ISSN No:-2456-2165

Antimicrobial Activities and Phytochemical Analysis of Ethanolic and Aqueous Leaf Extract of Pawpaw (Carica papaya) Grown in Maiduguri

Fanna Ali Baba, Hadiza Mala Musa, Zainab A Bello and Amina B Majama
Department of Science Laboratory Technology Ramat Polytechnic, Maiduguri, PO Box 1070, Borno State, Nigeria.

Abstract:- This study undertaken to determined the phytochemical screening and antimicrobial activity of ethanol and aqueous extract of Pawpaw leaf. A Fresh leaves of Pawpaw were collected from the Department Agricultural Technology, Ramat polytechnic Maiduguri, and allowed to dry under the shed for 10 days. The powdered plant materials were soaked in a conical flask and extracted with ethanol and aqueous. Phytochemical analysis was conducted by using specific standard procedure, Antimicrobial activity assays of all the extracts were performed by agar well diffusion methods and determined by measuring the zones of inhibition with transparent scale. The result of phytochemical screening revealed the presence of Tannin, Saponins, Alkaloids, Flavonoids and Phenol on ethanol extract while the Aqueous leaf extract shows the presence of Saponins, Tannin, Flavonoids, Alkaloids but Phenol was absent. The result of antimicrobial activity showed that E. coli has higher sensitivity of (10.0mm) zone of inhibition followed by Staphylococcus aureus (9.2mm) and Bacillus species has the least sensitivity of (7.5mm) amongst the organism ascertained with zone of inhibition on the ethanol extract and in aqueous extract the E. coli has the higher sensitivity of (8.2mm) zone of inhibition while Staphylococcus aureus and Bacillus species has not detected zone of sensitivity amongst the organism ascertained. The study revealed some of acclaimed medicinal importance of Pawpaw (Carica papaya) leaves and can be made available commercially for its medicinal value.

Keywords:- *Carica papaya, Antimicrobial, Phytochemical Screening, Zone of Inhibition*

I. INTRODUCTION

Pawpaw (Carica papaya) belongs to the family Caricaceae, which is a fast growing erect, usually unbranched tree or shrub 7-8m with copious latex, land trunk of about 20cm in diameter. Different parts of this plant which include the leaves, fruit seed, latex and root are used for medicinal purpose. Carica papaya plant produces natural compounds in leaf bark and twig tissue which have high anti-tumour and pesticidal properties (Ayandele and Bukola, 2015).

Medicinal plant extracts are endowed with several biologically active compounds which possess potent antimicrobial activity and could be used to effectively replace synthetic chemicals. Carica papaya can be observed as a potential source of antimicrobials and can be made available commercially for its medicinal value. Snigdha, S. et, al. (2019), Pawpaw is commonly known for its food and nutritional values throughout the world. The medicinal properties of papaya fruit and other parts of the plant are also well known in traditional system of medicine. Each part of papaya tree possesses economic value when it is grown on a commercial scale. Even though the active components are normally extracted from all parts of the plant, the concentration of these components vary from structure to structure.

However, antimicrobial is an agent either kills microorganisms or inhibits their growth but causes little or no damage to the host. Antimicrobial therapy refers to the use of antimicrobial medicines to treat infection while antimicrobial prophylaxis refers to the use of antimicrobial medicines to prevent infection (Amyes 1996). The leaves of papaya plants contain chemical compounds of karpain, substance which kills microorganisms that often interfere with the digestive function. Pawpaw (Carica papaya) can be observed as a potential source of antimicrobials and can be made available commercially for its medicinal value (Snigdha, S. et, al. 2019). The primary objectives of this study is to determine the antimicrobial activities and phytochemical analysis of Papaya leaves extract.

❖ *Statement of the Problem*

Majority of plant are medicinally important and it is necessary to investigate more on these plant through phytochemical screening with a view to revealing their medicinal value, based on the active compound that can be extracted from its part such as leaves, stems, barks, roots, bulks, rhizomes, woods, flowers, fruits or seeds for therapeutics purposes and pawpaw was found to available but unexploited for medical purposes on this region.

❖ *Significance of the Study*

Most plant are believed to possess medicinal important due to some phytochemicals present in them. The bioactive compounds from plants are important source of new drugs that are likely to lead to new and better medical treatment. The result

of this study will reveal some of acclaimed medicinal importance of Pawpaw (Carica papaya) leave.

