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Full Length Research Paper

Yield and quality of *Pycnanthus kombo* kernel butter as affected by temperature-moisture interactions

R. D. Nagre[1]*, W. O. Ellis[2] and I. Oduro[2]

[1]Department of Chemical Engineering, Kumasi Polytechnic, Kumasi, Ghana.
[2]Department of Food Science and Technology, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana.

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Preliminary studies of *Pycnanthus kombo* butter using petroleum ether as extractant have been reported. There is however, lack of data on the chemical composition of the kernels as well as an optimized method for expressing the butter from the kernels. This study discusses the chemical composition of the kernels of *P. kombo* as well as the optimisation of temperature-moisture interactions for Kombo butter expression using the low-pressure manual press and also the physicochemical properties of the butter extracted at the optimum conditions. A butter content of 74.13% was obtained by proximate analysis. Both processing temperature and paste moisture content significantly (p>0.05) affected the butter extraction efficiencies (BEE). Butter extraction efficiency generally increased at constant temperature as paste moisture content was increased. The study showed high butter extraction efficiencies under three conditions with a peak value of 86.1% at 100°C and 8.0% paste moisture. Physicochemical properties of the butter obtained at the three optimum conditions were not significantly (p>0.05) different and thus, showed no relative changes in butter quality. Hence, using a low-pressure manual (mechanical) press, a much higher yield of Kombo kernel butter can be obtained at a temperature of 100°C and paste moisture content of 8.0% than other conditions investigated in this study.

Key words: *Pycnanthus angolensis (Pycnanthus kombo),* Kombo butter, butter extraction efficiency, physicochemical properties.

INTRODUCTION

The demand for low priced common edible fats and oils have been on a steady increase in the markets of most countries. This is partly due to the competition between the industrial requirement for oil and their use for edible purposes. There is thus the need to find alternative sources of oils for industrial purposes, so as to overcome this competition. *Pycnanthus angolensis*, synonymously known as *Pycnanthus kombo* belongs to the myristicaceae family, it is commonly called an African nutmeg (Mapongmetsem, 2007). The trees grow widely in West and Central Africa (Eckey, 1954). In Ghana, the tree is known in the local Twi and Fante dialects as "Otie". They are widely distributed locally in most forest zones in the southern part of the country (Acquaye, 1999). The tree flowers between October and May and fruits from September to April. Parts of the plant are reported to have several medical applications (Akendengue and Louis, 1994; Agyare et al., 2009; Onocha and Ali, 2011). Kombo butter is documented to have industrial and medicinal applications (U.S. Patent No. 6,489,494). Its potential for the treatment of arthritis and for combating inflammations in the joints has been reported (Simon et al., 2005). Kombic acid, derived from Kombo butter functions as an anticancer and cholesterol-lowering agent (U.S. Patent No. 6,713,512). The crude fat is reported to exhibit anti-oxidising properties especially in the stabilisation of foods against rancidity,

*Corresponding author. E-mail: nagrerobert@yahoo.com. Tel: +233-243419741.

colour and odour development. It can also be used as stabilizers in cosmetic products (Lok et al., 1983). Other uses of Kombo butter include soap and candle production. Eckey (1954) and Irvine (1961) reported the fat content (determined by solvent extraction) of Kombo kernels as 54 to 62% and 54%, respectively.

Extraction methods for fats and oils vary depending on the source. The main traditional method for fats and oils extraction in Ghana has been the aqueous extraction technique, involving direct heating of the raw material and the use of large volumes of water. This is often time consuming and labour intensive. This process also affects the quality of the final product and results in low yields. The main industrial methods for extracting oils from oilseeds are the mechanical expression technique, direct solvent extraction and pre-press solvent extraction (Salunkhe et al., 1992). In most developing countries, the extraction of oil is carried out mostly by the traditional (expeller) method. The direct solvent extraction technique, even though efficient, is expensive and less appropriate for raw materials that have high percentage of oil. The mechanical expression on the other hand is more economical for high oil materials (>20%) relative to the solvent extraction process even though it is associated with relatively high oil losses (Gunstone et al., 1986). Optimizing the operational parameters such as moisture content and temperature can enhance the efficiency of oil extraction. Studies have been reported on the extraction of Kombo fat using petroleum ether; however, there is lack of data on the mechanical expression of the fat. This study was carried out to determine the chemical composition of the Kombo nuts and optimise conditions for extraction of Kombo kernel butter based on the mechanical expression technique

MATERIALS AND METHODS

Source of raw materials

The nuts used for this study were collected between January and February 2004 from a cluster of about ten trees of *Pycnanthus angolensis*, in Twifo Praso, in the Central Region of Ghana. The nuts were obtained from fully ripe fruits which split-open and fell to the ground.

