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A Review on Antifungal Activity of Neem Extract (*Azadirachta Indica*)

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ABSTRACT :

Study indicate that methanolic extracts from neem tree leaves have significant antifungal activity against dermatophyte isolates, with minimal inhibitory concentrations (MIC) ranging from 50 µg/mL to 200 µg/mL. In contrast, the seed oil extracts showed higher MIC values between 625 µg/mL and 2500 µg/mL. The positive control, Terbinafine, demonstrated much lower MIC values between 0.0019 µg/mL and 0.0313 µg/mL. High performance liquid chromatography analysis revealed different chemical profiles for the leaves and seed oil extracts, suggesting the presence of terpenoids, which may contribute to their antifungal properties.

Keyword: Antifungal agent azadirachta Indica, material of method , Benifits.

Introduction :

Azadirachta indica, commonly known as Neem, Nimtree, and Indian Lilac, is a tree belonging to the mahogany family Meliaceae. Native to India, Pakistan, and Bangladesh, it thrives in tropical and subtropical areas. The Neem tree is prevalent in India, growing 10 to 15 meters tall and featuring a girth of about 2-3 meters. Its leaves are compound with multiple leaflets. Neem has been significant in Ayurvedic medicine and agriculture for centuries, with historical references highlighting the medicinal benefits of its fruit, seeds, oil, leaves, roots, and bark.

The ancient texts Carak-Samhita and Susruta-Samhita outline the medicinal uses of the Neem tree, known in Sanskrit as sarva roga nivarini, or the curer of all ailments. Its twigs are commonly used as chewing sticks in the Indian subcontinent. Studies have identified Neem's various active substances with medicinal properties. Specifically, *Azadirachta indica* is noted for its potential in treating diabetes and demonstrating anti-diabetic activity, while the aqueous extract of Neem leaves shows promise as an anti-hyperglycemic agent.

Neem :



Azadirachta Indica (Neem)

Synonym:- Margosa oil

Biological Source :- Azadirachta Indica

Family :- Miliaceae

Part of use :- leaves , Seeds, Flower, Bark

Chemical Constituent :- Azadirachtin , Nimbin , Nimbidin

Geographical source:- India , Myanmar, Tropical countrical

Uses :

Moiscturing

Cooling

Antibacterial

Antiseptic

Immunomodulators

Insecticide

Bitter tonik

Antifredant

Materials and Method :

The study focused on the Neem plant (*Azadirachta indica*), with leaves collected from a college campus. The identification of the leaves was done using a taxonomic key from the departmental library and was verified through departmental herbaria. Leaf extract was prepared for further

Leaves Extract :

The dried material was powdered and subjected to successive extraction using varying concentrations of methanol and ethanol (25%, 50%, 75%, and 100%). The resulting liquid extracts were stored in airtight bottles at 4°C.

Disk Diffusion Method :

The Kirby Bauer method, established in 1966, is designed for organisms that grow quickly at 35-37°C overnight. A disc containing a specific concentration of fungicide absorbs moisture from the agar, allowing the fungicide to diffuse into the medium. The extraction rate from the disc surpasses the diffusion rate as distance increases, resulting in a logarithmic decrease in fungicide concentration. The susceptibility of the fungus is assessed by measuring the zone of inhibition around each disc.

Medium :

Sabouraud dextrose agar was prepared and autoclaved at 121 degrees Celsius for 15 minutes at 15 pounds of pressure. It was then placed into sterile petri plates to a uniform thickness of about 5-6 mm and allowed to set at room temperature for use

Analysis by High Performance liquid Chromatography of neem extract :

The HPLC analysis utilized a C18 column and an Agilent 1100 system for LC and LC/MS, which included an auto sampler and UV visible detector. Data analysis was performed using LCMS chemStation software. The separation used a specific LichroCART C18 column with a defined size and pore diameter. The gradient program initiated with a mobile phase flow of 1.0 mL/min, gradually changing the acetonitrile to water ratio from 35:65 to 45:55 at 10 minutes, then to 70:30 at 11 minutes, and returning to 35:65 from minutes 14 to 25. The study involved a sample volume of 50 µL and a concentration of 4 mg/mL, measured at a wavelength of 213 nm over 25 minutes. The resulting HPLC profiles were compared to those of a methanol extract from neem cultured cell suspensions.

