

Physico-Chemical Properties of Seed Oil from Papaya (*Carica papaya*) and the Kinetics of Degradation of the Oil During Heating

¹G. Bouanga-Kalou, ¹L. Matos, ^{1,4}J.M. Nzikou, ¹F.B. Ganongo-Po, ¹K.E. Malela,

¹M. Tchicailat-Landou, ¹R.M. Bitsangou, ³Th. Silou and ⁴S. Desobry

¹ENSP-UMNG, Laboratory of Food Physicochemistry and Biotechnology, Pole of Excellence in Nutrition and Food, P.O. Box 69 Brazzaville-Congo

² Faculty of Science of Health -University Marien Ngouabi P.O. box 69 Brazzaville-Congo

³Multi-field Team of Research in Food and Nutrition, Center IRD,
P.O. Box 1286 Pointe-Noire (Congo)

⁴ENSAIA-INPL, Laboratory of Engineering and Biomolecule, 2, avenue de la forêt de Haye,
54505 Vandoeuvre-lès-Nancy (France)

Abstract: The aim of the study was to investigate the physico-chemical properties and the kinetics of degradation of papaya seed oil during heating. The seed is a good source of oil (32%). The physical properties of the oil extracts showed the state to be liquid at room temperature and indicated that the oil had refractive index, 1.4680; the peroxide value, 0.05 (meq O₂/kg oil); free fatty acid, 1.2%; iodine value, 72.78 and saponification value, 198.5. Gas liquid chromatography technique has been developed for identification and quantitative determination of total unsaturated and saturated fatty acids shows that the crude oil had 80.44 and 19.56%, respectively. DSC indicates the presence of two components in oil extracted. The first peak at low melting point appears at -21.53°C (H_f = +1.81 J/g) and the second peak appears to -9.77°C (H_f = +41.53 J/g). The degradation kinetic of the oil was also investigated. The thermal oxidation of the double bonds of the oil showed a first-order thermal oxidation kinetic and the Arrhenius plot yielded a straight line with a slope equivalent to activation energy of 7.752 KJ/mol. There is the possibility of considering the seed as feed supplement and its oil for industrial application.

Key words: *Carica papaya* seed, degradation, DSC, essential fatty acid, kinetics

INTRODUCTION

Seed oils are important sources of nutritional oils, industrial and pharmaceutical importance. The characteristics of oils from different sources depend mainly on their compositions and no oil from a single source can be suitable for all purposes (Mohammed and Jorg, 2003). The study of these constituents is important for their effective uses.

Seed oils are known to deteriorate when processed inadequately with the principal decomposition reaction being oxidation. Oxidation of seed oil occurs through a free radical mechanism, initially characterised by the emergence of a sweetish and unpleasant odour which becomes progressively worse until it attains a characteristic smell of rancid fat (Gouveia De Souza *et al.*, 2004). Heating is one of the most commonly used methods of food preparation in the home and industries and prolong use of oil for this purpose causes change in its physical and chemical properties (Morette and Fett, 1998).

Under the influence of temperature, fat and oils are susceptible to oxidation primarily leading to the formation of hydroperoxides. Due to their high reactivity, these hydroperoxides especially at high temperatures rapidly react with secondary oxidative products e.g. aldehydes, ketones, peroxides, hydrocarbons as well as cyclic compounds that exhibits very different possible toxic or carcinogenic properties (Kowalki, 1995). The products formed during this oxidative process can be determined by chemical analysis and one of the frequently used tests employed to predict the quality of seed oils is the determination of peroxide value and iodine value. A number of seed oils have been characterised but the vast majority have not been adequately evaluated. Papaya (*Carica papaya*) is a plant that grows wild in many parts of the tropics. Papaya is important for its fruit and it is cultivated mainly for this purpose. Papaya is also grown for papain production. The seeds of papaya fruits are generally discarded. However, in order to make a more efficient use of papaya, it is worth investigating the use of the seeds as a source of oil.

