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Embryonic development of *Syagrus inajai* (Spruce) Becc. (Arecaceae, Arecoideae), an Amazonian palm

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Abstract. *Syagrus inajai* ('pupunharana') is a native palm of Brazil, with phytogeographic prevalence in the Amazon region. A morpho-anatomical analysis was undertaken in order to gain a better knowledge on the embryonic development and germinative process of the *S. inajai*. Plant material was collected from the Campus of the Universidade Federal do Amazonas – UFAM, Manaus, Amazonas, Brazil, and processed using standard morphological and anatomical techniques. The development process of the embryo takes ~220 days, and is divided into four stages: proembryo, globular embryo, lateral cordiform and torpedo. The embryo is small, linear, and derived from the terminal cell of the proembryo, arising from mitotic divisions in the apical cell. The embryonic axis is straight, located in the proximal region, aligned parallel to the length of the embryo. The single cotyledon is formed by the ground meristem, procambium and protoderm. The procambium supplies the embryonic axis and the haustorium.

Additional keywords: embryology, monocotyledons.

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Introduction

The Amazonian ecosystem hosts ~50% of the Neotropical genera and 30% of the species that comprise the Arecaceae family. Of these, eight genera (24%) are endemic to the Amazon region (Henderson 1995; Henderson *et al*. 1995).

The abundance and diversity of palms indicate that they play an important role the structure and functioning of the ecosystems (Duran and Franco 1992), and the importance of the Amazon rainforest in the maintenance and expression of this botanical family is evident. According to Miranda *et al*. (2001), the abundance of palms can be explained by their ability to adapt to various types of habitat.

Syagrus inajai ('pupunharana') is a native palm of Brazil, with phytogeographic prevalence in the Amazon region (Leitman *et al*. 2012). Belonging to the Cocoseae Mart. in Endl. tribe, its fruit is characterised by the presence of a hard endocarp with three visible pores (Dransfield *et al*. 2008). The fruits are popularly known as 'coquinho' and are widely used by the local communities. The pulp and seeds are consumed *in natura* and the endocarp is utilised in local handicrafts (Henderson *et al*. 1995; Miranda and Rabelo 2006).

However, there is a lack of research into the reproduction process of the species that make up this tribe. This shortfall is partly due to the morpho-anatomical characteristics of the fruits and seeds, including: the hardness of the endocarp, the slow development of the pericarp, the large quantity of fibres present in the mesocarp (Meerow and Broschat 2004; Dransfield *et al*. 2008), the liquid endosperm in which much or all of the development of the seed occurs, and finally, the small size of the embryo.

In order to gain a better understanding of the germinative process and the establishment and propagation of palms, further studies are needed on the reproductive organs, fruits and seeds. Haccius and Philip (1979) consider that the palm embryo appears to have more uniform development, without large variations, as cited in the literature in general. However, due to the lack of research in this area, further studies are needed to corroborate this claim. The majority of works carried out with zygotic embryos of palms were developed in the second half of the last century, e.g. those of: *Areca catechu* L.; tribe Sabaleae; *Chamaerops humilis* L.; *Phoenix*; *Livistona chinensis* (Jacq.) R.Br. ex Mart. and *Jubaeopsis caffra* Becc. (Rao 1955, 1958; Guignard 1961; Mahabale and Biradar 1967; Biradar 1968; Biradar and Mahabale 1968; Kulkarni and Mahabalé 1974; Robertson 1976). However, a growing number of works are being carried out with somatic embryos, cultivated *in vitro*,

with the aim of enabling the production and commercialisation of clones (Karunaratne and Periyapperuma 1989; Sane *et al.* 2006) as *Cocos nucifera* L. (Nunez *et al.* 2006) and *Phoenix dactylifera* L. (Othmani *et al.* 2009). The methods used in these studies have managed to overcome some of the difficulties imposed by the nature of the tissues of the plant.

Despite the efforts made so far, according to Ribeiro *et al.* (2012), we still do not have a detailed description of the embryonic axis of the embryo in palms. According to Natesh and Rau (1984) there are few studies on all the aspects of embryogenesis, zygote formation, and organisation of the meristems.

Therefore, using *S. inajai*, we seek to contribute information on the embryonic development of palms, which will further enhance our understanding of the germinative process of plants of the Arecaceae family.

Therefore, seeking to contribute information on the embryonic development of palms, which will further understanding of the germinative process of plants of the Arecaceae family, we decided to undertake the study of embryonic development of *S. inajai*.

Materials and methods

Plant materials were collected in 2009 and 2010 from a forest area at the Campus of the Universidade Federal do Amazonas – UFAM, Manaus, Amazonas, Brazil, in an area of baixio (gallery forest), around coordinates 03°05′45.84′S and 59°58′43.69′, under Afi climatic conditions (Köppen 1931). The material was analysed at the Laboratório de Botânica Agroflorestal (Agroforestry Botanical Laboratory), in the same University. For the morpho-anatomical description, adult individuals of *S. inajai* were selected, from which one rachilla was removed every 10 days, beginning 30 days after the opening of the spathe, when the pistillate flowers also opened.

The embryo length was measured every 10 days. Using these measurements, the ratio between the developmental periods and the embryo length was calculated by means of a regression study using Origin 8.6 software.

For the anatomical study, the collected material (whole fruit in various stages) was fixed in $FAA_{70\%}$ (formol: acetic acid: ethylic alcohol 70%) and conserved in 70% ethylic alcohol (Kraus and Arduin 1997). The samples were dehydrated in ethyl series (70–95%), embedded in 2-hydroxyethyl-methacrylate (Historesin, Leica Microsystems, Buffalo Grove, IL, USA), prepared according to the manufacturer's instructions), sectioned to thicknesses of 4–7 µm in a rotary microtome, and stained with 0.5% toluidine blue in citrate buffer, pH 4.0 (O'Brien *et al.* 1964). The slides were mounted in water.

Samples were fixed in buffered neutral formalin (Lillie 1965) solution, dehydrated in ethyl series, and dried using the critical point technique with CO_2 on a Balzers dryer (model CPD 030; Bal-Tec, Germany). The samples were collected in a metal support, covered with gold (Balzers SCD 050) and observed under a JEOL JSM 5800 LV (10-kV) Scanning Electron Microscope, at the Institute of Biology/Unicamp.

