Pakistan Journal of Marine Sciences, Vol.3(2), 107-113, 1994

## STUDIES OF MICROBIAL ABUNDANCES IN MANGROVE HABITATS ALONG THE KARACHI COAST

## M. Jalaluddin

Department of Botany, University of Karachi, Karachi-75270, Pakistan.

ABSTRACT: Three strains of bacteria, 8 species of fungi and 3 species of VAM-fungi were isolated from the soil substrata supporting *Avicennia marina* which comprises the majority of mangrove vegetation along the Karachi coast. The species abundances for fungi and bacteria were greater at one site (Sandspit) supporting healthy mangrove growth with soil pH 7.8, EC 16.2 mmhos/cm<sup>2</sup>, TSS 2.57 percent and available phosphorus 0.008% than at the other site (Korangi creek) with stunted growth of mangrove where the soil samples showed pH 7.9, EC 18.8 mmhos/cm<sup>2</sup>, TSS 1.45% and available phosphorus 0.001%. Symbiotic association by vasicular-arbuscular mycorrhizal (VAM) fungi in the roots of mangrove plants was also observed on a small scale at Korangi creek where the substratum was undergoing microbial degradation.

KEY WORDS: Mangrove substratum - physiochemical characterisite - microorganisms - VAM-infection - mangrove plants.

### **INTRODUCTION**

Mangrove along the Karachi coast is comprised of a distinct mangrove community dominated by *Avicennia marina* L. The soil substrata of the mangrove are usually firm to soft mud with aerial roots (pneumatophores) emerging from the substrata for the aeration of mangrove plants. The substrata supporting mangrove communities range from rich to poor in nutrient content depending on the location of site.

Two sites supporting mangrove vegetation along the Karachi coast were studied. One site situated at Korangi creek had sparse mangrove vegetation seemingly undergoing deterioration due to the discharge of municipal and chemical effluents in the area. The other site at Sandspit supports a healthy mangrove community. Soil constituting the substrata of mangrove vegetation from the two sites were collected and studied in respect of soil properties such as pH, electrical conductivity (EC), total soluble salts (TSS), total and available phosphorus, soil microorganisms (fungi and bacteria) and vasicular-arbuscular mycorrhizal (VAM) infection in roots of mangrove vegetation.

#### **MATERIALS AND METHODS**

#### SOIL SAMPLES

Soil samples were collected from two sites of mangrove habitat at Karachi coast, Korangi creek and Sandspit, during the year 1992-93. The two sites were equally subdivided into four subsites of  $100 \text{ m}^2$  each. From each subsite, three replicate soil samples each of 200 g were collected randomly from the vicinity of at least three mangrove plants.

The collected soil samples were air dried to 5 per cent moisture level (De, 1990) and ground in a grinder (Moulinex-2000) for 15 seconds. Ground soils were sieved through 350  $\mu$ m mesh size sieve and stored at 5°C for later estimation of total and available phosphorus. Total and available phosphorus were determined colorimeteri-

cally by the method of Olsen *et al.* (1954). Soil pH, EC and TSS were determined by following Singh (1988).

## **ROOT SAMPLES**

Root samples of mangrove plants along with fine feeder roots were collected to determine VAM-infection according to Phillips and Hayman (1970). Collections were made from four subsites of the two sites as described above.

#### VAM-SPORES AND INFECTION

Wet sieving and decanting method was followed for the extraction of VAM-spores which were counted using the eelworm counting slide method (Anon, 1982) and identified based on morphology following Schenck and Perez (1990). Common soil fungi were isolated and counted by method of Waksman and Fred (1922) and identified with the help of key published by Barnett and Hunter (1972).

VAM-infection in mangrove roots was determined using Phillips and Hayman (1970) technique and the percentage of VAM-infection was assessed by the method of Giovannetti and Mosse (1980).

#### **RESULTS AND DISCUSSION**

Both higher bacterial and fungus populations were observed for the Sandspit site compared to the Korangi creek site (Figs.1 and 2). The highest number of bacteria  $(3.5 \times 10^5 g^{-1} \text{ soil})$  were recorded at a subsite of Sandspit whereas the lowest number of bacteria  $(1.5 \times 10^5 g^{-1} \text{ soil})$  were recorded at a subsite of Korangi creek (Fig.1). All the isolated bacteria were rod-shaped, (Figs. 3A-C). Gram-positive bacteria were dominant at Sandspit whereas Gram-negative bacteria were dominant at Korangi creek (Table I).

