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Reserve Carbohydrates and Lipids from the Seeds of Four Tropical Tree Species with Different Sensitivity to Desiccation

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ABSTRACT

Considering the importance of water content for the conservation and storage of seeds, and the involvement of soluble carbohydrates and lipids for embryo development, a comparative study was carried out among the seeds of Inga vera *(ingá),* Eugenia uniflora *(pitanga), both classified as recalcitrant, and* Caesalpinia echinata *(brazilwood) and* Erythrina speciosa *(mulungu), considered as orthodox seeds. Low concentrations of cyclitols (0.3-0.5%), raffinose family oligosaccharides (ca. 0.05%) and unsaturated fatty acids (0-19%) were found in the seeds of ingá and pitanga, while larger amounts of cyclitols (2-3%) and raffinose (4.6-13%) were found in brazilwood and mulungu, respectively. These results, in addition to higher proportions of unsaturated fatty acids (53-71%) in orthodox seeds, suggested that sugars and lipids played important role in water movement, protecting the embryo cell membranes against injuries during dehydration.*

Key words: Desiccation tolerance, Fatty acids, Seeds, Starch, Sugars

INTRODUCTION

The storage of seeds is an important tool in the *ex situ* conservation of plant species. However, it is necessary to understand the seed behavior with respect to tolerance to desiccation and freezing in order to improve the preservation of seed viability during storage. In general, seeds accumulate large amounts of storage compounds such as carbohydrates, proteins and oils that represent the main source of nutrients for the initial seedling growth (Mayer and Poljakoff-Mayber, 1989) and also for the nutrition of animals and humans. The knowledge of seed biology provides tools for improving the agricultural practices and managing genetic resources (Santos-Mendoza et al., 2008).

Among various cellular attributes, desiccation and freezing tolerance have been focused on storage compounds. Compatible solutes, including many sugars and sugar alcohols, are synthesized by the plants in response to desiccation and low temperature, among other stressing conditions. These compounds are thought to stabilize the sensitive cellular components during drying and freezing (Hincha et al., 2006) and also act as osmoprotectans, improving the water status and

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the cell volume in the frozen state (Crowe et al., 1990).

Plant oils can provide renewable sources of high-value fatty acids for both the chemical and health-related industries (Dyer et al., 2008) and also can protect the plant cells against stressing conditions. For instance, an increase in unsaturated fatty acids was observed for the plants of *Vigna unguiculata* subjected to water stress (Paula et al., 1990). The effects of dehydration were firstly detected in the membrane composition (Navari-Izzo et al. 1989) and this was observed in the leaves of *Sporolobus stafianus*, known as resurrection plant. In this species, a drastic reduction in total lipids and in the amount of unsaturated fatty acids was observed after decreasing the water content (Quartacci et al., 1997).

The seeds of *Erythrina speciosa* Andr. (Leguminosae, Faboideae), like those from *E. caffra* (Nkang 2002), are remarkably tolerant to desiccation and can be stored for several years in a range of environmental conditions in the dried state (data not published). From the same plant family, the seeds of *Caesalpinia echinata* Lam. (Caesalpinioideae) are also tolerant to desiccation (Barbedo et al., 2002). However, these seeds loose their viability within three months when stored under the laboratory conditions and maintain high germinability under sub-zero conditions up to two years (Hellmann et al., 2006). The seeds of *Inga vera* Will. subsp. *affinis* (DC.) T.D. Pennington (Mimosoideae, Leguminosae) are classified as recalcitrant and have one of the shortest storability known (Bilia and Barbedo, 1997). Indeed, these seeds can be stored only up to seven days without utilization of artificial methods (Bilia et al., 1999). The storage of hydrated embryos in polyethylene glycol (PEG) solution was capable of maintaining high germinability until 30 days of storage at -1.7 MPa and 5 ºC (Faria et al., 2006) and 90 days at -2.4 MPa and 10 °C (Andréo et al., 2006). The seeds of *Eugenia uniflora* L. (Myrtaceae) also exhibit short storability, but higher than *I. vera*, and could be stored for six months under controlled conditions (Barbedo et al., 1998). Among other *Eugenia* species, *E. uniflora* seeds are the most tolerant, keeping their germinability with moisture content of 20% (Delgado and Barbedo, 2007).

