



ISSN: 2230-9926

Available online at <http://www.journalijdr.com>

IJDR

International Journal of
DEVELOPMENT RESEARCH

International Journal of Development Research
Vol. 6, Issue, 03, pp. 7041-7043, March, 2016

Full Length Research Article

ISOLATION AND IDENTIFICATION OF *PENICILLIUM* SPP., FROM KRISHNA RIVER, DISTRICT- SANGLI

*Andoji, Y.S. and Chougule, P.M.

Department of Botany, P.D.V.P. College, Tasgaon

ARTICLE INFO

Article History:

Received 17th December, 2015
Received in revised form
25th January, 2016
Accepted 10th February, 2016
Published online 31st March, 2016

Key Words:

River Krishna,
Soil Ecosystem,
Penicillium Spp.

ABSTRACT

The mycoflora from the bed of river Krishna at Sangli was studied at three different locations viz., Right Bank, Center and Left Bank from January 2014 to December 2015. Twenty six soil samples were collected from surface, 10, 15, and 25 cm depth. The mycoflora were isolated by using soil dilution and soil plate method. Out of the 75 strains of fungi isolated 10 species of *Penicillium* viz., *Penicillium funiculosum* (32.66) and *P. semitectum* (03.88%), *P. expensum* (2.33%), *P. chrysogenum* (16.33%), *P. Lilacinum* (09.63%), *P. notatum* (15.66%), *P. roseum* (1.62%), *P. tardum* (23.67%), *P. citrinum* (09.66%) and *P. rubrum* (2.67%). were identified. Greater number of species were isolated on soil plate technique as compared to dilution plate technique. Higher number of species were obtained from right bank as compared to left bank and very low frequency were obtained from centre.

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INTRODUCTION

Soil is a very complex environment in which the biological activity is mostly influenced by microorganisms. There are number of beneficial effects of soil microbes which includes nitrogen fixation and organic matter decomposition to breakdown of metabolic by-products and agrochemical, enhancing the bioavailability of nitrates, sulphates, phosphates and essential metals (Bridge & Spooner, 2001). Mycoflora is an important constituent of the soil microbiota typically constituting more of the soil biomass as compared to bacteria, depending upon the soil depth and nutrient conditions (Ainsworth & Bisby, 1995). The role of fungi in the soil is much complex one and fundamental to the soil ecosystem. They perform ecological services that highly impact on the quality of human welfare and give enormous potential for providing economic benefits, e.g., the isolation and identification of the soil fungus *Penicillium* led to a large pharmaceutical industry of antibiotics (Diana, 1994). It is recorded that there are 1.5 million fungal species on earth and out of which only about 70,000 have been described up to now (Hawksworth and Rossman, 1997). The present investigation is an attempt to study the variability of mycoflora from different depths at three locations of river Krishna at Sangli.

Apparently no report is available for fungi recorded from this site. This paper concentrates only on species of *Penicillium*.

Description of the research site: The study area is located at longitude 58.°21'E, latitude 21.°21'N. Air temperature ranges between 11°C to 44.7°C. There are significant variations in rainfall in the basin. The rainy months are from June to September end and the driest months are November to March end, during which the average monthly rainfall rarely exceeds 25 mm. The soil texture ranges from coarse to fine which is mostly favourable for irrigated agriculture. The pH value normally ranges from 7.5 to 8.30.

MATERIALS AND METHODS

The analysis of soil samples done in this study were collected from three different sites viz Left Bank, Right Bank and Center from the bed of river Krishna. Vertical samples were collected from surface, 10, 15 and 25cm depths with presterilized screw-cap vials. Vials were dipped perpendicularly to the vertical surface of the water. Three samples were collected from each depth. The samples were kept in pre-sterilized polyethylene bags surrounded by ice crystals until they brought to the laboratory. The samples were analysed by using the soil dilution plate (Waksman, 1922) and soil plate method (Warcup, 1950).

*Corresponding author: Andoji, Y.S.,
Department of Botany, P.D.V.P. College, Tasgaon.



