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Recognition of plant associations useful for conservation of *Ophioglossum nudicaule* L.f. and *Ophioglossum vulgatum* L. in the eastern lateritic part of India

Sumit Manna[1] and Anirban Roy[2]*

[1]West Bengal Biodiversity Board, Department of Environment, Government of West Bengal. Paribesh Bhawan, 10 A, LA Block, Sector- III, Saltlake City, Kolkata – 700098 West Bengal. India

[2]West Bengal Biodiversity Board, Department of Environment, Government of West Bengal. Paribesh Bhawan, 10 A, LA Block, Sector- III, Saltlake City, Kolkata – 700098 West Bengal. India

*Corresponding Author E-mail: aroy.wbbb@gmail.com

ABSTRACT

For the past few decades, populations of different species of *Ophioglossum* (Ophioglossaceae) including *Ophioglossum nudicaule* and *Ophioglossum vulgatum* have become more restricted in geographic range due to alteration of their actual or potential habitat conditions and therefore are designated as rare species of India. To find out the potential habitats of *Ophioglossum* for protection of their ancient gene pool, it is important to identify their plant associates as well as the nature of those associations. Structural parameters of the community such as density, abundance, frequency, relative abundance, relative frequency and importance value index of these two species of *Ophioglossum* and their co-existing plants were measured based on a quadrat study of three tropical deciduous forests of the lateritic part of West Bengal, India. This study reveals that there is a strong negative correlation in the association of *O. nudicaule* and *O. vulgatum*. Among the other associates, *Lindernia antipoda* (Scrophulariaceae), *Cyperus rotundus* (Cyperaceae) and *Phyllanthus niruri* (Euphorbiaceae) are positively associated with *O. nudicaule* where as *Phyllanthus niruri*, *Lindernia antipoda* and *Dioscorea bulbifera* (Dioscoreaceae) have strong negative association with *O. vulgatum*. These findings would help in *ex situ* and *in situ* conservation programs for both species of *Ophioglossum* through maintaining populations of *Lindernia antipoda*, *Cyperus rotundus*, and *Phyllanthus niruri* with *O. nudicaule* and removal of populations of *Lindernia antipoda*, *Phyllanthus niruri* and *Dioscorea bulbifera* found in association with *O. vulgatum*. It is also suggested that these two species of *Ophioglossum* be grown separately to maximize their population sizes.

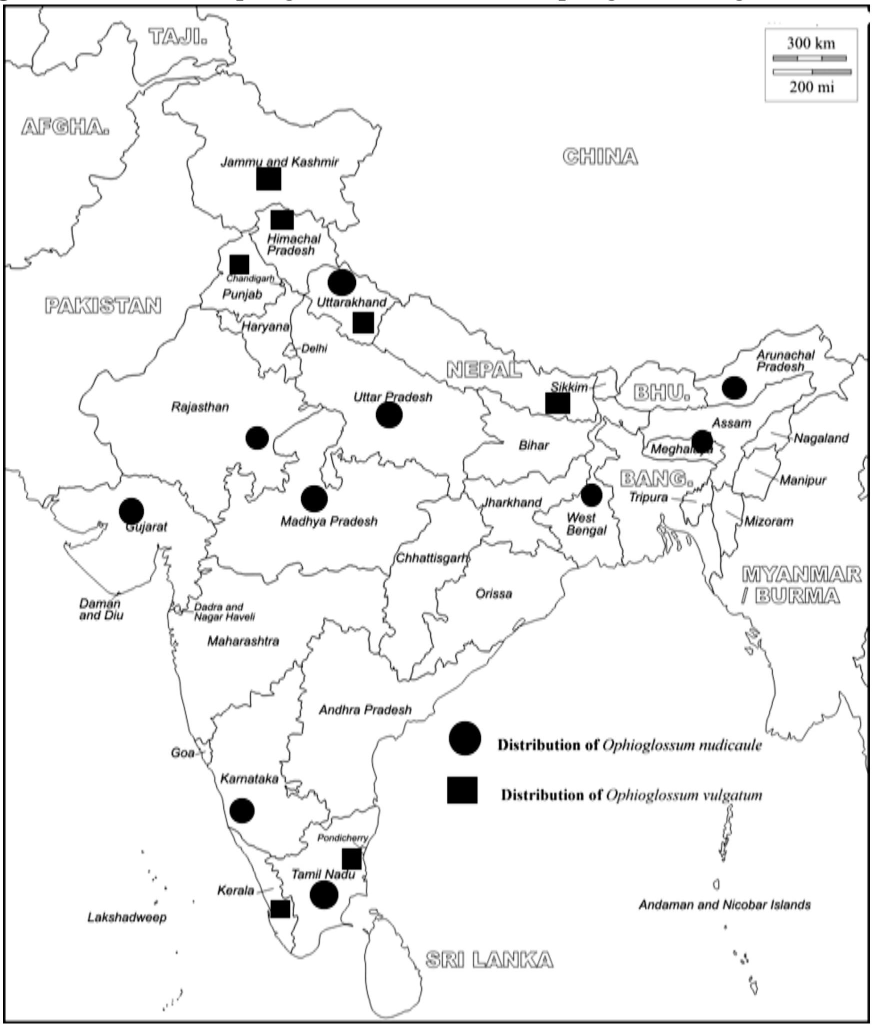
Key word: *Ophioglossum, plant association, co-existence, conservation, phytoassemblage.*

