Minor components in oils obtained from Amazonian palm fruits

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SUMMARY

Minor components in oils obtained from Amazonian palm fruits

This study deals with the characterization of minor compounds in oils obtained from the mesocarp of fruits of the main palm species from the State of Amapá, Brazil, i.e. bacaba (Oenocarpus bacaba), buriti (Mauritia flexuosa), inajá (Maximiliana maripá), pupuña (Bactris gasipaes) and tucumá (Astrocaryum vulgare). The concentration of minor glycericidal compounds, i.e. dimeric triacylglycerols (TAG), the oxidized TAG and diacylglycerols (DAG) related to oil quality, and the compounds of unsaponifiable matter, i.e. hydrocarbons, aliphatic alcohols, sterols and tocopherols have been determined.

The results indicate that the extracted oils had good initial quality, with DAG as the major glycericidal compound. The contents of hydrocarbons (50-734 mg·kg\textsuperscript{-1}) and aliphatic alcohols (80-490 mg·kg\textsuperscript{-1}) were highly variable with inajá oil containing the highest contents. In the case of tocopherols, buriti (1567 mg·kg\textsuperscript{-1}) and tucumá (483 mg·kg\textsuperscript{-1}) oils had the highest contents and the presence of significant amounts of tocotrienols was only detected in inajá oil. Finally, high concentrations of sterols were found in all the samples, particularly in the oils from pupuña (4456 mg·kg\textsuperscript{-1}) and tucumá (2708 mg·kg\textsuperscript{-1}), with \(\beta\)-sitosterol being the major sterol in all the samples with percentages between 65 and 83%.


1. INTRODUCTION

Oils and fats are complex mixtures containing a wide range of groups of compounds, where each of the groups in turn can contain a very wide range of individual components. Apart from the major compounds, i.e. triacylglycerols, there are two minor fractions of interest in fats and oils. On one hand, the unsaponifiable fraction is constituted by a high number of individual compounds belonging to different groups of components such as hydrocarbons, waxes, sterols, tocopherols, etc. Their importance in the characterization and detection of oil mixtures are of great interest (Apapacio and Aparicio-Ruiz, 2000; Janssen et al., 2009). Also, the nutritional value of edible oils depends on the content and composition of biological active compounds from this fraction such as phytosterols which prevent the intestinal absorption of cholesterol in humans, resulting in a lowering of serum cholesterol (Mackay and Jones, 2011) or tocopherols, vitamin E compounds only synthesized by plants, which are well recognized for their effective inhibition of lipid oxidation in foods and biological systems (Kamal-Eldin and Appleqvist, 1996).
On the other hand, there is a second fraction of compounds naturally occurring in oils and fats related to the TAG species. They are the mono and diacylglycerols, free fatty acids, oxidized acylglycerols and dimeric triacylglycerols. The determination of these minor components is of great value in establishing oil quality given the relationship between their contents and the incidence of the two main reactions undergone by fats and oils, i.e., hydrolysis and oxidation (Dobarganes et al., 1988).

Native palm trees belong to the Arecaceae family and are among the most useful plant resources for the Amazonian population. Previous studies starting from different species have shown that their fruits are of great potential interest due to their high lipid content and fatty acid composition (Clement et al., 2005). The information on minor compounds is scarce although studies have been carried out on some species (Yuyma et al., 2003; Bereau et al., 2003; Rodrigues et al., 2010; Montúfar et al., 2010; Vázquez-Ocmín et al., 2010) report the presence of bioactive compounds at considerable levels. Research remains to be done in order to know the specific composition of these sources and even the possibility of them being used as specialty oils (Madawala et al., 2012) in the same way as other oils extracted from fruits.

The aim of this study was to determine the minor compounds present in the oils obtained from the fruits of the main palms from the State of Amapá, Brazil, i.e., bacaba (Oenocarpus bacaba), buriti (Mauritia flexuosa), inajá (Maximiliana maripa), pupunha (Bactris gasipaes) and tucumá (Astrocaryum vulgare). A previous detailed study of the triacylglycerol species present in the five oils was carried out by means of HPLC and GC (Santos et al., 2013). In this paper, the contents of glyceridic compounds, i.e., dimeric triacylglycerols, oxidized triacylglycerols and diglycerides, as well as the main groups from the unsaponifiable matter, i.e., hydrocarbons, aliphatic alcohols, sterols and tocopherols have been quantified.

