

The Palms

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Variations in the microsporogenesis of monosulcate palm pollen

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Pollen morphology has been extensively studied in the Arecaceae, and pollen aperture organization is usually distal monosulcate, as in many monocot families. Much is known about the influence of microsporogenesis on aperture configuration, but the key processes during microsporogenesis responsible for aperture type, number and arrangement are still poorly understood. In order to clarify the developmental sequence underlying aperture type and organization in palm monosulcate pollen, a study of the characteristics of male postmeiotic development was carried out in representative species of four genera of subfamily Coryphoideae, and four genera of subfamily Arecoideae. We found evidence for the occurrence of successive cytokinesis in addition to simultaneous cytokinesis in three Coryphoideae species. Tetrad shape was highly diverse within all species. Our results reveal an unexpected diversity in microsporogenesis from which it may be possible to gain further insight into pollen evolution within the family. © 2006 The Linnean Society of London, *Botanical Journal of the Linnean Society*, 2006, **151**, 93–102.

ADDITIONAL KEYWORDS: aperture configuration – callose deposition – cytokinesis – evolution – postmeiotic development – tetrads.

INTRODUCTION

The Arecaceae comprise 2364 currently recognized species (Govaerts & Dransfield, 2005). The genera are distributed widely in the tropics and subtropics, and several genera are cool-tolerant. Palms are highly distinctive at the family level but, within the family, their morphological diversity is probably greater than that of any other monocotyledonous family (Uhl & Dransfield, 1987).

At the microscopic level this diversity is also found in the pollen morphology, for example, Sowunmi (1968, 1972), Thanikaimoni (1970), Dransfield, Ferguson & Uhl (1990), Ferguson & Harley (1993), Harley

(1990, 1999a), Harley & Baker (2001), Harley & Dransfield (2003). Shape, exine ultrastructure, exine ornamentation, pollen size, aperture form, number and arrangement all show a wide range of variation. Apertures are areas where the ectexine (outer layer of the exine) is usually very thin or more or less absent, thus providing greater flexibility for the mature pollen grain to contract or expand in response to variations in osmotic pressure, a phenomenon which Wodehouse (1935) termed 'harmomegathy'. The ability for pollen apertures to expand or contract, when exposed to differing environments, is important to the survival of the male gametes carried by the pollen grains. In transit from anther to stigma the apertures can contract to avoid desiccation of the internal cellular fraction while, on arrival on a receptive stigmatic surface, the apertures can expand to allow the swelling intine to

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emerge, and subsequent pollen tube growth to proceed rapidly.

Twelve aperture types, and inaperturate pollen in the small genus *Pigafetta*, have been described for the Areceaceae (Harley & Baker, 2001). All these aperture types are based on variations in the combination of a number of characteristics: aperture shape and size; aperture position in relation to polarity – distal polar, equatorial, or meridional; apertures with or without an operculum; number of apertures and symmetry of aperture position or spacing. However, the most widespread and presumed basic pollen type in palms, as elsewhere in the monocotyledons (Dahlgren & Clifford, 1982), is distal monosulcate with only one aperture in the shape of sulcus, located at the distal pole of the pollen grain.

Pollen morphology has been studied extensively in the Areceaceae, and much is known about the influence of microsporogenesis on different types of aperture configuration. On the basis of published data and new observations, Ressayre *et al.* (2002) developed a morphogenetic model that established a link between microsporogenesis and aperture pattern. The ontogeny of aperture pattern occurs during microsporogenesis and four features of this developmental process are particularly involved in aperture pattern definition. These features are: the type of cytokinesis, the way callose walls are formed among the four microspores, the tetrad shape and the position of the apertures within the tetrads. The model suggests that combining the variations in these characters accounts for most aperture types observed in angiosperms. Previously published observations of microsporogenesis in a range of angiosperm species offer notable support to this model (Periasamy & Swamy, 1959; Murty, 1964; Sampson, 1969, 1975; Blackmore & Barnes, 1995; Blackmore & Crane, 1998; Furness & Rudall, 1999; Ressayre, 2001; Ressayre *et al.*, 2002, 2003, 2005; Penet *et al.*, 2005). In particular, this model suggests

that distal monosulcate pollen grain could be obtained through different developmental pathways, a prediction confirmed by Penet *et al.* (2005) in a recent study focusing on the Asparagales. Exploring pollen aperture ontogeny in palms is particularly relevant from an evolutionary point of view because palms are isolated within the Commelinids, which are themselves sister to the Asparagales.

This paper presents a preliminary investigation of the characteristics of aperture pattern ontogeny in palms, describing microsporogenesis in eight species producing distal monosulcate pollen grains, four from subfamily Coryphoideae, and four from subfamily Arecoideae. In some of the Arecoideae examples, monosulcate pollen is sometimes associated with trichotomosulcate pollen grain (a triradiate variant of monosulcate) as recorded in the literature (Harley, 1999a). Three key developmental characteristics of microsporogenesis were observed: cytokinesis type, callose deposition between the microspores, and tetrad type.