The extraction and use of vegetable oils has for centuries played an important role in the manufacture of a large number of industrial products and food items (Puangsri *et al.*, 2005).

The chemical composition of *Carica papaya* seed oil in Congo Brazzaville, as well as their kinetics of degradation, has not yet been studied. Therefore, the objectives of this study were to investigate the physico-chemical properties of seed oil from *Carica papaya* and the kinetics of degradation of the oil during heating.

MATERIALS AND METHODS

This study was led to the laboratory of Engineering and Biomolecule of the ENSAIA-INPL, Vandoeuvre - lès-Nancy (France) for the period of Sept. 20, 2010 to Dec. 10, 2010.

Materials: *Carica papaya* seeds, grown in Diata district (Brazzaville-Congo), were bought from Total market of Bacongo. The seeds were dried in an oven at 60°C. The dried seeds were stored at -10°C until required for analysis.

Methods:

Oil extraction: For solvent extraction (soxhlet method), 50g of papaya seed flour were placed into a cellulose paper cone and extracted using light petroleum ether (b.p 40-60°C) in a 5-l Soxhlet extractor for 8 h (Pena *et al.*, 1992). The oil was then recovered by evaporating off the solvent using rotary evaporator Model N-1 (Eyela, Tokyo Rikakikal Co., Ltd., Japan) and residual solvent was removed by drying in an oven at 60°C for 1 h and flushing with 99.9% nitrogen. All experiments were done in triplicates and the mean and standard deviations were calculated.

Physico-chemical analysis of crude oil:

Thermal behaviour: The thermal property of the oil samples was investigated by differential scanning calorimetry using a Perkin-Elmer Diamond DSC (Norwalk, USA). The instrument was calibrated using indium and zinc. The purge gas used was 99.99% nitrogen with a flow rate of 100 mL/min and a pressure of 20 psi. Sample weights ranged from 5-7 mg and were subjected to the following temperature program: Frozen oil sample was heated at 50°C in an oven until completely melted. Oil sample was placed in an aluminium volatile pan and was cooled to -50°C and held for 2 min, it was then heated from -50 to 50°C at the rate of 5°C/min (normal rate) (Che Man and Swe, 1995), and held -50°C isothermally for 2 min and cooled from -50 to 50°C at the rate of 5°C/min. The heating and cooling thermograms for the normal and the fast (hyper DSC) scan rates were recorded and the onset, peak, and offset temperatures were tabulated. These values provide information on the temperature at which

the melting process starts, the temperature at which most of the TAG have melted, and the complete melting temperature of the oil, respectively.

Refractive indices: The refractive indices of the oil (at room temperature) were determined with Abbe refractometer (Pearson, 1976) and the specific gravity measurement (also carried out at room temperature) using specific gravity bottle. The state and color of the oil were noted using visual inspection at room temperature. The mean molecular mass was estimated from the relation $(56/SV) \times 1000$ (Akintayo and Bayer, 2002).

Chemical analysis: Determinations for peroxide, iodine, and saponification values, and free fatty acid contents were carried out using Pena *et al.* (1992) standard analytical methods. The fatty acid composition was determined by conversion of oil to fatty acid methyl esters prepared by adding 950 μ L of n-hexane 50 mg of oil followed by 50 μ L of sodium methoxide using the method of Cocks and Van Rede (1966). The mixtures were vortex for 5 s and allowed to settle for 5 min. The top layer (1 μ L) was injected into a gas chromatograph (Model GC- 14A, Shimadzu Corporation, Kyoto, Japan) equipped with a flame-ionisation detector and a polar capillary column (BPX70 0.25), 0.32 mm internal diameter, 60 m length and 0.25 μ m film thickness (SGE Incorporated, USA) to obtain individual peaks of fatty acid methyl esters. The detector temperature was 240°C and column temperature was 110°C held for 1 min and increased at the rate of 8 °C/min to 220°C and held for 1 min. The run time was 32 min. The fatty acid methyl esters peaks were identified by comparing their retention time with those of standards. Percent relative fatty acid was calculated based on the peak area of a fatty acid species to the total peak area of all the fatty acids in the oil sample. The minerals were determined by atomic absorption spectrophotometry. One gram samples, in triplicate, were dry, ashed in a muffle furnace at 550°C for 8 h until a white residue of constant weight was obtained. The minerals were extracted from ash by adding 20.0 mL of 2.5% HCl, heated in a steam bath to reduce the volume to about 7.0 mL, and this was transferred quantitatively to a 50 ml volumetric flask. It was diluted to volume (50 mL) with deionised water, stored in clean polyethylene bottles and mineral contents determined using an atomic absorption spectrophotometer (Perkin-Elmer, Model 2380, USA). These bottles and flasks were rinsed in dilute hydrochloric acid (0.10 M HCl) to arrest microbial action which may affect the concentrations of the anions and cations in the samples. The instrument was calibrated with standard solutions.