Sample preparation for extraction and analyses

The nuts were sun-dried for about two weeks at an average temperature of 27.5°C and relative humidity of 77%, and shelled to obtain the kernels. The kernels were sorted out by hand-picking to get rid of defective (spoilt/cracked) ones. The quartering sampling method was then used to select 150 g kernels sample (from a bulk of about 60 kg kernels) for the proximate analysis of chemical composition.

The remaining kernels, about 60 kg quantity were used in determining the oil extraction efficiency. Kernels used for the determination of chemical composition were stored under refrigerated conditions for one week before use; whereas, those used for mechanical expression were stored at room temperature for a month prior to extraction whilst the determination of chemical composition of the kernels was carried out.

Preliminary extraction

The kernels were milled to fine particle sizes (66.75% passing through a 1.18 mm standard sieve) using a plate mill prior to extraction.

A preliminary study was carried out using 8 × 3 factorial design: consisting of 8 temperature levels (55, 60, 65, 70, 80, 90, 100 and 110°C) against 3 moisture levels (4.1, 6.0 and 8.0%). The extraction at each temperature-moisture interaction was done in triplicates. For each replicate, 350 g of milled kernels sample was accurately weighed using an electronic balance and mixed (wetted up) with a calculated (based on initial moisture content of sample) quantity of warm water at 55 to 60°C to obtain the desired moisture level in the paste sample. The desired percentage moisture content (X) in milled Kombo kernel sample was calculated using:

$$X = 100(aw + y)/(w + y)$$

where, w = weight of sample, y = water added to sample to achieve desired moisture content, and a = initial moisture fraction in sample.

The samples were placed in linen cloth-bags and heated in a thermostatically controlled oven for 2 h at the selected temperature and the oil was then expressed using a low pressure manual press. The oil obtained was dried at 103°C for 6 h, weighed and the yield calculated.

The pressed cake was also dried at 103°C in the oven for 12 h. A 10 g portion of the powdered cake was then sampled (using quartering method) and the residual oil content in the cake was determined with petroleum ether as solvent using the soxhlet extraction method. The butter extraction efficiency (BEE) was calculated using the formula:

$$BEE = 100[1- (R(seed)/R(cake))]$$

where, R(seed) is the ratio of non-oil components to oil in the seed, R(cake) is the ratio of non-oil components to oil in the cake (Stafford, 1997; Tillekeratne and Ranasinghe, 1997; Obeng et al., 2010).

Optimum extraction

Based upon the preliminary results, 5 × 6 factorial design consisting of 60, 70, 80, 90 and100°C (based on average of high yields / BEEs) and 4.1, 6.0, 8.0, 10, 12, and 14% combined temperature-moisture interactions on extraction were further investigated. The yields and BEEs were then calculated and the respective graphs against moisture levels were then plotted.

Proximate composition of Kombo nuts

The recommended methods of the Association of Official Analytical Chemists (AOAC, 1990) were adopted for the determination of, ash, crude protein, crude fat, and crude fibre. Results are shown in Table 1.

Moisture content

Moisture content was determined in triplicate using 5.0 g of the milled seeds. The sample was dried to constant weight in a thermostatically controlled oven at 103°C for 12 h. It was then

Table 1. Chemical composition of Kombo kernels.

Component	Percent content (D)
Moisture	4.10 (0.16)
Crude fat	74.13 (2.21)
Crude protein	9.02 (0.32)
Crude fibre	8.35 (0.03)
Ash	2.11 (0.24)
Carbohydrate	2.29 (0.25)

Values were obtained in triplicates and the average values determined. D: Absolute deviation from mean.

cooled in a desiccator and weighed. The loss in weight expressed as a percentage of the initial weight of sample gives the percent moisture content on wet basis (AOAC Method 925.40, 1990).

The same procedure was also used to determine the moisture contents of the pressed fats obtained at different paste moisture levels (4.1 to 14.0%) on wet basis.

