GSC Biological and Pharmaceutical Sciences

eISSN: 2581-3250 CODEN (USA): GBPSC2

Cross Ref DOI: 10.30574/gscbps

Journal homepage: https://gsconlinepress.com/journals/gscbps/

(RESEARCH ARTICLE)

Check for updates

Physico-chemical characteristics of the fat of the flesh of the snail *Limicolaria flamméa*

Jean François Régis Bilo LOA [1,*], Envrin BOGUI [2], Siméon BROU [3] and Akissi Françoise KOUAME [3]

[1] *Laboratory of Biochemistry, Biotechnology and Food Sciences, Alassane OUATTARA University, UFR Sciences and Technologies, Department of Biosciences and Geologie; Côte d'Ivoire.*

[2] *Laboratory of Biotechnologies, UFR Bioscences Department of Biochemistry, Félix Houphouet-Boigny University, 22 BP 582 Abidjan 22 Côte d'Ivoire.*

[3] *Laboratory of Food Biochemistry and Technology of Tropical Products, UFR BIOSCIENCES, Department of Food Science and Technology, NANGUI ABROGOUA University, BP 801 Abidjan 02, Côte d'Ivoire.*

GSC Biological and Pharmaceutical Sciences, 2024, 29(01), 323–330

Publication history: Received on 06 August 2024; revised on 25 October 2024; accepted on 28 October 2024

Article DOI: https://doi.org/10.30574/gscbps.2024.29.1.0333

Abstract

Lipids are the most important energy-producing macromolecules in foods. The lipid level observed in the flesh of snails is conditioned by the species and their diet. To this end, studies were carried out on the chemical properties and physical characteristics of the fat coming from the flour obtained from the flesh of the *Limicolaria flamméa* snail. Standard analytical methods were used to assess certain physico-chemical properties (density, density, oleic acidity index.....). The analyses revealed that the fat from the flour derived from the flesh of the snail *Limicolaria flamméa* had an iodine value of 134.84 mg iodine/100g fat, a saponification value of 198.52 mg KOH/g fat, etc. The density, solidification point and saponification value of the fat were also measured. The density, solidification point and refractive index of the fat from the meal obtained from the flesh of the *Limicolaria flamméa* snail were 0.94 at 25 °C, 5 °C and 1.2 °B respectively. The fat from the meal obtained from the flesh of the *Limicolaria flamméa* snail is pure and non-flammable and contains virtually no free fatty acids.

Keywords: *Limicolaria flamméa* snail; Fat; Chemical properties; Physical characteristics

1. Introduction

The people of West Africa, particularly those of Côte d'Ivoire, appreciate the meat of giant African snails [1]. It is a significant source of protein, minerals and vitamins. The multiple nutritional potential offered by snail consumption means that the various snail species can be popularised [2]. The biochemical composition of the flesh of these animals generally reveals a broad spectrum of compounds whose energetic impact is also conditioned by the lipid content of the flesh of each species [3]. Lipids are the most important energy macromolecules in food. The lipid content observed in snail flesh is conditioned by the species and their diet. For example, the fatty acids contained in the flesh of giant snails vary [4]. This fat contains several essential fatty acids that are essential for human metabolism. The fatty acids identified in the flesh of the helix pomatia snail are palmitic acid, stearic acid, oleic acid, eicosadienoic acid and eicosatrienoic acid [4]. However, a good deal of scientific research has focused on snail farming. Most of this scientific work has focused on assessing the biological performance of snails in relation to the influence of diets and on characterising the biochemical and nutritional properties of snail meat [5]. In addition, very few studies have looked at the specific properties of the fat obtained from the flesh of snails, in particular the snail *Limicolaria flamméa*. The aim of this study was to evaluate the physical and chemical characteristics of the fat obtained from the flesh of the *Limicolaria flamméa* snail.

* Corresponding author: Jean François Régis Bilo LOA

2. Materials and methods

2.1. Materials

The biological material used in this study consisted of a single batch of *Limicolaria flamméa* snails collected in four communes of Abidjan, namely Yopougon, Abobo, Koumassi and Port-Boüet. The fatty matter derived from *Limicolaria flamméa* snail meat meal was used for the various analyses.

