

Bioactive Compounds in Cashew Nut (*Anacardium occidentale* L.) Kernels: Effect of Different Shelling Methods

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In the present study, the effects of various conventional shelling methods (oil-bath roasting, direct steam roasting, drying, and open pan roasting) as well as a novel "Flores" hand-cracking method on the levels of bioactive compounds of cashew nut kernels were investigated. The raw cashew nut kernels were found to possess appreciable levels of certain bioactive compounds such as β -carotene (9.57 μ g/100 g of DM), lutein (30.29 μ g/100 g of DM), zeaxanthin (0.56 μ g/100 g of DM), α -tocopherol (0.29 mg/100 g of DM), γ -tocopherol (1.10 mg/100 g of DM), thiamin (1.08 mg/100 g of DM), stearic acid (4.96 g/100 g of DM), oleic acid (21.87 g/100 g of DM), and linoleic acid (5.55 g/100 g of DM). All of the conventional shelling methods including oil-bath roasting, steam roasting, drying, and open pan roasting revealed a significant reduction, whereas the Flores hand-cracking method exhibited similar levels of carotenoids, thiamin, and unsaturated fatty acids in cashew nuts when compared to raw unprocessed samples.

KEYWORDS: Anacardium occidentale L.; cashew nut; bioactive compounds; carotenoids; tocopherols; thiamin; unsaturated fatty acids; shelling methods; "Flores" hand-cracking

INTRODUCTION

Nuts are recommended as an important constituent of a healthy diet, although their real intake varies remarkably in human populations in different regions of the world. Nuts constitute a good source of certain vital bioactive compounds that could elicit many health benefits in human beings. Results of several epidemiological studies suggested that there may be a connection between frequent nut consumption and reduced incidence of several chronic diseases (1). Long-term consumption of nuts has been associated with a lower risk of body weight gain and obesity (2). The consumption of nuts as a part of the healthy diet has a positive influence on the fatty acid profile of persons with type 2 diabetes (3). Analysis of the effects of the inclusion of cashew nut in the diet on the antioxidant status of human subjects with metabolic syndrome resulted in an increased antioxidant capacity (4). Furthermore, studies by Chisholm et al. (5) revealed that nut consumption has a cholesterol-lowering effect and also reduced the risk of lipoprotein-mediated cardiovascular disease, and recent emerging scientific findings have demonstrated that the bioactive constituents of whole nuts have cardioprotective, antiobesity, anticancer, and antioxidant effects by a number of different mechanisms (6).

Among the various types of commonly consumed nuts such as almond, Brazil nut, hazelnut, macadamia, peanut, pecan, pine, pistachio, kola nut, and walnut, the cashew nut (*Anacardium occidentale* L.) occupies a central position in the diets of the human population throughout the world. Cashew nut, native to Brazil, was introduced about two centuries ago into the Goa region of India, which became one of the major producers of cashew nuts, accounting for almost 50% of the total world export (7). The cashew nut is an extremely important agricultural trade product of Brazil, where cashew cultivation occupies an estimated 700,000 ha with a cashew nut production of 280,000 tons/year (8). Thus, cashew nut production has great social and economic importance in many developing countries, including Brazil, India, Indonesia, and some African countries.

The cashew nut kernels, the edible portion of the nut, which contain proteins, fats, and vitamins with high caloric value (7.32-7.76 kcal/g of DM) (9), are accepted worldwide as a nutritious food product. The cashew nut consists of an outer shell (epicarp), a tightly fitted inner shell (endocarp), and a strongly vesicant cashew nut shell liquid (CNSL). The CNSL is located between the inner and outer shells within a honeycomb matrix (10). It protects the nut from being destroyed by foragers that feed on the cashew apple. The kernel is slightly curved back on itself and forms two cotyledons, representing about 20-25% of the whole nut weight. It is wrapped within a thin, reddish-brown membrane called a testa, which is very difficult to remove (11).

The cashew nut represents one of the cheapest major sources of nonisoprenoid phenolic lipids, which have a variety of biological properties and medicinal applications and have also demonstrated a potential antioxidant activity. Previous research works indicated the presence of phenolic components such as anacardic acid, cardol, 2-methylcardol, and cardanol in CNSL (*12*). Quantitative determination of the major phenolic lipids in cashew apple,

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kernels, and shells of cashew nut at various stages of development suggested the possibility of fatty acid type biogenesis of these phenolic lipids (10). The presence of unsaturated fatty acids, tocopherols, squalenes, and phytosterols is also reported in cashew nuts by Ryan et al. (13). Recently the antioxidant activities of various bioactive compounds such as phenolics, flavonoids, phospholipids, sphingolipids, sterols, and tocopherols were reported in cashew nut samples (14, 15). Furthermore, the ethanolic extract of cashew nut testa has exhibited significant antioxidant activity (11), and the polyphenolic compounds present in the testa appear to contribute to the antioxidant activity (16).

