Notes on Economic Plants

Underutilized Annona Species from the Brazilian Cerrado and Amazon Rainforest: A Study on Fatty Acids Profile and Yield of Seed Oils¹

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Annona (Annonaceae) is an important source of fruits in the Brazilian Cerrado and the Amazon Rainforest. Some Annona species are widely commercialized as fresh fruit or as frozen pulp. Seeds are accustomedly discarded. Our main goal was to analyze fatty acids profile from seeds of *A. crassiflora* and *A. coriacea* from Cerrado, *A. montana* from Amazon Forest, and three cultivars (*A. cherimola* cv. Madeira, *A. cherimola* x *A. squamosa* cv. Pink's Mammonth and *A. cherimola* x *A. squamosa* cv. Gefner). The total oil yield ranged between 20 and 42% by weight of dry mass. The *A cherimola* x *A. squamosa* cv. Gefner has significantly higher total lipid yield than all other samples. 100 g of fruit of this species present 6-8 g of seeds. Considering the fruit production of Chile (over 221 ton of fruits/year), more than 1300 ton of seed/year could be obtained, which could provide at least 200 ton of seed oil. Oleic acid was predominant for most samples, but for *A. montana* linoleic acid was the most abundant FA. Phenotypic variation on FAME profile was observed. These new data are an urgent requirement for supporting conservation programs, mainly for Cerrado areas in Brazil.

Key Words: Lipids, fatty acids, seeds, Annona, Annonaceae.

Introduction

Annonaceae is a large family that is widely distributed in tropical and subtropical regions of the world, comprising 120 genera (Joly 2002). *Annona* L., the largest genus of this family, is an important source of fruits in the Brazilian cerrado and the Amazonian rainforest. Several species of this genus are economically important, such as *A. squamosa* L. (sugar apple), *A. muricata* L. (soursop), *A. reticulata* L. (custard apple), *A. cherimola* Miller (cherimoya), and the hybrid *A. cherimola* x *A. squamosa* (atemoya). In the cerrado, the wild species *A. crassiflora* Mart. and *A. coriacea* Mart. are commonly appreciated by the local population who use fruits *in natura* or prepared as juices,

desserts, and ice cream (Lorenzi and Matos 2001). However, these species are underutilized, and standard information related to nutritional properties is rare (Pinto et al. 2005).

Besides the fruit pulp, there are data that report the use of seeds of some domesticated and wild species of *Annona* (Pinto et al. 2005). Seeds are common sources of oils with nutritional, industrial, and pharmaceutical importance. However, for most nontraditional oleaginous species, seeds are normally discarded. Recently, the number of studies related to economic exploitation of seeds proceeding from new oleaginous vegetable sources has increased (Li et al. 2009).

Higher proportions of unsaturated fatty acids in the diet have been pointed out as important for human nutrition (Bruneton 1995). Linoleic and linolenic acids, commonly found on vegetable sources, are not synthesized in the human organism, yet are considered essential to human diet.

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Although some data have been published related to fatty acid composition of seed oil for *Annona* species, available profiles show unsaturated fatty acids as major components (Wélé et al. 2004).

This paper analyzes the fatty acids profile from seeds of the following species: *A. crassiflora* and *A. coriacea* from the cerrado, *A. montana* Macfad. & R. E. Fr. from the Amazon forest, and three widely cultivated cultivars (*A. cherimola* cv. Madeira, *A. cherimola* x *A. squamosa* cv. Pink's Mammonth, and *A. cherimola* x *A. squamosa* cv. Gefner).

Material and Methods

Fruit samples were harvested from domesticated and wild species of *Annona* at small farms in the state of São Paulo and national conservation areas in Brazil (Table 1).

Seeds were dried at 60° C and powdered; the extraction of oils was performed with *n*-hexane in a Soxhlet apparatus for six hours. The solvent was removed by using a rotator evaporator under reduced pressure at 40°C, and the residue was completely dried by a stream of nitrogen. Total lipid content was calculated by the gravimetric method.

The lipids were saponified for 2 hours under reflux with 10 mL of 10% KOH in methanol. After this period, samples were acidified with 10% HCl to pH 4.0–5.0. The fatty acids were washed with chloroform four times (Grunwald and Endress 1988); the extracts were combined and the solvent removed as described above. Fatty acids (either methylation or esterification) were developed using diazomethane solution (Vogel 1971).