2.2. Methods

2.2.1. Obtaining flour from the flesh of the snail Limicolaria flamméa

Collected snails are left in a pot for two days on an empty stomach to allow them to release their excrement. The aim is to produce a flour that will not drown because of the excrement. More than 340 *Limicolaria flamméa* snails were removed from the shells for analysis after washing. The snails were then washed to remove any saliva or slime. A small knife was used to separate the visceral mass from the snail meat. Following this operation, the snails were washed again and drained through a sieve. The snail meat was oven dried at 80 °C until it reached a constant weight and then ground using a moulinex PATENT PENDING TYPE LM 240 blender to form a flour after sieving on a series of five (5) sieve frames with decreasing mesh sizes. These sieves were made up of 1000, 750, 500, 250 and 150 µm diameter meshes. Sieving is carried out directly by moving the five (5) sieve frames back and forth. The sifting thus ensured that the particles making up the flour were distributed by refusal.

2.2.2. Lipid extraction

The fat content of each sample was extracted according to [6] method using the SOXHLET. Five grams of *Limicolaria flamméa* (PE) snail meal were weighed and introduced into a tared WHATMAN cartridge. A volume of 100 mL of hexane was introduced into an extraction flask that had been weighed empty. The flask containing (M1 in grams) was placed on the heating cap integrated into the SOXHLET for 8 hours. After this extraction time, the flask was removed from the SOXHLET apparatus and placed in an oven at 60 °C for one hour to ensure complete evaporation of the solvent. At the end of evaporation, the flask was weighed again (M2 in grams). The fat content (TMG) was then determined according to the following formula:

$$TMG = 100 \times (M1 - M2)$$

TMG: Fat content in g/100g of snail powder; M1= mass of the whole flask and sample after steaming; M2 = mass of empty flask; Me: mass of sample

2.3. Determination of fatty acids

2.3.1. Preparation of methyl esters

Fatty acid methyl esters were prepared using boron trifluoride reagent at a concentration of 14% in methanol, BF3/Met OH.

One hundred (100) mg of oil sample was weighed into a 10 mL screw-tube. 1.5 mL hexane and 1.5 mL BF3/Met OH are added. The tube was sealed under vacuum, shaken vigorously and heated to 100 °C for 1 hour. After cooling to room temperature, 1 mL of hexane and 0.5 mL of distilled water were added, followed by stirring in vacuo. Two phases separate after resting. The upper phase was recovered in another vacuum tube. Next, 5 mL of hexane is added to the mixture of methyl esters to make it suitable for gas chromatographic analysis.

2.3.2. Analysis by gas chromatography

The methyl esters were analysed on a gas chromatograph (HP 6890 series GC system) equipped with a flame ionisation detector. They were separated on an HP-5 capillary column (30 cm long, 0.32 mm internal diameter, with a film thickness of 0.25 µm) in the presence of 5% diphenyl and 95% dimethyl polysiloxane, with the oven temperature programmed to increase from 60 to 325 °C at a rate of 1 °C per minute. The injector temperature was set at 275 °C and the detector temperature at 325 °C. The pressure of the nitrogen at the inlet, used as a carrier gas, varied from 6.90 to 47.6 Kpa. The flow rate was maintained at 1 cm / minute and the dead time was 1 minute 15 seconds (hydrogen 40 cm / second). Peaks were identified using reference fatty acid methyl esters by comparing the retention distances of each peak in the chromatogram with those obtained from the standards.

2.4. Chemical characteristics of fat

2.4.1. Peroxide value

The peroxide value corresponds to the quantity of peroxide-active oxygen contained in 1 kg of fat, capable of being released under the conditions of the experiment. It is used to assess the degree of oxidation of the unsaturated fatty acids in the fat. The peroxide value was determined using the **[7]** method.

$$I_I = \frac{126{,}9 \times (V_T \times V_E) \times N}{PE}$$

With :
VE: volume of sodium thiosulphate (mL) needed to titrate the assay;
VO: volume of sodium thiosulphate (mL) required to titrate the blank;
N: Normality of the sodium thiosulphate solution; 126.9: Molar mass (g/mol) of iodine;
PE: mass (g) of the test sample.

2.4.2. Acid value and oleic acidity

The acid number is the number of milligrams of KOH needed to neutralise the free acidity of one gram of fat. It gives an assessment of the quantity of free fatty acids. The acid number was determined using the **[7]** method.