The highest population of fungi  $(321 \times 10^2 g^{-1} \text{ soil})$  was recorded for a subsite of Sandspit whereas a lowest population of fungi  $(128 \times 10^2 g^{-1} \text{ soil})$  was recorded for a subsite of Korangi creek (Fig.2). Altogether eight species of soil fungi were isolated from the two sites (Table II). Among the isolated fungal species, *Aspergillus niger* Van Tieghem was found to be predominant (Table II).

Colonies of fungi and bacteria were encountered at all four subsites of mangrove habitat but microbial populations were higher at the Sandspit site than at the Korangi creek site. This difference in the number of fungi and bacteria may be due to the physiochemical properties of the substrata (Table III) which were more condusive for the growth of mangrove vegetation at Sandspit than at Korangi creek. Schwartz *et al.* (1974) reported  $3.6 \times 10^6$  bacteria/g from the wet sediment of marine habitat. In saline soil, survival of microorganisms is difficult due to ex-osmosis in microbial cells (Austin, 1988). A higher population of Gram-positive bacteria at Sandspit and a higher population of Gram-negative bacteria at Korangi creek may be ascribed to the soil nutrient status of the substrata which was better at Sandspit and poorer at Korangi creek (Table III). My results suggest the survival of Gram-negative bacteria under lower nutrient conditions of the mangrove substratum. My results agree with Carlucci and Shimp (1974) who more frequently recovered Gram-negative bacteria from mangrove substrata at low levels of nutrient.

## Jalaluddin: Microbial abundances in mangrove habitats

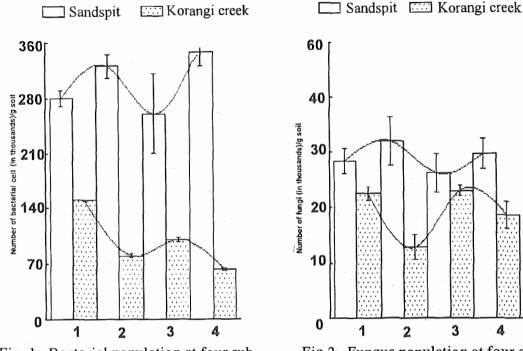


Fig. 1. Bacterial population at four subsites of Sandspit and Korangi creek of Karachi coast.

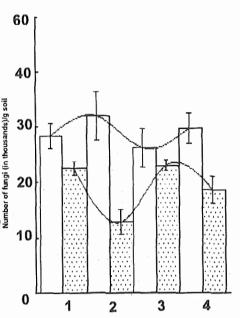


Fig.2. Fungus population at four subsites of Sandspit and Korangi creek of Karachi coast.

Table I: Cultural characteristics of the most abundant bacteria from mangrove substrata at Karachi coast.

Sub- sites		Sandspit		Korangi creek		
	Veg. cell	Gram reaction	Colony .characters	Veg. cell		Colony characters
1 2 3 4	Rod Rod Rod Rod	+ + - +	Transparent,slimy White,slimy Creamy,sputum like Pinkish,viscous	Rod Rod Rod Rod	- - -	White, watery Pinkish, undulated ends Transluscent, viscous Creamy round, viscous

Eight species of fungi and three species of VAM-fungi were isolated from the mangrove substrata at Karachi coast (Tables II and IV and Figs. 3D-F). Scanty VAM-infetion (5 to 8 per cent) in roots of A. marina was found at Korangi creek (Table IV). A maximum of 304 VAM-spores/100 g soil occurred in the soil substratum supporting mangrove vegetatation at both the sites of Karachi coast. Sengupta and Chaudhri (1990) isolated five species of VAM-fungi from a salt marsh sediment which had 204 VAM-spores/100 g soil. VAM-fungus species isolated by me from the saline soil substrata of Karachi coast differed from the salt marsh isolates of Sengupta and Chaudhri (1990). Khan (1974) isolated VAM-spores from saline soil supporting halophytic vegetation.