Determination of Antifungal Activity :

Dermatophyte fungi were isolated from patients at the Medical Mycology Laboratory, Universidad de Antioquia, Colombia. Classification involved microscopic and macroscopic analysis based on established criteria, alongside biochemical testing. The isolates included *Trichophyton rubrum* and *Trichophyton mentagrophytes* (five isolates each), *Epidermophyton floccosum* (three isolates), and *Microsporum canis* (one isolate). To create a conidial suspension for inoculum, the isolates of *T. Mentagrophytes*, *T. Rubrum*, and *E. Floccosum* were cultured in Potato-Dextrose-Agar or boiled rice for eight days. A sterile saline solution was added to stimulate conidial formation, and the conidial suspension was adjusted to a concentration of $1.0 - 3.0 \times 10^3$ CFU/mL. The antifungal activity tests were conducted using the M38-A2 broth microdilution method as per CLSI guidelines. RPMI 1640 medium was prepared with twofold serial dilutions of neem extracts in a MeOH:DMSO solvent and included a positive control with the commercial antifungal

Terbinafine and a solvent control. Negative and sterility controls contained no additives. The prepared medium and conidial suspension inoculum were placed in 96-multiwell microdilution plates and incubated at 35 °C for seven days without light or shaking. The neem extracts were tested at concentrations ranging from 0.0019 µg/mL to 7000 µg/mL. Only specific isolates of *T. Mentagrophytes* and *T. Rubrum* were evaluated due to a lack of *E. Floccosum* inoculum. Each experiment was performed three times with each extract and dermatophyte isolate. The Minimum Inhibition Concentration (MIC) was determined by the absence of turbidity, indicating complete fungal growth inhibition.

Application of Neem :

Neem oil is derived from the seeds of the neem tree and possesses both insecticidal and medicinal qualities, making it useful in rice pest control. Neem seed cake, a byproduct of oil extraction, enhances soil quality by adding organic matter and reducing nitrogen loss through inhibition of nitrification. Neem leaves are utilized as green leaf manure and in composting, while twigs serve as green manure when decomposed. Neem extracts from leaves and seeds have insecticidal effects and are applied as foliar sprays or seed treatments in rice farming. Additionally, the bark and roots have medicinal benefits and can control pests. Neem exhibits antibacterial, antifungal, and nematocidal properties, positively impacting disease management in rice cultivation, with many of its active components yet to be fully explored.

Neem used as Fertilizer :

The material remaining after oil extraction from seeds, known as seed cake, serves as a bio fertilizer providing essential nutrients to plants and is commonly used to enhance crop yields. Neem seed cake is effective for both food and cash crops, particularly rice and sugarcane. Its benefits include functioning as both a fertilizer and pesticide, enriching soil, reducing soil pests and bacteria, supplying essential macro nutrients for plant growth, boosting long-term yield, being biodegradable and eco-friendly, and serving as an excellent soil conditioner.

Neem as area coating agent :

Neem and its components are used to create a urea coating agent that enhances soil fertility by reducing the activity of bacteria that cause denitrification. This helps retain urea in the soil and controls various pests, including caterpillars and beetles. Available in liquid or powdered forms, Neem Urea Coating has properties such as antifeedant and pest growth regulation. Its benefits include serving as an effective soil conditioner, being a natural pesticide, environmentally friendly, non-toxic, reducing urea consumption, and boosting crop yield.

Neem as Fumigant :

The Neem tree is utilized for controlling household, storage, and crop pests. Its natural pest fumigant is available in gas form and is used as both a pesticide and disinfectant, and it is commercially used by farmers in many countries. As a non-toxic product, it is environmentally friendly and particularly significant in developing nations where synthetic fumigants cause numerous accidental poisonings. Neem fumigant not only eradicates pests but also disrupts their feeding, mating, and growth. Studies show it effectively protects stored rice grains from pests. A key advantage is that pests do not develop resistance to neem. With a growing preference for bio-based agricultural products, neem cultivation is increasing globally for its active ingredient azadirachtin, which halts pest growth. Neem is gaining traction in crop management as

Benefits :

Treats pigmentation
Exfoliation
Heals scars
Clear and glowing skin
Moisturizer
Treats care infection
Treats scalp infection
Oil control
Fights Acne
Anti aging

Conclusion :

Neem, or *Azadirachta indica*, is a significant plant known for its various benefits to both the human and animal kingdoms. It is recognized for its powerful properties as a blood purifier, detoxifier, and immune system booster, with numerous diseases showing positive responses to neem. The leaves can be consumed as tea or capsules, while neem oil can be used externally or ingested in small amounts. Additionally, neem contributes to improving soil

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