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The ripe pulp of *Mangifera indica* L.: A rich source of phytosterols and other lipophilic phytochemicals

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ABSTRACT

The chemical composition of the lipophilic extracts of the ripe pulp of mangoes from twelve cultivars of *Mangifera indica* L. from Madeira Island was investigated by gas chromatography–mass spectrometry (GC–MS) for the first time. The ripe pulp of these mango cultivars showed analogous amounts of lipophilic extractives, as well as similar qualitative chemical compositions. The predominant compounds were free and glycosylated sterols and fatty acids, representing 44.8–70.7% and 22.6–41.9%, respectively, of the total amount of lipophilic components. Smaller amounts of long chain aliphatic alcohols and α-tocopherol were also identified. These data indicate that the investigated mango cultivars are a rich source of valuable phytochemicals, contributing to the intake of at least 9.5–38.2 mg of phytosterols (free and glycosylated) and 0.7–3.9 mg of fatty acids ($\omega-3$ and $\omega-6$) per 100 g of fresh mango, with recognizable beneficial effects on human nutrition and health.

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1. Introduction

The mango fruit, one of the most important tropical fruits in the world, enjoys the status of "the king of fruits" as a result of its unique flavor, fragrance and appearance (Singh, Singh, Sane, & Nath, 2013). The *Mangifera indica* L. species, belonging to the *Mangifera* genus, Anacardiaceae family and Sapindales order, is the most important edible species and its fruit shows a pronounced diversity in size, shape, color, flavor, seed size, and chemical composition (Stafford, 1983), depending on the cultivar (Othman & Mbogo, 2009), edaphoclimatic conditions (Léchaudel & Joas, 2006) and postharvest storage (Nunes, Emond, Brecht, Dea, & Proulx, 2007). 'Kent', 'Tommy Atkins', 'Haden', and 'Keitt' are the most popular export mango cultivars (Sauco, 2004).

M. indica L. species, native from Southeast Asia, are widely cultivated at both tropical and subtropical latitudes (Kaira, Tandon, & Singh, 1995; Ueda, Sasaki, Utsunomiya, Inaba, & Shimabayashi, 2000), over a harvested area of approximately 5 million ha in 94 countries (FAOSTAT, 2011). Over the last decade, mango cultivated area increased by 41.8%, and is expected to increase even more due to the growing consumption of fresh fruit and processed products. The annual world production accounted in 2011 for ca. 38 million tonnes, with India as the major producer (15 million tonnes), Mexico and India as the major exporters (275 and 260 thousand tonnes in 2010, respectively), while the European Union and United States of America were the main importers (369 and 320 thousand tonnes in 2010, respectively) of mango fruits (FAOSTAT, 2011). In Madeira Island, a Portuguese Mediterranean subtropical region with adequate climate to the growth cycle of tropical and subtropical fruits (*e.g.* banana (*Musa acuminata* Colla), avocado (*Persea americana* Mill.) and annona (*Annona cherimola* Mill.)), mango was introduced after the second half of the eighteenth century. Nevertheless, it was only in the twentieth century that the plantations of this fruit attained a commercial dimension and became an important crop for the island's economy.

The mango fruit, with an average annual worldwide per capita consumption of 3.42 kg (CBI Report, 2011), is one of the nutritionally richest fruits, providing about 64–86 cal per 100 g (Rathore, Tariq, Shehla, & Soomro, 2007), with 32–200 mg per 100 g of vitamin C (Akinyele & Keshinro, 1980). When regularly consumed, this fruit can be a valuable dietary source of many phytochemicals (Haard & Chism, 1996) that provide several human health benefits (Singh et al., 2013). Several studies have addressed the phytochemical composition of diverse mango plant tissues, namely leaves, stem bark, peel, pulp and kernel, given their medicinal applications (Masibo & He, 2009). For example, Garido et al. (2001) found different polyphenols, steroids, flavonoids and tannins with antinociceptive and anti-inflammatory actions in *M. indica* L. stem bark extracts that could be used to improve the life quality in patients suffering from high stress levels. The mango seeds of *M. indica* L., with a broad antimicrobial spectrum (Kabuki

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et al., 2000) and a significant anti-diarrheal activity (Sairam et al., 2003), showed potential as food additives for extending the shelf-life of a variety of food products. Nevertheless, there is still a lack of detailed studies on the phytochemical composition of mango pulp, particularly on the lipophilic components, with only a study reporting the sterol composition of mango from China (Han, Yang, & Feng, 2008).

