ULTRASTRUCTURE OF SIEVE-ELEMENT PLASTIDS OF MYRTALES AND ALLIED GROUPS[1]

H.-DIETMAR BEHNKE[2]

ABSTRACT

The core families of Myrtales (69 species investigated) contain S-type sieve-element plastids. In these, the presence of several medium-sized spherular starch grains is probably a common trend in the order. Among those families usually closely associated or more or less distantly related, the great majority also have S-type plastids, some with the same characteristics. However, Rhizophoraceae, in contrast to the close allies and Connaraceae, Gunneraceae, and Rhabdodendraceae of the distantly related families, have developed P-type plastids. That subtype PV-plastids, which exclude all starch, are found in all tribes of Rhizophoraceae except Anisophylleae supports family recognition of the latter.

Sieve-element characters investigated with the transmission electron microscope have repeatedly contributed to the circumscription and classification of higher taxa, e.g., Leguminosae (Behnke & Pop, 1981), Monocotyledoneae (Behnke, 1981b). Of these, the most intensively studied feature is the ultrastructure of sieve-element plastids. The great number of investigated angiosperms (some 2,000 species from nearly 400 families—as of 1983-12-31) allow a subdivision of the plastids into P- and S-type, a number of subtypes, and a great many of characteristic forms by using both chemical and morphological composition at the ultrastructural level (see Behnke, 1981a, for more detail). The successful delimitation of the order Caryophyllales to the PIII plastid families (Behnke, 1976; Mabry, 1977) and the use of sieve-element plastid data in separating Vitidales from Rhamnales (Behnke, 1974; Dahlgren, 1980) and Buxaceae from Simmondsiaceae and Euphorbiaceae (Behnke, 1982a) stimulated, among others, the screening of all the core families of Myrtales and of those closely or more distantly related to the order, with the results to be included in this symposium report.

MATERIALS AND METHODS

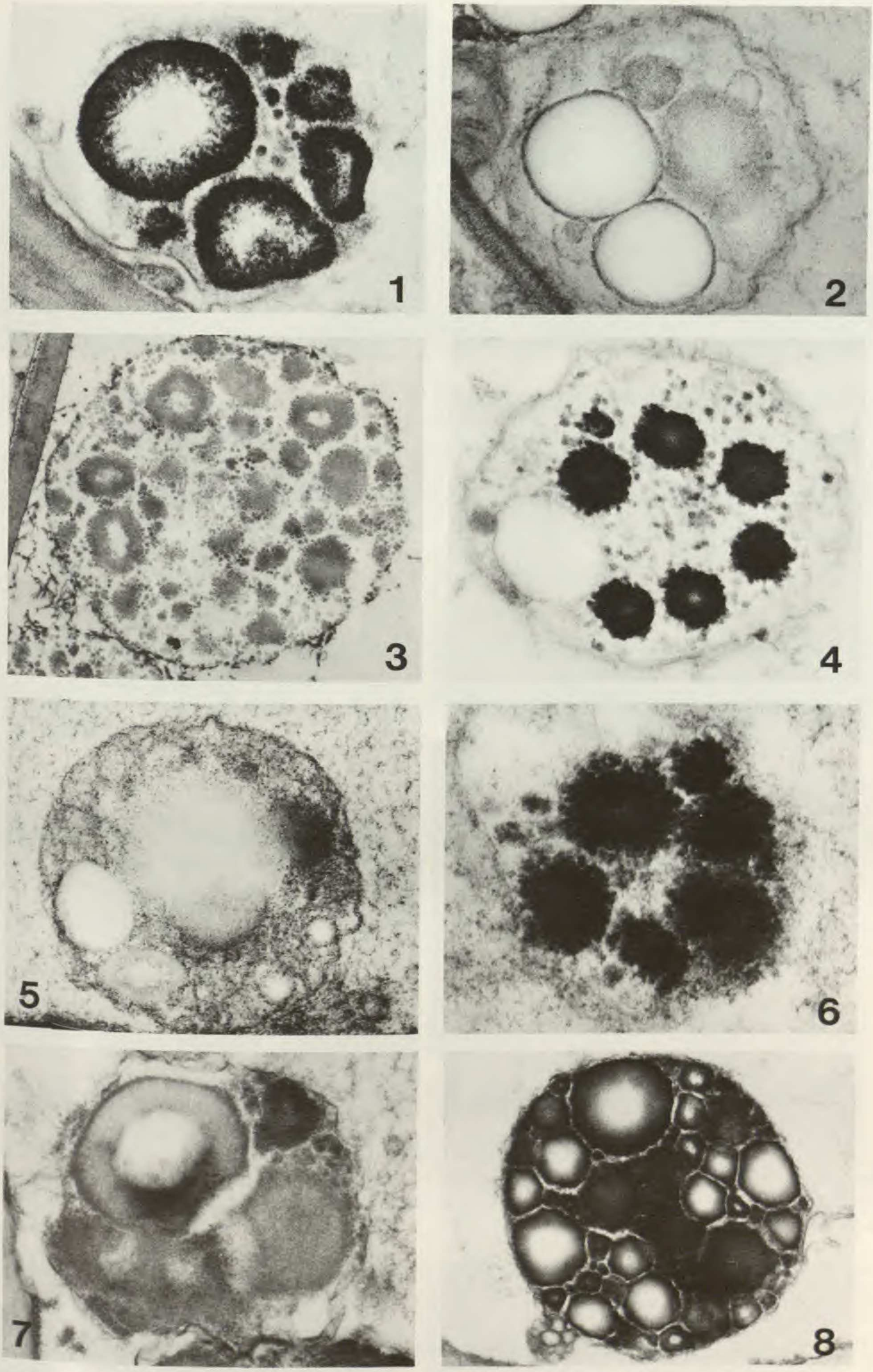
Preferentially young stem pieces of the plant species listed in Table 1 were cut into longitudinal sections fixed in a formaldehyde-glutaraldehyde mixture followed by 1% buffered OsO_4 and dehydrated in acetone. Small pieces containing phloem tissue were embedded and polymerized in epoxy resins, cut with an ultramicrotome, and the final ultrathin sections viewed and photographed with a transmission electron microscope (for exact procedures see Behnke, 1982a).

