INNSPUB

International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print), 2222-5234 (Online)
http://www.innspub.net
Vol. 8, No. 6, p. 93-102, 2016

RESEARCH PAPER OPEN ACCESS

Comparison of some physicochemical properties of oil extracted from *Ricinodendron heudelotii* (Bail.) kernels by UV spectrophotometer

Horliane Ghomdim Nzali[1], Clergé Tchiegang[1*], Bernard Sandjon[2], Marc Meurens[3]

[1]*Laboratoire des Bio-Procédés, Unité de Recherche en Biochimie-Technologie Alimentaires et Nutrition, Université de Ngaoundéré, Ngaoundere, Cameroon*

[2]*Laboratoire Phytorica, Douala, Cameroon*

[3]*Institut des Sciences de la Vie, Université Catholique de Louvain, Croix du Sud 2 (bte 8), Louvain la Neuve, Belgium*

Key words: *Ricinodendron heudelotii*, Oil, Physicochemical characteristics α-eleostearic acid, UV, GC,

http://dx.doi.org/10.12692/ijb/8.6.93-102 Article published on June 20, 2016

Abstract

In order to contribute to research on nutrients with therapeutic value, the oil rate, fatty acid composition and the physicochemical and quality characteristics of *Riconodencron heudelotii* kernels oil obtained by pressing and solvent extraction were determined. The KOMET Screw Oil Expeller was used. To achieve this, UV Spectrophotometry and Gas Chromatography (GC) were used to quantify the amount of α-eleostearic acid (ESA) in the oils. The results indicate that the extraction temperature has an influence on the rate of extraction. Thus the extraction rate for cold pressing (30°C) was 34.51±1.98% compared to hot pressing (80°C) with 44.44±2.46%. But for the quality of the oil, cold pressing gives the indices of peroxide and acid below standard. There is no significant difference ($p < 0.05$) of α-tocopherol according to extraction mode. The proportion of ESA of oil obtained by cold pressing (30°C) was the highest (49.02±0.64%). The UV confirmed that the press had no influence on the structure of the ESA. The calibration test for the determination of ESA by UV spectrophotometry was obtained with a Regression coefficient (R) of 0.95. The UV spectrophotometry technique is a rapid and less expensive method for determining the quality and the quantity of ESA in *R. heudelotii* oil than GC. The cold pressing almonds of *R. heudelotii* gives an oil with good physicochemical properties and is rich of α-eleostearic acid. Moreover eleostearic acid is a conjugated linolenic acid and these acids have benefic effects for the management of metabolic diseases.

* **Corresponding Author:** Clergé Tchiegang ✉ tclerge@yahoo.fr

Introduction

Ricinodendron heudelotii is a specie which belongs to the Euphorbiaceae family. It grows in tropical Africa from Guinea to Angola, East Africa and Madagascar. In Cameroon, it is found in the equatorial forest and even in the savannas of West perished forest (Vivien and Faure, 1996). It is known as *njansan* in Cameroon. Kernels are used as spices in many dishes in Cameroon (Tchiegang *et al.*, 1997; Mosso *et al.*,1998). These kernels are characterized by their high oil content (45- 55 %) and crude proteins (55.37 % in the defatted cake). The fatty acids composition of this oil indicates a high α- eleostearic acid content (52 % of total fatty acids) (Kapseu and Tchiegang, 1995; Tchiegang *et al.*, 1997).

α- eleostearic acid (9c 11t 13t -18:3) is a conjugated linolenic acid (CLNA). This conjugated acid has been shown to have a cytotoxic effect on cultured human tumor cells (Suzuki *et al.,* 2001; Igarashi and Miyazawa, 2005; Yasui *et al.*, 2005), to inhibit carcinogenesis (Kohno *et al.*, 2002; Tsuzuki *et al.,* 2004; Tsuzuki and Kawakami, 2008), and alter the lipid metabolism in animals (Yang *et al.*, 2005; Yamasaki *et al.*, 2006; Koba *et al.*, 2007; Lam *et al.*, 2008). Tsuzuki *et al.* (2004); Tsuzuki *et al.* (2003) and Yuan *et al.* (2009) showed that CLNA (α-eleostearic) is converted to Conjugated Linoleic Acid (CLA) in liver of rats and mice. Although the CLNA are known for their nutritional, pharmaceutical and nutraceutical values, they are also very sensitive to oxidation. For extraction, it is important to develop an extraction method which give a good oil yield but also protects fatty acid sensitivity, due to the fact that extraction conditions could influence the oil quality (Orthoeffer, 1995; Kone, 1998).

In addition, to identify the presence of fatty acids, the chromatographic techniques that are often used include Gas Chromatography (GC) , Gas chromatography–Mass Spectrometry (GC-MS), High Performance Liquid Chromatography (HPLC). These techniques are very long and expensive (Poulli *et al.*, 2006; Bernuy *et al.*, 2009; El-Abassy *et al.*, 2009). Considering the nutraceutical and therapeutic importance attached to the α-ESA.. However, the presence of conjugated double bonds in their molecules confer spectral properties that may allow their detection by the spectrophotometric techniques such as UV. Because UV spectrophotometry is a technique that could be used to control this parameter online.

