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Systematic importance of rhizome stelar anatomy in selected Monilophytes from Thailand

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ABSTRACT: The present research aimed to investigate a possible relationship between rhizome stelar anatomy and current phylogenetic fern classifications. Forty-eight species of Thai ferns belonging to 46 genera in 25 families were collected from the field and their stelar anatomy was investigated using the paraffin method. Haplostele, actinostele, solenostele, dictyostele and arthrostele rhizome types were observed, but these anatomical features appear to be more closely correlated with the traditional morphological classifications, rather than recent molecular phylogenetic ones.

KEY WORDS: Classification, Ferns, Paraffin method, Phloem, Stele, Xylem.

INTRODUCTION

Ferns, or the monilophytes, are a group of vascular plants that similar to lycophytes in being reproduced by spores and undergo an alternation of generations (Christenhusz and Chase, 2014). Recent phylogeny suggested that ferns are sister to gymnosperms and angiosperms (seed plants), whereas lycophytes are sister group to ferns and seed plants (Schuettpelz and Pryer, 2007; Lehtonen, 2011). Ferns comprise about 318 genera and 10,400–12,000 species worldwide (Addo-Fordjour *et al.*, 2007). They are distributed in all parts of the world, but are especially rich in tropical rain forests (Judd *et al.*, 2007). On the basis of sprorangial development, botanists divided ferns into two groups; eusporangiate and leptosporangiate. The former is regard as plesiomorphy (Christenhusz and Chase, 2014). Most ferns families produced only one size of spore and are called homosporous ferns, which presented as a plesiomorphic condition, except in Marsileaceae and Salviniaceae which produced two different size and type of spores (heterospory) (Christenhusz and Chase, 2014).

Early attempts to classify the ferns relied on morphological and anatomical characters (Bower, 1922, 1926, 1928). More recently, a phylogenetic classification of ferns based on both molecular and morphological data has been introduced by Smith *et al.* (2006). Their analysis included data from one nuclear gene (18S rDNA), seven chloroplast markers (*rbcL*, *atpA*, *atpB*, *accD*, *rps*4, 16S rDNA, ITS), and three mitochondrial genes (*atp*1, *nad*2, *nad*5) (Hasebe *et al.*, 1994, 1995; Kranz and Huss, 1996; Manhart, 1994, 1995; Pahnke *et al.*, 1996; Pryer *et al.*, 1995, 2001, 2004; Sano *et al.*, 2000; Schneider *et al.*, 2004; Tsutsumi and Kato, 2005; Vangerow *et al.*, 1999; Wikström and Pryer, 2005; Wolf, 1997; Wolf *et al.*, 1998), and brought about several changes in family and genus circumscriptions. Specifically, they identified 4 monophyletic classes, 11 monophyletic orders, and 37 families, of which 32 families were strongly supported as monophyletic (Smith *et al.*, 2006). These findings allow anatomical and other morphological characters used in previous classifications to be reappraised in the light of a robust phylogeny for the entire group.

In addition to the "true ferns", the "fern allies" have traditionally been recognised as an associated division. The fern allies are usually considered to comprise the families: Psilotaceae, Equisetaceae, Lycopodiaceae, Selaginellaceae and Isoetaceae (Bower, 1922, 1926, 1928; Foster and Gifford, 1989). However, Smith *et al.* (2006) placed Psilotaceae and Equisetaceae together with the eusporangiate ferns, while Lycopodiaceae, Selaginellaceae and Isoetaceae forming the "Lycophytes". This classification has been broadly adopted by many botanists, and Christenhusz *et al.* (2011) have subsequently provided a linear classification of extant ferns and lycophytes based on this and other phylogenetic studies. The classification of Christenhusz *et al.* (2011) corresponds directly to the findings of Smith *et al.* (2006) in recognising 3 families and 5 genera in the Lycophytes, but differs from the latter in its treatment of the Monilophytes in recognising 45 families and 280 genera, as well as in the circumscription of several subclasses and orders.

Several morphological and anatomical characters of ferns are regards as significant characters for fern classification and identification. Common diagnostic morphological characters are such as indusium, annulus orientation, scale on rhizome and spore characters (Christenhusz and Chase, 2014). The anatomical characters are focused on stele in rhizomes and stipe characters. Structure and organization of stele as well as

vascular bundle arrangement in stipe are commonly used in fern classification (Hernandez *et al.*, 2012; Christenhusz and Chase, 2014). Hernandez *et al.* (2012) studied anatomical details of the stipe in ferns. They concluded that the circumendodermal band (CB) that surrounds the leaf trace could have two possible functions, namely, a protective function, in behaving like an endodermis, and a biomechanical function. They also found that the presence of the CB was correlated with leaf trace evolution. It is generally accepted that the morphology and anatomy of vascular bundles in the rhizome of ferns are conserved, with a minimal affects from the environment (Srivastava and Chandra, 2009). Therefore, this study aimed to explore diversity in stelar anatomy of Thai ferns and investigate the possible relationship with previous morphology-based and current molecular phylogenetic classifications. Stelar anatomy probably has some taxonomic significance and could provide information useful to support or further refine details of the recently proposed classifications.