2. MATERIALS AND METHODS

2.1. Materials

Five samples of fruits from five palm species, i.e., bacaba (Oenocarpus bacaba), buriti (Mauritia flexuosa), inajá (Maximiliana maripa), pupunha (Bactris gasipaes) and tucumá (Astrocaryum vulgare), were collected in the State of Amapá, Brazil. The mesocarp of the fruits was separated, moisture was eliminated by freeze-drying before lipid extraction and the lyophilized samples were maintained at −30 °C until extraction and analysis.

2.2. Lipid extraction

The total amount of lipids in the samples was obtained by Soxhlet applying an extraction period of 6 h and diethyl ether as solvent (AENOR, 1991). Then, the solvent was evaporated under vacuum and the extracted oil was dried to constant weight using a stream of nitrogen.

2.3. Analytical determinations

2.3.1. Minor glyceridic compounds

The contents of minor glyceridic compounds were determined gravimetrically according to the IUPAC Standard Method 2.507 (IUPAC, 1992) with slight modifications. Thus, the non-polar and polar fractions were separated from 1 g of oil by silica column chromatography. The non-polar fraction, which contains the non-polar TAG, was eluted with 150 mL of n-hexane/diethyl ether (90:10, v/v). A second fraction, which comprises the total polar compounds, was eluted with 150 mL of diethyl ether. The solvents were evaporated and the contents of the non-polar and polar fractions were determined gravimetrically. The efficiency of the separation was checked by thin layer chromatography using hexane/diethyl ether/acetic acid (80:20:1, v/v/v) for the development of plates and exposure to iodine vapor to reveal the spots. The polar fraction was analyzed by HPSEC to determine the content of dimeric and oxidized monomeric triacylglycerols (TAG), as well as diglycerides (DG) and fatty acids. A chromatograph equipped with a Rheodyne 7725i injector with a 10 μL sample loop, a Knauer 1200 HPLC pump (Knauer, Germany) and a Merok refractive index detector was used. The separation was performed on two 100 and 500Å PL gel columns (30 cm x 0.75 cm I.D.) packed with porous, highly cross-linked polystyrene-divinylbenzene copolymers (film thickness 5 μm) (Agilent Technologies, Santa Clara CA, USA) connected in a series. Tetrahydrofuran (1 mL min⁻¹) was used as the mobile phase and samples were analyzed at concentrations between 15 and 20 mg·mL⁻¹ in tetrahydrofuran (Dobarganes et al., 2000).

2.3.2. Unsaponifiable matter

Unsaponifiable matter was determined according to the AOCS Official Method Ca 6b-53 (AOCS, 2001).

2.3.3. Hydrocarbons

Hydrocarbons were obtained by adsorption chromatography starting from 1 g of oil according to the IUPAC Standard Method 2.507 (IUPAC, 1992) for silica column preparation except that the glass column was filled with a slurry of silica gel and hexane, the elution solvent for hydrocarbons. The elution of hydrocarbons is carried out with 150 mL of hexane. One mL of a solution of squalane (1,025 mg·mL⁻¹) was initially added to the samples as internal standard for quantitative purposes. GC was performed using an Agilent 7890A chromatograph.
2.3.4. Tocopherols and aliphatic alcohols

The composition of free sterols in the oils was determined in the unsaponifiable fractions obtained from 2 g of samples according to the IUPAC Standard Method 2.404 (IUPAC, 1992). Previously, for quantification purposes, 1 mg of α-cholestanol and 0.5 mg of heneicosanol were added to the samples as internal standard for free sterols and aliphatic alcohols, respectively. Both groups of compounds were isolated by thin layer chromatography before silylation and analysis. Separation by GC was performed using an Agilent 7890A chromatograph equipped with a flame ionization detector, a PTV injector and a HP-5 fused silica capillary column (30 m length, 0.32 mm i.d., 0.20 μm film thickness). Hydrogen was used as carrier gas. The detector temperature was 325°C. The initial oven temperature was 75°C (1 min) and a ballistic temperature gradient of 40°C min⁻¹ to 250°C and then maintained for 25 min for free sterols. For aliphatic alcohols, the initial oven temperature was 75°C (1 min), a first ballistic temperature gradient of 40°C min⁻¹ to 180°C and then a second ramp to 300°C at 5°C·min⁻¹. The maximum temperature was maintained for 30 minutes.

2.3.5. Tocopherols

Tocopherols were determined by HPLC with fluorescence detection (excitation at 290 nm and emission at 330 nm), following the IUPAC Standard Method 2.432 (IUPAC, 1992). The column was a LiChrosorb Si 60 packed with silica (5 μm particle size) (Merck, Darmstadt, Germany). Sample solutions of 50 mL·min⁻¹ were used and the mobile phase was n-hexane/isopropanol (99:1, v/v), with a flow rate of 1 mL·min⁻¹.

