Characterization of Yam Bean (*Pachyrhizus* spp.) Seeds as Potential Sources of High Palmitic Acid Oil

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ABSTRACT: Seeds from 22 accessions of the yam bean species *Pachyrhizus ahipa* (14 accessions), *P. erosus* (5), and *P. tuberosus* (3) were investigated for oil and protein contents, fatty acid composition of the seed oil, and the total tocopherol content and composition. Plants from the accessions were grown under greenhouse conditions during one (*P. erosus* and *P. tuberosus*) or two years (*P. ahipa*). The pattern of the investigated seed quality traits was very similar in the three species. Yam bean seeds were characterized by high oil (from about 20 to 28% in one environment) and protein contents (from about 23 to 34%). Seed oil contained high concentrations of palmitic (from about 25 to 30% of the total fatty acids), oleic (21 to 29%), and linoleic acids (35 to 40%). Levels of linolenic acid were very low, from about 1.0 to 2.5%. Total tocopherol content was relatively low in *P. erosus* (from 249 to 585 mg kg^{-1} oil) and *P. tuberosus* (from 260 to 312 mg kg^{-1} oil) compared with the levels found in *P. ahipa* grown under identical conditions (508 to 858 mg kg^{-1} oil). In all the samples, γ-tocopherol was predominant, accounting for more than 90% of the total tocopherol content. The combination of high oil and protein contents, together with high palmitic acid, low linolenic acid, and high γ-tocopherol concentration, makes these crops an interesting alternative as sources of high palmitic acid oil for the food industry.

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KEY WORDS: Fatty acids, legumes, *P. ahipa*, *P. erosus*, palmitic acid, protein, *P. tuberosus*, tocopherols, yam bean.

The neotropical genus *Pachyrhizus* Richard ex DC. (yam bean) is a close relative to the soybean and phaseolus bean (1). The genus comprises three cultivated species with different ecogeographical origins: *P. ahipa*, from the highland tropics of Bolivia and northern Argentina; *P. erosus,* from the semiarid tropics of Central America; and *P. tuberous*, from the tropical lowlands of both sides of the Andean mountain range. *Pachyrhizus erosus* was introduced to the Philippines during the 16th century, and it is nowadays a popular crop in many countries of Asia, as well as West Africa. The yam beans are exclusively used as a tuber crop and the crisp and fruity tubers are consumed raw as a refreshment (2,3).

The use of yam bean seeds is limited by the presence of high rotenone content (about 1% seed weight). Nevertheless, the 1000-seed weight is high (from 180 to 230 g) and seed yields of up to 600 kg/ha have been reported in addition to a tuber yield of 7000 to 10,000 kg/ha (4). Studies on yam bean seed composition are scarce and concentrated exclusively on *P. erosus*. For this species, Duke (4) reported an approximate composition of 6.7% moisture, 26.2% protein, 27.3% oil, 20.0% carbohydrates, 7.0% fiber, and 3.6% ash. Santos *et al.* (5) reported similar values, as well as the amino acid profile of the flour, which was deficient in sulfur amino acids, as in other legumes. Potential uses of yam bean seed have mainly focused on the extraction of rotenone as a source of a natural insecticide (4). Additionally, Santos *et al.* (5) pointed out the potential value of yam bean meal for human consumption after the elimination of rotenone.

According to Duke (4) and Santos *et al*. (5), the seeds of *P. erosus* contain one of the highest oil contents reported to date in a cultivated legume species, second only to groundnut (*Arachis hypogaea*, 38 to 50% seed weight) (6). The fatty acid composition of the seed oil in yam beans has hardly been studied. Broadbent and Shone (7) reported the fatty acid composition of one accession of *P. erosus*, which contained 26.7% palmitic, 5.7% stearic, 33.5% oleic, and 34.2% linoleic acids. The fatty acid composition of other species has not been evaluated. Similarly, the total tocopherol content and composition of yam bean seeds has not been studied to date.

The objective of the present study was to evaluate the oil and protein contents, the fatty acid composition of the seed oil, the total tocopherol content, and composition in a collection of 22 yam bean accessions of contrasting ecogeographical origins.

MATERIALS AND METHODS

Germplasm accessions. Fourteen accessions of *P. ahipa* were obtained during a collection trip in Bolivia (3). Three accessions of *P. erosus* originated from Mexico and two from Indonesia. *Pachyrhizus tuberosus* accessions originated from Peru. For *P. ahipa*, 37 and 38 individual plants were grown in 1996 and 1997, respectively, and the seeds from each individual plant were bulked for seed quality analyses. For the other two species, between 5 and 10 individual plants per accession were grown in 1996. Because of the lower seed production, the seeds from all the plants of each accession were bulked for the seed quality analyses. In both years, the plants were cultivated under greenhouse conditions, with temperatures not below 23°C.