Proximate analysis: Proximate analysis was carried out as described by the Association of Official Analytical Chemists (AOAC, 1995).

Heat treatment: Thermal degradation of *Carica papaya* oil was carried out by heating the oil up to 200°C for a period of 0-240 min. The peroxide value, refractive index and the iodine values were determined respectively at 100, 150 and 200°C using the Association of Official Analytical Chemists method.

Kinetic calculations: A general reaction rate expression for the deterioration kinetic can be written as follows (Ramaswami *et al.*, 1989; Van, 1996):

$$-d[C]/dt = k[C]^m$$

where 'C' is the quantitative value of the concentration of the molecule under consideration, 'k' is the reaction rate constant and 'm' is the order of the reaction. For first order reaction where m = 1 the equation can be written as:

$$\ln ([C_t] / [C_o]) = -kt$$

where [C_o] is the concentration of the reactants under consideration at time zero and [C_t] is the concentration of the reactants at time 't'. Arrhenius relationship of the reaction rate to temperature is generally given as:

$$K = A_o \exp (-E_a / RT)$$

where 'E_a' is the activation energy of the reaction, 'R' is the gas constant, 'T' is absolute temperature and 'A_o' is a pre-exponential constant. Each experiment was performed in triplicates and the average values were taken for the parameters determined. Kinetic data were analysed by regression analysis using MS Excel 8.

Statistical analysis: Values represented are the means and standard deviations for three replicates. Statistical analysis was carried out by Excel Version 8.0 software. Significance was defined at p<0.05.

RESULTS AND DISCUSSION

Proximate analysis: The oil content is high, it was found to be 32±1.1% (Table 1) which shows that the processing of the oil for industrial or edible would be economical. The obtained fat content agree with the value (28.3%) reported by Marfo *et al.* (1986). slight variation in oil yield may be due to the differences in variety of plant, cultivation climate, ripening stage, the harvesting time of the seeds and the extraction method used. State of papaya seed oil at room temperature is liquid.

Physico-chemical properties of oil:

Physical properties:

Differential Scanning Calorimetry (DSC): DSC is suitable to determine these physical properties. The results of thermal analysis of oils are presented in

Table 1: Physical and chemical properties of papaya (*Carica papaya*) seed

Properties	Obtained values
Colour	slightly orange
Oil ^a (%)	32±1.1 ^a
PV	0.05±0.24 ^a
FFA (as % oleic acid)	1.2±0.2 ^a
IV (wijs)	72.78±0.22 ^a
Saponification value	198.5±0.21 ^a
Mean molecular mass	282.12
Refractive index (at 25°C)	1.4680
E _a (KJ/mol)	7.752
State at room temperature	Liquid

Means for the determined values in the same row followed by the same superscript; letter are not significantly different (p<0.05); ^a: Oil = weight of extracted oil x 100/weight of seed; PV: Peroxide Value; FFA: Free Fatty Acid; IV: Iodine Value

Table 2: Melting behaviour of papaya (*Carica papaya*) seed oil using different scan rates. Experimental conditions: temperature program set at -50°C for 10 min, rising to 50°C at rate of 5 °C/min