This technique gives an excellent indication of the total conjugated double bonds (Reaney *et al.*, 1999). UV spectrophotometry has been used in the determination of conjugated linoleic acids (CLA) in cow milk (Bernuy *et al.*, 2008); in the rapid determination of olive oil oxidative Stability (Cayuela Sánchez *et al.*, 2013). This shows the importance of UV in the rapid characterization of oils.

The aims of this research was to evaluate the variation of chemical properties of *R. heudelotii* oil using different methods of extraction and to evaluate the potential of UV method for the determination of α-elostearic acid in oil.

Materials and methods

R. heudelotii almonds were collected in Douala (Littoral, Cameroon).

Commercial α- eleostearic acid (C18:3-9c,11t,13t), and the fatty acid methyl esters mixtures standards were purchased from Larodan fine chemicals (Limhamn, Sweden).

Extraction of R. heudelotii

Extraction of oil from almonds was performed by pressing. Soxhlet solvent extraction with hexane was used as standard. Extraction rates and yields were calculated. The extraction yield is defined as the amount of oil (in grams per 100 g dried almonds) while the extraction rate is the ratio of the amount of oil extracted by pressing on the amount of available oil extracted by solvent (Wiemar and Altes, 1993).

Solvent extraction

Solvent extraction was performed with hexane using Soxhlet method (IUPAC, 1979).

Pressing extraction

Cold pressing

R. heudelotii almonds were pressed at 30 ± 3 °C using a KOMET screw oil expeller DD85G, number 200666 manufactured in 1991 (Germany). Before extraction, almonds were crushed in a mill brand KOMET, type Crusher, number 200666 manufactured in 1991 (Germany). KOMET oil expeller features a special cold pressing system with a single conveying screw to squeeze the oils from various oil-bearing seeds.

The machines operate on a gentle mechanical press principle that does not involve mixing and stirring of the almonds. Fig. 1 shows the diagram of laboratory KOMET oil expeller for pressing operation. One kilogram of *R. heudelotii* almonds was filled into the feeding hopper and pressed by screw press with nozzle sizes of 6 mm and with rotational speed of 21 rpm. The extracted oil was kept away from light, placed in a dark container (wrapped with aluminum foil) and stored in a chiller (4°C). After 48 h, the oil was filtered using a sieve to remove other fine particles and weighed. The sample vials were purged with nitrogen and stored at -18°C.

Hot pressing

Samples were pressed at 80 ± 3°C using nozzle sizes: 6 mm, diameter; 21 rpm rotational speed. At this temperature proteins coagulate and facilitate the release of all the fat. The screw press was first run for 30 min. by heating it using electrical resistance-heating ring attached around the press head. A digital thermometer was used to measure the temperature between the heating ring and the press head. Pressed oils were stored away from light in a dark container as previously described before been filtered after 48 h. The sample vials were purged with nitrogen and stored at -18°C.

Physicochemical characteristics

Physico-chemical characteristics of oils (acid, peroxide, iodine, chlorophylls and carotenoids values) were performed using UICPA standard methods (IUPAC, 1979). The density of oils was performed with pycnometer.

A quantitative determination of α-tocopherol was established by colorimetry method (Kivçak and Mert, 2001).

Gas chromatography (GC)

The quantification of fatty acids methyl esters (FAME) of the oils was also determined by gas chromatography. Briefly, the FAME were performed by treatment of a 500 mg sample wit h 10 mL of KOH 3 in methanol during 1 h at 70°C. The addition of 4 mL of HCl (1.2 M) in methanol and further incubation during 15 min at the same temperature completed the methyl esterification. The extraction of FAME was done after the addition of 20 mL of hexane and 10 mL of demineralized water (Focant *et al.*, 1998). After extraction of methyl ester, the chromatograph using to identify fatty acids is Thermo Finnigan, type TRACE GC (Milan, Italy). The capillary column used was RESTEK Rt- 2560 (100 m length, 0.25 mm internal diameter, 0.20 µm film thickness) (Supelco, Bellefonte, PA, USA). Gas chromatography conditions were a flow rate of 1mL/min of He with an initial temperature of 140°C held for 5 min. The column temperature was then increased to 250°C at a rate of 2°C/min, and then held at 250°C for 15 min. Fatty acid peaks were identified using pure methyl ester standards.

UV spectrophotometry

The spectra values of various oils were performed between 200 and 500 nm using UV Spectrophotometer S2000, Ocean Optics (Dunedin, Florida). These spectra were compared to the spectrum of standard of α- eleostearic acid. Samples were diluted with pure hexane and analysed in triplet with a quartz vial of 3 ml and 1 cm optical path. The spectra were acquired using a software based OII 32 (S2000, Ocean Optics, Dunedin, Florida). For each sample, ten spectra were averaged to provide the final spectrum.

Data treatment

All experiments were done in triplicate. Results were expressed as means ± standard deviation. All results were analysed using a one-way analysis of variance.