On the world market, large, white, and whole cashew nut kernels of high quality achieve the best prices. However, the processing of cashew samples is very expensive due to the specific characteristics of the shell. The irregular shape in conjunction with the wood-expressed, rugged enclosure makes manual or mechanical cracking very difficult. To win a high percentage of whole kernels, it is necessary to break the shell and remove the testa by means of high-temperature treatments (17). The highly nutritious kernel of the cashew nut is removed from the shell by a process known as shelling, which can be achieved by various methods such as drying, steam roasting, oil-bath roasting, or cooking under highpressure steam (18). During these conventional shelling processes, the cashew nut samples are exposed to extremely high levels of temperature (ranging from 75 to 200 °C), which may affect the heat-sensitive bioactive compounds. Furthermore, the liberated CNSL during the shelling process also seriously complicates the processing of cashew nut and extraction of its kernel for food use.

To overcome these constraints, a novel shelling method ("Flores" hand-cracking method) was introduced by PT. Profil Mitra Abadi (PT. PMA), Tangerang, Jakarta, Indonesia, by using a simple hand-cracking machine. In this process, cashew nut kernels were freed from the shell individually with the help of a new, specially designed hand-cracking machine. Furthermore, the shelling process occurs without any contact with a heat source at any point throughout the cracking process. Subsequently, the cashews are dried for 3 h at a mild temperature of 45 °C.

Even though few reports are available on the presence of certain bioactive compounds in cashew nut kernels, to our knowledge there is no information on the effect of various conventional shelling methods as well as the newly developed Flores handcracking method on the levels of bioactive compounds in cashew nut kernels. Hence, the present study was carried out to analyze the levels of β -carotene, lutein, zeaxanthin, α -tocopherol, γ -tocopherol, thiamin, stearic acid, oleic acid, and linoleic acid in raw and differentially processed cashew nut kernels with a view to identify the more suitable and effective shelling process, which minimizes the loss of health beneficial bioactive compounds.

MATERIALS AND METHODS

Sample. The cashew nut samples were collected from the agricultural farms located at four different villages, Rowa, Ile Padung, Ilenmedo, and Kringa, in central and eastern Flores Island, Indonesia. To ensure comparability of different processing methods with each other, all of the cashew nut samples were obtained from the same harvesting time and the same drying and storage conditions were used. The collected samples were randomly categorized and divided into six batches, and different shelling processes were carried out as described below. Autoxidation and isomerization of the vitamins were prevented by working under yellow light.

Chemicals. Chemicals such as oleic acid, α -tocopherol, and boron trifluoride in methanol were purchased from Fluka, Taufkirchen, Germany; linoleic acid, stearic acid, and thiamine were obtained from Sigma-Aldrich, Steinheim, Germany; lutein, γ -tocopherol, zeaxanthin, and β -carotene were procured from Roche, Basel, Switzerland; isolute-HM-N was purchased from Separtis, Grenzlach-Wyhlen, Germany; and all other chemicals were purchased from Merck, Darmstadt, Germany. **Raw Cashew Nuts.** To determine the initial content of bioactive ingredients of cashew nuts, this group was left untreated, cracked manually by using a wooden hammer, but not dried. Throughout the experiment, these samples were not exposed to heat at any point.

Oil-Bath Roasting. The oil-bath roasting process was carried out on the basis of the methodology described by Mandal (18). In the first step, the water content of the cashew nut samples was increased from 6 to 16% by conditioning. The cashew nuts were individually weighed, added in accordance with the required amount of water, and shaken in Falcon tubes (Sarstedt, Nürnberg, Germany) for 4 days at 9 °C. Then the cashew nut samples were roasted in an oil bath at 200 °C for 90 s and then cooled to room temperature. For roasting, locally available vegetable oil (100% sunflower oil, Biskin) was used. Subsequently, the cashew nuts were cracked and dried at 75 °C in a dry heating block for 3 h. In the dry heating block, a plastic bowl was lined with aluminum foil, so that the cashew nuts did not come in contact with the walls of the heating block.

