proportions in the tropics. The enormous leaves have a diameter of 20 to 28 centimetres and a length of 50 to 70 centimetres. Papaya trees have unbranched flowers that resemble plumerias but are smaller and waxier if they aren't clipped. Giant fruits of 10 to 30 centimetres in diameter and 15 to 45 centimetres in length grow on the leaf axils. This means that the fruit is ripe when its skin turns from amber to orange in colour and the flesh is pliable to the touch. In size and shape, papayas are similar to melons, and they hang on long, thick peduncles. In addition to [9, 10]., Papaya latex's antifungal and antibacterial properties are dependent on the plant's medicinal potential.

1.1 Plant Profile of *Carica papaya*



Fig. 1. *Carica papaya*

Synonyms: papaya, papayer, tinti, pepol

Biological Source: It is cultivated fruiting tree known as *Carica papaya* belongs to family Caricaceae and It is one of the most popular and commercially important plants in the world for its nutritional and medicinal value [9].

Table 1. Different species of *Carica papaya* Linn

<i>C. candamarcensis</i>	<i>C. glandulosa</i>
<i>C. Mexicana</i>	<i>C. goudotiana</i>
<i>C. caudate</i>	<i>C. heterophylla</i>
<i>C. cauliflora</i>	<i>C. candicans</i>
<i>C. chilensis</i>	<i>C. longiflora</i>
<i>C. horovitziana</i>	<i>C. crassipetala</i>
<i>C. cundinamarcensis</i>	<i>C. weberbaueri</i>

[10, 11, 13, 14, 15]

1.1.1 Taxonomical classification

Kingdom : Plantae
 Subkingdom : Tracheobionta
 Division : Magnoliophyta
 Class : Magnoliophyta
 Family : Caricaceae
 Genus : Carica L.
 Species : *Carica papaya* L [9].

Varnacular Names

Hindi : Papita
 English : Papaya
 Brazil : Mamao [12].

1.1.2 Morphological characteristics

Carica papaya plant: It is a dicotyledonous, polygamous, diploid species of papaya plants and huge perennial herbs with a quick growth rate. The papaya tree's trunk has a diameter of 6 to 20 feet and is composed of soft wood (1.8-6.1m) [11].

Leaf: The diameter of the leaves ranges from 50 to 70 centimetres, with long petioles and 5-7 lobes. The leaves are severely cut and divided. The mature leaves are palmate with deep lobes and big in size [11].

Flower- Female flowers are large, solitary, or in small racemes, whilst male flowers are highly pubescent, many-flowered cymes at the ends of the pendulous [13].

Fruit- It features a large hollow core with a lengthy, elongated form whose dimensions vary widely.

Seed- The seeds are tuberculous, black, and encased in an aril that is transparent.

Latex- Latex is a milky substance found in unripe fruit that includes the protein papain that has been fermented [11, 13].

1.1.3 Chemical constituents

Fruit, juice, seeds, roots, leaves, bark, and latex are only some of the papaya's various chemical components.

Fruit- An example of a volatile substance includes amino acids, citric acid and benzylisothiocyanate. Important nutrients include calcium, iron, vitamin C, thiamine, riboflavin, niacin, and carotene [16].

Juice- A wide variety of lipids and myristic acids as well as linolenic acid, linlenic acid, oleic acid, and linolenic acid [17].

Seed- Fats, protein and fibre from papaya, caricin, carpain, and the enzyme myrosin are all found in papaya [17].

Root- Enzyme called myrosin and a carbohydrate called carboside

Leaves- In addition to carpain and pseudocarpain, alkaloids include choline, dehydrocarpaine I and II, dehydrocarpaine III, and the vitamins C and E [18].

Bark- Glucose, fructose, sucrose, xylitol, and sitosterol are all types of sugars.

Latex- Peptidase A and B, papain, chymopapain, lysozymes, and lysozymes [18].

Table 2. *Carica papaya* L. (papaya) macronutrient and fibre content per 100 g of ripe fruit pulp, seeds, and leaves

Composition	Pulp	Seeds	Leaves
Protein	0.6g	2.6g	5.8g
Lipids	0.1g	3.1g	1.8g
Carbohydrates	7.2g	43.6	74.2g
Fibres	0.8g	2.1g	13.1g
Energy	32.1kcal	212.7kcal	348.6 kcal

[12,17]

1.1.4 Application

1.1.4.1 Ripe fruit extracts

It is used to treat ringworm, malaria, and hypertension, among other medical conditions.