MATERIALS AND METHODS

Plant materials of 48 species from 46 genera were selected as being representative of each of 25 fern families (Table 1). Voucher specimens were deposited at BCU, BKF and QBG. Stelar anatomy was studied using a modified paraffin method based on the protocol described by Johansen (1940). Mature rhizomes from fertile fronds were cut into ca. 5 mm long and fixed in 70% EtOH and glycerol. The samples were then slowly dehydrated in a TBA series, and then transferred into melted paraplast. They were then embedded in paraffin blocks, trimmed and sectioned using a sliding microtome. The resulting ribbons were double stained in Safranin-O and Fast-green. Anatomical observations were made under a light microscope and light micrographs were taken as needed.

RESULTS AND DISCUSSIONS

Five stelar forms were observed in this study: actinostele, haplostele, solenostele, dictyostele and arthrostele. Dictyostele was the most common stele type, being found in 32 species. A solenostele type was found in 10 species, and a haplostele arrangement was found in 4 species. Actinostele and arthrostele arrangements were rather uncommon, being found only in *Psilotum* and *Equisetum*, respectively (Figs 1–4; Table 2).

Comparison of our results with the recent phylogenetic classification of Smith *et al.* (2006)

Considering the leptosporangiate ferns, siphonostele (dictyostele and solenostele) was found in all families of the Polypodiales, Cyatheales and Osmundales, whereas protostele (haplostele) was observed in the Hymenophyllales, Schizaeales and Gleicheniales (Table 2). These 3 orders had a primitive stele (protostele is regarded as more primitive than siphonostele) (Wardlaw, 1944). Therefore, they should be merged at the same rank, as was proposed earlier by Foster and Gifford (1989). In the heterosporous ferns (Salviniales), each family (Salviniaceae and Marsileaceae) had a different stele type. Salviniaceae had a haplostele, whereas Marsileaceae had a solenostele. The morphological characters of Salviniaceae were primitive, including a dichotomous branch stem and stele (haplostele) (Smith *et al.*, 2006). The solenostele observed in the family Marsileaceae was also more advanced than that of the Salviniaceae (Worsdell, 1902). Therefore Salviniales sensu Smith should be separated into 2 orders, the Salviniales and the Marsileales, which correspond to the previous morphological classification of Foster and Gifford (1989).

Comparison of our results with the previous morphological study of Foster and Gifford (1989)

In leptosporangiate ferns, all families in the order Filicales had a siphonostele (dictyostele and solenostele), except for 2 families, Schizaeaceae and Hymenophyllaceae (Table 2), which had a protostele (haplostele). Based on our results, the 3 families with a protostele should be excluded from the Filicales, as shown in the phylogenetic classification of Smith *et al.* (2006). Among the heterosporous ferns, stelar anatomy of the Salviniaceae was more primitive than in the Marsileaceae. These 2 families are separated into 2 orders in the morphological classification of Foster and Gifford (1989), and our findings support this scheme. Stelar anatomy was variable among the eusporangiate ferns. Stelar anatomy in Psilotaceae was actinostele, while in Equisetaceae it was arthrostele. Marattiaceae and Ophioglossaceae had dictyostele and solenostele, respectively.

Stele type in eusporangiate ferns

Our comparison of stele type with different classifications of eusporangiate ferns had the same outcome. Stelar anatomy supports the eusporangiate ferns as being more primitive than the leptosporangiate ferns. However, the stele types observed in this study revealed the eusporangiate ferns (Marattiales, Equisetales, Psilotales and Ophioglossales) to be variable, exhibiting forms that range from primitive to more advanced. Though, some families in this group had the same stele type as certain leptosporangiate ferns, they cannot be grouped with the leptosporangiate ferns because other morphological characters are anomalous, in particular spore structure. Stele type in the Ophioglossaceae (solenostele) was more advanced than that of the Psilotaceae. This result corresponds with other morphological characters, with the Ophioglossaceae having true leaves (megaphylls) whereas the Psilotaceae just has enations.

Table 1. List of species used in this study. Family classification is based on Smith *et al.* (2006).