Steam Roasting. The steam roasting of cashew nut samples was performed as described by Jain et al. (17). The apparatus for steam generation was developed by Giovanni Migliore (Dairy Research and Training, University of Hohenheim). In this method, the vessel containing the cashew nut samples was hermetically sealed, so that the steam did not escape and the pressure could build up completely. The cashew nuts were processed for 15 min at 120 °C and 15 psi of pressure. Subsequently, the steam-roasted cashew nuts were cooled to room temperature, cracked, and dried at 75 °C in a dry heating block for 3 h.

Drying. To analyze whether the two previous methods (oil-bath and steam roasting) or the subsequent drying is responsible for nutrient loss, the cashew nut samples were directly dried after cracking at a temperature of 75 °C for 3 h in a dry heating block and then cooled to room temperature.

Open Pan Roasting. The cashew nut samples were processed by using an open pan roasting treatment as described by Mandal (18). Before the roasting process was begun, the container was sparked by fire and logs created in the wake flame. Then the cashew samples were placed in a perforated pan, which was placed on the fire for 2 min. The heat caused the withdrawal of CNSL, which led to ignition of cashew samples. After 2 min, the cashew nuts were removed from the fire, slowly cooled to room temperature, cracked with wooden hammers, and stored in the dark.

Flores Hand-Cracking. The hand-cracking of cashew nuts was conducted on the basis of a detailed description of production given by Rudolf Heering (President Director, PT. PMA, Tangerang, Jakarta, Indonesia). First, by visual inspection, cashew samples of too small size were removed from the cracking process and only acceptable cashew samples were passed into the breakers. It is inevitable that only a minimum amount of CNSL emerges from the mesocarp of the shell as a result of the cracking process. Thus, the cashew nut kernels were protected themselves from exiting CNSL. The outer surface of the cashew nut kernel was not damaged during the cracking process, so that the cashew nuts in the pan could be removed precisely without violating the testa. Furthermore, the cashew nut kernel, which is protected by testa, was not in contact with CNSL. The samples were cracked and dried for 3 h at a temperature of 45 °C in a heating block.

Preparation of the Samples. Processed sample (50 g) per category was obtained at the end of each shelling process. All of the processed as well as raw samples were frozen at -80 °C and freeze-dried for 48 h, and then the testa was removed manually. The cashew nut kernels without testa were homogenized by using a mortor and pestle, freeze-dried for 24 h, and stored at 9 °C until further use.

Carotenoids and Tocopherols. The β -carotene, lutein, zeaxanthin, and α - and γ -tocopherol contents in cashew nut kernels were analyzed by RP-HPLC after saponification and extraction as follows: 2 mL of ethanol containing pyrogallol (2.5%) and β -apo-8'-carotenal-methyloxime and 1 mL of 50% KOH solution were added to 100 mg of the sample in a screwcapped glass tube. The internal standard β -apo-8'-carotenal-methyloxime was synthesized as described by Sommerburg et al. (19). For saponification, the contents were covered by argon gas, the tubes were closed with screw caps, and then the mixture was stirred for 4 h in a water bath at 38 °C. After the addition of 2 mL of saline solution (15%), the fat-soluble substances were extracted twice with 1 mL of hexane. The combined hexane phases were washed (15% saline solution), evaporated by nitrogen gas, and finally redissolved in 200 μ L of ethanol eluent (1:3, v/v) in order to

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be analyzed on a Varian HPLC (Prostar-210) equipped with UV–vis and fluorescence detector (Waters-2487, Waters-474) with following chromatographic conditions: a Spherisorb ODS-2 analytical column (3 μ m, 250 × 4.6 mm, Trentec, Germany) at 40 °C and a mobile phase consisting of acetonitrile (82%), dioxane (15%), and methanol (3%, containing 100 mM ammonium acetate and 0.1% triethylamine) in a recirculation mode with a flow rate of 1.6 mL/min. The carotenoids were detected at 450 nm, whereas α - and γ -tocopherols were measured by fluorescence with an excitation/emission set at 298/328 nm, respectively.

Thiamin. The determination of the thiamin content of cashew nut samples was performed by precolumn derivatization and reverse-phase liquid chromatography and fluorescence detection as described by Gerrits et al. (20) with modifications. In brief, 100 mg of the sample was mixed with 7.5 mL of 0.1 M HCl solution and stirred for 1 h at 30 °C in dark. Subsequently, an aliquot of 1.5 mL of this mixture was centrifuged at 5000g, and the clear supernatant was derivatized as follows: 500 μ L of the clear solution was mixed with $100 \,\mu\text{L}$ of freshly prepared oxidation reagent (12.1 mM K₃Fe(CN) in 3.35 M NaOH solution). The "thiochrome reaction" was stopped by the addition of 20 μ L of 6 M orthophosphoric acid, and 20 μ L of the aliquot was analyzed on a Merck-Hitachi HPLC (LaChrom) equipped with column oven (set at 40 °C), fluorescence detector (L-7480), and Clarity chromatographic station (DA-C50, DataApex Ltd., Praha). Separation was achieved by using a 5 μ m analytical column (Grom-Sil 120 ODS-4 HE, 125 × 4 mm, Grom, Rottenburg-Hailfingen, Germany) and a mobile phase consisting of methanol (27.5% v/v) and phosphate buffer (pH 7.0) at a flow rate of 0.8 mL/min. Thiamin was detected by excitation/emission set at 367/435 nm.

