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# Screening of Phylloplane Fungal Flora of Some Medicinal Plants in Durg- Bhilai Region of Chhattisgarh State

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***Abstract:*** ***The leaf surface fungal flora is subjected not only to the influence of the host, but also to its own factors. An important aspect is the production of self incubatory products as well as self stimulating products by the fungal organisms present on the leaf surface. Amongst microorganisms fungi can be a sole cause of spoilage and substantially decrease the quality, grade and price of plant materials. In the present studies leaves were found mainly infected with Aspergillus niger, A. favus , Curvularia lunata, Chaetomium globosom, Penicillum citrinum and Fusarium oxysporum as these six were found on the leaves throughout the study and their incidence degree were quite high.***
***Keywords: Phylloplane, Fungal flora, IVI***

## I. INTRODUCTION

Plants are the most important sources of medicine. Throughout the world several thousand plants have been and are still used for medical purposes. These medicinal plants have their values in the substance or substances present in various plant tissues. The important of these substances are alkaloids, compounds of carbon, hydrogen, oxygen and nitrogen. Besides these glycosides, essential and fatty oils, resins, gums, tannin etc. are also of large use.

The leaf surface of plants represents a colorful arrange of mycoflora compsising heterogeneous population of both polygenic and non polygenic fungal species. (Chandrol, 2008). Deuteromycetous fungi were found to be the highest in number and *Aspergillus & Penicillium* were dominant among all fungal species (Hasin and Islam, 2010). The term “phyllosphere” was proposed by Gregory and phylloplane was proposed by Karling. The use of term “phyllosphere” should be restricted to the zone near the leaves and term “phylloplane” should be used to refer to actual surface of the leaf. Plant leaves provide suitable substrate for the growth of much diverse fungal organisms, which in turn cause heavy damage to these leaves and decrease its market value to a greater extent or render it completely unusable. The associations of fungi and other kinds of micro organisms with leaves are known since long. A large number of fungal organisms are known today and well documented mycological records are available.

The present studies aim to evaluate qualitatively and quantitatively fungal flora from some medicinal plants to get ways to save medicinal value of plant.

## II. MATERIAL & METHOD

1) *Collection of Samples:* Fresh harvested leaves of *Ocimum, Eucalyputs, Datura, Calotropis and Catharanhus* species were collected at random from herbs, shrubs and trees growing at Durg Bhilai region of Chhattisgarh State and brought to the laboratory under aseptic conditions.
2) *Isolation of Fungal Organisms:* The isolation of leaf surface fungi area done by culturing the leaf surface washing and by direct incubation of leaves in moist chamber (Sharma, 1974). 15 leaves from each sample were randomly selected of which 5 leaves were kept in moist chamber for 5-10 days for growth of fungal flora remaining 10 leaves were surface sterilized using a sterile cork borer, disks 5mm diameter were cut from lamina. The disks were transferred to a conical flask with 100 ml sterilized distilled water and shaken thoroughly for 10 minutes. After proper shaking, 1ml aliquot of each sample was poured into culture plates in fine replicates containing potato dextrose agar (P.D.A.) media supplemented with rose Bengal (Antibacterial). The culture plates were incubated for 5-10 days at 28 ± $1^{o}$ c temperature and fungal colonies developed on plates were recorded. The mixed cultures were purified through single spore culture method. After ensuring the purity, the cultures were stored in the slants for further studies.

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3) *Identification of Fungal Organisms:* The purified fungal cultures were identified by using mycological techniques and were compared with the available authentic literature, reviews and mycological manuals (*Damatiaceous Hyphomycetes* - Ellis, 1976; *Introductory mycology* - Alexopoulos, 1978; *Illustrated genera of imperfect fungi* - Barnett and Hunter, 1987 and *A manual of soil fungi* - Gilman, 1995 etc.).