The present study is part of a global project concerning sub-tropical fruits' nutritional and functional values, aiming to add value to the fruits and by-products, by promoting an increase in consumption and market competitiveness of this sector. To the best of our knowledge, no studies about the chemical composition of lipophilic extracts of ripe mango pulp of 'Tommy Atkins', 'Rosa', 'OTT', 'Anderson', 'Rubro Brasil', 'Osteen', 'Tolbert', 'Irwin', 'Gleen', 'Gomera I', 'Gomera II' and 'Gomera III' cultivars, growing in Madeira Island, have been published until now. In this context, this work aims at establishing the lipophilic extractives profile (fatty acids, sterols, long chain aliphatic alcohols and other compounds) of mango pulp by gas chromatography–mass spectrometry (GC–MS) analysis and to link them with the potential health benefits of these mango pulp cultivars growing under the Mediterranean subtropical climate conditions of that island. From a commercial point of view, the evaluation of the selected mango cultivars could provide information to farmers about the cultivars with a higher commercial added-value, in order to compete favorably for local and export markets.

2. Materials and methods

2.1. Chemicals

Dichloromethane (99% purity), pyridine (99% purity), trimethylchlorosilane (99% purity), *N,O*-bis(trimethylsilyl)trifluoroacetamide (99% purity), stigmasterol (95% purity), octadecanoic acid (99% purity), nona decanol (99% purity), ferulic acid (99% purity) and tetracosane (99% purity) were supplied by Sigma Chemicals Co. (Madrid, Spain).

2.2. Sample preparation and physicochemical parameters

Mango fruits (*M. indica* L.) without evidence of physical or pathological injuries were selected from Centro de Fruticultura Subtropical do Funchal, Madeira Island, Portugal (32° 38′ 52″ N, 16° 57′ 44″ W). The mature green fruits from 'Tommy Atkins', 'Rosa', 'OTT', 'Anderson', 'Rubro Brasil', 'Osteen', 'Tolbert', 'Irwin', 'Gleen', 'Gomera I', 'Gomera II' and 'Gomera III' cultivars were hand harvested and then left to reach full ripeness at room temperature (20–23 °C). Fruit firmness was determined after removing the skin on two opposite sides in the middle of each fruit using a pressure-testing instrument (Model FT 327) fitted with an 11.3 mm cylindrical plunger. The force required to penetrate into the flesh was expressed in Newtons (N). The fruits were immediately peeled (peel was fully discarded), sliced, quick-frozen in liquid nitrogen and lyophilized. Fresh slices of each sample were used to measure fruit water content through a Gibertini-Eurotherm balance, at 105 °C and Brix using a digital Brix refractometer from ATAGO. The frozen samples were lyophilized and milled to pass through a 40–60 mesh sieve and stored (humidity c.a. 5%) in dark at −18 °C for further analyses.

2.3. Extraction

Three powdered samples (20 g) of each cultivar were Soxhlet extracted with dichloromethane for 6 h. The solvent was evaporated to dryness, the lipophilic extracts were weighted and the results were expressed in percent of dry weight (% dw). Dichloromethane was selected as a fairly specific solvent for lipophilic extractives isolation for analytical purposes.

2.4. GC–MS analysis

Before GC–MS analysis, two aliquots of each dried extract (20 mg each) and an accurate amount of internal standard (tetracosane, 0.50 mg) were dissolved in 250 μL of pyridine. The compounds containing hydroxyl and carboxyl groups were converted into trimethylsilyl (TMS) ethers and esters, respectively, by adding 250 μL of *N,O*-bis(trimethylsilyl)trifluoroacetamide and 50 μL of trimethylchlorosilane, standing the mixture at 70 °C for 30 min (Freire, Silvestre, Neto, & Cavaleiro, 2002). The derivatized extracts were analyzed by GC–MS following previously described methodologies (Freire et al., 2002; Oliveira, Freire, Silvestre, & Cordeiro, 2008) on a TRACE Gas Chromatograph 2000 Series, equipped with a Thermo Scientific DSQII single-quadrupole mass spectrometer and a DB-1 J&W capillary column (30 m × 0.32 mm inner diameter, 0.25 μm film thickness). The chromatographic conditions were as follows: initial temperature, 80 °C for 5 min; temperature gradient, 4 °C min^{-1}; final temperature, 260 °C; temperature gradient, 2 °C min^{-1}; final temperature, 285 °C for 8 min; injector temperature, 250 °C; transfer-line temperature, 290 °C; and split ratio, 1:33.