Fatty Acids. The quantification of fatty acids (stearic acid, oleic acid, and linoleic acid) of raw and processed cashew nut samples was carried out by following the method of Thurnhofer et al. (21). Five hundred milligrams of the sample was extracted by using an ASE 200 system (Dionex, Idstein, Germany). For this purpose, 22 mL extraction cells were used, which were filled with Isolute-HM-N, where each cell was extracted twice. The azeotropic mixture of cyclohexane and ethyl acetate (46:54, v/v) was used as extractant (22). Two extracts were combined, concentrated by using a rotary evaporator with vacuum controller at 60 °C temperature, and precisely adjusted to a definite volume. For the gravimetric determination of fat content, after thorough vortexing, 70 μ L of aliquot was evaporated in a heating block at 60 °C under nitrogen gas and the mass was determined.

To determine the fatty acids, the extracted oil was first transesterified. Ten microliters of the first internal standard (10,11-dichloroundecanoic acid), produced according to the method of Thurnhofer et al. (21), was pipetted into the mixture and extracted with 500 μ L of 0.5 M methanolic KOH solution at 80 °C and then saponified. After a reaction time of 5 min, the mixture was cooled in an ice bath. The subsequent methylation was started by adding 1 mL of boron trifluoride in methanol and kept for 5 min at 80 °C. After cooling, 2 mL of hexane and 2 mL of a saturated NaCl were added and shaken. The contents were centrifuged at 8000g, and 180 μ L of organic phase was mixed with 20 μ L of the second internal standard, oleic acid ethyl ester, which was prepared according to the method of Thurnhofer et al. (21). The solution was diluted in 1:4 ratio and analyzed by gas chromatography in combination with electron ionization mass spectrometry (GC-EI/MS), which consists of a 5890 series II gas chromatograph and a 5971 mass selective detector, MS Data analysis version C.00.07 HP 1989-1992 from Hewlett-Packard, Waldbronn, Germany. For GC analysis, helium gas (99.999% purity) was used as carrier with a flow rate of 1 mL/min. A fused silica capillary column (100% cyanopropylpolysiloxane, 50 m \times 0.25 mm i.d. \times 0.20 μ m film thickness, CP-Sil 88 from Chrompack, Middelburg, The Netherlands) was installed in the GC oven. Injection of a 1 μ L volume of aliquot was used at a temperature of 250 °C and analyzed for 38.81 min. Under selected ion monitoring (SIM) mode, nine fragment ions were detected, and seven of them were identified during the whole run at (1) m/z 74 and (2) m/z 87 for methyl esters of saturated and monounsaturated fatty acids, (3) m/z 81 and (4) m/z 79 for methyl esters of polyunsaturated fatty acids, (5) m/z 88 and (6) m/z 101 for ethyl esters of saturated and monounsaturated fatty acids, and (7) m/z 55.

Statistical Analysis. The statistical analysis was carried out by using SPSS for Windows (SPSS Inc., Chicago, IL, version 11.0). Values of analyzed compounds were found to be normally distributed by using the Kolmogorov–Smirnov test and were described by their mean. Means of

the groups regarding different shelling methods were compared by oneway ANOVA and Dunnett post hoc test using the raw unprocessed cashew nuts as a control. Two-tailed p values of < 0.05 were considered to be statistically significant.