The results were documented using an Olympus (BX51) photomicroscope with Olympus (DP71) photographic camera (Olympus America, Melville, NY, USA), and a Leica (M125) stereomicroscope with Leica (DFC 490) photographic camera. Indian ink drawings were also made.

Results

The growth of the embryo of *S. inajai* follows the sigmoid function model of Boltzmann (Fig. 1).

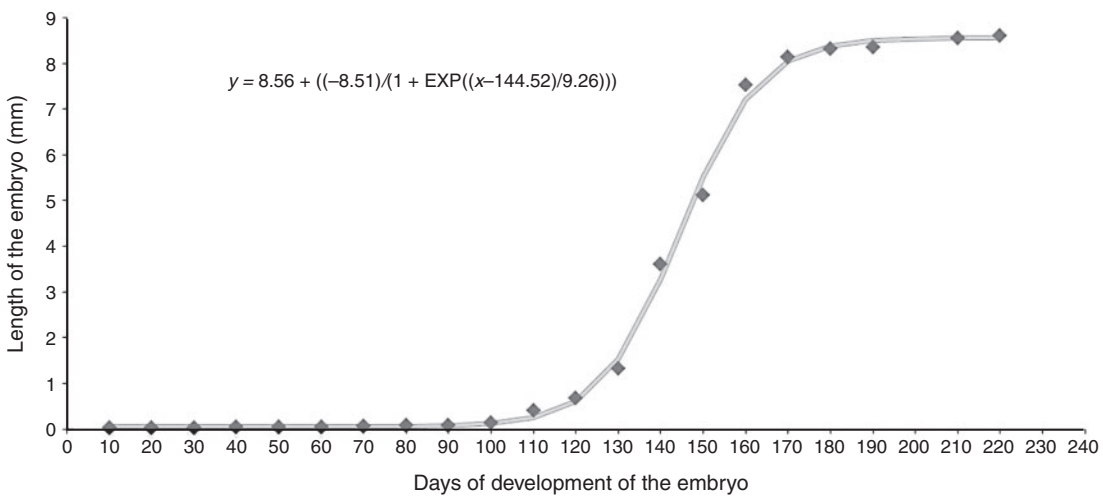
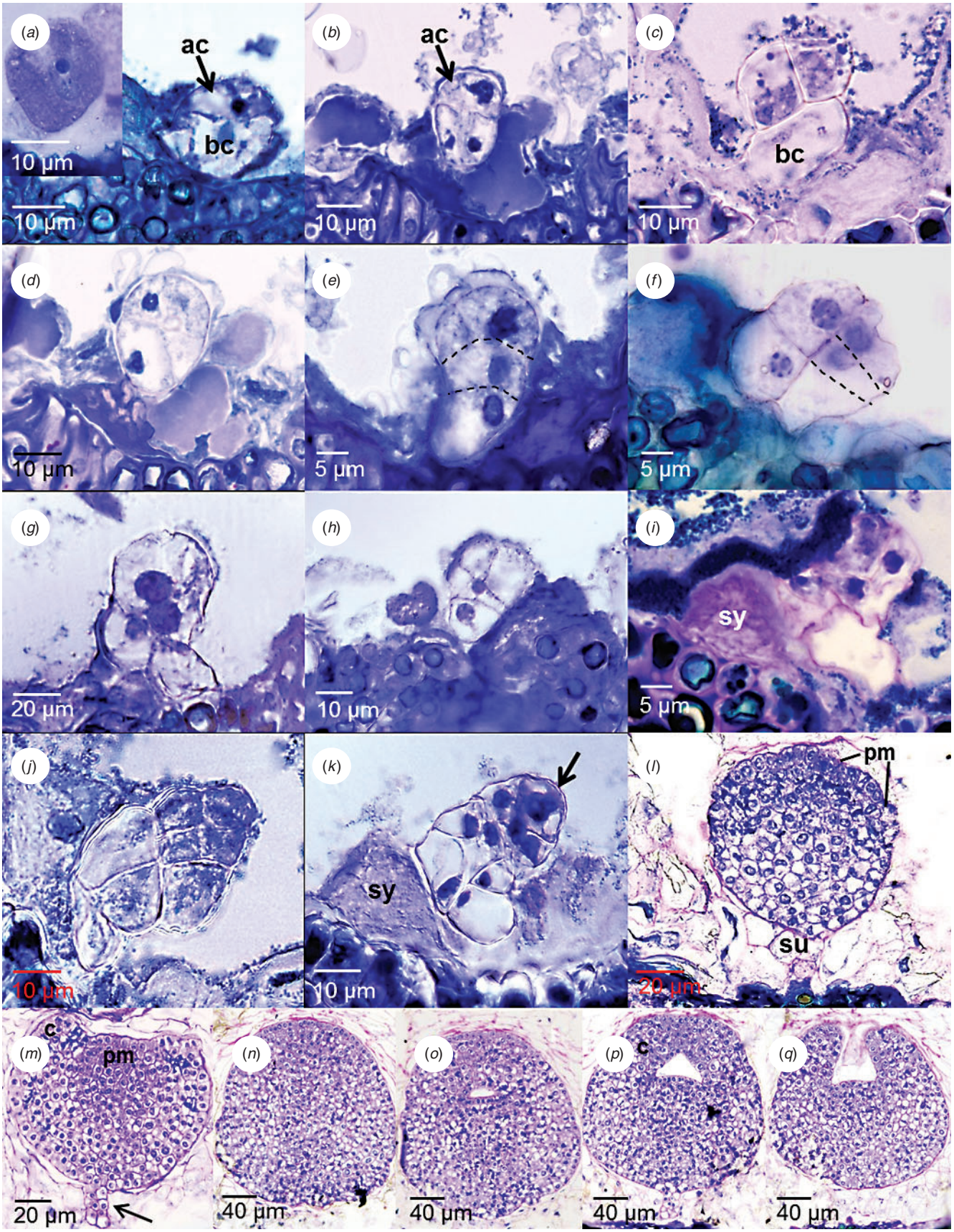


Fig. 1. Regression analysis of the growth of the embryo of *Syagrus inajai*. Measurements of embryo length at 10-day intervals.