❖ *Limitation*

This study primarily focused on the antimicrobial activities and phytochemical analysis of papaya leaf to determine some active metabolites on some pathogen and restricted to E. Coli, Bacillus Species and Staphylococcus Aureus.

II. MATERIALS AND METHOD

❖ *Materials Include*

Incubator, measuring cylinder, Petri dish, Autoclave, Wire loop, Cotton wool, Aluminum foil paper, distilled water, Hands gloves, Bunsen burner, Sprit, Pipette, Beaker, masking tape and Weighing balance, Nutrient agar, conical flask and culture media.

➢ *Reagent Used*

Gram reagent, Ethanol, Aqueous, and *Pawpaw leaves* extract.

➢ *Sample Collection*

Fresh leaves of Pawpaw were collected from the Department Agricultural Technology, Ramat polytechnic Maiduguri, Borno State.

❖ *Method*

➢ *Preparation of plant materials:*

Freshly collected leaves of Pawpaw were washed with distilled water and dried under the shade at normal room temperature for 10 days. After drying, the plant material was pounded using mortar and pestle into smaller particles and then blended to powder. 50grams of the powdered samples were stored in airtight containers and kept under normal room temperature for subsequent analysis.

➢ *Preparation of Ethanol extracts*

50grams of the dried powered samples were weighed and soaked in 250ml of ethanol contained in two different 250ml flasks and stirred for five minutes. The flask was covered with foil and then allowed to stand for 24 hours. After 24 hours, the suspensions were shaken well and filtered using filter paper. Meanwhile the same procedure used for Aqueous extract.

phytochemical analysis of Pawpaw leaves extract was carried out using the standard protocol method (Oti and Olivia 2017).

➢ *Preparation of Mayer's reagent*

1.3g of mercuric chloride and 5.0g of potassium iodide were dissolved in distilled water in a 100ml volumentary flask and the solution was made up to 100ml.

➢ *Test for alkaloids*

- *Mayer's test*

2ml of the extract was pipette into a test tube and to the extract was added 1ml of 1% HCl, it was heated for twenty minutes in a water bath and then allowed to cool. After cooling 0.5ml of Mayer's reagent was added. The appearance of creamy white color indicates the presence of alkaloid.

➢ *Test for flavonoid*

- *Lead acetate test*

2mls of the extract was pipette into a test tube to it was added 10mls of distilled water, and then to it was added 5mls of distilled water, then 1ml of 10% NaOH. The change in the color of the mixture to yellowish indicates positive.

➢ *Test for saponins*

- *Foam test*

2ml of the extract was pipette into a test tube, to it was added 2ml of distilled water, it was shaking vigorously, and a formation of the foam indicates the presence of saponins.

➢ *Test for tannins*

- *Braymer's test*

2mls of the extract was pipette into a test tube and was boiled in a water bath for twenty minutes, after cooling 3 drops of 1% ferric chloride was added to it; a bluish precipitate indicates the presence of tannin.

➢ *Test of phenols*

- *Ferric chloride test*

2ml of extract add 1ml of 5 % ferric chloride solution, formation of blue-black color indicates the presence of phenols.

➢ *The micro-organism used*

Antimicrobial activity of pawpaw leaves (*Carica papaya*) on *E. coli, Bacillus species and Staphylococcus aureus.* The pure culture was obtained locally from Microbiology Unit University of Maiduguri Teaching Hospital (U.M.T.H) Borno State. The isolates were grown on blood agar on which is in urine sample were used the *E. coli, Bacillus species and Staphylococcus aureus* were obtained, the bacteria were maintained on nutrient agar.

➢ *Preparation of media*

5mm of nutrient agar powder was weight and dissolved in a conical flask containing 500ml of distilled water and sterilized by autoclaving at 121°C for 15 min the media was allowed to cool at 45°C, when poured into astride Petri dish and allowed to solidify at room temperature.

ISSN No:-2456-2165

➢ *Sensitivity testing of pawpaw leaves (Carica papaya)*

Pawpaw leaves (Carica papaya) powder and the ethanolic extract. Were used to test the Sensitivity or resistance of E. Coli, Bacillus Species and Staphylococcus Aureus about 5mm of the powder and10mg the extract was used onto earth newly Inoculated plate of E. coli, Bacillus species and Staphylococcus aureus the plate were then incubated at 37°C for 24 hours the presence of the ability of the powder or extract to inhibit the growth of the organism.