Duncan's Multiple Range test was performed to evaluate differences between the results using the software Statgraphics 5.0 (1998). Differences between means were considered to be significant at $p < 0.05$. Predictive equation was developed using regression from the software Sigma Plot.

Results and discussion

Oil content of almonds, extraction yield and *extraction* rate

From fig. 2, almonds of R. heudelotii has 51.03 ± 0.23 % of oil. The extraction yield is higher for hot pressing (22.68 ± 1.26) compared to cold extraction (17.61 ± 1.01). It is the same for the extraction rates which varied from 44.44 ± 2.46 % to 34.51 ± 1.98 % respectively for hot and cold extraction. Indeed, heating the almonds reduces the affinity of oil for the solid particles of almonds, which leads to an easy release of the oil during pressing (Norris, 1982). The same trends were observed by Tchiegang *et al.* (2004) on almonds of *R. heudelotii* treated dry and wet in 90 ° C. But the values obtained in this study (44.44%) are lower than those reported by Tchiegang *et al.* (2004) (58.47%). The difference may be due to press model. Tchiegang *et al.* (2004) used a hydraulic vertical screw press; compared to the horizontal electric screw press used in this study.

Table 1. Some chemical characteristics of oils from *R. heudelotii* kernels with respect to extraction methods.

	Extraction methods		
	Hot	Cold	Hexane
Densities (g/ml)	1.007 ± 0.005^{b}	1.002 ± 0.003^{b}	0.990 ± 0.002^{a}
Acid values	0.90 ± 0.05^{c}	0.70 ± 0.01^{a}	0.80 ± 0.01^{b}
Peroxide value (meq O_2 per kg of oil)	16.56 ± 0.37^{b}	14.43 ± 0.40^{a}	14.79 ± 0.50^{a}
Iodine values	103.46 ± 0.79^{a}	105.38 ± 0.37^{a}	104.65 ± 0.39^{a}
Carotenoid values (µg/g)	8.43 ± 0.95^{a}	8.53 ± 0.25^{a}	8.57 ± 0.57^{a}
Chlorophyll values (µg/g)	0.50 ± 0.01^{b}	0.45 ± 0.01^{a}	0.40 ± 0.06^{a}
α-tocopherol values (mg/g)	$0,31 \pm 0.01^{a}$	$0,33 \pm 0.01^{a}$	$0,33 \pm 0.01^{a}$

Values on the same line with different superscripts are significantly different at $p < 0.05$ (Duncan's test).

Physicochemical characteristics of oils

Some physicochemical characteristics of extracted oils are given in table 1. The oil extracted with hexane has the lowest density (0.990 ± 0.002). The values found were lower than those obtained by Tchankou Leudeu (2006) (0.806 ± 0.001) on oil from a plant of the same species.

Oil extracted by the cold press had the lowest value acid (0.70 ± 0.01) compared to that oil extracted with hexane (0.80 ± 0.01) and hot press (0.90 ± 0.05). This difference is due to the fact that high temperature using hexane extraction and hot pressing could lead to the degradation of α- eleostearic acid of *R. heudelotii*. The unsaturated fraction (over 50 %) in the almonds of *R. heudelotii* oil makes it a very fragile and therefore thermosensitive (Aboubakar Dandjouma, 2004). The recorded values are similar to those found by Tchiegang *et al.* (2005) on the oils of almond R. heudelotii extracted with hydraulic vertical screw press. But the acid values obtained were below the recommended limit for edible oil as recommended by Codex Alimentarius (1992) which is 4. Cold pressing gave oil containing less free fatty acids than the hot pressing and the control.

The peroxide value was measured to assess the oxidation levels of oils. There was a significant difference ($P < 0.05$) between the oils obtained by pressing compared to the control extracted with hexane value. The peroxide value, obtained for the cold pressing was lower (14.43 ± 0.07 meq O_2 / kg of oil) compared to the maximum value of peroxide allowed for virgin oils which is 15 meq O_2 / kg of oil

(CODEX, 1999).

There is no a significant change in iodine value. The iodine value (103 g/100 g) (table1), the index of the degree of fat unsaturation, was consistent with values noted by Granados *et al.* (2003).

All obtained parameters prove the good quality of the oil, indicating that the cold pressing of *R. heudelitii* oil could be stored for longer time without any additional purification.

Carotenoids values are not affected by the extraction method. Hot pressing provided oil with more chlorophyll. the free radical chain breaking reaction and protecting lipids from oxidation by quenching the anion-radical superoxide (Schneider, 2005).