The acid number was determined as follows:

$$I_A = \frac{V_{KOH} \times N_{KOH} \times 56{,}1}{PE}$$

With :
V: volume of KOH (mL) required to neutralise the free fatty acids;
N: normality of the KOH solution;
56.1: molar mass (g/mol) of KOH;
PE: mass (g) of the test sample.

Free acidity is the quantity of free fatty acids expressed as a percentage of oleic acid (molar mass: 282). Oleic acidity was determined by the following relationship:

$$\text{Oleic acidity} = \frac{I_A \times 282}{56{,}1} \times 0{,}1 = \frac{I_A}{1{,}9}$$

2.4.3. Saponification number

The saponification number corresponds to the number of milligrams of KOH required to saponify 1 g of fat. It is an indicator of the quantity of total fatty acids present in a fatty substance. The saponification number was estimated using the AOAC method, standard 920.160, 1997 **[7]**. The saponification index (SI) was expressed by the following formula:

$$I_S = \frac{(V_E \times V_T) \times N \times 56{,}1}{PE}$$

With :
VT: volume of hydrochloric acid required to titrate the blank ;
VE: volume of hydrochloric acid needed to titrate the sample;
N: normality of hydrochloric acid (HCl) ; PE: mass (g) of the test sample; 56.1: molecular mass (g/mol) of potassium.

2.4.4. Refractive index

The refractive index was determined using the method described by [8].

2.4.5. Determination of unsaponifiable matter content

The unsaponifiable matter content was determined by deduction using the following mathematical method:

$$\text{Unsaponifiable content} \quad \frac{(I_S \times I_A)}{PE} \times 100$$

2.5. Physical characteristics of fat

2.5.1. Density, appearance and colour of fat

The density of a body in relation to water is the ratio between the density of this body and that of water. It has no unit. Ten (10) mL of water and 10 mL of fat are taken using a tared beaker. Each sample is weighed with the beaker. A mass difference is made to determine the mass of each sample taken.

2.5.2. Colorimetric indices

Colorimetric indices have been determined according to the method defined by the CIE International Commission on Illumination (1931). To measure colour, a colorimetric device illuminates a sample, captures the quantity of light transmitted or reflected in the wavelength range 380 to 780 nm and quantifies it as a spectral measurement.

2.5.3. Fat density

The density of fat has been determined using the [9] method. It represents the quotient of its mass by its volume.

$$\textbf{density} = \frac{M1 - M0}{V} \times 100$$

M0: mass (g) of the empty volumetric flask
M1: mass (g) of the volumetric flask filled with fat,
V: volume (ml) of fat contained in the volumetric flask at room temperature (25 °C).

2.5.4. Solidification point

The solidification point was determined by cryoscopy in accordance with standard NFEN ISO 5764. This is the temperature at which the fat solidifies.

2.5.5. Flash point

The flash point was determined in accordance with ISO 15267:1998, NF in ISO 2592, ASTM D 92 or the Pensky-Martens closed cup method.

2.5.6. Melting point

The slip melting point was determined using the ISO 6321: 2002 method or the automatic capillary method, which involves immersing a capillary tube containing a column of crystallised fat.

3. Results

3.1. Chemical characteristics of the fat

The chemical characteristics of the fat from the meal obtained from the flesh of the *Limicolaria flamméa* snail are shown in Table 1. Analysis of the data obtained provides an inventory of chemical indices at varying levels. The peroxide, iodine and saponification indices were respectively 11.52 meq O2/Kg fat, 134.84 mg iodine/100g fat and 198.52 mg KOH/g

fat. The fat showed an acid value of 11.79 mg KOH/g fat, an oleic acid value of 6.2 % and an unsaponifiable matter content of 6.68 %.

Table 1 Chemical characteristics of fat

Parameters	Values
Peroxide value (meq oxygen/kg fat)	11,52 ± 0,21
Iodine value (mg iodine/100g fat)	134,84 ± 0,53
Saponification index (mg KOH/g fat)	198,52 ± 0,19
Acid number (mg KOH/g fat)	11,79 ± 0,19
Oleic acidity index (%)	6,20 ± 0,06
Unsaponifiable matter content (%)	6,68 ± 0,45

3.2. Physical characteristics of the fat

Table 2 shows the physical characteristics of the fat from the meal obtained from the flesh of the snail *Limicolaria flamméa*. The results showed that the fat had a brown colour deduced from the chromatic indices (L = 51.1; a* = -1.9; b* = + 17.9). Snail meal from *Limicolaria flamméa* has a fat content of 8.64%. This fat has a density of 0.94 with a density of 0.86 g/ml. The soluble solids content, measured in Brix degrees, is 1.2 °B. The fatty matter derived from *Limicolaria flamméa* snail meal has specific temperature points that can modify the state of this matter. The solidification point (Ps), flash point (Pe) and melting point (Pf) are 5, 92 and 6 °C respectively.