Fig. 2. Photomicrograph of longitudinal section of the development of the proembryo and zygotic embryo of *Syagrus inajai*. (*a*) Zygote, first division, detail of the basal cell, apical cell (arrow); (*b*) second basal cell division, apical cell (arrow); (*c*) second apical cell division; (*d*) quadratic proembryo; (*e*) periclinal division in the apical cell; (*f–g*) second division, cells derived from apical cell; (*h*) third division, cells derived from apical cell; (*i*) proembryo with six cells; (*j*) proembryo with nine cells; (*k*) proembryo with polarised cell (arrow); (*l*) globular stage; embryo at 80 days; (*m*) longitudinal sagittal section of the embryo at 90 days, suspensor arrow; (*n–q*) sequence of sections on the longitudinal sagittal plan of the embryo at ~95 days; (*n*) central region of the embryo; (*o*) start of the cotyledonary cavity and shoot apical promeristem; (*p*) edges of the cotyledon; and (*q*) lateral with free borders. Initials: ac, apical cell; bc, basal cell; co, cotyledon; pm, promeristem; su, suspensor; sy, syndergids.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Proembryo stage

The start of embryogenesis was observed at ~30 days after the start of endosperm formation. The zygote is approximately the same size as the egg cell (Fig. 2*a*) and is located in the micropilar region. Its first mitotic division occurs on the periclinal or oblique plane, resulting in a larger basal cell and smaller apical cell than those that comprise the proembryo (Fig. 2*a*). Next division occurs on the anticlinal plane, which may be initiated with the basal cell or the apical cell (Fig. 2*b–d*). The apical cell may also divide primarily in the periclinal plane (Fig. 2*e*). The two cells originating from the apical cell subsequently divide in the periclinal, oblique or anticlinal planes, with no particular pattern being observed (Fig. 2*f–j*). When the cells derived from the apical cell reach nine in number, it is possible to see a larger polarised terminal cell, precursor of the embryo, which is marked by a densely stained nucleus and dense cytoplasm (Fig. 2*k*). The other cells derived from basal and apical cell form the small suspensor, comprised of approximately eight cells (Fig. 2*l*, *m*). The two conspicuous synergids persist until the start of the globular stage of the embryo (Fig. 2*i*).

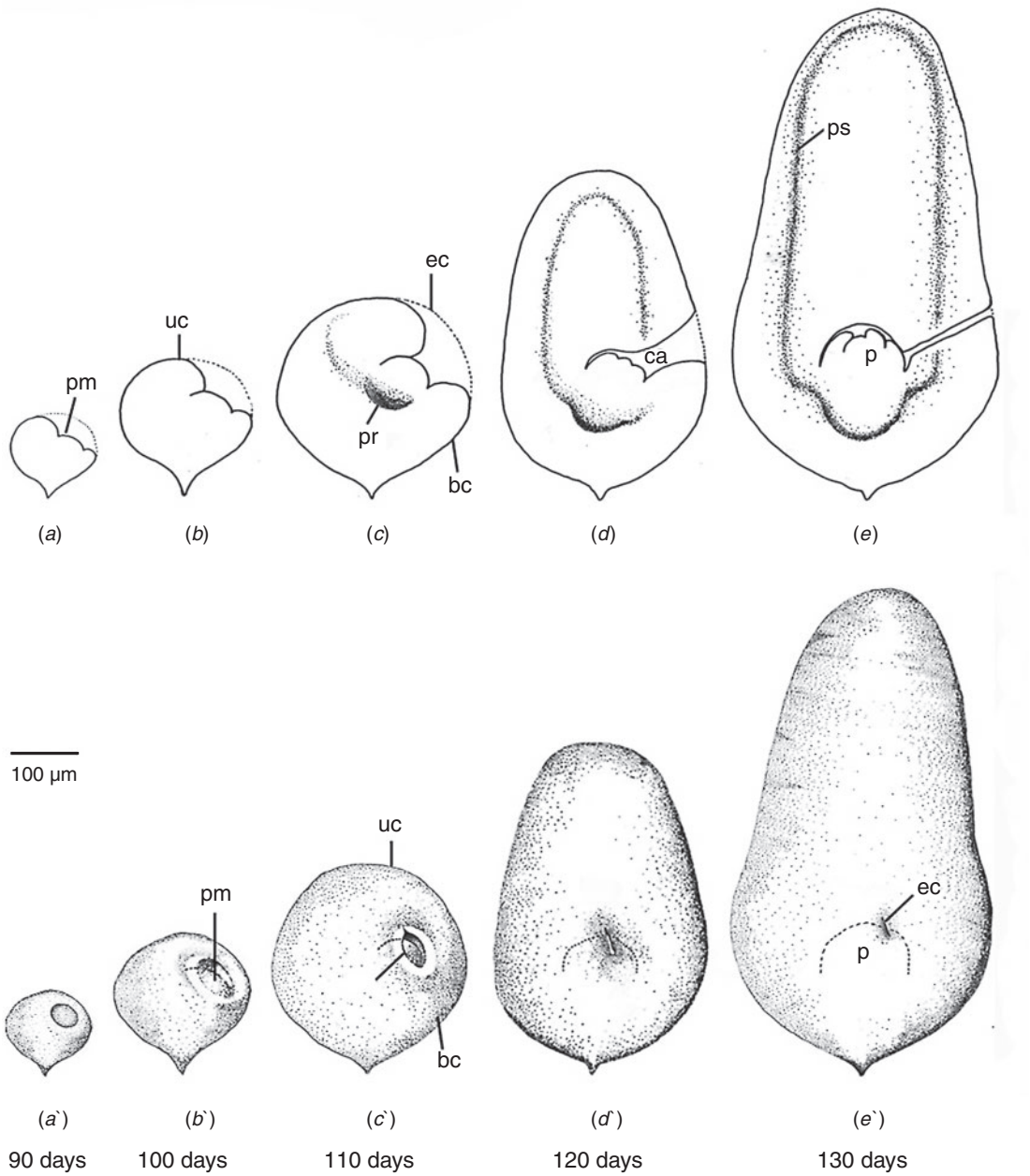


Fig. 3. Development and internalisation of the shoot apical promeristem and formation of the procambium in the embryo of *Syagrus inajai*. (*a–e*) View in longitudinal section; (*a*′*–e*) view of the surface; (*a*) start of shoot promeristem; (*b*) start of cotyledonary growth; (*c*) cotyledon growing around the shoot promeristem; (*d*) elongation of the cotyledon; and (*e*) internalisation of the plumule. Initials: bc, cotyledonary base; ca, cotyledonary cavity; ec, cotyledonary edge; p, plumule; pm, promeristem; pr, procambium; ps, procambial strand; uc, upper cotyledon.