Table 2. Fatty acids composition (% of total fatty acids) of R. heudelotti oils with respect to the extraction methods

Fatty acids	Extraction methods		
	Hot	Cold	Hexane
Palmitic (C16 : 0)	6.86 ± 0.30^{b}	5.46 ± 0.16^{a}	6.51 ± 0.17^{b}
Stearic (C18: 0)	6.92 ± 0.31^{a}	6.67 ± 0.11^{a}	6.77 ± 0.38^{a}
Oleic (9c- 18 : 1)	7.50 ± 0.14^{a}	7.36 ± 0.28^{a}	7.43 ± 0.13^{a}
Linoleic (9c 12c– 18 : 2)	32.27 ± 0.57^{a}	31.17 ± 0.67^{a}	31.54 ± 0.44^{a}
α-eleostearic (9c 11t 13t- 18 : 3)	45.95 ± 0.37^{a}	49.02 ± 0.64^{c}	47.64 ± 0.66^{b}
Arachidonic (C20 :0)	0.14 ± 0.01^{b}	0.13 ± 0.01^{a}	0.14 ± 0.01^{b}
α-linolenic (9c 12c 15c– 18 : 3)	0.19 ± 0.20^{a}	0.17 ± 0.08^{a}	0.17 ± 0.08^{a}
Eicosapentaenoic (5c 8c 11c 14c 17c- 20 : 5)	0.17 ± 0.01^{a}	0.16 ± 0.08^{a}	0.16 ± 0.06^{a}
Total saturated	13.92	12.26	13.42
Total monounsaturated	7.50	7.36	7.43
Total Polyunsaturated	78.58	80.52	79.51

Values on the same line with different superscripts are significantly different at $p < 0.05$ (Duncan's test).

The oils are one of the major sources of tocopherols (Bernal *et al.*, 2011). In the oil of *R. heudelotii*, the content of α-tocopherol ranged from 0.31mg/g to 0.33mg/g showing the richness of the oil of α-tocopherol. But The extraction method does not affects α-tocopherol content.

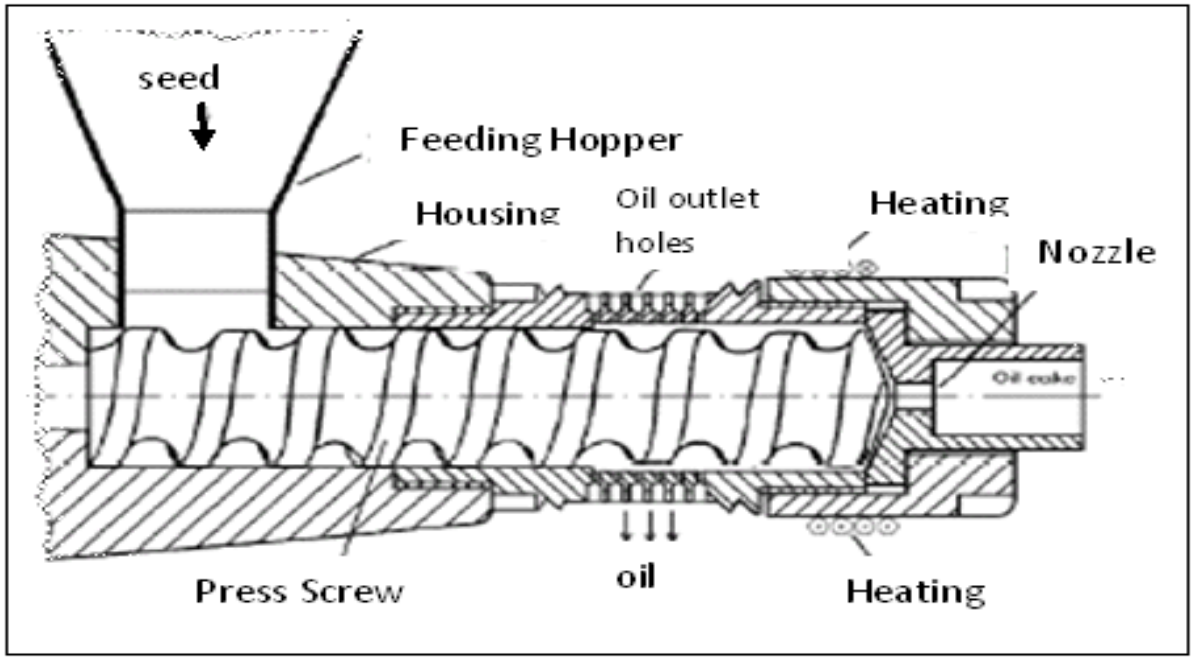

Fig. 1. Hole cylinder press of a KOMET oil expeller

Chromatographic analysis

The quantification of the fatty acids of the *R. heudelotii* oil was possible with the use of GC.

The fatty acids compositions of oils obtained from different methods of extraction are presented in Table 2. In general, hot pressing leads to increase of the total saturated fatty acids compare to cold pressing. The increase in these saturated fatty acids was compensated by a decrease in relative percentage of polyunsaturated fatty acid. It should be noted that the greatest variation was noted with α- eleostearic acid which is the major acid found in these oils. Cold pressing increased relatively the value of α-eleostearic acid (49.02 ± 0.64 % of total fatty acids**)** compared to hot pressing (45.95 ± 0.37 % of total fatty acids).