In the present work, the content and composition of reserve carbohydrates and lipids from the seeds of these four tropical tree species presenting different degrees of tolerance to desiccation, were analyzed aiming at a better understanding of the involvement of these compounds with the cellular protection against drying.

MATERIALS AND METHODS

Mature fruits of *I. vera* Will subsp. *affinis* (DC.) T. D. Pennington, *E. uniflora* L., *C. echinata* Lam. (Borges et al., 2005) and *Erythrina speciosa* Andr. were harvested in 2006 from plantations located at the Botanical Garden in São Paulo (SP, Brazil) – 23°38´S-46°37´W – and in the "Reserva Biológica e Estação Experimental de Moji-Guaçu" (SP, Brazil) – 22°15´S-47°8´W. After the harvest, the fruits were transferred to the "Instituto de Botânica" and the seeds were extracted and mixed at random. The water content (g water per g dry matter - g g^{-1}) was determined (four replicates of five seeds) gravimetrically (oven at 103 ºC ± 3 ºC for17h) according to ISTA (1985). The germination tests were carried out in germination chambers (Marconi MA400) at 25 ± 1 ºC, under 12 h photoperiod, in paper roll moistened previously with tap water, with four replications of 12 seeds (Mello and Barbedo, 2007). The germination was evaluated every two days from the sowing day, by registering the protrusion of the primary root (at least 5 mm). The number of seedlings with normal development of both radicular system and shoots was also registered.

Three replications of five embryos from the seeds of each species were separated and analyzed for soluble carbohydrates and starch. For *C. echinata* and *E. speciosa*, the tissues were separated into embryonic axis and cotyledons. Immediately after the excision, both the tissues were weighed, boiled in 80% ethanol (v/v), homogenized with a mortar and pestle, and heated at 80 °C for 10 min. After centrifugation for 5 min at 1000 *g*, the residue was re-extracted twice with 80% ethanol for 5 min at 80 °C. The extracts obtained were analyzed colorimetrically by the phenol-sulfuric method (Dubois et al., 1956) for total carbohydrate determination, using glucose as standard and were expressed as mg per g of dry mass (mg g^{-1} DM). Neutral soluble carbohydrates were analyzed by high performance anion exchange chromatography coupled with pulsed amperometric detection (HPAEC/PAD) after deionization through a column consisting of equal amounts of Dowex–1 (Cl^- form) and Dowex–50W (H^+ form). The HPAEC/PAD system (Dionex ICS-3000) used consisted of a 4 x 250 mm CarboPac PA-1 column and a gradient of 250 mM sodium hydroxide (eluent B) and water (eluent A) with the following programme: 0-2 min, 7% eluent B; 2.1-25 min, 4% eluent B; 25.1-30 min, 80% eluent B; 30.1-35 min, 7% eluent B, and the rate flow of 0.2 mL min^{-1}. The residue of soluble carbohydrate extraction was subjected to enzymatic determination of starch according to Amaral et al. (2007).

Total lipids were extracted from the powdered material (0.2-2 g DM) using soxhlet and hexane as solvent during four hours. After the extraction, the solvent was evaporated and yield was calculated. The oil extracted was saponified and methylesterified according to Mayworm et al. (1998), with modifications. The methyl esters were separated and analyzed in a gas chromatograph (Agilent 6890 series) with a mass spectrometry (Agilent 5973 Network) on a 30 m X 0.35 mm column HP5 with silica, temperature range from 160 °C increasing 5 °C min^{-1} during three minutes and 5 °C min^{-1} until 175 °C, followed an increase of 1.5 °C min^{-1} until 195 °C and finally, an increase of 10 °C min^{-1} until 300 °C for five minutes. The peaks of concentration in each case from the unknown samples were compared with the library and were matched. The percentage of fatty acid constituents were obtained by the display unit of the instrument.

