Assiut J. of Agric. Sci., 42 (Special Issue)(The 5th Conference of Young Scientists Fac. of Agric. Assiut Univ. May,8, 2011) (212-221)

Assessment of Fatty Acids Composition, Total Lipid Fractions and Oil Characteristics of Four Nut Kinds

Mokhless A. M. M. Abd El-Rahman*, M. Kamal E. Youssef, Mohamed N. A. El-Rify and El Sayed A. Ramadan.

Food Sci.& Tech. Dept., Fac. Agric., Assiut Unvi, Assiut, Egypt

Abstract

This study was planed to evaluate separation and identification of lipids fractions in four nuts namely: Almonds, Hazelnuts, Walnuts and Peanuts. The study included the fractionation and determinations of the compounds of nuts oil as well as the fatty acids composition by GLC technique. Besides, the seeds contained significant amounts of crude oil recorded 55.18, 67.52, 71.40 and 46.93 % (dry matter) in Almonds, Hazelnuts, Walnuts and Peanuts; respectively. The refractive index, acid number, peroxide values, iodine number and saponification number were determined in nuts oil. Using TLC technique the total lipids of nuts were fractionated to eight fractions. Triglycerides fraction recorded 85.17, 80.44, 71.85 and 70.11% of total lipids in Almonds, Hazelnuts, Walnuts and Peanuts; respectively, while the polar lipids fraction represented 1.18, 0.70, 1.73 and 1.50% at the same nuts kinds; respectively. The data revealed that the unsaponifiable matters of the four studied nuts recorded 0.48, 0.55, 0.48 and 0.48; respectively. The fatty acids analysis indicated the presence of 9 fatty acids in nuts oil namely: myristic palmitic, palmitoleic, oleic, stearic, eicosenoic, linolenic, linoleic and arachidic. The predominant unsaturated fatty acid was oleic acid (C18:1), while the predominant saturated fatty acid was palmitic acid in all studied nut kinds. The total unsaturated fatty acids recorded 75.66, 59.75, 86.60 and 69.15%, while the total saturated fatty acids recorded 24.34, 40.25, 13.40 and 30.85% in Almonds, Hazelnuts, Walnuts and Peanuts; respectively.

Keywords: Refractive index, Acid number, Acidity, Peroxide value, Iodine number, Saponification number, Unsaponifiable matter, Fatty acids, Total lipids, Almonds, Hazelnuts, Walnuts, Peanuts.

*Corresponding author: E-mail addresses:

mokhless2000@yahoo.com

1- Introduction

Nuts had constituted a part of mankind's diet since preagricultural times (Eaton & Konner, 1985). While the amount of nuts in the human diet in the distant past was unknown, consumption data from industrialised nations indicated a downward trend for

Received on:29/3/2011 **Accepted for publication on:** 5/4/2011

Referees: Prof.Dr.Abdalah S. Abdel-Gouad Prof.Dr Mohamed R. Abdelaal

most of the 20th century, although nut consumption in countries following a more Mediterranean diet was twice that of the American diet (Sabaté, 1993; Dreher *et al.*, 1996). Vegetarians and other health-conscious populations, such as Seventh Day Adventist, tend to consume nuts more often than their counterparts (Sabaté, 1999). Whether for custom, economy, apprehension or simple lack of knowledge, large segments of the world population do not consume nuts on a regular basis, since, nuts contribute a small proportion of their total caloric intake. Nuts are consumed either as snacks or part of a meal. Nuts are eaten whole (fresh or roasted), in spreads (peanut butter, almond paste) or hidden (e.g. commercial products, mixed dishes, sauces, baked goods, and oils). Nuts are nutrient dense foods. They contain high amounts of protein and fat, mostly unsaturated fatty acids. Nuts are also dense in a variety of other nutrients and provide dietary fiber, vitamins (e.g. folic acid, niacin, vitamin E, and vitamin B_6), minerals (e.g. copper, magnesium, potassium, zinc) and many bio-active constituents such as antioxidants, phytosterols and other phytochemicals (Dreher *et al.*, 1996). The most popular edible tree nuts include almonds, Brazil nuts, cashews, hazelnuts, macadamias, pecans, pine nuts, pistachios and walnuts. Ground nuts, commonly known as peanuts, are actually legumes but are identified by consumers as part of the nuts food group. Peanuts share a similar nutrient profile with tree nuts.