Globular stage

Approximately 30 days after mitosis of the zygote, the terminal cell, precursor of the embryo, derived from apical cell division, undergoes successive divisions, in different planes, until it forms the globular body of the embryo, a process that takes ~50 days. Concomitantly with these divisions the formation of the protoderm occurs, which is established by anticlinical divisions. With the end of this stage comes the formation of the embryo, with globular formation and radial symmetry (Fig. 2*l*).

Lateral cordiform stage

It was possible to observe, in the lateral terminal region of the embryo at ~90 days, the start of differentiation of the stem promeristem, which is characterised by a group of cells with dense cytoplasm and large, densely stained nuclei (Figs 2*l*, 3*a*, *a*′). The position of the shoot apical promeristem, dislocated from the centre, shows asymmetry, with uneven distribution of the tissues surrounding it, the larger side corresponding to the developed cotyledon (Fig. 2*n–q*) and the smaller side to the undeveloped cotyledon (Fig. 3*a*). The presence of this vestigial tissue relates to

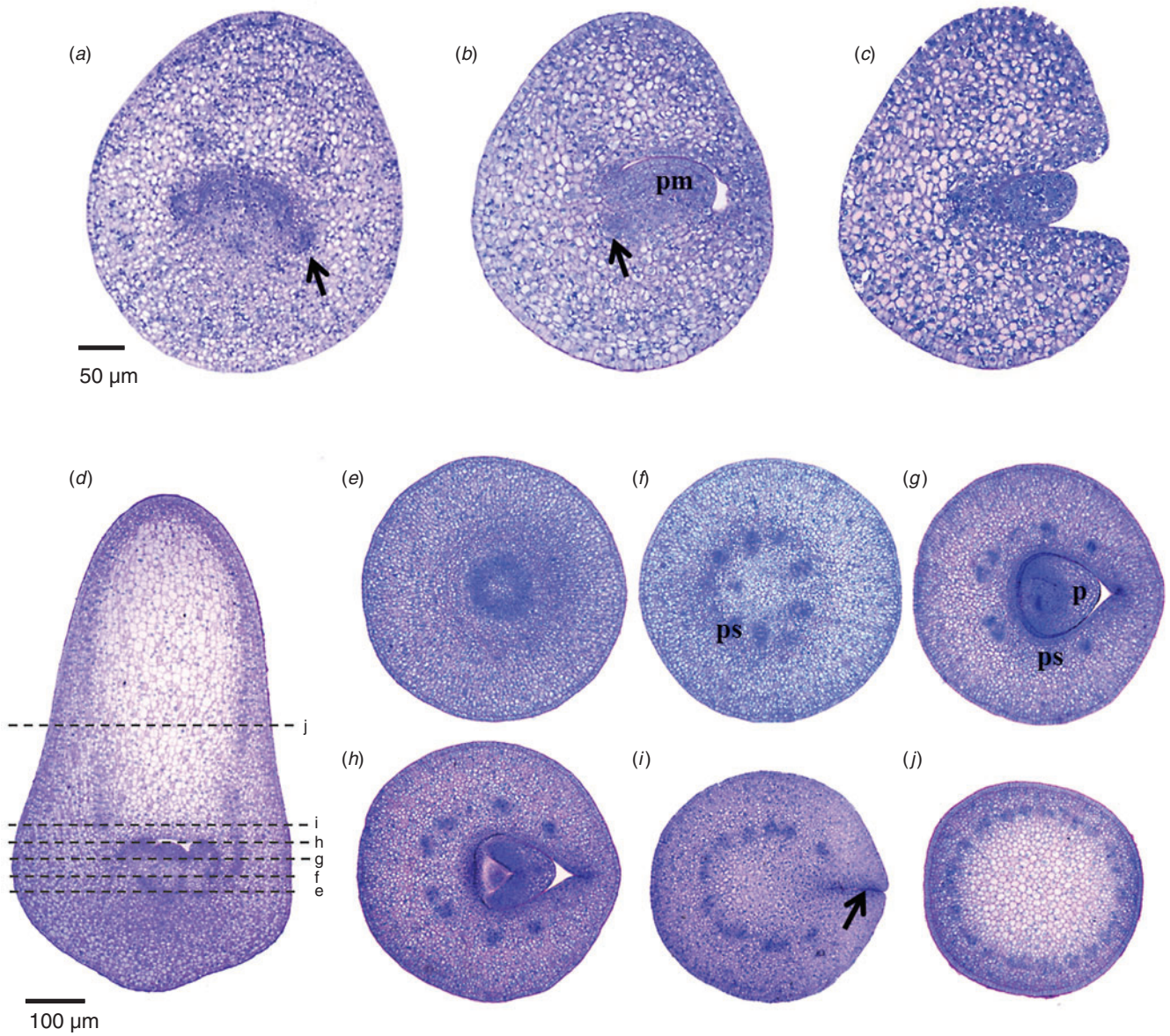


Fig. 4. Photomicrograph of the embryo of *Syagrus inajai*. (*a–c*) Embryo at 110 days; (*a*) longitudinal sagittal section, procambium (arrow); (*b–c*) frontal longitudinal section; (*b*) growth of the border around the shoot apical promeristem, procambium (arrow); (*c*) region of the embryo with shoot apical promeristem; (*d–j*) embryo of *S. inajai* at ~120 days. (*d*) General appearance of the embryo in longitudinal section, line indicating position of the transversal sections; (*e–j*) sequence in transversal section of the body of the embryo; (*e*) procambium of the radicle; (*f*) procambial strands of the hypocotyl; (*g*) base of the plumule; (*h*) apex of the plumule; (*i*) cotyledonary gap (arrow); and (*g*) distal region of the embryo. Initials: p, plumule; pm, promeristem; ps, procambial strand.

the second cotyledon of the dicotyledons (Fig. 3*a*). The differentiation of the promeristem marks the end of the globular phase; the radial symmetry is replaced with bilateral symmetry, which will continue until the end of its development (Fig. 3*a*′).