Fatty acid methyl esters (FAME) were analyzed by gas chromatography-mass spectrometry (GC-MS HP 5890 ser.II Plus/ HP 5989B), employing a capillary column (DB-5HT—30 m×0.32 mm i.d.). Initial oven temperature was 120°C, increasing 10°C/min up to 280°C, remaining 10 minutes. The injector temperature was 300°C. Helium was used as carrier gas at 1.2 mL/min. Sample injection volume was 1 μ L and the split ratio was 50. The source temperature was 250°C, the quadrupole temperature was 150°C, and the electron multiplier voltage adjusted for 70 eV. FAMEs were identified by comparison of the obtained mass spectra with the Wiley 275 database and standard samples.

The total lipid content was submitted to One Way ANOVA. When significant variations ($p \le 0.05$) were detected, the samples were submitted

	Species/cultivars	Z	Localities
Wild species	A. crassiflora	3	Estação Experimental e Ecológica de Jataí e Reserva Biológica e Estação Experimental de Mogi Guaçú (São Paulo), Parque Estadual Rola Moça de Belo Horizonte (Minas Gerais)
	A. coriacea	μ,	Reserva Biológica e Estação Experimental de Mogi Guaçú (São Paulo)
	A. montana	1	Centro de Projetos e Estudos Ambientais da Amazônia (Amazonas)
Domesticated species	A. cherimola cv. Madeira	ĉ	Pedra Bela (São Paulo)
	A. cherimola x A. squamosa cv. Gefiner	2	Indaiatuba (São Paulo)
	A. cherimola x A. squamosa cv. Pink's Mammoth	2	Santa Isabel (São Paulo)

to the Method of Bonferroni using SigmaStat software (Neter et al. 1996).

Results and Discussion

The total lipids amount of Annona species ranged from 203.42±18.8 g/kg of dry mass to 421.57±7.87 g/kg of dry mass, which means that 20-40% of dry weight of seeds of these species are lipids (Table 2). Two different cultivars of the commercial hybrid A. cherimola x A. squamosa were investigated. While seeds of the hybrid A. cherimola x A. squamosa cv. Gefner have significantly higher total lipid content (421.57±7.87 g/kg dry mass) than all other samples, A. cherimola x A. squamosa cv. Pink's Mammoth presented total lipid yield similar to other species (Table 2). Lalas and Tsanknis (2002) described significant differences between seed oil yield of both Moringa oleifera Lam. cv. Periyakulam and Moringa oleifera cv. Mbololo.

Despite limited data related to quantitative analyses of seed oil for species of Annona, previous research of A. muricata and A. squamosa suggested around 20% of dry seed weight is due to lipid content (Andrade et al. 2001; Onimawo 2002). Higher yields have been detected for A. crassiflora and A. coriacea, 35% (Caramori et al. 2004) and 46% (Agostini et al. 1995), respectively. Similar amounts were observed for these species in the present study. A. crassiflora presented 345.8±7.3 g/kg and A. coriacea had 447.0 ±0.28 g/Kg (Table 2).

Oil amounts found in seeds of Annona suggest a potential use for these materials, which are commonly discarded. Many traditional oleaginous species that are consumed world-wide contain even less oil in the seeds; for example: corn-Zea mays L. (90-170 g/Kg) (Reynolds et al. 2005), olive—Olea europaea L. (150 g/kg), soybean-Glycine max (L.) Merr. (150-200 g/ kg), and sunflower-Helianthus annuus L. (200-300 g/kg) (Bruneton 1995).

Low qualitative variation in fatty acid profiles among Annona species was observed. Palmitic (16:0), stearic (18:0), oleic (18:1), and linoleic (18:2) acids were detected in all analyzed species (Table 2). Other unsaturated (linolenic-18:3, and gondoic-20:1) and saturated (arachidic-20:0) fatty acids were rare.

