

# Discovery

# In vitro antimicrobial activity of medicinal plants Ashwagandha (Withania Somnifera) and Papaya (Carica Papaya) with commercial antibiotics

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## **ABSTRACT**

The present study aimed to examine a comparative analysis of the antimicrobial efficiency of methanol root and leaf extracts Ashwagandha (Withania Somnifera) and Papaya (Carica Papaya) with commercial antibiotics against the microbial strains was assessed by agar well diffusion method. The methanolic extracts of W. somnifera and Carica papaya found to show potent antimicrobial activity against the pathogenic strains as compared to antibiotic discs. Methanol root extracts of all samples showed an effective antibacterial activity might be due to presence of numerous bioactive compounds. The extracts of Withania somnifera and Carica Papaya might be exploited as natural drug for the treatment of several infectious diseases.

Keywords: Withania somnifera, Carica Papaya, Methanolic Extract, Antimicrobial activity.

Abbreviation: Dengue Shock Syndrome (DSS).



#### 1. INTRODUCTION

# 1.1. Withania somnifera (Ashwagandha)

The increasing failure of chemotherapeutics and antibiotic resistance exhibited by microbial pathogens has led to the screening of medicinal plants for their potential antimicrobial activity (Singh and Sharma, 2010). Withania somnifera (L) Dunal, also known as Ashwagandha, Indian ginseng, and Winter Cherry is a small medium under shrub belonging to the Solanaceae family. The plant has been found useful in the treatment of burns, wounds, and dermatological disorders, and gastrointestinal diseases, dysfunctions of the respiratory system, asthma, bronchitis, cancer and geriatric problems (Betsy and Chawla, 2012). The major biochemical constituents of Ashwagandha root are steroidal alkaloids and steroidal lactones is a class of constituents called withanolides (Verma Sitansu, Kumar Ajay, 2011). Withaferin A and Withanolide D are the two main Withanolides that contribute to most of the biological activity of Withania somnifera. The total alkaloid content of Indian root varies between 0.13% to 0.31%. Withania somnifera has been used as an anti-oxidant, adaptogen, aphrodisiac, liver tonic, anti-inflammatory agent, astringent and more recently as antibacterial agent (Pujari and Gandhi, 2012).

# 1.2. Carica Papaya (Papaya)

Carica papaya L. (Caricaceae) is valuable as food and is also used in traditional medicine. Papaya fruits contain components that can increase the total antioxidant power in blood and reduce the lipid peroxidation level (Octoo Augustine, Nwokolo, 2012; Osanaiye Tkin, 2005). These components include  $\alpha$ -tocopherol, ascorbic acid, beta carotene, flavonoids, vitamin B1, and niacin. Carica papaya (pawpaw) is one of the fruits commonly used as food and medicine in India (Krishna and Paridhavi, 2008). It is eaten as fresh fruit or processed into deserts. The unripe matured pawpaw fruit is used for the production of papain by the making of incisions on the back of the fruits to get the latex for the papain production. Papaya is a powerhouse of nutrients and is available throughout the year. It is a rich source of threes powerful antioxidant vitamin C, vitamin A and vitamin E; the minerals, magnesium and potassium; the B vitamin pentothenic acid and folate and fiber. Ayurvedic literature reveals that papaya (Carica papaya) leaf extract has haemostatic and other medicinal properties, the beneficial effects of papaya leaf extract in Asian patients who have dengue hemorrhagic fever and Dengue Shock Syndrome (DSS). This pilot study was undertaken to find out whether carica papaya leaf extract had any effect on increasing the lowered platelet count (Pandey and Kumar, 2011). Carica papaya leaves have been use traditionally to treat indigestion, as a vermifuge. Carica papaya leaves have also been shown to possess anti-tumor and immunomodulatory effects.

#### 2. EXPERIMENTAL PROCEDURE

#### 2.1. Selection of Plant

The plants Papaya (*Carica papaya*) and Ashwagandha (*Withania Somnifera*) were selected for study. Fresh leaves and roots of Papaya and Ashwagandha were collected from S. D. College of engineering & technology Garden in Muzaffarnagar District, Uttar Pradesh.

#### 2.1.1. Leaf extracts

Fresh leaves of papaya and Ashwagandha were washed and macerated with distilled water in mortar and pestle. Crude leaf extracts were obtained by filtering the macerated mixture with muslin cloth. The obtained extracts were subjected to rotary evaporator and subsequently concentrated under reduced pressure (in vacuum at 40°C) and evaporated to dryness and stored at 4°C in air tight bottle.

