

## INFLUENCE OF TANNINS, AMINO ACIDS AND SUGARS ON FUNGI OF MARINE HALOPHYTES

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### ABSTRACT

The fungal load on leaves of 11 marine halophytes was studied along with the contents of tannins, sugars and amino acids in leaf tissues and their leakiness. The fungal load was more on leaf litter than that on fresh leaves. These higher densities on litter were found associated with lower content of tannins and sugars, higher content of amino acids, lower leakiness of tannins and sugars, and higher leakiness of amino acids.

*Key-words* : Mangroves, fungi, tannins, amino acids, sugars

Fungi are known to colonize leaves of marine halophytes either as 'phylloplane fungi' on fresh leaves or as 'litter fungi' (Kohlmeyer, 1984). These fungi are important in the marine environment as decomposers of dead organic substrates which serve as a source of detritus (Fell and Master, 1980). The fungal population depends on the interaction between the environmental matrix and the type of nutrient source available in the substrates (Kohlmeyer, 1984) which received very little attention. We have found earlier that phylloplane fungi are influenced by tannin content of leaves in seven marine halophytes (Sivakumar and Kathiresan, 1990). The present study deals with both phylloplane and litter fungi as influenced by amino acids, sugars and tannins in 11 marine halophytes.

Fresh leaf samples, third position from shoot apex, were collected from *Avicennia marina* (Forsk) Vierh., *A. officinalis* Linn., *Bruguiera cylindrica* (L.) Bl., *Ceriops decandra* (Griff) Ding Hou, *Lumnitzera racemosa* Willd., *Rhizophora apiculata* Blume, *R. mucronata* Lamk., *Salicornia brachiata* Roxb., *Sonneratia apetala* B. Ham, *Suaeda monoica* Forsk and *Xylocarpus granatum* J. Koen. from Pichavaram mangrove forest (Lat. 11°26'N; Long. 79°48'E), southeast coast of India, during August 1992. Leaf litter from the various species were also collected in bamboo baskets. The fresh leaves and litter were transferred to the laboratory in polythene bags and immediately used for fungal isolation.

Fungi from the upper surface of the leaf sample were isolated by using the leaf impression method on potato dextrose agar medium and incubated at 27±2°C and identified after three days. Areas of the leaves used for fungal isolation were imme-

diately measured using graph sheet and then analysed spectrophotometrically using 1% ninhydrin in acetone as a reagent for amino acids (Moore and Stein, 1948), using phenol-sulphuric acid method for sugars (Dubois, Gills, Hamilton, Rebers and Smith, 1951) and using 1% ferric chloride reagent for tannins (Hagerman and Butler, 1978). The chemicals were also estimated in the leachates obtained from the fresh leaf and litter samples allowed to leak for one hour in distilled water at  $28 \pm 2^\circ\text{C}$ . Three replicate samples were used for enumerating fungi and for the biochemical analyses.

The fungi *Aspergillus niger*, *A. flavus*, *Penicillium funiculosum* and *P. expansum* were isolated from both fresh leaves and leaf litter. *Mucor racemosus* was recorded only in leaf litter. Maximum number of fungi in fresh leaves was recorded from *Xylocarpus granatum* (149/100 cm<sup>2</sup> leaf area) and in leaf litter from *Sonneratia apetala* (939/100 cm<sup>2</sup> leaf area) (Table I). Both these plants are about to extinct in the Pichavaram mangrove forest. No fungi were isolated from fresh leaves of *Suaeda monoica* and *Lumnitzera racemosa*, but were recorded from their litter. Higher densities of fungi were generally recorded on leaf litter than that on fresh leaves. The number of fungal colonies ranged from 10.6 to 149 per m<sup>2</sup> area of fresh leaves and from 67.7 to 939 per m<sup>2</sup> area of leaf litter (Table I). The increase in fungal colonies ranged from 70 to 98% in litter as compared to fresh leaves. There was an average of 90% higher number of fungi in leaf litter than those on fresh leaves of 11 mangrove species. This implicates that the fungi are active in plant litter decomposition in the aquatic environment (Kohlmeyer and Kohlmeyer, 1979). The significance of occurrence of fungi on fresh leaf surface is not totally unknown (Preece and Dickinson, 1971). The fungal infestation is influenced by the exudations on the surface of leaves and their contents in leaf tissues (Tables II, III).

Table I – Fungal population in fresh leaves and leaf litter of marine halophytes (values are average of triplicate samples).

Plant Species	Number of fungi/100 cm <sup>2</sup> leaf area		% increase
	Fresh leaf	Leaf litter	
<i>A. marina</i>	9.0	293.8	97
<i>A. officinalis</i>	10.6	238.0	96
<i>B. cylindrica</i>	34.4	156.5	78
<i>C. decandra</i>	15.5	213.3	93
<i>L. racemosa</i>	0.0	449.0	—
<i>R. apiculata</i>	67.7	227.2	70
<i>R. mucronata</i>	10.9	166.1	93
<i>S. apetala</i>	16.0	938.5	98
<i>S. brachiata</i>	36.0	556.0	94
<i>S. monoica</i>	0.0	111.0	—
<i>X. granatum</i>	149.0	67.7	-55
Mean value	38.8	310.6	90
'F' Value Between			
	Samples (d.f.21,1)	3.9**	
	Plant species (d.f.21, 10)	2.3*	

\* Significant at P<0.05    \*\* Significant at P <0.01

Table II – Contents of tannins, sugars and amino acids in fresh leaves and litter of 11 marine halophytes (values are average of triplicate samples and those in parentheses show per cent increase or decrease in dead leaves over fresh ones).