RESULTS

The water content of mature seeds of *E. speciosa* and *C. echinata* at harvesting were 0.138 and 0.388 g g^{-1} DM, respectively, and the water content of seeds from *E. uniflora* and *I. vera* were 1.236 and 1.214 g g^{-1} DM, respectively. The germination percentages of *E. speciosa*, *E. uniflora* and *I. vera* did not differ among them but were significantly higher than that of *C. echinata* (Table 1). However, the development of normal seedling did not differ among the four analyzed species. The analysis of total soluble carbohydrates showed higher amounts in the embryonic axis of *E. speciosa* seeds (28% DM), followed by the cotyledons of the same species and cotyledons and axis of seeds from *C. echinata* (*ca.* 14%). Intermediate values were obtained for the embryos of *E. uniflora* and the lowest amount was found in the embryos (0.6%) from *I. vera* (Table 2).

The quantification of starch showed that the seeds of *E. speciosa* were poor in this reserve compound, as also the axis of *C. echinata* (0.7-2.3% DM). However, the cotyledons of *C. echinata* and embryos of *I. vera* presented more than 42% DM as starch. Among the analyzed species, the highest content of starch was found in the embryos of *E. uniflora*, representing *ca.* 64% of the seed dry matter (Table 2). The cotyledons of *E. speciosa* contained high amounts of lipids (more than 23% DM) when compared to the other analyzed species and tissues, followed by *C. echinata* cotyledons (more than 17% DM). The embryonic axes of both the species showed lower contents of lipids (7-12%) than those of their cotyledons. In contrast, the embryos of *E. uniflora* and *I. vera* presented less than 1% of lipids in the seed dry mass basis (*ca.* 0.8 and 0.3%, respectively).

The analysis of soluble carbohydrates by HPAEC/PAD showed similar composition in both the axis and cotyledons of seeds from the four tropical tree species. Sucrose was the major sugar present in all the seeds (Fig. 1), except *E. speciosa* with high amounts of the raffinose family oligosaccharides (RFOs). Sucrose contents in the seeds of *E. uniflora* and *I. vera* were lower when compared to the other analyzed species (Table 3), but was the main sugar found in the seeds of *C. echinata*, presenting the highest amounts in the cotyledons (*ca.* 12% DM). Table 3 also shows that raffinose was the predominant carbohydrate in *E. speciosa*, representing more than 13% DM of the embryonic axis. RFOs were absent or were detected in minute proportions in the other studied species. Cyclitols were present in higher proportions in *C. echinata* tissues (2-3%), being 10 times the values found in the other species. Little amounts of monosaccharide (lower than 0.2%) such as glucose and fructose were found in all the analyzed tissues (Table 3), except *E. uniflora*.

Table 1 - Physiologycal parameters of seeds from four tropical tree species harvested at maturity. Means±sd.

Species	Moisture content (g g $^{-1}$DM)	Fresh mass (g seed^{-1})	Dry mass (g seed^{-1})	Germination (%)	Seedling (%)
Erythrina speciosa	0.138±0.020	0.31±0.04	0.27±0.03	100.0±0.0	100.0±0.0
Caesalpinia echinata	0.388±0.018	0.28±0.03	0.19±0.22	89.6±8.0	85.4±10.5
Eugenia uniflora	1.236±0.062	0.39±0.09	0.18±0.05	100.0±0.0	95.0±6.4
Inga vera	1.214±0.034	0.65±0.07	0.29±0.02	100.0±0.0	100.0±0.0

Table 2 - Overall soluble carbohydrate, starch (mg g^{-1} DM) and lipids (%) in seeds of four tropical tree species with different sensitivity to desiccation. Within each column, means followed by the same letter do not differ significantly (Tukey's test, P=0.05, n=3)