RESULTS AND DISCUSSION

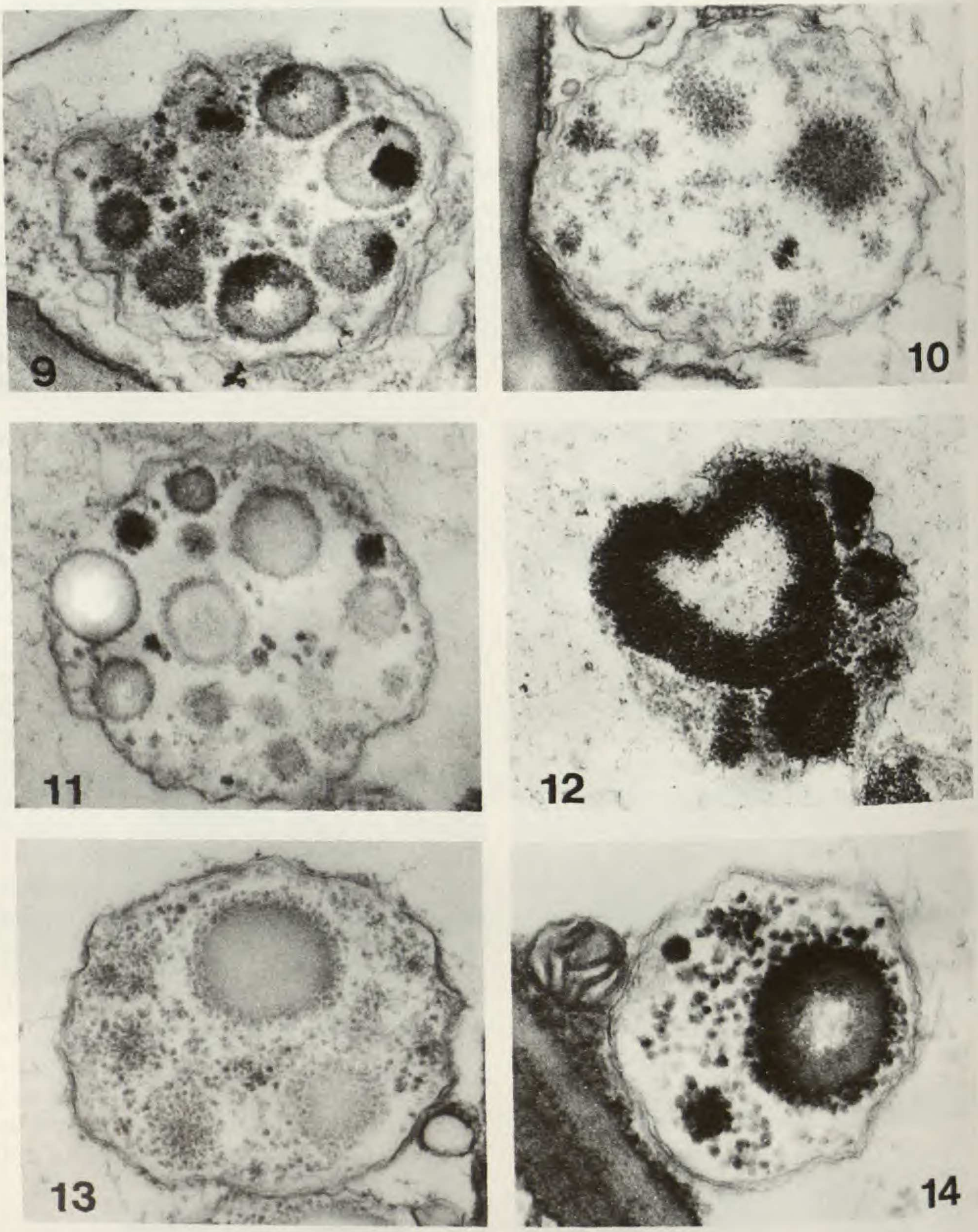
Sieve-elements of many of the myrtalean families are extremely difficult to fixate in a close-to-natural condition. There is less plasmatic content which is more labile and, during preparation, sieve-element plastids burst more often than in the average dicotyledon family. Nevertheless, all of the 69 investigated species of the core families of Myrtales (cf. Dahlgren & Thorne, 1984) could be shown to contain S-type plastids (cf. Table 1). Number, size, and shape of the starch grains in these plastids certainly vary (Figs. 1–14), but some common trends may be recognized. There are always several starch grains in one plastid—often six or more (Figs. 1–4, 6, 8–11), their size being variable (sometimes all of the same size) but never very large and their shape almost exclusively is a spherule. Considerably smaller starch grains are present in ad-

[1] For permitting collection of living material used in this study the author wishes to thank the directors and curators of the Botanical Gardens in Berlin-Dahlem, Bogor, Bonn, Copenhagen, Edinburgh, Heidelberg, Kew, Leiden, Mainz, and Utrecht. I am particularly indebted to A. Assi (Abidjan), P. Berry (Caracas), D. Clark (Kirstenbosch), I. Dorr (Kiel), D. M. C. Fourie (Pretoria), I. S. Gottsberger (Botucatu), A. Irvine (Atherton, Qld), A. Juncosa (Petersham), K. Kubitzki (Hamburg), Lee Ying Fah (Sandakan, Sabah), G. Merz (Heidelberg), H. B. Nicholson (St. Michaels-on-Sea, RSA), H. Téhé (ORSTROM), R. Tracey (Indooropilly) for collecting and shipping living plant material, A. P. Bennell (Edinburgh) and L. B. Jørgensen (Copenhagen) for helping with fixations, and A. R. A. Noel (Pietermaritzburg) and P. H. Raven (St. Louis) for making contact to collectors. I gratefully acknowledge the expert technical help of Mrs. L. Pop and Mrs. D. Laupp (both Heidelberg). This work was supported by grants from the Deutsche Forschungsgemeinschaft.

