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Nutritional and antinutritional compositions of processed Avocado (*Persea americana* Mill) seeds

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ABSTRACT

Research has been on-going on many plant materials, especially those discarded as waste, to exploit their nutritional and antinutritional properties. This is as a result of the United Nations' millennium development goal to eradicate extreme poverty and hunger. Avocado (Persea americana Mill) seeds are often discarded after taking the pulp of the fruit. This study is aimed at evaluating the effect of processing methods (soaking and boiling for various periods) on the nutritive values and antinutritional factors of Persea americana seed as a step towards establishing purposeful utilization of the seed. The processed avocado seeds were evaluated for proximate, mineral, antinutritional factors, and vitamin compositions. Raw avocado seed had the lowest moisture content (73.69%) while the soaked seeds had the highest moisture content (80.73%). Analysis of variance (ANOVA) showed significant differences ($p < 0.05$) in moisture, crude protein, crude fat, ash contents, crude fibre, carbohydrate, vitamins C and E and mineral contents at different processing methods. Boiling of the seeds for 15, 20 and 25 minutes reduced the vitamin A content of the seed by 16%, 13% and 15% as compared with the raw seeds. Soaking of seeds only reduced vitamin A content by 3%. There was high percentage reduction of vitamin C content (81%) in seeds soaked in water as compared with raw seeds. Percentage reduction of vitamin E contents ranged between 4.0% - 7.0%. The raw seeds had the highest content of sodium, calcium and potassium. Soaking reduced the tannin, phytic acid, alkaloid, saponin and oxalate contents of the seeds by 65%, 58%, 64%, 48% and 49% respectively. Boiling for 25 minutes also reduced the tannin, phytic acid, alkaloid, saponin and oxalate contents by 75%, 53%, 79%, 21% and 32% respectively. In general, soaking and boiling of the avocado seeds reduced the antinutritional factors to a large extent.

Keywords: Lauraceae, antinutritional factors, pear, waste, proximate analysis, soaking, boiling

INTRODUCTION

Persea americana Mill, commonly known as avocado pear, is an evergreen tree belonging to the family Lauraceae. It is a tropical tree native to Mexico, Central America and South America but it is now grown worldwide (Adodo, 1995). *Persea americana* is high in antioxidants, vitamins (e.g. resveratrol and vitamin D), heart-friendly monounsaturated fatty acids, fibre and potassium, with variable data suggesting a role in anticancer, anti-infection, anti-inflammatory, and life-prolonging effects (Asha *et al.*, 2009; Kim and Uhl, 2011). Avocado fruit has a multipurpose value as food, medicine, source of high quality oil and numerous industrial uses (Human, 1987, Bergh, 1992). The edible fleshy part of avocado is amongst the most nutritious of all salad fruits. It is served as a salad vegetable. It can also be eaten raw or on bread and tortills. When seasoned, the flesh is also used as a sandwich filling (Morton, 1987). The young leaves of *Persea americana* are used for hair treatment (Verheij and Coronel, 1992).

Persea americana have been applied ethnomedically, most especially in some African countries. It is believed to possess aphrodisiac properties (Asha *et al*., 2009). The skin of the fruit is employed as a vermifuge and remedy for dysentery. The aqueous extract from the leaves are used to correct hypertension and irregular heart beat (Odugbemi, 2006) and the leaves, roasted and ground seeds are used for diarrhea and dysentery. Avocado seed oils are used in healing skin eruptions (Swisher, 1988). The edible flesh is low in sugar content and thus serves as a high energy source for diabetics.

