

## PHYSICOCHEMICAL, PHYTOCHEMICAL AND CHROMATOGRAPHIC INVESTIGATION OF NEEM (*AZADIRACHTA INDICA*) SEED OIL

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### ABSTRACT

The *Azadirachta indica* (Neem) plant is a very common folklore medicine used since ancient times for the treatment of various diseases. Neem plant is mainly known for its antimicrobial and insecticidal activities. The pharmacological actions of neem are due to different chemical constituents present in different parts of the plant. Neem plant is a rich source of different compounds like nimbin, nimbidin, quercetin and azadirachtin which are responsible for its pharmacological actions. Most chemical constituents of neem are present in leaves and seeds of the plant. Neem leaves are mainly used in many dentifrices and cosmetics for their antimicrobial activity and neem seed oil is mainly used in insecticidal preparations due to its potent insecticidal activity. This study comprises of physicochemical, phytochemical and chromatographic evaluation of neem seed oil.

**Keywords:** *Azadirachta indica*, Neem oil, Phytochemical evaluation and TLC.

### INTRODUCTION

Traditional medicines are gaining popularity in this era where the patients are more concerned about side effects of any medication. In Ayurvedic system of medicines plants are used as the source of most medicines. Each plant has specific Pharmacological activity based on the active principals present in that plant. Plants consist of number of different chemical constituents which gives the pharmacological action. While studying any plant drug the Phytochemistry of the drug should be studied to gain knowledge about the class of chemical compounds present in that plant.

The *Azadirachta indica* A. Juss plant belonging to Mahogany family Meliaceae is a subtropical plant indigenous to Indo-Pak subcontinent. Neem plant is used in various traditional medicines in Ayurvedic system of medicines for its different pharmacological activities like Antiviral, Antimicrobial, Antiulcer and Antibacterial. The pharmacological activities of neem plant are mainly due to presence of different terpenoids in different parts of the plant. The concentration of terpenoids in neem

plant varies from 0.3-0.8% in different plant parts. Seeds of neem plant consist of relatively high concentrations of terpenoids than other parts of the plant.

Neem oil is mainly isolated from neem seeds and is used for different purposes like in cosmetics, insecticides, pesticides, Antibacterial and Antiviral. Neem oil consists of a triterpenoid Azadirachtin which is a potent antimicrobial compound. Azadirachtin acts by inhibiting the formation of microtubules. Due to presence of various limonoids and triterpenoids, neem oil is commonly used in various insect repellent formulations and pesticides.

### MATERIALS AND METHODS

#### Collection

Neem seed oil was procured from local market of Nashik area

#### Sample preparation

Sample was prepared by dissolving neem oil in methanol and then it was used for physicochemical and phytochemical evaluation.

### Physico chemical evaluation of neem seed oil

Different Physicochemical parameters such as Viscosity, Relative density, Specific gravity, Refractive index, Iodine value, Saponification value and Acid value were studied for neem seed oil.

### Phytochemical evaluation

The Neem oil dissolved in methanol was used for phytochemical evaluation; tests for different classes of chemical constituents were performed.

### Chromatographic evaluation

Thin Layer chromatography was performed for neem oil the sample for TLC was prepared by dissolving oil in methanol. The mobile phase used for TLC was Chloroform: Acetone (8:2). Spot for Azadirachtin (Triterpenoid) was obtained at Rf 0.36, Visualized under 366nm.

### RESULT AND DISCUSSION

The Physicochemical evaluation of neem seed oil indicated that the oil is greenish brown in

colour with unpleasant odour and very bitter test. Neem seed oil is found to be soluble in polar organic solvents methanol, ethanol and ethyl acetate. The other physicochemical parameters such as viscosity, relative density, specific gravity, refractive index, iodine value, acid value and saponification value define physicochemical nature of the oil (Table no.1). The Phytochemical investigation of oil shows the presence of terpenoids, phenolic compounds, saponins, tannins and flavonoids (Table no. 2). Chromatographic investigation confirmed the presence of triterpenoid Azadirachtin in the oil (Table no 3).

### CONCLUSION

Physicochemical, Phytochemical and Chromatographic investigation of Neem seed oil was performed. The results indicate that the neem seed oil consists of variability of chemical constituents. The Thin Layer Chromatography of neem seed oil indicates the presence of triterpenoid Azadirachtin at Rf 0.36. This investigation is helpful in determining the nature of Phytoconstituents present in neem seed oil.