RESULTS AND DISCUSSION

Bioactive Compounds. In general, the raw cashew nut kernels were found to possess appreciable levels of carotenoids such as β -carotene (9.57 μ g/100 g of DM), lutein (30.29 μ g/100 g of DM), and zeaxanthin (0.56 μ g/100 g of DM) (**Table 1**). The β -carotene content of cashew nut was found to be higher than that of peanut $(2 \mu g/100 g)$ but lower when compared to chestnut $(24 \mu g/100 g)$, hazelnut ($29 \,\mu g / 100 \,g$), almond ($120 \,\mu g / 100 \,g$), pecan ($80 \,\mu g / 100 \,g$), pistachios (150 μ g/100 g), walnut (48 μ g/100 g), and previously reported values in cashew nut (60 μ g/100 g) (23). The lutein content of cashew nut $(30.29 \,\mu g/100 \,g)$ was higher than that of an earlier report on red apple ($15 \mu g/100 g$), red grape ($24 \mu g/100 g$), mango (6 μ g/100 g), peach (11 μ g/100 g), and water melon (4 μ g/ 100 g) and comparable with that of orange juice $(33 \mu g/100 g)$ and tomato $(32 \mu g/100 g)(24)$. Nonetheless, the zeaxanthin content of cashew nuts (0.56 μ g/100 g) was lower than that of endive (3 μ g/ 100 g), red grape (4 μ g/100 g), and peach (3 μ g/100 g) (24).

In addition to the importance as provitamin A, β -carotene has a photobiological effect, that is, a protective function against UV light. β -Carotene protects against oxidative stress through deactivation of singlet oxygen and by inhibition of lipid peroxidation (25). Functions of carotenoids are discussed as a precursor of vitamin A, which is required for adaptive immunity and plays a significant role in the development of both T-helper cells and B cells (26). Furthermore, carotenoids are known to play a role in the prevention of diseases such as cancer and atherosclerosis (27, 28).

The levels of α - and γ -tocopherols in raw cashew nut samples were found to be 0.29 and 1.10 mg/100 g of DM, respectively (Table 1). The α -tocopherol content of the present study was comparable with that of an earlier paper (0.26 mg/100 g of DM), but the γ -tocopherol level was found to be lower when compared to a previous paper on cashew nut (5.2 mg/100 g of DM) (23). Furthermore, the level of tocopherols in cashew nut samples of the present study was lower in comparison with the value given for other kinds of nuts (29), which may be due to the oxidation sensitivity of the tocopherols. It is well-known that tocopherols exhibit a protective role on lipid peroxidation of membrane lipids, lipoproteins, and depot fats (25) and, therefore, protect against atherosclerosis. The ability of vitamin E to induce apoptosis in tumor cells and modulate oncogenes is probably the reason for the low occurrence of cancer as a result of a vitamin E-rich diet, which was demonstrated in a variety of epidemiological studies (30). However, tocopherols are sensitive to light and oxygen, and the most important degradation reaction is the oxidation of tocopherols into tocopherylchinon.

The raw cashew nut kernels were found to contain a considerable amount of thiamin (1.08 mg/100 g of DM) (**Table 1**). This value was found to be higher when compared to an earlier report on the thiamin content of cashew nut (0.63 mg/100 g of DM), chestnut (200 μ g/100 g), peanut (900 μ g/100 g), hazelnut (390 μ g/ 100 g), coconut (61 μ g/100 g), kola nut (60 μ g/100 g), macadamia (280 μ g/100 g), almond (220 μ g/100 g), Brazil nut (1 mg/100 g), pecan (860 μ g/100 g), pistachio (690 μ g/100 g), and walnut (340 μ g/ 100 g) (23). Various enzymes of intermediary metabolism, including pyruvate dehydrogenase, α -ketoglutarate dehydrogenase, and transketolase, require thiamin pyrophosphate as an essential cofactor. Thiamin is thermolabile in a neutral or alkaline solution and sensitive to oxidation and ionizing radiation.

The results of quantification of the fatty acids such as stearic acid (4.96 g/100 g of DM), oleic acid (21.87 g/100 g of DM), and