[2] Zellenlehre, Universität Heidelberg, Im Neuenheimer Feld 230, D-6900 Heidelberg, Federal Republic of Germany.

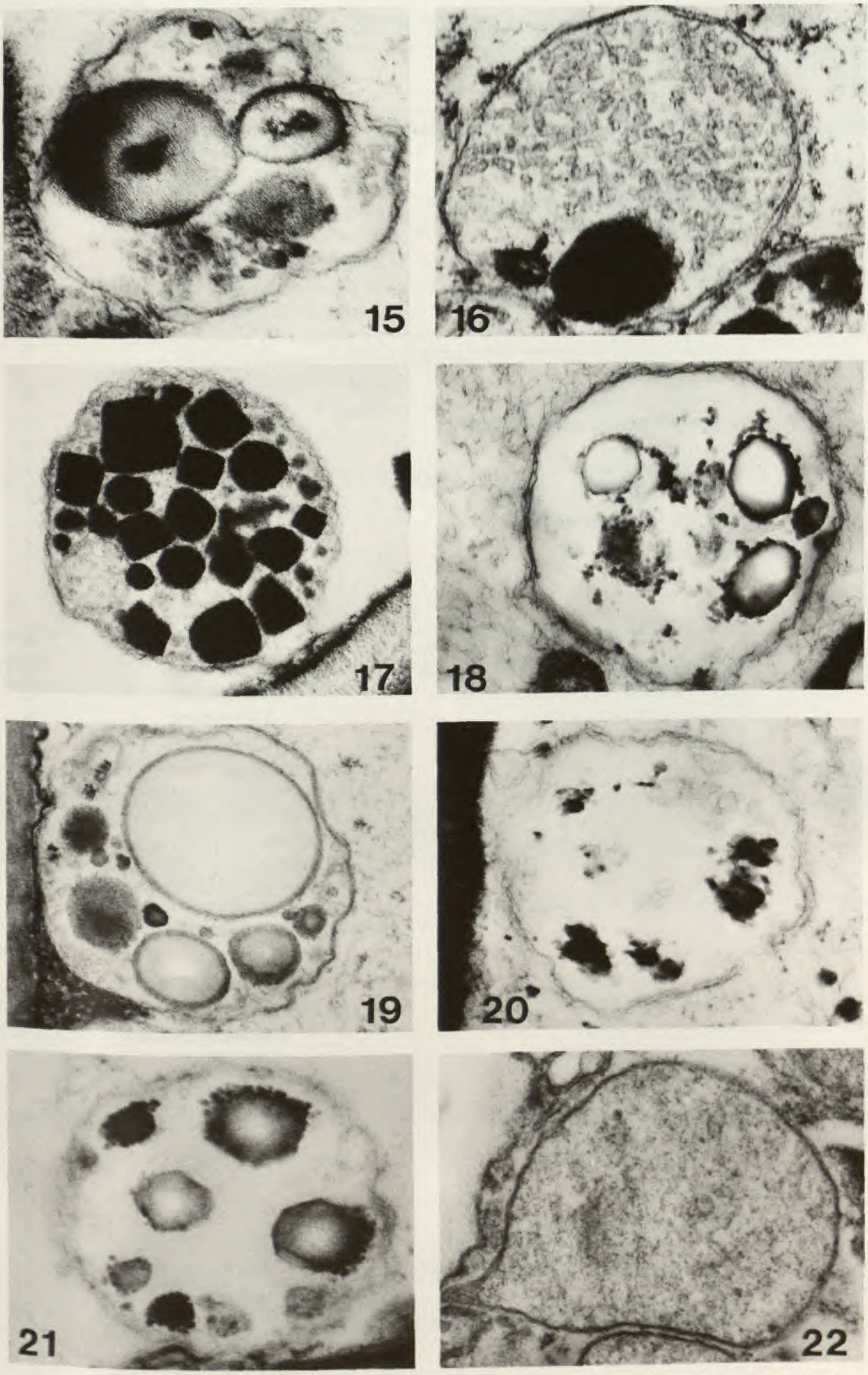


FIGURES 1–8. Sieve-element plastids of the core families of Myrtales I. All plastids are S-type, with examples from Melastomataceae.—1. *Schizocentron elegans.*—2. *Bertolonia maculata.*—3. *Medinilla magnifica,* Penaeaceae.—4. *Saltera sarcocolla,* Combretaceae.—5. *Combretum racemosa,* Rhynchocalycaceae.—6. *Rhynchocalyx lawsonioides,* Lythraceae.—7. *Lagerstroemia indica.*—8. *Duabanga sonneratioides.* Plastids reproduced to cover the same area, actual magnifications diverge around 20,000×.