One of the millennium development goals initiated by the United Nations in 2000 is to eradicate extreme poverty and hunger. Thus, research has been on-going on many plant materials, especially those discarded as waste, to exploit their nutritional and antinutritional properties. *Persea americana* seeds is often discarded after taking the pulp of the fruit. Research has shown that the avocado seed is a good source of carbohydrate, protein, fat and some mineral elements such as calcium, phosphorus, potassium and magnesium as well as high concentration of antinutritional factors such as phytate, oxalate and cyanogenic glycosides making the seed to appear potentially toxic (Nwaogu *et al*., 2008). This study is, therefore, aimed at evaluating the effect of processing methods (soaking and boiling) on the nutritive values and antinutritional factors of *Persea americana* seed as a step towards establishing purposeful utilization of the seed.

MATERIALS AND METHODS

2.1 *Materials*
Mature avocado fruits were purchased from Oje market in Ibadan East Local Government Area of Oyo State, Nigeria and authenticated by at I.A.R.&T., Moor Plantation, Ibadan. The seeds of the fruits were removed and those of uniform size were randomly picked. All reagents used were of analytical grade.

2.2 *Preparation of plant materials*
The seeds of *Persea americana* were manually chopped and aliquots was ground and used for moisture content determination. The chopped seeds were also subjected to two processing methods: boiling and soaking. For boiling, the chopped seeds were boiled at 100 ^{o}C for 10, 15, 20 and 25 minutes respectively. Aliquots were ground for moisture content determination. For soaking, the chopped seeds were soaked in water for 24 hours with occasional stirring. Aliquots were ground for moisture content determination. All samples (raw, boiled and soaked) were drained, oven-dried at 60 ^{o}C for 10 hours and milled in a laboratory type hammer mill with a mesh screen size of 0.25 mm into fine powder. The powder (which served as the control) was sealed in polythene bag and stored at 4 ^{o}C until needed for analysis. The waters used for boiling and soaking were also analyzed respectively.

2.3 *Proximate analysis*
The seed samples were analyzed for moisture, percentage ash, crude protein (micro-Kjeldhal method), crude fat (ether extract), crude fibre, total carbohydrate and gross energy according to AOAC (1990).

2.4 *Determination of mineral contents*
Potassium and sodium were estimating using a modified method of Bonire *et al*., (1990). The samples were digested with perchloric acid and nitric acid and analyzed with digital flame photometer (spectronic 20). Phosphorus was determined by vanado-molybdate colorimetric method. Calcium and iron content was achieved with Atomic Absorption Spectrometry (Buck Scientific, Norwalk).

2.5 *Determination of vitamins*
Vitamin A (β-carotene$_{eq}$) and vitamin E (tocopherol) contents were determined according to AOAC (1984). Vitamin C (ascorbic acid) content was determining according to the method of Barros *et al.* (2007).

2.6 *Determination of antinutrients*
Oxalate content was determined by a modified method of Libert and Franceschi (1987). Briefly, 1 g of the sample was weighed into a 250 ml conical flask, soaked in 100 ml distilled water and allow to stand for 3 hours, followed by filtration through a double layer of filter paper. Absorbance was determined spectrophotometrically at 420 nm. The oxalate concentration was extrapolated from a standard curve. Phytate content was estimated by the titration method of Sudarmadji and Markakis (1977). Tannin content was determined by the spectrophotometric method of Griffiths and Jones (1977). Saponin content was determined by the method of Makkar and Becker (1996) and alkaloid content was estimated by the distillation and tritrimetric method of Henry (1973).

2.7 *Statistical analysis*

The results obtained were expressed as mean ± standard error of mean (SEM) and subjected to one-way analysis of variance (ANOVA). Least significance difference test was used to determine the significant difference between the samples.

RESULTS AND DISCUSSION

The proximate composition of avocado seed subjected to soaking and boiling is as presented in Table 1. Raw avocado seed had the lowest moisture content (73.69%) while the soaked seeds had the highest moisture content (80.73%). Analysis of variance (ANOVA) showed significant difference in moisture content of the seed ($p < 0.05$), the moisture content values of seeds boiled for 15 minutes and 20 minutes were the same but significantly different ($p < 0.05$) from the seeds boiled for 10 minutes, 20 minutes and the raw seed likewise the values of seeds boiled for 25 minutes and soaked seeds were in close range and there was no significant difference ($p > 0.05$) between them.

