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# Morphological Studies of the *Euterpe oleracea* Mart. Seeds

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**Abstract:** The aim of this research is to describe the morpho-anatomy of *Euterpe oleracea* Mart. seed. The samples with 30 seeds (0, 10, 30 days after the beginning of the germination process), in which the seeds were placed in plastic boxes with sand as substrate and immediately introduced in germination chamber under 30°C, being irrigated daily with distilled water. The morphological and anatomical observations of the seed were carried out in the times 0, 10 and 30 and 0 days of germination, respectively. This seed has ruminated endosperm, solid, besides being constituted by large cellular walls, with several pit primary fields. The testa is ruminated, constituted by fine cellular walls, divided in three layers (external, medium and internal), it being that the medium layer has vascular bundles. The embryo is conic, basal and axial linear, with two regions, being a proximal (hypocotyl-radicle axis) and other distal (cotyledon).

**Key words:** *Euterpe oleracea*, seed, morphology, anatomy

## INTRODUCTION

The species *Euterpe oleracea* Mart. has economical importance to the Amazon region, because with the fruit pericarp several food products and energy drinks with economic potential in the Brazil and world are manufactured (Oliveira *et al*., 2002). The palm and its derivatives might be utilized in the pulp production, animal ration, forest, medicine, natural colorants, besides producing heart of palm, commercialized mainly for the export, in which 95% of all hearts of palm produced in the Brazil is extracted of this palm (Oliveira and Muller, 1998).

The distribution of this palm in the Brazil occur mainly in the states of the Pará, Amazonas, Maranhão and Amapá, besides to be found also in the Colombia, Equator, Guiana and Venezuela (Nascimento, 2006). Moreover, *Euterpe oleracea* has with habitat humid environment and soil partially flood, present pined leaf, flowers intrafoliars and the fruits are globosa drupes with pericarp green or lilac, when immature or mature, respectively (Jardim *et al*., 2004; Oliveira *et al*., 2002; Rogez, 2000).

The seed is the result of the development of the fertilized ovule through the process of double fecundation, being in Angiosperms, the mature seed is basically constituted of the embryo, endosperm and testa, in which the embryo develops through the diploid zygote formed by the fusion of one gametic nucleus and the oosfere, besides triploid endosperm originated from the combination of two polar nucleus with the second gametic nucleus (triple fusion) and the tegument or testa, formed by the integuments that involve the ovule (Cardoso, 2004).

The palm seeds are round or elliptic, are sticked to pericarp or simply free. When they are free, the seminal tegument is relatively thick and shows one opercule, in which it corresponds to the scar on the surface, next the micropyle that becomes visible during the germination (Alves and Demattê, 1987; Nascimento *et al*., 2007). Generally the cavity of the palm fruits is filled in by only one hard and dense seed constituted of the tegument derived from the ovule wall that covers the endosperm, in which it is a mass of nutritive tissue that is fitted in one small and soft embryo (Ferreira and Borghetti, 2004; Lorenzi *et al*., 1996).

The objective of this research is to describe the morpho-anatomy of *Euterpe oleracea* Mart. seed, it aimed to contribute subsequent studies with the structure function showed.

## MATERIALS AND METHODS

**Plant material and procedures:** The *Euterpe oleracea* Mart. seed were harvested of fruits and transported to the Laboratório de Fisiologia Avançada of the Universidade Federal Rural da Amazônia (UFRA), city of Belém, state of Pará, Brasil (01°27'S and 48°26"W), it being the experiments were conducted during the months January

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and February of 2006. The seeds were washed in neutral liquid detergent and running water, for to remove impurities and fruit residues in pericarp and immediately the mesocarp was removed with the objective to avoid fermentations provoked by fruit residues and fungus proliferation (Carvalho *et al*., 1998). The samples with 30 seeds (0, 10, 30 days after the beginning of the germination process, in which it was utilized plastic boxes with the dimensions (length×width×height; 35×25×7 cm, respectively) previously sterilized with sodium hypochlorite at 2% and the substrate was washed and autoclaved sand (120°C, 1 atm by 20 min). The seeds were uniformly distributes and placed in the substrate with seed opercule in the horizontal position and profundity of 2 cm (Aguiar and Mendonça, 2002). The plastic boxes containing the seeds and substrate were placed in germination chamber under temperature of 30°C, being irrigated daily with distilled water.