INTRODUCTION

Species of *Ophioglossum* are distributed all over the world, inhabiting a range of climatic conditions and habitat conditions including calcareous soil barrens, limestone glades, dry limestone, dolomite prairies, savannas, rock openings of upland forests on dolomite bogs, fens, damp sand, pastures, wet meadows, grassy swales, moist woods, rich swamplands, mud creeks, and cedar swamps [1-8]. Some *Ophioglossum* species also inhabit dry, sandy beaches or hillsides and the subterranean gametophytes may have adapted to seasonal drying and fire. *Ophioglossum vulgatum* L. and *Ophioglossum nudicaule* L. f. are restricted to moist mineral-humic mesotrophic soils with neutral or alkaline pH, moderate light and warm climatic conditions. *Ophioglossum vulgatum* also tolerates relatively high NaCl content [10]. Out of 40 species of *Ophioglossum* in the world only 12 of them have distributions that include India [11-15]. In India, *O. vulgatum* and *O. nudicaule* grow in only a few areas and have distinct habitat preference. *Ophioglossum vulgatum* was found to be restricted to a few populations in Sikkim, Uttarakhand, Kashmir, Himachal

Pradesh, Punjab, Kerala and Tamilnadu whereas *O. nudicaule* was reported from Madhya Pradesh, Meghalaya, Arunachal Pradesh, Uttarakhand, West Bengal, Uttar Pradesh, Gujarat, Rajasthan, Karnataka and Tamilnadu (Fig. 1)[16,17]. In order recommend ways to conserve these two rare species of *Ophioglossum* and to increase their population sizes, we use phytoassemblage analysis of *Ophioglossum* and co-existing plant associates in order to identify and conserve strong positive associates and to remove strong negative associates.

Fig. 1. Distribution of *Ophioglossum nudicaule* and *Ophioglossum vulgatum* in India



MATERIALS AND METHODS

Study area

The eastern lateritic part of India is one of the preferred habitats for many novel species like *Ophioglossum vulgatum, O. nudicaule, Drosera indica* L.*, Drosera burmannii* Vahl*, Utricularia bifida* L.*, Euphorbia fusiformis* Buch.-Ham. ex D. Don etc. which are restricted in few pockets throughout the world. Heavy rainfall in this area promotes luxuriant growth of these species during monsoon months (July to September).

A reconnaissance survey was done in different parts of forests to identify the habitats of *O. vulgatum* and *O. nudicaule* and their other plant associates. The study sites are located in regenerated and managed *Shorea robusta* Gartn. dominated forest. Three study sites were selected mainly on the basis of pedological condition and vegetation pattern. The Ghaga forest of western part of the state West Bengal, India (24º 04' 57.52" N 87º 40' 43.93" E) was selected as site 1 and was comprised of laterite soil admixed with sand and older alluvium[18]. Trees like *Shorea robusta* C.F. Gaertn*, Madhuca latifolia* J.F. Macbr*, Buchanania lanzan* Spreng.*, Semecarpus anacardium* L.f.*, Gardenia latifolia* Schlecht. ex Hook.f. were important components of the canopy and reduced light intensity at ground level (322.16 Lx st.dev.

200.59). The Ulpahari forest in the western part of West Bengal, India (24º 04' 14.90" N 87º 39' 18.35" E) was selected as site 2 and was composed of laterite to red soil with the similar forest composition of site 1 with the addition of some climbers and lianas (*e.g. Butea superba* Roxb., *Aristolochia indica* L., *Cissus adnata* Roxb., *Ichnocarpus frutescens* R. Br., *Ventilago denticulata* Willd.) which also contributed to the reduced light intensity at ground level [(157. 66 Lx st. dev. 91.15)[19]]. The Choupahari forest in the western part of West Bengal, India (23º 37' 44.98" N 87º 34' 57.53" E) was selected as site 3 which was located 13 km from the river Ajoy and comprised of older to younger alluvium. It has a floristic composition similar to that of site 2 except for less canopy cover by climbers and lianas on the trees resulting in comparatively greater light penetration at ground level (415.5 Lx st.dev. 203.37).