Table 1: Effect of processing methods on the proximate composition (on dry matter basis) of *P. americana* seeds

Sample Treatments	Moisture (%)	Dry Matter (%)	Ash (%)	Crude Protein (%)	Ether Extract (crude fat) (%)	Crude Fibre (%)	**Carbohydrate (%)	Gross Energy (Kcal/100 g)
T_1	$73.69^{a}\pm0.02$	$26.30^{a}\pm0.03$	$5.21^{a}\pm0.03$	$6.34^{a}\pm0.34$	$16.81^{a}\pm0.02$	$3.97^{a}\pm0.01$	$67.68^{a}\pm0.31$	$4.49^{a}\pm0.18$
T_2	$74.25^{a}\pm0.05$	$25.75^{a}\pm0.05$	$4.68^{b}\pm0.01$	$15.58^{d}\pm0.18$	$28.90^{d}\pm0.03$	$3.29^{b}\pm0.02$	$47.56^{b}\pm0.23$	$5.48^{c}\pm0.09$
T_3	$76.79^{b}\pm0.01$	$23.21^{b}\pm0.01$	$3.77^{c}\pm0.01$	$15.23^{cd}\pm0.18$	$28.74^{c}\pm0.01$	$2.95^{c}\pm0.01$	$49.32^{c}\pm0.17$	$5.30^{bc}\pm0.09$
T_4	$77.35^{b}\pm0.15$	$22.65^{b}\pm0.15$	$3.38^{d}\pm0.02$	$14.97^{bc}\pm0.09$	$28.69^{c}\pm0.00$	$2.16^{d}\pm0.01$	$50.81^{d}\pm0.09$	$5.12^{bc}\pm0.09$
T_5	$78.75^{c}\pm0.75$	$21.25^{c}\pm0.75$	$2.62^{e}\pm0.01$	$14.44^{b}\pm0.09$	$28.62^{b}\pm0.01$	$1.58^{e}\pm0.02$	$52.75^{e}\pm0.09$	$4.94^{b}\pm0.10$
T_6	$80.73^{c}\pm0.01$	$19.27^{c}\pm0.01$	$2.25^{f}\pm0.05$	$23.54^{e}\pm0.09$	$30.83^{e}\pm0.01$	$13.76^{f}\pm0.02$	$29.66^{f}\pm0.11$	$6.01^{d}\pm0.09$

Results are presented as mean ± standard error of mean

Means for each attribute followed by the same letter are not significantly different (p>0.05) by least significant different test (LSD)

***Calculated by difference*

T_1: Raw seeds, T_2: Seeds boiled for 10 minutes, T_3: Seeds boiled for 15 minutes, T_4: Seeds boiled for 20 minutes, T_5: Seeds boiled for 25 minutes, T_6: Soaked seeds.

Table 2: Effect of processing on vitamin composition (dry matter basis) of *Persea* seeds

Sample treatment	Vitamin A (μg/100 g)	Loss (%)	Vitamin C (mg/100 g)	Loss (%)	Vitamin E (mg/100 g)	Loss (%)
T_1	$207.02^{d}\pm3.40$	-	$14.63^{f}\pm0.21$	-	$0.65^{c}\pm0.00$	-
T_2	$186.63^{bc}\pm3.39$	10.0	$9.12^{e}\pm0.22$	38.0	$0.61^{a}\pm0.01$	7.0
T_3	$173.08^{a}\pm3.39$	16.0	$7.00^{d}\pm0.22$	52.0	$0.61^{a}\pm0.00$	7.0
T_4	$179.85^{ab}\pm3.38$	13.0	$6.15^{c}\pm0.21$	58.0	$0.63^{b}\pm0.00$	4.0
T_5	$176.46^{ab}\pm3.40$	15.0	$4.88^{b}\pm0.22$	67.0	$0.63^{b}\pm0.00$	4.0
T_6	$200.23^{cd}\pm3.40$	3.0	$2.76^{a}\pm0.22$	81.0	$0.63^{b}\pm0.01$	4.0

Results are presented as mean ± standard error of mean

Means in the same column followed by the same letter are not significantly different (p > 0.05).