4) *Myco-Ecological Parameters:* Various myco-ecological characters have been calculated using the following formulae-

a) *Relative frequency (RF %)*

RF = frequency of a specific organisms / Total frequency of all organisms X100

b) *Relative density (RD %)*

RD = Density of each organisms / Total Density of all X 100

c) *Relative Abundance (RA %)*

RA= Abundance of each organisms / Total Abundance of all organisms x 100

d) *Importance Value Index (IVI)*

IVI = Relative frequency + Relative Density + Relative Abundance

## III. RESULT & DISCUSSION

### A. *Phylloplane Fungal flora of Ocimum Sanctum*

Screening studies revealed that out of 09 fungal species isolated, 06 species namely *Aspergillus niger, A. flavus, Curvularia lunata, Chaetomium globosum, Penicillum spp. and Fusarium oxysporum* were present with high IVI and most frequent throughout the study. *Aspergillus niger* was found most frequent followed by *Penicillium citrinum, A. flavus, Fusarium oxysporum* respectively while *Penicillum rotundum* was found least frequent. From density point of view, *Aspergillus niger* was most dominant followed by *Aspergillus flavus, Fusarium oxysporum, Curvularia lunata* and *Chaetomium globosum* respectively. *Chaetomium globosum* was found most abundant over other fungal organisms. (Table No.1)

### B. *Phylloplane Fungal Flora of Eucalyptus rostrata*

Screening studies revealed that out of 06 fungal species isolated, 04 species i.e., *Aspergillus niger, A. flavus, Curvularia lunata* and *Chaetomium globosum* were present with high IVI. *Aspergillus niger, Chaetomium globosum* and *A. flavus were* most frequent. *Alternaria crassa* was found least frequent throughout the study. From density point of view, *Aspergillus niger, Curvularia lunata* and *Aspergillus flavus* were found dominated over other fungal organisms. *Aspergillus niger* was most abundant followed by *A. flavus, Chaetomium globosum* and *Curvularia lunata* respectively. (Table No. 2)

### C. *Phylloplane fungal flora of Datura Stramonium*

Screening studies revealed that out of 10 fungal species isolated, 06 species i.e., *Chaetomium globosum, Fusarium oxysporum, Curvularia lunata, Aspergillus niger, Penicillium citrinun* and *A. flavus*, were present with high IVI. *Chaetomium globosum, Aspergillus niger, Curvularia lunata, Fusarium oxysporum* and *Penicillium citrinun* were found most frequent. From density point of view, *Chaetomium globosum* was most dominant followed by *Fusarium oxysporum, A. niger* and *Curvularia lunata* respectively. *A. flavus* was found most abundant over other fungal organisms while *Aspergillus sulphureus* was found least frequent during the study. (Table No. 3)

### D. *Phylloplane fungal flora of Calotropis procera*

Screening studies revealed that out of 07 fungal species isolated, 04 species namely *Aspergillus niger, Fusarium oxysporum, Penicillium citrinum* and Rhizopus nodosus were present with high IVI. *Aspergillus niger* and *Fusarium oxysporum* were found most frequent. From density point of view, *Aspergillus niger* was most dominant over other fungal organisms. *Aspergillus niger* and *Fusarium oxysporum* were found most abundant throughout the study. *Aspergillus terreus* was found least frequent. (Table No. 4)

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*E. Phylloplane Fungal Flora of Catharanthus Rosea*

Screening studies revealed that out of 09 fungal species isolated, 04 species i.e., *Aspergillus niger, Fusarium oxysporum, Curvularia lunata* and *A. flavus* were found with high IVI. *Aspergillus niger, A. flavus* and *Penicillium citrinum* were found most frequent over other fungal organisms. From density point of view, *Aspergillus niger* was most dominant followed by *Curvularia lunata* and *fusarium oxysporum* respectively. *Aspergillus niger* was most abundant while *Aspergillus tamarii* was found least frequent. **(Table No. 5)**

Some of the fungal species were found confined to a particular phyllosphere only. For example *Aspergillus ochraceus* and *Penicillium rotundum* were found confined to a phyllosphere of *Ocimum sanctum.* Some fungal species are also highly confined to a particular phyllosphere only such as *Alternaria crassa* (*Eucalyptus rostrata*), *Aspergillus sulphureus (Datura stramonium)* and *Aspergillus tamarii* (*Catharanthus rosea).* Those confinements of fungal species certainly depend on floristic pattern of the area. Much diversity in fungal flora indicates more diversity in the vegetation composition of that particular area.