**Table 1: Physico chemical evaluation of neem oil**

S. No.	Parameters	Observations
1.	Color	Greenish brown
2.	Odour	Unpleasant, Characteristic
3.	Taste	Bitter
4.	Solubility	Soluble in methanol, ethanol and Ethyl acetate
5.	Viscosity	7 centistoke
6.	Relative density	0.88 g/cm <sup>3</sup>
7.	Specific gravity	0.928
8.	Refractive index	1.4638
9.	Iodine Value	70
10.	Saponification Value	190
11.	Acid Value	40

**Table 2: Phytochemical Evaluation**

Sr. No.	Class of Compounds	Tests	Observation	Result
1.	Carbohydrates	Molisch's test	Violet ring at junction of two liquids	Negative
		Felhings test	Brick red precipitate	Negative
2.	Alkaloids	Mayer's test	Precipitate	Negative
		Wagner's test	Reddish brown precipitate	Negative
3.	Glycosides	Borntragers test	Pink color to the ammonical layer	Negative
4.	Terpenoids	Salkowski test	Chloroform layer appears red and acid layer gives greenish yellow fluorescence	Positive
		Liebermann's reaction	Blue color	Positive
5.	Flavonoids	Shinoda test	Orange, pink, red to purple color	positive
6.	Phenolic compounds	Ferric chloride test	Dark green color	positive
		Lead acetate test	Bulky white precipitate	positive
7.	Saponins	Frothing test	Foam lasting for 1 min	positive
8.	Tannins	Gelatin test	White precipitate	positive

**Table 3: Chromatographic evaluation of neem seed oil**

Sample	Mobile phase	Visualization	Rf value
Neem seed oil in methanol(Azadirachtin)	Chloroform: Acetone (8:2)	UV (366nm)	0.36

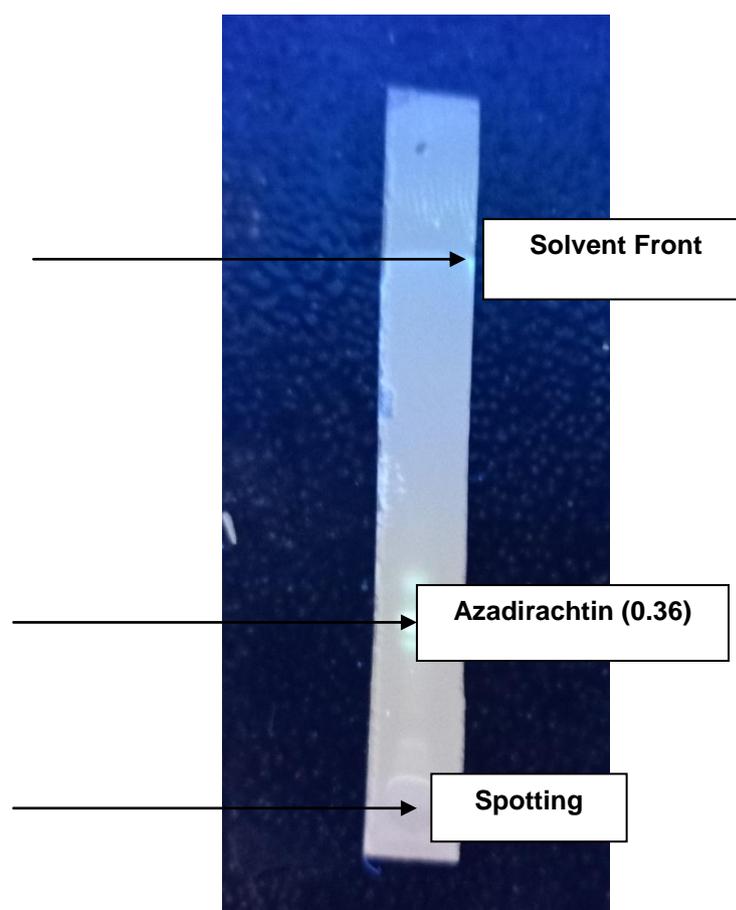


Fig. 1: TLC of Neem Seed Oil

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