Methods

One voucher specimen of each species of *Ophioglossum* and of all other plants within the area of the study site where these pteridophytes were found were collected, dried and preserved following standard field procedures [20]. The specimens were identified with the help of standard taxonomic keys, references [21, 22] and by comparison with specimens deposited at Central National Herbarium (CAL). The voucher specimens were deposited at the same Herbarium (CAL).

For community analysis, eight sampling units (SUs = quadrats 0.304 m x 0.304 m) were located in each of the three selected forest sites. Quadrats were placed randomly in each site to avoid biased sampling. Parameters such as density (D), abundance (A), frequency (F) %, relative density (RD), relative frequency (RF) and importance value index (IVI, the summation of relative density and relative frequency of each species) were estimated by using standard procedures[23, 24]. To study the inter-specific associations (if any) between the two target species of *Ophioglossum* and the 20 other plant species present in the community, a 2 x 2 contingency/species-association table (Fig. 2) was prepared from the presence-absence data matrix for pair wise comparisons [25]. The presence-absence data matrix was derived from the data of 24 sampling units (quadrats) and were used for structural analysis of *Ophioglossum* and other plant associates in the three study sites. Large numbers of sampling units (24) were taken throughout the three patches to avoid biased chi-square values as much as possible (Eq. 1 of Table 1). For further continuity a correction to ensure a closer approximation to the theoretical continuous Chi-square distribution, Yates's correction formula (Eq. 2 of Table 1) was adopted. Association was measured by computing Ochiai (*OI*), Dice (*DI*) and Jaccard (*JI*) Indices (Eq. 3, 4 & 5 respectively of Table 1)[25].

Fig. 2. Structure of 2 x 2 contingency/species-association table.

		Species B		
		Present	Absent	
Species A	Present	a	b	m = a+b
	Absent	c	d	n = c+d
		r = a+c	s= b+d	

a= the number of SUs where both species occur, b = the number of SUs where species A occurs but not B, c = the number of SUs where species B occurs but not A, d = the number of SUs where neither A nor B are found and N = the total number of SUs (N = a+b+c+d)

Table. 1 Equation used for association study between two species of *Ophioglossum* and twenty other co-existing plants.

Equation Number	**Name of the Equation**	**Equations**
1	Chi-square	$\chi_c^2 = \frac{N(ad-bc)^2}{mnrs}$
2	Yates's correction	$\chi_c^2 = \frac{N[\lvert(ad)-(bc)\rvert-(N/2)]^2}{mnrs}$
3	Dice (*DI*) Index	$DI = \frac{2a}{2a+b+c}$
4	Jaccard (*JI*) Index	$JI = \frac{a}{a+b+c}$
5	Ochiai (*OI*) Index	$OI = \frac{a}{\sqrt{a+b}\sqrt{a+c}}$

a= the number of SUs where both species occur, b = the number of SUs where species A occurs but not B, c = the number of SUs where species B occurs but not A, d = the number of SUs where neither A nor B are found and N = the total number of SUs (N = a+b+c+d)

RESULTS

Two species of *Ophioglossum* (*O. vulgatum* and *O. nudicaule*) were found to co-exist with 20 other plant species of the moist forest floor covered with dense mat of mosses (Table 2, 3 and 4). Of the 20 plant species associated with these two species of *Ophioglossum*, 15 species were common to all these three study sites (Tables 2, 3 & 4). Structural parameters of the community of these two species of *Ophioglossum* and other associated plants revealed that both species of *Ophioglossum* were not found at a high relative density, abundance or relative frequency at a single site (Table 2, 3 & 4). In site 1, *O. nudicaule* showed its highest importance value index in contrast to sites 2 and 3 where *O. vulgatum* showed its peak importance value index (Fig. 3). Of the 20 plant species co-existing with these 2 species of *Ophioglossum*, *Lindernia antipoda* (L.) Alston had its highest importance value index followed by *Mazus pumilus* (Burm. f.) Steenis and *O. nudicaule* in site 1 whereas *Mazus pumilus* followed by *Ophioglossum vulgatum* represented their extreme peak of importance value index in site 2 and 3 (Fig. 3 & 4). It is very interesting to note that *Lindernia antipoda*, with its highest importance value index in site 1 was totally absent in site 2 and 3. Pair-wise association analysis between two species of *Ophioglossum* and other 20 co-existing plants (Table 5) indicated a true negative association between *O. vulgatum and O. nudicaule* (at 10% probability level). *Ophioglossum nudicaule* also showed the possibility of a strong negative association with *Lygodium flexuosum* (L.) Sw. (at 0.1% probability level). Strong positive association with *Lindernia antipoda* (at 1% probability level), *Cyperus rotundus* L. (at 5% probability level) and *Phyllanthus niruri* L. (at 10% probability level) was also observed to *O. nudicaule* (Fig. 5). On the other hand there does seem to be some strong possibilities of true negative association between *O. vulgatum* and *Lindernia antipoda* (at 1% probability level) followed by *Phyllanthus niruri* and *Dioscorea bulbifera* L. (at 10% probability level) (Fig. 6).