To check the presence of lower volatility esterified structures, samples were also analyzed with a DB-1 J&W capillary column (15 m × 0.32 mm inner diameter, 0.25 μm film thickness); the chromatographic conditions were as follows: initial temperature, 100 °C for 3 min; temperature gradient, 5 °C min^{-1}; final temperature, 340 °C for 12 min; injector temperature, 290 °C; transfer-line temperature, 290 °C; and split ratio, 1:33.

Compounds were identified as TMS derivatives by comparing their mass spectra with the GC–MS spectral library (Wiley-NIST Mass Spectral Library 1999) and their retention times with published data obtained under the described experimental conditions (Oliveira et al., 2006, 2008), and also by comparing their fragmentation profiles with published data or by injection of standards.

For semi-quantitative analysis, GC–MS was calibrated with pure reference compounds, representative of the major lipophilic extractive families (stigmasterol, octadecanoic acid, ferulic acid and nonadecan-1-ol) relative to tetracosane. The respective response factors were calculated as an average of six GC–MS runs. For tocopherol the response factor of stigmasterol was used. Each aliquot was injected in triplicate. The presented results are the average of the concordant values obtained for the six aliquots (less than 5% variation between injections of the same aliquot and between aliquots of the same mango cultivar extracts).

3. Results and discussion

The physicochemical characteristics, namely weight, length, pulp/seed ratio, water content, pulp firmness, total soluble solids (TSS) and pH, of the twelve mango cultivars investigated in this study, are given in Table 1. All the values obtained are comparable to values previously reported for other mango varieties/cultivars (Charoensiri, Kongkachuichai, Suknicom, & Sungpuag, 2009; Liu et al., 2013; Pleguezuelo, Zuazo, Fernández, & Tarifa, 2012). Mangoes have high water content, with the cultivar 'Tolbert' and 'Gomera II' presenting the highest (86.4%) and the lowest (75.1%) values, respectively. Firmness was evaluated when the mangoes reached the mature stage, and 'Rosa' presented the highest pulp firmness with 1.52 N. Total soluble solids (TSS) determination expressed as °Brix, is usually used as an estimation of the sugar content of fruit. Generally, the TSS in mangoes range from 7.0 to 17.4 °Brix, depending on the variety, the production place and maturity stage (Lucena, Assis, Alves, Silva, & Enéas, 2007), and good quality mango for fresh consumption should have a TSS between 13 and 15 °Brix (Rovira & Alvarez, 1990). In the present study, the lowest °Brix was observed for 'OTT' (11.0 °Brix), the highest one for 'Gomera III' (19.3 °Brix), and except for 'OTT', all cultivar showed TSS above 12 °Brix. Finally, the pH of the studied samples varied between 5.02 for 'Anderson' and 3.41 for 'OTT'. The differences among them can be attributed to the different

Table 1
Physicochemical characteristics of the twelve mango cultivars.[a]

Cultivar	Weight (g)	Length (mm)	Pulp/seed (%)	Moisture (%)	Firmness (N)	TSS (°Brix)	pH
'Tommy Atkins'	246.8 ± 46.9	120.3 ± 20.5	95.0 ± 1.4	81.2 ± 1.6	0.96 ± 0.19	14.4 ± 0.9	3.88 ± 0.05
'Rosa'	236.0 ± 32.9	132.5 ± 8.0	90.7 ± 1.4	83.7 ± 1.2	1.52 ± 0.35	12.0 ± 0.8	3.52 ± 0.09
'OTT'	301.7 ± 32.4	141.3 ± 8.1	95.3 ± 1.4	83.6 ± 1.6	0.72 ± 0.12	11.0 ± 1.0	3.41 ± 0.18
'Anderson'	299.0 ± 60.3	104.7 ± 11.9	92.2 ± 1.3	78.1 ± 1.0	0.74 ± 0.22	18.4 ± 0.3	5.02 ± 0.20
'Rubro Brasil'	276.1 ± 34.2	162.3 ± 6.7	91.8 ± 1.1	85.8 ± 1.6	0.78 ± 0.19	14.0 ± 1.2	4.75 ± 0.17
'Osteen'	425.9 ± 77.0	158.8 ± 9.6	97.1 ± 0.9	82.8 ± 1.7	0.92 ± 0.18	15.0 ± 1.3	4.64 ± 0.06
'Tolbert'	340.8 ± 36.2	134.2 ± 9.6	94.5 ± 0.4	86.4 ± 2.1	1.28 ± 0.25	14.5 ± 0.9	3.97 ± 0.05
'Irwin'	373.2 ± 62.6	148.8 ± 10.2	94.3 ± 0.8	82.1 ± 1.4	0.86 ± 0.19	12.3 ± 1.5	4.22 ± 0.07
'Gleen'	218.2 ± 20.4	86.0 ± 12.0	96.4 ± 3.3	79.2 ± 0.9	1.12 ± 0.08	18.2 ± 0.3	4.64 ± 0.12
'Gomera I'	168.7 ± 35.1	73.5 ± 8.3	90.2 ± 1.6	78.7 ± 0.2	0.98 ± 0.12	17.8 ± 2.4	4.73 ± 0.04
'Gomera II'	130.5 ± 19.9	105.0 ± 7.3	86.2 ± 1.1	75.1 ± 1.1	0.75 ± 0.15	16.0 ± 1.6	4.38 ± 0.11
'Gomera III'	130.2 ± 24.2	71.5 ± 6.2	87.9 ± 2.1	77.5 ± 0.8	0.93 ± 0.19	19.3 ± 0.4	4.29 ± 0.20