Results so far reveal a diversity in microsporogenesis from which we may be able to gain further insight into pollen evolution within the family.

MATERIAL AND METHODS

Pollen material was sampled from the Montgomery Botanical Centre (FL, USA) in January 2004 and January 2005. Eight species of palms were selected for this study, representing eight genera, four that belong to the Coryphoideae and four from the Arecoideae. Species and Montgomery accession numbers are listed in Table 1.

Fresh flower buds were collected at different developmental stages and the anthers were extracted and immediately squashed. Depending on the stage observed, different staining techniques were used. Cytokinesis type was identified by staining with aceto-

Table 1. List of the species examined in this study. Systematic position is according to Dransfield *et al.* (2005). Accession numbers of living material used in this study at the Montgomery Botanical Centre are given

Subfamily and tribe	Genus & species	Accession number
Coryphoideae		
Livistoneae	<i>Serenoa repens</i> (W. Bartram) Small	20040680.A; Wild
	<i>Copernicia hospita</i> Mart.	94607.A, B
Caryoteae	<i>Caryota mitis</i> Lour.	87319.A, C; 89225.D; 89899.C
Borasseae	<i>Hyphaene coriacea</i> Gaertn.	951155.C, F, J
Arecoideae		
Cocoseae	<i>Allagoptera arenaria</i> (M. Gómez) Kuntze	97807.A, D, M, N; 97805.A, C; 71442.H, L
	<i>Butia</i> sp.	976.H
Areceae	<i>Dypsis decaryi</i> (Jum.) Beentje & J. Dransf.	951204.B, D
	<i>Veitchia spiralis</i> H. Wendl.	961336.N, Q

Table 2. Number of observed individuals, flower buds and stamens per species, and levels of microsporogenesis variation in each species. AC, aceto-carminic; AB, aniline blue

Species	Number of		
	Individuals observed (AC-AB)	Flower buds observed (AC-AB)	Stamens observed (AC-AB)
<i>Serenoa repens</i>	3-3	9-16	21-42
<i>Copernicia hospita</i>	1-2	6-16	9-44
<i>Caryota mitis</i>	1-1	1-3	4-6
<i>Hyphaene coriacea</i>	3-3	8-22	18-62
<i>Allagoptera arenaria</i>	4-3	5-7	13-33
<i>Butia</i> sp.	1-1	3-5	5-12
<i>Dypsis decaryi</i>	1-1	1-6	4-17
<i>Veitchia spiralis</i>	2-1	2-3	12-18

carminic (67.5 mL acetic acid, 0.75 g carminic, 0.025 g ferrous acetate, made up to 150 mL with distilled water). A dyad stage is characteristic of successive cytokinesis while four nuclei in the same cytoplasm is characteristic of simultaneous cytokinesis. The progression of intersporal wall formation was followed by staining with aniline blue, modified from the method of Arens (1949), with the addition of glycerol to 15% of the final volume. With this method, the callose contained in the Golgi vesicles forming the cell plates becomes fluorescent when illuminated by UV light. Tetrad shape was also identified using this staining method.

Whenever possible, several individuals, several flower buds per individual and several stamens per flower bud were sampled and observed for each developmental stage. Table 2 gives the details of the material sampled for each species. Owing to problems in the availability of plant material and technical constraints, sometimes only one individual could be examined, as in the case of *Butia* sp. Furthermore, several developmental stages, such as the formation of the cell plates, take place during a very short time and are therefore difficult to find.

Aceto-carminic preparations were observed using a Zeiss Axiophot microscope (light microscopy). Aniline blue preparations were observed using the epifluorescence option of this microscope with a DAPI filter (filter set at 01; excitation at 345, emission at 425 nm long pass) or confocal microscopy with the same type of filter.

RESULTS

Our initial observations reveal that during microsporogenesis, characteristics of cytokinesis, intersporal wall formation and the form of the resulting tetrads are variable in palms (Table 3), although only one pollen type, bisymmetrical to highly asym-

metrical distal monosulcate, is produced in the species studied: *Veitchia spiralis* (Figs 1-4), *Dypsis decaryi* (Figs 5-7), *Allagoptera arenaria* (Figs 8-11), *Butia* sp. (Figs 12-16), *Copernicia hospita* (Figs 17-22), *Serenoa repens* (Figs 23-28), *Hyphaene coriacea* (Figs 29-36) and *Caryota mitis* (Figs 37-42).

VARIATION IN CYTOKINESIS

In Angiosperms male meiotic cell division can be simultaneous or successive. In the first case, the cytoplasmic division occurs simultaneously, after the second meiotic division. The presence of four nuclei in the same cytoplasm constitutes the characteristic stage of this type of cytokinesis. Successive cytokineses are characterized by a dyad stage that corresponds to the cytoplasmic division taking place after the first meiotic division.