Crude fat

Crude fat was determined in triplicate based on the soxhlet extraction method. For each replicate, 2.0 g of the dried and milled sample was weighed into a muslin thimble. The thimble was then placed in a soxhlet extractor and the fat was extracted using petroleum ether (B.P 40 to 60°C). The flask containing the fat and residual solvent was placed on a water bath to evaporate the solvent followed by a further drying in the oven at 103°C for 30 min. It was then cooled in a dessicator and weighed. The fat obtained was expressed as a percentage of the initial weight of the sample (AOAC Method 948.22, 1990).

Crude fibre

The defatted sample (from crude fat determination) was transferred to a 750 ml Erlenmeyer flask and approximately 0.5 g of asbestos was added. 200 ml of boiling 1.25% H_2SO_4 was added and the flask was immediately set on a hot plate and a condenser was connected to it. The content was left to boil within 1 min and the sample was digested for 30 min. At the end of the 30 min, the flask was removed and the contents filtered through a linen cloth in a funnel and subsequently washed with boiling water until the wash was no longer acidic. The charge and asbestos were washed back into the flask with 200 ml boiling 1.25% NaOH solution. The condenser was again connected to the flask and the content was boiled for exactly 30 min. It was then filtered through the linen cloth and thoroughly washed with boiling water. The residue was then transferred to a crucible with a spatula and the remaining particles washed off with 15 ml ethanol into the crucible. It was then dried in an oven overnight and cooled in a desiccator and weighed. The sample in the crucible was then ignited in a furnace at 600°C for 30 min, cooled and reweighed. The loss in weight gave the crude fibre content. Values were determined in triplicate and each expressed as a percentage of the initial weight of the sample (AOAC Method 962.09, 1990).

Ash content

2.0 g of the dried and milled seeds was weighed into a previously ignited and weighed crucible. The crucible and content were then placed in a furnace at 600°C for 2 h. The sample was allowed to cool in the furnace to 250°C. The crucible and the ash were then transferred into an oven at 100°C for 30 min cooling and finally cooled to room temperature in a desiccator and weighed. The ash content was determined in triplicate. The weight of the ash was expressed as a percentage of the initial weight of the sample (AOAC Method 923.03, 1990).

Protein content

The crude protein was determined in triplicate using the macro Kjeldahl method. 2.0 g of the dried and milled nuts was weighed into a digestion flask and 0.5 g of selenium catalyst was added. 25 ml of conc. H_2SO_4 was added and the flask was shaken to mix the contents. The flask was then placed on a digestion burner for 8 h and heated until the solution turn green and clear. The sample solution was then transferred into a 100 ml volumetric flask and made up to the mark with distilled water. 25 ml of 2% boric acid was pipetted into a 250 ml conical flask and 2 drops of mixed indicator (20 ml of bromocresol green + 4 ml of methyl red) solution added. 10 ml of the digested sample solution was then introduced into Kjeldahl flask. The condenser tip of the distillation apparatus was then dipped into the boric acid contained in the conical flask. The ammonia in the sample solution was then distilled into the boric acid until it changed completely to bluish green. The distillate was then titrated with 0.1 NHCl solution until it became colorless. The percent total nitrogen and crude protein were calculated using a conversion factor of 6.25 (AOAC Method 955.04, 1990).

Carbohydrate content

The percent carbohydrate content of the Kombo kernels was obtained from the aforementioned results by difference. That is:

Percentage carbohydrate = 100 - (moisture + crude fat + crude protein + crude fibre + ash)%

Physicochemical properties

From the results of the extraction trials, the temperature-moisture combinations which gave high average BEEs or yields of butter were selected and the physicochemical properties determined.

Refractive index

The refractive value was measured in triplicate at 60°C. Several drops of the melted fat sample were placed on the lower prism of an Abbe refractometer (which was also adjusted to the same temperature as the sample). The prisms were closed and tightened firmly with the screw head, ensuring that the sample came to the same temperature as the instrument. The instrument was adjusted until the most distinct reading possible was obtained and the refractive index noted.