[a] Values are expressed as mean ± SD (n = 3).

cultivars, edaphoclimatic conditions and fruit maturity. In fact, it is known that during mango ripening process the acidity decreased and pH increased, due to the cell metabolization of volatile organic acids and non-volatile constituents (Tucker, 1993).

The lipophilic extractives yields from the ripe pulp of mango cultivars were quite similar, with values ranging from 0.56 to 1.34% of dry material for 'Gomera I' and 'Tommy Atkins', respectively, as shown in Fig. 1. These lipophilic extractives contents are similar to those found in other tropical fruits *e.g.* in the unripe pulp of banana (Oliveira et al., 2008).

The composition of the lipophilic extracts of the ripe mango pulp was analyzed in detail by GC–MS, and the identities and abundances of the identified compounds are summarized in Table 2. The predominant lipophilic compounds were a series of free sterols that accounted for nearly 32.8–54.2% of all identified compounds. Free fatty acids (C12–C25) were also very abundant accounting for 22.6–41.9% of all lipophilic compounds. Additionally, minor amounts of long chain aliphatic alcohols (C14–C30) were also identified in the extracts. The relative abundance of the identified compounds and their families differs somewhat between cultivars, as illustrated in Table 2 and Fig. 2. The presence of these classes of compounds was already reported in other tissues (*e.g.* stem bark, peel, kernel) of *M. indica* L. (Gaydou, 1984; Masibo & He, 2009; Muchiri, Mahungu, & Gituanja, 2012).

Free sterols are the most abundant class of lipophilic compounds present in the ripe mango pulps, accounting for 343 and 1030 mg kg^{-1} of dry material for 'Gomera I' and 'OTT', respectively (Table 2). β-Sitosterol is definitely the major component of this family in all pulp samples, representing between 51.0 ('Irwin') and 69.1% ('Gomera I') of total sterol contents and between 20.1 ('Osteen') and 36.4% ('Rubro Brasil') of the total lipophilic extractives (Table 2). Other identified free sterols include campesterol (52–174 mg kg^{-1} of dry material), fucosterol (23–146mg kg^{-1} of dry material), stigmasterol (24–82 mg kg^{-1} of dry material), 24-methylenecholesterol (1–50 mg kg^{-1} of dry material) and 24-methylenecycloartanol (7–108 mg kg^{-1} of dry material). Generally, the human intake of phytosterols varies from about 145 to 405 mg per day (Sánchez-Moreno, De Pascual-Teresa, De Ancos, & Cano, 2012), and, although fruits in general are not considered good sources of sterols, 'Gomera II' (*e.g.*) can contribute to the intake of ca. 23.4 mg of free phytosterols per 100 g of fresh mango. This value is in agreement with the average value of 24.4 mg per 100 g of edible portion of mango reported by Han et al. (2008). Hence, the ripe mango pulps from these twelve cultivars can contribute to the intake of natural phytosterols in the human diets, which appear to be a practical and safe option for reducing cholesterol levels in the population (Piironen, Lindsay, Miettinen, Toivo, & Lampi, 2000; Quílez, García-Lorda, & Salas-Salvadó, 2003).

