

## Antibacterial effect of *Carica papaya* against *Salmonella typhi*, causative agent of Typhoid fever.

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**Abstract:** Investigation on the antibacterial effect of *Carica papaya* was carried out using the agar dilution method. The extract used was gotten from the plant using different diluents which are methanol, ethanol and aqueous hot water. However, the methanolic extracts of the plant showed the highest bactericidal effect on the test isolates at a low concentration of 4.5 mg/ml while that of ethanolic extract was 6.0mg/ml and hot aqueous extract is 7.5mg/ml therefore the Minimum Inhibitory Concentration (MIC) of all the extracts ranged from 4.5-7.5mg/ml.

**Keywords:** Antibacterial, *Carica papaya*, Typhoid fever.

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### I. Introduction

Salmonella belong to the family *Enterobacteriaceae*. The genus is composed of gram -ve bacilli that are facultative and flagellated (motile) [1]. *Salmonella* posses three major antigens the 'H' or flagella antigen phase 1 and 2, the 'O' or somatic antigen and the 'V' or capsular antigen. Each serotype is characterized by a particular combination of these antigens [1]. The disease produced by the different specie of *Salmonella* are collectively known as Salmonellosis. These diseases occur world wide and are most generally manifested as a self-limiting gastroenteritis which causes acute intestinal distress with sudden onset of headache, fever, abdominal pain, diarrhea, nausea and sometimes vomiting [2]. According to [3] in 1982, medicinal plant is any plant in which one or more of its organs contain substances that can be used for therapeutic purposes or which are precursors for the synthesis of useful drugs. Plants synthesizes a large variety of chemical substances which include phenolic compounds, alkaloids, steroids, hydroxychaviol, glycosides, saponnins, tannins and a host of other chemical compounds referred to as secondary metabolites. They continue to be important to people that do not have access to modern medicines and moreover, modern pharmaceuticals rely heavily on the same active principles be they natural or synthetic. The active principles differ from plant to plant due to their biodiversity, that is, to the plant's genetic coding and ability to produce them [4]. Most of the known medicinal plants have been reported to contain some active chemicals which are toxic and inhibitory to some microorganism causing diseases in human. Traditional medicine employs the use of medicinal plants for the treatment of diseases caused by microorganisms within an organized indigenous system. However, traditional herbalists employ two kinds of treatment which are the real treatment and the physiological treatment. The former is for patients who require no incantations or other ceremonies while the latter requires incantations or other ceremonies such as sacrifices before the medicine acts[6].

#### Justification of this study

Scientific Justification based on the fact that;

*Carica papaya* leave extract is used in treating malaria caused by the parasite *Plasmodium* specie.

It is one of the cheapest medicinal leaves that is easily found within the local environment.

It is the leave that can be gotten in every season of the year.

#### Therefore the objectives of this work are to determine:

1. The antibacterial activities of the leaves of *Carica papaya* using different means of extraction.
2. The bactericidal activities and the minimum inhibiting concentration (MIC) of each means of extraction.

### II. Materials And Methods

#### Sterilization of materials used

All glass wares were thoroughly washed in water containing detergent and rinsed with distilled water, they were air dried and sterilized in the oven at 160<sup>0</sup>c for one hour. Inoculating chamber and growth chamber were fumigated using formaldehyde and then irradiated on exposure to UV lamp for one hour. Laboratory

benches were cleaned with absolute alcohol while the inoculating loop was flamed to redness and allowed to cool prior to use.

#### **Preparation of agar**

Nutrient agar, nutrient broth, E (LABM) and sensitivity test agar were prepared according the manufacturer's instruction.

#### **Collection and identification the plant used**

The leaves of *Carica papaya* used were plucked in Osogbo, Osun State, Nigeria and they were identified and authenticated in the herbarium unit of the Department of Forestry and Wild Life in the Federal University of Technology, Akure.

#### **Processing of plant materials**

The freshly collected leaves were air dried completely. The air dried leaves were macerated and ground into powdery form using a sterile electric blender and stored in a clean airtight container until further use.

#### **Extraction of plant materials**

##### **Alcoholic extraction**

40 grams of the grounded leaves was accurately weighed twice and added to 200mls of ethanol and methanol in separate conical flasks and allowed to stay for 5 days after which the extract was filtered through Whatman filter paper no 1. The filtrate was air dried and the residue was scraped and kept in clean bottles.

##### **Reconstitution of the extracts**

2 g of the extract was weighed and dissolved in 20ml of 50% Dimethyl sulfoxide (DMSO) to make a stock concentration of 100mg/ml from which the various concentrations used were calculated.

##### **Test organism and source**

Isolates of *Salmonella typhi* from five different samples were used. They were collected from Nigeria Institute of Medical research Yaba (NIMIR), where they were maintained on slants.

##### **Bacteriological assay**

The test organisms were removed from stock, streaked on freshly prepared Nutrient agar plates and incubated at 37<sup>o</sup>c for 48 hours to resuscitate the organisms.

##### **Sterilization of the extracts**

This was effected by preparing stock solution of all the extract at 100mg/ml concentration. The reconstitution was followed by sterilization at 121<sup>o</sup>C for in an autoclave for 15 minutes Membrane filter (disposable) of 0.2mm milipore was also used.