Specific gravity

The specific gravity was determined using the specific gravity bottle. The bottle was then placed in a water bath maintained at 25°C and filled with distilled water. It was removed, wiped dry and weighed. The bottle was emptied, dried and placed in a water bath at 60°C and allowed to attain a temperature of 60°C for about 30 min. The bottle was then refilled with the melted fat sample. It was then cleaned and wiped completely dry and weighed. The specific gravity was calculated using the formula (AOCS method Cc 10a-25, 1993):

$$Specific\quad Gravity\quad at\ 60\ /\ 25\ ^{0}C\ =\ \frac{(weight\quad of\quad bottle\quad +\ oil\ )\ -\ (weight\quad of\quad bottle\quad)}{w\left[1\ +\ 0.000025\quad (t\ -\ 25\ )\right]}$$

w = weight of water at 25°C, t = temperature = 60°C.

Free fatty acids (FFA)

The fat was melted at 50°C, well mixed and 1.4 g of it was weighed into a flask containing 15 ml of hot neutralized alcohol and 0.4 ml of phenolphthalein indicator was then added. The content was titrated with 0.5 N NaOH. The free fatty acid value was calculated (as oleic acid) using the formula (AOCS Method Ca 5a-40, 1993):

$$FFA(as\ oleic\ (\%)) = \frac{V \times N \times 28.2}{W}$$

Where N = normality of NaOH solution, v = volume (ml) of NaOH solution, W = weight of oil sample.

Acid value

5 g of the melted fat was dissolved in a well mixed neutral solvent consisting of 25 ml of diethyl ether and 25 ml 95% ethanol. It was then titrated with aqueous 0.1 N NaOH using 1 ml of phenolphthalein indicator solution and shaking constantly until a pink color that persisted for at least 15 s was obtained. The acid value was then calculated using the formula (Kirk and Sawyer, 1991):

$$Acid\ value mg\ KOH\ /\ g\ of\ sample = \frac{56.1N \times V}{W}$$

where, N = normality of NaOH solution, v = volume (ml) of NaOH solution, W = weight of oil sample.

Peroxide value

5 g of the fat was weighed into a 250 ml-conical flask with a glass stopper. 30 ml of 3:2 v/v glacial acetic acid-chloroform solvent was added and swirled to dissolve the sample. 0.5 ml of saturated KI solution was added. The solution was left in the dark with occasional shaking for exactly 1 min, and 30 ml of distilled water added immediately. The mixture was titrated with 0.1 N sodium thiosulphate using 0.5 ml of starch indicator solution. A blank was also performed at the same time. The test was done in triplicate. The peroxide value was calculated using the formula (AOCS Method Cd 8-53, 1993):

$$\text{Peroxide}\quad \text{value}\ = \frac{1000(V_1 - V_2) \times N}{W}$$

where, N = normality of $Na_2S_2O_3$ solution, v_1 = volume (ml) of $Na_2S_2O_3$ solution used in test, v_2 = volume (ml) of $Na_2S_2O_3$ solution used in blank, W = weight of oil sample.

Statistical analyses

The butter extraction efficiencies as well as results of the physicochemical properties of the butter samples were statistically analysed using ANOVA, Genstat Software version 7.0. The software basically, calculated the average values, compared them and established significant ($p<0.05$) differences or otherwise. Graphs were plotted using Microsoft Excel spreadsheet software.

RESULTS AND DISCUSSION

Composition of Kombo nuts

The weight per seed was found to range from 1.15 to 1.21 g. Respective average diameter and length of the seeds after sorting out were 1.21 and 2.18 cm. The average composition with respect to seed and shell of the nut were 82.9 and 17.1%, respectively. The results of the study showed that the crude fat content of the Kombo seeds was 74.13%, categorizing Kombo seeds as high yielding oilseeds. The protein and fiber contents of the Kombo kernels were however very low and thus, the kernels are not a good source of proteins. The ash content of the Kombo seeds was 2.11% and this was within the range of ash content for nuts (0.8 to 3.5%) reported by Aurand et al. (1987). The carbohydrate content (2.29%) was low, confirming previous report that Kombo seeds are starch free (Irvine, 1961).

Yield and butter extraction efficiency

The results of the extraction trials in Table 2 indicated that BEE generally increased at constant temperature as paste moisture content was increased. The BEEs at 55°C were significantly ($p<0.05$) lower than the corresponding BEEs at the other temperatures. The BEEs at 110°C were also observed to be lower than the corresponding BEEs at 100°C. Significantly ($p< 0.05$) lower BEEs were also obtained at the moisture level of 4.1% for all temperatures between 55 and 110°C compared to BEEs at other moisture levels (6.0 and 8.0%).