Table 3: Vitamin content in processing water of *Persea americana* seeds

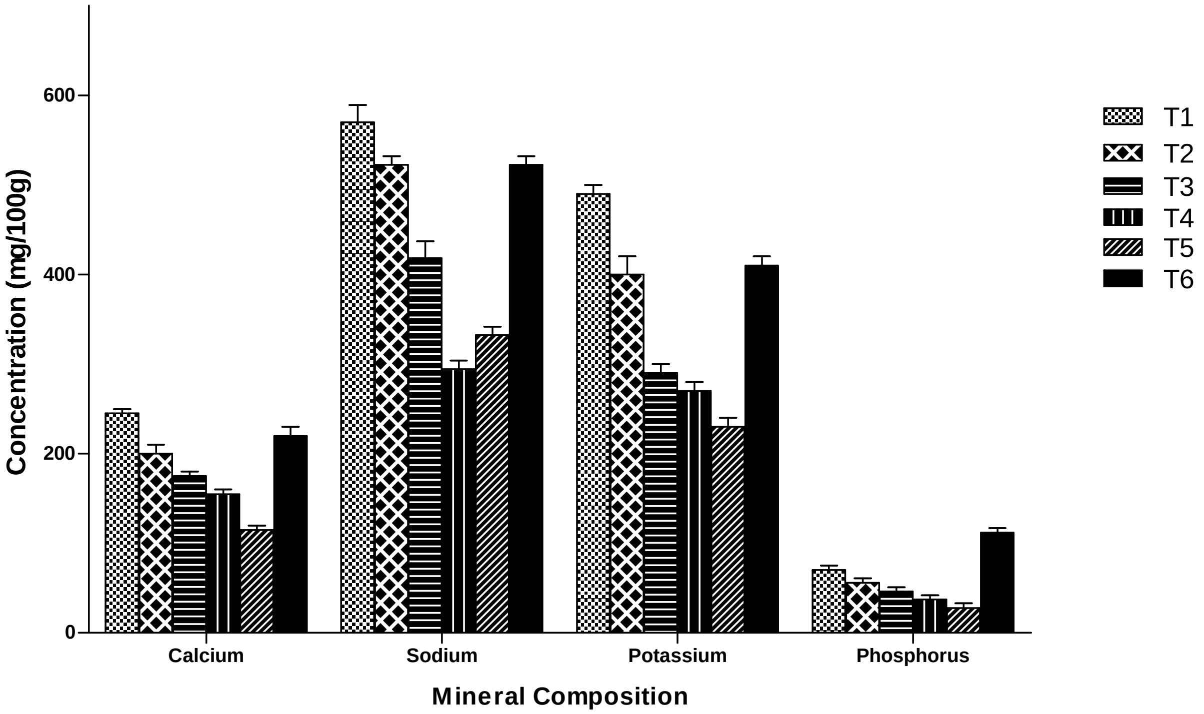
Sample treatment	Vitamin A (μg/100 ml)	Vitamin C (g/100 ml)	Vitamin E (mg/100 ml)
Soaked water	-	$4.03^{c}\pm0.21$	-
Boiled water (10 mins)	-	$0.36^{a}\pm0.02$	-
Boiled water (15 mins)	-	$0.40^{a}\pm0.02$	-
Boiled water (20 mins)	-	$1.06^{a}\pm0.02$	-
Boiled water (25 mins)	-	$1.49^{b}\pm0.22$	-

-: No trace

Table 4: Antinutritional factors (g/100 g on dry matter basis) in *Persea americana* seeds