3. RESULTS AND DISCUSSION

Table 1 summarizes the fatty acid composition as a reference of the major compounds for these palm species. Triacylglycerols and fatty acid compositions have been detailed previously (Santos et al., 2013). As can be observed, monounsaturated fatty acids were the most abundant in all the species and, consequently, they could be considered healthy oils which give protection against cardiovascular diseases.

Table 2 shows the quantitative results for the minor glyceridic compounds other than TAG, i.e. dimeric TAG, oxidized monomeric TAG and diacylglycerols (DAG). Their concentration is indicative of oil quality as they are compounds originated by thermal polymerization, oxidation and hydrolysis, respectively. And consequently, the

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**Table 1**

<table>
<thead>
<tr>
<th>Fatty acid methyl esters (FAME)</th>
<th>Bacaba</th>
<th>Buriti</th>
<th>Inajá</th>
<th>Pupunha</th>
<th>Tucumã</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saturated FAME</td>
<td>30.6</td>
<td>22.4</td>
<td>42.0</td>
<td>41.3</td>
<td>26.0</td>
</tr>
<tr>
<td>Monounsaturated FAME</td>
<td>47.3</td>
<td>72.3</td>
<td>40.8</td>
<td>51.4</td>
<td>64.7</td>
</tr>
<tr>
<td>Polyunsaturated FAME</td>
<td>20.6</td>
<td>3.9</td>
<td>14.4</td>
<td>5.3</td>
<td>7.2</td>
</tr>
</tbody>
</table>

**Table 2**

<table>
<thead>
<tr>
<th>Group of compounds</th>
<th>Bacaba</th>
<th>Buriti</th>
<th>Inajá</th>
<th>Pupunha</th>
<th>Tucumã</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dimeric TAG</td>
<td>0.2 ± 0.0</td>
<td>0.1 ± 0.0</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Oxidized monomeric TAG</td>
<td>1.1 ± 0.2</td>
<td>0.7 ± 0.1</td>
<td>1.2 ± 0.1</td>
<td>0.8 ± 0.2</td>
<td>1.2 ± 0.1</td>
</tr>
<tr>
<td>Diacylglycerols</td>
<td>2.9 ± 0.1</td>
<td>1.5 ± 0.1</td>
<td>2.0 ± 0.0</td>
<td>1.6 ± 0.2</td>
<td>2.8 ± 0.0</td>
</tr>
<tr>
<td>Fatty acids*</td>
<td>1.0 ± 0.1</td>
<td>0.9 ± 0.0</td>
<td>0.5 ± 0.0</td>
<td>1.3 ± 0.1</td>
<td>1.2 ± 0.0</td>
</tr>
<tr>
<td>Total (mg·kg⁻¹)</td>
<td>5.0 ± 0.1</td>
<td>3.3 ± 0.1</td>
<td>3.7 ± 0.1</td>
<td>3.6 ± 0.5</td>
<td>5.2 ± 0.1</td>
</tr>
</tbody>
</table>

mean ± standard deviation (n = 3). TAG, triacylglycerols. *also including polar unsaponifiable matter.
content of these groups of compounds should be as low as possible. As can be observed, the major compounds in all the samples were DAG. They were present in the range of 1 to 3%, similar to those found in virgin olive oils and they are not only attributed to the enzymatic or chemical hydrolysis of TAG occurring before or during the oil extraction process but also to the incomplete biosynthesis of TAG (Vázquez-Roncero et al., 1965; Castellani et al., 2008). Oxidized monomeric TAG were around 1% in all the samples, which are normal levels in good quality oils, while the level of dimeric TAG was very low, as expected for crude oils due to the absence of high temperatures in their extraction (Ruiz-Méndez et al., 1997). Finally, the total values found, between 3.3 and 5.2%, were low and denote good quality oils (Lumley, 1988).

Table 3 shows unsaponifiable matter and the total concentrations of hydrocarbons and aliphatic alcohols. To our knowledge, there is no previous information on the content of these two groups of compounds, probably because the composition is very complex. As can be observed, the concentrations were highly variable for both groups. Thus, hydrocarbons ranged from 50 to 734 mg·kg⁻¹, and alcohols from 80 to 490 mg·kg⁻¹. The high concentration of hydrocarbons and alcohols in inajá oil stands out where octacosanol and triacontanol were the most abundant aliphatic alcohols in the samples. As for the unsaponifiable fraction, the contents vary from 0.8 to 1.8% and were similar to the values found in most edible vegetable oils.