The significantly low BEEs at 55°C may be due to the high viscosity of oil, since oil generally seeps through the cake structure less easily at high viscosities (Fellows, 1992). The low BEEs at moisture level 4.1% compared to higher moisture levels was probably due to the low moisture availability in the paste to displace the oil from the surface of the “meat” particles of the oil containing material. The significantly ($p< 0.05$) lower BEEs at 110°C relative to the corresponding values at 100°C may be due to high moisture loss resulting in low moisture availability in the paste; leading to a decrease in the amount of oil displaced from the “meat” particles of the oil containing material.

Figures 1 and 2 generally showed a sharp increase in butter yield or BEE from a moisture level of 4.1 to 6.0% followed by a gradual increase to a maximum and then a slight decrease. This trend was quite similar to BEEs reported for shea butter extraction under similar conditions (Obeng et al., 2010). Maximum butter extraction efficiencies (the peaks of the graphs) 83.6, 83.0, and 86.1% were obtained at temperature of 60°C

Table 2. Percent butter extraction efficiencies (% BEE) at different temperature- moisture interactions (Preliminary extraction).

Temperature (°C)	Moisture content (%)		
	4.1	6.0	8.0
55.0	63.90	74.47	76.13
60.0	65.80	80.90	83.60
65.0	66.83	80.17	80.17
70.0	67.03	81.67	81.30
80.0	68.40	80.80	81.40
90.0	70.83	81.23	81.83
100.0	75.20	80.53	86.10
110.0	74.73	78.27	79.77
SED		(0.480)	
LSD		(0.966)	

LSD: Least significant differences of means (5%). SED: Standard errors of differences of means. Values with differences greater than the LSD value are significantly different ($p < 0.05$). Values were obtained in triplicates and the average values determined using ANOVA, Genstat software version 7.0.

and 8.0% moisture content, 80°C and 12%, and 100°C and 8.0%, respectively. The maximum yields or BEEs at the aforementioned conditions was due to optimum temperature-moisture interactions. Thus, at the right moisture content and temperature, the oil droplets unite to form a continuous phase and flow out resulting in maximum yield or BEE.

Therefore, to obtain maximum yield of Kombo fat, the extraction should be carried out at the optimum temperature-moisture conditions, particularly at a temperature of 100°C and moisture content of 8.0%. However, this differed from the optimum extraction temperature of 60°C and moisture content of 12% established by Obeng et al. (2010) for shea butter extraction, at which an extraction efficiency of 68.5% was obtained. The differences in optimum extraction conditions and extraction efficiencies were possibly due to the differences in chemical compositions between the Kombo kernel and shea kernel. The BEE is a measure of the efficiency of the extraction process, thus, obtaining BEEs ranging from 63.9 to 86.1% indicates that the technique of extraction is efficient.

Residual moisture content in butter

There was a slight increase in moisture content of the mechanically expressed Kombo butter with an increase in the moisture level of the paste (milled Kombo nuts) as shown in Figure 3. However, the relatively low increase in residual moisture in the butter compared to the quantity of water added to paste prior to pressing was an indication that much of the water added to the paste was retained in the cake. The additional moisture might have displaced the oil component from the “meat” particles of the paste leading to an increase in oil yield. Hence, a significant increase in yield or BEEs particularly between moisture levels of 4.1 and 6.0% compared to levels between 6.0 and 14.0% (Figures 1 and 2).

Butter extraction efficiencies (BEEs) and physicochemical properties

Four conditions, A, B, C and D which gave relatively high BEEs/yields were selected (Table 3) and physico-chemical analysis carried out on the oils from these conditions. The results showed that the oil samples obtained under the four conditions were not significantly ($p>0.05$) different in terms of specific gravity and refractive index. Although, sample C had a significantly ($p<0.05$) lower % FFA and acid value than the rest, its peroxide value was significantly ($p<0.05$) higher. The peroxide values of samples A, B, and C were not significantly ($p>0.05$) different.