**Seed morphology:** The observations were carried out for 0, 10 and 30 days of germination. The seeds were removed of the substrate, washed in neutral liquid detergent and running water, after immersing in sodium hypochlorite at 2% by the period of 72 h. The observations were carried out at stereoscope photographic (model Motic Digital Microscope DM 148) with the software Motic imagens plus 2,0 ML of the Museu Paraense Emílio Goeldi (MPEG), Belém, Brasil.

**Seed anatomy:** The observation of the seeds was carried out in 0 days of germination, in which were removed of the substrate, washed in neutral liquid detergent and transversal histologic cuts were carried out in endosperm. In the confection of the histological blades aqueous glycerin at 50% was utilized (Purvis *et al*., 1964). The photomicrographs were carried out at three increases (100, 200 and 400X), utilizing stereomicroscope (model Olympus BX 41-FL-III) with the software Applied Spectral Imaging in the Universidade Federal do Pará (UFPA), Belém, Brasil and photomicroscope (model Zeiss XZ-305) of the Museu Paraense Emilio Goeldi (MPEG), Belém, Brasil.

## RESULTS

The *Euterpe oleracea* Mart. seed with 0 days after the beginning of the germination process present testa of the ruminated type (Fig. 1a), in which this rumination is not restricted to the testa, but also catches the endosperm, leaving this structure with irregular aspect, being denominated ruminated endosperm (Fig. 1b). This rumination appears as radial lines that arise in the seed outskirt and goes in the direction toward center (Fig. 1c and d).

The embryo is small, conic, basal, being of the axial linear type and is involved by abundant endospermatic tissue (Fig. 1e). It has two regions, one being proximal, more wide and dark described as hypocotil-radicle axis of yellow-white color and the other is distal more narrow and bright of white-pale that corresponding to simple cotyledon (Fig. 1f).

It were showed in the seeds with 10 days the breakthrough of the seed opercule (circular testa) at function of the protrusion of the cotyledonal bud, being a bulbose structure originated of the dilatation of the radicle-hypocotil axis that loads the meristematic pole in the extremity (Fig. 2a). In the seeds with 30 days after the beginning of the germination process was showed the primary root and subsequently the secondarys are originates from besides seedling stem that is responsible of originating the first leaf denominated eophyll (Fig. 2b), as well as the embryo distal portion, the growth and dilatation of the cotyledon originating the haustorium, a globule structure with spongy aspect, in which is responsible of the absorption of the sugars coming from degradation of the endosperm reserve wall occurs (Fig. 2c).

The optical microscopy demonstrated that the center region of the seed testa projection is constituted of extended cells, with thin and irregular walls, besides abundant cytoplasmatic content (Fig. 3a). However the general visualization of the testa projection in the optical microscope, reveals that this is formed by the center region, with pale color and approximately eight layers of parenchymatical cells, coved in the top part that in the lower by an epiderm formed by small cells, in which probably great amount of tannin cell content accumulate, in which it is responsible for the dark aspect of this region (Fig. 3b).

The transversal section of seed endosperm reveal that this is voluminous (Fig. 3c), constituted by cells with irregular and large walls, rich in pit primary fields and that takes up greater part of total cell volume.