Sample Treatments	Alkaloid	Loss (%)	Tannin	Loss (%)	Phytic acid	Loss (%)	Saponin	Loss (%)	Oxalate	Loss (%)
T_1	$2.05^{e}\pm0.04$	-	$11.29^{a}\pm0.11$	-	$12.87^{a}\pm0.02$	-	$0.92^{e}\pm0.00$	-	$4.07^{a}\pm0.00$	-
T_2	$1.61^{d}\pm0.03$	21.0	$8.44^{b}\pm0.11$	25.0	$11.35^{b}\pm0.02$	11.0	$0.83^{d}\pm0.00$	9.0	$3.86^{b}\pm0.02$	5.0
T_3	$1.33^{c}\pm0.05$	35.0	$7.89^{c}\pm0.00$	30.0	$9.54^{c}\pm0.03$	25.0	$0.80^{c}\pm0.00$	12.0	$3.57^{c}\pm0.01$	12.0
T_4	$1.05^{b}\pm0.05$	45.0	$5.37^{d}\pm0.11$	51.0	$7.19^{d}\pm0.02$	44.0	$0.76^{c}\pm0.01$	14.0	$3.12^{d}\pm0.05$	26.0
T_5	$0.42^{a}\pm0.05$	79.0	$2.74^{e}\pm0.11$	75.0	$5.99^{e}\pm0.02$	53.0	$0.72^{b}\pm0.00$	21.0	$2.77^{e}\pm0.00$	32.0
T_6	$0.32^{a}\pm0.05$	64.0	$3.60^{f}\pm0.11$	65.0	$5.29^{f}\pm0.02$	58.0	$0.48^{a}\pm0.01$	48.0	$2.09^{f}\pm0.01$	49.0

-: No trace



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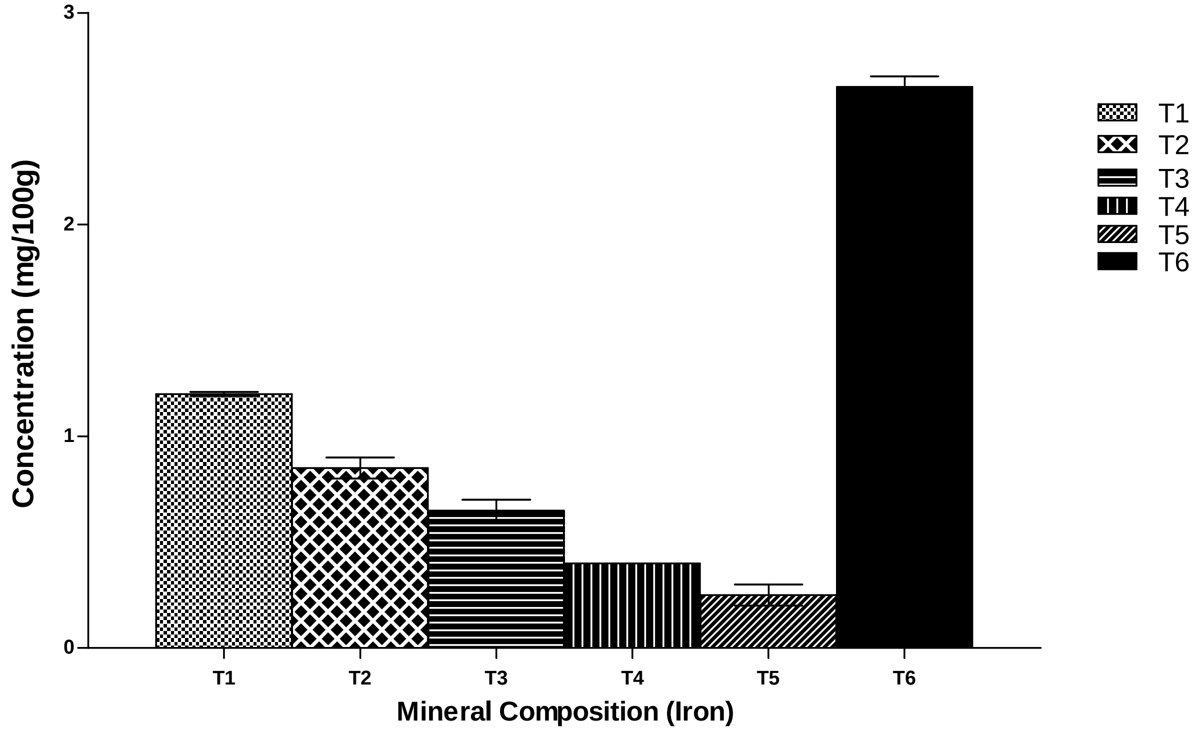