In the present study, cytokinesis was of the typical simultaneous type only in *V. spiralis* (Fig. 1) and *C. hospita* (Fig. 17). In the other Arecoideae (*D. decaryi*, *A. arenaria* and *Butia* sp.), the cytokinesis is neither simultaneous nor successive. It is an intermediate situation described further on. In the three other Coryphoideae, simultaneous cytokinesis was observed in association with successive cytokinesis within the same anther, as shown by the record of dyads in addition to microsporocytes with all nuclei in the same cytoplasm in *H. coriacea* (Figs 29, 30) and *C. mitis* (Figs 37-39). Dyads were also recorded in *S. repens* (Fig. 23), in which the occurrence of simultaneous cytokinesis is suggested by the simultaneous formation of the intersporal walls observed with aniline blue staining (Fig. 24). Although no statistics were made, we estimated the occurrence of successive cytokinesis at around two percent.

A particular situation was observed in *D. decaryi*, *A. arenaria*, *Butia* sp. and *H. coriacea*, where a single

Table 3. Summary of microsporogenesis characteristics for the species examined. An asterisk indicates that cytokinesis is simultaneous, but one cell plate is initiated before the other three. Only stages with four nuclei in the same cytoplasm were observed with aceto-carmine staining

Species	Additional callose Cytokinesis deposits	Cell wall formation	Tetrad shape
<i>Serenoa repens</i>	Simultaneous + successive Present	Centrifugal	Irregular tetrahedral, Tetragonal, Decussate, Linear, T
<i>Copernicia hospita</i>	Simultaneous Absent	Centripetal	Rhomboidal, Irregular tetrahedral
<i>Caryota mitis</i>	Simultaneous + successive Present	Centrifugal	Rhomboidal, Irregular tetrahedral, Tetragonal, Decussate
<i>Hyphaene coriacea</i>	Simultaneous (+ ephemeral cell plate) + successive Present	Centripetal	Rhomboidal, Irregular tetrahedral, Tetragonal, Decussate
<i>Allagoptera arenaria</i>	Simultaneous (+ ephemeral cell plate) Absent	Centripetal	Rhomboidal, Irregular tetrahedral
<i>Butia</i> sp.	Simultaneous* Present	Centrifugal	Rhomboidal, Irregular tetrahedral, Tetragonal, Decussate
<i>Dypsis decaryi</i>	Simultaneous* Present	Centrifugal	Rhomboidal, Irregular tetrahedral
<i>Veitchia spiralis</i>	Simultaneous Present	Centrifugal	Rhomboidal, Irregular tetrahedral

isolated cell was observed in the centre of several microsporocytes with aniline blue staining (Figs 5, 10, 14, 31). In *A. arenaria* and *H. coriacea* this cell, which was thickly covered with callose, was not observed in later stages. No dyad stage (characteristic of successive cytokinesis) was recorded with aceto-carmine staining in *D. decaryi*, *A. arenaria* and *Butia* sp. Only microsporocytes in which the four nuclei were visible in the same cytoplasm (characteristic of simultaneous cytokinesis) were recorded in *A. arenaria* and *Butia* sp. (Figs 8, 12). In *H. coriacea* however, both dyads and microsporocytes with four nuclei in the cytoplasm were recorded (Figs 29, 30).

VARIATION IN INTERSPORAL WALL FORMATION

In the species examined, cytokinesis was achieved by either centrifugal or centripetal cell wall formation. Cell wall formation achieved by centrifugally developing cell, was recorded in *V. spiralis* (Fig. 2), *D. decaryi* (Fig. 6), *Butia* sp. (Fig. 13), *S. repens* (Figs 24, 26) and *C. mitis* (Fig. 40). Centripetal wall formation was observed in *A. arenaria* (Fig. 9), *C. hospita* (Fig. 18) and *H. coriacea* (Fig. 32).