	Families (Smith *et al.*, 2006)	Species	Collected sites	Voucher specimens
1	Aspleniaceae	*Asplenium ensiforme* Wall. ex Hook. & Grev.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 079 (BCU)
2	Blechnaceae	*Blechnum orientale* L.	Bangkok, Chatuchak market	AR 101 (BKF)
3		*Stenochlaena palustris* (Burm. f.) Bedd.	Bangkok, Suan Luang Rama IX	AR 045 (BKF)
4	Cibotiaceae	*Cibotium barometz* (L.) J. Sm.	Phetchabun, Lom Sak	AR 092 (BCU)
5	Cyatheaceae	*Cyathea gigantea* (Wall. ex Hook.) Holttum	Phetchabun, Lom Sak	AR 093 (BCU)
6	Davalliaceae	*Humata repens* (L.f.) Diels	Phitsanulok, Phu Hin Rong Kla N.P.	AR 076 (BCU)
7		*Araiostegiella faberiana* (C. Chr.) M. Kato & Tsutsumi	Phitsanulok, Phu Hin Rong Kla N.P.	AR 078 (BCU)
8	Dennstaedtiaceae	*Microlepia herbacea* Ching & C. Chr. ex Tardieu & C. Chr.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 063 (BCU)
9		*Histiopteris incisa* (Thunb.) J. Sm.	Phetchabun, Lom Sak	AR 091 (BCU)
10	Dryopteridaceae	*Bolbitis sinense* (Baker) K. Iwats.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 068 (BCU)
11		*Dryopteris diffracta* (Baker) C. Chr.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 061 (BCU)
12		*Leucostegia immersa* Wall. ex C. Presl	Phitsanulok, Phu Hin Rong Kla N.P.	AR 054 (BCU)
13		*Elaphoglossum subellipticum* Rosenst.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 066 (BCU)
14	Equisetaceae	*Equisetum ramosissimum* Desf. subsp. *debile* (Roxb. ex Vaucher) Hauke	Phitsanulok, Phu Hin Rong Kla N.P.	AR 087 (BCU)
15	Gleicheniaceae	*Dicranopteris linearis* (Burm. f.) Underw.	Chiang Mai, Mae Rim	AR 036 (QBG)
16	Hymenophyllaceae	*Hymenophyllum denticulatum* Sw.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 083 (BCU)
17	Lindsaeaceae	*Lindsaea cultrata* (Willd.) Sw.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 084 (BCU)
18		*Sphenomeris chinensis* (L.) Maxon	Phetchabun, Lom Sak	AR 088 (BCU)
19	Lomariopsidaceae	*Cyclopeltis crenata* (Fée) C. Chr.	Phangnga, Mueang Phangnga	AR 097 (BCU)
20	Lygodiaceae	*Lygodium circinatum* (Burm. f.) Sw.	Bangkok, Suan Luang Rama IX	AR 037 (BKF)
21	Marattiaceae	*Angiopteris evecta* (G. Forst.) Hoffm.	Bangkok, Chatuchak market	AR 103 (BCU)
22	Marsileaceae	*Marsilea crenata* C. Presl	Bangkok, Suan Luang Rama IX	AR 051 (BKF)
23	Oleandraceae	*Oleandra musifolia* (Blume) C. Presl	Phitsanulok, Phu Hin Rong Kla N.P.	AR 074 (BCU)
24	Ophioglossaceae	*Helminthostachys zeylanica* (L.) Hook.	Bangkok, Chatuchak market	AR 095 (BKF)
25		*Ophioglossum petiolatum* Hook.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 089 (BCU)
26	Osmundaceae	*Plenasium javanicum* (Blume) C. Presl	Bangkok, Chatuchak market	AR 104 (BKF)
27	Polypodiaceae	*Belvisia henryi* (Hieron. ex C. Chr.) Tagawa	Phitsanulok, Phu Hin RongKla N.P.	AR 059 (BCU)
28		*Drynaria rigidula* (Sw.) Bedd.	Phitsanulok, Phu Hin RongKla N.P.	AR 055 (BCU)
29		*Goniophlebium microrhizoma* (C. B. Clarke ex Baker) Bedd.	Phitsanulok, Phu Hin RongKla N.P.	AR 075 (BCU)
30		*Lecanopteris sinuosa* (Hook.) Copel.	Bangkok, Chatuchak market	AR 102 (BKF)
31		*Leptochilus decurrens* Blume	Chiang Mai, Mae Rim	AR 011 (QBG)
32		*Microsorum punctatum* (L.) Copel.	Chiang Mai, Mae Rim	AR 001 (QBG)
33		*Phymatosorus scolopendria* (Burm. f.) Pic. Serm.	Chiang Mai, Mae Rim	AR 013 (QBG)
34		*Pyrrosia lingua* (Thunb.) Farw.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 060 (BCU)
35		*Selliguea hirsuta* S. Linds.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 056 (BCU)
36	Psilotaceae	*Psilotum nudum* (L.) Beauv.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 096 (BCU)
37	Pteridaceae	*Calciphilopteris ludens* (Wall. ex Hook.) Yesilyurt & H. Schneider	Chiang Mai, Mae Rim	AR 028 (QBG)
38		*Acrostichum speciosum* Willd.	Bangkok, Suan Luang Rama IX	AR 043 (BKF)
39		*Ceratopteris thalictroides* (L.) Brongn.	Prachin Buri, Ban Sang	AR 099 (BCU)
40		*Pteris ensiformis* Burm. f.	Chiang Mai, Mae Rim	AR 009 (QBG)
41		*Adiantum lunulatum* Burm. f.	Loei, Chiang Khan	AR 098 (BCU)
42	Salviniaceae	*Salvinia cucullata* Roxb.	Bangkok, Suan Luang Rama IX	AR 052 (BKF)
43	Tectariaceae	*Tectaria laotica* Tardieu & C. Chr.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 070 (BCU)
44		*Heterogonium pinnatum* (Copel.) Holttum	Phangnga, Mueang Phangnga	AR 100 (BCU)
45	Thelypteridaceae	*Cyclosorus lakhimpurense* B. K. Nayar & S. Kaur	Phitsanulok, Phu Hin RongKla N.P.	AR 065 (BCU)
46		*Cyclosorus parasiticus* (L.) Farwell	Chiang Mai, Mae Rim	AR 038 (QBG)
47	Woodsiaceae	*Diplazium dilatatum* Blume	Bangkok, Suan Luang Rama IX	AR 044 (BKF)
48		*Diplazium esculentum* (Retz.) Sw.	Phitsanulok, Phu Hin Rong Kla N.P.	AR 050 (BCU)

In the primitive fern family, Marattiaceae, various morphological characters are primitive, but the dictyostele is advanced. Rothwell (1999) investigated the fossil of ferns to resolve land plant phylogeny. He recognized *Psaronius* as an extinct genus in Marattiaceae, inferring some affinity between *Marattia* and *Angiopteris*. He also noted that *Psaronius* had a dictyostele, in common with *Marattia* and *Angiopteris*. Therefore, we can conclude that Marattiaceae has an advanced stele type (dictyostele), and as such represents the most advanced family as compared with other eusporangiate ferns. However, spore structure (eusporangiate) and the initiated of radial symmetry in their embryo indicate them to be primitive as compared with all members in the leptosporangiate ferns (Bower, 1926).