Composite results indicates that in all five leaf phylloplane, damages of leaves were mainly caused by *Aspergillus niger, A. flavus, Curvularia lunata, Chaetomium globosum, Penicillium citrium* and *Fusarium oxysporum* throughout the study due to their high degree of frequency, density and abundance. Importance value index (IVI) is the pool value of the percentage relative frequency, relative density and relative abundance. IVI of a particular fungal species gives the idea of its relative importance in the fungal community. (Table No. 1-5)

The species of *Aspergillus, Fusarium, Alternaria, Penicillium and Rhizopus* were reported as the most common fungi isolated from drug plants (Aziz and Youssef 1991). A large number of Ascomycota species are economically important (James et al., 2006; Kirk et al., 2008; Crous, 2009). A variety of relationships exist between fungal organisms and their host plants, ranging from mutualistic or symbiotic to antagonistic or slightly pathogenic (Arnold, 2007). Variation in the fungal population on leaf surface depends upon the climatic factors. The variation in the composition of leaf surface fungal flora depends upon the biochemical nature of the host plants.

Table NO. 1: MYCO- Ecological Parameters - *Ocimum sanctum* L.

| S. No. | Name of Organisms | Relative Frequency % | Relative Density % | Relative Abundance% | IVI |
|---|---|---|---|---|---|
| 1 | *Aspergillus niger* Vantieghem. | 25.23 | 18.65 | 15.04 | 58.92 |
| 2. | *A. flavus* link. | 18.38 | 15.32 | 13.55 | 47.25 |
| 3. | *A. ochraceus* Wilhelm. | 10.84 | 4.93 | 03.37 | 19.14 |
| 4. | *Curvularia lunata* Boedijn. | 15.38 | 12.69 | 13.16 | 41.23 |
| 5. | *Chaetomium globosum* Kunze. | 16.00 | 10.68 | 15.88 | 42.56 |
| 6. | *Penicillum rotundum* Raper & Fennell. | 6.84 | 3.15 | 6.39 | 16.38 |
| 7. | *Rhizopus nodosus* Namystowski. | 11.53 | 9.28 | 10.49 | 31.3 |
| 8. | *Fusarium oxysporum* Link. | 16.38 | 13.25 | 14.48 | 44.11 |
| 9. | *Penicillium citrinum* Thom. | 18.66 | 9.77 | 13.72 | 42.15 |

Table NO. 2 : MYCO- Ecological Parameters - *Eucalyptus rostrata* schlecht.

| S. No. | Name of Organisms | Relative Frequency % | Relative Density % | Relative Abundance% | IVI |
|---|---|---|---|---|---|
| 1. | *Aspergillus niger* Vantieghem. | 21.83 | 18.01 | 17.60 | 57.44 |
| 2. | *A. flavus* Link. | 19.66 | 13.97 | 15.70 | 48.33 |
| 3. | *A. Sydowi* Thom & Church. | 10.33 | 8.78 | 7.97 | 27.08 |
| 4. | *Alternaria crassa* Rands. | 7.16 | 5.44 | 4.55 | 17.15 |
| 5. | *Curvularia lunata* Boedijn. | 16.66 | 15.51 | 13.07 | 45.24 |
| 6. | *Chaetomium globosum* kunze. | 20.66 | 11.18 | 13.36 | 45.50 |

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Table NO. 3 : MYCO- Ecological Parameters - *Datura stramonium* L.