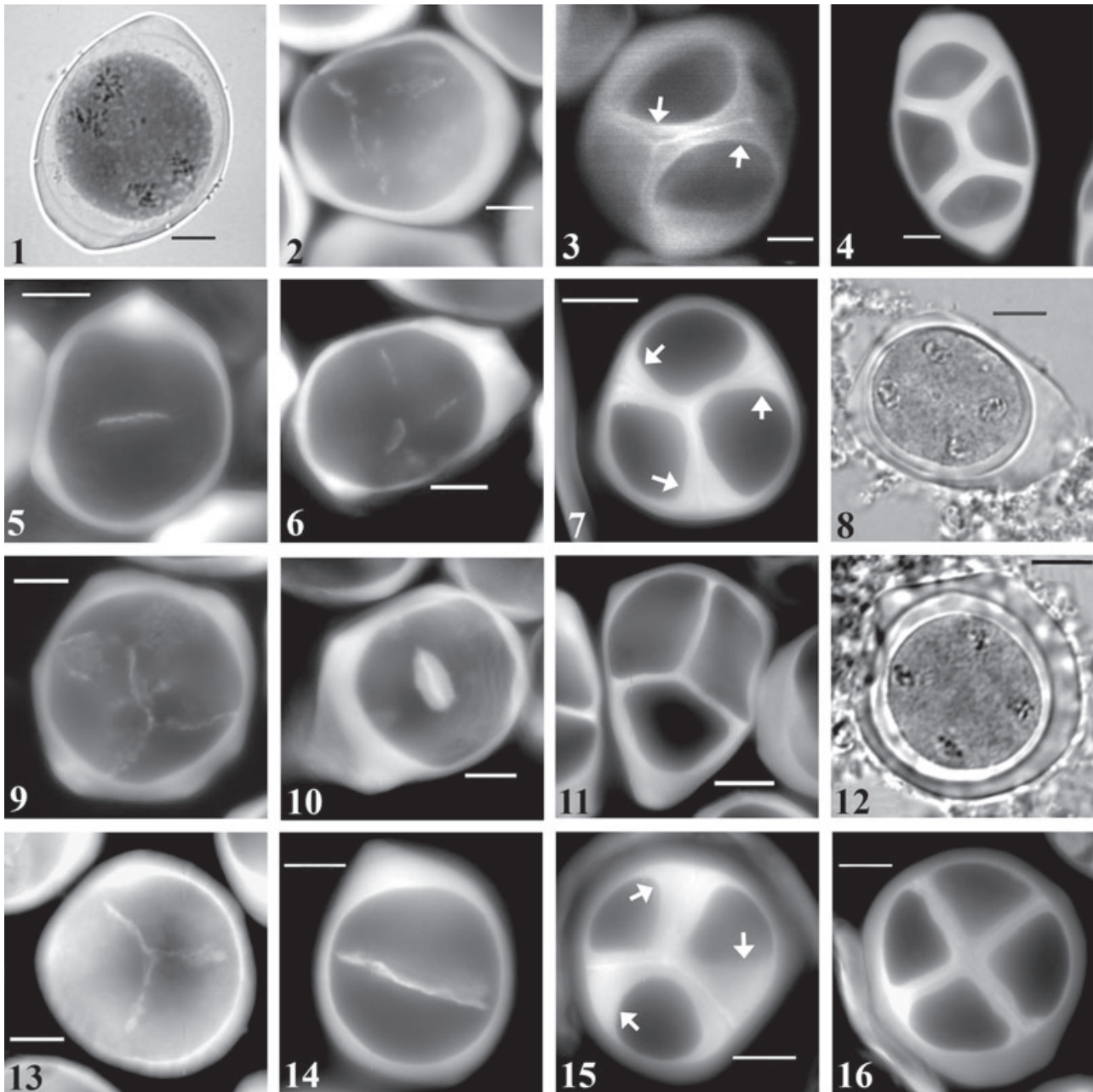
In some cases, additional callose was laid onto the cell walls. These callose deposits were located at the junction between the intersporal walls and the outer wall of the tetrad in *V. spiralis* (Fig. 3), *D. decaryi*

(Fig. 7), *Butia* sp. (Fig. 15), *S. repens* (Fig. 25), *H. coriacea* (Fig. 34) and in *C. mitis* (Figs 41, 42). In *H. coriacea* and *C. mitis*, there were also callose deposits in the centre of the tetrad (Figs 33, 42). The thickness of the deposits varied among species; they were particularly thick in *D. decaryi* (Fig. 7) and *Butia* sp. (Fig. 15). In *A. arenaria* and *C. hospita* additional callose deposits were absent.