The genus *Equisetum* has a specific type of stele termed an "arthrostele". This type of stele has been described by Takhtajan (1959). The word "arthro" refer

Table 2. Comparison between three classification systems of ferns.

Foster and Gifford (1989) **Class** **Order** **Family** **Genus**	**Smith *et al.* (2006)** **Class** **Order** **Family** **Genus**	**Christenhunz *et al.* (2011)** **Subclass** **Order** **Family** **Genus**	**Stele type**	**Fern group**
Sphenopsida	*Equisetopsida*	Equisetidae		
Equisetales	*Equisetales*	Equisetales		
Equisetaceae	*Equisetaceae*	Equisetaceae		
Equisetum	*Equisetum*	*Equisetum*	Arthrostele	
Psilopsida	*Psilotopsida*	Ophioglossidae		
Psilotales	*Psilotales*	Psilotales		
Psilotaceae	*Psilotaceae*	Psilotaceae		
Psilotum	*Psilotum*	*Psilotum*	Actinostele	
Filicopsida				
Ophioglossales	*Ophioglossales*	Ophioglossales		
Ophioglossaceae	*Ophioglossaceae*	Ophioglossaceae		
Helminthostachys	*Helminthostachys*	*Helminthostachys*	Solenostele	
Ophioglossum	*Ophioglossum*	*Ophioglossum*	Solenostele	
	Marattiopsida	Marattiidae		
Marattiales	*Marattiales*	Marattiales		
Marattiaceae	*Marattiaceae*	Marattiaceae		
Angiopteris	*Angiopteris*	*Angiopteris*	Dictyostele	
	Polypodiopsida	Polypodiidae		
Salviniales	Salviniales	Salviniales		
Salviniaceae	Salviniaceae	Salviniaceae		
Salvinia	*Salvinia*	*Salvinia*	Haplostele	
Marsileales				
Marsileaceae	Marsileaceae	Marsileaceae		
Marsilea	*Marsilea*	*Marsilea*	Solenostele	
	Polypodiopsida	Polypodiidae		
Filicales	Osmundales	Osmundales		
Osmundaceae	Osmundaceae	Osmundaceae		
Osmunda	*Osmunda*	*Osmunda* (*Plenasium*)	Dictyostele	
	Hymenophyllales	Hymenophyllales		
Hymenophyllaceae	Hymenophyllaceae	Hymenophyllaceae		
Hymenophyllum	*Hymenophyllum*	*Hymenophyllum*	Haplostele	
	Schizaeales	Schizaeales		
Schizaeaceae	Lygodiaceae	Lygodiaceae		
Lygodium	*Lygodium*	*Lygodium*	Haplostele	
	Gleicheniales	Gleicheniales		
	Gleicheniaceae	Gleicheniaceae		
	Dicranopteris	*Dicranopteris*	Haplostele	
	Cyatheales	Cyatheales		
Cyatheaceae	Cyatheaceae	Cyatheaceae		
Cyathea	*Cyathea*	*Cyathea*	Dictyostele	
	Cibotiaceae	Cibotiaceae		
	Cibotium	*Cibotium*	Solenostele	
	Polypodiales	Polypodiales		
	Lindsaeaceae	Lindsaeaceae		
	Lindsaea	*Lindsaea*	Solenostele	
	Sphenomeris	*Sphenomeris*	Solenostele	
Dennstaedtiaceae	Dennstaedtiaceae	Dennstaedtiaceae		
Microlepia	*Microlepia*	*Microlepia*	Solenostele	
	Histiopteris	*Histiopteris*	Solenostele	

Eusporangiate ferns

Leptosporangiate ferns

Table 2. Comparison between three classification systems of ferns. (continued)