Thermogram	5 °C/min
Peak 1 [°C]	-21.53
H _r [J/g]	+1.81
Peak 2 [°C]	-9.77
H _r [J/g]	+41.53

Table 3: Relative percent composition of fatty acid in papaya (*Carica papaya*) seed oil

Fatty acid	Determined values
	Soxhlet
Palmitic acid (C16:0)	15.05±0.18 ^a
Stearic acid (C18:0)	4.51±0.32 ^b
Oleic acid (C18 : 1)	76.64±0.23 ^a
Linoleic acid (C18 :2)	3.81±0.42 ^a
Saturated acid	19.56
Unsaturated acid	80.44

Means for the determined values in the same row followed by the same superscript letter; are not significantly different (p<0.05)

Table 2. The obtained peaks were asymmetries and may indicate the presence of two components. The first peak at low melting point appears at -21.53 °C (H_r = +1.81 J/g). This peak corresponds to triglycerides formed by Poly Unsaturated Fatty Acids (PUFA). The second melting point is at -9.77°C (H_r = +41.53 J/g). This is suggesting the presence of mixed triglycerides groups with different melting points.

Chemical properties: The chemical properties of oil are amongst the most important properties that determines the present condition of the oil. Free fatty acid and peroxide values are valuable measures of oil quality. The iodine value is the measure of the degree of unsaturation of the oil. The low free fatty acids content is indicative of low enzymatic hydrolysis. This could be an advantage as oil with high free fatty acids develop off flavour during

Table 4: Effect of heating on PV and IV of papaya (*Carica papaya*) seed oil

Temp. (°C)	Time (mins)	PV (mg/g)	IV (mg/iodine)	RI (25°C)
100	30	0.39±0.12	72.59±0.42	1.4302
	60	0.80±0.24	72.34±0.20	1.3702
	120	1.20±0.18	72.03±0.16	1.3010
	180	1.63±0.22	71.82±0.21	1.2501
	240	2.09±0.31	71.59±0.12	1.1701
150	30	0.68±0.14	72.47±0.28	1.3590
	60	1.02±0.27	71.87±0.12	1.3180
	120	1.38±0.32	71.17±1.20	1.2572
	180	1.70±0.18	70.67±0.31	1.1851
	240	2.05±0.20	70.17±0.25	1.1650
200	30	1.28±0.15	72.23±0.24	1.2410
	60	1.49±0.24	71.75±0.25	1.2191
	120	1.68±0.20	71.14±0.20	1.1943
	180	1.98±0.23	70.79±0.15	1.1851
	240	2.20±0.12	70.29±0.22	1.1810

Values are mean ± standard deviation of triplicate determinations; RI: Refractive Index

storage (Bailey, 1954). The refractive index reflects the degree of unsaturation and chain length. The value obtained here (1.4692) is expected of on oils with low iodine value and the presence of Oleic acid fatty in the proportion observed (Table 1). The saponifiable percentage is 198,5 and this percentage is in conformity with the value (197%) reported by Marfo *et al.* (1986).

The saponification value is high and this suggests the use of the oil in production of liquid soap, shampoos and lather shaving creams. The peroxide Table 4 also shows that the iodine value of the oil decreases as it was heated over a period of time. This suggests the loss of unsaturation in the fatty acids of the triacylglycerols. This loss of unsaturation was also reflected by the decrease in the value of refractive index, value is 0.05 ± 0.24 mg/g oil, this value is lower than that expected of rancid oil which ranges from 20.00 to 40.00 mg/g oil (Oderinde and Ajayi, 1998). This shows that the oil is not rancid and considered stable (Ajayi *et al.*, 2002). The refractive index which is the ratio of the velocity of light in a medium was found to be 1.4680 (at room temperature). This refractive index is an indication of the level of saturation of the oil.