III. RESULTS

Table 1: Phytochemical screening of Pawpaw (Carica papaya) leaves on Ethanol and Aqueous Extract.

TEST	ETHANOL EXTRACT	AQUEOUS EXTRACT
Flavonoids	+ve	+ve
Tannins	+ve	+ve
Saponins	+ve	+ve
Alkaloids	+ve	+ve
Phenol	+ve	-ve

Key: +ve= positive, -ve= negative

The result of table 1**,** showed that Tannin, Saponins, Alkaloids, Flavonoids and Phenol are all positive in ethanol extract. While in aqueous extract Tannin, Saponins, Alkaloids, Flavonoids are positive while Phenol is negative as indicated.

Table 2: zones of inhibition for the effect of ethanol leaf extract of *Carica papaya leaves* on clinical isolated Organisms

Organisms	Activity	Zones of inhibition(mm)
E. coli	+	10.0mm
Bacillus species	+	7.5mm
Staphylococcus aureus	+	9.2mm

Key: +ve= Positive, -ve= Negative, ND= Not Detected

The result on table 2 showed that *E. coli* is highly sensitivity with (10.0mm) zone of inhibition followed by *Staphylococcus aureus* (9.2mm) and *Bacillus species* has the least sensitivity amongst the organism ascertained with (7.5mm) zone of inhibition on the ethanol extract.

Table 3: zones of inhibition for the effect of Aqueous leaf extract of *Carica papaya leaves* on clinical isolates Organism

Organisms	Activity	Zones of inhibition (mm)
E. coli	+	8.2mm
Bacillus species	-	ND
Staphylococcus aureus	-	ND

Key: +ve= Positive, -ve= Negative, ND= Not Detected

The result of table 3 showed that *E. coli* is highly sensitivity with (8.2mm) zone of inhibition while *Staphylococcus aureus* and *Bacillus species* has not detected sensitivity amongst the organism ascertained on the aqueous extract.

IV. DISCUSSION

The result of present study showed that, the phytochemical analysis of the pawpaw leave contains saponin, Tannin, Alkaloids, phenol and Flavonoids on ethanol extract and on aqueous extract phenol was absent. This finding can be similar to the work of Sikanda et al. (2013) who also reported similar finding and also stated the effect of these phytochemical as a good antimicrobial agent on different test organism. All the phytochemicals compounds that were detected are known to have industrial and medicinal importance. For example, saponin is useful in medicine as antioxidant, anticancer and for treatment of hypercholesterolemia and hyper glycaemia Tannin exhibits antiviral, antibacterial, antitumor activities. It was also reported that certain tannins are able to inhibit HIV replication selectivity (Drabble and Nierenstein, 2000). Flavonoid has inherent ability to modify the body's reaction to allergies, virus and carcinogens (Erdman *et al.*, 2007). They show anti allergic, anti-inflammatory, antimicrobial and anticancer activities. Alkaloid has the potency to correct serious disorders such as heart failure, cancer and blood pressure. Alkaloids were also detected in the leaves of *Carica papaya*, alkaloids are toxic against cells of foreign organisms. These activities have been widely studied for their potential use in the elimination and reduction of human cancer cell lines. Just *et al*. (1998) reported the inhibitory effect of saponins on inflamed cell and this has supported the usefulness of this plant in managing inflammation.

The result of antimicrobial activity showed that E. coli is highly sensitivity with (10.0mm) zone of inhibition followed by Staphylococcus aureus (9.2mm) and *Bacillus species* has the least sensitivity amongst the organism ascertained with (7.5mm) zone of inhibition on the ethanol extract and the result also showed that the E. coli is highly sensitivity with (8.2mm) zone of inhibition on the aqueous extract while Staphylococcus aureus and *Bacillus species* has not detected sensitivity amongst the organism ascertained in the aqueous extract.

The present study showed that the ethanol extract is very effective than aqueous extract on both phytochemical and antimicrobial activity. The present study is similar to Marshall, et., al. (2015). the antimicrobial activity of the plant showed that the plant had a broad spectrum of activity against the test isolate with varying zones of inhibitions ranging from 16.0mm, 18.4, 15.0, 16.5, and 13.0mm for S. aureus, E. coli, S. typhi, P. aeroginosa, and C. albican respectively at 100% concentration of the ethanolic extract. 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 may be because different solvents have different polarities, hence different degrees of solubility for the various phytoconstituents (Uwah et al., 2013; Dougahri, 2008).