Foster and Gifford (1989) **Class** **Order** **Family** **Genus**	**Smith *et al.* (2006)** **Class** **Order** **Family** **Genus**	**Christenhunz *et al.* (2011)** **Subclass** **Order** **Family** **Genus**	**Stele type**	**Fern group**
Pteridaceae	Pteridaceae	Pteridaceae		
Adiantum	*Adiantum*	*Adiantum*	Solenostele	
	Pteris	*Pteris*	Dictyostele	
	Ceratopteris	*Ceratopteris*	Dictyostele	
	Calciphilopteris	*Calciphilopteris*	Solenostele	
	Acrostichum	*Acrostichum*	Dictyostele	
Aspleniaceae	Aspleniaceae	Aspleniaceae		
Asplenium	*Asplenium*	*Asplenium*	Dictyostele	
	Thelypteridaceae	Thelypteridaceae		
	Cyclosorus	*Cyclosorus*	Dictyostele	
	Thelypteris	*Thelypteris* (*Cyclosorus*)	Dictyostele	
	Tectariaceae	Tectariaceae		
	Tectaria	*Tectaria*	Dictyostele	
	Heterogonium	(*Heterogonium*)	Dictyostele	
Blechnaceae	Blechnaceae	Blechnaceae		Leptosporangiate ferns
Blechnum	*Blechnum*	*Blechnum*	Dictyostele	
	Stenochlaena	*Stenochlaena*	Dictyostele	
Aspidiaceae	Dryopteridaceae	Dryopteridaceae		
	Bolbitis	*Bolbitis*	Dictyostele	
Dryopteris	*Dryopteris*	*Dryopteris*	Dictyostele	
	Elaphoglossum	*Elaphoglossum*	Dictyostele	
		Hypodematiaceae		
	Leucostegia	*Leucostegia*	Dictyostele	
	Woodsiaceae	Athyriaceae		
Athyrium	*Athyrium*	*Athyrium* (*Diplazium*)	Dictyostele	
	Diplazium	*Diplazium*	Dictyostele	
	Lomariopsidaceae	Lomariopsidaceae		
	Cyclopeltis	*Cyclopeltis*	Dictyostele	
	Oleandraceae	Oleandraceae		
	Oleandra	*Oleandra*	Dictyostele	
	Davalliaceae	Davalliaceae		
	Humata	*Davallia (Humata,*	Dictyostele	
	Araiostegiella	*Araiostegiella)*	Dictyostele	
Polypodiaceae	Polypodiaceae	Polypodiaceae		
Polypodium	*Goniophlebium*	*Goniophlebium*	Dictyostele	
(*Goniophlebium*)	*Drynaria*	*Drynaria*	Dictyostele	
	Belvisia	*Lepisorus (Belvisia)*	Dictyostele	
	Lecanopteris	*Lecanopteris*	Dictyostele	
	Leptochilus	*Leptochilus*	Dictyostele	
	Microsorum	*Microsorum*	Dictyostele	
	Phymatosorus	*Phymatosorus*	Dictyostele	
	Pyrrosia	*Pyrrosia*	Dictyostele	
	Selliguea	*Selliguea*	Dictyostele	