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Figure 1: Effect of processing on mineral composition (mg/100 g on dry matter basis) of *Persea americana* seeds

Analysis of variance (ANOVA) showed significant difference ($p < 0.05$) in crude protein values. There was also significant differences ($p < 0.05$) obtained for crude fat, ash contents, crude fibre, and carbohydrate contents at different processing methods. The protein contents of the samples boiled for 10, 15, 20 and 25 minutes were in close range irrespective of the boiling time. The crude fat value obtained for seeds boiled for 15 and 20 minutes were not significantly different from each other but were significantly different from the values obtained for raw seeds, seeds boiled for 10 minutes and 25 minutes and soaked seeds. There was no significant difference ($p > 0.05$) in the gross energy values of seeds boiled for 10, 15, 20 and 25 minutes, but they were significantly different from the raw seeds and soaked seeds.

The result of vitamin contents on the various processed *Persea americana* seeds is as shown in table 2. The raw seeds had the highest vitamins A, C and E contents. There was no significant difference ($p > 0.05$) in vitamin A contents of all the samples. However, boiling of the seeds for 15, 20 and 25 minutes reduced the vitamin A content of the seed by 16%, 13% and 15% as compared with the raw seed value. Soaking of seeds only reduced vitamin A content by 3%. There was high significant difference in the vitamin C content of the seeds when subjected to the different processing methods. There was high percentage reduction of vitamin C content (81%) in seeds soaked in water for 24 hours as compared with raw seeds and a low percentage reduction was observed in seeds boiled for 10 minutes (38%). There was significant difference in vitamin E content of all processed seeds. Vitamin E contents of seeds boiled for 20 and 25 minutes and soaked seeds were not significantly different from one another ($p > 0.05$). Moreover, percentage reduction of vitamin E contents was low, ranging between 4.0% - 7.0%. The water used for processing the seeds showed no significant values of both vitamins A and E. the was significant amount of vitamin C present in the water used for boiling and soaking avocado's seeds as presented in table 3.

The result of the mineral composition of processed *Persea americana* seeds is presented in Figure 1. Analysis of variance revealed a significant difference in the mineral contents of the seeds when subjected to different processing methods. The raw seeds (control) had the highest content of sodium, calcium and potassium.

The result of antinutritional factors (alkaloid, tannin, phytic acid, saponin and oxalate) on the processed seeds of *P. americana* is presented Table 4 while the result of antinutritional factors in the processing water of *P. americana* are as presented in Figure 3. Soaking and boiling of the avocado seeds reduced the antinutritional factors to a large extent. Water used in processing the soaked seeds had the highest contents of alkaloid, tannin, phytic acid, saponin and oxalate. The water used in processing the boiled seeds (for 10 minutes) had the least values of antinutritional factors' contents.