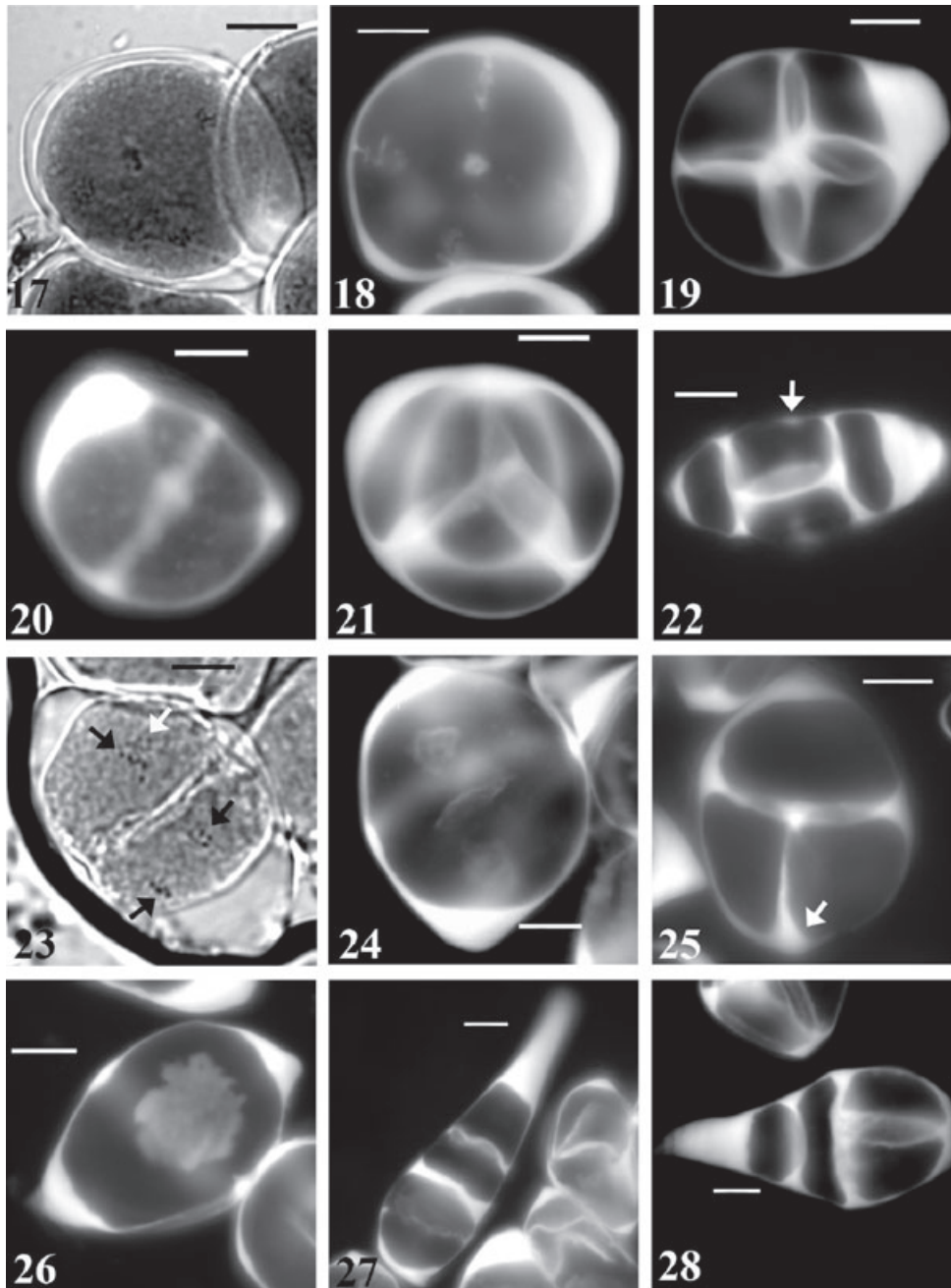
TETRAD SHAPE VARIATIONS

Tetrad shape varied between tetragonal, for example in *Butia* sp. (Fig. 16), *H. coriacea* (Fig. 36) and *C. mitis* (Fig. 42), rhomboidal, for example in *V. spiralis* (Fig. 4) and *C. hospita* (Fig. 22), and tetrahedral, for example in *A. arenaria* (Fig. 11) and *C. hospita* (Figs 19, 21). Although the proportions of the different shapes were not assessed, this last form was observed most frequently. It should be noted that all the tetrahedral tetrads observed were irregular, i.e. with six unequal walls separating the four microspores. All these forms co-occur within most species and furthermore within a single anther. In *S. repens* linear tetrads (Fig. 27) and T-shaped tetrads (Fig. 28) were recorded in addition to the shapes already mentioned.

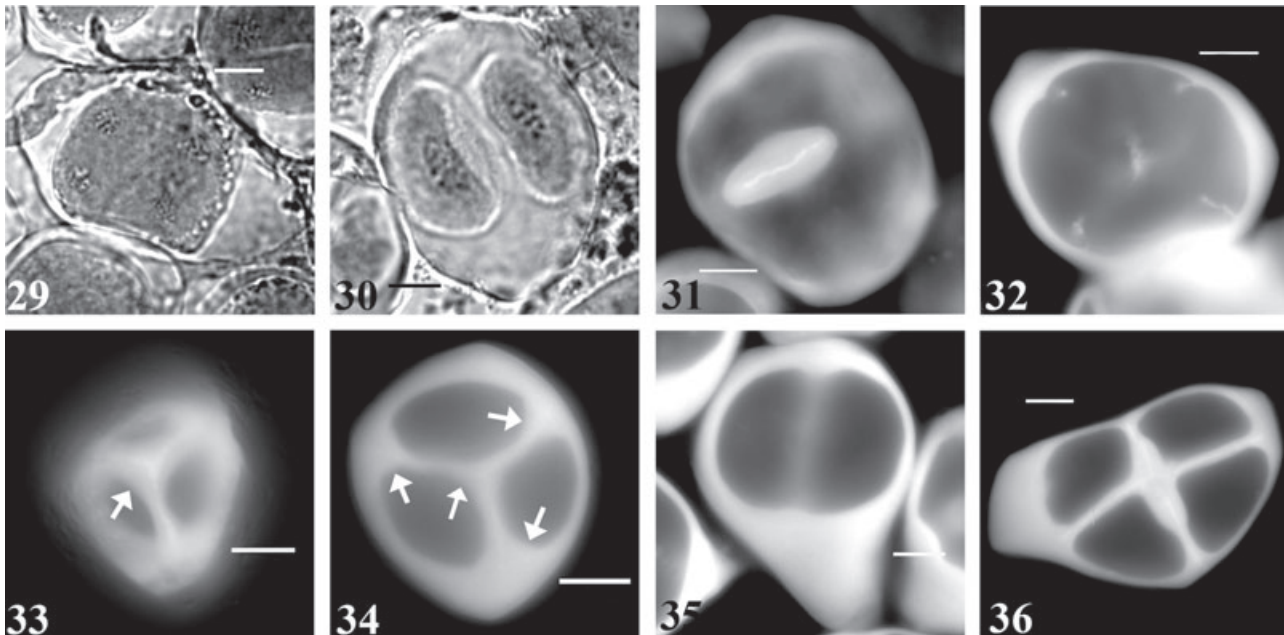
In *C. hospita*, *S. repens*, and *H. coriacea* the presence of a callose ring was noted (Figs 22, 26, 35). This callose ring is present at an early stage. It is already