Species	Tissues	Soluble carbohydrate (mg g^{-1}DM)	Starch (mg g^{-1}DM)	Total lipids (%)
Erythrina speciosa	Cotyledons	137.76 b	7.77 c	23.29 a
	Axis	285.18 a	15.40 c	12.31 c
Caesalpinia echinata	Cotyledons	148.37 b	420.35 b	17.57 b
	Axis	140.63 b	22.79 c	7.04 d
Eugenia uniflora	Whole embryo	70.10 c	641.03 a	0.84 e
Inga vera	Whole embryo	60.90 c	514.61 b	0.27 f
CV (%)		14.21	16.67	2.22

The relative composition of fatty acids varied among the analyzed species, showing higher diversity in the tissues from *C. echinata* and *E. speciosa* seeds. In the embryos of *I. vera* and *E. uniflora* predominantly palmitic (C16:0) and stearic (C18:0) (saturated fatty acids) were detected. The main fatty acid found in *C. echinata* tissues was the linoleic acid (C18:2), representing *ca.* 25% of the total lipid, while in *E. speciosa* the oleic acid (C18:1) constituted *ca.* 50% of the total lipids from the cotyledons (Table 4).

Figure 2 showed that the proportion of unsaturated fatty acids increased with the potential tolerance to desiccation presented by the analyzed seeds. The embryos of the recalcitrant seeds of *I. vera* and *E. uniflora* showed 100% or more than 80% of the total lipid content as saturated fatty acids, respectively. The proportions of unsaturated and saturated fatty acids were similar in the seeds of *C. echinata* while in the cotyledons of *E. speciosa*, the amounts of unsaturated fatty acids were much higher, figuring more than 70% of the total fatty acids detected.

Table 3 - Composition of soluble neutral carbohydrates (mg g^{-1} DM) in seeds of four tropical tree especies with different sensitivity to desiccation. Cyl = cyclitols; Glu = glucose; Fru = fructose; Suc = sucrose; Raf = raffinose; Sta = stachyose. Within each column, means followed by the same letter do not differ significantly (Tukey's test, P=0.05, n=3).

Species	Tissues	Soluble carbohydrates (mg g^{-1} DM)					
		Cyl	Glu	Fru	Suc	Raf	Sta
E. speciosa	Cotyledons	3.42 d	0.22 b	0.19 b	37.27 d	45.86 b	37.17 a
	Axis	9.76 c	0.64 b	0.55 b	106.28 b	130.79 a	13.04 b
C. echinata	Cotyledons	20.52 b	2.04 b	1.13 b	120.64 a	2.12 c	1.72 c
	Axis	32.27 a	2.32 b	1.21 b	95.90 b	5.46 c	3.91 c
E. uniflora	Whole embryo	3.54 d	5.98 a	10.25 a	50.22 c	0.49 c	0.11 c
Inga vera	Whole embryo	5.94 cd	1.32 b	1.57 b	51.67 c	0.52 c	0.00 c
CV (%)		15.39	43.66	33.18	5.69	8.07	30.58