FIGURES 9–14. Sieve-element plastids of the core families of Myrtales II. S-type plastids from Myrtaceae.—9. *Metrosideros excelsa*.—10. *Callistemon phoeniceus*.—11. *Psidium littorale*, Trapaceae.—12. *Trapa natans*, Onagraceae.—13. *Epilobium fleischerei*.—14. *Circaea cordata*.—Plastids reproduced to cover the same area, actual magnifications diverge around 20,000 ×.

FIGURES 15–22. Sieve-element plastids of families often allied to Myrtales. S-type plastids in Haloragaceae.—15. *Haloragis erecta*.—16. *Myriophyllum brasiliense*, P-type plastids in Rhizophoraceae.—17. *Rhizophora* cf. *conjugata*, S-type plastids in Thymelaeaceae.—18. *Daphne mezereum*, Lecythidaceae.—19. *Couroupita gui-*



anensis, Coridaceae.—20. *Coris monspeliensis* and Chrysobalananceae.—21. *Chrysobalanus icaco*, S_0-plastids in Elaeagnaceae.—22. *Elaeagnus angustifolius*, young sieve element. Plastids reproduced to cover the same area, actual magnifications diverge around 20,000 ×.

TABLE 1. Sieve-element plastids in Myrtales and allied groups.

Family	Species	Source	Type of Plastids
Melastomataceae	*Amphiblemma cymosum* (Schrad.) Naud.	Utrecht	S
	Bertolonia maculata (Mart.) DC.	Edinburgh	S
	B. marmorata Naud.	Heidelberg	S
	Bredia tuberculata (Guillamin) Diels	Edinburgh	S
	Calvoa orientalis Taub.	Copenhagen	S
	Centradenia floribunda Planch.	Edinburgh	S
	Clidemia hirta (L.) D. Don	Bonn	S
	Dissotis rotundifolia (Sm.) Triana	Copenhagen	S
	Gravesia guttata (Hook.) Triana	Edinburgh	S
	Heterocentron subtriplinervium (Link & Otto) R. Br.	Edinburgh	S
	Medinilla magnifica Lindl.	Heidelberg	S
	Melastoma sanguineum D. Don	Edinburgh	S
	Memecylon sp.	Utrecht	S
	Monochaetum humboldtianum Walp.	Edinburgh	S
	Monolena primuliflora Hook. f.	Copenhagen	S
	Pachycentria constricta Bl.	Bogor	S
	Sakersia africana Hook.	Parc Nat. Comoé, Côte d'Ivoire (Leg. Merz, Téhé & Assi)	S
	Schizocentron elegans (Schl.) Meissn.	Edinburgh	S
	Tibouchina semidecandra (Schrank & Mart.) Cogn.	Heidelberg	S
	Tococa guianensis Aubl.	Heidelberg	S
	Triolena pustulata Triana	Heidelberg	S
	T. scirpioïdes Naud.	Copenhagen	S
Crypteroniaceae	*Crypteronia paniculata* Bl.	Bogor	S
Penaeaceae	*Penaea mucronata* L.	Kirstenbosch (J. P. Rourke)	S
	Saltera sarcocolla (L.) Bullock	Cape Point Nature Reserve (leg. D. Clark)	S
Oliniaceae	*Olinia emarginata* Burtt-Davy	B. I. Pretoria (D. M. C. Fourie)	S
Combretaceae	*Combretum racemosa*	Kew	S
	C. quadrangulare Kurz.	Bogor	S
	C. sundaicum Miq.	Bogor	S
	Quisqualis indica L.	Heidelberg	S
	Terminalia catappa L.	Heidelberg	S
Lythraceae			
subfam. Lythroideae	*Cuphea micropetala* H.B.K.	Berlin	S
	Lafoensia punicaefolia DC.	Bogor	S
	Lagerstroemia indica L.	Copenhagen	S
	Lawsonia inermis L.	Copenhagen	S
	Lythrum salicaria L. cv Robert	Heidelberg	S
	Rotala rotundifolia Buch.-Ham.	Mainz	S
subfam. Duabangoideae	*Duabanga sonneratioïdes* Buch.-Ham.	Rio de Janeiro (leg. K. Kutitzki)	S
subfam. Sonneratioideae	*Sonneratia caseolaris* (L.) Engl.	Bogor	S
subfam. Punicoideae	*Punica granatum* L.	Heidelberg	S
Rhynchocalycaceae	*Rhynchocalyx lawsonioïdes* Oliv.	Natal (leg. H. B. Nicholson)	S