Long chain fatty acids represent about 22.6–41.9% of the lipophilic components of ripe mango pulps. Whereas the cultivars 'Tommy Atkins' and 'Osteen' presented the higher amounts of fatty acids (940 and 1108 mg kg^{-1} of dry material, respectively), 'Gomera I' presented the lowest one (353 mg kg^{-1} of dry material). The identified fatty acids ranged from dodecanoic to pentacosanoic acids, including five unsaturated structures (C16 and C18), one diacid (nonadioic acid) and one ω-hydroxy fatty acid (Table 2). Hexadecanoic acid is the most abundant

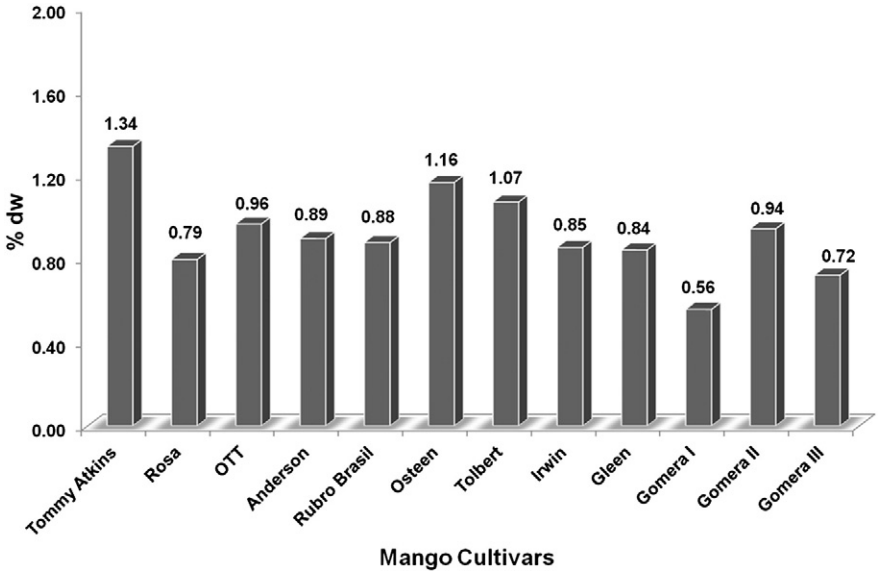

Fig 1. Lipophilic extractives yields in (%) of dry weight for each ripe pulp from the studied mango cultivars.

Table 2
Compounds identified in the lipophilic extracts of ripe pulp from mango cultivars expressed in mg kg^{-1} of dry material.[a]

