RESEARCH ARTICLE

ADAPTIVE VARIABILITY IN LEAF TRAITS OF MANGROVE FERN ACROSTICHUM AUREUM L. IN RELATION TO ECOLOGICAL VARIATIONS

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Abstract: The macro and micro morphological traits of lamina (length and maximal width, the ratio of length & width (L: W), area, and stomatal index) are studied in the mangrove fern *Acrostichum aureum* L. (golden leather fern) grown naturally in mangrove and non-mangrove regions in West Bengal, India. Jharkhali, and Bhagbatpur are two selected sites in mangrove region of Indian Sundarbans from where the mature leaves are collected for study. Besides, the collection sites also include three non-mangrove regions namely, E.M. Bypass, and Garia of South 24 Parganas district and Majherchar, Kalyani of Nadia district of West Bengal. Individual parameter has been assessed and statistically analyzed. One-way analysis of variance (ANOVA) test significant variations in most of the leaf traits between and among the sites, but the percentage of variations in stomatal indices are less which signifies more stability. Results suggest that ecological conditions measured through soil attributes have some important role in controlling leaf traits adaptability.

Keywords: Indian Sundarbans, E.M. Bypass, Garia, Majherchar, Kalyani, macro and micro morphology, ANOVA

INTRODUCTION

Adaptive mechanisms of water use efficiency by mangrove plants have enormous importance in human welfare (Tomlinson 1986; Kathiresan 2012). As of conservation priorities, explicit design of experimental studies is performed to generate results that can be applied to emerging global problems; however, reports are rather meagre about the influence of habitat fragmentation on adaptive measures of mangrove plants (Ellison 2000, 2004; Hazra *et al.* 2002; Mitra *et al.* 2014). As such, there are ample scopes of research to explore the uncertainties of how component traits can be integrated into whole plant phenotypes to combat the water stress. Furthermore, the plasticity of mangroves enables them to colonize in a wider range of habitats (Tomlinson 1986).

The present research work has been designed to determine phenotypic variations in macro and micro-morphological leaf structural traits (length, width, area, length: width ratio, and stomatal index) of mangrove fern *Acrostichum aureum* L., commonly known as golden leather fern, grown along the gradients from sea to land that is both in mangrove and non mangrove zones. The objective of the work is to ascertain the key trait(s) for adaptation. Further, the leaf structural attributes have been compared with corresponding soil attributes to find out the relationships between the variables offering scope for ecological adaptations.

MATERIAL AND METHOD

A total of five study sites (Fig. 1a), both from mangrove regions of Indian Sundarbans (Jharkhali, Bhagabatpur) and non-mangrove regions (Garia, E.M. Bypass of South 24 Parganas and Majherchar, Kalyani, Nadia) have been chosen for collection. At least 50 matured pinnae collected from *A. aureum* plants (Fig. 1b) having almost same height (1.5-2.0m) representing same age of the plants were assessed.

The attributes studied are pinna length (PL), maximal pinna width (PW), length and width ratio (L:W), pinna area (PA), and stomatal index (SI). For studying the first four traits, the pinnae were placed on graph paper and measurements were taken. For stomatal analysis, the pinnae were subjected to standard technique by using 10% potassium hydroxide (KOH) to peel off the cuticle and the preparations were observed under compound microscope. Microscopic study and microphotographs were performed by Leitz Laborlux S trinocular compound microscope with attached Leica EC3 scientific camera. The count of stomatal cell (S) and epidermal cell (E) were done from at least ten microscopic fields. The SI is calculated by using the formula [(S / (S +E)) *100] as suggested by Salisbury (1927). Individual parameter was assessed between and among the study sites by statistical tool, ANOVA, considering $P<0.05$ level of significance.

To observe the ecological variations in study sites, collected soil samples were analysed considering the attributes of salinity (EC in mS/cm), pH, percentage of carbon (%C), and organic matter (%OM).

RESULT AND DISCUSSION

The occurrence of *A. aureum* is rather not restricted to mangrove ecosystem only as it can survive in non-saline conditions of non-mangrove regions (present study and Jayatissa et al. 2002; Medina et al. 1990; Sun et al. 1999; Taylor 1986; Tomlinson 1986).