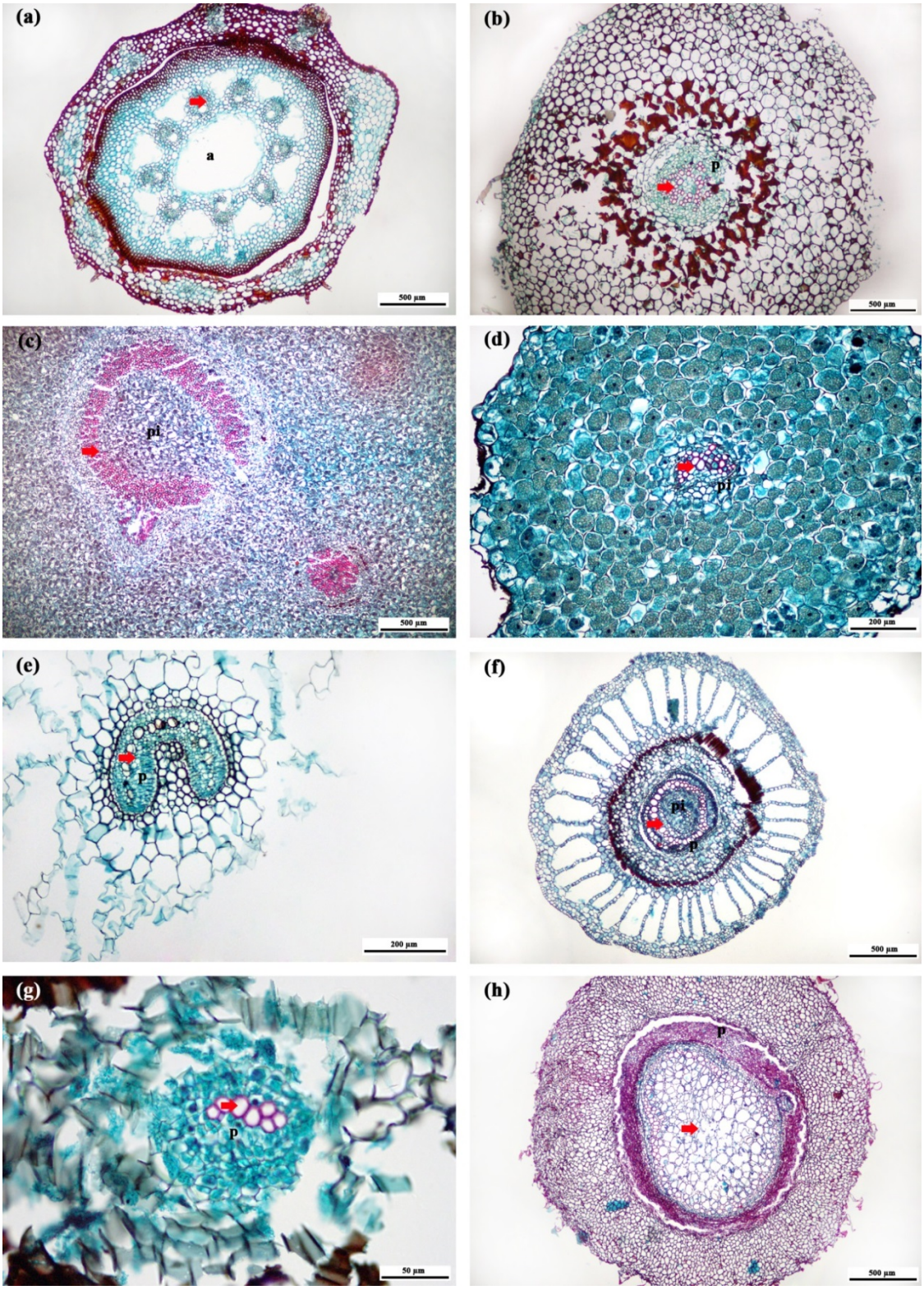

Fig. 1. Rhizome stelar anatomy in a selection of Thai fern families, seen in transverse section. **(a)**, Arthrostele in ***Equisetum ramosissimum*** subsp. ***debile*** (Equisetaceae); **(b)**, Actinostele in ***Psilotum nudum*** (Psilotaceae); **(c)**, Solenostele in ***Helminthostachys zeylanica*** (Ophioglossaceae); **(d)**, Solenostele in ***Ophioglossum petiolatum*** (Ophioglossaceae); **(e)**, Haplostele in ***Salvinia cucullata*** (Salviniaceae); **(f)**, Solenostele in ***Marsilea crenata*** (Marsileaceae); **(g)**, Haplostele in ***Hymenophyllum denticulatum*** (Hymenophyllaceae); **(h)**, Haplostele in ***Lygodium circinatum*** (Lygodiaceae); (arrow showing xylem); p = phloem; a = air space; pi = pith.