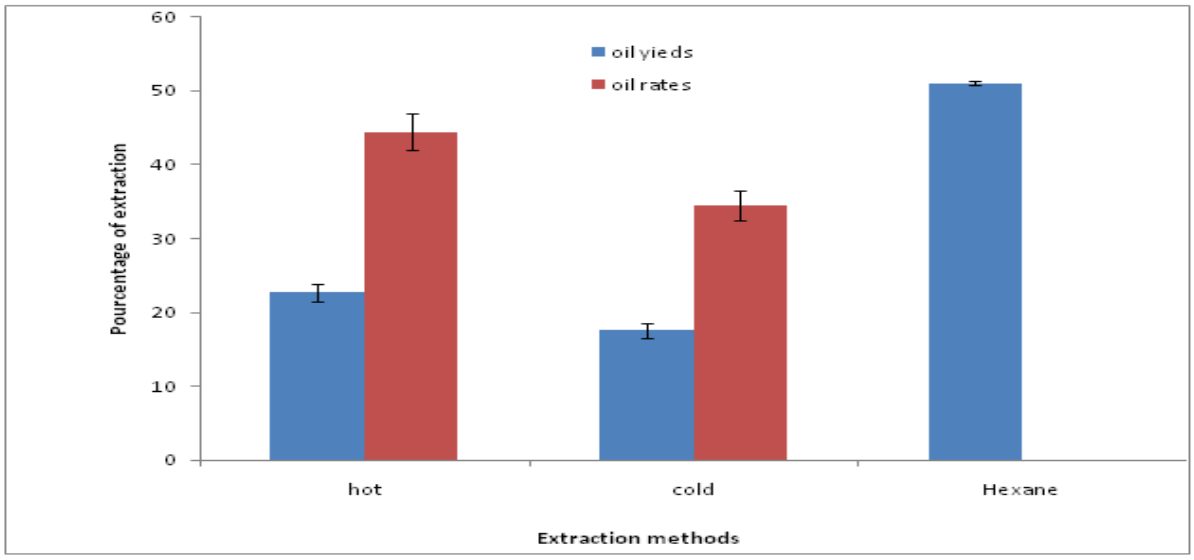

Fig. 2. The extraction Yield and rates of oil of almonds of R. heudelotii. Histograms with the same letter are not significantly different (p<0,05).

This difference is due to the High temperatures during hot pressing leads to degradation of α-eleostearic of oil. High proportions of this fatty acid makes very fragile and there for thermosensitive. The values found here are superior to those mentioned by Aboubakar Dandjouma *et al*. (2008) (43.56 ± 0.04%) during the extraction oil of *R. heudelotii* by enzymatic pathways. This shows that the fatty acid content of an oil depends on the extraction method.

UV spectra of R. heudelotii oil

The standards of α- eleostearic acid and the sample of *R. heudelotii* oils were analyzed by UV spectrophotometry. Fig. 3 shows these spectra. The spectra of α- eleostearic acid and *R. heudelotii* oils overlap. The spectra recorded presented three peaks between 260 nm and 280 nm. The characteristic peak was at 270 nm which characterizes the conjugated trienes (Karleskind, 1996). The spectra of the oils and the α- eleostearic overlap shows that the pressing had no effect on the structure of our α- eleostearic acid. The oil obtained by cold extraction has the highest absorbance (0.57± 0.01) than oil obtained from in hot condition (0.43 ± 0.01). The same trends were observed by GC. This difference could be due to the fact that the extraction temperature (80 °C) could result to the degradation of α- eleostearic acid. Torrecilla *et al.* (2010) have also showed that UV-vis is the rapid Method to quantify the adulteration of extra virgin olive.

Comparison of optical spectrophotometry analysis with capillary gas chromatography

The calibration test for the determination of α-eleostearic acid by UV spectrophotometry based on analysis by gas chromatography of the three oils resulted in the linear model using spectral data of the wavelength 270 nm. The calibration graph is shown in fig. 4.