Fatty acid composition: The major saturated fatty acids in *Carica papaya* seed oil were palmitic (15.05%) and stearic (4.51%) acids and the main unsaturated fatty acids are linoleic (3.81%) and oleic (76.64%) acids (Table 3). The oil sample of *Carica papaya* contained saturated and unsaturated acids (19.56% and 80.44%) respectively. The proportion of unsaturated fatty acids was greater than the saturated fatty acids. The results obtained are in agreement with those of the literature (Marfo *et al.*, 1986).

Kinetic data for the degradation of *Carica papaya* oil:

The rate of production of peroxide in *Carica papaya* oil increases as the temperature increases as shown in Table 4. This shows that the prolong heating of this oil makes it to undergo thermal degradation resulting in oxidative rancidity, formation of hydroperoxides and other products of degradation that can liberate volatile compounds. since double bonds (unsaturation) increase

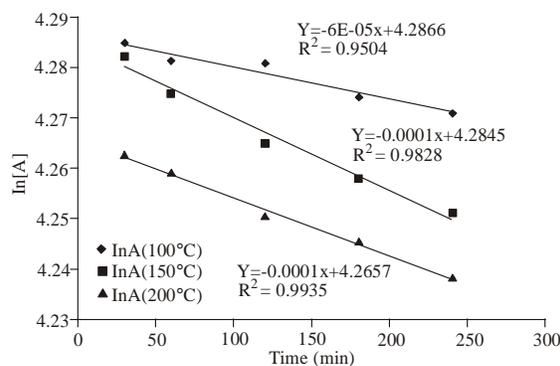


Fig. 1: Graph of rate of change in concentration of iodine value against time, [A] Represents iodine value

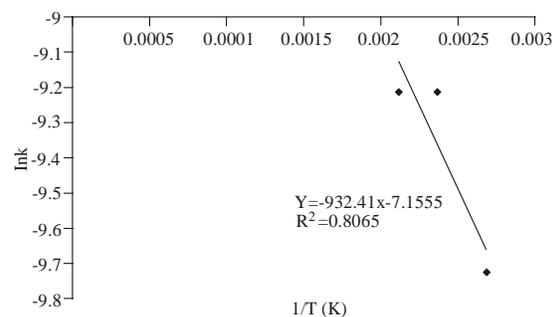


Fig. 2: Arrhenius plot for oxidation rate of the double bonds of fatty acids in papaya (*Carica papaya*) seed oil

the refractive index of organic compounds by reducing the angle of refraction in relation to the angle of incidence and the higher the unsaturation the greater the effect of reducing the refraction angle (Oderinde *et al.*, 2009).

In order to obtain the reaction rate constant ('k'), a first order degradation of the oil was presumed (Labuza and Riboh, 1982). Accordingly 'ln [C]_t/[C]₀' was plotted against 't'(time) from which rate constant 'k' was calculated from the slope of the line (Anthon and Barrett, 2002) as shown in Fig. 1. A correlation

Table 5: Kinetic parameters for degradation of papaya (*Carica papaya*) seed oil

Kinetic parameters	100°C	150°C	200°C
$k_1 (s^{-1})$	0.00006	0.0001	0.0001
$t_{1/2} (s)$	11550	6930	6930

coefficient >0.9 in all the cases confirmed the assumption of the degradation (loss of unsaturation) to follow the first order kinetic. The half life for the degradation was calculated from the rate constant as $'0.693/k'$ and is given Table 5.

Fig. 2 shows the Arrhenus plot of $\ln k$ versus $1/T$ for the reduction of unsaturation (Iodine value) in *Carica papaya* oil. The linear nature of the plot obtained gave the activation energy of the reaction to be 7.752 KJ/mol (Table 1).

CONCLUSION

This study showed that the *Carica papaya* seed is a good source rich in oil. *Carica papaya* seed oil was obtained from the seed with good yield (32%), allowing the possibility of economical exploitation, and its fatty acid composition is comparable to that of some conventional oils.

The thermal oxidation of *Carica papaya* oil follows a first order reaction. This oxidation is temperature and time dependent. The process quality assurance of this oil can be monitored using iodine value and peroxide value.