Table 2 Physical characteristics of fat

Parameters	Values
Colour indexes	L = 51,1 a* = -1,9 b* = + 17,9
Hue (°)	H = 180,39
Colour	Brown
Extraction rate (%)	6,99 ± 0,2
Density (25 °C)	0,94 ± 0,005
Density (g/ml)	0,86 ± 0,03
Solidification point (°C)	5 ± 0,09
Flash point (°C)	92 ± 1
Melting point (°C)	6 ± 2
Fat content (g/100g)	8,6 ± 0,6
Refractometric index (°B)	1,2 ± 0,03

3.3. Determination of fatty acids

The fatty acid composition of *Limicolaria flamméa* snail meal is shown in Table 3. This composition shows a varied presence of fatty acids such as myristic acid (7.55 mg/100g), arachidonic acid (13.11 mg/100g), oleic acid (3.75 mg/100g), linolenic acid (5.96 mg/100g) and linoleic acid (2.58 ± 0.70 mg/100g).

Table 3 Fatty acid composition

Fatty acids	Content (mg/100g)
Myristic acid	7,55 ± 2,12
Arachidonic acid	1,31 ± 1,41
Oleic acid	3,75 ±1,14
Linolenic acid	5,96 ± 0,71
Linoleic acid	2,58 ± 0,70

4. Discussion

The acid number (Ia) of a lipid is the mass of potassium hydroxide (KOH), expressed in milligrams, required to neutralise the free acidity contained in one gram of fat. The acid number is therefore used to judge the state of deterioration. The Ia of the fat of the *Limicolaria flamméa* snail is 11.79 mg KOH/g fat. This suggests that the fat contains very few free acids. This would be due to the fact that the acidity would be caused by bodies other than fatty acids due to their intimate intermingling. This value is in agreement with that highlighted by [10] in Limicolaria undulata snails, which is 9.23 ± 0.03. Similarly, the oleic acid index of the fat in the meal obtained from the flesh of the *Limicolaria flamméa* snail is low at 6.2 ± 0.06%, and the unsaponifiable matter content is 6.68 ± 0.06%, 45 respectively would indicate that the fat of the *Limicolaria flamméa* snail would contain very little free fatty acid due to the low fat content of the flesh of the *Limicolaria flamméa* snail. The concentration of fatty acids (FA) increases with the lipid content of the flesh. There are major differences in muscle lipid content (from less than one gram to more than 15g/100g of flesh) between species. These differences are not linked to origin (collection or farming), but to the capacity of the species' muscle tissue to store fat. For the same species, muscle lipid content can vary over the year depending on the animal's physiological state and the quantity of food available.

The saponification index obtained in the fat of the snail *Limicolaria flamméa* (198.52 ± 0.19 mg KOH/g fat) is higher than that of the fat in the snail Limicolaria torquata highlighted by Roslizawati et al., (2017). In dairy products such as Roquefort cheese 15.43 ± 6.81. The same applies to the caterpillar Imbrasia oyemensis (151.79 mg KOH/g fat) and is almost equal to the fat content of the larvae of Rhynchophorus pheonicis (198.90 mg KOH/g fat) and Oryctes rhinoceros (190 mg KOH/g fat) obtained by [11].

The value of the iodine index (134.84± 0.53 mg/100g of fat) obtained after analysis of the fat is based on the presence of unsaturated fatty acids in the snail's flesh. The higher the iodine value of a body, the higher its unsaturated fatty acid content. The presence of iodine in the fat is thought to be strongly linked to the preference for certain species of plants that have it and which make up the spectrum of the snail's diet [12].

Unsaponifiables (6.68%), made up of bio-active substances including hydrocarbons, tocopherols, sterols and terpene alcohols, are present in small quantities in this fat from the larva of Oryctes owariensis. The presence of tocopherols is important in foods, as they protect fats against autoxidation and thus increase the shelf life and nutritional value of food [13].

