

**ANTIMICROBIAL ACTIVITY AND PHYTOCHEMICAL ANALYSIS OF  
*NELUMBO NUCIFERA* LEAVES**

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**ABSTRACT**

Medicinal plants play an important role in the discovery of novel drugs used in modern medicine. The medicinal plant of *Nelumbo nucifera* (lotus) possesses wide range of medicinal properties which were confirmed through literature reviews. But there are no reports on *Nelumbo nucifera* leaves for anti microbial studies. The objective of present study is to determine whether *Nelumbo nucifera* leaf have any antimicrobial proprieties. The leaves of *Nelumbo nucifera* is extracted using different solvents such as ethanol, methanol and water by various extraction methods. The presence of chemical constituents in the extracts was studied by preliminary phytochemical analysis. The antimicrobial susceptibility studies conducted against gram (-) bacteria such as *E. coli*, *P. aeruginosa*, *K. pneumonia* and gram (+) bacteria such as *Staphylococcus aureus*. Among the tested extracts, ethanolic extract shows good anti microbial activity. However further studies need to isolate the active constituents from the ethanolic extract and to study the antimicrobial activity in cellular level.

**Keywords:** *Nelumbo Nucifera*, *E. coli*, *S. aureus*, *P. aeruginosa*, *K. Pneumonia*

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## **INTRODUCTION:**

Many common plants seen in the Kitchen gardens or in the compound or in the forests are used by the tribal as medicines. The objectives of the present investigation are to record the anti-microbial activity of the medicinal plant *Nelumbo Nucifera* (family - *Nelumbonaceae*).<sup>[1]</sup> Among the herbal medicine, lotus leave is one of the plants which have some medicinal properties in it. The lotus type which used in this research is known as sacred lotus (*N.nucifera*), very native to Asian, it is commonly cultivated and also used in Chinese medicine. The aim of this research is to determine whether the extract of the lotus leaf have any anti-microbial properties. There are many ways to treat or heal the disease. One of the safe methods is by treating with herbal medicine, which are derived from plants and are normally known free from side effect, toxicity effect and are very economical. Most of the people are not aware of such information. There is a belief that plants have compounds such as glycosides, carbohydrates, alkaloids, tannins, saponins, and flavonoids in them. Therefore to prove the presence of these compounds in the *N. nucifera* leaves certain scientific tests will be carried out on the plant, to know whether the

constituent are really present in the leaves of the plant<sup>[2]</sup>.

This research also helps to discover new form of medication for the upcoming severe diseases such as Microbial infection such as tuberculosis, diabetics, hypertension and inflammatory disorders and others. The active ingredients from the lotus leaf is more natural and less chemical so it consist less toxicity, side effects and safe compared to synthetic drugs. This new discovery of lotus leaf product will create awareness regarding the advantage of the lotusleaf usage. This medication well tolerated by patients, with fewer unintended consequences than pharmaceutical drugs because it less toxic and side effects.

## **MATERIALS AND METHODS:**

### **Collection and drying of plant materials:**

Fresh plant materials (leaves) were collected from Taman Tasik Permaisuri, Kuala Lumpur, Malaysia. The leaves of *Nelumbo Nucifera* which floats on water were washed to remove soil debris and cut into small pieces dried for four weeks at room temperature (25<sup>0</sup>C to 30<sup>0</sup>C).

### **Micronization and storage:**

The dried leaves of *Nelumbo Nucifera* were further reduced into small pieces and then

pulverized into fibrous powder using clean electric blender. The dried plant materials should be placed in plastic containers or tightly covered bottles; brown colored bottles are preferred as they minimize deterioration due to sunlight<sup>[3]</sup>.

#### **Preparation of plant extracts:**

Ethanol extraction of the air-dried and powdered leaves (100g) of plants material was carried out by suspending 100 grams of the powder with *nelumbo nucifera* in around 500ml of 95% ethanol and extraction was allowed to stand for 72 hours at 27°C±1°C. The extracts were filtered through Watman filter paper No.1 extracts were evaporated to paste at 45°C<sup>[4]</sup>. They were transferred into sterile bottles and kept in refrigerator until used.