The high moisture content values observed in the soaked and boiled seeds as compared with the raw sample might be due to soaking and boiling of the seeds. Boiling and soaking in water softened the cell tissues of the seed, increasing the water-absorbing and retention capacities of the seeds due to increased permeability of the cell membrane to water. Increased moisture content as a result of boiling and soaking has been reported for mango seed kernels (Ravindran and Sivakanesan, 1996). The ash content o the raw seed (5.21%) was higher than the value obtained for mango seed kernel (1.72%) (Morton, 1987), African pear, *Dacryodes eddulis* (1.26%) (Obasi and Okolie, 1993). The loss of ash contents observed in boiled and soaked seed samples as compared to the raw seeds may be due to leaching of soluble inorganic salts into the processing water. The crude protein content of the raw seed was lower than the value obtained for *Opuntia ficus indica* (prickly pear) (Kossori *et al*., 1998) and *Dacryodes edulis* (African native pear) seeds (Obasi and Okolie, 1993), but was much higher when compared to the value obtained with its pulp (Morton, 1987). Increase in protein content observed during boiling and soaking of *P. americana* seeds may be due to high tannin, oxalate, alkaloid and phytate in the seed that were lost drastically during the processing techniques employed, rendering the protein content of the seeds more bioavailable (Osagie, 1998). Boiling and soaking increased the ether extract content of *P. americana* seeds. It is possible that these techniques enhanced the efficiency of oil extraction from the samples by the solvent. High ether extract content of avocado seed is indicative of the fact that the seed is rich in oil which makes it a good source of vegetable oil for nutritional and industrial purposes. Raw avocado seed showed a carbohydrate content of 67.68%, which is comparable with starchy foods such as cereals. Carbohydrate content is greatly reduced during boiling and soaking of the seeds which may be as a result of leaching of soluble carbohydrates like sugars. Also, carbohydrate loss may be partly attributed to non-enzymic browing reactions between the amino acids and sugars contained in appreciable amounts in the seed during thermal processing. This reaction may account, in part, for the light brown colour of the seeds. The low gross energy values obtained for avocado seeds imply presence of soluble sugars in large quantity suggesting that the seed could be exploited in feed formulations as an excellent source of low energy for ruminants (Enujiugha and Agbede, 2000).

Vitamin C content value obtained for *Persea americana* raw seed (14.63 mg/100 g) falls within the range of values obtained for its pulp (4.5–21.3 mg/100 g) (Morton, 1987). Vitamin C is a water soluble vitamin and heat labile, thus, 38–67% was lost during boiling and 81% during soaking of the seeds. Losses of vitamin C are mostly as a result of leaching into the processing water, thermal destruction and oxidation (Fellows, 2000). The low ascorbic acid value of avocado seed gives an indication that the seeds may be poor source of ascorbic acid when compared to citrus fruits (Adepoju, 2004). Vitamin A (β-carotene equivalent) contents of the seeds samples were high (173.08–207.00 μg/100 g). The seeds also contain appreciable amount of vitamin E. Antioxidants such as ascorbic acid, carotenoids and tocopherol have been associated with prevention of nutritionally associated diseases such as cancer, coronary heart disease and obesity (Larrauri *et al*., 1996). Vitamins A and E are oil soluble vitamins that are mostly contained within the dry matter of the seed. This is the reason they are not leached into the water used in processing.

Macro minerals like calcium, potassium and sodium had moderately high values. Phosphorus and iron, on the other hand, had low values. Phytic acids are the principal form of phosphorus in many seeds; 60–90% of phosphorus in seeds is present as phytic acid (Mohammed and Oloyede, 2004) and this lowers the total phosphorus value in avocado. Moreover, phytic acid is a strong chelator of many divalent minerals such as iron, magnesium and calcium and renders these minerals biologically unavailable.