Table 2 Structural parameters of two species of *Ophioglossum* and 20 other co-existing plants in Site 1.

Name of the Species (Family)	NO.	D	RD	FQ%	AB	RF	IVI
Study species							
Ophioglossum nudicaule L. f. (Ophioglossaceae)	1	285.33	29.29	87.50	30.00	10.30	39.60
Ophioglossum vulgatum L. (Ophioglossaceae)	2	10.87	1.12	62.50	1.60	7.40	8.50
Co-Existing Species							
Pteridophytes							
Lygodium flexuosum (L.) Sw. (Lygodiaceae)	5	2.72	0.28	12.50	2.00	1.50	1.70
Selaginella rupestris (L.) Spring (Selaginellaceae)	17	0.00	0.00	0.00	0.00	0.00	0.00
Graminoids							
Cyperus rotundus L. (Cyperaceae)	12	21.74	2.23	87.50	2.29	10.30	12.50
Eragrostis tenella (L.) Roem. & Schult. (Poaceae)	15	1.36	0.14	12.50	1.00	1.50	1.60
Forbs							
Antidesma ghaesembilla Gaertn. (Euphorbiaceae)	18	0.00	0.00	0.00	0.00	0.00	0.00
Cissus adnata Roxb. (Vitaceae)	21	0.00	0.00	0.00	0.00	0.00	0.00
Commelina benghalensis L. (Commelinaceae)	19	0.00	0.00	0.00	0.00	0.00	0.00
Curculigo orchioides Gaertn. (Hypoxidaceae)	8	4.08	0.42	25.00	1.50	2.90	3.40
Cyanotis axillaris (L.) Sweet (Commelinaceae)	22	0.00	0.00	0.00	0.00	0.00	0.00
Desmodium triflorum (L.) DC. (Fabaceae)	9	8.15	0.84	62.50	1.20	7.40	8.20
Dioscorea bulbifera L. (Dioscoreaceae)	11	2.72	0.28	25.00	1.00	2.90	3.20
Drosera burmannii Vahl (Droseraceae)	16	1.36	0.14	12.50	1.00	1.50	1.60
Evolvulus alsinoides L. (Convolvulaceae)	7	2.72	0.28	25.00	1.00	2.90	3.20
Hemidesmus indicus (L.) R.Br. (Asclepiadaceae)	4	1.36	0.14	12.50	1.00	1.50	1.60
Ionidium suffruticosum DC. (Violaceae)	13	1.36	0.14	12.50	1.00	1.50	1.60
Lindernia antipoda (L.) Alston (Scrophulariaceae)	14	302.99	31.10	100.00	27.88	11.80	42.90
Mazus pumilus (Burm. f.) Steenis (Scrophulariaceae)	3	281.25	28.87	100.00	25.88	11.80	40.60
Oldenlandia corymbosa L. (Rubiaceae)	6	27.17	2.79	75.00	3.33	8.80	11.60
Phyllanthus niruri L. (Euphorbiaceae)	10	12.23	1.26	75.00	1.50	8.80	10.10
Ziziphus oenoplia (L.) Mill. (Rhamnaceae)	20	1.36	0.14	12.50	1.00	1.50	1.60

D/Sq. mt.: Density/Square meter; RD: Relative Density; Ab: Abundance; F%: Frequency; RF: Relative Frequency; RBA: Relative Basal Area; IVI: Importance Value Index; No. refers to the unique identifying number assigned to the species for analysis and which will be used in Table 5 and the Plexus diagrams in Figures 5 and 6.

Table 3 Structural parameters of two species of *Ophioglossum* and 20 other co-existing plants in Site 2.

