

## **ASPERGILLUS MYCOFLORA ISOLATED FROM SOIL OF KOTRI BARRAGE SINDH, PAKISTAN**

**M. SUHAIL, FOZIA IRUM, T. JATT, FARZANA KOREJO AND H. ABRO**

*Institute of Botany,  
University of Sindh, Jamshoro, Sindh, Pakistan.*

### **Abstract**

Mycoflora from the soil of Kotri barrage at down stream was investigated at three locations viz., Right Bank, Centre and Left Bank from July 2005 to May 2006. Thirty samples were collected from surface, 10, 20, 30 and 50cm depth. The fungi were isolated by using soil dilution and soil plate method. Out of 90 strains of fungi isolated, 21 species of *Aspergillus* viz., *A. niger* (21.07), *A. flavus* (18.26), *A. ochraceus* (17.28), *A. wentii* (7.47), *A. flavus oryzae* (7.85), *A. fumigatus* (8.49), *A. sulphureus* (2.29), *A. ustus* (3.48), *A. violaceofuscus* (1.82), *A. flavipes* (2.97), *A. terreus* (0.80), *A. clavatus* (0.72), *A. restrictus* (0.93), *A. versicolor* (0.55), *A. candidus* (1.35), *A. nidulans* (0.38), *A. citrisporus* (0.04), *A. granulosis* (1.95), *A. sparsus* (0.55), *A. elegans* (0.50), *A. giganteus* (1.14) were identified. Greater number of species were isolated on soil plate method than on dilution plate method. Higher number of species were recovered from Right bank as compared to left bank while in centre isolates were in low frequency.

### **Introduction**

Fungi are an important component of the soil micro biota. (Ainsworth & Bisby, 1995). The role of fungi in the soil is an extremely complex one and is fundamental to the soil ecosystem (Diana, 1994). Aspergilli are ubiquitous in nature. They are geographically widely distributed and have been observed in a broad range of habitats principally in soils and decaying vegetation. Species of *Aspergillus* are an important microorganism, both medically and commercially. Some of these fungi are important pathogens of plants and animals (Gregory *et al.*, 1997).

In a previous study, the occurrence of *Penicillium* spp., from the bank of the river Indus has been reported (Suhail *et al.*, 2006). The taxonomy of *Aspergillus* is primarily based on morphological features, rather than the physiological features of *Aspergillus* spp., (Anon., 1997). The present research is an attempt to study the diversity from various depths and different locations of Kotri barrage, Sindh, Pakistan.

### **Material and Method**

Kotri barrage originally called the lower Sindh barrage was renamed as Ghulam Muhammad barrage constructed in 1955 (Mushtaq, 1975). The area is located at longitude 68.°22'E, latitude 25.°22'N. It is situated on the right bank of the river Indus. Air temperature ranges between 9.3°C to 40.4°C. There are significant extremes of rainfall in the basin (Suhail *et al.*, 2006). The soil texture ranges from coarse to fine with 85% in the moderately fine categories, mostly suitable for irrigated agriculture. The pH value generally ranges from 8 to 8.50 (Coleman, 2004).

Thirty soil samples used for the isolation and identification were collected from various depths (Surface, 10, 20, 30 and 50cm) of Kotri barrage, Sindh Right Bank, Left Bank and Center with two intervals of approximately 100 yards distance by using quadrat method

from the bed of Indus at the kotri barrage. Vertical samples were taken from surface, 10, 20, 30 and 50cm depths. Nine samples were collected at each depth. The samples were stored in sterilized polyethylene bags. The samples were processed using the soil plate method (Warcup, 1950) and Soil dilution plate Method (Waksman, 1922).

Soil plate method: About 1g of soil was scattered on the bottom of a sterile Petri dish and molten cooled (40-45°C) agar medium (PDA) & (CZEP) was added, which was then rotated gently to disperse the soil particles in the medium. The plates were then incubated at 28°C for three days.

Soil dilution plate method: The soil samples were mixed with sterile distilled water and a series of dilutions were made. From the dilutions, 1ml volumes were pipetted onto Potato Dextrose agar and Czapek Dox agar and incubated at 28°C for three days.

Fungal colonies were counted and screened for *Aspergillus* species. Identification was performed according to Raper & Thom (1945) and Gilman (1957).

### Result and Discussion

Out of 2354 colonies of *Aspergillus* species, a total of 21 species were obtained from 45 soil samples. The total number of species isolated decreased with increased sampling depth. A greater number of species and colonies were isolated on soil plates than on dilution plates and were recovered from right bank (49.02) where the population rate was high as compared to left bank (34.40) while in centre, isolates were lowest (16.56) in frequency and variation.

No previous report on the prevalence and distribution of this genus in the soil of Kotri barrage is reported (Sultan *et al.*, 1997).