The high antinutritional factors present in the raw seeds of *Persea americana* could be recognized as a potential threat in the use of these seeds in animal and human nutrition, in spite of its nutritional composition. However, the processing techniques employed in this study reduced these natural toxicants greatly. Tannins are known to bind irreversibly to proteins of forming insoluble complexes with them and thus rendering them indigestible by intestinal enzymes thereby interfering with their bioavailability (Meshansho *et* al., 1987; Liener, 1994). Seventy-five percent of tannin in *Persea americana* raw seeds was removed after boiling the seed for 25 minutes and 65% after soaking the seeds for 24 hours, which was much higher than the loss reported for mango seed kernel after soaking (44%) and boiling (67%) (Ravindran and Sivakanesan, 1996). The phytic acid content of avocado raw seed (12.87 g/100 g) was higher than that of African wild mango (2.9 g/100 g) (Oboh and Ekperigin, 2003). The antinutritional nature of phytic acid lies in its ability to chelate divalent minerals such as iron, calcium, copper and zinc, rendering them biologically unavailable. The high phytic acid content in raw avocado seed is of nutritional significance. It cannot be broken down by humans and monogastrics (Osagie, 1998). Processing techniques (soaking and boiling) significantly ($p < 0.05$) reduced the phytic acid content of avocado seeds.

Saponin levels in all the seed samples were generally low and thus could not produce adverse effects on the growth of animals. The oxalate level in the seed samples were below the lethal dosage of soluble oxalate intake (2 – 5 g) (Ezeagu, 1997).

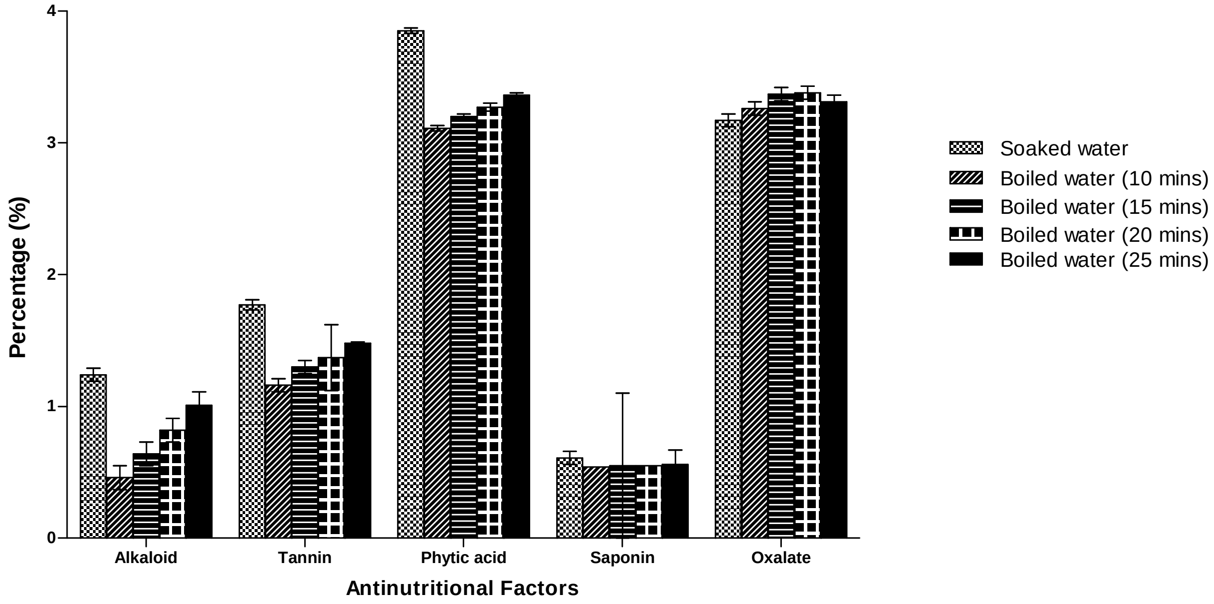

Figure 2: Anti-nutritional factors (g/100 ml) in processing water of *Persea americana* seeds

CONCLUSION

Persea americana is a good source of dietary protein and its high fat content could contribute calories to man and animal ration. The limitation to the full utilization of *Persea americana* seeds is the high concentrations of antinutritional factors (tannin, phytic acid and alkaloids) which renders it useless for human and animal nutrition. However, processing methods, such as soaking and boiling, reduced the levels of these antinutrients present in the raw seeds.

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