Name of the Species (Family)	NO	D	RD	FQ%	AB	RF	IVI
Study species							
Ophioglossum nudicaule L. f. (Ophioglossaceae)	1	1.36	0.25	12.50	1.00	2.10	2..3
Ophioglossum vulgatum L. (Ophioglossaceae)	2	99.18	18.16	100.00	9.13	16.70	34.80
Co-Existing Species							
Pteridophytes							
Lygodium flexuosum (L.) Sw. (Lygodiaceae)	5	9.51	1.74	50.00	1.75	8.30	10.10
Selaginella rupestris (L.) Spring (Selaginellaceae)	17	12.23	2.24	37.50	3.00	6.30	8.50
Graminoids							
Cyperus rotundus L. (Cyperaceae)	12	27.17	4.98	50.00	5.00	8.30	13.30
Eragrostis tenella (L.) Roem. & Schult. (Poaceae)	15	119.57	21.89	12.50	88.00	2.10	24.00
Forbs							
Antidesma ghaesembilla Gaertn. (Euphorbiaceae)	18	1.36	0.25	12.50	1.00	2.10	2.30
Cissus adnata Roxb. (Vitaceae)	21	2.72	0.50	25.00	1.00	4.20	4.70
Commelina benghalensis L. (Commelinaceae)	19	2.72	0.50	25.00	1.00	4.20	4.70
Curculigo orchioides Gaertn. (Hypoxidaceae)	8	1.36	0.25	12.50	1.00	2.10	2.30
Cyanotis axillaris (L.) Sweet (Commelinaceae)	22	1.36	0.25	12.50	1.00	2.10	2.30
Desmodium triflorum (L.) DC. (Fabaceae)	9	0.00	0.00	0.00	0.00	0.00	0.00
Dioscorea bulbifera L. (Dioscoreaceae)	11	0.00	0.00	0.00	0.00	0.00	0.00
Drosera burmannii Vahl (Droseraceae)	16	0.00	0.00	0.00	0.00	0.00	0.00
Evolvulus alsinoides L. (Convolvulaceae)	7	0.00	0.00	0.00	0.00	0.00	0.00
Hemidesmus indicus (L.) R.Br. (Asclepiadaceae)	4	0.00	0.00	0.00	0.00	0.00	0.00
Ionidium suffruticosum DC. (Violaceae)	13	0.00	0.00	0.00	0.00	0.00	0.00
Lindernia antipoda (L.) Alston (Scrophulariaceae)	14	0.00	0.00	0.00	0.00	0.00	0.00
Mazus pumilus (Burm. f.) Steenis (Scrophulariaceae)	3	213.32	39.05	87.50	22.43	14.60	53.60
Oldenlandia corymbosa L. (Rubiaceae)	6	42.12	7.71	50.00	7.75	8.30	16.00
Phyllanthus niruri L. (Euphorbiaceae)	10	0.00	0.00	0.00	0.00	0.00	0.00
Ziziphus oenoplia (L.) Mill. (Rhamnaceae)	20	1.36	0.25	12.50	1.00	2.10	2.30

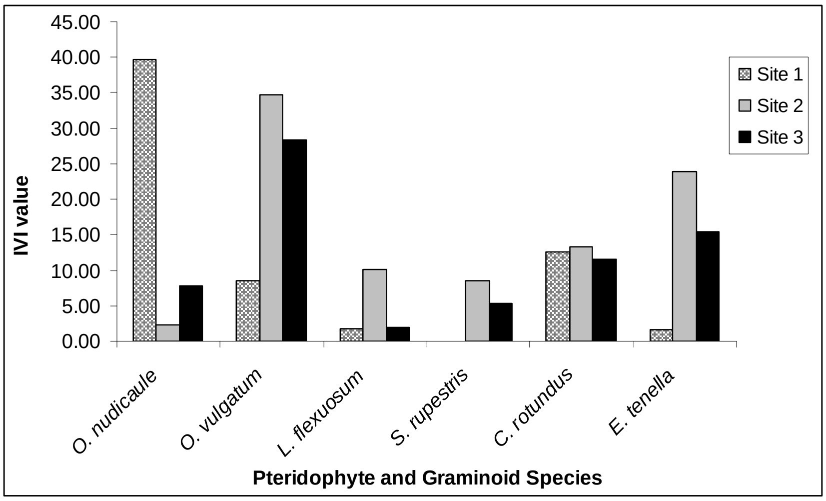
D/Sq. mt.: Density/Square meter; RD: Relative Density; Ab: Abundance; F%: Frequency; RF: Relative Frequency; RBA: Relative Basal Area; IVI: Importance Value Index; No. refers to the unique identifying number assigned to the species for analysis and which will be used in Table 5 and the Plexus diagrams in Figures 5 and 6.

Table 4 Structural parameters of two species of *Ophioglossum* and 20 other co-existing plants in Site 3.