#### **Antimicrobial susceptibility testing:**

Antibacterial activity was determined using agar diffusion method of Boakye-yiadon. Five millimetre (ml) discs containing 1000µg/ml and 1500µg/ml of extract were placed on cultured pathogenic bacteria on agar plates and incubated at 37°C. The plates were checked for bacterial growth after a minimum of 18 hours and occasionally till 24 hours<sup>[5]</sup>. The diameter of the zone of inhibition was then measured. Commercial disc of Chloramphenicol

(30µg) was used as positive control (standard) and experiment was done thrice for each extract.

#### **Determination of Minimum Inhibitory Concentration (MIC):**

MICs are considered as the “gold standard” for determining the susceptibility of the organisms to antimicrobials. MIC of antibiotics was evaluated (thrice) using standard microbroth dilution method against *Escherichia coli* (gram negative) and *Staphylococcus aureus* (gram positive) organisms<sup>[6,1]</sup>.

#### **Phytochemical analysis:**

Phytochemical analysis to screen the plants for the presence of alkaloids, glycosides, saponins, tannins, flavonoids and carbohydrates, was performed according to the literature [7].

#### **RESULTS:**

The antibacterial activity of the ethanolic extract of pink *Nelumbo nucifera* leaf extracts had shown (Table 1). Ethanolic extract at two different concentrations such as 1500 & 1000µg (D. Brindha et al.,) were performed antibacterial activity against four bacterial strains by the streaking method and the results are summarized in Table 2.

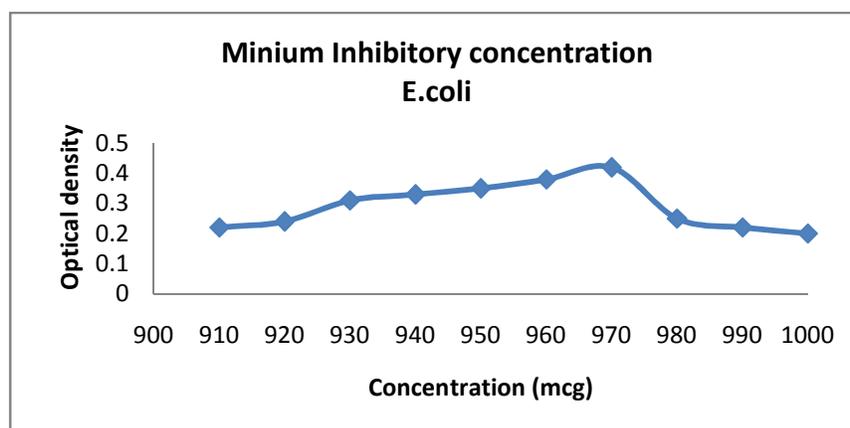
**Table-1:** Antibacterial activity pink *Nelumbo nucifera* leaf extracts

Microorganism	Solvent- Ethanol
Escherichia coli	+ve
Klebsiella pneumonia	+ve
Pseudomonas aeruginosa	+ ve
Staphylococcus aureus	+ ve

**Table-2:** Antibacterial activity pink *Nelumbo nucifera* leaf by streaking method

Microorganism	Zone of Inhibition (mm)		
	Pink <i>nelumbo nucifera</i>		Chloremphenicol (Control)
	1500µg/ml	1000µg/ml	30µg/ml
Escherichia coli	12	8	28
Klebsiella pneumonia	9	7	22
Pseudomonas aeruginosa	10	5	29
Staphylococcus aureus	12	8	27