Compound	'Tommy Atkins'	'Rosa'	'OTT'	'Anderson'	'Rubro Brasil'	'Osteen'	'Tolbert'	'Irwin'	'Gleen'	'Gomera I'	'Gomera II'	'Gomera III'
Fatty acids	940	704	519	360	504	1108	565	801	729	353	607	587
Saturated	324	476	270	175	225	559	354	458	390	189	391	351
Dodecanoic acid	4	51	2	3	2	40	18	12	32	11	45	37
Tetradecanoic acid	26	33	21	21	39	81	29	109	32	12	37	27
Pentadecanoic acid	2	4	3	1	4	5	2	4	4	2	2	3
Hexadecanoic acid	228	203	186	117	160	311	160	294	216	107	183	138
Heptadecanoic acid	8	13	8	7	4	16	14	5	11	8	8	13
Octadecanoic acid	28	125	33	15	7	70	81	15	66	34	72	102
Eicosanoic acid	2	12	5	2	2	11	18	4	9	4	10	12
Heneicosanoic acid	n.d.	4	1	n.d.	n.d.	1	1	n.d.	1	1	1	1
Docosanoic acid	8	16	5	3	2	15	16	3	11	7	22	11
Tetracosanoic acid	10	9	5	5	4	5	9	7	7	3	8	5
Pentacosanoic acid	8	5	2	2	2	3	6	4	2	1	3	1
Unsaturated	612	225	245	180	276	546	209	340	331	158	214	228
Hexadec-9-enoic acid	91	40	50	19	34	110	41	74	71	21	26	33
Heptadec-9-enoic acid	16	36	25	30	20	35	31	19	52	34	20	48
Octadeca-9,12-dienoic acid	48	13	57	11	13	28	13	10	11	21	27	21
Octadeca-9,12,15-trienoic acid	131	33	30	48	115	198	34	46	75	29	47	33
cis-Octadec-9-enoic acid	205	74	49	39	62	84	63	127	89	38	56	54
trans-Octadec-9-enoic acid	122	30	35	34	31	91	27	64	33	15	38	40
Diacids	1	1	2	1	1	1	1	2	1	1	1	n.d.
Nonadioic acid	1	1	2	1	1	1	1	2	1	1	1	n.d.
ω-Hydroxy acids	3	3	2	4	3	3	1	1	7	5	2	7
22-Hydroxydocosanoic acid	3	3	2	4	3	3	1	1	7	5	2	7
Long chain aliphatic alcohols	104	86	68	67	52	107	69	82	99	49	58	79
Tetradecan-1-ol	1	2	1	1	2	2	2	1	3	1	1	1
Hexadecan-1-ol	20	22	22	15	14	26	18	16	41	23	14	28
Octadecan-1-ol	7	11	10	7	7	13	9	8	11	7	7	11
Docosan-1-ol	2	2	n.d.	1	n.d.	1	n.d.	1	3	2	1	4
Octacosan-1-ol	27	14	13	19	16	44	15	26	18	6	16	18
Triacontan-1-ol	47	35	22	24	13	21	25	30	22	9	19	18
Sterols	947	1023	1030	607	852	872	856	819	600	343	941	655
24-Methylenecholesterol	37	19	16	23	13	32	50	45	13	1	15	12
Fucosterol	80	54	54	114	91	146	71	137	44	23	54	46
β-Sitosterol	571	618	692	335	573	531	480	418	369	237	607	413
Campesterol	149	165	174	89	106	113	124	120	110	52	131	110
Stigmasterol	68	59	81	27	46	32	82	51	41	24	80	47
24-Methylenecycloartanol	41	108	12	18	22	19	49	49	23	7	54	27
Steryl glucosides	201	575	595	98	96	421	506	167	220	102	593	188
Campesteryl 3β-D-glucopyranoside	21	47	43	9	6	38	53	19	27	8	44	18
Stigmasteryl 3β-D-glucopyranoside	4	13	24	1	3	6	21	3	6	4	19	7
Sitosteryl 3β-D-glucopyranoside	176	515	528	88	87	377	432	145	187	90	530	163
Others	195	133	86	87	69	135	90	150	183	67	118	94
1-Monohexadecenoin	15	8	9	4	4	9	8	7	16	8	5	9
1-Monohexadecanoin	70	13	33	12	14	39	22	10	55	28	13	21
1-Monooctodecenoin	45	19	11	9	22	17	19	37	27	13	12	12
trans-Ferulic acid	n.d.	2	n.d.	1	n.d.	1	n.d.	1	1	n.d.	n.d.	2
Tricosane	1	1	1	6	2	3	3	1	10	6	3	8
α-Tocopherol	64	90	32	55	27	66	38	94	74	12	85	42

[a] Results are the average of the concordant values (less than 5% variation between injections) obtained for the two aliquots of each sample injected in triplicate. n.d., not detectable.

saturated fatty acid, with the highest content observed in the cultivar 'Osteen' (311 mg kg^{-1} of dry material) and the lowest in the 'Gomera I' (107 mg kg^{-1} of dry material). Unsaturated fatty acids were also present in high amounts (158–612 mg kg^{-1} of dry material), with octadec-9-enoic acid as the major compound of this group, with the highest content observed in the cultivar 'Tommy Atkins' (327 mg kg^{-1} of dry material) and the lower in the 'Gomera I' (53 mg kg^{-1} of dry material), followed by octadeca-9,12,15-trienoic acid (an $\omega-3$ fatty acid) with 29–198 mg kg^{-1} of dry pulp, hexadec-9-enoic acid with 19–110 mg kg^{-1} of dry pulp and octadeca-9,12-dienoic acid (an $\omega-6$ fatty acid) with 10–57 mg kg^{-1} of dry pulp. Minor amounts of 22-hydroxydocosanoic (1–7 mg kg^{-1} of dry pulp) and nonadioic acids (1–2 mg kg^{-1} of dry pulp) were also found in all twelve extracts of ripe mango pulps.