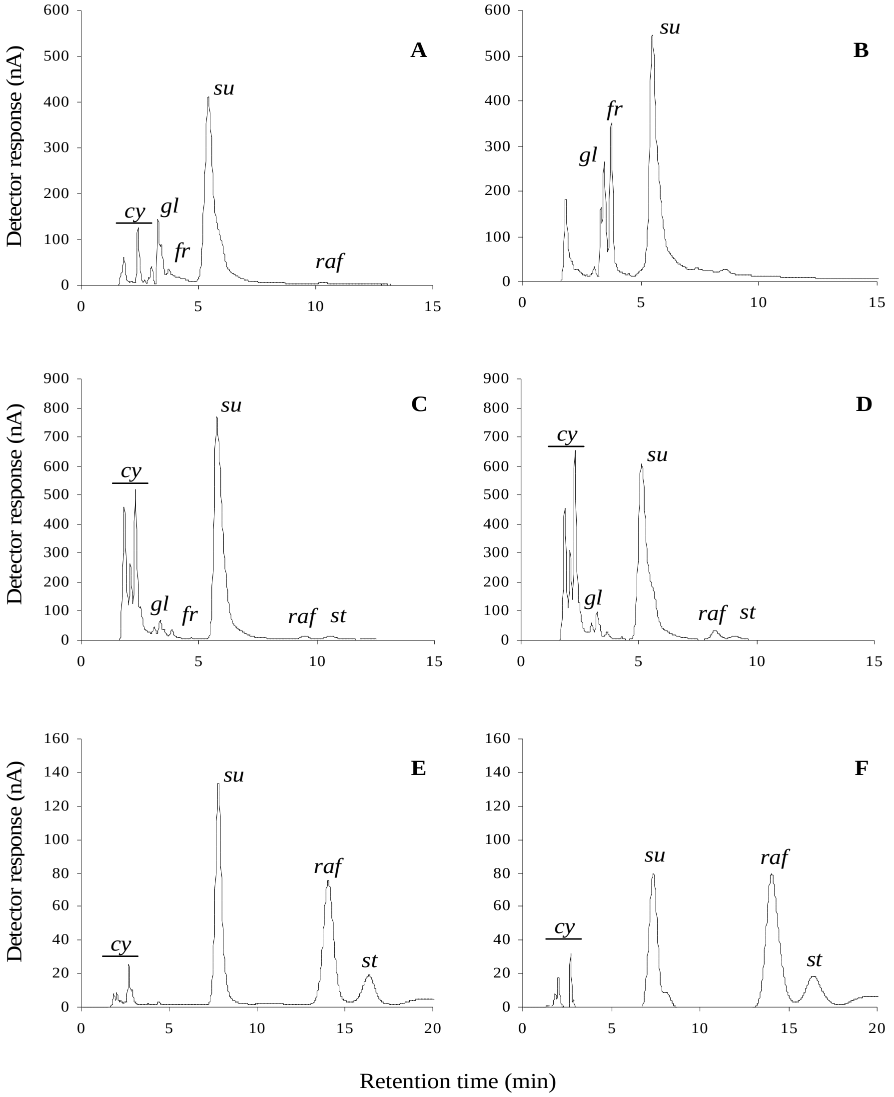

Figure 1 - HPAEC/PAD of neutral soluble carbohydrates of mature seeds from *Inga vera* (A), *Eugenia uniflora* (B), *Caesalpinia echinata* - embryonic axis (C) and cotlyledons (D) – and *Erythrina speciosa* - embryonic axis (C) and cotyledons (D). *cy* – cyclitols, *gl* – glucose, *fr* – fructose, *suc* – sucrose, *raf* – raffinose and *st* – stachyose.

Table 4 - Relative composition of fatty acids (%) from storage lipids in seeds of four tropical tree species with different sensitivity to desiccation.

Fatty acid (%)	***E. speciosa* ***	***C. echinata* ***	***I. vera***	***E. uniflora***
Palmitic (C16:0)	18.95	13.00	66.55	57.38
Stearic (C18:0)	6.47	25.70	26.66	10.10
Oleic (C18:1)	51.77	7.90	0.00	17.23
Linoleic (C18:2)	18.56	45.40	0.00	2.21
Arachidic (C20:0)	1.84	4.10	1.38	7.89
Eicosenoic (C20:1)	0.68	0.10	0.00	0.00
Behenic (C22:0)	1.48	3.40	0.95	2.89
Lignoceric (C24:0)	0.24	0.40	4.46	2.30

*Values correspond to cotyledons only.

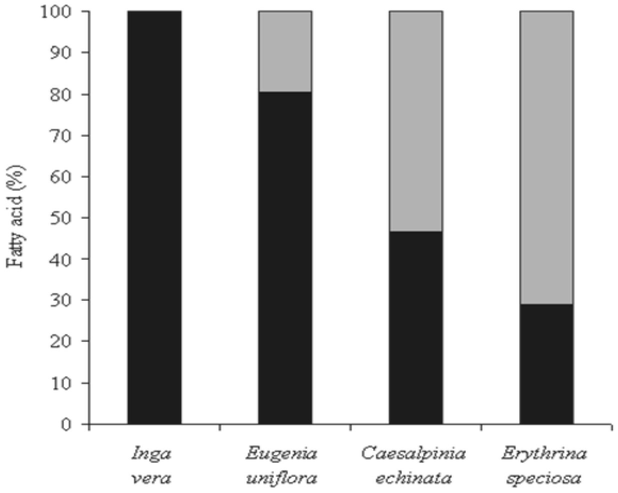

Figure 2 - Percentage of saturated (black column) and unsaturated (gray column) fatty acids in storage lipids from seeds of tropical tree species with different sensitivity to desiccation.

