RESEARCH ARTICLE

Antifungal Activity of Medicinal Plants Cymbopogon sprenge L., Mentha spicata L., and Murraya koenigii L. against Growth of Fungi Aspergillus niger and Mucor spp.

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ABSTRACT

Medicinal plants are distributed all over the world. Plants are affected by the various types of the diseases as bacterial, nematodal, viral, fungal etc. These all are the infectious diseases caused to any part of plants and damage it. These medicinal plants extracts are used to stop the fungal infections hence; it is called as ‘antifungal’. An antifungal is a drug produced from the extract of the medicinal plants and use to cure the fungal infections. From the above result it concludes that M. koenigii and C. sprenge has highly antifungal activity in aqueous extract than n-butanol solvent against A. niger and Mucor spp. Therefore, now a days use of natural plant derived fungicide is evolved as an alternative to synthetic fungicides.

Keywords- Antifungal activity, Aspergillus niger, Mucor spp.

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1. Introduction

All these medicinal plants showed the phytochemical compounds as alkaloids, tannins, flavanoids and it showed the antioxidants, antidibetic, antibacterial, antifungal activities due to this properties these plants are used as medicinal plants. These phytochemical compounds of the International Journal of Pharmacy and Natural Medicines plants are used to cure the various infectious diseases. (Monoorkar and Gachande, 2014). Plants are affected by the various types of the diseases as bacterial, nematodal, viral, fungal etc. These all are the infectious diseases caused by any part of plants and damage it. These medicinal plant
extracts are used to stop the fungal infection hence, it is called as ‘antifungal.’ An antifungal is a drug produced from the organic extract of the medicinal plants and use to cure the fungal infections. It does not caused any type of side effects to plants, and animals. Now a days the uses of synthetically produced chemical compounds are used in large amount to cure the fungal infection. But it caused side effects to plant and animal. The various types of the locally available medicinal plants are used to cure fungal infections. *Murraya koenigii* L., *Mentha spicata* L., *Cymbopogon spreng* L these plants are easily available and are used as an antifungal drug. *Murraya koenigii* L is called as herbal antifungal drug because it having same antifungal effect as like synthetically produced fungicides of imidazole. This plant having the ability to synthesis of lipase because lipase is most important for entry of fungi into the host plant. The use of plants as an antifungal agent to cure various fungal diseases and disorders has a great significance over the synthetically and chemically prepared antifungal drugs. Therefore now a day’s most of the scientist attracted towards antibiotic that can easily obtained from living medicinal plant species.

2. Materials and Methods

Materials:

In order to carry out research work following three medicinal plants species were selected.

**Plant Materials:**

- *Murraya koenigii* L. (Curry leaves),
- *Mentha spicata* L. (Mint),
- *Cymbopogon spreng* L (Lemon grass)

**Fungi:** *Test organism:* *Aspergillus niger* and *Mucor spp.*

These two fungi *Aspergillus niger* and *Mucor spp.* are isolated from fruit and bread by bating method. These fungi are grown on petriplates up to pure culture is produced.

**Preparation of Potato Dextrose Agar medium (PDA medium):**

PDA medium were prepared for pure culture of fungi and to study the effect of plant extract.

1. Weight of 200gms of peeled potato and washed it with water.
2. Cut it into small pieces and boiled in flask for about 30 min in 500ml distilled water.
3. It allows cooling down and decanting the supernatant potato extracts using muslin cloths.
4. In a separate 1liter conical flasks take 500ml distilled water. And add 20 gm of agar –agar powder. Boiled it up to the brownish solution is formed. Add potato extract in the agar solution and also add 20gm dextrose sugar into this mixture up to it completely homogenous. Boil the mixture needed add the distilled water and make the final volume 1 liter.
5. Adjust the pH of medium by adding 0.1N NaOH by up to 6.2.
6. Sterilized petriplates, medium by using Autoclaved this autoclaved medium was poured it into the sterilized petriplates. All process is carried into the laminar air flow to avoid the contamination.

7. After this incubation solidified petriplates are used for to grow pure culture of fungi.

**Methods**

**Preparation of extracts**

*Murraya koenigii* L.: This dried powder of *Murraya koenigii* L.(Curry) leaves weight it as 0.5gm,1gm, 2gm, 4gm, 8gm and 10gm respectively. Pure solvents as 9.5ml, 9ml, 8ml, 6ml, 2ml and 10ml was used and add this plant powder into these solvents and makes the different concentration grades (5%,10%,20%,40%,80%,100%) of plant extract using solvents as n-butanol and aqueous.

*Cymbopogon spreng* L.: This *Cymbopogon spreng* L. (lemon grass) weight it as 0.5gm, 1gm, 2gm, 4gm, 8gm, and 10gm respectively and pure solvents as 9.5ml, 9ml, 8ml, 6ml, 2ml and 10ml was used and add this plant powder into these solvents and makes the different concentration grades (5%,10%,20%,40%,80%,100%) of plant extract using solvents as n-butanol and aqueous.

*Mentha spicata* L.: This *Mentha spicata* L. (mint) weight it as 0.5gm, 1gm, 2gm, 4gm, 8gm, and 10gm respectively. Pure solvents as 9.5ml, 9ml, 8ml, 6ml, 2ml and 10ml was used and add this plant powder into these solvents and makes the different concentration grades (5%,10%,20%,40%,80%,100%) of plant extract using solvents as n-butanol and aqueous.