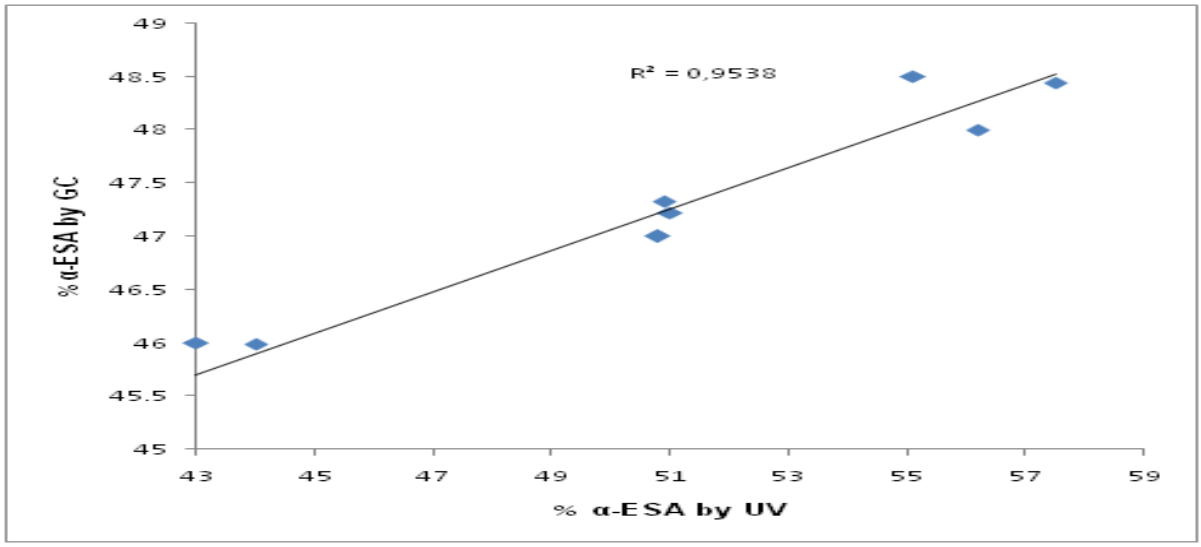

Fig. 3. Calibration graph for the determination of α- eleostearic acid % by UV and GC.

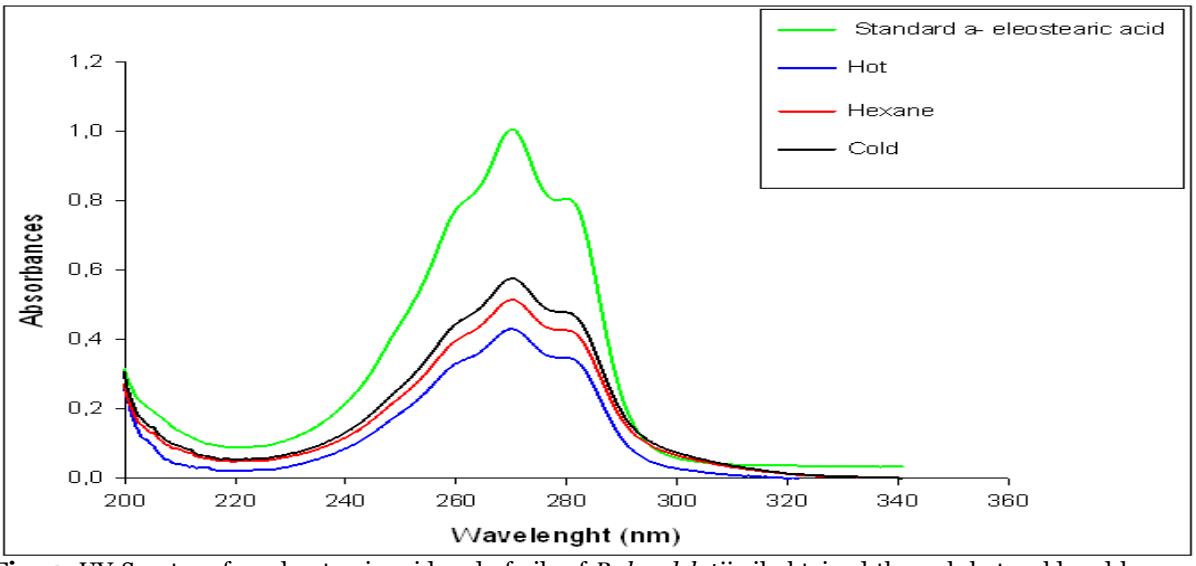

Fig. 4. UV Spectra of α- eleostearic acid and of oils of *R. heudelotii* oil obtained through hot, cold and hexane exact.

The correlation coefficient R (0.95) obtain for this calibration analysis of α-eleostearic acid by UV spectrophotometry clearly indicates the possibility offered by UV spectrophotometry to monitor the quality of *R. heudelotii* oil with benefits to be easier, faster and less expensive than gas chromatography. Bernuy *et al.* (2009) also showed that the correlation coefficient was positive and significant (0.97) between the Fourier Transform Raman Spectroscopy and gas chromatogrphy when quantifying the conjugated linoleic acid in a total of 22 samples synthesized by photoisomerization of linoleic acid contained in soybean oil. This shows the efficiency and fastness of spectroscopic techniques.

Conclusion

All results obtained in this work to suggest that the extraction of virgin oil of R. heudelotii with the Komet cold press (30 °C) gives a good quality oil that meets the standards. The high conjugated C18: 3 (α-eleostearic acid) is pleased of this oil of Particular interest from a nutritional point of of view.

Further research on the *R. heudelotii* oil extraction should make use the convenience and cost benefits offered by UV spectrophotometry compared to gas

chromatography for rapid quality control of the oil and especially the direct determination of α-eleostearic acid.

Acknowledgements

We thank the AUF (Agence Universitaire de la Francophonie) to finance this work.

References

Aboubakar Dandjouma AK. 2004. Contribution a l'étude des conditions de production et des aptitudes technologiques des huiles des oléagineux non – Conventionnels: cas de R*icinodendron heudelotii* (Bail.) Pierre ex Pax et de *Canarium schweinfurthii* Engl.. PhD thesis, Université of Ngaoundere, Cameroun, 203p.

Aboubakar Dandjouma AK., Tchiegang C, Kapseu C, Linder M, Parmentier M. 2008. Enzyme-assisted hexane extraction of *Ricinodendron heudelotii* (Bail.) Pierre ex Pax seeds oil. International Journal of Food Science and Technology **43**, 1169–1175.