The Fig. 4a reveal that the seed have the testa as main mechanical layer that cover and protect the endosperm, however this structure has not tegmen. According the classification of Corner (1976) this seed is testal and with one tegument. The Fig. 4b reveal a general vision of the endosperm longitudinal section, testa and projection testa of *Euterpe oleracea* seed. It was showed a penetration of testa projection in the seed endosperm, probably carrying transport function. The Fig. 4c reveal

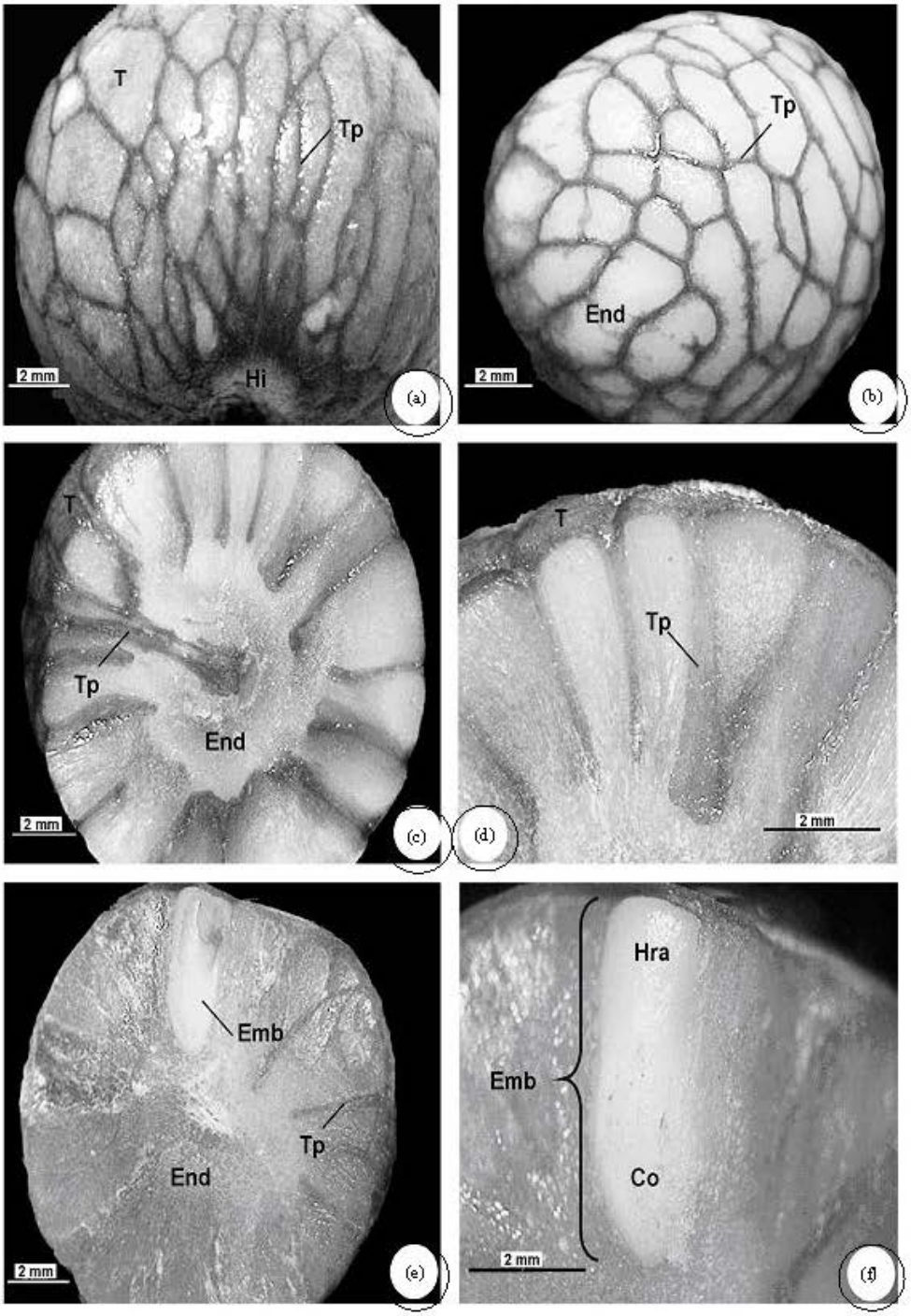


Fig. 1: Aspects of *Euterpe oleracea* mature seed. a: Seed without mesocarpic fibers, reveal details of testa and testa projections (ruminated testa); b: Ruminated endosperm of seed penetrated by testa projections; c and d: Seed sections showing the endosperm tissue and ruminate endosperm; e: Embryo encased