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Analysis of protein content. Nitrogen concentration was determined for about 20 mg flour by Dumas combustion method using an automated CN analyzer (Heraeus CN-Rapid, Hanau, Germany) and standard procedures (8). Total nitrogen was converted to percent protein using a factor of 6.25.

Analysis of oil content and fatty acid composition. The seed oil content was determined on milled samples of about 200 mg by Soxhlet oil extraction with petroleum ether. The fatty acid composition of the seed oil was determined by gas–liquid chromatography (GLC) of fatty acid methyl esters (FAME). FAME were prepared following the procedure developed by Thies (9), and analyzed on a Perkin Elmer gas chromatograph model 8600 (Perkin Elmer Corp., Norwalk, CT) equipped with a fused-silica capillary column FFAP, 25 m × 0.25 mm × 0.25 µm film thickness (Macherey & Nagel GmbH + Co KG, Düren, Germany). The oven, detector, and injector temperatures were 200, 250, and 250°C, respectively. The carrier gas was hydrogen, at a pressure of 100 kpa. Two microliters of sample was injected at a split rate of 1:70. Individual fatty acids were expressed as percentage of the total fatty acids, including other minor fatty acids.

Analysis of tocopherols. About 50 mg of ground seeds was analyzed for tocopherol content by HPLC as described by Thies (10), with a fluorescence detector (excitation = 295 nm; emission = 330 nm), a C 18 diol column (250 × 3 mm), and *i*-octane/*t*-butyl-methylether (94:6) as eluent at a flow rate of 0.7 mL/min^{-1}. β-Tocopherol was used as the external standard. Total tocopherol content was expressed as mg/kg^{-1} seed oil. The concentration of the individual tocopherols was expressed as a percentage of the total tocopherol content.

RESULTS AND DISCUSSION

The oil and protein contents, fatty acid composition of the seed oil, and the total tocopherol content and composition of the 83 yam bean samples analyzed in the present study are listed in Table 1. The three species were characterized by a high oil content, with average values of above 20%, and high protein content, with average values of above 25%, confirming for the three species the results reported by Duke (4) and Santos *et al.* (5) for *P. erosus*.

The fatty acid composition of the seed oil was characterized by high palmitic acid (>24% of the total fatty acids in all the samples evaluated), high oleic acid (>20%), and high linoleic acid concentration (>35%). Conversely, linolenic acid concentration was very low, between 1.0 and 2.5%. These results demonstrate that the fatty acid profile found by Broadbent and Shone (7) for one accession of *P. erosus* is characteristic of the genus, with low variability being detected in the material included in the present study. The fatty acid compo-

TABLE 1
Mean and Range of Oil and Protein Contents, Fatty Acid Composition of the Seed Oil, Total Tocopherol Content, and Tocopherol Composition[a]

	P. erosus 1996	*P. tuberosus* 1996	*P. ahipa* 1996	*P. ahipa* 1997
No. of samples	5	3	37	38
Oil content	24.0 22.0–25.9	21.8 20.2–22.7	25.8 24.1–28.1	22.0 18.7–24.8
Protein content	29.9 28.9–31.8	32.2 30.1–33.8	25.7 23.2–27.6	26.5 23.7–30.8
Palmitic acid	27.8 25.0–31.4	24.2 24.0–24.5	29.2 27.5–31.05	29.3 26.8–31.1
Stearic acid	4.5 3.9–5.2	5.2 4.7–5.5	5.5 4.7–7.5	5.4 4.7–6.4
Oleic acid	25.4 22.2–28.6	26.6 25.7–27.2	23.1 21.0–24.9	23.7 22.3–25.8
Linoleic acid	37.0 35.9–38.1	37.6 36.7–39.0	36.9 35.0–40.7	36.2 34.9–39.4
Linolenic acid	1.3 0.9–1.6	2.0 2.0–2.1	1.6 1.3–2.5	1.6 1.3–2.3
Total tocopherols	443.7 249.3–585.4	285.2 259.9–311.9	631.2 508.5–858.0	684.4 465.9–896.2
α-Tocopherol	4.8 2.3–9.7	4.1 0–8.3	0.3 0.1 –0.7	0.4 0.1–0.9
γ-Tocopherol	94.5 90.1–97.5	95.7 91.4–100	98.5 97.7–99.8	98.4 97.7–99.3
δ-Tocopherol	0.7 0.1 –2.5	0.2 0–0.4	1.2 0.1–1.9	1.3 0.6–2.0

[a]Oil and protein contents are expressed as % seed weight; fatty acid composition is expressed as % total fatty acids; total tocopherol content is expressed as mg/kg; and tocopherol composition is expressed as % total tocopherol content. Means and ranges are for 83 samples of yam beans (*Pachyrhizus* spp.) grown in 1996 or 1997.

sition of the *P. ahipa* accessions was very similar during the two consecutive years (Table 1).