Alongside with the development of the shoot apical promeristem, the cotyledonary primordium surrounding it begins to grow over the promeristem (Fig. 2*n*) up to the junction of its borders (Fig. 3*a*–*e*′), a process that takes ~30 days and culminates with the intrusion of the promeristem and the formation of the cotyledonary cavity (Fig. 3*a*–*e*). The basal region of the cotyledon grows around the shoot apical promeristem, and shifts towards the centre, repositioning it so that it becomes central, with an inclination of 90°C, parallel to the body of the embryo (Fig. 3*a*–*e*). The upper region of the cotyledon includes the terminal portion of the shoot apical promeristem (Fig. 3*c*, *d*).

The primary cells of the procambium are observed in the embryo at ~100 days, when they begin to differentiate around the shoot apical promeristem, within the confines of the cotyledon (Fig. 4*a*, *b*). These cells present dense cytoplasm and large, densely stained nuclei (Fig. 5*a*).

Torpedo stage

At 110 days, an increase in embryo length is observed (Fig. 6), with the maximum value at between 190 and 220 days (Figs 1, 6).

At the end of the process of intrusion of the shoot apical promeristem, intense cell division is observed in the upper region of the cotyledon. This gives rise to lengthening of the embryo, which expands in the opposite direction to that of the micropile by cell division, leading to the development of the distal region of the embryo and exerting a haustorial function (Figs 2*n*, 3*c*, *d*, c′–*e*′). With the lengthening of the cotyledon, the embryo becomes conical in shape (Figs 2*q*, 3*d*, *e*). The protoderm accompanies the lengthening by anticlinal divisions, and is continuous in the cotyledonary cavity surrounding the shoot apical promeristem.

At ~120 days, the shoot apical promeristem is internally located (Fig. 4*d*, *h*), forming the cotyledonary cleft (Figs 3*c*–*e*, 4*h*, *i*), which is imperceptible externally in the developed embryo and can only be visualised in cross section (Figs 5*l*, 6). The presence of the cotyledonary cleft shows the point of meeting of the borders of the basal and upper regions of the growing cotyledon (Fig. 3*a*–*e*′). Concomitantly with the lengthening of the upper portion of the cotyledon, the formation of the primary leaf primordium is observed, its differentiation occurring subsequently to the intrusion of the shoot apical promeristem (Figs 3*d*, 4*d*, 5*b*). In this phase, two morpho-anatomical regions are distinguished in the embryo, namely, the distal and the proximal (Fig. 4*d*). From the cotyledonary node, the procambium emits eight traces to the proximal region, which will irrigate the leaf primordia and the radicle, and eight traces to the distal region, which will irrigate the haustorium (Figs 3*c*–*e*, 4*a*–*j*).

At ~130 days, it is possible to discern, on the opposite border of the shoot apical promeristem of the embryo, the initial cells of the second leaf primordium (Fig. 5*c*). Next to the micropile, in the opposite direction to the plumule, we observed that the cells of the procambium are organised in a semicircle, with a rudimentary embryo axis clearly visible (Fig. 5*c*).

The cells of the procambium, arranged in a semicircle, are elongated (Fig. 5*b*). At the apex there is a set of juxtaposed, tiny cells, comprising the radicular promeristem (Fig. 5*d*–*f*).

The first cells of the third leaf primordium are visible in the embryo at ~140 days.

During the formation of the distal region of the embryo, the cells of the ground meristem located inside the procambial strands increase in number and size. At the end of the lengthening, these cells are characterised by the presence of large vacuoles (Fig. 4*d*).