Name of the Species	NO.	D	RD	FQ%	AB	RF	IVI
Study species							
Ophioglossum nudicaule L. f. (Ophioglossaceae)	1	6.79	1.25	50.00	1.25	6.60	7.80
Ophioglossum vulgatum L. (Ophioglossaceae)	2	82.88	15.25	100.00	7.63	13.10	28.40
Co-Existing Species							
Pteridophytes							
Lygodium flexuosum (L.) Sw. (Lygodiaceae)	5	1.36	0.25	12.50	1.00	1.60	1.90
Selaginella rupestris (L.) Spring (Selaginellaceae)	17	10.87	2.00	25.00	4.00	3.30	5.30
Graminoids							
Cyperus rotundus L. (Cyperaceae)	12	27.17	5.00	50.00	5.00	6.60	11.60
Eragrostis tenella (L.) Roem. & Schult. (Poaceae)	15	74.73	13.75	12.50	55.00	1.60	15.40
Forbs							
Antidesma ghaesembilla Gaertn. (Euphorbiaceae)	18	2.72	0.50	25.00	1.00	3.30	3.80
Cissus adnata Roxb. (Vitaceae)	21	2.72	0.50	25.00	1.00	3.30	3.80
Commelina benghalensis L. (Commelinaceae)	19	2.72	0.50	25.00	1.00	3.30	3.80
Curculigo orchioides Gaertn. (Hypoxidaceae)	8	1.36	0.25	12.50	1.00	1.60	1.90
Cyanotis axillaris (L.) Sweet (Commelinaceae)	22	2.72	0.50	25.00	1.00	3.30	3.80
Desmodium triflorum (L.) DC. (Fabaceae)	9	2.72	0.50	25.00	1.00	3.30	3.80
Dioscorea bulbifera L. (Dioscoreaceae)	11	0.00	0.00	0.00	0.00	0.00	0.00
Drosera burmannii Vahl (Droseraceae)	16	6.79	1.25	37.50	1.67	4.90	6.20
Evolvulus alsinoides L. (Convolvulaceae)	7	1.36	0.25	12.50	1.00	1.60	1.90
Hemidesmus indicus (L.) R.Br. (Asclepiadaceae)	4	5.43	1.00	25.00	2.00	3.30	4.30
Ionidium suffruticosum DC. (Violaceae)	13	2.72	0.50	25.00	1.00	3.30	3.80
Lindernia antipoda (L.) Alston (Scrophulariaceae)	14	0.00	0.00	0.00	0.00	0.00	0.00
Mazus pumilus (Burm. f.) Steenis (Scrophulariaceae)	3	205.16	37.75	87.50	21.57	11.50	49.20
Oldenlandia corymbosa L. (Rubiaceae)	6	46.20	8.50	75.00	5.67	9.80	18.30
Phyllanthus niruri L. (Euphorbiaceae)	10	0.00	0.00	0.00	0.00	0.00	0.00
Ziziphus oenoplia (L.) Mill. (Rhamnaceae)	20	1.36	0.25	12.50	1.00	1.60	1.90

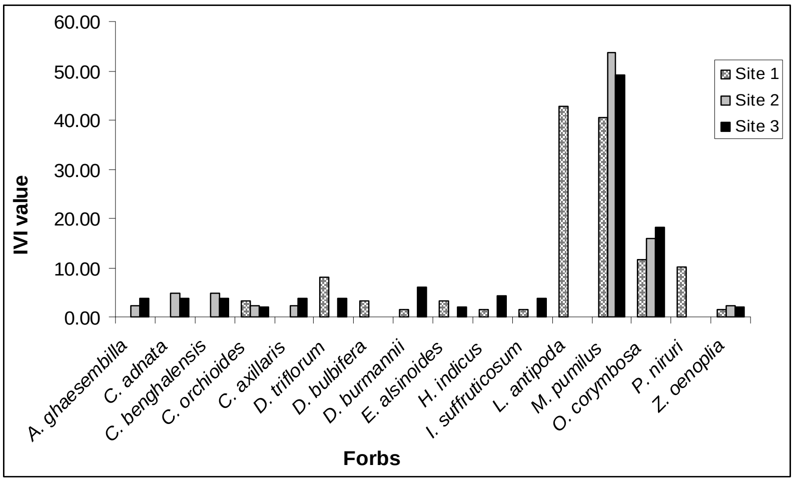
D/Sq. mt.: Density/Square meter; RD: Relative Density; Ab: Abundance; F%: Frequency; RF: Relative Frequency; RBA: Relative Basal Area; IVI: Importance Value Index; No. refers to the unique identifying number assigned to the species for analysis and which will be used in Table 5 and the Plexus diagrams in Figures 5 and 6.

Fig. 3. Comparative account of Importance Value Index of pteridophytes and graminoids in the three study sites



IVI: Importance Value Index.