ACKNOWLEDGMENT

We thank Carole Jeandel and Carole Perroud, technicians at the Liblio laboratory for their assistance during the trials.

REFERENCES

Akintayo, E.T. and E. Bayer, 2002. Characterization and possible uses of *plukenatia conophora* and *Adenopus breviflorus* seed and seed oil. Bores. Technol., 85: 95-97.

Ajayi, I.A., F.A. Dawodu, K.O. Adebowale and R.A. Oderinde, 2002. Chemical composition of Pentaclethra macrophylla seed and seed oil grown in Nigeria. Riv. Ital. Sostanze Grasse., 74: 183-185.

Anthon, G.E. and D.M. Barrett, 2002. Kinetic parameter for the thermal inactivation of quality-related enzymes in carrots and potatoes. J. Agric. Food Chem., 50: 4119-4125.

AOAC, 1995. Official Methods of Analysis. 14th Edn., Association of Official Analytical Chemist, Arlington, VA, 67: 1-45.

Bailey, E.A., 1954. Industrial Oil and Fat Products. 3rd Edn., Interscience Publishers, London.

Che Man, Y.B. and P.Z. Swe, 1995. Thermal analysis of failed-batch Palm oil by differential scanning calorimetry. J. Am. Oil Chem. Soc., 72(12): 1529-1532.

Cocks, L.V. and C. Van Rede, 1966. Laboratory Handbook For Oil and Fats Analysis. Academic Press, London, pp: 88.

Gouveia De Souza, A., J.C. OLivera Santos, M.M. Conceicao, M.C. Dantas Silva and S. Prasad, 2004. A thermoanalytic and kinetic study of sunflower oil. Braz. J. Chem. Eng., 21(2): 265-273.

Kowalki, B., 1995. Determination of oxidative stability of edible vegetable oil by Pressure differential scanning calorimetry. Thermo chim. Acta, 250: 197-205.

Labuza, T.P. and D. Riboh, 1982. Theory and application of Arrhenius kinetics to the prediction of nutrient losses in foods. Food Technol., 36: 66-74.

Marfo, E.K., O.L. Oke and O.A. Afolabi, 1986. Chemical composition of papaya (*Carica papaya*) seeds. Food Chem., 22: 259-266.

Mohammed, R.F. and M. Jorf-Thomas, 2003. Determination of the lipid classes and fatty acid profile of Niger seed (*Guizotia abyssinica* Cass). Phytochem. Anal., 14: 366-370.

Morette, E. and R. Fett, 1998. Oils and Food Technology in Industrial Station of Purification. Sao Paulo, Varele.

Oderinde, R.A. and I.A. Ajayi, 1998. Metal and oil characteristics of *Terminalia catappa*. Riv. Ital. Sostanze Grasse, 75: 361-362.

Oderinde R.A., I.A. Ajayi and A. Adewuyi, 2009. Characterization of seed and seed oil of *Hura crepitans* and the kinetics of degradation of the oil during heating. EJEAFChe, 8(3): 201-208.

Pearson, D., 1976. The Chemical Analysis of Foods. 7th Edn., ChurchillLivingstone, Edinburgh, U.K., pp: 488-496.

Pena, D.G., R.G.L. Anguiano and J.J.M Arredondo, 1992. Modification of the method 1 AOAC (CB-method) for the detection of aflatoxins. Bull. Environ. Contam. Toxicol., 49: 485-489.

Puangri, T., S.M. Abdulkarim and H.M. Ghazali, 2005. Properties of *Carica papaya* seed oil. J. Food Lipids, 12: 62-76.

Ramaswami, H.S., F.R. Van De Voort and S. Ghazada, 1989. Analysis of TDT and Arrhenius method for handling process and kinetic data. J. Food Sci., 54: 1322-1326.

Van Boekel, M.A., 1996. Statistical aspect of kinetic modeling for food science problems. J. Food Sci., 61: 477-485.