				processed cashew nut kernels		
bioactive compound	raw cashew nut kernels	oil-bath roasting ^b	direct steam roasting	drying	open pan roasting	Flores hand-cracking
β -carotene (μ g/100 g of DM)	9.57 ± 0.42	$8.50 \pm 0.27^{**} (-11\%)$	$7.90 \pm 0.15^{**} (-17\%)$	$7.73 \pm 0.49^{**} (-19\%)$	$4.29\pm0.17^{**}(-55\%)$	$8.96\pm0.39^{*}~(-6\%)$
lutein (µg/100 g of DM)	30.29 ± 3.41	$26.33 \pm 1.43^{*} (-13\%)$	$26.68 \pm 3.00^{*} (-12\%)$	$24.35 \pm 2.29^{**} (-20\%)$	$12.77 \pm 1.26^{**} \ (-58\%)$	$29.16 \pm 2.67 \ (-4\%)$
zeaxanthin (µg/100 g of DM)	0.56 ± 0.22	$0.59\pm 0.19~(+4\%)$	$0.57\pm 0.17\ (+1\%)$	$0.44\pm 0.09~(-22\%)$	$0.20 \pm 0.06^{**} (-65\%)$	$0.71\pm0.23~(+20\%)$
α -tocopherol (mg/100 g of DM)	0.29 ± 0.04	$0.24 \pm 0.01^{*} (-14\%)$	$0.26 \pm 0.02 \ (-10\%)$	$0.25\pm0.02^{*}(-14\%)$	$0.20 \pm 0.03^{**} (-29\%)$	$0.24\pm0.04^{*}~(-16\%)$
γ -tocopherol (mg/100 g of DM)	1.10 ± 0.12	$0.96\pm0.04^{**}(-13\%)$	$0.98 \pm 0.04^{*} \ (-11\%)$	$0.85\pm0.03^{**}(-23\%)$	$0.93 \pm 0.07^{**} (-15\%)$	$1.01 \pm 0.06^{*} (-8\%)$
thiamin (mg/100 g of DM)	1.08 ± 0.12	$0.81 \pm 0.08^{**} (-25\%)$	$0.52 \pm 0.04^{**} (-52\%)$	$1.03 \pm 0.07 \; (-5\%)$	$0.51 \pm 0.03^{**} (-53\%)$	$1.07 \pm 0.10 (1\%)$
stearic acid (g/100 g of DM)	4.96 ± 0.67	$4.30 \pm 0.57^{*} (-13\%)$	$3.59\pm0.36^{**}~(-27\%)$	$5.37 \pm 0.51 \ (+8\%)$	$2.81 \pm 0.31^{**} (-43\%)$	$5.05\pm0.69~(+2\%)$
oleic acid (g/100 g of DM)	21.87 ± 1.77	$35.02 \pm 2.01^{**} (+37\%)$	$14.95 \pm 1.48^{**} (-32\%)$	$26.72 \pm 3.40^{**} (+18\%)$	$12.29 \pm 1.28^{**} (-44\%)$	$20.05\pm2.57~(-8\%)$
linoleic acid (g/100 g of DM)	5.55 ± 0.73	$4.98 \pm 0.79 \ (-10\%)$	$4.36 \pm 0.46^{*} (-21\%)$	$6.89 \pm 0.65^{**} (+44\%)$	$3.35 \pm 0.42^{**} (-40\%)$	$5.70 \pm 0.82 \ (+3\%)$
total lipids (g/100 g of DM)	66.21 ± 7.87	53.83 ± 3.21	50.74 ± 1.23	62.74 ± 3.29	52.80 ± 8.50	53.81 ± 9.52
a Values are mean and \pm SD of nin	e separate determinations $(n = 9)$. V	/alues given within parentheses with I	negative/positive sign indicate the pe	rcentage of reduction/increase compa	ared to control. *, $p < 0.05$; **, $p < 0.00$	01, significantly different from

Table 1. Levels of Various Bioactive Compounds in Raw and Differentially Processed Cashew Nut Kernels a

mean level of the control (raw unprocessed cashew). ^b The sunflower oil (Biskin) used for oil-bath roasting contained 3.5% stearic acid. 22.6% olicic acid, and 65.6% linoleic acid

linoleic acid (5.55 g/100 g of DM) in raw cashew nut samples are shown in Table 1. The level of stearic acid (4.96 g/100 g of DM) of the cashew nut was found to be higher when compared to previous reports on cashew nut (3.43 mg/100 g), chestnut (20 mg/100 g), peanut (1300 mg/100 g), hazelnut (940 mg/100 g), coconut (1100 mg/100 g), macadamia (1470 mg/100 g), almond (553 mg/ 100 g), pecan (1510 mg/100 g), pistachio (987 mg/100 g), and walnut (1370 mg/100 g) (23). The oleic acid content of cashew nut was higher than that of chestnut (981 mg/100 g), coconut (2100 mg/ 100 g), Brazil nut (18.5 g/100 g), and walnut (11.4 g/100 g), whereas the linoleic acid level of cashew nut was higher than that of coconut (680 mg/100 g) and macadamia (1740 mg/100 g) (23). Both oleic and linoleic acids are sensitive to oxidation, which

means that they must be protected from autoxidation. They need also to be protected against very high temperatures, because on heating cytotoxic products such as trans-isomers may arise (31). Linoleic acid was assessed as a precursor of arachidonic acid and eicosapentaenoic acid in the eicosanoid metabolism (32). It is also a component of phospholipids responsible for the structure and function of biological membranes. In recent years a variety of studies have demonstrated that the dietary polyunsaturated fatty acids have protective effects on metabolic syndrome diseases such as type 2 diabetes, hyperlipidemia, or cardiovascular disease. Equally promising results are now available regarding their positive effects on inflammatory diseases, cancer, and osteoporosis (33). In view of the afore-said facts and the rising obesity data, the industrialized nations are perfectly suitable for cashew nut consumption to prevent those chronic diseases due to the presence of appreciable levels of unsaturated fatty acids in cashew nuts.