##### **Determination of Antibacterial Efficacy (MIC)**

A 2cm streak of the standardized isolates were made radially on dried sensitivity test agar containing increasing concentrations (1.5-12.0(mg/ml) of the extracts .The bactericidal activities of the extracts were observed visually and recorded against each concentration of the extract used. Likewise the lowest concentration that gave no visible growth after 24 hours of incubation at 37<sup>o</sup>C was noted and recorded as the Minimum Inhibitory Concentration (MIC) of each extract. The petri -dishes without the extracts served as the control.

### **III. Results And Discussion**

#### **Results**

The results obtained showed that the extracts of the leaves of *Carica papaya* were active against the test isolates of *Salmonella typhi*, the Causative agent of typhoid fever.

The methanolic extracts of the plant showed the highest bactericidal effect on the test isolates at low concentration of 4.5mg/ml (table 1). Also the ethanolic extracts of the plant leaves had a bactericidal effect on the test isolates at concentration of 6.0m/ml (table 2) whereas the hot aqueous solution showed susceptibility at 9.0mg/ml(table 3). None of the test isolates were susceptible at concentrations of 1.5-3.0mg/ml. 100% of all the isolates were susceptible at the remaining higher concentrations. Table 4 shows the Minimum Inhibitory Concentrations (MIC) of all the extracts against the test isolates. It was observed that the methanolic extracts exhibited the lowest range of concentrations (4.5-6.0mg/ml) and the hot aqueous extracts showed the highest range of concentrations 9.0-12.5mg/ml to inhibit the growth of *Salmonella typhi*.

**Table 1: The Bactericidal Activities of *Carica papaya* Using Methanol Plant Extract**

Concentration (mg/ml)	1.5	3.0	4.5	6.0	7.5	9.0	10.5	12.0
TEST ISOLATES	1.5	3.0	4.5	6.0	7.5	9.0	10.5	12.0
ST-1	-	-	-	+	+	+	+	+
ST-2	-	-	-	+	+	+	+	+
ST-3	-	-	-	+	+	+	+	+
ST-4	-	-	+	+	+	+	+	+
ST-5	-	-	+	+	+	+	+	+

**Key:**

ST- Isolates of *Salmonella typhi*

-: No inhibition

+: Inhibition

**Table 2: Ethanolic Extracted *Carica papaya* Plant Extract Concentration (mg/ml)**

TEST ISOLATES	1.5	3.0	4.5	6.0	7.5	9.0	10.5	12.0
ST-1	-	-	-	-	+	+	+	+
ST-2	-	-	-	-	+	+	+	+
ST-3	-	-	-	+	+	+	+	+
ST-4	-	-	-	-	+	+	+	+
ST-5	-	-	-	-	-	+	+	+

**Table 3: Hot Aqueous Extracted *Carica papaya* Plant Extract concentration (mg/ml)**

TEST ISOLATES	1.5	3.0	4.5	6.0	7.5	9.0	10.5	12.0
ST-1	-	-	-	-	-	-	-	+
ST-2	-	-	-	-	-	-	+	+
ST-3	-	-	-	-	-	+	+	+
ST-4	-	-	-	-	-	-	+	+
ST-5	-	-	-	-	-	-	-	+

**Table 4: The Minimum Inhibitory Concentrations (MIC) of the Extracts Against the Test Isolates (mg/ml).**

TEST ORGANISM	METHANOLIC EXTRACT	ETHANOLIC EXTRACT	AQUEOUS EXTRACT
ST-1	6.0	7.5	10.5
ST-2	4.5	7.5	9.0
ST-3	6.0	6.0	10.5
ST-4	4.5	7.5	12.5
ST-5	4.5	9.0	9.0

#### IV. Discussion

According to [4] plants were the sole source of active principles capable of curing man's ailments before the development of chemistry and synthesis of organic compounds in the 19<sup>th</sup> century. From the results it was observed that the crude extracts of the plant contain active principles that inhibited the growth of the test organism (tables 1, 2, 3 & 4 ) this is in accordance with [7] who reported the antimicrobial activities of some medicinal plants found in the south west region of Nigeria to be active against the six bacteria tested. [8] worked on the anti- diarrhoeal activities leaf extract of *Ocimum gratissum* using disc diffusion and tube dilution methods and found out that it was active *Salmonella typhi*, *Aeromonas sorbis*, *Escherichia coli*, *Plesiomonas shigelloides* and *Shigella dysenteriae*. [9] reported that organic (ethyl acetate) extracts of roots and stem of *Byrsonimacrassifolia*(L. H. B. K was found to be active against *Klebsiella flexneri*, *Pseudomonas Aeruginosa*, *Salmonella typhi*, *Staphylococcus epididimis* and *Micrococcus luteus*. [10] reported that the ethanolic extracts of *Mitracarpus villosus* leaves was found to produce antifungal activities against *Trichophyton rubrum*, *Candida albicans* and *Mirosporium gypseum* but aqueous extract and the glycerol vehicle control did not inhibit any of the fungi tested.

#### V. Conclusion

The fact that crude extract of this plant exhibited an inhibitory effect on *Salmonella typhi*, if properly researched and processed hygienically, this plant could serves as a cheap source of antimicrobial compound for the treatment of typhoid fever.

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