Figures 1–16. Features of microsporogenesis in four species of Arecoideae: *Veitchia spiralis* (Figs 1–4), *Dypsis decaryi* (Figs 5–7), *Allagoptera arenaria* (Figs 8–11) and *Butia* sp. (Figs 12–16) (Figs 1, 8, 12: aceto-carmine; Figs 2–7, 9–11, 13–16: aniline blue). Fig. 1. Simultaneous cytokinesis with four nuclei in the same cytoplasm. Fig. 2. Cell plates developing from the centre of each cleavage plane. Fig. 3. Upper section, cell plates are covered by additional callose deposits located at the intersection between the cleavage planes and the outer wall of the tetrad (specified by arrows). Fig. 4. Rhomboidal tetrad in which the cell plates are covered with callose. Fig. 5. Single cell plate growing in the centre of the mother cell. Fig. 6. Cell plates developing centrifugally from the centre of each cleavage plane. Fig. 7. Middle section of a tetrahedral tetrad. Additional callose deposits are thicker towards the edges of the tetrad (arrows). Fig. 8. Simultaneous cytokinesis. Fig. 9. The six walls of a tetrahedral tetrad form from the edges of each cleavage plane towards their centre. Fig. 10. Early stage: a plate, thickly covered with callose, forms centrifugally in the centre of the cell. Fig. 11. Irregular tetrahedral tetrad without any additional callose deposits. Fig. 12. Simultaneous cytokinesis with four nuclei in the same cytoplasm. Fig. 13. Developing cell walls (only three walls are visible here) in a future tetrahedral tetrad. Fig. 14. Ephemeral cell plate developing centrifugally. Fig. 15. Middle section of an irregular tetrahedral tetrad. Additional callose deposits are thicker towards the junction of intersporal walls and the mother cell wall (arrows). Fig. 16. Tetragonal tetrad. Scale bars = 10 μ m.



Figures 17–28. Cytokinesis, the way in which callose walls and additional callose deposits are formed in two species of Coryphoideae: *Copernicia hospita* (Figs 17–22) and *Serenoa repens* (Figs 23–28) (Figs 17, 23: aceto-carmin; Figs 18–22, 24–28: aniline blue). Fig. 17. Simultaneous cytokinesis with four nuclei in the same cytoplasm. Fig. 18. Six cell plates progress from the edges of each cleavage plane. Wall formation begins simultaneously from the tetrad wall and from the centre of the tetrad. Fig. 19. Irregular tetrahedral tetrad at a later stage without callose deposits. Fig. 20. Early stage: presence of a callose ring surrounding the mother cell. Fig. 21. Irregular tetrahedral tetrad. Fig. 22. Rhomboidal tetrad on which a shadow of a callose ring is visible (arrow). Fig. 23. Successive cytokinesis, telophase II with two cells including both nuclei. The two pairs of nuclei are orthogonal (arrows). Fig. 24. Three visible plates grow from the centre of each cleavage plane, a weak callose ring is visible. Fig. 25. Middle section, additional callose deposits progress from the outer wall of the tetrad towards its centre in an irregular tetrahedral tetrad. Therefore, the additional callose deposits are thicker at the intersections between each cleavage planes and the outer wall (arrows). Fig. 26. Formation of a centrifugal cell plate in the centre of the mother cell, presence of a callose ring. Fig. 27. Linear tetrad. Fig. 28. T-shaped tetrad. Scale bars = 10 μ m.



Figures 29–36. Cytokinesis, the way in which callose walls and additional callose deposits are formed in one species of Coryphoideae, *Hyphaene coriacea* (Figs 29–30: aceto-carmin; Figs 31–36: aniline blue). Fig. 29. Simultaneous cytokinesis with four nuclei in the same cytoplasm. Fig. 30. Dyad stage, successive cytokinesis. Fig. 31. Early stage – a plate, thickly covered with callose, forms centrifugally in the centre of the cell. Fig. 32. Six cell plates progress simultaneously from the tetrad wall and from the tetrad centre. Figs 33–34. Irregular tetrahedral tetrad with additional callose deposits thicker (Fig. 33: upper section; Fig. 34: middle section). So, they are most important at the intersections between the cleavage planes (arrows). Fig. 35. Early stage: presence of a callose ring. Fig. 36. Tetragonal tetrad with additional callose deposits only on the first formed plate. Scale bars = 10 μ m.

visible before the initiation of cell formation and persists throughout microsporogenesis.