Selection of optimum condition of extraction

In terms of processing conditions, BEE at 100°C and 4.1% moisture (A) was significantly ($p<0.05$) lower whilst the BEE at 100°C and 8.0% (D) was significantly ($p<0.05$) higher than that the BEEs at 60°C and 8.0% and 80°C and 12%. The processing condition of 100°C and 8% moisture level was considered most suitable for Kombo butter extraction relative to the other conditions due to the following reasons; (1) The temperature-moisture interactions of 100°C and 8% gave a significantly ($p< 0.05$) higher BEE than the other conditions. (2) The viscosity of oil at 100°C was lower than the viscosity at

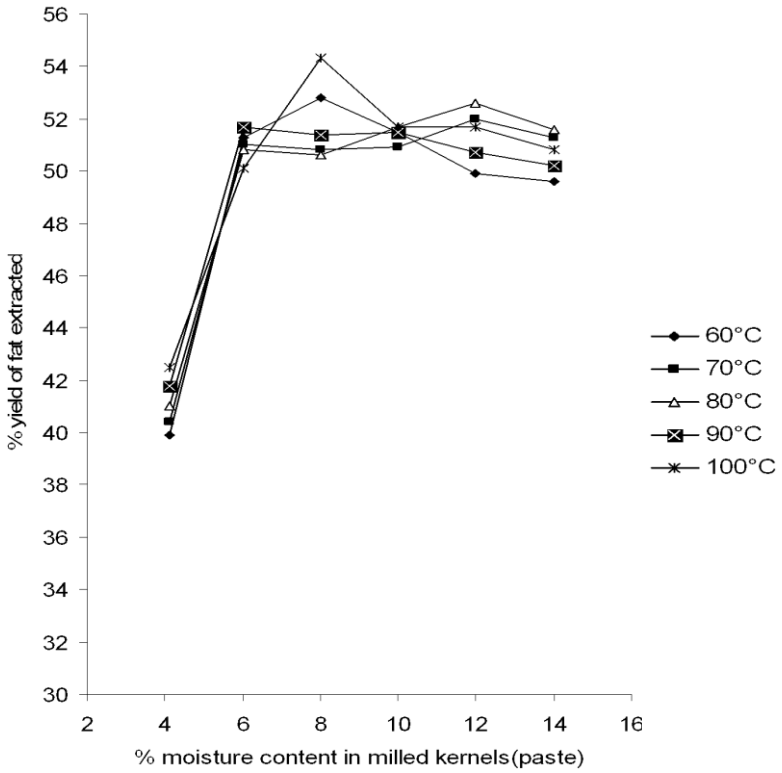

Figure 1. Average yield of pressed Kombo butter at varying moisture and temperature conditions.

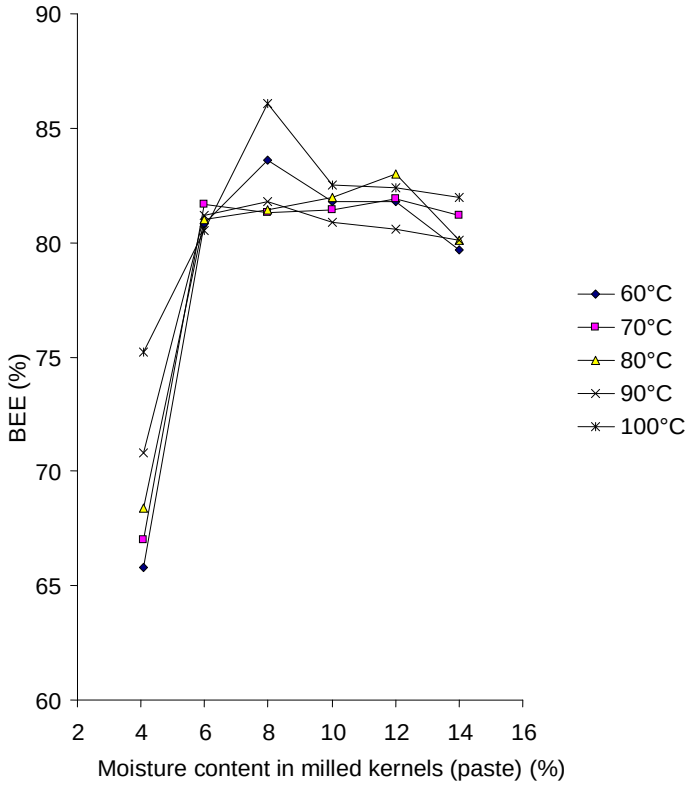

Figure 2. Average butter extraction efficiency (%BEE) at varying moisture and temperature conditions.