**Figure 1** Minimum inhibitory concentration pink *Nelumbo nucifera* leaf against *Escherichia coli*

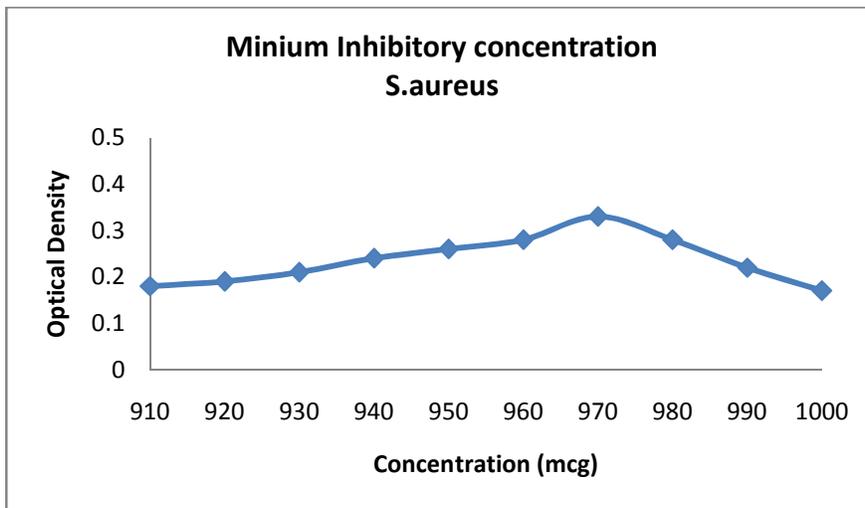


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The antibacterial activity of *Nelumbo nucifera* leaf extracts was found to be increased in dose dependent manner. The maximum zone of inhibition was exhibited by pink *Nelumbo nucifera* leaf against *Escherichia coli* (12mm), *Staphylococcus aureus*(12mm). The moderate zone of inhibition was found *Klebsiella pneumonia* (9mm) and *Pseudomonas aeruginosa* (10 mm). Gram-negative bacteria were more susceptible to the *Nelumbo nucifera* leaf extracts than gram-positive bacteria which contradict the previous reports that plant extracts are more active against gram-positive bacteria than gram-negative bacteria. However, the results revealed that the methanolic extract of pink *Nelumbo nucifera* leaf showed effective antibacterial

activity. This may be due to its variation in phytochemical constituents like flavonoids (nuciferine), alkaloids and tannins which were also reported by Bose *et al.*, [14] and these results were compared with the standard antibiotic chloramphenicol (30µg/ml). Antibacterial activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* showed that the plant can be used in the treatment of gastrointestinal infection and diarrhea in human. The minimum inhibitory concentration for *Nelumbo nucifera* leaf extract against *Escherichia coli* and *Staphylococcus aureus* was found to be 925µg and 965µg respectively (Figure 1 & 2). The lowest MIC was exhibited by against both the microorganisms.

**Figure-2:** MIC pink *Nelumbo nucifera* leaf against *Staphylococcus aureus*



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## CONCLUSION:

As a conclusion, through the research has carried that proven *Nelumbo Nucifera* leaf have antimicrobial activity since the extract of *Nelumbo Nucifera* inhibits the growth of microorganism from the species of *E.coli*, *S.aureus*, *P.aeruginosa* and *K.pneumoniae*. Various research such as obesity, diabetic, anti-tussive, expectorant and anti-spasmodic effects are made on pink lotus but our emerging idea induce us to do antimicrobial activity against *E.coli*,

*S.aureus*, *P.aeruginosa* and *K.pneumoniae*. Actually herbal extracts have less toxicity compared to synthetic drugs and also reduces the side effects. Through this research, we have found that pink lotus contains flavonoids (quercetin), alkaloids (nuciferine) carbohydrate, tannins and glycosides activity against microorganism. This plant can be used in the treatment of gastrointestinal infection and diarrhoea in human.

## ACKNOWLEDGEMENT:

The authors are thankful to the management of Masterskill University College of Health Sciences, Malaysia, for providing infrastructure facilities to conduct this work.

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