All above mixture were shaked frequently and kept for 74hrs in order to complete extraction of active principles in solvent used. Then filtered using muslin cloths and filtrate was used as extract of particular concentration of that plant species for antifungal activity.

**Identification of fungi:**

The use of fruit of banana, papaya and bread to grow the fungi for about 15 days into the incubation chamber. After 15 days the various types of fungi were grown on this fruits. This fungi using forceps and niddle taken on slides and stained with cotton blue and mount in lactophenol. These slides were observed under the compound microscope. For the identification of this fungi book of “Introductory Mycology” (C.J. Alexopolous and C.W. Mins.) 3rd edition was referred. This identified fungi of *Aspergillus niger* and *Mucor spp.* were used for further pure culture on PDA medium.

**Fungal suspension preparations:**

The suspension preparation worked under laminar air flow. Fungal spores from pure culture plate of these fungi mixed with the 50ml distilled water.

**Preparation of wells:**

The wells were made using sterilized cork borer on the agar petriplates. First the borer was deeped into the alcohol for sterilization and then it used to make the wells on petriplates for zone of inhibition test.

**Inoculation of plant extract into the wells:**

i. This fungal suspension is spread by dropper on petriplates.

ii. Using micropipette the extract was added into each well which was made on petriplate.

iii. These plates were incubated for 24 hours at room temperature.

iv. The next day the zone of inhibition were observed.
v. Record observations of this zone and observed the effect of three medicinal plants on two fungi.

3. Results and Discussion

In the present study the antifungal activity of medicinal plants extract growth of fungi Aspergillus niger and Mucor spp was examined and result obtained are given in table no. 1. Medicinal plants having pharmacological properties like anabolic, hypotensive, cardiac depressant and smooth muscle relaxant, antifertility and antistress activity, its extract show antifungal activity. (Manoorkar and Gachande, et.al 2014) from the above observation it is observed than zone against Aspergillus niger and Mucor spp at different concentrations i.e. 5%, 10%, 20%, 40%, 80% and control in n-butanol and aqueous solvents.

In Cymbopogon ssp L. zone observed at 40%, 80% and not observed in 5%, 10%, 20%, and control against Mucor spp. Antifungal activity of Mentha spicata L. against Mucor spp. showed zone only 40%, 80% in n-butanol. In aqueous extract zone observed at 10%, 20%, 40%, and 80% but not shown in any effect at 5% concentration. In Murraya koenigii L. showed inhibition activity against Mucor spp. in n-butanol extract, zone observed at concentration 5%, 10%, 20%, 40%, 80% and not in control. In aqueous extract observed at 5%, 10%, 40%, and 80% but does not shown in 20% and control.

Then we observed zone against Aspergillus niger at different concentration i.e. (5%, 10%, 20%, 40%, 80% and control) in n-butanol and aqueous solvent. In Cymbopogon ssp L. zone observed at each concentration in aqueous extract but did not show any zone in extract of n-butanol against Aspergillus niger. Lemongrass oil shows high significant antifungal activity against Aspergillus niger (Yousef, 2013).

The antifungal activity of Mentha spicata L. against Aspergillus niger shows zone only 5% concentration of n-butanol but other concentration did not shows any effect. In aqueous extract zone observed at each concentration but except control. Peppermint oil has been also shown to be fungistatic (Yousef, 2013). In Murraya koenigii L. showed inhibition activity against Aspergillus niger in n-butanol extract zone observed 40% and 80% except 5%, 10%, 20% and control. In aqueous extract zone observed in 5%, 10%, 20%, 40%, and 80% except control.
Table 1: Antifungal activity of plant species against fungal test species

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Plant species</th>
<th>Fungi test Species</th>
<th>Solvent used for extraction</th>
<th>zone of inhibition at different concentrations</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>5%</td>
<td>10%</td>
</tr>
<tr>
<td>1.</td>
<td><em>Cymbopogon Spreng. L.</em></td>
<td><em>Mucor spp</em></td>
<td>n-butanol</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Aqueous</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Aspergillus Niger</em></td>
<td>n-butanol</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Aqueous</td>
<td>+</td>
</tr>
<tr>
<td>2.</td>
<td><em>Mentha spicata L.</em></td>
<td><em>Mucor spp</em></td>
<td>n-butanol</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Aqueous</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Aspergillus Niger</em></td>
<td>n-butanol</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Aqueous</td>
<td>+</td>
</tr>
<tr>
<td>3.</td>
<td><em>Murraya koenigii L.</em></td>
<td><em>Mucor spp</em></td>
<td>n-butanol</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Aqueous</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Aspergillus Niger</em></td>
<td>n-butanol</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Aqueous</td>
<td>+</td>
</tr>
</tbody>
</table>

4. Conclusion
From the above result we concluded that *Murraya koenigii* L. and *Cymbopogon spreng. L.* highly antifungal activity in aqueous extract than n-butanol solvent against *Aspergillus niger* and *Mucor spp.* but in *Murraya koenigii* L. highly activity in different concentration of n-butanol solvent than aqueous extract. Therefore it is recommended that, in agricultural field the use of natural plant derived fungicide for alternative source of synthetic fungicides is best and ecofriendly option for chemical fungicides.

5. References