TABLE 1. Continued.

Family	Species	Source	Type of Plastids
Myrtaceae	*Acca sellowiana* Burret	Heidelberg	S
	Agonis flexuosa (Willd.) Lindl.	Kew	S
	Angophora cordifolia Cav.	Kew	S
	Callistemon phoeniceus Lindl.	Heidelberg	S
	Calothamnus rupestris Schau	Mainz	S
	Eucalyptus diversifolius Bonpl.	Heidelberg	S
	Eugenia myrcianthes Niedenzu	Copenhagen	S
	Hexaclamys edulis Kausel ex D.	Mainz	S
	Kunzea ambigna (Sm.) Hochr.	Kew	S
	Leptospermum laevigatum F. Muell.	Heidelberg	S
	Lophomyrtus obcordata (Raoul) Burret	Utrecht	S
	Melaleuca acuminata F. Muell.	Heidelberg	S
	Metrosideros excelsa Soland ex Gaertn.	Heidelberg	S
	Myrceugenia luma Berg.	Mainz	S
	Myrtus communis L.	Heidelberg	S
	Pimenta racemosa (Mill.) J. W. Morre	Bonn	S
	Psidium littorate Raddi	Berlin	S
	Rhodamnia cinerea Jack.	Leila Forest, Sandakan, Sabah (leg. Behnke & Lee 83-07-22)	S
	Tristania conferta R. Br.	Leiden	S
Trapaceae	*Trapa natans* L.	Bonn	S
Onagraceae	*Circaea cordata* Royle	Heidelberg	S
	C. × *intermedia* Ehrh.	Zastler, Germany (leg. Behnke & Cole 80-08-26/1)	S
	Epilobium fleischeri Hochst.	Heidelberg	S
	Fuchsia arborescens Sims.	Heidelberg	S
	Godetia amoena G. Don.	Kew	S
	Oenothera biennis L.	Heidelberg	S
	O. missouriensis Sims.	Heidelberg	S
	Zauschneria californica Presl.	Heidelberg	S
Families allegedly allied (after Dahlgren & Thorne, 1984)			
Haloragaceae	*Haloragis erecta* (Banks & Murr) Eichl.	Copenhagen	S
	Myriophyllum brasiliense Cambess	Bonn	S
Rhizophoraceae	*Bruguiera gymnorrhiza* Lam.	Berlin	PVc
	Carallia brachyata	Brisbane, Australia (leg. R. Tracey)	PVc
	Cassipourea elliptica (Sw.) Poir.	Finca La Selva Costa Rica (Juncosa 26 VIII 74)	PVc
	C. cf. *killipii* Cuatrecasas	Chocó, Colombia (Juncosa 2540)	PVc
	Ceriops tagal var. *australis* C. T. White	Cape Ferguson, Queensland (leg. G. J. Muller)	PVc
	Crossostylis biflora Forst.	Edinburgh	PVc
	C. grandiflora Brongn. & Gris.	Mt. Panié, New Caledonia (Juncosa 20 IX 81A)	PVc
	Kandelia rheedii Wight & Arn.	Brisbane, Australia (Leg. R. Tracey)	PVc
	Rhizophora cf. *conjugata*	Bonn	PVc

TABLE 1. Continued.