Oleic acid was the main fatty acid in all samples, followed by linoleic acid. Palmitic and stearic acids were detected at similar proportions. Oleic acid (18:1) has already been described in

		Fatty acids							
Species	Yield	16:0	18:0	20:0	Sat ^a	18:1	18:2	18:3+20:1	$\mathrm{Uns}^{\mathrm{a}}$
A. crassiflora	345.8±7.3	8	9	1	15	50	34	1	85
A. coriacea	447.0	13	4		17	51	30	2	83
A. montana	212.5	16	С		19	30	49	2	81
A. cherimola cv. Madeira	203.4 ± 18.8	13	7	1	21	45	33	1	79
A. cherimola x A. squamosa cv. Pink's Mammoth	204.3 ± 12.6	22	6		31	51	15	\mathcal{C}	69
A. cherimola x A. squamosa cv. Gefner	421.6	13	10	1	24	49	27		76
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TABLE 2. SEED TOTAL LIPIDS YIELD (G/KG) AND FATTY ACID PROFILES (%) OF SOME ANNONA SPECIES

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seed oils of other *Annona* species, such as *A. muricata*, *A. senegalensis* Pers. (Wélé et al. 2004), and *A. squamosa* (Andrade et al. 2001).

High concentration of monounsaturated fatty acids has been indicated as a desired characteristic in food oil, and has an increased economic value on the international market. Olive oil, a traditional source of oleic acid, has been suggested as responsible for lowering the risk of several types of cancers (Colomer and Menéndez 2006). Unsaturated fatty acids from the *omega*-family are also important for the human diet. Linoleic acid (*omega*-6) is classified as one of the two essential fatty acids for humans. Many investigators have suggested that oleic (O'Byrne et al. 1997) and linoleic (Lorgeril et al. 2001) acids decrease plasma cholesterol and minimize the risk of coronary heart disease.

Based on fatty acids composition, *Annona* seed oil could be used for human diet. However, an initial plant breeding is needed, since acetogenins, alkaloids, and saponins (Chen et al. 1999) have been detected in *Annona* seeds. These substances are considered toxic for human health.

Besides food application, *Annona* seed oil could be applied in formulation of new types of coatings and/or in the cosmetology industry. In recent years, interest in new vegetable oils for paint industries has increased. The high proportion of unsaturated fatty acids on *Annona* seed oil allows its classification as a semi-secant oil, which is useful for paint industries. For cosmetology application, rich oleic acid oils are considered a good vehicle for drug solubility-partitioning (Davis et al. 2002), and this fatty acid has been used as an emollient, surfactant, and thickening agent (Gonzalez 2007).

Biodiesel has become more attractive because of its environmental benefits as compared to nonrenewable fuels. This fuel source is said to reduce engine wear and produce less harmful emissions (Demirbas 2005). The world has been confronted with the crises of fossil fuel depletion and environmental degradation. The indiscriminate extraction and consumption of fossil fuels has led to a reduction in petroleum reserves. Alternative fuels, energy conservation and management, energy efficiency, and environmental protection have become important social and political issues in recent years. Biodiesel obtained from vegetable oils, which is an environmentally friendly fuel that can be used in any diesel engine without modification, is considered a promising option (Barnwal and Sharma 2005). Several oleaginous sources, such as sunflower, soybean, and peanut, characterized by higher proportions of unsaturated fatty acids, have gained importance as potential sources of raw material for biodiesel production (Bruneton 1995). Crude seed oils of *Annona* species should also be considered as potential sources for biodiesel production.

Our data suggest a wide range of alternative economic uses for seeds of *Annona* species. *A. cherimola* fruit production, for example, reaches 5–20 ton/ha per year (FAO/Ecocrop 2010). Seeds of this species are usually discarded. Currently, Chile is one of largest producers of fruits of *A. cherimola*, surpassing 220 tons of fruits per year (ICUC 2002). Each 100 g of cherimoya fruit provides 6–8 g of seeds. Therefore, considering the fruit production of Chile, more than 1,300 tons of seed/year could be obtained from this crop, which could provide at least 200 tons of crude seed oil.

Potential economic uses are also possible from wild Annona species. A. crassiflora, A. coriacea, and A. montana reveal high amounts of oleic and linoleic acids in their seed oils, important fatty acids for food and cosmetic industries. Besides fatty acid content, these species occur in important Brazilian ecosystems. A. crassiflora and A. coriaceae are widespread in Brazilian cerrado, and A. montana occurs in the Amazonian forest. These new data may contribute to selection of desired genotypes in breeding programs and/or germplasm conservation banks of Annona.

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