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Leaf attributes

The observed data show wide range of variations in all the studied traits of pinnae (Table 1; Figs. 1b-1e). Mean values of mature pinnae length range from 23.19 cm ± 0.94 to 13.8 cm ± 1.53. Maximum pinnae length is noted from collection of Garia (23.20 cm ± 0.943) followed by Jharkhali (22.45 cm ± 2.14), E.M. Bypass (20.30 cm ± 1.27), Bhagabatpur (19.51 cm ± 2.73), and Majherchar (13.80 cm ± 1.53). Pinnae width is calculated highest (3.13 cm ± 0.30) in lamina of plants of E.M. Bypass. The differential variations in length and width ratio are also reflected in observations. The ratio of L:W is measured utmost in the pinnae of Garia (12.47 ± 1.94) followed by other sites of collection. Interestingly, though the pinnae of the region Garia possesses highest length and L:W ratio, it shows lower laminal area. Lamina area of pinnae collected from Majherchar (30.40 cm^2 ± 3.83) is more or less similar to that of Garia. Such observation is important in term of photosynthetic capacity. The most expanded laminal area is observed in the plants of E. M. Bypass region (74.80 cm^2± 7.33 cm). The anomocytic stomata (Fig. 1f) recovered in the pinna of various study sites also vary in calculation of stomatal index which is ranges from 49.33 ± 0.18 to 24.01 ± 2.86 of which pinnae of Jharkhali show highest value (49.33±0.18).

Statistical analysis of leaf attributes

Individual leaf trait assessed following one-way ANOVA (Probability level 0.05) reveals significant variations mostly between sites (Table 1). By analysing the results through ANOVA test (Tables 2; Fig. 1g), it reveals that the relationships of all the macro- and micro- morphological leaf traits (pinna length, width, length: width, area, stomatal index) between and among the collection sites show higher percentage (78.0%) of significantly variable traits than non-significantly variable (22.0%) traits when measured collectively. Individual macro-morphological trait i.e. pinna length, width, area, length and width ratio are found to show 80.0%, 70.0%, 90.0%, 90.0% variations respectively; it is 60.0% in micro-morphological trait (stomatal index) indicating more plasticity in macro-morphology than micro-morphology.

Soil attributes

The soil samples collected from different studied sites are found to vary considerably in various attributes (Table 3). Among the soil attributes, salinity differs markedly in the sites whereas soil pH is less variable (7.46 to 8.58). The salinity ranges from 6.58 mS/cm in Majherchar to 82.4 mS/cm in Jharkhali which is well in conformity with the geographical location of the sites (Figure 1a). The salinity in remaining three areas i.e. Bhagabatpur, Garia, and E.M. Bypass are 45.7, 30.1, and 14.4 mS/cm respectively also corroborate with the successive remoteness from seaward to landward. Such trend is not reflected while calculating the organic matter and carbon content of soils in collection sites. The percentage of OM (9.62%) and C (5.58%) are measured highest in Garia and the consecutive values are 3.72% and 2.16% in Majherchar, 2.59% and 1.50% in E.M. Bypass, 1.76% and 1.02% in Jharkhali, and 1.50% and 0.87% in Bhagabatpur. This result suggests that the nearness or remoteness of a locality from sea is not related with the organic matter and carbon content.

Comparison of leaf traits with soil attributes

The studied leaf traits indicate that the variations in macro-morphological traits are not mangrove or non mangrove zone specific (Table 1). By comparing the observations of leaf traits with that of soil attribute it can be suggested that the macro-morphological variations observed in *A. aureum* are more dependent on OM and C content than the soil salinity. However, the studied micro-morphological trait that is stomatal index is likely to be dependent on salinity as the values of SI are directly proportional to the values of salinity in the collection sites i.e. Majherchar<E.M. Bypass<Garia<Bhagabatpur<Jharkhali.