Family	Species	Source	Type of Plastids
	R. mangle L.	Heidelberg	PVc
	R. sexangula	Copenhagen	PVc
	R. stylosa	Australia (leg. R. Tracey)	PVc
	Sterigmapetalum heterodoxum Steyem. & Liesner	Sierra de San Luis (Berry & Wingfielá 4304)	PVc
Anisophylleaceae	*Anisophyllea trapezoidales* Baill.	Sepilol Forest Res., Sandakan, Sabah (leg. Behnke & Lee 83-7-21)	PVc
	Combretocarpus cf. *motleyi* Hook. f.	Sri Aman, Sarawak (Leg. Othman Ismawi)	S
Thymelaeaceae	*Dais cotonifolia* L.	Mainz	S
	Daphne mezereum L.	Heidelberg	S
	Passerina filiformis L.	Bonn	S
	Phaleria disperma Baill.	Kew	S
	Pimelea ferruginea Labill.	Bonn	S
	Wikstroemia sp.	Kew	S
Lecythidaceae	*Barringtonia acutangula* (L.) Gaertn.	Bogor	S
	B. speciosa Forst.	Copenhagen	S
	Chydenanthus excelsus (Bl.) Miers	Bogor	S
	Couroupita guianensis Aubl.	Berlin	S
	Gustavia angusta L. f.	Berlin	S
	G. gracillinea Miers	Rio de Janeiro (K. Kubitzki)	S
	Napoleonaea imperialis Beauv.	Kew	S
	Planchonia vallida Bl.	Bogor	S
Elatinaceae	*Elatine hydropiper* L.	Berlin	(S_o)
Coridaceae	*Coris monspeliensis* L.	Berlin	S
Chrysobalanaceae	*Chrysobalanus icaco* L.	Berlin	S
	Couepia grandiflora (Mart. & Zucc.) Benth. ex Hook f.	Botucatu, Brazil (leg. I. S. Gottsberger)	S
	Moquilea tomentosa	Kew	
Elaeagnaceae	*Elaeagnus angustifolius* L.	Heidelberg	S_o
	E. umbellata Thunb.	Heidelberg	S_o
	Hippophae rhamnoïdes L.	Heidelberg	S_o
Families distantly related (all families mentioned under this heading by Dahlgren & Thorne (1984) contain S-type plastids except for:)			
Gunneraceae	*Gunnera magellanica* Lam.	Copenhagen	P
	G. manicata Lindl. ex André	Copenhagen	P
	G. tinctoria Mirbel	Bonn	P
Connaraceae	*Agelaea macrophylla* (Zoll.) Leenh.	Bogor	P
	Connarus conchocarpus	Atherton, Qld, Australia (leg. A. Irvine)	P
	C. oblongus Schellenb.	Bogor	P
	C. suberosus Planch.	Botucatu, Brazil (leg. I. S. Gottsberger)	P
	Rourea mimosoïdes (Val.) Planch.	Bogor	P
	Roureopsis emarginata (Jack.) Merr.	Bogor	P
Rhabdodendraceae	*Rhabdodendron amazonicum* (Spruce ex Benth.) Hub.	Manaus, Brazil (leg. G. T. Prance)	P
	R. macrophyllum (Spruce ex Benth.) Hub.	Manaus, Brazil (Prance 20187)	P

dition (see e.g., Figs. 3, 8), and all grains may show a tendency to disintegrate at their periphery into tiny particles—most prominent, e.g., in *Callistemon* (Fig. 10), *Epilobium* (Fig. 13), and *Circaea* (Fig. 16). These features make the sieve-element plastics of the core families in Myrtales not very distinctive, but exclude at least some starch forms, like the one-large-grain, the club-shaped, and the completely particulated starches.