V. CONCLUSION

The results of the present study showed the presence of the following bioactive compounds, flavonoid, Saponins, tannins, phenol and alkaloids which make it to have valuable antibacterial activities. From the result obtained, it could be inference that the potency of active ingredient from leaves is very effective against the selected test organisms i.e E. coli, Bacillus species and Staphylococcus aureus. The ethanol extracts of *Carica papaya leave* have sensitivity activity on E. coli and Staphylococcus aureus and Bacillus spps. This finding justifies the medicinal uses of these plant parts for therapeutic purposes.

REFERENCES

[1]. Amyes, S., (1996). Antimicrobial Chemotherapy: Pocketbook. CRC Press, p 25.
[2]. Aruljothi, S., Uma, C., Sivagurunathan, P., and Bhuvaneswari, M. (2014). Investigation on Antibacterial Activity of Carica Papaya Leaf Extracts against Wound Infection-Causing Bacteria International Journal of Research Studies in Biosciences (IJRSB) Volume 2, PP 8-12 ISSN 2349-0357.
[3]. Ayandele Abiodun Ayanfemi and Ayandele Oluwaseun Bukola (2015). Antibacterial Activity of Carica Papaya Leaves and Seeds Extracts On Some Bacteria and Them Phytochemical Characterization International Journal of Botany and Research (IJBR) ISSN (P): 2277-4815; ISSN (E): 2319-4456 Vol. 5, 15-22.
[4]. Bhushan Kaavimandan, (2016). Studies on biological efficacy of various leaf extracts of Carica papaya L., *ICGTETM*. 2(3): 510-516.
[5]. Doughari JH, El Mahmud AM, Manzara S (2008). Studies on the antibacterial activities of root extract of *Carica papaya* L. *Afri. J. Microbiol. Res.* Pp. 037-041.
[6]. Drabble, E., and Nierenstein, M. (2000). On the Role of Phenols, Tannic Acids, and Oxybenzoic Acids in Cork Formation. *Biochemical Journal* 2 (3): 96-102.
[7]. Erdman, J.W., Balentine, D. and Arab, L. 2007. Flavonoids and Heart Health: Proceedings of the ILSI North America Flavonoids Workshop, May 31-June 1, 2005, Washington DC. *J Nutri.*137 (3): 718S-737S.
[8]. J.O. Oti Wilberforce and Eze-Ilochi Nkechinyere Olivia, (2017). Phytochemical Screening and Antimicrobial Activity of Leaves Extracts of Mangifera indica and Carica papaya. *International Journal of Current Microbiology and Applied Sciences* ISSN: 2319-7706 Volume 6 pp. 3253-3259.
[9]. Just, M. J., Recio, M. C., Giner, R. M., Cueller, M. J, Manez, S., Bilia, A. R. and Rios, J. L. (1998). Anti-inflammatory activity of unusual lupine saponins from Bupleurum fruticescens. Plant Medicine. 64(2):404-407.
[10]. Ekaiko Marshall U, Chiwendu Stephen, Ukpabi Emmanuel O and Ezikpe Chizaram A, (2015). Antimicrobial screening and phytochemical analysis of Carica papaya Leaf extracts. Standard Research Journal of Microbiological Sciences Vol 2(1): (ISSN: 2409-062X). http://www.standresjournals.org/journals/SRJMS.
[11]. Nirosha. N and R. Mangalanayaki (2013). Antibacterial Activity of Leaves and Stem Extract of Carica papaya L. *International Journal of Advances in Pharmacy*, Biology and Chemistry Vol. 2(3), Issn: 2277 – 4688.
[12]. Sikandar KS, Tasveer ZB, Kanwal N, Syed AG, Shahama UK (2013). Qualitative phytochemical screening and antifungal activity of *Carica papaya* leaf extract against human and plant pathogenic fungi. *Int. Res. J. Pharm.* 4 (7).
[13]. Snigdha, S. Ravish, M. Narayan, G. Manisha, N. Nilotpol, K. and Kishore, D. (2019). Phytochemical Analysis of Papaya Leaf Extract: Screening Test. *EC Dental Science 18.3*: 485-490.
[14]. Uwah AF, Otitoju O, Ndem JI, Peter AI (2013). Chemical composition and antimicrobial activities of adventitious root sap of Musanga cecropioides. Der Pharmacia Lettre. 5 (2):13-16.