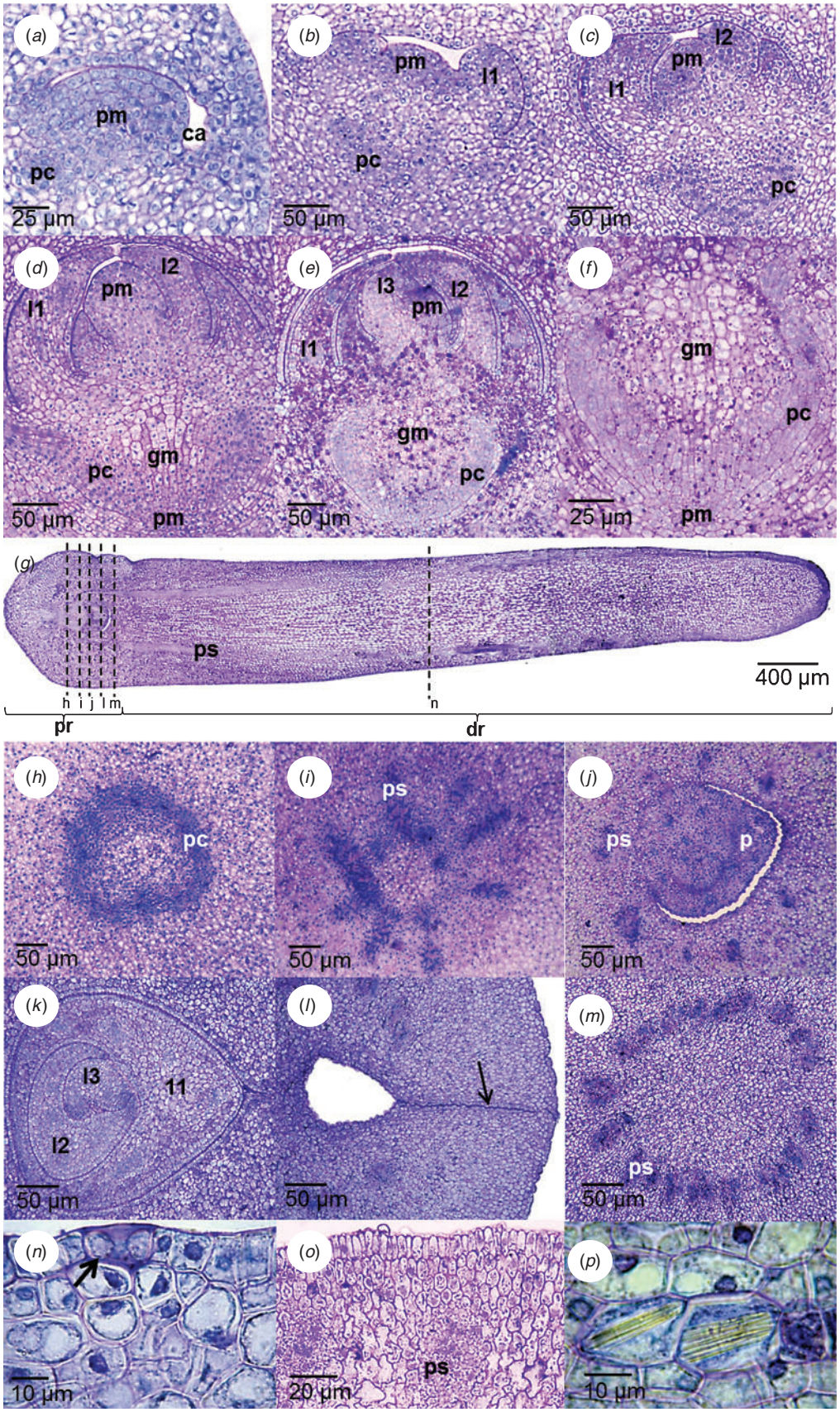
Embryo

At the moment of dehiscence of the fruit, ~270 days after the opening of the spathe, the embryo is at ~220 days, ~8 mm in length, straight, lateral, oblique and small (Figs 1, 6).

The proximal region of the embryo is cylindrical, with the funnel-shaped apex in contact with the micropile. Its surface is smooth (Fig. 7*b*), except for a protuberance on the apex, which is a vestige of the suspensor (Fig. 7*c*). It is composed of the cotyledon sheath, leaf primordia and embryo axis (Fig. 5*g*). The protoderm presents cuboid cells and one two stomata are also visible (Fig. 5*n*. The cells of the ground meristem are heterodimensional (Fig. 7*d*) with idioblasts containing raphides (Fig. 5*p*). The embryo axis is short, and parallel to the length of the embryo (Fig. 5*g*). The hypocotyl-radicle is short, with a radicle presenting a procambium, ground meristem and radicular promeristem, characterised by small, juxtaposed cells, without a differentiated protoderm (Fig. 5*e*, *f*). The three-leaf primordia and shoot apical promeristem form the plumule, located in the cotyledonary cavity and formed by three-leaf primordia and the shoot apical promeristem with a protoderm of isodiametric cells, procambial strands and ground meristem (Figs 5*e*, *j*–*k*, 7*d*).

The distal region of the embryo has laminar appearance (Figs 6, 7*a*). It has a sinuous surface, forming elevations that increase the contact surface with the endosperm and can be observed along the embryo (Figs 6, 7*a*). The protodermal cells are radially elongated and larger than those of the proximal region (Fig. 5*o*). The cells of the ground meristem have large vacuoles (Fig. 5*g*, *m*, *o*). The procambial strands become gradually more peripheral as they distribute ~18–26 strands to the outer edges of the distal region, and parallel to the length of the embryo (Figs 4*d, i, j*, 5*g*, 6).

Fig. 5. Photomicrograph of the embryo of *Syagrus inajai*. (*a*–*f*) Development of the embryo axis in longitudinal section; (*g*–*q*) fully-formed embryo; (*a*) shoot apical promeristem (arrow); (*b*) first leaf primordium; (*c*) start of formation of the second leaf primordium; (*d*) visible procambium; (*e*) fully-formed embryo axis; (*f*) detail of the radicle; (*g*) general appearance of the embryo at 210 days; (*h*) procambium of the radicle; (*i*) procambial strand; (*j*) start of the plumule; (*k*) plumule; (*l*) cotyledonary border forming the cotyledonary gap (arrow); (*m*) region of the haustorium; (*n*) protoderm of the proximal region, stomata (arrow); (*o*) epidermis in the distal region; and (*p*) idioblasts with raphides. Initials: ca, cotyledonary cavities; dr, distal region; l1, first leaf primordium; l2; second leaf primordium; l3, third leaf primordium; gm, ground meristem; p, plumule; pc, procambium; pm, promeristem; pr, proximal region; ps, procambial strand.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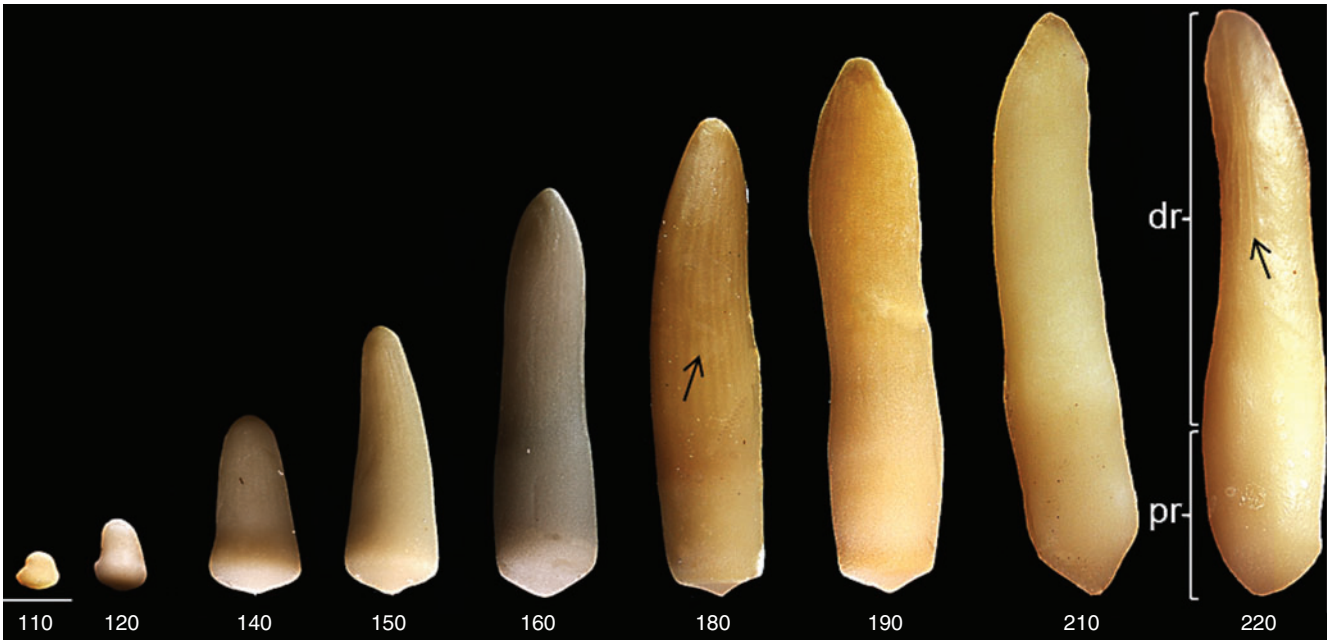

Fig. 6. Morphological appearance of the development of the embryo of *Syagrus inajai*, days after fertilisation, relief formed by the procambial strands (arrow). Initials: dr, distal region; pr, proximal region; bar 3 mm.