1.1.4.2 Leaves extract

Patients with dengue should increase their platelet count. It is also used as an anthelmintic, antibiotic, antifertility, and antiamoebic drug.

1.1.4.3 Papaya seed extract

Candida albicans exerts an antifungal effect. It is also used as an analgesic, an amebicide, a pectoral, a cardi tonic, a cholagogue, a digestive, a stomachic, an anticancer febrifuge, a hypotensive, a laxative, a wound-healing agent, etc.

1.1.4.4 Unripe fruits extract

It is used in traditional medicine to cure ulcers. nutritional function, the cardiovascular system, and colon cancer prevention.

1.1.4.5 Leaf extracts

Extract from leaves inhibits the formation of sickle cells.

1.1.4.6 Latex

Antifungal properties Anti-inflammatory, analgesic, amebicide, digestive, febrifuge, and laxative properties. Dermatitis and psoriasis, among others Abortion.

1.1.4.7 Papaya juice

Carotene, vitamin C, and vitamin B are antioxidant substances.

1.1.4.8 Flowers

Concoction, infusion Jaundice, a cough, hoarseness, bronchitis, laryngitis, and tracheitis are all signs of tracheitis.

1.1.4.9 Roots/barks

Decoction, infusion, and poultice toothache, gastrointestinal, tonic, and contraceptive [2, 5, 6, 7, 17-22].

1.1.5 Pharmacological activity

This fruit, in addition to being an excellent source of antioxidants and free radical scavenging properties as well as anti-inflammatory and hypolipidemic capabilities as well as wound-healing, anti-sickling as well as hypotension and hypoglycemia.

1.1.6 Antibacterial activity

Papaya seed extracts are antibacterial against *P. vulgaris*, *S. faecalis*, *S. aureus*, *B. cereus*, and *E. coli*. As well as for *B. subtilis*, it was found that the maximum inhibitory zones for *B. typhi* and *Staph aureus* were also detected, as well as for *aeruginosa* (15.0 mm). Papaya latex has antifungal effects that reduce *Candida albicans* infection by 60% [15].

1.1.6.1 Wound healing activity

Both papaya latex coagulation and mammalian coagulation have a comparable component. Cloth development during the healing process in animals. Some plant metabolites may have a similar function during plant healing. Papain contains anti-burning properties [23].

1.1.6.2 Anti-inflammatory activity

Rats with paw oedema were used to test an ethanolic extract of *Carica papaya* L. leaves for anti-inflammatory effects. From the fourth to the tenth day of the trial, the extract significantly reduced persistent oedema in a formaldehyde arthritis model [24].

1.1.6.3 Anti-diabetic activity

Carica papaya L. leaf extract was tested for its anti-diabetic properties in an experimental rat model. *Carica papaya* L. leaf chloroform extract significantly reduces blood glucose, transaminases, and lipid levels in diabetic rats [25].

1.1.6.4 Antimicrobial activity

The antibacterial activities of *Carica papaya* L. leaf preparations have been demonstrated. Various solvents, including ethanol (300, 600, and 900L):methanol (300, 600, and 900L):ethyl acetate (300, 600, and 900L):acetone, chloroform (300, 600, and 900L):petroleum ether, hexane, and aqueous extract, were used to produce the extract, which was effective against bacteria and fungi. *Candida albicans* was more resistant to acetone extract than other fungi [2].

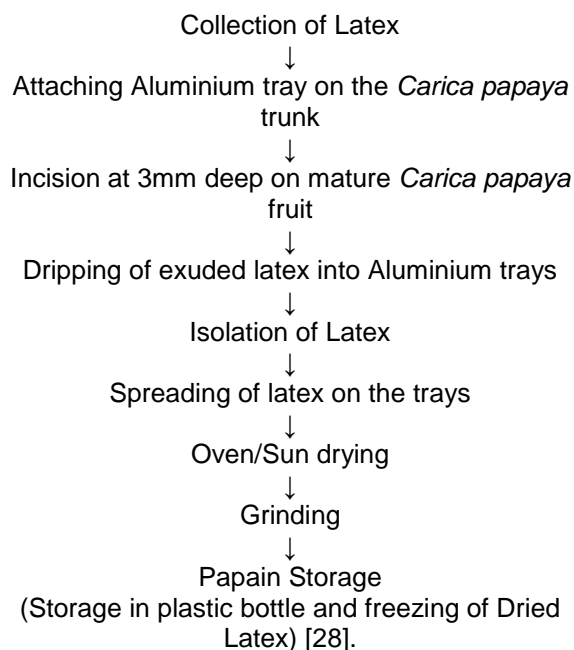
1.1.6.5 Analgesic activity

Three *Carica papaya* L. leaf extracts were evaluated for their analgesic potential in a mouse model of acetic acid-induced pain. Analgesic activity was established by n-hexanes, Ethyl Acetates, and Ethanol in comparison to aspirin [26].