Plate 1. Culture plate showing *Penicillium* culture

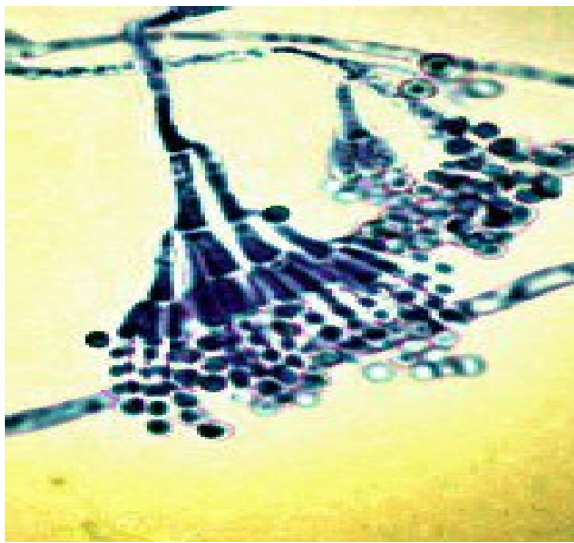


Plate 2. *Penicillium notatum*

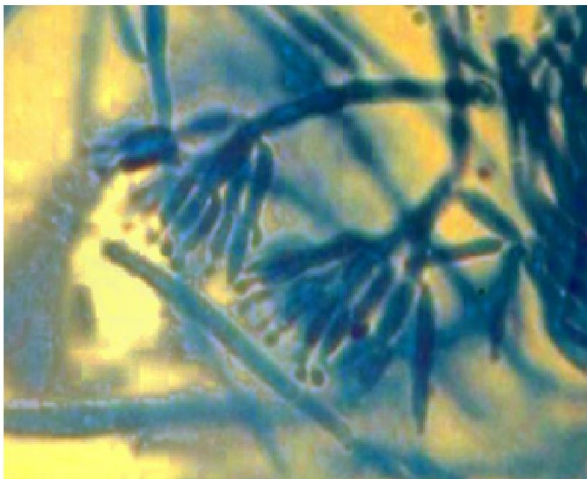


Plate 3. *Penicillium funiculosum*



Plate 4. *Penicillium chrysogenum*

From such dilutions, 1ml samples were spread on Potato Dextrose Agar medium and incubated at 28°C for four days.

Soil Plate Method: About 0.05 g of soil sample was spread randomly on the bottom of a sterile Petri plate and pre sterilized PDA medium was poured, which was then spread gently to scatter the soil particles in medium. The plates were then incubated at 28°C for four days. Fungal colonies were counted and isolated for *Penicillium* species was made from mixed isolates and subcultured on PDA. Sub culturing of *Penicillium* spp. were continued up till pure culture was obtained. Identification of *Penicillium* spp. were done by following standard manuals Raper & Thom (1949) and Gilman (1945), (Ellis,1960), (Ellis,1963), (Ellis,1976), (Subramanian,1971).).

RESULTS AND DISCUSSION

A total of 10 species were isolated and identified from forty six soil samples. From the three study sites, Right bank, Center and Left bank, a total of 1433 colonies of *Penicillium* spp., were isolated of which 723 is of *Penicillium funiculosum* (32.66) and *P. semitectum* (03.88%), *P. expansum* (2.33%), *P. chrysogenum* (16.33%), *P. Lilacinum* (09.63%), *P. notatum* (15.66%), *P. roseum* (1.62%), *P. tardum* (23.67%), *P. citrinum* (09.66%) and *P. rubrum* (2.67%). Rate of occurrence of species isolated in terms of percentage from each of the three sites is given in Table 1.

Table 1. Name of isolated *Penicillium* spp. from Left bank, Center and Right bank

Sr.No	Name of isolated <i>Penicillium</i> spp.	Left bank, Center and Right bank frequency percentage
1	<i>Penicillium semitectum</i>	03.88
2	<i>Penicillium expansum</i>	2.33
3	<i>Penicillium chrysogenum</i>	16.33
4	<i>Penicillium funiculosum</i>	32.66
5	<i>Penicillium lilacinum</i>	09.63
6	<i>Penicillium notatum</i>	15.66
7	<i>Penicillium rubrum</i>	2.67
8	<i>Penicillium roseum</i>	1.62
9	<i>Penicillium tardum</i>	23.67
10	<i>Penicillium citrinum</i>	09.66

Soil dilution plate Method: The soil samples were mixed with sterile distilled water in equal volume and a series of dilutions were made.

From the collected during present investigation samples of left bank, *P. tardum* was the predominant fungus isolated and accounted for 23.67% of the total fungi isolated from left bank whereas from samples of right bank, *P. funiculosum*, was the predominant fungal species which accounts for 32.66%. Not a single fungal species were predominated from the center part of the bed instead of *P. chrysogenum* (16.33) and *Penicillium expansum* (2.33). The study shows that the total number of species of *Penicillium* isolated decreased with increased sampling depth. A greater number of species and colonies were isolated on soil plates as compared to dilution plates and were recovered from right bank (51.63%) where the soil is rich in organic matter and vegetation as compared to right bank (39.43%) while in center, isolates were lowest (13.33) in frequency and diversity.

Conclusion

No previous report on the predominance and diversity of *Penicillium* from the soil of River Krishna were found. This preliminary data focused on the predominance of soil fungi in the bed of river Krishna. A detailed survey including many more samples to confirm the distribution of soil fungi in river Krishna with particular emphasis on the ecological factors determining the predominance in various soil types and habitats is very essential tool.

Acknowledgement

The authors are thankful to Dr. Narendra A. Kulkarni, Head department of botany and Principal Dr. Ramesh R. Kumbhar, P.D.V.P. college Tasgaon for moral support and providing lab facility during present research work.

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