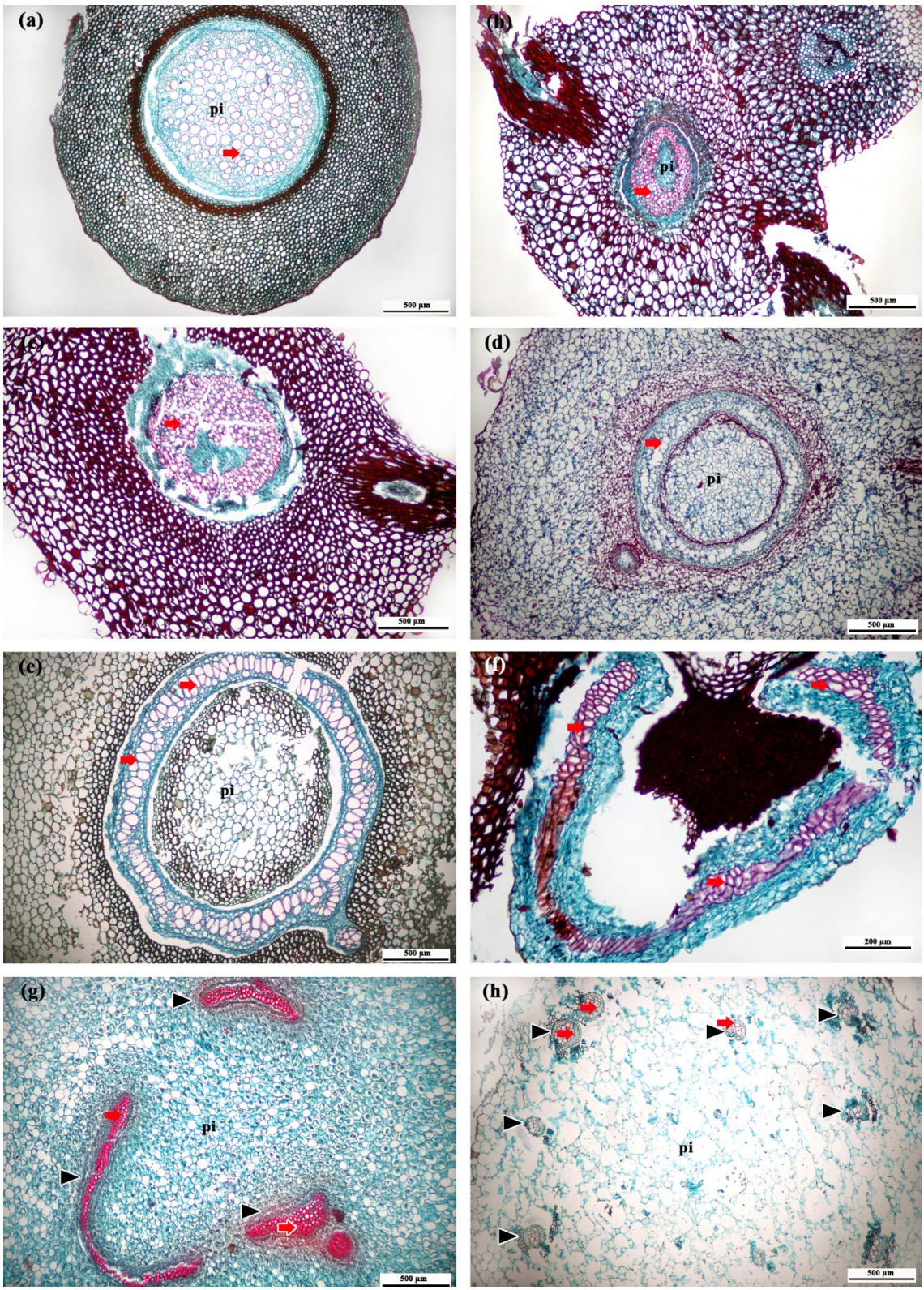

Fig. 2. Rhizome stelar anatomy in a selection of Thai fern families, seen in transverse section. **(a)**, Haplostele in ***Dicranopteris linearis*** (Gleicheniaceae); **(b)**, Solenostele in ***Lindsaea cultrata*** (Lindsaeaceae); **(c)**, Solenostele in ***Sphenomeris chinensis*** (Lindsaeaceae); **(d)**, Solenostele in ***Microlepia herbacea*** (Dennstaedtiaceae); **(e)**, Solenostele in ***Histiopteris incisa*** (Dennstaedtiaceae); **(f)**, Solenostele in ***Adiantum lunulatum*** (Pteridaceae); **(g)**, Dictyostele in ***Pteris ensiformis*** (Pteridaceae); **(h)**, Dictyostele in ***Ceratopteris thalictroides*** (Pteridaceae); (arrow showing xylem, triangle indicating meristele); pi = pith.

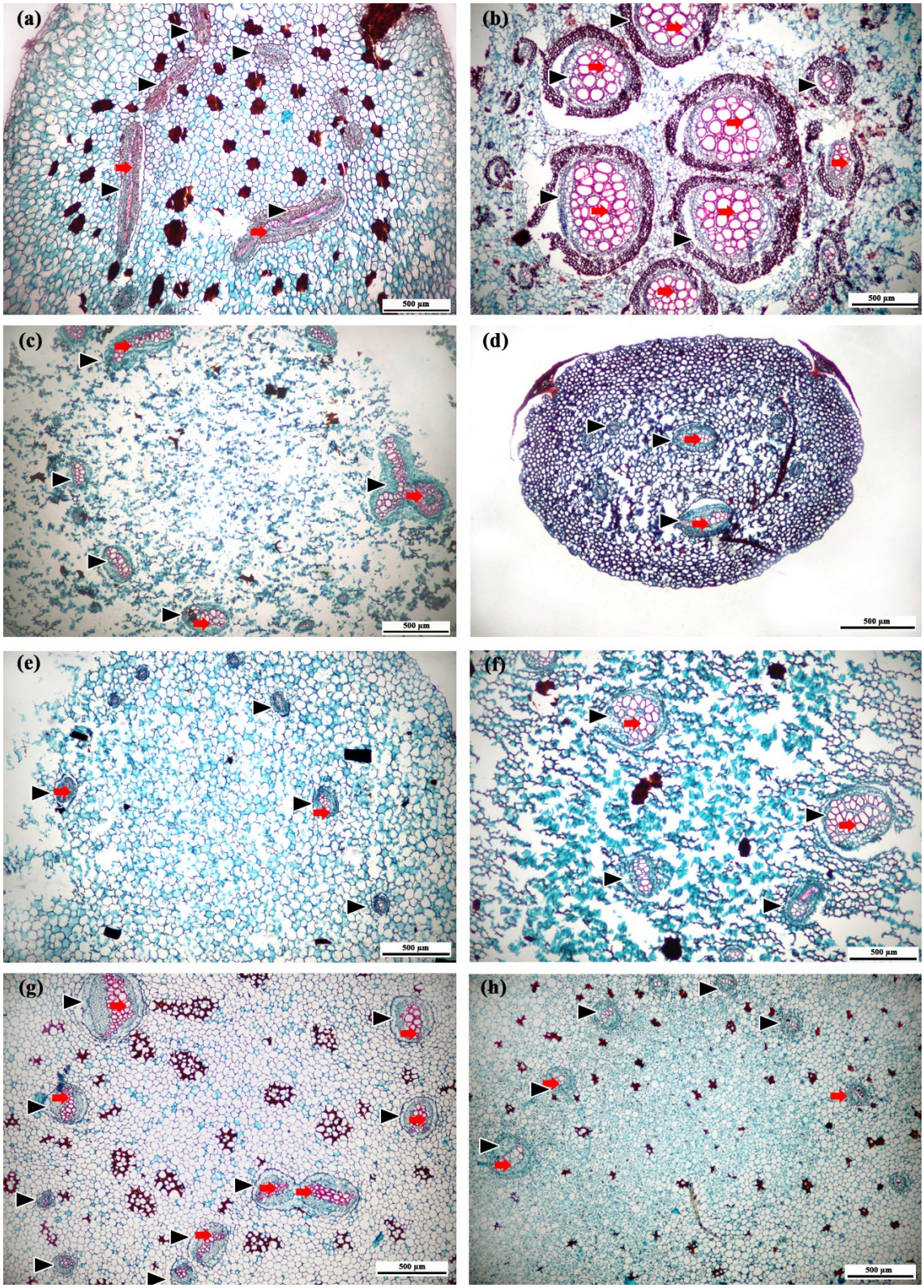

Fig. 3. Rhizome stelar anatomy in a selection of Thai fern families, seen in transverse section. **(a)**, Dictyostele in ***Asplenium ensiforme*** (Aspleniaceae); **(b)**, Dictyostele in ***Stenochlaena palustris*** (Blechnaceae); **(c)**, Dictyostele in ***Oleandra musifolia*** (Oleandraceae); **(d)**, Dictyostele in ***Humata repens*** (Davalliaceae); **(e)**, Dictyostele in ***Goniophlebium microrhizoma*** (Polypodiaceae); **(f)**, Dictyostele in ***Drynaria rigidula*** (Polypodiaceae); **(g)**, Dictyostele in ***Leptochilus decurrens*** (Polypodiaceae); **(h)**, Dictyostele in ***Microsorum punctatum*** (Polypodiaceae); (arrow showing xylem, triangle indicating meristele).