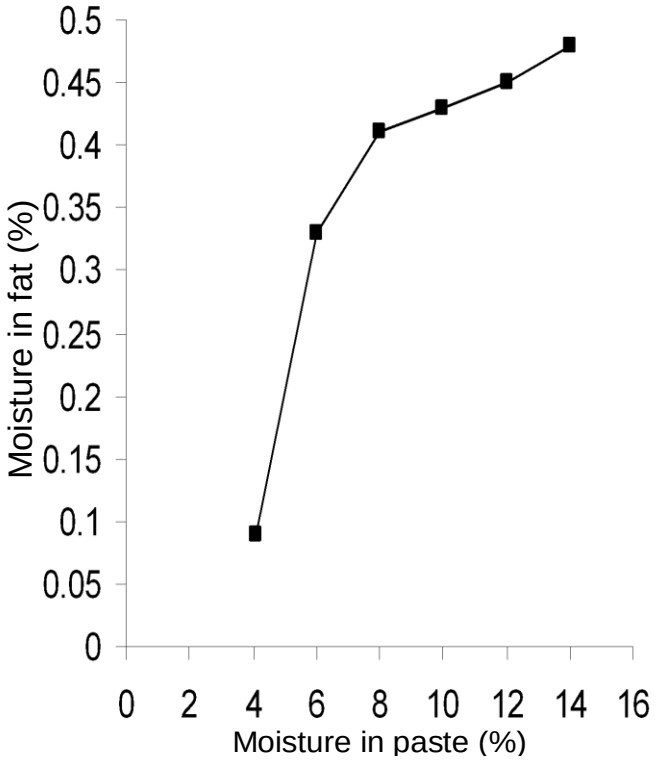

Figure 3. Effects of moisturising of Kombo paste on the moisture content of the pressed crude fat.

Table 3. Percent butter extraction efficiencies (% BEEs), yield and physicochemical properties of butter from the four selected samples.

Parameter	Samples*				LSD 5%	SED
	60°C at 8% (A)	80°C at 12% (B)	100°C at 4.1% (C)	100°C at 8% (D)		
%Yield	52.83^{a}	52.63^{a}	42.50^{b}	54.27^{c}	0.782	0.339
%BEE	83.57^{a}	83.06^{b}	75.20^{c}	86.10^{d}	0.261	0.113
Specific gravity (60°C/25°C)	0.9265^{a}	0.9283^{b}	0.9270^{ab}	0.9269^{ab}	0.001	0.001
%FFA (calc. as oleic acid)	17.77^{a}	17.88^{b}	17.32^{c}	17.84^{ab}	0.098	0.043
Acid value (mEq/kg fat)	36.50^{a}	36.64^{b}	36.00^{c}	36.55^{ab}	0.091	0.039
Peroxide value	8.82^{a}	8.88^{b}	9.0^{c}	8.9^{b}	0.038	0.017
Refractive index at 60°C	1.465^{a}	1.465^{a}	1.466^{a}	1.466^{a}	0.002	0.001

*Values with different superscripts (a, b, c, d) in the same row are significantly different. Values were obtained in triplicates and the average values determined using ANOVA, Genstat Software version 7.0. A: Butter sample obtained at 60°C and 8% paste moisture content. B: Butter sample obtained at 80°C and 12% paste moisture content. C: Butter sample obtained at 100°C and 4.1% paste moisture content. D: Butter sample obtained at 100°C and 8% paste moisture content.

the lower extraction temperatures. (3) The refractive index, specific gravity and percent FFA of sample D (100 and 8% moisture) were not significantly ($p>0.05$) different from that of samples A and B. (4) Even though sample C has the least %FFA, its peroxide value which is a measure of oxidative rancidity was significantly ($p<0.05$) higher than the other samples. Thus, the optimum processing condition for mechanical expression of Kombo butter extraction is 100°C and 8% paste moisture.

Conclusion

The study has shown that Kombo seeds (kernels) are high yielding (74.13%) oil seeds and can serve as a commercially rich source of fat. It has been established in this study that in extracting Kombo butter using the low pressure manual screw press, both temperature and paste moisture content of Kombo kernels have significant ($p<0.05$) effect on butter extraction efficiency or yield.

Although, the physicochemical properties of the three samples (A, B, and D) from the moisturized paste were similar, they were however significantly different from fat samples obtained from unmoisturized paste. For small scale processing of Kombo seeds using a low pressure manual press, optimum yield of butter is achieved at a paste (milled kernels) temperature of 100°C and moisture content of 8.0%.

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