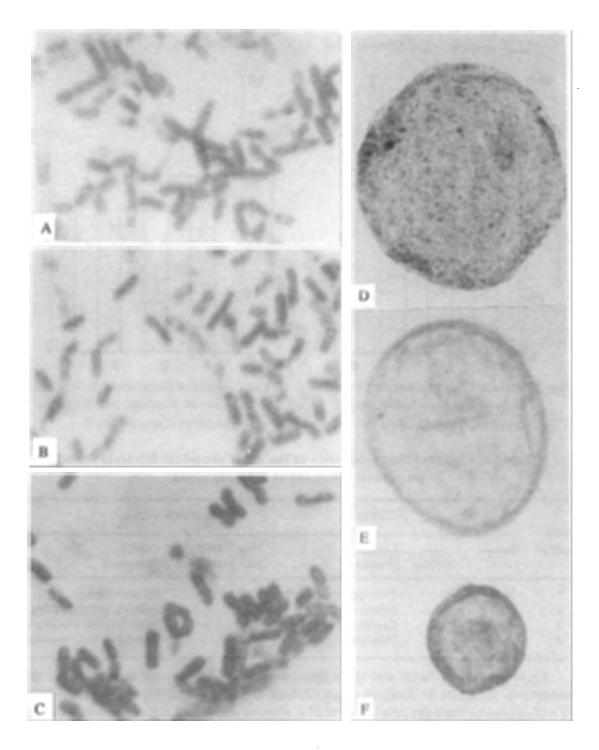


Fig.3. Rod-shaped bacterial cells (A-C) and VAM-spore (D-F) from rhizospheres of mangrove. A. Gram-negative bacterial cells from Korangi creek; B. Gram-negative bacterial cells from Sandspit; C. Gram-positive bacterial cells from Sandspit; D. VAM-spore (*Acaulospora gadanskensis*) from Sandspit; E. VAM-spore (*A. gadanskensis*) from Korangi Creek; F. VAM-spore (*Entrophospora* sp.) from Korangi creek. x 1000 for A-C; x 400 for D-F.

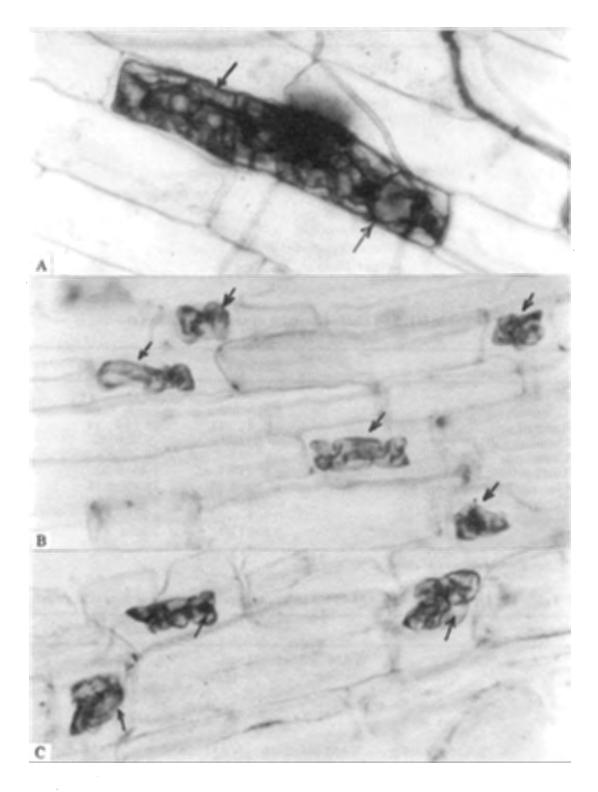


Fig.4. Intracellular VA-mycorrhizal fungi in cortical root tissues of mangrove at: (A) Korangi creek; (B) and (C) Sandspit. x 400.

Subsites	Sandspit	Korangi creek	
1	Aspergillus niger	A. flavus	
2	A. flavus Penicillium sp. A. niger	Cladosporium herbarum	
3	A. niger A. flavus	A. sulphurus Alternaria maritima	
4	Mucor mucedo Rhizopus varians Penicillium sp.	A. niger	

Table II. Isolated fungi from mangrove substrata at Karachi coast.

# Table II1. Soil characteristics of mangrove habitat.

Characteristics	Sandspit	Korangi creek	
Physical:	- - -	•	
Texture	Sandy loam	Sandy loam	
Ratio of soil (sand;silt;clay)*	63:19:18	67 : 19 : 14	
Ratio of soil (sand:silt:clay)* Moisture (at 110°C)	0.003%	0.01%	
Chemical:			
Total soluble salts (%)	$2.57 \pm 1.05$	$1.45 \pm 0.02$	
Electrical conductivity (mmhos/cm <sup>2</sup> )	$16.20 \pm 0.13$	$18.10 \pm 0.58$	
Total phosphorus (%)	$0.008 \pm 0.01$	$0.005 \pm 0.00$	
Available phosphorus	$0.003 \pm 0.001$	$0.001\pm0.00$	
pH	$7.80 \pm 1.02$	$7.90 \pm 0.89$	

\* Average of three replicate samples.