into the seed; f: General aspect of embryo. Co: cotyledon, Emb: embryo, End: endosperm, Hi: hilum, Hra: hypocotyl radicle axis, T: testa, Tp: testa projection

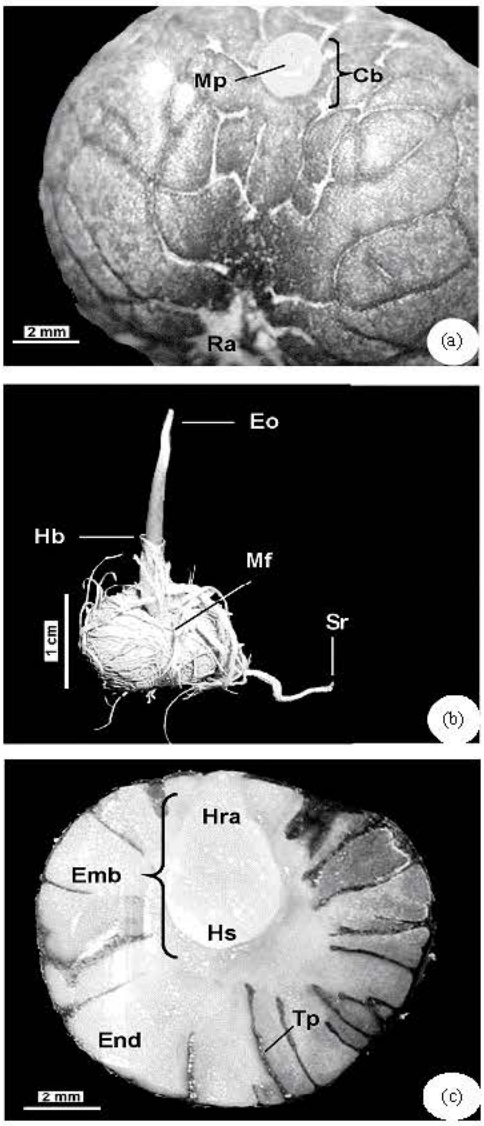


Fig. 2: Germination of *Euterpe oleracea* seed. a: Seed with 10 days after of the germination, reveal the emergence of cotyledonal bud containing the meristematic pole by rupture of the seed opercule (testa in disc format); b: Seed morphology with 30 days after of the germination, reveal the emergency of eophyl; c: General aspect of seed morphology with 30 days after of the germination: embryo and haustorium development. Cb: cotyledonal bud; Emb: embryo; End: endosperm; Eo: eophyl; Hb: hypocotyl base; Hra: hypocotyl radicle axis; Hs: haustorium; Mf: meristematic fiber; Mp: meristematic pole; Ra, rafe; Sr: secondary root; Tp: testa projection