#### 2.1.2. Root extract

Fresh papaya and Ashwagandha root were washed thoroughly and crushed with distilled water in mortar and pestle and crushed mixture was filtered through muslin cloth. The obtained root extract was subjected to rotary evaporator and subsequently concentrated under reduced pressure (in vacuum at 40°C) and evaporated to dryness and stored at 4°C in air tight bottle.

# 2.1.3. Microorganism

The pathogenic strain of unknown environment bacteria isolated from Air, soil and tap water.



# 2.2. Antimicrobial activity assay

# 2.2.1. Agar well diffusion method

Screening of leaf extract of *Withania Somnifera* and *Carica Papaya* tested using agar well diffusion method of microbial strains. Nutrient agar medium was used to assess the antibacterial activity of the prepared extracts. 20 ml of the inoculated nutrient agar were distributed into sterile Petri dishes. The agar was left to set and in each of these plates, 5 mm in diameter, were cut using a sterile cork borer No. 4 and the agar discs were removed. Alternate cups were filled with 20  $\mu$ L of each extracts using micro titerpipette and allowed to diffuse at room temperature for two hours. The antibacterial were evaluated at antibiotic susceptibility testing using the disc diffusion method. For the agar diffusion method, wells were filled with essential antibiotics. The plates were then incubated in the upright position at 37°C for 18 hours. The diameters of the growth inhibition zones were measured at 24 hours of incubation averaged and the mean values were tabulated.

# 2.2.2. Statistical analysis of data

The data obtained were subjected to statistical analysis to determine the significance extraction antimicrobial activity of Carica papaya and Withania somnifera.

**Table 1**Zone of inhibition of bacteria against root and leaf extracts of Papaya and Ashwagandha

Methanolic Extract	Soil bacteria (in mm)	Water bacteria ( in mm)	Air bacteria (in mm)
Papaya leaf	26	30	26
Ashwagandha leaf	26	25	25
Papaya root	22	23	24
Ashwagandha root	15	15	26

**Table 2**Zone of inhibition of bacteria against different antibiotics

Essential antibiotics	Soil bacterial (in mm)	Water bacteria (in mm)	Air bacteria (in mm)
Ampicillin	21	28	24
Ofloxacin	35	15	16
Tetracycline	24	29	33
Chloramphenicol	25	26	20

**Table 3**Comparison between zone of inhibition of bacteria against root and leaf extracts of Papaya and Ashwagandha and different antibiotics

Essential antibiotics	Soil bacteria	Water bacteria	Air bacteria
(in mm)	(in mm)	(in mm)	(in mm)
Papaya leaf	26	30	26
Ashwagandha leaf	26	25	25
Papaya root	22	23	24
Ashwagandha root	15	15	26
Ampicillin	21	28	24
Ofloxacin	35	15	10
Tetracycline	34	29	33
Chloramphenicol	25	26	20



**Graph 1**Zone of inhibition bacteria against extract of Papaya and Ashwagandha

#### 3. RESULTS

Refer Tables 1-3 & Graph 1-3

#### 4. DISCUSSION

Plants contain many biologically active compounds which have potential for development as medicinal agents. Many of the existing synthetic drugs cause various side effects. Hence development plant based compounds could useful in meeting this demand for newer drugs with minimal side effects

W. somnifera (Solanaceae), also known as Ashwagandha, is one of the most valuable plants in the traditional Indian systems of medicine. This plant is used in more than 100 formulations in Ayurveda. The ethno pharmacological properties of the plant include adaptogenic, anti-sedative and anti-convulsion activities, and the plant is used to treat various neurological disorders, geriatric debilities, arthritis, stress and behavior-related problems (Singh and Kumar, 2012). Papaya fruit is a rich source of nutrients such as provitaminA, carotenoids, vitamin C, vitamin B complex, dietary minerals and dietary fiber. Papaya skin, pulp and seeds also contain a variety of phytochemical, including natural phenols (Hamman O Wilson and Musa A Sunday, 2011). Daniel one is a phytoalexin found in the papaya fruit and showed high antifungal activity against Colletotrichumglo esporioides, pathogenic fungus of Papaya. Papaya contains many biologically active compounds. Two important compounds are Papain and Chymopapain, which aid indigestion. Papain also used to treat arthritis (Doughari and Elmahmood, 2007).