Table 1. Mean value of studied leaf traits, value wise site ranges, and C.D. value analysed through (ANOVA) of *Acrostichum aureum* L. collected from varying habitats

Leaf Traits	Mean values in different collection sites					Value wise site ranges	C.D. value at 5% level
	J	BH	G	E	M		
PL (cm)	22.45± 2.14	19.51 ± 2.73	23.19± 0.94	20.30 ± 1.27	13.80 ± 1.53	M<B<E<J<G	**1.41**
PW (cm)	2.52 ± 0.47	1.84 ± 0.168	1.86 ± 0.34	3.13 ± 0.30	1.80 ± 0.31	M<B<G<J<E	**0.26**
L: W	8.91 ± 1.78	10.60 ± 1.50	12.47± 1.94	6.49 ± 0.30	7.67 ± 1.17	G<M<B<J<E	**1.14**
PA (cm^2)	49.80± 10.90	31.80 ± 3.42	29.20± 4.49	74.80 ± 7.33	30.40 ± 3.83	E<M<J<B<G	**6.55**
SI	49.33± 0.18	49.25 ± 0.34	47.21± 0.48	45.06 ± 3.13	24.01 ± 2.86	M<E<G<B<J	**2.52**

Pinna Length **[PL]**; Pinna Width **[PW]**; L. Length/L. Width **[L: W]**; Pinna area **[PA]**; Stomatal Index **[SI]**; Jharkhali **[J]**; Bhagabatpur **[BH]**; Garia **[G]**; E. M. Bypass **[E]**; Majherchar **[M]**

Table 2. Total score and percentage of significantly variable traits (SVT) and non-significantly variable traits (NSVT) as analysed through ANOVA result

Correlation between sites	Scores of SVT	Name of SVT	Scores of NSVT	Name of NSVT
J/BH	4	PL, PW, L: W, PA	1	SI
J/G	3	PW, L: W, PA	2	PL, SI
J/E	5	PL, PW, L: W, PA, SI	0	NA
J/M	5	PL, PW, L: W, PA, SI	0	NA
BH/G	3	PL, L: W, PA	2	PW, SI
BH/E	4	PW, L: W, PA, SI	1	PL
BH/M	4	PL, L: W, PA, SI	1	PW
G /E	4	PL, PW, L: W, PA	1	SI
G/M	3	PL, L: W, SI	2	PW, PA
E/M	4	PL, PW, PA, SI	1	L: W
Total Scores & percentage of variations	**39 (78%)**		**11 (22%)**	
	PL : 8 (80%)		PL : 2 (20%)	
	PW : 7 (70%)		PW : 3 (30%)	
	PA : 9 (90%)		PA : 1 (10%)	
	L: W : 9 (90%)		L: W : 1 (10%)	
	SI : 6 (60%)		SI : 4 (40%)	

Pinna Length **[PL]**; Pinna Width **[PW]**; L. Length/L. Width **[L: W]**; Pinna area **[PA]**; Stomatal Index **[SI]**; Jharkhali **[J]**; Bhagabatpur **[BH]**; Garia **[G]**; E. M. Bypass **[E]**; Majherchar **[M]**

Table 3. Data of soil attributes collected from different study sites

Collection Sites	Soil attributes			
	Percentage of C (%)	Percentage of OM (%)	Salinity (mS/cm)	pH
Jharkhali	1.02	1.76	82.4	7.58
Bhagabatpur	0.87	1.50	45.7	8.58
Garia	5.58	9.62	30.1	7.58
E. M. Bypass	1.50	2.59	14.4	8.10
Majherchar	2.16	3.72	6.58	7.46

[C: Carbon; OM: Organic matter]