Plant Species	Content in leaf tissue ( $\mu\text{g/g}$ dry weight)					
	Tannins ( $\mu\text{g}$ wattle tannin equivalent)		Sugars ( $\mu\text{g}$ D-glucose equivalent)		Amino acids ( $\mu\text{g}$ glycine equivalent)	
	Fresh	Litter	Fresh	Litter	Fresh	Litter
<i>A. marina</i>	9432	8470(-10)	11225	12517(+12)	668	3670(+449)
<i>A. officinalis</i>	8218	5011(-39)	14720	5572(-62)	1102	2443(+122)
<i>B. cylindrica</i>	40498	25224(-38)	19110	10778(44)	2767	5938(+115)
<i>C. decandra</i>	18663	4428(-76)	19093	4693(-75)	2086	1878(-10)
<i>L. racemosa</i>	62925	5986(-90)	17468	7253(-58)	788	1808(+129)
<i>R. apiculata</i>	20288	1186(-94)	22499	6001(-73)	1356	1701(+25)
<i>R. mucronata</i>	51768	2376(-95)	29335	4498(-85)	4024	1221(-70)
<i>S. apetala</i>	78441	12782(-84)	17543	20130(+15)	4464	9633(+115)
<i>S. brachiata</i>	50078	6981(-86)	21144	15987(-24)	2843	4505(+58)
<i>S. monoica</i>	102043	3759(-96)	41821	15410(-63)	5261	1684(-70)
<i>X. granatum</i>	23806	13278(-44)	37466	11049(-71)	2994	3364(+13)
Mean value	42378	8135(-80)	22857	10353(-55)	2578	3440(+33)
'F' Value Between						
Samples (d.f. 21, 1)	2.36*		6.44**		2.58*	
Plant species (d.f. 21, 10)	1.96*		5.30**		7.70**	

\* Significant at  $P < 0.05$     \*\* Significant at  $P < 0.01$

Table III – Leakiness of tannins, sugars and amino acids in fresh leaves and litter of marine halophytes (values are average of triplicate samples and those in parentheses show increase and decrease in leaf litter over fresh leaves).

Plant Species	Leakiness of biochemicals $\mu\text{g/g}$ dry sample/hour of leakiness in distilled water					
	Tannins ( $\mu\text{g}$ wattle tannin equivalent)		Sugars ( $\mu\text{g}$ D-glucose equivalent)		Amino acids ( $\mu\text{g}$ glycine equivalent)	
	Fresh	Litter	Fresh	Litter	Fresh	Litter
<i>A. marina</i>	5265	180(-97)	5516	2785(-50)	73	126(+73)
<i>A. officinalis</i>	7227	372(-95)	6556	1223(-81)	203	445(+119)
<i>B. cylindrica</i>	4476	155(-97)	3824	759(-80)	679	1547(+128)
<i>C. decandra</i>	1975	321(-84)	3733	1186(-68)	69	58(-16)
<i>L. racemosa</i>	7643	358(-95)	2107	1693(-42)	39	31(-21)
<i>R. apiculata</i>	4400	391(-91)	3416	2586(-24)	71	540(+661)
<i>R. mucronata</i>	10476	685(-94)	7832	7174(-72)	168	1068(+536)
<i>S. apetala</i>	10258	284(-97)	2632	810(-69)	118	199(+69)
<i>S. brachiata</i>	8624	345(-96)	5841	959(-82)	147	238(+62)
<i>S. monoica</i>	3406	488(-86)	3392	839(-74)	108	304(+181)
<i>X. granatum</i>	7227	110(-99)	4095	5199(+27)	92	320(+248)
Mean value	6453	335(-93.7)	4450	1837.5(-59)	161	443(+185)
'F' Value Between						
Samples (d.f. 21, 1)	6.89**		0.28*		2.96**	
Plant species (d.f. 21, 10)	2.19*		0.48**		1.89*	

\* Significant at  $P < 0.05$     \*\* Significant at  $P < 0.01$

In general, tannin and sugar contents were lower in leaf litter than those in fresh leaves (Table II). There was an average decrease of 80% tannins and 55% sugars in leaf litter as compared to those in fresh leaves of 11 mangrove species. These low levels of sugars and tannins in litter (Table II) may be attributed to the rapid leaching of soluble organics (Table III) and also to the absence of photosynthetic production of sugars in leaf litter. However, the opposite was true with amino acid contents which was higher by 33% in leaf litter than fresh leaves as a result of increased microbial biomass or proteolysis in the litter (Table II). Thus, the fungal population appears to be associated with high level of amino acids and low levels of tannins and sugars in leaf litter.

It is interesting to note that fungi were not isolated from fresh leaves of *Suaeda monoica* and *Lumnitzera racemosa* (Table I) having the high tannin content (Table II). But fungi were isolated from the same plant species in the leaf litter which contained low tannin content by about 90% (Table I). Tannins are well known for antifungal activity (Lakshmanan, 1986). Leaf tannin content was found to have a negative correlation of 0.95 with fungal load on mangrove leaves (Sivakumar and Kathiresan, 1990).

Like the contents, leakiness of tannins and sugars was lower in leaf litter than that in fresh leaves. There was an average decrease of 94% tannins and 59% sugars in leaf litter as compared to those from fresh leaves of 11 mangrove species. However, the opposite was true with amino acid leakiness which was higher by 185% in leaf litter than fresh leaves, which might provide nitrogen to the growing fungal population. Thus the fungal population appears to be associated with higher leakiness of amino acids and lower leakiness of tannins and sugars from leaf litter.

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