1.1.6.6 Anti-hypertensive activity

As an antihypertensive agent, papaya leaf decoction can be utilised. The papaya plant has hypotensive action when taken orally [27].

1.1.7 Isolation of latex



2. MATERIALS AND METHODS

2.1 Collection of Plant Materials

Latex is extracted from *C. papaya* fruits by incisions performed using a knife made of stainless steel. The fruit exuded fresh latex, which was collected in a beaker [1]. To evenly dissolve the latex in the methanol, shake 500 l of latex for 5 minutes.

2.2 Latex Extraction and Collection

Early in the months of June and July, the latex of mature unripe fruit was extracted with a stainless-steel knife. Collecting the latex in a beaker or plastic plate. The latex was mixed with potassium meta-bisulphite ($K_2S_2O_5$) at a weight-to-weight ratio of 0.5 percent [6].

2.3 Latex Drying and Storage

Latex was collected in aluminium trays and dried in a tray drier at 40°C and 746, 6 mbar pressures for two hours. After drying, the latex was transferred to a plastic bottle and kept at 20°C until analysis [7].

2.4 Collection of Microorganisms

Bacteria with a Gram stain Gram-negative bacteria and *Bacillus subtilis* In the microbiology lab, strains of *Escherichia coli*, *Agrobacterium* sp., and *Rhizobium* sp. were grown. This was coded in C. investigation. *C. albicans* *C. glabrata* *C. Krusei* in addition to *C. tropical* mushroom types. The fungi are *Fusarium* spp., *Colletotrichum gloeosporioides*, and *Rhizopus stolonifer*. These test cultures were maintained on nutrient agar and subcultured in LB broth medium every two weeks [3, 7].

2.5 Preparation of Culture Medium

Except for agar, all of the materials were properly dissolved in 1 L of double-distilled water to produce 1 L of broth medium. To produce agar plates, 15 g/L of agar is added to the solidifying medium of choice [5]. After adjusting the pH of the medium to 7.0 with 1 N NaOH, it was autoclaved for 20 minutes at roughly 121°C [6].

2.6 Disk Diffusion Assays

Using the disk-diffusion technique, the antibacterial activity of leaf extracts was analysed. 20 mL of sterile agar medium containing 100 mL of microbial cell suspension are injected into 100 mL of sterile Petri plates [2, 3]. The Whatman No. 1 was used to cut discs of six different diameters. Different concentrations of papaya latex (1, 2, 5, 7, 10, 15, and 20 mg/disk) were applied to paper discs using a micropipette and dried under laminar airflow for 5–10 minutes. Spreader used to inoculate agar solid medium in plates with test organisms before agar plates were inoculated [4].

2.6.1 Anti-fungal activity

The greatest growth inhibition produced by the Ethanolic extract of papain is reported to be greater than the positive control. The outcome indicates that papain has substantial antifungal activity against *Candida albicans*. Papain was isolated from crude latex and derived from unripe papaya fruit, and the presence of protein was verified using the ammonium per sulphate precipitation method.

Table 3. The basic confirmatory test shows the presence of papain [29]

Fungi	Papain (conc.) µl				
	10	20	30	40	50
<i>Candida albicans</i>	-	-	13mm	15mm	18mm

2.6.2 Confirmation of papain

The sample of 0.01g should be heated to 37 degrees Celsius. The liquid solidifies. The solution of papain displays maximum absorbance between 270 and 280 nm. Twenty milligrammes of protein, or 20 milligrammes per decilitre, were detected in the sample [30].

2.7 Physiochemical Parameters

Yellow essential oil was produced from papaya seeds. Overall, the yield (w/w) was 0.2 percent. Essential oil has a specific density of 1.124 g•mL⁻¹ at 25°C and a refractive index of 1.6026 at 20°C [31].

2.7.1 Papaya seed oil extraction using the Soxhlet technique with ultrasound assistance

n-hexane was used as the solvent during an ultrasound-assisted extraction of papaya seed oil. In a supersonic bath, the mixture was sonicated for 30 minutes at 50 degrees Celsius. In a 250 mL Soxhlet extractor, 10 g of seed powder was put in a cellulose paper bag and extracted with n-hexane for eight hours. The liquid phase was then separated from the solvent using a rotary evaporator (RE-801, Yamato Scientific). The liquid phase was then used to extract the lipid.