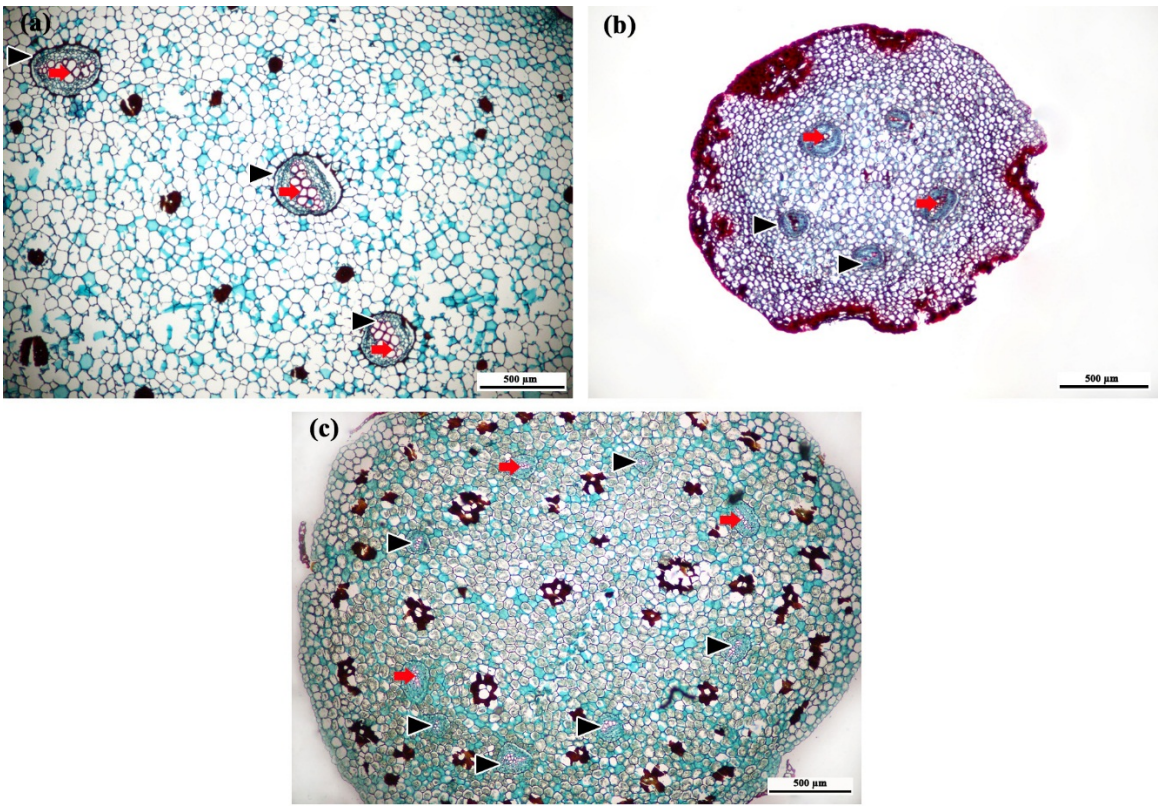

Fig. 4. Rhizome stelar anatomy in a selection of Thai fern families, seen in transverse section. **(a)**, Dictyostele in ***Phymatosorus scolopendria*** (Polypodiaceae); **(b)**, Dictyostele in ***Pyrrosia lingua*** (Polypodiaceae); **(c)**, Dictyostele in ***Selliguea hirsuta*** (Polypodiaceae); (arrow showing xylem, triangle indicating meristele).

to the internodes that look like the stem of *Equisetum*. Radford *et al.* (1974) presented a scheme for the evolution of the arthrostele. They suggested that the arthrostele is developed from a dictyostele. However, *Equisetum* is more primitive than other leptosporangiate taxa. Therefore, this conflict must be clarified in further studies using more species and other techniques. The stelar anatomical characters observed in this study correspond to the morphological characters described for the ferns elsewhere, and as such are rather more correlated with morphological classifications (Foster and Gifford, 1989) than with more recent phylogenetic classifications (Smith *et al.*, 2006). Our findings suggest that Psilotaceae and Equisetaceae should be treated as separate from the monilophytes, in conflict with the conclusions of Smith *et al.* (2006).

Variation within the Pteridaceae

The representative genera of ferns examined here generally exhibited the same stele type within each of their respective families (Table 2). Only in the Pteridaceae was a different stelar anatomy observed: the genus *Adiantum* and *Calciphilopteris* had a solenostele, whereas *Pteris*, *Ceratopteris* and *Acrostichum* had a dictyostele. More genera of the examined families should be observed to clarify variation in stele type.

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