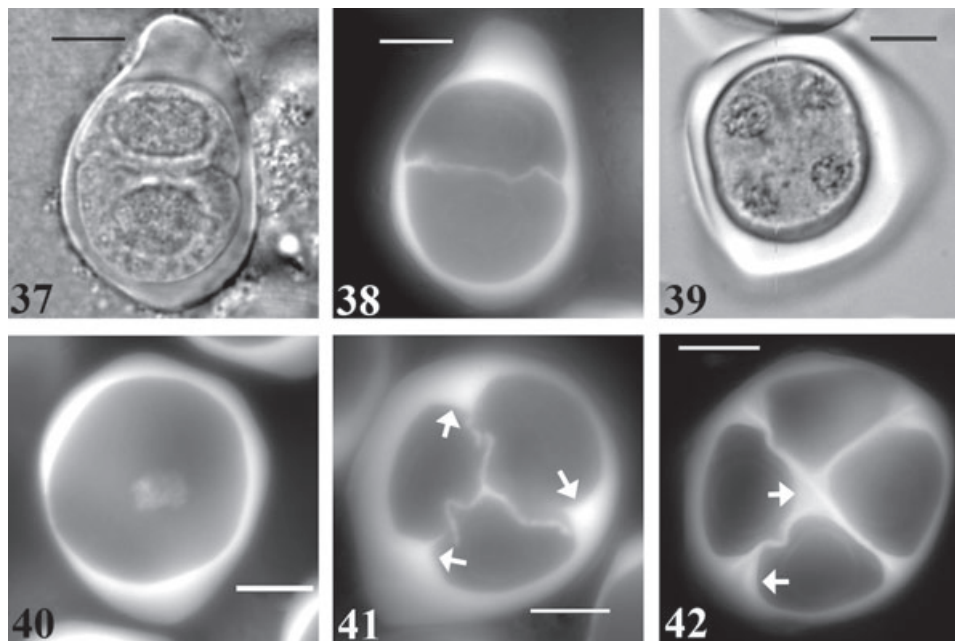
DISCUSSION

COMPARISON WITH OTHERS ANGIOSPERMS

In monocots, meiotic cytokinesis is predominantly successive and associated with centrifugal cell plates (Dahlgren & Clifford, 1982; Furness & Rudall, 1999), and tetragonal tetrads. Simultaneous cytokinesis has been recorded in several families of the Asparagales, Dioscoreales and Cyperales (Furness & Rudall, 1999). The situation we describe within the palm species examined is unusual because meiotic cytokinesis can be strictly simultaneous or both simultaneous and successive. The co-occurrence of successive and simultaneous cytokinesis within the same species, such as our examples in Coryphoideae, is occasionally described elsewhere, for example, in *Trichopus* (Dioscoreaceae, Rao 1955) where dyad stages are recorded, but the resultant tetrads are isobilateral, decussate, linear or tetrahedral in arrangement. It is generally admitted that the latter tetrad shape can be obtained only with simultaneous cytokinesis, suggest-

ing that cytokinesis can be simultaneous as well as successive in *Trichopus*. In *Tritonia* (Iridaceae) true dyad stages, as well as four cell plates developing simultaneously, have been observed (Penet *et al.*, 2005). In palms, the two types of cytokinesis occur within the same stamen. The variation is intra-flower bud but also intra-stamen, and the position of the flower bud along the inflorescence plays no role in this variation. The proportions of both cytokinesis types are not the same, because successive cytokinesis occurs in around 2% of the microsporocytes.

It is generally thought that simultaneous cytokinesis with centripetal cell wall formation, resulting in tetrahedral tetrads, is typical of the eudicots (Ressayre, 2001). However, this combination has been described recently in the Asparagales (Penet *et al.*, 2005) and occurs also in palms, as shown by this study, where both centrifugal (*S. repens*, *C. mitis*, *Butia* sp., *D. decaryi* and *V. spiralis*) and centripetal (*C. hospita*, *H. coriacea* and *A. arenaria*) cell wall formation were observed. Palms (this study) and Iridaceae (Penet *et al.*, 2005) are so far the only monocot families where variation in cell wall formation has been described. Variants of modified simultaneous cytokinesis with 'ephemeral cell plates',



Figures 37–42. Cytokinesis, the way in which callose walls, and additional callose deposits, are formed in one species of Caryoteae, *Caryota mitis* (Figs 37–38, 40–42: aniline blue; Fig. 39: aceto-carmine). Figs 37, 38. Dyad stage observed with light microscopy (Fig. 37) and epifluorescent microscopy (Fig. 38), successive cytokinesis with a cell wall separating the two sister cells. Fig. 39. Simultaneous cytokinesis with four nuclei in the same cytoplasm. Fig. 40. centrifugal formation of a cell plate in the centre of the cell. Fig. 41. Three visible walls grow from the centre of each cleavage plane and, at the same time, additional callose deposits progress from the edges of these cleavage planes (arrows). Fig. 42. Tetragonal tetrad with additional callose deposits growing from the edges of each cleavage plane (arrows). Scale bars = 10 μ m.