Most of the suggested closer allies and the more or less distantly related families of Myrtales also contain S-type plastids (see Table 1), very few of which, however, fit the starch features described for the core families (e.g., Figs. 15, 18, 21; probably also Fig. 19 but note very large grains). More informative is the absence of S-type plastids in some of the families. While its quantity is much decreased in *Myriophyllum* (Fig. 16), starch is completely lost in Elaeagnaceae (Fig. 22) and probably also in Elatinaceae. Four families contain P-type sieve-element plastids—Rhizophoraceae (Fig. 17) of the closer allies and Connaraceae (Behnke, 1982c), Gunneraceae (see Behnke, 1981a), and Rhabodendraceae (Behnke, 1976) among the groups sometimes related. All of the latter families containing either subtype S_0 or P-type plastids should be excluded from the Myrtales.

Elaeagnaceae (S_0-plastids) contain crystalline P(hloem)-protein bodies, another sieve-element character which among the myrtalean core families is only found in Myrtaceae. However, the ultrastructural composition of the crystalline P-protein is unlike that of Myrtaceae, but comes very close to crystalline P-protein in Proteaceae, which is also among the distantly related group. However, sieve-element plastids in Proteaceae are of S-type.

The family Rhizophoraceae needs special mention, since according to traditional treatments it incorporates both S-type and P-type genera. While the 12 species examined from the tribes Gynotrocheae, Macarisieae, and Rhizophoreae (see Table 1) contain very specific sieve-element plastids (Fig. 17) that include numerous protein crystals but no starch at all (subtype-PV; the only other families recorded to have this subtype are Cyrillaceae and Erythroxylaceae, Behnke, 1982b), *Anisophyllea* and *Combretocarpus* were found to have S-type plastids. If further investigations on other genera and other characters (e.g. from vegetative and generative morphology, chromosome cytology) would corroborate the plastid diversity, a separation of the Anisophylleae as a distinct family, Anisophyllaceae, could be substantiated.

Certainly sieve-element plastids cannot be used as a critical character to positively shape the order Myrtales, but are helpful in negating close relationships of some of the peripheral families.

Literature Cited

Behnke, H.-D. 1974. P- und S-Typ Siebelement-Plastiden bei Rhamnales. Beitr. Biol. Pflanzen 50: 457–464.

———. 1976. Ultrastructure of sieve-element plastids in Caryophyllales (Centrospermae), evidence for the delimitation and classification of the order. *In* T. J. Mabry & H.-D. Behnke (editors), Evolution of Centrospermous Families. Pl. Syst. Evol. 126: 31–54.

———. 1981a. Sieve-element characters. *In* H.-D. Behnke (editor), Ultrastructure and Systematics of Seed Plants. Nord. J. Bot. 1: 381–400.

———. 1981b. Siebelement-Plastiden, Phloem-Protein und Evolution der Blütenpflanzen: II. Monokotyledonen. Ber. Deutsch. Bot. Ges. 94: 647–662.

———. 1982a. Sieve-element plastids, exine sculpturing and the systematic affinities of the Buxaceae. Pl. Syst. Evol. 139: 257–266.

———. 1982b. Sieve-element plastids of Cyrillaceae, Erythroxylaceae and Rhizophoraceae. Presentation and significance of subtype-PV plastids. Pl. Syst. Evol. 141: 31–39.

———. 1982c. Sieve-element plastids of Connaraceae and Oxalidaceae. A contribution to the knowledge of P-type plastids in dicotyledons and their significance. Bot. Jahrb. Syst. 103: 1–8.

——— & L. Pop. 1981. Sieve-element plastids and crystalline P(hloem)-protein in Leguminosae: micromorphological characters as an aid to the circumscription of the family and subfamilies. Pp. 707–715, *in* R. M. Polhill & P. H. Raven (editors), Advances in Legume Systematics. Academic Press, London.

Dahlgren, R. M. T. 1980. A revised system of classification of the angiosperms. Bot. J. Linn. Soc. 80: 91–124.

Dahlgren, R. & R. F. Thorne. 1984 [1985]. The order Myrtales: circumscription, variation, and relationships. Ann. Missouri Bot. Gard. 71: 633–699.

Mabry, T. J. 1977. The order Centrospermae. Ann. Missouri Bot. Gard. 64: 210–220.