The total fat content of cashew nut samples of the present study (66.21 g/100 g of DM) (**Table 1**) was comparable with that of an earlier paper on different kinds of nuts (47.1-76.2 g/100 g) (29). Our results confirms the previous observations that nuts are sources of high energy due to their total lipid content. Therefore, nuts have been treated with caution in most of the food pyramids. However, this point of view has now been reconsidered, and moderate nut consumption is recommended, because their unsaturated fatty acids have been shown to exert a cardioprotective function (34).

Oil-Bath Roasting. Cashew nuts processed by using an oil-bath roasting method showed a significant level of reduction of β -carotene (11%), lutein (13%), α -tocopherol (14%), γ -tocopherol (13%), thiamin (25%), and stearic acid (13%), which may be mainly due to the impact of high temperature of 200 °C (Table 1). Chen et al. (35) reported that long-chain carbon-carbon double bonds in carotenoids are sensitive to high temperature and light. Furthermore, Zepka and Mercadante (36) reported the degradation of carotenoids in cashew apple juice during heat treatment. However, in general, relatively higher levels of β -carotene, lutein, zeaxanthin, thiamin, oleic acid, and linoleic acid in cashew nut kernels processed by an oil-bath roasting method suggest that this treatment could be the mildest in proportion to other conventional processing methods. This method is very similar to that of the direct steam roasting process, but shorter in time due to high-temperature effect. The highly significant loss of α - and γ -tocopherol during oil-bath roasting was comparable to that of other conventional processing methods. The major difference of this method from all other batches was the highest content of oleic acid. However, there was no clear evidence that the oil used for oil-bath roasting had an impact on the fatty acid composition of the cashew nuts.

Steam Roasting. The cashew nut kernels obtained by direct steam roasting were significantly reduced in their content of bioactive substances due to the strong influence of temperature (120 °C) under steam pressure as compared to the unprocessed

samples (**Table 1**). Only the content of zeaxanthin was unaffected. Here the duration of heat is a critical factor for the loss of nutrients in addition to the higher level of temperature. Furthermore, the effect of direct steam roasting caused lower loss of carotenoids (12-17%) and tocopherols (10-11%), because in this treatment the temperature of 120 °C was reached with the pressure of 15 psi in the presence of moisture. However, a drastic loss of thiamin was noted (52%) during this treatment.

The analysis of fatty acids, which has been carried out in the direct steam roasted cashew nut kernels, showed that the content of individual fatty acids was significantly reduced in relation to that of unprocessed cashew nuts as a result of the influence of temperature and pressure. In comparison to the oil-bath roasted samples, a significantly lower concentration of fatty acids was noted in steam roasted cashew nuts, which might be due to a prolonged effect of temperature, and also lipid peroxidation may have been taken place as a result of increased pressure.

Drying. To investigate which stage of the oil-bath and steam roasting methods has the highest impact on the nutrient loss in cashew nut kernels, the samples were subjected to drying alone for 3 h at 75 °C, and the levels of bioactive compounds are shown in **Table 1.** The most similar β -carotene, lutein, and α -tocopherol contents of the dried batch sample when compared to the oil-bath and direct steam roasted batches suggested that the loss of these nutrients took place mainly during the 3 h drying at a relatively high temperature of 75 °C. This is confirmed by the fact that the concentrations of β -carotene, lutein, and α -tocopherol of the dried batch were significantly reduced by 19, 20, and 14%, respectively, when compared to raw cashew nut samples. However, the reason for the presence of a higher level of β -carotene content in the oil-bath roasted sample than in the dried sample remains unclear. Dried cashew sample was found to contain a significantly lower level of γ -tocopherol content (0.85 mg/100 g of DM) with respect to oil-bath roasted and direct steam roasted batches. Such a drastic level of reduction of γ -tocopherol (23%) in dried samples might be due to a nonuniform processing and storage of dried samples. This assumption is supported by the fact that, in general, the β -carotene, lutein, zeaxanthin, and α - and γ -tocopherol levels in the oil-bath and direct steam roasted batches were located just above the values of dried samples, which could be due to the heating of the samples in the presence of moisture in those treatments.