The physical properties of the fat from the meal obtained from the flesh of the *Limicolaria flamméa* snail analysed reveal that this fat has a viscous appearance at room temperature and a brown colour. The refractive index is 1.2 ± 0.03 with a density of 0.94. These parameters, which highlight the purity of the fat from the meal obtained from the flesh of the *Limicolaria flamméa* snail, are similar to those determined for snails of the helix genus, which are pale brown in colour, with a density of 0.76 and a refractive index of 1.3 [14].

The density of the fat in the meal from the flesh of the *Limicolaria flamméa* snail is 0.86 ± 0.03 g/ml. This density is in line with that obtained from the giant African snail, which is 0.82 g/ml [15]. The brown colour of the fat of the *Limicolaria flamméa* snail is thought to be due to the synthesis of melanin, which contributes to the recognition of individuals of different species in the same biotope [16].

The refractive index of the fat would indicate that the soluble solids content is virtually non-existent and that this fat would be pure, which would underlie the fat extraction technique.

The melting point or melting temperature of a substance is the temperature at which a pure element or chemical compound melts, i.e. passes from the solid state to the liquid state at a given pressure, while the solidification point, depending on the substance, is the temperature of the opposite transition. It is usually measured under normal atmospheric pressure (1 atmosphere) and there is coexistence between solid and liquid states between these two points. The flash point is the minimum temperature at which a liquid generating flammable vapours can be ignited in air by a flame above its surface [17]. The melting point of *Limicolaria flamméa* snail fat is 6 °C and the solidification point is 5 °C. The flash point is 92 °C. The melting and solidification temperatures are virtually identical because the fat has a high purity level. The flash point of snail fat indicates that it is non-flammable (EC, 2008). The non-flammable nature of snail fat (*Limicolaria flamméa*) is due to the fact that it is derived from living organic matter, in this case animal, as described by [17].

Fat is a form in which energy is stored in the living organism. Lipids are the most energetic macronutrients present in foods [18]. The presence of fatty acids from the flesh of the snail *Limicolaria flamméa*, namely myristic acid, arachidonic acid, oleic acid, linolenic acid and linoleic acid, highlights medium-chain (myristic acid) and long-chain fatty acids.

The relative presence of medium- and long-chain fatty acids illustrates the possibility of synthesis or accumulation of several fatty acids by the snail *Limicolaria flamméa*. The synthesis of these fatty acids would be due to interesterification by enzymatic catalysis corresponding to an exchange of acyl groups between a fatty acid ester and a sugar ester and would have the advantage of generating neither water nor alcohol in the environment [19]. The fatty acid content, in particular oleic acid (3.75 ± 1.414mg/100g), linolenic acid (5.965 ± 0.707 mg/100g) and linoleic acid (258.5 ± 0.707mg/100g) from the meat of the snail *Limicolaria flamméa* remains respectively below the fatty acids from the meat of wild snails studied by [4], 11.47%; 11.92%; and 14.24% in helix pomatia. The essential fatty acid (linolenic acid) present in the fat from the flesh of the *Limicolaria flamméa* snail is the precursor of omega-6 fatty acids, which are responsible for cardiovascular and immune balance through their action on cholesterol [20].

This fat also contains oleic acid, a monounsaturated fatty acid involved in regulating the heart rhythm and limiting the risk of heart attack [21]. In fact, oleic acid has no disadvantages; on the contrary, it is involved in the synthesis of good cholesterol. Highly insensitive to oxidation, oleic acid is a factor in the stability of oils during frying or cooking [22].

5. Conclusion

The fat in the flour derived from the flesh of the *Limicolaria flamméa* snail contains saturated and unsaturated fatty acids. Essential fatty acids such as linolenic acid and oleic acid are precursors of omegas 3 and 6, which are essential for heart activity. The fat in the flour derived from the flesh of the *Limicolaria flamméa* snail is pure and non-flammable, and contains virtually no free fatty acids. The flesh of the *Limicolaria flamméa* snail has a great capacity to store fat in its muscles. The fat from the meal obtained from the flesh of the *Limicolaria flamméa* snail has excellent chemical and physical properties, making it suitable for incorporation into certain food formulations.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

References

[1] Owen D.F. & Reid J.C. The white snails in Africa:the significance of man in the maintenance of a striking polymorphism, *OIKOS*. 1986; **46**, 267269.