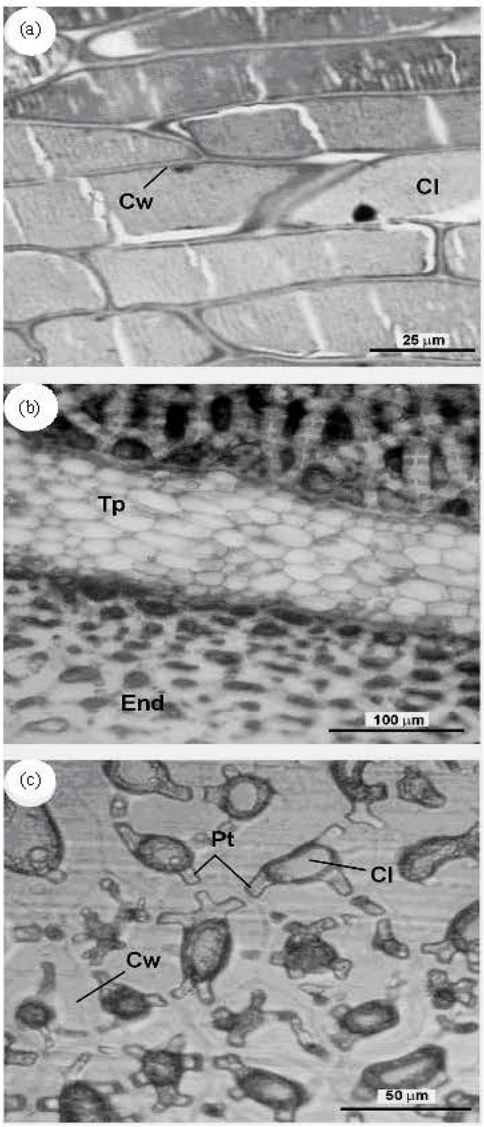


Fig. 3: Photomicrographs of *Euterpe oleracea* seed. a: Detail of the testa projections, with thin cellular walls and cytoplasm rich in tannin cell content; b: Endosperm throughout by the testa projection; c: Detail of morphology of the cell endospermatic, with irregular and large walls, with pits that take up greater part of the total cell volume. Cw: cellular wall; Cl: cellular lumen; End: endosperm; Pt: pits; Tp: testa projection

the longitudinal section of seed endosperm, in which this tissue is formed by elongated cells, with thick cellular wall and greater number of pit primaries and cytoplasm that was strongly stained.

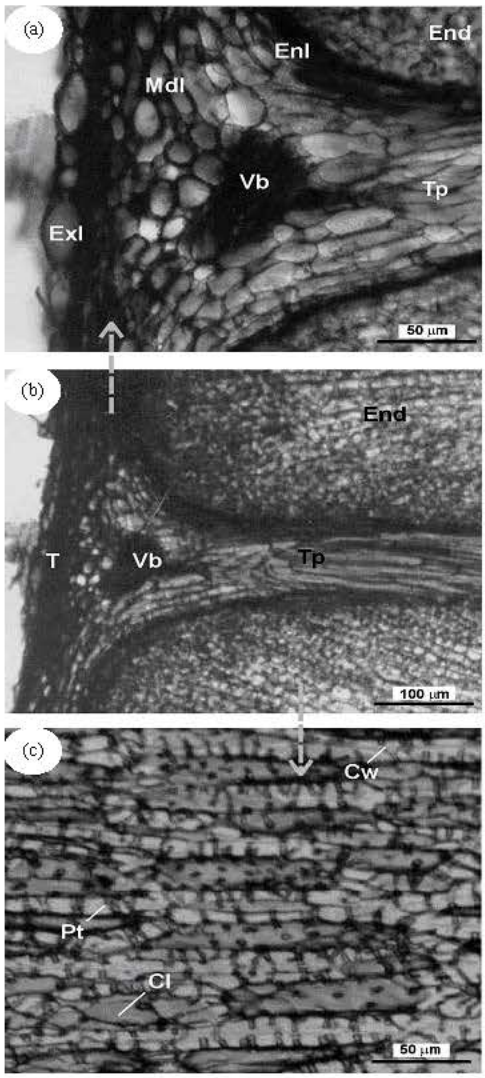


Fig. 4: Photomicrographs of *Euterpe oleracea* seed. a: Mesophyll projection of testa with vascular bundle and layers of composition. b: Endosperm penetrated by testa projection; c: Detail of endosperm with cellular components and structure: pits and large cellular wall. Cl: cellular lumen; Cw: cellular wall; End: endosperm; Enl: internal layer; Exl: external layer; Mdl, medium layer; Pt: pits; T: testa; Tp: testa projection; Vb: vascular bundles

## DISCUSSION

The ruminated endosperm showed occurred by consequence of the growth or by to have folds in the seed testa that originate projections that penetrate in the young endosperm interior under development phase. Similar results were showed by Beltrati and Paoli (2003) and Neto (2004) in several palm seeds, in which both testa and seed endosperm of the *Euterpe oleracea* are ruminated. Moreover, the description of embryo carried out in this study corroborate the research of Beltrati and Paoli (2003).