such as we observed in *A. arenaria* and *H. coriacea*, have also been observed elsewhere in the monocots, for example in *Cyanotis* (Commelinaceae, Rau 1930), and in the magnoliids, for example, Magnoliaceae (Dinis & Mesquita, 1993; Furness, Rudall & Sampson, 2002), and Winteraceae (Sampson, 1963, 1970; Prakash, Lim & Sampson, 1992). However, the situation observed in *Butia* sp. and in *D. decaryi* is somewhat different: the second division cell plates are initiated before the end of the first cell plate formation. In monocots, this situation has also been recorded, but in association with successive cytokinesis, in *Stemona* (Stemonaceae) and *Eriocaulon* (Eriocaulaceae) (Furness & Rudall, 1999). *Blandfordia* (Blandfordiaceae) has ‘irregular’, simultaneous cytokinesis (Furness & Rudall, 1999), and in *Hemerocallis fulva* L. (Hemerocallidaceae) cytokinesis is described as intermediate between successive and simultaneous partitioning. Resulting tetrads are tetragonal or irregular tetrahedral (Penet *et al.*, 2005). These observations are unusual exceptions to the view of Blackmore & Crane (1998) who note that the resultant postmeiotic tetrad stage of modified simultaneous cytokinesis is usually tetragonal, decussate or T-shaped.

The diversity of tetrad form in palms has already been described by several authors, for example, Dahl-

gren & Clifford (1982), Harley (1999b) and Harley & Baker (2001). However, a striking feature of the pollen tetrad formation in the palm species examined is the high diversity found within each species. Although the proportions of the different categories of tetrads have not been assessed statistically, our results show that this intraspecific diversity occurs in all eight species examined. An unexpected feature is that rhomboidal and tetragonal tetrads were usually observed in addition to tetrahedral tetrads, although in lower frequency. Rhomboidal tetrads are not frequently recorded in angiosperms. The tetrahedral tetrads were generally irregular with six intersporal walls of unequal size. The association between simultaneous cytokinesis and irregular tetrahedral tetrads has been described in several monocotyledonous families, for example, Blandfordiaceae (Furness & Rudall, 1999), Asphodelaceae (Huynh, 1976; Nadot *et al.*, in press) and Asparagales (Nadot *et al.*, in press). This contrasts with the situation in eudicots, where tetrahedral tetrads are normally regular in form (Wodehouse, 1935; Erdtman, 1952).

An intriguing callose ring was observed in *C. hospita*, *S. repens* and *H. coriacea*. A similar phenomenon has been described in a few species of the Iridaceae with simultaneous cytokinesis, where it seems

to constrain the tetrad shape to tetragonal (Penet *et al.*, 2005). The situation in palms appears different because the callose ring was observed in tetrads with different shapes, such as rhomboidal and tetrahedral. Therefore, the role of this callose ring remains to be explained.

The diversity of microsporogenesis observed in palms contrasts with the apparent uniformity found in the rest of the Commelinids, where cytokinesis is mostly successive with a centrifugal progression of the cell plates.

CONCLUSIONS

Our results show that behind the uniform aperture pattern in the eight species of Arecaceae examined, lies a notable diversity in the ontogenic pathway. We do know that there are other species in the four arecoid genera represented where the pollen shape, and sometimes the aperture shape as well, is often highly asymmetrical, and trichotomosulcate pollen is known to co-occur in some species of *Dypsis* (Harley, 1999a). These findings raise the question of how aperture configuration is determined in palm species producing monosulcate pollen grains.

The impact of cytokinesis type, callose wall formation among the microspores, tetrad form and aperture position within the tetrad on final aperture configuration was demonstrated by Ressayre (2001) and Ressayre *et al.* (2002, 2005). But, concerning palms, further investigations will be necessary to understand exactly how aperture configuration is determined. In particular, investigations are needed concerning the relative position of the apertures within the tetrad. At present, our results do not allow us to conclude if the situations observed correspond to the predictions of the model of Ressayre *et al.* (2002).

In the pollen of Arecaceae some of the variations in aperture type, position and number are very unusual, and it is difficult to access fresh material in order to study pollen ontogeny in some of the most interesting examples, although we look forward to doing so when opportunities arise. However, we surmise that most of the more unusual and extreme aperture configurations in palms are rare outcomes of developmental processes similar to those governing bisymmetrical monosulcate, asymmetrical monosulcate, trichotomosulcate, or equatorial disulcate. Therefore, future studies will include microsporogenesis, and its impact on aperture position, in calamoid bisymmetrical monosulcate, and equatorial disulcate pollen. Additional studies of aperture development in monosulcate and trichotomosulcate pollen in the other four currently recognized subfamilies of palms will be reported soon (Dransfield *et al.*, 2005). We hope that by continuing our research into microsporogenesis in

palms we will be able to elucidate the dynamics of aperture development and explain the reasons for so much variation in aperture types in this remarkable monocot family.

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