[2] Okon, B., Ibom, L. A., Ina-Ibor, O.B. and Owai, P.U. Nutritional evaluation of African land snail (*Archachatina marginata saturalis)* fed diet containing full fat rubber as a replacement for soybean. Nigerian Journal of Agriculture, Food and Environment. 2016 ; 12(2): 1-8.

[3] Chen X, Feng Y, Chen Z. Common edible insects and their utilization in China: invited review. Entomological Research. 2009 ; 39:299–303.

[4] Daina I., AleKsandrs J., Vita S. Evaluation of nutrion value of roman snail's (Helix pomata) meat obtained in Latvia.FOODBALT. 2014.

[5] Kouassi K.D., Otchoumou A. & Dosso H. Les escargots comestibles de Côte d'Ivoire : Influence de substrats d'élevage sur les paramètres de croisszнces *d'Archachatina ventricosa* (Gould, 1850) en élévage hors-sol. *Livestock Res. Rural Dev.* 2007 ; **19**, 16-20.

[6] AOAC : Association of Official Analytical Chemists. *Official Method of Analysis.* 16th Edition. AOAC Arlington. 1995; 250p.

[7] AOAC: Association of Official Analytical Chemists, norme 920.160. *Official Method of Analysis* Washington D.C. 1997

[8] International Union of pure and applied chemistry and International Union of Biochimical nomenclature (IUPAC). The nomenclature of lipids. *J. Lipid Res*., 1978, **19**, 114-129.

[9] ISO 15267:1998 NF-Animals and vegetal fats and oils flash point limit test using pensky Martens closed cup flash tester (Identical with ISO 15267: 1998) ICS: 67.200.10 Published by Ethiopian Standards Agency ©ESA.

[10] Roslizawati Ab L, Smith J, Savins D, Ashley Dowell, Bucher D. Investigation of nutritional properties of three species of marine turban snails for human consumption Food Science & Nutrition. 2017; 5(1): 14–30 doi: 10.1002/fsn3.360

[11] Ekpo K.E. & Onigbinde A.O. Characterization of lipids in winged reproductives of the termite *Macrotermes bellicosus*. *Pakistan Journal of Nutrition*. 2007 ; 6: 247-251.

[12] Alais. C, Linden. G. Biochimie alimentaire. Publisher, Masson Paris ISBN,2225828539 ; 97822258539. Length ; Export citation BiBTex. 1997 pp : 248.

[13] Kamal-Eldin and Appelqvist, L.A. The chemistry and antixydant properties of tocopherol and tocotrienols'.*Lipids.* 1996; (31) 671-701.

[14] Gomot A. Biochimical composition of *Helix snails :* influence of genetic and physiological factors. Journal of Mollus studies. 1997; 64, 173-181.

[15] Kalia G. A., Etela I. Nutritional and sensory profiling of the Africain giant snail fed commercial-type and leaf-based diet in Rain Forest Ecology. African Journal of food, Agriculture,*Nutrition and Development,* 2011. Vol.23, p. 453-458.

[16] Friday E. Uboh, Ima O. Williams, Nesssie C. Essien. Effect of processing on the proximate and Mineral Composition of *Archachatina marginata* and *Achatina achatina.* Food and Public Health. 2014; 4(1): 10-14.

[17] Janes A, Chaineaux J, Douglas Carson, Wassila Benaissa. Experimental study of ignition of bulk storage by hot points. AIChE Spring Meeting. 2011; 7.

[18] Reece JB, Urry LA, Cain ML, Wasserman SA, Minorsky PV. Campbell biology. Yorkshire: Pearson Benjamin Cummings. 2011

[19] Akoh C.C, Swanson Dan. B. G. One- stage synthesis of raffinose fatty acid polyester. Journal of Food Science. 1987; 52 : 1570 -1578.

[20] Demaison L., Moreau D.: Dietary n-3 poly- unsaturated fatty acids and coronary heart disease -related mortality: a possible mechanism of action. Cellular and Molecular Life Sciences. 2002; 59, 463-477

[21] Dommels Y, Alink GM, Van Bladeren PJ & van Ommen B. Dietary n-6 and n-3 polyunsaturated fatty acids and colorectal carcinogenesis: results from cultured colon cells, animal models and human studies. Environmental Toxicology and Pharmacology. 2002; 11 297–308.

[22] FAO/OMS : Les graisses et huiles dans la nutrition humaine. Rapport d'une consultation mixte d'experts, Rome. 1993., 26 p.