Contrary to saturated and monounsaturated fatty acids that are non-essential dietary lipids, polyunsaturated fatty acids, like octadeca-9,12-dienoic ($\omega-6$) and octadeca-9,12,15-trienoic ($\omega-3$) acids, are essential nutrients that must be obtained from the diet because they are not synthesized in the human body (Sánchez-Moreno et al., 2012). Hence, these mango pulps can also contribute to the intake of the above $\omega-3$ and $\omega-6$ fatty acids, with 'Osteen' contributing to the higher intake of octadeca-9,12,15-trienoic acid with ca. 3.4 mg per 100 g of fresh mango, and 'Tommy Atkins' and 'Osteen' to the higher intake of octadeca-9,12-dienoic acid with ca. 0.9 mg per 100 g of fresh mango. The role of fatty acids in the human health, especially $\omega-3$ and $\omega-6$ fatty acids, is mainly associated with the prevention, delay, or treatment of chronic and acute diseases, such as cancer, cardiovascular diseases, osteoporosis, and immune disorders (Chen, McClements, & Decker, 2013; Sánchez-Moreno et al., 2012; Simopoulos, 1999, 2008).

Long-chain aliphatic alcohols (LCAA) were also detected in the ripe mango pulps (49–107 mg kg^{-1} of dry material), representing only a small fraction (2.5–5.5%) of the total amount of lipophilic extractives (Table 2). The most abundant LCAA found are triacontan-1-ol, octacosan-1-ol and hexadecan-1-ol, with 9–47, 6–44 and 14–41 mg kg^{-1} of dry material, respectively. Reports on the role of LCAA in human health suggest a decrease of the low-density lipoprotein (LDL) cholesterol and an increase of the high-density lipoprotein (HDL) cholesterol (Hargrove, Greenspan, & Hartle, 2004), with their regular consumption.

Finally, other compounds like monoglycerides, α-tocopherol, *trans*-ferulic acid and tricosane were also detected in smaller amounts (Table 2). Only three monoglycerides were identified (25–131 mg kg^{-1}

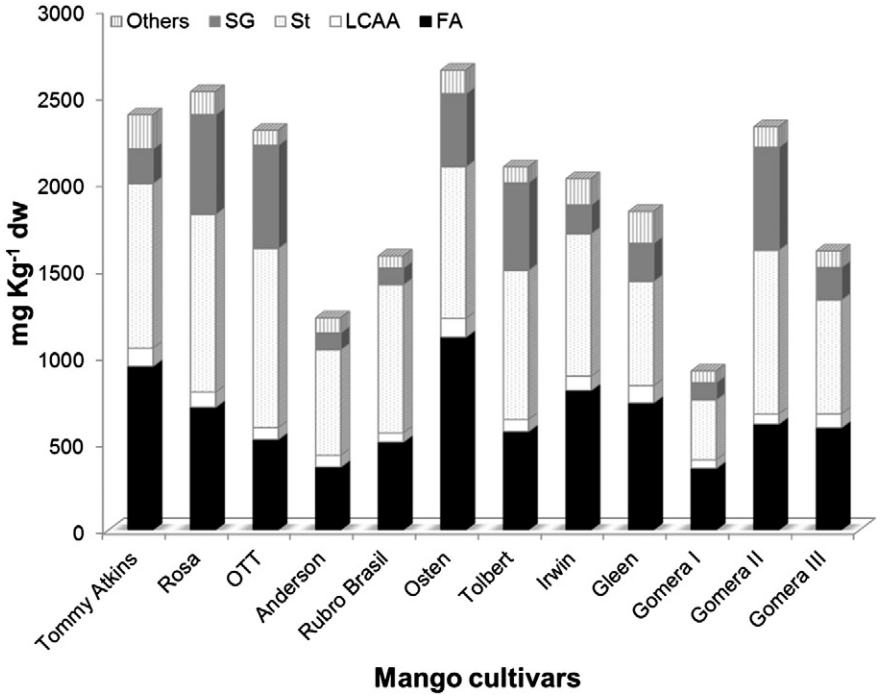

Fig. 2. Major families of lipophilic components identified in the mango pulp extracts. SG, steryl glucosides; St, sterols; LCAA, long-chain aliphatic alcohols; FA, fatty acids.

of dry material, Table 2), from which one was saturated (C16) and two were unsaturated (C16 and C18), with the exclusive presence of the isomer in position 1. α-Tocopherol, the most bioactive form of vitamin E, was the only tocopherol detected in all the studied mango pulps, accounting for 12–94 mg kg^{-1} of dry material, with the extreme values recorded for 'Gomera I' and 'Irwin', respectively (Table 2). In terms of edible portion, the studied mango pulps presented an α-tocopherol content of about 0.3–2.1 mg per 100 g of fresh mango, which is in agreement with previously published data for other mango varieties (Charoensiri et al., 2009). Albeit their small α-tocopherol content compared to vegetable oils, nuts and grains (Tiwari & Cummins, 2013), the consumption of ripe mango together with other plant-derived foods represents an important source of vitamin E (Eitenmiller & Lee, 2004), which has been associated with the prevention of cardiovascular diseases, cancer, inflammatory diseases, neurological disorders, cataract and age-related macular degeneration, as well as to the maintenance of the immune system (Bramley et al., 2000).