Similarly, a significantly lower level of thiamin concentration of oil-bath and direct steam roasted batches when compared to dried samples indicates that the loss of thiamin occurs mostly due to high roasting temperature and not due to the drying step. This is confirmed by the fact that the thiamin concentration of the dried samples was similar when compared to that of the raw cashew sample. It is very interesting to note that the content of individual fatty acids of the dried batch was higher than in the raw cashew nut. However, the ratio of the individual fatty acids was very similar and was not affected during processing of the samples by 3 h of drying at 75 °C. The highly significant loss of stearic acid during oil-bath roasting and maximum level of loss of all three unsaturated fatty acids during direct steam roasting when compared to dried cashew nuts were most likely due to the high processing temperatures of 200 or 120 °C.

Open Pan Roasting. The open pan roasting method exhibited by far the strongest reduction of bioactive compounds in cashew nut samples (**Table 1**). The open pan roasted cashew nut kernels exhibited a significantly lower concentrations of all the bioactive compounds when compared to raw as well as other processed batches. The highly significant decrease in all of the investigated compounds including β -carotene (55%), lutein (58%), zeaxanthin (65%), α -tocopherol (29%), γ -tocopherol (15%), thiamin (53%), stearic acid (43%), oleic acid (44%), and linoleic acid (40%) might be due to the direct action of fire, which also caused the scorching of a high proportion of cashew nuts. It is especially impressive to recognize that the γ -tocopherol concentration was reduced by at least 15%, whereas α -tocopherol was decreased by 29% in relation to raw cashew sample. Furthermore, open pan roasting is the only procedure that caused a significant loss of zeaxanthin.

Flores Hand-Cracking. Among the various shelling methods implemented in the present study, the Flores hand-cracking method recorded by far the least loss of bioactive compounds in relation to conventional methods (**Table 1**). Because the contents of lutein (29.16 μ g/100 g of DM), zeaxanthin (0.71 μ g/ 100 g of DM), stearic acid (5.05 g/100 g of DM), oleic acid (20.05 g/ 100 g of DM), and linoleic acid (5.70 g/100 g of DM) but above all the highly thermolabile thiamin content (1.07 mg/100 g of DM) of the cashew nut kernels obtained by Flores hand-cracking process were found to be comparable to that of raw samples without any significant difference, it can be considered as an extremely gentle method.

It is noticeable that the levels of zeaxanthin (20%), stearic acid (2%), and linoleic acid (3%) were slightly higher in the Flores hand-cracked cashew nuts in comparison to raw samples. In the Flores hand-cracking method, the 3 h drying at 45 °C has a very low impact on the contents of all the analyzed bioactive compounds. This is confirmed by the fact that an only meager level of reduction of β -carotene (6%) and γ -tocopherol (8%) in the handcracked cashew nut kernels appeared in comparison to a highly significant loss of these compounds during conventional processing methods. The cashew nut samples processed by the careful hand-cracking method were found to contain relatively high levels of antioxidants and resulted in a good protection of unsaturated fatty acids against autoxidation. Furthermore, the content of quantified fatty acids in the Flores hand-cracked cashew nut kernels was more or less comparable to that of raw sample, because 45 °C is the usual temperature applied to evaporate the solvents in fatty acid analysis. Therefore, it was expected that such a mild temperature causes no degradation of fatty acids present in the sample. However, the significant reduction of α - and γ -tocopherol was found to be similar to that of the other processing methods. In general, under the Flores hand-cracking method the cashew samples were processed in a very gentle manner, so that only a very low level of loss of bioactive compounds was noted when compared to other conventional processing methods.

The results of the present study indicate that the cashew nut kernels constitute a viable source of certain health-beneficial bioactive compounds. Among the various conventional shelling methods employed in the present study, the open pan roasting exhibited a drastic level of reduction of bioactive compounds in cashew nut samples due to the direct action of fire and therefore represents the most aggressive practice. Oil-bath roasting, which was followed by direct steam roasting, also caused significant losses of bioactive compounds in cashew nut samples. Furthermore, we could show that the reduction of thiamin and fatty acids of the cashew samples took place mainly during the oil-bath, steam, and open pan roasting processes and the significant decreases of tocopherols and carotenoids were found only during the subsequent 3 h drying at 75 °C. Alternatively, the recently developed Flores hand-cracking method exhibited only very low level of reduction of bioactive compounds in cashew nut kernels and is considered to be an extremely gentle and suitable shelling process. Hence, such a potential shelling process could be adapted at the industrial level for the retention of higher

levels of health-promoting/disease-preventing bioactive compounds in cashew nut kernels. The economic and sensory evaluation of differentially processed cashew nut samples deserves future research.

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