The accessions of *P. ahipa* showed higher tocopherol content than the accessions from the other two species, with an average value of 631 mg kg^{-1} oil in 1996 compared with 444 mg kg^{-1} in *P. erosus* and 285 mg kg^{-1} in *P. tuberosus* grown under the same conditions (Table 1). In all cases γ-tocopherol was predominant, accounting for more than 90% of the total tocopherol content. Relatively high levels of α-tocopherol (up to 9.7%) were detected in some samples of *P. erosus* and *P. tuberosus,* but not in *P. ahipa*, were the concentration of α-tocopherol was in all the accessions below 1% of the total tocopherol content during 2 yr.

The results of this study reveal that the levels of oil and protein of yam bean seeds resemble those typical of soybean seeds, which contain about 34% protein and 19% oil (11). Conversely, the fatty acid composition of yam bean oil is very different from that of soybean, which is characterized by a considerably lower concentration of palmitic acid and a higher concentration of linolenic acid (6). Previous studies have shown the existence of legume species with either high seed oil content with a low concentration of saturated fatty acids (soybean, groundnut) (6) or low seed oil content with a high concentration of saturated fatty acids (e.g., *Acacia* spp., *Cajanus* spp., *Vigna* spp.) (4,12). In the case of yam beans, the high seed oil content is paralleled by a high concentration of saturated fatty acids, representing an exception within the family Leguminoseae and an interesting potential source of oil rich in saturated fatty acids.

Vegetable fats with high concentrations of saturated fatty acids are desired by the food industry, especially to avoid the need for hydrogenation and transesterification processes in the production of margarine and related products (13). In fact, there are currently breeding programs focusing on the development of varieties of the major oilseed crops with a high concentration of saturated fatty acids in the seed oil (14–17). It is worth noting that the palmitic acid concentration of *P. ahipa* is considerably higher than that of the high palmitic acid mutants of soybean (15 to 20%) (18–21), and similar to that of the high palmitic acid mutants of flax (about 28%) (15) and sunflower (25 to 30%) (16,19,20). Furthermore, yam bean oil has the additional advantage of possessing a considerably lower linolenic acid concentration than the mentioned mutants of soybean (4 to 8% linolenic acid) and flax (43% linolenic acid).

Table 2 shows the fatty acid composition and the tocopherol content and composition of *P. ahipa* seed oil as compared with current sources of high palmitic acid oil. Total tocopherol content of yam bean oil is higher than that of palm oil and cocoa butter, but lower than tocopherol content in cottonseed oil. However, γ-tocopherol in *P. ahipa* represents more than 98% of the total tocopherols, compared with a concentration of 58% γ- and 41% α-tocopherol in cottonseed. Although the results of the *in vitro* antioxidant effectiveness of the different tocopherols are somewhat contradictory, there is evidence supporting the higher antioxidant strength of γ- vs. α-tocopherol (10,21–23). Additionally, Demurin *et al.* (24) reported the existence of synergism of fatty acid and tocopherol composition. In this way, the combination of a high palmitic acid concentration with such high levels of γ-tocopherol is unique, and its implications for oil stability will have to be analyzed.

Currently, yam beans are exclusively grown on small scale for tuber production and the seeds remain as crop residuals on the field. However, the seeds could be used by farmers for small-scale oil processing with additional by-products in form of protein and rotenone. Nevertheless, evaluation of the seed vs. tuber yield potential and selection for eliminating the rotenone content of the seeds are still needed before the crop can be extended as a major-scale production oil crop. It is also worth noting that *P. ahipa* is short-day insensitive and comes from the cold highland tropics, so it might be successfully grown in higher degrees of latitude.

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TABLE 2
Major Fatty Acids (% of the total fatty acids), Total Tocopherol Content (mg kg^{-1} oil) and Concentration of Individual Tocopherols (% of total tocopherol content) of Oils Rich in Palmitic Acid[a]

	Fatty acids					Tocopherols				
Oil	16:0	18:0	18:1	18:2	18:3	Total	α	β	γ	δ
Cocoa[b]	26.0	34.4	34.8	3.0	0.2	198	5.5	0.0	85.9	8.6
Cottonseed[b]	27.3	2.0	18.3	50.5	0.0	983	40.9	0.2	58.2	0.8
Palm[b]	44.3	4.6	38.7	10.5	0.3	617[c]	13.8	0.0	20.1	2.9
Yam bean[d]	29.3	5.5	23.4	36.6	1.6	655	0.4	0.0	98.2	1.4

[a]Values for major fatty acids and total tocopherol content are expressed as in Table 1. Concentration of individual tocopherols is expressed as % total tocopherol content.
[b]Data from Gunstone *et al.* (6).
[c]Includes 20.1% α-, 51.4% γ-, and 11.7% δ-tocotrienol.
[d]Average data for *Pachyrhyzus ahipa* in the two environments included in this study (see Table 1).

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