2.7.2 Zone of inhibition

The antifungal activity of papaya seed essential oil was evaluated using the filter paper disc

diffusion technique. EO reduced the development of every fungal strain tested. EO was the most effective antifungal agent against *C. parapsilosis* [31].

Table 4. The product's ethanolic extracts prevent *Carica papaya* L. from developing mycelium (radial diffusion)

Fungal strain	Zone of inhibition	
	EO (100µg)	FLZ (25µg)
<i>C. albicans</i>	29.6±1.5	43.3±0.3
<i>C. glabrata</i>	29.6±0.8	24.1±0.9
<i>C. krusei</i>	32.9±1.1	21.7±0.4
<i>C. parapsilosis</i>	33.2±0.8	40.9±0.3
<i>C. tropical</i>	27.2±0.8	42.1±0.9

2.7.3 Antimicrobial activity measurement

The cup plate agar diffusion technique was used to evaluate the antibacterial activity of aqueous and organic extracts of the *Carica papaya* plant. The bacterial cultures were adjusted to a turbidity standard of 0.5 McFarland, and 100 l extract dilutions reconstituted in 50 percent DMSO and distilled water were added at concentrations of 200, 150, 100, and 50 mg/ml to each well of the previously seeded culture plates [9].

For 24 hours, the cultures were incubated at 37 °C. A well was created in each of the agar growth plates, and it was then filled with 20 l of the conventional antibiotic, ciprofloxacin, and 20 l of streptomycin, the positive control [8]. Antibacterial activity was measured in each well and the zone of inhibition.

Table 5. SUE seed (unripe) extract, SRE seed (ripe) extract, and LE Leaf extract [5]

Fungi spp.	Mycelial inhibition (%)			
	LE	SUE	SRE	Ridomil
<i>Fusarium spp.</i>	18.17±1.8	8.99±1.5	1.60±08	60.82±0.3
<i>Colletotrichum gloeosporioides</i>	21.84±1.3	0	0	51.20±1.3
<i>Rhizopus stolonifer</i>	0	0	0	32.56±0.9

Table 6. The antibacterial efficacy of *Carica papaya* leaf acetone extracts

Sr. No	Wound pathogen	Diameter of zone of inhibition (mm)			
		25mg/ml	50mg/ml	75mg/ml	100mg/ml
1	<i>Staphylococcus aureus</i>	0	0	10	14
2	<i>E. coli</i>	0	12	13	17
3	<i>Klebsiella pneumonia</i>	0	10	12	15
4	<i>Proteus vulgaris</i>	0	11	14	16
5	<i>Pseudomonas aeruginosa</i>	0	13	24	26

[8, 9]

Table 7. Antibacterial activity of *Carica papaya* leaf methanol extract

Sr. No	Wound pathogen	Diameter of zone of inhibition (mm)			
		25mg/ml	50mg/ml	75mg/ml	100mg/ml
1	<i>Staphylococcus aureus</i>	0	0	10	12
2	<i>E. coli</i>	0	10	12	13
3	<i>Klebsiella pneumonia</i>	0	10	11	14
4	<i>Proteus vulgaris</i>	0	12	13	15
5	<i>Pseudomonas aeruginosa</i>	0	14	17	24

[7, 11]

Table 8. The leaf extract of *Carica papaya* possesses antibacterial action

Sr. No	Wound pathogen	Diameter of zone of inhibition (mm)			
		25 mg/ml	50 mg/ml	75 mg/ml	100 mg/ml
1	<i>Staphylococcus aureus</i>	0	0	0	10
2	<i>E. coli</i>	0	0	11	11
3	<i>Klebsiella pneumonia</i>	0	0	10	11
4	<i>Proteus vulgaris</i>	0	0	12	13
5	<i>Pseudomonas aeruginosa</i>	0	0	14	15

[8, 9, 6]

2.8 Minimum Bactericidal Concentration (MBC) and Minimum Inhibitory Concentration (MIC) Determination (MIC)

Using test organisms, the Minimum Inhibitory Concentration of the extracts was estimated. After adding 2 ml of nutrient broth and 0.5 ml of different extract concentrations (5, 25, 50, 75, 100, 125, 150, 175 and 200 mg/ml): a loopful of the test organism diluted to the 0.5 McFarland turbidity standard was added to the test tubes. Ciprofloxacin is routinely used to treat test organisms. In a test tube, the test organisms were grown in nutritional broth. The culture tubes were then placed in an incubator at 37 degrees Celsius for 24 hours. After incubation, the tubes were inspected for microbial growth by observing turbidity [9].