| S. No. | Name of Organisms | Relative Frequency % | Relative Density % | Relative Abundance% | IVI |
|---|---|---|---|---|---|
| 1. | *Aspergillus niger* Vantieghem. | 16.88 | 10.39 | 9.55 | 36.82 |
| 2. | *A. flavus* link. | 13.11 | 9.90 | 11.41 | 34.42 |
| 3. | *A. sulphureus* Thom & Church. | 7.77 | 5.49 | 2.15 | 15.41 |
| 4. | *A terreus* Thom. | 7.55 | 4.99 | 4.63 | 17.17 |
| 5. | *Alternaria solani* Sorauer . | 11.33 | 8.32 | 8.77 | 28.42 |
| 6. | *Curvularia lunata* Boedijn. | 16.11 | 10.24 | 11.40 | 37.75 |
| 7. | *Chaetomium globosum* Kunze. | 17.11 | 11.99 | 9.50 | 38.6 |
| 8. | *Penicillium citrinum* Thom. | 15.11 | 8.88 | 11.04 | 35.03 |
| 9. | *Rhizopus nodosus* Namystowski. | 10.33 | 8.02 | 6.50 | 24.85 |
| 10 | *Fusarium oxysporum* link. | 16.11 | 11.26 | 11.18 | 38.55 |

Table No. 4 : MYCO- Ecological Parameters - *Calotropis procera* Br. Vern.

| S. No. | Name of Organisms | Relative Frequency % | Relative Density % | Relative Abundance% | IVI |
|---|---|---|---|---|---|
| 1. | *Aspergillus niger* VanTieghem. | 30.73 | 17.50 | 20.60 | 68.83 |
| 2. | *A. sydowi* Thom & Church. | 10.69 | 8.00 | 8.80 | 27.49 |
| 3. | *A. terreus* Thom. | 8.69 | 8.50 | 6.94 | 27.49 |
| 4. | *Alternaria solani* Sorauer. | 15.04 | 10.20 | 12.07 | 37.31 |
| 5. | *Penicillium citrinum* Thom. | 19.39 | 14.83 | 15.16 | 49.38 |
| 6. | *Rhizopus nodosus* Namystowski. | 15.04 | 12.52 | 13.05 | 40.61 |
| 7. | *Fusarium oxysporum* Link. | 21.39 | 14.42 | 16.34 | 52.15 |

Table No. 5 : MYCO- Ecological Parameters - *Catharanthus rosea* L.

| S. No. | Name of Organisms | Relative Frequency % | Relative Density % | Relative Abundance% | IVI |
|---|---|---|---|---|---|
| 1. | *Aspergillus niger* VanTieghem. | 19.12 | 11.00 | 15.97 | 46.09 |
| 2. | *A. flavus* link . | 15.90 | 8.63 | 11.10 | 35.63 |
| 3. | *A. tamarii* Kita. | 5.22 | 3.12 | 4.56 | 12.9 |
| 4. | *Alternaria solani* Sorauer. | 13.67 | 7.33 | 7.25 | 28.25 |
| 5. | *Curvularia lunata* Boedijn . | 14.90 | 10.94 | 10.62 | 36.46 |
| 6. | *Chaetomium globosum* Kunze. | 13.90 | 8.92 | 6.50 | 29.32 |
| 7. | *Penicillium citrinum* Thom. | 15.90 | 7.43 | 7.98 | 31.31 |
| 8. | *Fusarium oxysporum* Link. | 14.90 | 10.26 | 11.52 | 36.68 |
| 9. | *Cladosporium herbarum* link. | 9.45 | 7.46 | 6.77 | 23.68 |

According to Chandel (1990), the frequency and abundance are directly or indirectly correlate with climatic conditions. The associated microorganisms use plant material as a source of nutrients for their own growth and reproduction. Thus they increase in number, utilize nutrients and cause certain changes in quality of products (Jain et al., 1994).

In developing and under developing countries losses can reach up to 15-20% or even more (Jain et al., 1994). Bilgrami et al. (1981) reported a number of fungi including *Alternaria, Penicillium , Rhizopus* and *Pestalotia* species on grapes, Chopra (1982), studied rot of *Alubukhara* caused by *Aspergillus niger*. Similar results were also obtained by Krishnaiah and Thirupathaiah (1990). Patel and Pathak (1993) also found that *Rhizopus* and *Botryodiplodia* species were responsible for *guava rot.* According to Dutta and Roy (1987) *Aspergillus niger*. *A. flavus* and *Pencillium citrinum* had maximum percentage of incidence. Earlier workers have reported dominance of *Aspergillus, Chaetomium , Fusarium , Penicillium* and *Curvularia* species in the leaf surface of different plant in India also (Mishra and Tiwari, 1978 & Chandrol, 2002, 2003, 2007).