The lipophilic extracts of the twelve mango pulps were also analyzed by GC–MS with a short length (15 m) column, in order to verify the presence of lower volatility esterified structures, such as steryl esters and steryl glucosides. As an example, Fig. 3 shows the typical GC–MS chromatogram of the derivatized lipophilic extract of 'Rosa' cultivar. Here, steryl glucosides, namely campesteryl 3β-D-glucopyranoside, stigmasteryl 3β-D-glucopyranoside and sitosteryl 3β-D-glucopyranoside (42.00, 42.22 and 42.72 min, respectively) were detected in significant amounts, representing ca. 6.1–25.9% of the lipophilic components of ripe mango

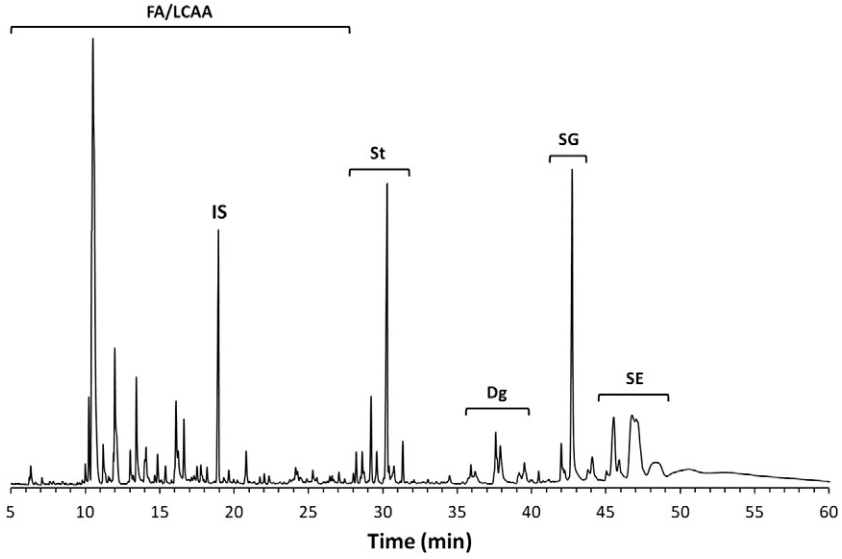

Fig. 3. GC–MS chromatogram of the derivatized lipophilic extract from the ripe pulp of 'Rosa' cultivar, obtained by a DB-1 15 m column. FA, fatty acids; LCAA, long-chain aliphatic alcohols; St, sterols; Dg, diacylglycerols; SG, steryl glucosides; SE, steryl esters; IS, internal standard.

pulps. Sitosteryl 3β-D-glucopyranoside is the most abundant steryl glucoside accounting for 87–530 mg kg^{-1} of dry material, with the extreme values recorded for 'Rubro Brasil' and 'Gomera II', respectively (Table 2). On the other hand, steryl esters were also found to be considerably abundant, as illustrated in Fig. 3. Nevertheless, their content was not assessed since they were detected as broad peaks corresponding to complex mixtures of compounds. Reports on the role of these bioactive compounds (together with free sterols) on human health suggest an important contribution to the improvement of lipid metabolism and immune function (Moreau, Whitaker, & Hicks, 2002).

4. Conclusions

The present work represents the first study on the lipophilic components from the ripe pulp of twelve mango cultivars of the *M. indica* L. species, namely 'Tommy Atkins', 'Rosa', 'OTT', 'Anderson', 'Rubro Brasil', 'Osteen', 'Tolbert', 'Irwin', 'Gleen', 'Gomera I', 'Gomera II' and 'Gomera III', cultivated in Madeira Island. The major groups of compounds identified in the lipophilic fraction of the extractives consisted mainly of sterols and fatty acids, followed by long-chain aliphatic alcohols. Considerable amounts of steryl glucosides and steryl esters were also detected. Among all the identified compounds, the presence of phytosterols (and derivatives) and $\omega-3$ and $\omega-6$ fatty acids with well-established beneficial nutritional and health effects, contributes to the valorization of these mango cultivars as sources of valuable phytochemicals.

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