Fig. 4. Comparative account of Importance Value Index of different Forbs in the three study sites



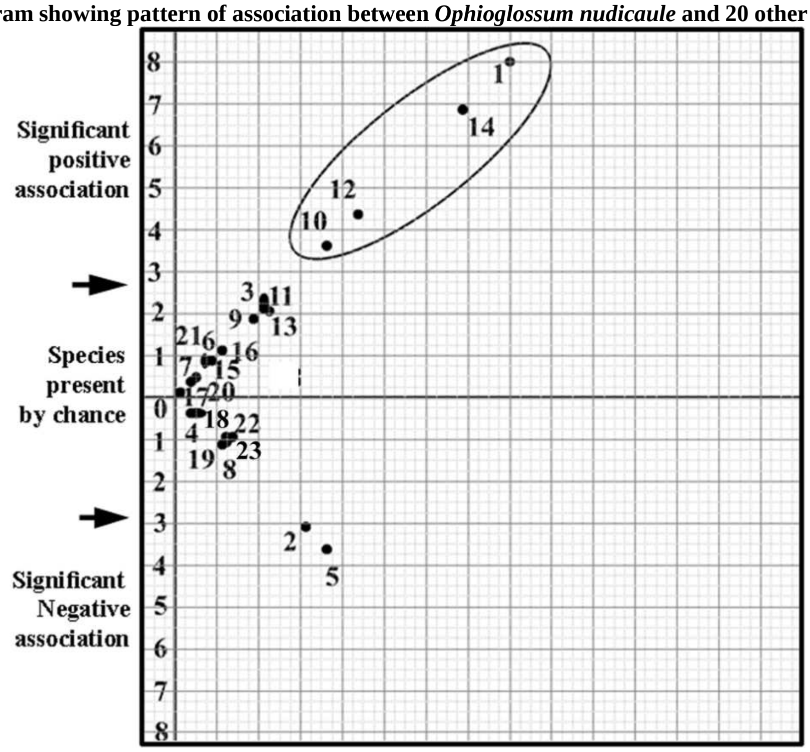
IVI: Importance Value Index.

Table 5 Inter-specific association indices and test statistics between two species of *Ophioglossum* and 20 other co-existing plants

Ophioglossum nudicaule 1							*Ophioglossum vulgatum* 2						
			Yate's	Association Index						Yate's	Association Index		
Sp. Pair	AT	Chi-Square	Chi-square	OI	DI	JI	Sp. Pair	AT	Chi-Square	Chi-square	OI	DI	JI
1-2	−	3.16	1.34	0.58	0.56	0.34	2-3		0.31	0.31	0.08	0.08	0.79
1-3		2.18	0.55	0.74	0.71	0.55	2-4		0.49	0.05	0.38	0.25	0.14
1-4		0.38	0.00	0.17	0.13	0.07	2-5		0.13	0.13	0.45	0.37	0.23
1-5	−	3.56	2.00	0.12	0.11	0.06	2-6		1.71	0.43	0.71	0.70	0.54
1-6		0.75	0.19	0.65	0.64	0.47	2-7		0.31	0.31	0.31	0.17	0.10
1-7		0.38	0.00	0.33	0.27	0.15	2-8		0.69	0.00	0.44	0.32	0.19
1-8		1.20	0.30	0.14	0.13	0.07	2-9		2.33	0.72	0.41	0.36	0.22
1-9		1.82	0.81	0.55	0.53	0.36	2-10	−	3.17	1.14	0.36	0.30	0.17
1-10	+	3.56	2.00	0.59	0.56	0.38	2-11	−	2.81	0.31	0.15	0.09	0.50
1-11		2.18	0.55	0.41	0.29	0.17	2-12		2.06	0.63	0.68	0.67	0.50
1-12	+	4.44	2.84	0.75	0.74	0.59	2-13		0.31	0.31	0.31	0.17	0.10
1-13		2.18	0.55	0.41	0.29	0.17	2-14	−	6.86	3.86	0.39	0.34	0.21
1-14	+	6.75	4.69	0.71	0.70	0.54	2-15		1.36	0.05	0.25	0.17	0.09
1-15		0.38	0.00	0.33	0.27	0.15	2-16		0.69	0.00	0.44	0.32	0.19
1-16		1.20	0.30	0.43	0.38	0.23	2-17		0.69	0.00	0.44	0.32	0.19
1-17		0.25	0.00	0.39	0.35	0.21	2-18		0.49	0.05	0.38	0.25	0.14
1-18		0.38	0.00	0.17	0.13	0.07	2-19		0.69	0.00	0.44	0.32	0.19
1-19		1.20	0.30	0.14	0.13	0.07	2-20		0.31	0.31	0.31	0.17	0.10
1-20		0.38	0.00	0.33	0.27	0.15	2-21		0.69	0.00	0.44	0.32	0.19
1-21		1.20	0.30	0.14	0.13	0.07	2-22		0.31	0.31	0.31	0.17	0.10
1-22		0.38	0.00	0.17	0.13	0.07							