DISCUSSION

Recalcitrant seeds are usually shed at water contents greater than 0.40 g water per g dry matter (g g^{-1}) and lose vigor with relatively little drying, while orthodox seeds are shed with low water content and can withstand the dehydration to as low as 0.05 g g^{-1} (Sershen et al., 2008). The present results showed higher water contents in mature seeds of *I. vera* and *E. uniflora*, the more sensitive species to desiccation, in contrast to what was found in the less sensitive ones from *C. echinata* and *E. speciosa*, both previously considered as orthodox seeds (Barbedo et al., 2002; Nkang, 2002). While seed longevity appears to be a consequence of metabolic activity, desiccation sensitivity can be ameliorated by protection mechanisms that may not affect the storage life span of a given species (Sershen et al., 2008). Thus, at the same environmental condition, it would be possible to find the desiccation tolerant seeds with shorter lifespan than desiccation intolerant ones, as found between the tolerant *C. echinata* and both intolerant *E. involucrata* and *E. brasiliensis* seeds (Barbedo et al., 2002; Kohama et al., 2006). Therefore, associations between both the processes deserve more attention.

The differences found in water content were not related with the germination potential of the seeds analyzed in the present work. The seeds of *C. echinata* presented germination percentage lower than that of other analyzed species. However, Hellmann et al. (2006) have reported higher values for the seeds harvested from the same plantation of *C. echinata*. Therefore, the differences found could be attributed to the seasonal differences in environmental conditions rather than to genetic factors. The high germinability found in the seeds of *E. uniflora* and *I. vera* seeds was consistent with the current literature and with the fact that they were commonly indicated for reforestation (Bilia and Barbedo, 1997).

Concerning the carbohydrate quantification, it was possible to separate the studied species into two groups, one including the desiccation tolerant seeds, *E. speciosa* and *C. echinata* (Nkang, 2002; Barbedo et al., 2002), both with more than 14% DM as soluble carbohydrates, and the other group represented by *E. uniflora* and *I. vera* seeds, with recalcitrant behavior (Delgado and Barbedo, 2007; Bonjovani and Barbedo, 2008) and lower amounts of soluble carbohydrates. A variety of orthodox seeds, including mature legumes, can contain high levels of soluble carbohydrate. *Glycine max* (soybean) comprised 14 and 28% of the dry matter of the axis and cotyledons, respectively (Górecki et al., 2001). According to Pritchard et al. (1995), the intolerance of recalcitrant seeds to desiccation could be related to the low values of soluble carbohydrates contained by the seeds, which was reinforced by the present results relative to *E. uniflora* and *I. vera*, containing 7 and 6% sugars, respectively.