**Table 1. Per day growth in mm at room temperature.**

S. NO.	Name of species	Czapek medium	PDA medium	Temperature °C
1.	<i>Aspergillus niger</i>	5.2	6.5	25-35
2.	<i>A. flavus</i>	5.3	5.0	25-35
3.	<i>A. ochraceus</i>	6.1	6.5	25-35
4.	<i>A. wentii</i>	3.1	4.8	25-35
5.	<i>A. flavus oryzae</i>	4.25	5.25	25-35
6.	<i>A. fumigatus</i>	2.5	2.8	25-35
7.	<i>A. sulphurus</i>	2.05	4.8	25-35
8.	<i>A. ustus</i>	2.04	2.0	25-35
9.	<i>A. violaceo-fuscus</i>	2.9	3.4	25-35
10.	<i>A. flevipes</i>	2.9	4.7	25-35
11.	<i>A. terreus</i>	3.8	4.9	25-35
12.	<i>A. clavatus</i>	1.9	3.3	25-35
13.	<i>A. restrictus</i>	1.95	2.5	25-35
14.	<i>A. versicolor</i>	4.0	5.2	25-35
15.	<i>A. candidus</i>	3.8	4.0	25-35
16.	<i>A. nidulans</i>	1.9	2.4	25-35
17.	<i>A. citrisporus</i>	2.5	2.0	25-35
18.	<i>A. granulosis</i>	1.5	1.9	25-35
19.	<i>A. sparsus</i>	1.8	4.5	25-35
20.	<i>A. elegans</i>	3.9	4.9	25-35
21.	<i>A. giganteus</i>	2.9	3.5	25-35

Table 2. *Aspergillus* species with number of colonies and percentage.

S. No.	Name of isolates & identified species	Sample from right bank	% age	Samples from left bank	% age	Samples from center	% age	Total colonies	% age
1.	<i>Aspergillus niger</i>	226	9.60	174	7.37	96	4.07	496	21.07
2.	<i>A. flavus</i>	209	8.87	133	5.64	88	3.73	430	18.26
3.	<i>A. ochraceus</i>	187	7.94	149	6.32	71	3.01	407	17.28
4.	<i>A. wentii</i>	103	4.37	64	2.71	09	0.36	176	7.47
5.	<i>A. flavus oryzae</i>	87	3.69	41	1.74	57	2.42	185	7.85
6.	<i>A. fumigatus</i>	112	4.75	67	2.84	21	0.89	200	8.49
7.	<i>A. sulphureus</i>	---	0.00	46	1.95	08	0.33	54	2.29
8.	<i>A. ustus</i>	51	2.16	31	1.31	---	0.00	82	3.48
9.	<i>A. violaceo-fuscus</i>	18	0.76	23	0.97	02	0.08	43	1.82
10.	<i>A. flavipes</i>	39	16.56	21	0.89	10	0.42	70	2.97
11.	<i>A. terreus</i>	07	0.29	12	0.50	---	0.00	19	0.80
12.	<i>A. clavatus</i>	16	0.67	---	0.00	01	0.04	17	0.72
13.	<i>A. restrictus</i>	20	0.84	02	0.08	---	0.00	22	0.93
14.	<i>A. versicolor</i>	---	0.00	09	0.38	04	0.16	13	0.55
15.	<i>A. candidus</i>	27	1.14	---	0.00	05	0.21	32	1.35
16.	<i>A. nidulans</i>	02	0.08	07	0.29	---	0.00	09	0.38
17.	<i>A. citrisporus</i>	---	0.00	01	0.04	---	0.00	01	0.04
18.	<i>A. gramulosus</i>	19	0.80	22	0.93	05	0.21	46	1.95
19.	<i>A. sparsus</i>	01	0.04	---	0.00	12	0.50	13	0.55
20.	<i>A. elegans</i>	04	0.16	08	0.33	---	0.00	12	0.50
21.	<i>A. giganteus</i>	26	1.10	---	0.00	01	0.04	27	1.14
<b>Total</b>		<b>1154</b>	<b>49.02</b>	<b>810</b>	<b>34.40</b>	<b>390</b>	<b>16.56</b>	<b>2354</b>	

**References**

- Ainsworth, G.C. and G.R. Bisby. 1995. *Dictionary of the fungi*. Commonwealth Mycological Institute Kew, Surrey, pp. 445.
- Anonymous. 1997. *Aspergillus niger* Final Risk Assessment. Biotechnology program under toxic substances control act (TSCA) U.S. Environmental Protection Agency U.S.A. 3171.
- Coleman, J. 2004. *Indus River Delta, Pakistan, Asia*. In Darinage basin of the Indus River system, World Delta.
- Diana, W.F. 1994. *Soil biodiversity: its importance to ecosystem processes*. Report of a workshop held at the natural history museum, London, England.
- Gilman, J.C. 1957. *A manual of soil fungi*. The Iowa State University Press. Iowa USA.
- Gregory, S. and H.A. Thomas. 1997. Molecular Approaches to Controlling Cancer. The Importance of Fungi to Man. Baylor College of Medicine, Houston, Texas. *USA.Genome Research*, 7(11): 1041-1044.
- Mushtaq-ur-Rahman. 1975. A Geography of Sindh Province, Pakistan. Tha Karachi Geographers association (Ed.) Oxford University Press, Karachi,. 93-96.
- Raper, K.B. and C. Thom. 1945. *Manual of Aspergilli*. Williams and Wilkins Co. Baltimore, USA.
- Suhail, M., S. Akhund, T. Jatt, A.M. Mangrio and H. Abro. 2006. *Isolation and identification of Penicillium spp., from the bed of River Indus at kotri*. *Pak. J. Bot.*, 38(4): 1289-1292.
- Sultan, A., S.H. Iqbal and A.N. Khalid. 1997. *Fungi of Pakistan*. Dept of Botany, University of the Punjab, Quaid-e-Azam Campus, Lahore Pakistan. 248pp.
- Waksman, S.A. 1922. A method of counting the number of fungi in the soil. *J. Bact.*, 7: 339-341.
- Warcup, J.H. 1950. The soil plate method for isolation of fungi from soil. *Nature*, 166: 117-118.

(Received for publication 2 February 2007)