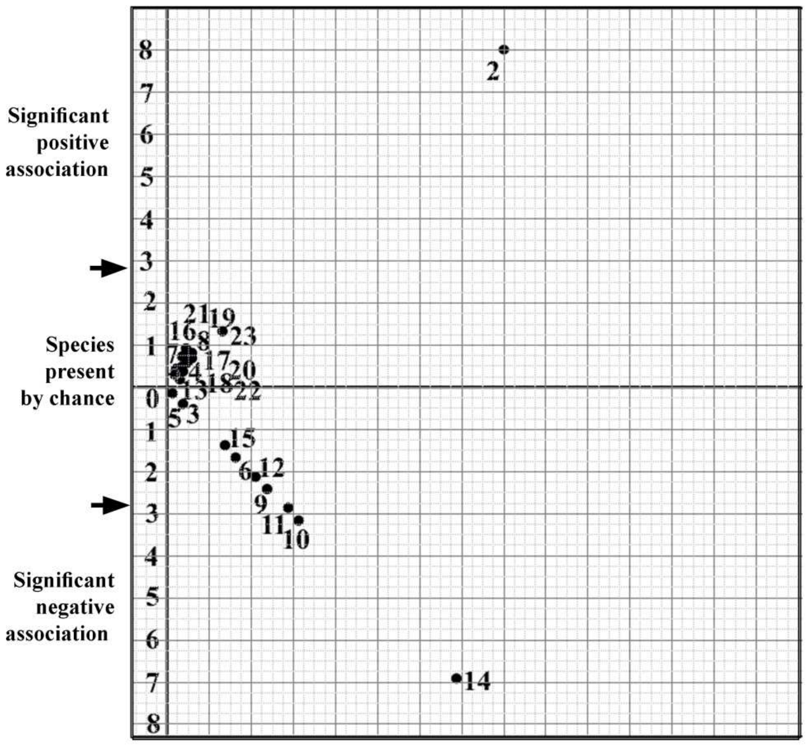
(OI: Ochia; DI: Dice and JI: Jaccard Indices; Species Pair numbers are as pairs of unique identifying number assigned to each species - See Tables 2, 3, 4)

Fig. 5. Plexus diagram showing pattern of association between *Ophioglossum nudicaule* and 20 other co-existing plants



1 large box = 1 Chi- square value; Numbers in the diagram are as per unique identifying number assigned to each species - See Tables 2, 3, 4.

Fig. 6. Plexus diagram showing pattern of association between *Ophioglossum vulgatum* and 20 other co-existing plants.



1 large box = 1 Chi- square value; Numbers in the diagram are as per unique identifying number assigned to each species - See Tables 2, 3, 4.

DISCUSSION

Vegetative as well reproductive (development of spike) growth during rainy season and perennation through subterranean rhizome for the rest of the year may explain the preference of *Ophioglossum vulgatum* and *O. nudicaule* for moss-covered substrates. Moss beds help to retain moisture which probably favors the growth and development of *Ophioglossum* in tropical non monsoon season [26]. Co-existence of 15 out of 20 plant species with both study species of *Ophioglossum* in all three study sites indicates similar habitat preference of these two species. This result can be corroborated with the findings of [27, 28]. Therefore the negative association between *O. vulgatum* and *O. nudicaule* may be due to interspecific competition because of the overlapping nature of their niches [29]. Competition rather than the need for specialized habitat may therefore explain the lower relative density, abundance and relative frequency of both species of *Ophioglossum* (*O. vulgatum* and *O. nudicaule*) at any sampling units of all three studied sites.

Lindernia antipoda followed by *Phyllanthus niruri* have some strong function (positive and negative) in the establishment of *O. nudicaule* as well as *O. vulgatum* in the community as *O. nudicaule* prefers to grow with *Lindernia antipoda* and *Phyllanthus niruri* but *O. vulgatum* does not. Therefore, these two species of *Ophioglossum* were not co-existing together at a high frequency in any sampling unit of the three study sites.

CONCLUSION

In addition to natural calamities, anthropogenic pressure like alteration of habitat through deforestation etc. causes serious threats to the extinction of numerous ancient evolutionary significant novel plants. *Ophioglossum*, being an ancient lineage land vascular plant with the highest plant chromosome number is significantly under serious threat all over the world. The present study suggests that co-existence of the population of *Lindernia antipoda*, *Cyperus rotundus* and *Phyllanthus niruri* will help to increase the population size of *O. nudicaule*. In contrast, populations of *Lindernia antipoda*, *Phyllanthus niruri* and *Dioscorea bulbifera* should be avoided to increase population size of *Ophioglossum vulgatum*. It is also suggested that *O. nudicaule* and *O. vulgatum* not be grown together during *ex situ* conservation programs as they have strong negative association with each other in the competitive niche. Based on our study results, when growing these species outside regenerated and managed forests conservation efforts many be more effective if careful attention is paid to associated species.

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