Bernal J, Mendiola JA, Ibánez E, Cifuentes A. 2011. Advanced analysis of nutraceuticals. Journal of Pharmaceutical and Biomedical Analysis **55**, 758–774.

Bernuy B, Meurens M, Mignolet E, Larondelle Y. 2008. Performance Comparison of UV and FT-Raman Spectroscopy in the Determination of Conjugated Linoleic Acids in Cow Milk Fat. Journal of Agricultural and Food Chemistry **56(4),** 1159–1163.

Bernuy B, Meurens M, Mignolet E, Turu C, Larondelle Y. 2009. Determination by fourier transform raman spectroscopy of conjugated linoleic acid in i2-photoisomerized soybean oil. Journal of Agricultural and Food Chemistry **57,** 6524-6527.

Cayuela Sánchez, JA., Moreda W, García JM. 2013. Rapid Determination of Olive Oil Oxidative Stability and Its Major Quality Parameters Using Vis/NIR Transmittance Spectroscopy. Journal of Agricultural and Food Chemistry **61(34)**, 8056–8062.

Codex Alimentarius. 1992. Programme mixte FAO/OMS sur les normes alimentaires ». FAO, Rome (Italie).

CODEX. 1999. Norme codex pour graisses et les huiles comestibles non visées par des normes individuelles ». Codex STAN 19-1981 (Rév. 2-1999), 5p.

El-Abassy RM, Donfack P, Materny A. 2009. Visible Raman spectroscopy for the discrimination of olive oils from different vegetable oils and the detection of adulteration. Journal of Raman Spectroscopy **40,** 1284-1289.

Focant M, Mignolet E, Marique M, Clabots F, Breyne T, Dalemans D, Larondelle Y. 1998. The effects of vitamin E supplementation of cow diets containing rapeseed and linseed on the prevention of milk fat oxidation. Journal of Dairy Science **81**, 1095-1101.

Igarashi M, Miyazawa T. 2005. Preparation and fractionation of conjugated trienes from alpha-linolenic acid and their growth inhibitory effects on human tumor cells and fibroblasts. Lipids **40,** 109–113.

International Union Of Pure And Applied Chemistry (IUPAC). 1979. Méthodes d'analyses des matières grasses et dérivées, 6 éme édition, Lavoisier Tec et Doc, Paris, France, 190p.

Kapseu C, Tchiegang C. 1995. Chemical properties of *Ricinodendron heudelotii* (Bail.) seed oil. Journal of Lipids **2,** 87-98.

Karleskind A. 1996. Oils and Fats Manual. Vol. 1, Lavoisier TEC and DOC, Paris, France.

Kivçak B, Mert T. 2001. Quantitive determination of α-tocopherol in *Arbutus unedo* by TLC-densitometry and colorimetry. Fitoterapia **72,** 656 –

661.

Koba K, Imamura J, Akashoshi A, Kohno-Murase J, Nishizono S, Iwabuchi M, Tanaka K, Sugano M. 2007. Genetically modified rapeseed oil containing *cis*-9,*trans*-11,*cis*-13-octadecatrienoic acid affects body fat mass and lipid metabolism in mice. Journal of Agricultural and Food Chemistry. **55,** 3741–3748.

Kohno H, Suzuki R, Noguchi R, Hosokawa M, Miyashita K., Tanaka T. 2002. Dietary conjugated linolenic acid inhibits azoxymethane-induced colonic aberrant crypt foci in rats. Jpn. Journal of Cancer Research **93,** 133–142.

Kone S. 1998. Unités modernes de transformation des oléagineux à petite et très petite échelle: performances et limites . In C. Kapseu & G.J. Kayem (Eds.), Actes du 2éme Séminaire Safou, 3–5décembre 1997. Ngaoundéré, Cameroun: Presse Universitaires de Yaoundé I., 285–301p.

Lam CK, Chen J, Cao Y, Yang L, Wong YM, Yeung SY, Yao X, Huang Y, Chen ZY. 2008. Conjugated and nonconjugated octadecaenoic acids affect differently intestinal acyl coenzyme A: cholesterol acyltransferase activity. Atherosclerosis **198**, 85–93.

Mosso K, Kouadio N, Koulibaly S, Kouman KG. 1998. Utilisations alimentaires des amandes du *Ricinodendron heudelotii* ssp *heudelotii* en Côte d'Ivoire. *In:* Kapseu C. et Kayem G.J. [eds.]. *Actes du 2ème Séminaire Safou*, 3-5 décembre 1997, Ngaoundéré (Cameroun), Presse Universitaires de Yaoundé I, 325-341. ISBN 2-911541-10-3.

Norris FA. 1982. Extraction of fats and oils. In D. Swern (Ed.), Baileyés industrial oils and fat products (Vol. 2, 4th ed.). New York (USA): John Wiley and Sons, 603p.

Orthoeffer T. 1995. Rice bran. Processing and nutritional considerations. Institute of Food Technologists Annual meeting, 7p.