Similar germinative behavior was found in seeds of the *Euterpe precatoria* (Aguiar and Mendonça, 2001, 2002, 2003) and *Phoenix roebelenii* (Iossi, 2002). These results corroborate the studies carried out by De Paula (1975), in which the testa projections are formed by 8 to 13 cell layers, with fine and pectic-cellulosic walls and rich reserve substance.

According to Lorenzi *et al.* (1996) the folds or invaginations of testa projections of palm seeds located in endosperm leave this tissue with irregular aspect, denominate ruminated endosperm (Beltrati and Paoli, 2003). These testa invaginations in the endosperm of *Euterpe oleracea* seed originate cell radial lines (testa projections) that distribute through the tissue and carry out some function of carbohydrate conduction coming from the degradation of the endosperm reserves in the direction to embryo haustorium during the germination (Neto, 2004), however this author describe that only the seed testa is ruminated, not the endosperm. This way, future studies on the endosperm and testa ontogeny of this seed are important to confirm the origin of rumination. The results of this investigation are similar with the findings in the description seeds of *Euterpe precatoria* (Aguiar and Mendonça, 2003), *Euterpe edulis* (Panza *et al.*, 2004), as well as *Euterpe oleracea* (Neto, 2004), in which belonging to same genus of *Euterpe oleracea*. This way, this study reveals that not exist morphological differences among these seeds.

According to De Paula (1975) the immature seeds, have the endosperm with fine walls, pectic-cellulosics, storage of lipid bodies and rare starch grains. However in mature seeds the endosperm is constituted of large walls, rich in celluloses and hemicelluloses, with pits that have the capacity of lipid and hemicellulose storage.

The seed testa is constituted by several cell layers that might be organized in three groups: external layer, medium layer and internal layer. The external layer is formed by the external epiderm of testa and corresponds the layer with greater cells, large walls, lignified and rich in tannin, phenolic compost that is responsible for the seed color, increase of the testa stiffness and protection against predatories and microorganisms (Beltrati and Paoli, 2003).

The medium layer is the testa mesophyll, constituted by greater cells, with large walls, lignified and that probably has mainly the characteristic of cell growth by elongation, originating testa projections, which penetrate

in endospermatic tissue. A strong evidence that confirms the cell elongation might the fact of the mesophyll cells that has greater and roundly aspect, besides large and lignified walls, compared to the testa external layer. Whereas the cells that constitute the testa projections are elongated, with fine and irregular walls. Other important characteristic of the testa mesophyll is the presence of vascular bundles in the projection regions, being showed that the testa projections has probable function of carbohydrate transport, coming from the endosperm reserve degradations in the direction to embryo haustorium during the germination.

The internal layer is an internal epiderm, constituted by an irregular single file of the small cells, with fine and lignified walls and rich in tannin cell content. As well as it covers internally all the testa mesophyll and the projections penetrated in the endosperm. Similar results were found in *Euterpe precatoria* (Aguiar and Mendonça, 2003), however in this study the mesophyll cells and seed internal layer do not originate projections.

The *Euterpe oleracea* Mart. seed has solid ruminated endosperm, besides is constituted by large cellular walls, with several pit primary fields. The testa is ruminated, constituted by fine cellular walls, divided in three layers (external, medium and internal), being that the medium layer has vascular bundles. The embryo is conic, basal and axial linear, with two regions, being a proximal (hypocotil-radicle axis) and other distal (cotyledon).

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