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Isozymic variation studies on the selected species of *Tectaria* from India

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ABSTRACT

Isozymic variation studies on Tectaria coadunata (J. Smith) C.Chr., Tectaria wightii (Clarke) Ching and Tectaria paradoxa (Fee) Sledge was carried out to reveal the diversity existing at molecular level. PAGE was employed for the isoperoxidase analysis. A total of seventeen bands with five active regions were expressed in this enzyme system. Present study revealed hundred percentage genetic differentiations between the three Tectaria species. The isoperoxidase banding profiles confirmed the classification based on morphology. The present study confirms that all the three species are morphologically and genetically distinct, but cytologically uniform.

Keywords: *Tectaria*, Isozyme, PAGE

INTRODUCTION

Biological diversity is important gift to human beings, as it provides food medicine and industrial raw materials along with an immense potential for accruing many unknown benefits to future generations. As far as plants are concerned there are an estimated 2700,000 known species of vascular plants, which include ferns, fern allies, gymnosperms and flowering plants. Ferns and fen allies, collectively called Pteriodophytes, are first successful group of land plants without flowers and seeds. India is a megabiodiversity country with the presence of about 1200 species of Pteridophytes which are growing on the high altitude mountains like the Western Ghats and Himalayas.

Biodiversity should be analyzed accurately and in details in each group and in each level of taxa, and at the same time it is to be observed in synthetic standpoint as the whole. Systematic should always be elucidated in analyzing method and at the same time observed in integrated point of view. As far as Indian Pteridophytes are concerned, biodiversity has not been studied completely at any level. Modern floristic account on ferns of the Western Ghats

and Himalayas has been completed. Next to the morphological diversity, the cytological diversity on Indian Pteridophytes has been known considerably due to the hard risks of several dedicated workers from south India and North India and this is the successful field of biodiversity research on Indian Pteridophytes. Chromosome atlas of Indian Pteridophytes is being prepared by Prof. S. S. Bir, founder of Indian Fern Society, Punjabi University, Patiala, India. Other field of biodiversity research like, chemical diversity and molecular diversity are still at infant stage in India. On phytogeographical point of view south Indian ferns are the well studied ferns in India. After Beddome [1], revised flora has been published after a long gap by Manickam and Irudayaraj [2]. South Indian ferns have also been studied cytologically and for about 80% South Indian ferns cytological detail is available [3 and 4]. After morphotaxonomical and cytotaxonomical studies several important large families have been studied cytotaxonomically. Molecular taxonomic studies have carried out recently.

As the first step, isozyme analysis has been done on about 75 rare, endangered and common South Indian ferns and the results for the species of *Tectaria* is communicated here. *Tectaria* (Dryopteridaceae) is a temperate genus and it is poorly represented in the tropical areas like South India. There are about 20 speceis of *Tectaria* in India [5]. Recent survey shows the presence of three species of *Tectaria* in South India [2]. Isozymes constructed a path way to study the genetic differentiation in plant populations [6]. The presence and absence of band has been found useful in the placement of the plant in taxonomic categories. Isozyme differentiation is one such important and powerful procedure which has often employed for this purpose. Each and every isozyme has its own cell specificity and metabolic activity in the plant metabolism. In the present study three common species viz., *Tectaria coadunata* (J. Smith) C.Chr., *Tectaria wightii* (Clarke) Ching and *Tectaria paradoxa* (Fee) Sledge have been subjected to isozyme analysis in order to know their genetical distinctness.

EXPERIMENTAL SECTION

Plants of *Tectaria coadunate* (J. Smith) C.Chr., *Tectaria wightii* (Clarke) Ching and *Tectaria paradoxa* (Fee) Sledge were collected from the natural habitats and established in the green house attached to Kodaikanal Botanic Garden, Kodaikannal, Tamil Nadu and India. Croziers were harvested from the mother plants and served as explants for the isoperoxidase analysis. The isoperoxidase isolation, separation and staining was followed by Smila et al., method [7]. The isoperoxidase profiles were visualized and documented in Vilber Lobermat (Germany) and similarity and variation between the selected species were carried out by Biogene software (Germany).

RESULTS AND DISCUSSION

Isoperoxidase profile expressed multiple regions of activity for this system (Fig. 1). Regions three and four contained only one band $PRX3^1$ and $PRX4^1$ respectively. Both showed their unique presence in *T. coadunata*, other two species were failed to express these bands. Region seven contained six bands in different positions. $PRX7^1$, $PRX7^3$ and $PRX7^5$ were present only in *T. wightii*. $PRX7^2$ was restricted to *T. paradoxa*. $PRX3^4$ and $PRX7^6$ were illustrated in *T. coadunata*. Region seven played a key role in the biochemical characterization of the selected three species of *Tectaria*. Region eight showed three bands in three different positions with varied MW - Rf values. $PRX8^1$ was restricted to *T. coadunata*, other two bands $PRX8^2$ and $PRX8^3$ were present only in *T. paradoxa*; *T. wightii* failed to express in this region. Region nine showed six bands with varied Rf values; $PRX9^{1and\ 3}$ showed their presence in *T. paradoxa* and $PRX9^{2,\ 4\ \&\ 6}$ were restricted to *T. coaduanata* and

PRX9[5] was present only in *T. wightii*. A total of seventeen bands in five active regions were observed in this enzyme system (Table - 1).