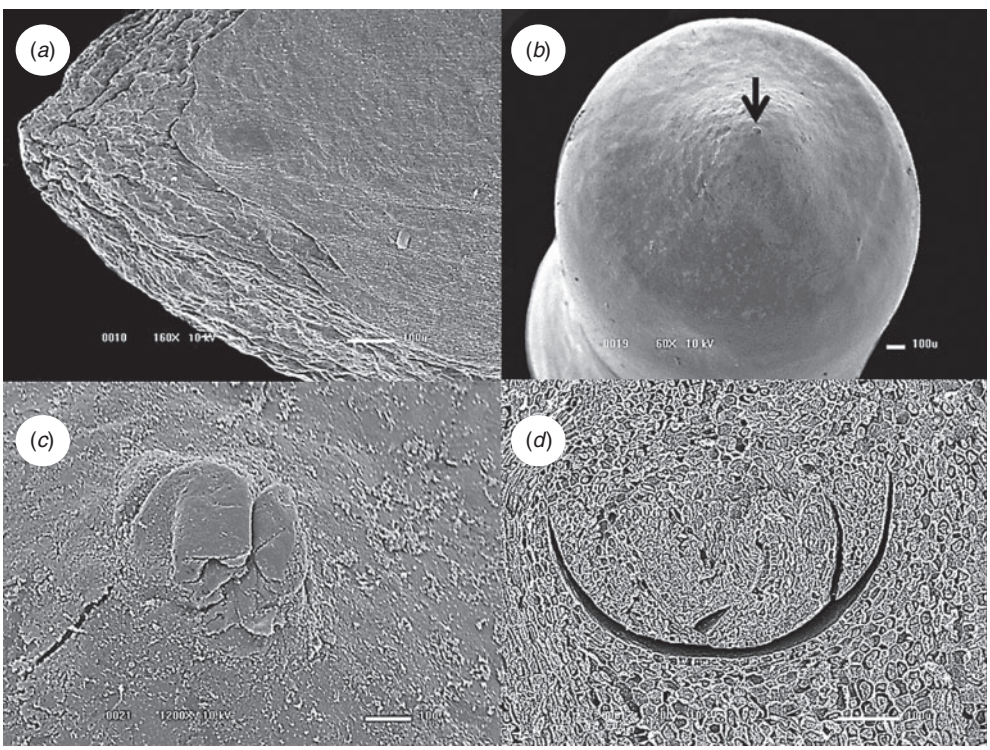

Fig. 7. The micrographs from the scanning electron microscope of the appearance of the embryo of *Syagrus inajai* at the moment of dispersion. (*a*) Haustorial region; (*b*) proximal region, suspensor (arrow); (*c*) vestige of the suspensor; and (*d*) appearance of the plumule.

The procambium is arranged along its length, forming two loops, emerging from the cotyledonary node (Figs 3*e*, 4*d*). A larger loop with eight traces branches into 18–26 strands (Figs 4*h–j*, 5*n*) peripherally providing with vascular bundles in the distal region of the cotyledon, which are responsible for the translocation of substances from the endosperm to the

embryo; and another smaller loop in the proximal region (Fig. 5*f*) comprising the radicular procambium, with eight traces issued to the plumule (Figs 4*e*, *f*, 5*h–j*).

Discussion

The sigmoid growth curve observed for the embryo of *S. inajai* has also been reported for *Elaeis guineensis* Jacq. (Fernandino *et al*. 1985), and is similar to the typical growth curve of dicotyledons.

The embryo of *S. inajai* is comprised only of cells derived from the apical cell, characterising it as the Onagrad type, according to Johansen (1950). The same characteristic was observed in *A. catechu*, *Sabaleae* and *C. nucifera* (Rao 1955; Haccius and Philip 1979). In studies carried out with *Phoenix* and *L. chinensis* (Mahabale and Biradar 1967; Biradar 1968; Biradar and Mahabale 1968; Kulkarni and Mahabalé 1974), it was found that the embryo was of cells derived from apical cell and basal cell, classified as the Asterad type.

The shoot apical promeristem was described as lateral terminal in *S. inajai*, as although its origin is terminal, it does not occur in the centre of the embryo axis, but it is displaced. Haccius and Philip (1979) describe the origin of the shoot apical promeristem as terminal in *C. nucifera*, although they report that the subsequent development of the cotyledon causes the shoot apical promeristem to become lateral. Guignard (1984) argues that there is a transition from dicotyledons to monocotyledons in relation to the position of the shoot apical promeristem, and sees this character as transitory in palms. This author believes that in *C. nucifera*, the quiescent central axial zone is positioned between the two meristem zones, but only one of them continues its development. In other monocotyledons, the position of the shoot apical promeristem becomes progressively more lateral, until it is entirely lateral, as observed in *Cyperus fuscus* L. Therefore, the other meristematic zone of *S. inajai* does not develop, because it is a vestigial cotyledon.

The lateral cordiform stage is characterised by the formation of the shoot apical promeristem in the lateral terminal region of the embryo, and by the growth of the edges of the cotyledon around it. The longitudinal section of this region reveals the embryo, with cordiform appearance similar to the cordiform stage in dicotyledons. However, what we see in *S. inajai* is the edges of a single cotyledon that grows around the shoot apical promeristem. According to Guignard (1984) and Cronk (2009), the embryos of monocotyledons have a single cotyledon, which initiates its development early. Mahabale and Biradar (1967), studying the embryo of *Phoenix sylvestris* (L.) Roxb. in longitudinal plane, describe two cotyledons. However, Haccius and Philip (1979), studying the embryo of *C. nucifera* in the frontal longitudinal and sagittal longitudinal planes, observed that this was an error caused by the section plane.

In *S. inajai* the growth of the cotyledonary regions is not uniform in space and time. The basal portion plays a more active role in the intrusion of the shoot apical promeristem, with intense cell division at the start of embryo development, while the upper region elongates in the opposite direction to the micropile, forming the haustorial region, with intense cell division after the intrusion of the shoot apical promeristem. The same sequence of events was observed in *C. nucifera* (Haccius and Philip 1979).