Poulli KI, Mousdis GA. Georgiou CA. 2006. Synchronous fluorescence spectroscopy for quantitative determination of virgin olive oil adulteration with sunflower oil. Analytical and Bioanalytical Chemistry **386,** 1571–1575.

Reaney MJT, Liu YD, Westcott ND. 1999. Commercial production of conjugated linoleic acid. In Advances in Conjugated Linoleic Acid Research, Press Champaim, IL **1,** 39 -54. ISBN : 1-893997-02-2.

Schneider C. 2005. Review: chemistry and biology of vitamin E. Molecular Nutrition and Food Research **49,** 7–30.

Suzuki R, Noguchi R, Ota T, Abe M, Miyashita K, Kawada T. 2001. Cytotoxic effect of conjugated trienoic fatty acids on mouse tumor and human monocytic leukemia cells. Lipids **36,** 477–482.

Tchankou Leudeu BC. 2006. Etude sur des jeunes rats mâles des propriétés nutritionnelles des huiles de *Canarium schweinfurthii* Engl., *Dacryodes edulis* (G. Don) H. J. Lam, *Ricinodendron heudelotii* (Bail.) Pierre ex Pax, et *Tetracarpidium conophorum* Müll. Arg. Hutch. and Dalz. PhD thesis, Université of Ngaoundere, Cameroun, 259p.

Tchiegang C, Kapseu C, Ndjouenkeu R, Ngassoum MB. 1997. Amandes de *Ricinodendron heudelotii* (Bail.): Matière première potentielle pour les industries agro-alimentaires tropicales. Journal of Food Engineering **32,** 1-10.

Tchiegang C, Ngo Oum M, Aboubakar Dandjouma AK, Kapseu C. 2004. Qualité et stabilité de l'huile extraite par pressage des amandes de *Ricinodendron heudelotii* (Bail.) Pierre ex Pax pendant la conservation à température ambiante. Journal of Food Engineering **62,** 69-77.

Tchiegang C, Aboubakar Dandjouma A K, Kapseu C, Parmentier M. 2005. Optimisation de

l'extraction de l'huile par pressage des amandes de *Ricinodendron heudelotii* Pierre ex Pax ». Journal of Food Engineering **68,** 79–87.

Torrecilla JS, Rojo E, Dominguez JC, Rodriguez F. 2010. A Novel Method To Quantify the Adulteration of Extra Virgin Olive Oil with Low-Grade Olive Oils by UV-Vis. Journal of Agricultural and Food Chemistry **58,** 1679-1679. http://dx.doi.org/10.1021/jf903308u.

Tsuzuki T, Igarashi M, Komai M, Miyazawa T. 2003. The metabolic conversion of 9,11,13-eleostearic acid (18:3) to 9,11-conjugated linoleic acid (18:2) in the rat. Journal of Nutritional Science and Vitaminology **49,** 195–200.

Tsuzuki T, Tokuyama Y, Igarashi M, Nakagawa K, Ohsaki Y, Komai M. 2004. Alpha-eleostearic acid (9Z11E13E-18:3) is quickly converted to conjugated linoleic acid (9Z11E-18:2) in rats. Journal of Nutrition **134**, 2634–2639.

Tsuzuki T, Kawakami Y. 2008. Tumor angiogenesis suppression by alpha-eleostearic acid, α linolenic acid isomer with a conjugated triene system, via peroxisome proliferator-activated receptor gamma. Carcinogenic. **29,** 797–806.

Vivien J, Faure JJ. 1996. Fruitiers sauvages d'Afrique. Espèces du Cameroun. Coopération Française, *CTA*., 416 p.

Wiemar HJ, Altes KFW. 1993. Méthodes artisanales de transformation des oléagineux. Gate/GTZ, Eschborn (Allemagne Fédérale), 134p.

Yamasaki M, Kitagawa T, Koyanagi N, Chujo H, Maeda H, Kohno-Murase J, Imamura J, Tachibana H, Yamada K. 2006. Dietary effect of pomegranate seed oil on immune function and lipid metabolism in mice. Nutrition **22,** 54–59.

Yang L, Leung KY, Cao Y, Huang Y, Ratnayake WM, Chen ZY. 2005. Alpha-linolenic acid but not conjugated linolenic acid is hypocholesterolaemic in hamsters. British. Journal of Nutrition **93**, 433–438.

Yasui Y, Hosokawa M, Sahara T, Suzuki R, Ohgiya S, Kohno H, Tanaka T, Miyashita K. 2005. Bitter gourd seed fatty acid rich in 9 *c*, 11*t* - conjugated linolenic acid induces apoptosis and up-regulates the GADD45, p53 and PPARgamma in human colon cancer Caco-2 cells. Prostaglandins Leukotrienes and Essential Fatty Acids **73**, 113–119.

Yuan GF, Sinclair AJ, Zhou CQ, Li D. 2009. Alpha eleostearic acid is more effectively metabolized into conjugated linoleic acid than punicic acid in mice. Journal of the Science of Food and Agricture **89,** 1006-1011.