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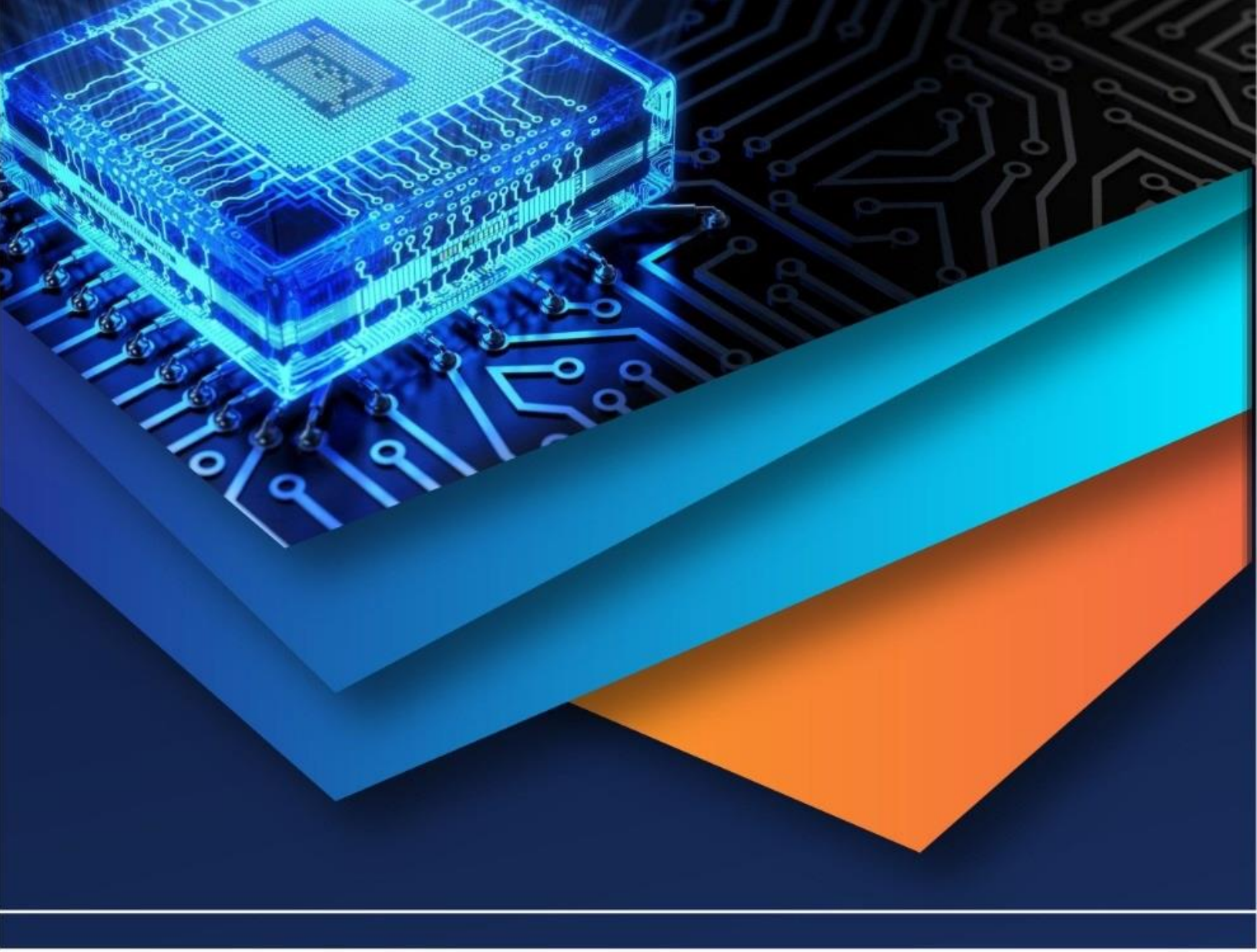
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