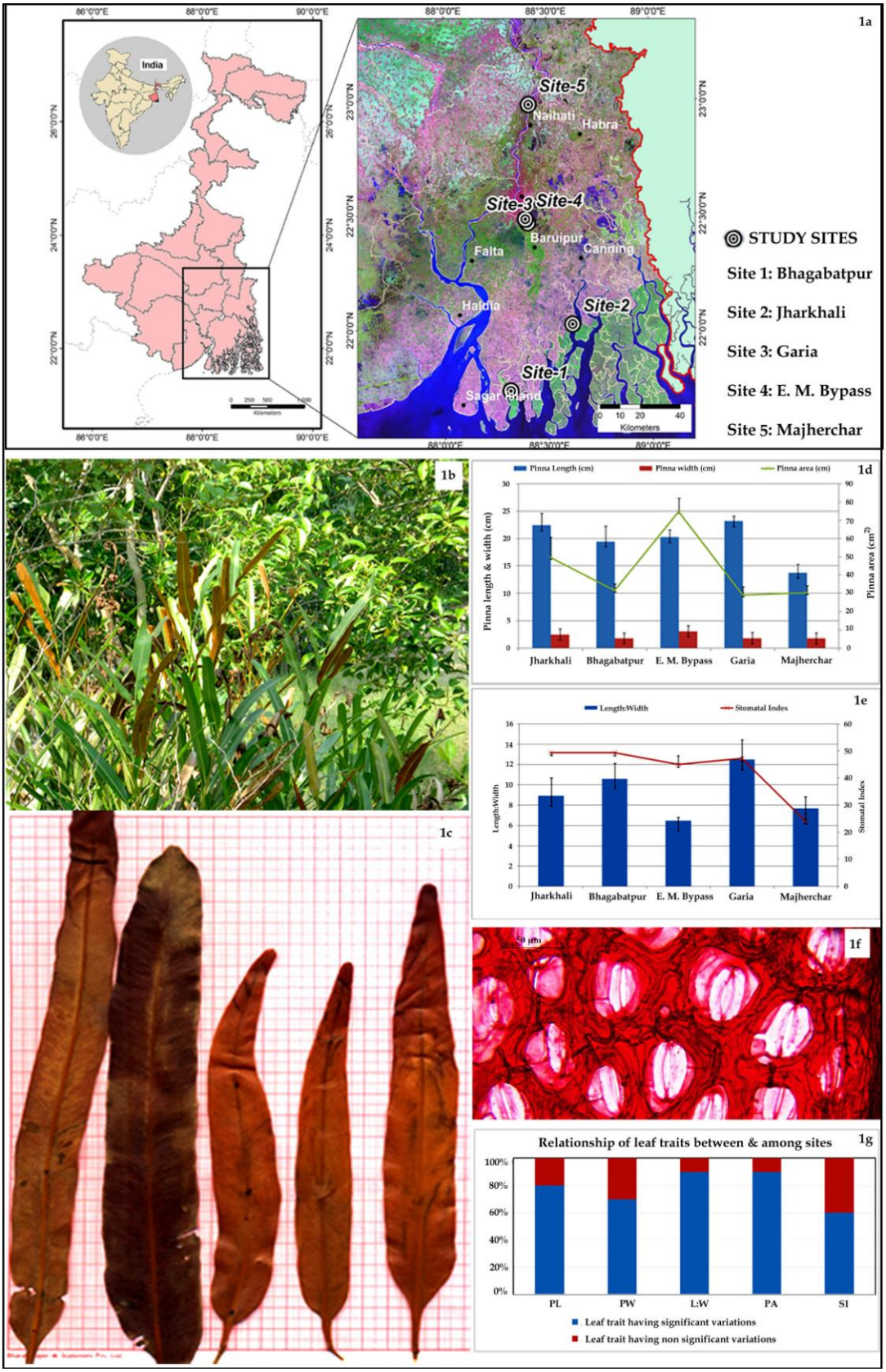

Figure Legends

Fig. 1 (a) Map of West Bengal showing study sites; (1b) golden leather mangrove fern *Acrostichum aureum* L.; (1c) matured pinnae of the plant; (1d-e) graphical representation of variations in macro- and micro-morphological leaf traits (d) traits: pinna length, width, area (e) traits: length & width ratio, stomatal index; (1g) graphical representation of percentage of variations in studied leaf traits as analysed through ANOVA result

CONCLUSION

The golden leather fern *Acrostichum aureum* L., commonly grown in mangrove habitat faces a succession of ecological challenges when they grow and develop in non-mangrove habitats also. The plant gets equipped with an adaptive plasticity in the phenotypic expression as revealed through various leaf traits which afford a kind of biological insurance or flexibility to varying environments.

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