Among the soluble carbohydrates, sucrose is mostly accumulated in the seeds and is thought to play a major role in cellular desiccation tolerance (Hincha et al., 2006). However, this sugar was found in large proportions in the seeds of the four species analyzed in the present study, independently of their tolerance to desiccation and storage behavior. Other compounds have been involved in the acquisition of tolerance to dehydration, including LEA proteins (Barbedo and Marcos Filho, 1998 and references therein), amino acids (e.g. proline), raffinose family oligosaccharides (RFO) and sugar alcohols (Yancey et al., 1982). In *C. echinata* seeds, RFO was present in very low proportions (less than 0.5%), confirming previous reports (Garcia et al., 2006; Hellmann et al., 2008), while cyclitols were found in larger amounts, mainly in the embryonic axis (*ca.* 3%), and might play a protective role during the desiccation and loss of water during seed maturation (Borges et al., 2006), replacing water and stabilizing membranes and other sensitive systems, as proposed for other legume seeds (Peterbauer and Richter, 2001). *E. speciosa* seeds presented RFO as the principal soluble carbohydrates, especially raffinose, in proportion equivalent to that of sucrose (*ca.* 4%). In general, there are differences in the quantities of RFO members in seeds from different species, particularly in legumes (Górecki et al., 2001) and this could be related to their behavior during storage. According to Horbowicz and Obendorf (1994), seed storability depends on the ratio of sucrose to oligosaccharides. Seeds with sucrose: oligosaccharide ratio lower than 1.0 have a storability half-viability period higher than 10 years, whereas those with ratio higher than 1.0 have storability half-viability period lower than 10 years. Therefore, the interactions of these carbohydrates during the storage in desiccation tolerant tissues might promote the hydration of membrane phospholipids, maintaining the conformation and functionality of cell membranes during water loss (Hoekstra et al., 2001). In addition to sucrose (5%), desiccation intolerant seeds of *I. vera* and *E. uniflora* presented only traces of cyclitols and RFO, which probably contributed to the loss of viability after drying and storage of the seeds. In combination with data in the literature these results showed that the ratio of sucrose to oligosaccharides in seed tissues could provide useful information on the seed storage category.

The small proportion of starch in the embryonic axis of the orthodox seeds (less than 2%) could be compensated by the larger amounts of soluble carbohydrates found (14-28%), which were more easily metabolized and could be readily used in the germination process. This positive association between the soluble carbohydrates and seed storability/desiccation tolerance could not be applied to starch contents since the seeds with extremely different sensitivity to desiccation (*C. echinata*, *I. vera* and *E. uniflora*) presented relatively high (42 to 64%) and similar percentage of starch. For most grain and legumes, the largest part of the carbohydrate fraction was starch (Table 5), accounting for about 35-45% of the seed weight (Hedley, 2001).

The oil content in the seeds of the starch storing legumes is not usually higher than about 2%. On the other hand, in the seeds storing high levels of oil, such as soybean, starch only makes up about 1.5% of the seed weight (Hedley, 2001). However, the lipid amounts found in the embryonic axis of *C. echinata* and *E. speciosa* were relatively high (7-12%) and were similar to the values obtained in the seeds of *E. velutina*, a species from the Brazilian Caatinga (Mayworm et al., 1998); the cotyledons of *E. speciosa* showed the same contents (*ca.* 23%) as the seeds of *C. pyramidalis* (Mayworm et al., 1998). The orthodox seeds of some species of *Citrus* can contain more than 50% of lipids (Hor et al., 2005), which has also been reported for the seeds of *Dalbergia miscolobium* from the Cerrado which are very rich in oil (Silva et al., 1998). On the other hand, the desiccation intolerant seeds of *Inga* sp. presented only traces of lipids (Pritchard et al., 1995), which was consistent with the very low values found in the present work for the seeds of *E. uniflora* and *I. vera* (lower than 0.8%). These results reinforced the suggestion that high proportions of lipids could be involved in the mechanism of tolerance to desiccation.

Table 5 – Starch, lipids and fatty acid composition of seeds from some tropical legumes.