A loopful of broth from the tubes in which no bacteria had grown was placed on sterile nutritional agar medium. To act as controls, only test organisms were streaked over nutrient agar plates. The plates were then incubated at 37°C for 24 hours [9].

3. RESULTS AND DISCUSSION

Papaya fruit is produced by papaya plants belonging to the Caricaceae family and the *Carica papaya* genus. The papaya plant is simple to cultivate and yields more per acre. The findings of *Carica papaya* plant materials with a

greater phytoconstituent concentration have been reported. Papaya leaves, fruit, and seed extracts include protein, vitamin C, carbohydrates, alkaloids, flavonoids, tannin, steroids, phenolic compounds, vitamin C, and saponins. The antibacterial action may be due to the presence of bioactive compounds such as phenols, saponins, steroids, alkaloids, and flavonoids [32,33].

In addition to reducing drug-induced hematotoxicity and nephrotoxicity, *C. papaya* possesses antibacterial, antimalarial, antiparasitic, anticancer, anti-inflammatory, and wound-healing activities [34,35]. *C. papaya* exhibits hypoglycemic, hypolipidemic, and antihypertensive potential as well as elevated antioxidant activity in relation to metabolic dysfunctions in both in vitro and in vivo experimental settings. Various components of *C. papaya* will be necessary for the development of innovative natural products for the treatment and prevention of obesity and metabolic illnesses [36,37].

The bactericidal activity of acetone, methanol, and aqueous leaf extracts of *Carica papaya* is summarised in Tables 2, 3, and 4. At dosages of 25mg/ml, 50mg/ml, 75mg/ml, and 100mg/ml, the methanolic extract of papaya leaf had greater wound healing efficacy against *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus vulgaris*, and *Pseudomonas aeruginosa*. Antifungal and antibacterial effects are provided by this concentration. The aforementioned

activities were diminished as the concentration increased or decreased. Table 1 summarises the papaya leaf ethanolic extract's antifungal effectiveness. The antifungal efficacy against *Colletotrichum gloeosporioides* is greater, but papain inhibits fungi with a diameter of 18 mm at concentrations of 50 I against *Candida albicans*. The antifungal action of papaya seed-derived essential oil is more potent against *Candida parapsilosis*.

4. CONCLUSION

Carica papaya fruits are abundant in antioxidant compounds, including beta-carotene, vitamin C, vitamin B, flavonoids, folate, panthotenic acids, and minerals. Plant components display analgesic, amebicide, antibacterial, cardiotoxic, cholagogue, digestive, emmenagogue, hypotensive, laxative, and stomachic properties. As a dengue treatment, papaya leaves from the *Carica* genus were dissolved in palm oil. It has been demonstrated that both papaya seed extract and oil has antifungal properties against *Candida albicans*. In addition to wound-healing, free radical scavenging, anti-sickling, anti-hypertension, hypoglycemia, and hypolipidemic properties, *carica papaya* also possesses antiviral, protozoan, antibacterial, antifungal, antiviral, anti-inflammatory, and antiviral properties. Papaya latex's antifungal effect on *Candida albicans* inhibits 60 percent of the fungal infection. In both in vitro and in vivo experimental settings, *C. papaya* has improved antioxidant activity, hypolipidemic, hypoglycemic, and antihypertensive potential. The bactericidal activity of acetone, methanol, and aqueous leaf extracts of *Carica papaya* is summarised in Tables 2, 3, and 4. At dosages of 25mg/ml, 50mg/ml, 75mg/ml, and 100mg/ml, the methanolic extract of papaya leaf had greater wound healing efficacy against *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus vulgaris*, and *Pseudomonas aeruginosa*. Antifungal and antibacterial effects are provided by this concentration. The aforementioned activities were diminished as the concentration increased or decreased. Essential oil made from papaya seeds has a stronger antifungal effect against *Candida parapsilosis*.

At dosages of 50 mg/ml, 75 mg/ml, and 100 mg/ml, methanolic and acetone leaf extracts were more effective than aqueous leaf extract against *Colletotrichum gloeosporioides*, *Fusarium spp.*, and *Rhizopus stolonifer*.

CONSENT AND ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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