#### 5. CONCLUSION

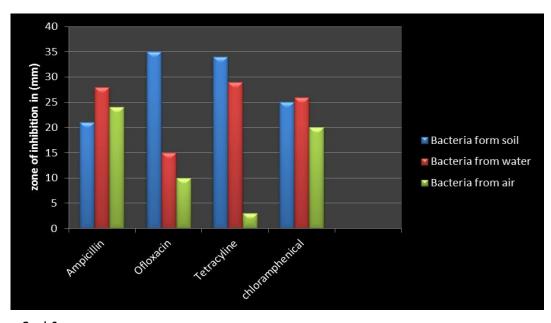
The study was conducted to evaluate the antibacterial activity of Papaya and Ashwagandha. In this study, the results obtained showed that the extract of Ashwagandha leaf inhibited the growth of soil, air, and water culture while root shows less antimicrobial activity. This study is a preliminary evaluation of antimicrobial activity of *Carica papaya* and *W. somnifera* (Jeyanthi and Subramanian, 2013). We used both the Papaya and Ashwagandha for the extraction of extracts from leaves and roots of plants. In case of Papaya, the root and leaf extract show high antimicrobial activity against soil, water, air culture. The antibacterial activity was expressed at varying degrees with the activity being both strain and dose dependent. Many previous works also concluded that papaya contains the effective active constituents responsible for eliminating the bacterial pathogens.

Finally, it can be concluded that the active chemical compounds present in *Carica papaya* and *W. somnifera* should certainly find place in treatment of various bacterial infections. The results from the present study are very encouraging and indicate this herb should be studied more extensively to explore its potential in the treatment of infectious diseases as well. Plant based antimicrobial compounds have enormous therapeutical potential as they can serve the purpose without any side effects that are often associated with synthetic antimicrobials (Kumar Vivek and Sharma Shivesh, 2009).

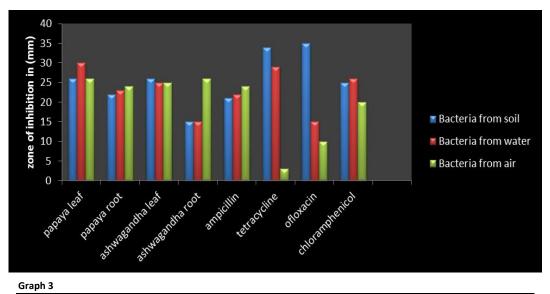
The results show that the methanol extract of Papaya showed more inhibitory effect than the other plant extracts. This tends to show that the active ingredients of the plant parts are better extracted with methanol than other solvents. Also antimicrobial effects of plant extracts have been demonstrated against air, soil, water. These might be due to presence of triterpenoids, phenolic compounds, carotenoids, steroids, valavinoids, ketones and tetratriterpenoids (Singaria and Kumar, 2012). Plant extracts posses a broad spectrum of activity against a panel bacteria responsible for the most common bacterial diseases. These extracts activities open the possibility of finding new clinically effective antibacterial compounds.

Further research is necessary to determine the identity of the antibacterial compounds from within these plants and also to determine their full spectrum of efficacy. However the present study of in vitro antibacterial evaluation of some plants forms a primary platform for further phytochemical and pharmacological studies to discover new antibiotic drugs (Velu and Baskaran, 2011).





Zone of inhibition bacteria against different antibiotics



Zone of inhibition b/w herbal antibiotics and different antibiotics

# **SUMMARY OF RESEARCH**

In this connection, the study was conducted to evaluate the antibacterial activity of Papaya and Ashwagandha. In this study, the results obtained showed that the extract of Ashwagandha leaf inhibited the growth of soil, air, and water culture while root shows less antimicrobial activity. This study is a preliminary evaluation of antimicrobial activity of *Carica papaya* and *W. somnifera*. It indicates that *Carica papaya* and *W. somnifera* have the potential to generate novel metabolites. The plant extracts demonstrated antibacterial activity could result in the discovery of novel anti-bacterial agents. Besides, the same way, also be used for self medication in domestic settings. All over the world scientific research is getting momentum to evaluate the pharmacological activities, side effects and medicinal uses of *Carica papaya* and *W. somnifera* against different diseases.

#### **FUTURE ISSUE**

The ultimate purpose of endophytes research is to find new antibiotics or pesticides, thus the following aspects should be intensively studied:

- INLOLAINOIT
- 1.To find better bioactive antimicrobial substances without any side effect to human, plant and environment.
- 2.To optimize conditions of endophyte fermentation that has been found to show bioactivity in order to enhance the yield of active substances synthesized by endophyte.
- 3.To search for the regulatory gene in synthesis path of antimicrobial compound, and use genetic engineering technology to increase the production of antibacterial substances.

#### **DISCLOSURE STATEMENT**

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