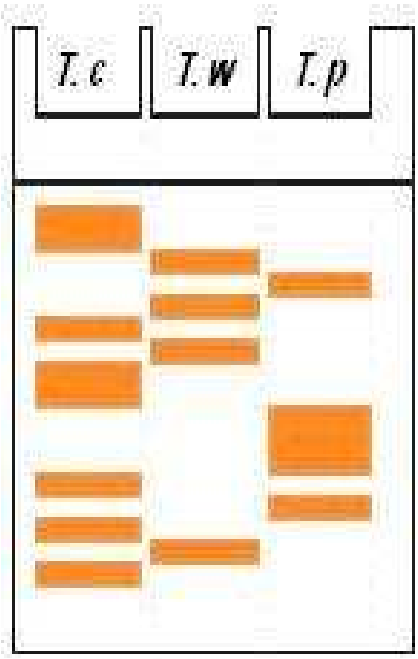

Fig.1. Isoperoxidase profiles of *Tectaria* sps. *T. c - Tectaria coaduanata,*
T. w - Tectaria wightii. T. p - Tectaria paradoxa

All bands showed variations between the species, similarity between the selected species were not observed among the species due to the presence of hundred percentage of diversified banding profile. Present study confirms the real distinctness of *Tectaria* species. These three species of *Tectaria* from South India are morphologically very distinct, particularly in type of lamina, but they are cytologically same. All the three species are diploid sexual (n=40; 2n=80). *Tectaria paradoxa* is with bipinnate fronds, *Tectaria wightii* is with typically simply pinnate lamina and in *Tectaria coadunata* the lamina is palmately lobed. Based on the type of fronds *Tectaria coadunata* is considered to be advanced species and *Tectaria paradoxa* is a primitive species and *T. wightii* is an intermediate one. The venation pattern also shows the same kind of evolutionary trend by having the primitive free venation type in *T. paradoxa* and anastamosing type with included veinlets in the areoles in *T. wightii* and the anastamosing type without included veinlets in the areoles in *T. coadunata*. In ferns, the evolution in venation pattern is to cope with environment. The trend of evolution in plants is from the aquatic environment towards terrestrial and from high altitude towards the plains. The results of isozyme analysis on these three species also reflect more or less the same trend of evolutionary differences among these three species with the presence of eight different bands in *T. coadunata*, five bands in *T. paradoxa* and only four bands in *T. wightii*. Isozymic variation has been chosen here to reveal the diversity existing at molecular level in *Tectaria coadunata, Tectaria wightii* and *Tectaria paradaxa*. Present study revealed the genetic differentiation between the three species and banding profiles of the selected three species expressed with diversified banding pattern in this enzyme system. The active regions occupied by a particular isozyme in the form of bands are the representatives of the expression of a particular gene locus coding for that isozyme. In certain regions, more than one distinct band are resolved, these bands could represent allelic isozymes, coded by different alleles of the same gene at a locus and thus occupy that particular zone on the system. In the present study also similar kind of banding profiles are obtained in regions

three, four and five and expressing the presence of multiple alleles. Similar to the present study, genetic distinctness of medicinal plants was carried out by Onus and Pickergill [8]; Sabu *et al.* [9]; Irudayaraj *et al.* [10], Liu *et al.*[11] Werth [12] Soltis *et al.* [13] and Irudayaraj *et al.* [14]. In addition the present study provided the biochemical marker for the three important ferns of Western Ghats, South India.

Table 1: Isoperoxidase profiles of *Tectaria* species of South India

MW-Rf Values	Isoform	*T. coadunata*	*T. wightii*	*T. paradaxa*
0.272	$PRX3^1$	+	-	-
0.339	$PRX4^1$	+	-	-
0.609	$PRX7^1$	-	+	-
0.649	$PRX7^2$	-	-	+
0.653	$PRX7^3$	-	+	-
0.666	$PRX7^4$	+	-	-
0.681	$PRX7^5$	-	+	-
0.693	$PRX7^6$	+	-	-
0.755	$PRX8^1$	+	-	-
0.770	$PRX8^2$	-	-	+
0.792	$PRX8^3$	-	-	+
0.824	$PRX9^1$	-	-	+
0.837	$PRX9^2$	+	-	-
0.842	$PRX9^3$	-	-	+
0.849	$PRX9^4$	+	-	-
0.856	$PRX9^5$	-	+	-
0.886	$PRX9^6$	+	-	-

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