Species	**Starch** (%)	**Lipids** (%)	**Fatty acid** (%)		**Reference**
Vigna radiata	-	1.40	18:1	26.4	Anwar et al., 2007
			18:2	41.2	
Phaseolus vulgaris	41.50	1.50	-		Hedley, 2001
Cassia floribunda	-	2.60	-		Vadiel & Janardhanan, 2001
Pisum sativum	45.00	2.70	-		Hedley, 2001
Hymenaea stigonocarpa	-	5.30	16:0	8.9	Matuda & Maria Netto, 2005
			18:0	4.7	
			18:1	31.6	
			18:2	52.3	
			18:3	1.2	
			20:0	0.8	
Cicer arietinum	44.40	5.20	-		Sotelo, 1996
Hymenaea courbaril	-	8.59	-		Pinto et al., 2005
Bauhinia variegata	-	16.61	16:0	19.69	
			18:0	13.98	
			18:2	56.98	
			20:0	1.08	
Bauhinia forficata	-	19.45	18:2	46.47	Faria et al., 2004
			16:0	19.27	
			18:0	13.89	
			20:1	0.51	
Glycine max	1.50	19.70	-		Hedley, 2001
Glycine max	-	20.60	-		Moraes et al., 2006
Glycine max	-	16.62	-		Marega Filho et al., 2001
Mimosa grahamii	-	23.50	-		Ortega-Nieblas et al., 1996
Lonchocarpus muehlbergianus	-	26.80	16:0	11.65	Vallilo et al., 2001
			18:0	5.80	
			18:1	56.00	
			18:2	10.90	
			18:3	12.90	
Dipteryx alata	-	38.20	18:0	5.40	Takemoto et al., 2001
			18:1	50.40	
			18:2	28.00	
			20:0	1.07	
			20:1	2.70	
			22:0	2.60	
			24:0	2.10	
Caesalpinia peltophoroides	-	50.00	-		Corte et al., 2006

The value and application of particular oil is determined largely by its fatty acid composition, and while most vegetable oils contain just five basic fatty acid structures, there is a rich diversity of fatty acids present in nature, many of which have potential usage in industry (Dyer et al., 2008). In the present study, the unsaturated fatty acids, oleic (C18:1) and linoleic acid (C18:2) were the major components of the oil from the seeds of *E. speciosa* and *C. echinata*. The seeds of *E. speciosa* presented *ca.* 50% of oleic acid, similar to *E. velutina* seeds (Mayworm et al., 1998) and the proportion of linoleic acid in *C. echinata* seeds (45%) was similar to that reported for the seeds of *C. pyramidalis* (Mayworm et al., 1998). In addition to the taxonomic features, these similarities could be related with seed behavior as observed by Liu et al. (2006). They emphasized that the orthodox seeds presented higher proportion of linoleic acid (C18:2) than the recalcitrant seeds, in which the predominant fatty acid was the linolenic acid (C18:3). Additionally, recalcitrant seeds exhibited higher fatty acid saturation in the phospholipids and higher phase transition temperature when compared to orthodox seeds (Liu et al., 2006). Thus, it would be interesting to study the differences in the lipid composition among the seeds from the species with different desiccation tolerance levels, but into the same genus, *e.g. Eugenia* species as demonstrated by Delgado and Barbedo (2007). Taking altogether, the present results suggested that lipids stored in the analyzed seeds could play, in addition to sugars, an important role in the water movement, protecting the embryo cell membranes against the injuries during seed dehydration.

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RESUMO

Os compostos de reserva das sementes, além de suprirem energia para o desenvolvimento embrionário, desempenham importantes funções relacionadas à proteção celular contra secagem e congelamento. Considerando a importância do teor de água para a conservação e para o armazenamento de sementes de espécies arbóreas e o envolvimento dos carboidratos solúveis e de lipídios nesses processos, foi realizada uma análise comparativa desses compostos em sementes de *Inga vera* (ingá) e *Eugenia uniflora* (pitanga), consideradas recalcitrantes, e em *Caesalpinia echinata* (pau-brasil) e *Erythrina speciosa* (mulungu), com comportamento ortodoxo. Baixas concentrações de ciclitóis (0,3-0,5%), de oligossacarídeos da série da rafinose (0,05%) e de ácidos graxos insaturados (0-19%) foram encontradas em sementes de ingá e pitanga, enquanto maiores quantidades de ciclitóis (2-3%) e de rafinose (4,6-13%) foram encontradas nas sementes de pau-brasil e mulungu, respectivamente. Estes resultados, juntamente com as altas proporções de ácidos graxos insaturados (53-71%) presentes em sementes de pau-brasil e mulungu, sugerem que os açúcares e os lipídios poderiam desempenhar papel importante no movimento de água, protegendo as membranas celulares embrionárias contra injúrias provocadas durante a dessecação.

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