The position of the embryo axis parallel to the length of the embryo, as observed in *S. inajai*, *Borassus flabellifer* L. and *Jubaea chilensis* (Molina) Baill., *Phoenix roebelenii* O'Brien and *Allagoptera leucocalyx* (Mart.) Kuntze (Dassanayake and Sivakadachchan 1973; Iossi *et al*. 2003; Henderson 2006), is mainly due to the growth of cells close to the shoot apical meristem, in the basal region of the cotyledon. The embryo axis, arranged obliquely to the length of the embryo, as observed in species of *Euterpe precatoria* Mart., *Oenocarpus minor* Mart., *Acrocomia aculeata* (Jacq.) Lodd. ex. Mart. (Aguiar and Mendonça 2003; Oliveira *et al*. 2010; Ribeiro *et al*. 2012) may be the result of a greater participation of the upper region of the cotyledon in the process of intrusion of the shoot apical promeristem, with a smaller addition of cells in the basal region, as observed in *C. nucifera* (Haccius and Philip 1979).

The development of the shoot apical promeristem in *S. inajai* occurs at the start of embryogenesis, after the formation of the globular stage of the embryo. The same sequence of events was observed in the zygotic embryo of *C. nucifera* and *Elaeis guineensis* (Haccius and Philip 1979; Kanchanapoom and Domyoas 1999) and in the somatic embryo of *P. dactylifera* L. (Sane *et al*. 2006). The development of the radicular promeristem occurs after the intrusion of the shoot apical promeristem and start of differentiation of the primary leaf primordium, in accordance with Haccius and Philip (1979). The radicle can be seen visualised in sections 3–5-μm thick, due to the small size of the cells that comprise it. It is characteristic of cells of the procambium, arranged to form a cap in longitudinal plane, by cells of the ground meristem and by the radicular promeristem, comprised of small, densely blue stained cells. The presence of the radicle was also reported in embryos of *Allagoptera arenaria* (Gomes) Kuntze, *A. aculeata*, *Borassus flabellifer*, *Euterpe edulis* and *L. chinensis* (Dassanayake and Sivakadachchan 1973; Kulkarni and Mahabalé 1974; Sylvestre *et al*. 1989; Panza *et al*. 2004; Ribeiro *et al*. 2012). However, in *E. precatoria* Mart. and *O. minor* (Aguiar and Mendonça 2003; Oliveira *et al*. 2010), the presence of a radicle pole was described. The fact that the plumule is located in a cotyledonary cavity, being covered by protoderm makes it easier to observe than the radicular hypocotyl axis, which is inserted in the ground meristem without a protoderm, which can make it difficult to distinguish.

In the embryo at 140 days, ~80 days before the dispersal of the fruit of *S. inajai* it is possible to observe their distal and proximal regions and its embryonic axis. According to Tomlinson (1990), it is possible to see two morpho-anatomical regions in the embryo of palms; the proximal region, known as the cotyledonary petiole, and the distal region, with haustorial function. These two regions were observed in mature embryos of various palm species, such as *Attalea maripa* (Aubl.) Mart., *E. precatoria*, *Archontophoenix alexandrae* (F.Muell.) H. Wendl. & Drude, *A. aculeata* and *O. minor* (Araújo *et al*. 2000; Aguiar and Mendonça 2003; Charlo *et al*. 2006; Moura *et al*. 2008; Oliveira *et al*. 2010; Ribeiro *et al*. 2012).

According to Orozco-Segovia *et al*. (2003) anatomical immaturity is common in the embryo of palms during dispersion of the fruit, attributing morphological dormancy to

the seed. Baskin and Baskin (1998) suggested that morphological dormancy occurs when the embryo is undifferentiated, rudimentary, or in the torpedo stage at the moment of dispersion. However, the embryo of *S. inajai* does not present these characteristics, as its embryo axis differentiates ~50 days before dispersion of the fruit. At ~190 days, the embryo is ~8 mm in length, close to the size observed in dispersion. Haccius and Philip (1979) report that the plumule in *C. nucifera* initiates its development before the cotyledon reaches its full length. Hemanthakumar *et al*. (2012) managed to produce germination of the zygotic embryo of *Calamus thwaitesii* Becc.; by removing the fruit months before dispersion, they promoted the extraction of young embryos of the fruit for cultivation *in vitro*, when the tissue damage is reduced and the embryo is viable.

In the protoderm of *S. inajai*, the presence of apparently inactive stomata was observed, and the same has been reported for *A. aculeata* (Ribeiro *et al*. 2012). However, the authors believe that its presence may be related to the oxygen demand in the initial phases of germination. The presence of idioblasts with raphides in the ground meristem was also reported by Zona (2004) and Oliveira *et al*. (2010). According to Zona (2004), the presence of raphides is common in the Areceae tribe. However, in the species of the *Syagrus* genus studied by the author, these were not observed, and the author reports the need for additional sampling. For the author, the probable function of the raphide crystals in the embryos is the storage of calcium oxalate and or hydrogen peroxide. Moura *et al.* (2010) and Ribeiro *et al*. (2012) observe raphides in *A. aculetata* only after germination.

The arrangement of the procambium in the embryo resembles two loops, the larger with traces issued to the distal region, and the smaller with the traces to the plumule and embryo axis. The same was also observed in *C. nucifera* and *A. aculetata*. However, in these two species, the occurrence of a procambium around the plumule was observed, which was not observed in *S. inajai* (Haccius and Philip 1979; Moura *et al*. 2008; Ribeiro *et al*. 2012).

Conclusion

The embryo of *S. inajai* presents four specific stages of development: the proembryo stage, the globular stage, the lateral heart stage, and the torpedo stage. These stages are also observed in other species of palms. We therefore propose the existence of a pattern of embryo development in this family.

The greater participation of the basal region of the cotyledon in the process of intrusion of the shoot promeristem and formation of the cotyledonary cavity results in the central positioning of the embryo axis, parallel to the length of the embryo. Therefore, the final position of the embryo axis, whether parallel or oblique, is related to the growth dynamic of the basal and upper regions of the cotyledon during the process of intrusion of the shoot promeristem.

Finally, we found that the embryo of *S. inajai* does not present morpho-anatomical immaturity.

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