	Table IV. Occurrence of	VAM-fungi in mangrov	e substrata at Karachi coast.
--	-------------------------	----------------------	-------------------------------

Subsites	Sandspit		Korangi cree	k	
		VAM-species iso	lated		
1	Acaulospo		Entrophospora sp.		
2	A. mellea		Acaulospora gadanskensis		
3	Entrophospora sp.		A. mellea		
4	A. mellea	Ĩ	A. gadanski	х Х	
	VAM-spores/g	VAM-infection	VAM-spores/g	VAM-infection	
1	1	1.2%	2	5.2%	
2	3	1.5%	2	5.6%	
3	2	2.0%	3	8.7%	
4	1	0.1%	0	1.4%	

These halophytic plants were not mangrove plants and the VAM-fungi were not further identified. The higher percentage of VAM-fungus infection in *A. marina* at Korangi creek as compared to Sandspit may be due to the lack of nutrients at Korangi creek site. This deficiency of nutrition at Korangi creek site could be compensated for by the uptake of nutrients by means of symbiotic association of VAM-fungi. A greater degree of VAM-association at Korangi creek, with mangrove plants under nutrient stress, agrees with the findings of Sengupta and Chaudhari (1990) who observed VAM- infection in salt marsh halophytes growing under nutrient stressed condition which ordinarily do not have VAM-associations, reported to have by Sengupta and Chaudhri (1990).

#### ACKNOWLEDGEMENTS

The work was carried out with support from the Faculty of Science, University of Karachi (1992-93) which is gratefully acknowledged.

#### REFERENCES

Austin, B. 1988. Marine Microbiology. Cambridge University Press, London. Pp.216.

- Barnette, H.L. and B.H. Hunter, 1972. Illustrated Genera of imperfect fungi. 3rd Ed. Burgess Publishing Co., Menneapolis. Pp.241.
- Carlucci, A.F. and S.L. Shimp, 1974. Isolation and growth of a marine bacterium in low concentration of substrata. In: *Effect of ocean environment on microbial activities*. (Eds. Colvwell R.R. and R.Y. Morgta) University Park Press, Baltimore. Pp.363-367.
- De, S.K. 1990. Method of soil analysis. Narayan Publishing House, Allahabad, India. Pp.204.

Gerdemann, J.W. and T.H. Nicolson, 1963. Spores of mycorrhizal Endogone species extracted from soil by wet sieving and decanting. *Transaction British Mycological Society* 46: 234-235.

Giovannetti, M. and B. Mosse, 1980. An evaluation of techniques for measuring vesicular arbuscular mycorrhizal infection in roots. New Phytology 84: 489-500.

Khan, A.G. 1974. The occurrence of mycorrhizae in halophytes and xerophytes and of Endogone spores in adjacent soil. Journal of General Microbiology 81: 7-14.

Olsen, S.R., C.V. Cole, F.S. Watanable and L.A. Dean, 1954. Estimation of available "P" in soil by extraction with NaHCO3. United States Department of Agriculture No.939. Pp.19.

Phillips, J.M. and D.S. Hayman, 1970. Improved procedures for clearing and staining parasitic and vesicular arbuscular mycorrhizal fungi for rapid assessment of infection. *Transaction of British Mycological Society* 55: 158-161.

Schenck, N.C. 1982. Methods and principles of Mycorrhizal Research. The American Phytopathological Society, St. Paul, Minnesota. Pp.33.

Schenck, N.C. and Y. Perez, 1990. Manual for the identification of VA-mycorrhizal fungi. Synergistic Publications, Gainesville, USA. Pp.286.

Schwartz, J.R., J.D. Walker and R.R. Colmwell, 1974. Growth of deep sea bacteria on hydrocarbons at ambient and *in situ* pressure. *Development in Industrial Microbiology* 15: 239-249.

Sengupta, A. and S. Chaudhri, 1990. Vesicular arbuscular mycorrhiza (VAM) in pioneer salt marsh plants of the Ganges river delta in West Bengal (India). *Plant and Soil* 122: 111-113.

- Singh, L. 1988. Practical Agricultural Chemistry and Soil Science. Gajendra Singh Gahlot at Shiva Printers, Dehra Dun. India.
- Waksman, S.A. and B. Fred, 1922. A tentative outline of the plate method for determining the number of microorganisms in the Soil. Soil Science 14: 27-28.

(Received: 2 October 1993)