The tocopherol composition is given in Table 4. As can be observed, the content is also highly variable, with the mean value ranging from 85 mg·kg⁻¹ for inajá oil to 1567 mg·kg⁻¹ for buriti oil, which seems to indicate that this oil has an promising potential as a dietary source due to its high vitamin E content. α-tocopherol was the major compound in all the oils and the presence of tocotrienols in significant amounts was only detected in inajá oil. Figure 1 shows the chromatographic profile of

![Figure 1](image)

**Figure 1** Gas chromatographic profiles of the tocopherols of oils from the mesocarp of fruits from Amazonian palm trees.

Table 3

<table>
<thead>
<tr>
<th>Group of compounds</th>
<th>Bacaba</th>
<th>Buriti</th>
<th>Inajá</th>
<th>Pupunha</th>
<th>Tucumá</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unsaponifiable matter</td>
<td>1.0 ± 0.1</td>
<td>1.3 ± 0.3</td>
<td>0.8 ± 0.1</td>
<td>1.3 ± 0.1</td>
<td>1.8 ± 0.2</td>
</tr>
<tr>
<td>Hydrocarbons</td>
<td>50 ± 0.5</td>
<td>145 ± 1.3</td>
<td>734 ± 126</td>
<td>44 ± 3.0</td>
<td>110 ± 0.4</td>
</tr>
<tr>
<td>Fatty alcohols</td>
<td>80 ± 14.7</td>
<td>149 ± 8.3</td>
<td>490 ± 54.4</td>
<td>202 ± 29.9</td>
<td>428 ± 75.1</td>
</tr>
</tbody>
</table>

mean ± standard deviation (n = 3).

Table 4

<table>
<thead>
<tr>
<th>Compound</th>
<th>Bacaba</th>
<th>Buriti</th>
<th>Inajá</th>
<th>Pupunha</th>
<th>Tucumá</th>
</tr>
</thead>
<tbody>
<tr>
<td>α-tocopherol</td>
<td>148 ± 41</td>
<td>1100 ± 198</td>
<td>26 ± 16</td>
<td>117 ± 18</td>
<td>480 ± 40</td>
</tr>
<tr>
<td>β-tocopherol</td>
<td>466 ± 26</td>
<td>3 ± 1</td>
<td>3 ± 2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>γ-tocopherol</td>
<td>18 ± 1</td>
<td>ND</td>
<td>ND</td>
<td></td>
<td></td>
</tr>
<tr>
<td>δ-tocopherol</td>
<td>37 ± 1</td>
<td>ND</td>
<td>ND</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total (mg·kg⁻¹)</td>
<td>148 ± 41</td>
<td>1567 ± 205</td>
<td>85 ± 16</td>
<td>117 ± 18</td>
<td>483 ± 40</td>
</tr>
</tbody>
</table>

mean ± standard deviation (n = 3); tr, traces; ND, not detected; * also containing 26 and 24 mg·kg⁻¹ of α- and γ-tocotrienols, respectively.
tocopherols and tocotrienols for the five samples. Previous results reported concentrations of a similar order for buriti oil (Silva et al., 2009; Costa et al., 2010) and pupunha (Bereau et al. 2003), lower concentrations for buriti, inajá and tucumã oils (Rodrigues et al., 2010) and higher concentrations for bacaba oil (Montúfar et al., 2010).

Table 5 shows the percentages of phytosterol species and the total concentrations in the oils. As can be observed, β-sitosterol was the major compound in the group in all the oils ranging from 65 and 83% followed by campesterol (6.6-18.8%) and stigmasterol (4.2-16.8%). The total sterol concentrations were variable, ranging from around 1000 mg·kg⁻¹ in bacaba oil to more than 4000 mg·kg⁻¹ in pupunha oil. Similar contents have been reported for burití (Costa et al., 2010), tucumã (Bereau et al., 2003; Costa et al., 2003; Costa et al., 2003; Costa et al., 2003) and inajá oils (Bereau et al., 2003). Given the interest of phytosterols as functional ingredients, buriti, pupunha and tucumã oils would be good edible oils from a nutritional point of view.

In summary, from the analysis of the oils from the Amazonian fruits it can be deduced that they have a great potential as edible vegetable oils. Apart from the high content of monounsaturated fatty acid, bioactive compounds may give them an added value as healthy oils. In particular, concerning the two groups of bioactive compounds analyzed, i.e. tocopherols and sterols, pupunha and buriti oils stand out. Pupunha oil is an excellent oil due to its high content of phytosterols while buriti oil has also a high vitamin E content.

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REFERENCES


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