The present investigation was carried out in an attempt to clarify the lipids profile as well as the fatty acid composition of studied four nut kinds. Meanwhile their oil characteristics were assessed as well.

2- Materials and Methods

2.1. Materials:

2.1.1. Nut samples:

Nut kinds namely: Almonds (*Prunus dulcis*); Hazelnuts (*Corylus avellana*); Walnuts (*Juglans regia*) and Peanuts (*Arachis hypogaea*) were used in the present study. The samples were obtained from local Cairo market, during the season 2009. 5 Kg of each nut kind were obtained.

2.2. Analytical Methods:

2.2.1. Preparation of samples:

Randomly picked whole nuts fruit were weighed. The edible portion was manually removed. The respective weights of whole fruit, edible portion, and shell were recorded (Agunbiade & Olanlokun, 2006). The nuts were shelled manually and screened to remove bad seeds (Christian & Ukhun, 2006). The edible portions of nuts were ground using a blender to a fine powder and dried as described by (Christian & Ukhun, 2006). The resulting powder was preserved in closed plastic bags under freezeing conditions until analysis.

2.2.2. Lipids analysis:

2.2.2.1. Lipids extraction:

Lipids were extracted from the dried samples of nuts by Chloroform: Methanol mixture (2: 1, v: v) according to the method described by (Folch *et al.*, 1957).

2.2.2.2. Physical and chemical constants of oils:

Refractive index at 25°C, acid value, peroxide value, iodine number and saponification number were determined as outlined in **(AOAC, 1995)**.

2.2.2.3. TLC- separation and identification of total lipids:

Silica gel (Gf 254, type 60) plates (20×20 cm) were used for qualitative and quantitative determinations of lipid fractions. Plates were developed in a mixture of petroleum ether: diethyl ether and glacial acetic acid (80: 20: 1, v/ v/ v) (Kates, 1972).

The lipid fractions were visualized by exposure to iodine vapor. The isolated fractions were identified on thin layer plates by comparing their Rf values with those of known lipid standards. For quantitative analysis, the TLC chromatograms were scanned using densitometer model (Seroscan elvi 146) and the data were analyzed by QS computer program analysis J scans.

2.2.2.4. Unsaponifiable matters determination:

Unsaponifiable matters were determined according to the procedures described by (Kornsteiner *et al.*, 2006).

2.2.2.5. Preparation of methyl ester of fatty acids:

The methyl esters of fatty acids were prepared from aliquots of total lipids using 5 ml 3% H_2SO_4 in absolute methanol and 2 ml benzene as mentioned by (Rossell *et al.*, 1983).

2.2.2.6. GLC-of methyl ester of fatty acids:

The methyl esters of fatty acids were separated using a PYE Unicam Pro-GC gas liquid chromatography with a dual flame ionization carried out on (1.5m × 4 mm) SP-2310 column, packed with 55 % cyanopropyl phenyl silicone dimensions. Column temperature: At first increasing the temperature from 70 190°C at the rate of 8°C/ minute and then isothermal for 10 minute at 190°C. The injector and detector temperature were 250°C and 300°C, respectively. Carrier gas: Nitrogen at the rate 30 ml/ minute, hydrogen and air flow rate were 33 and 330 ml/ minute; respectively. The chart speed was 0.4 cm/ minute. Peak identifications were established by comparing the retention times obtained with standard methyl esters. The areas under chromatographic peak were measured with electronic integrator as mentioned by (Rossell *et al.*, 1983).

3. Results and Discussion

3.1 Physical and chemical properties of nuts oil

The data presented in Table (1) revealed the refractive index, acid number, peroxide value, iodine number and saponification number of nuts oil for the four studied kinds. From such data it could be noted that slight differ-

ences between the refractive index of the four studied kinds were detected. Among the four studied kinds walnuts had the highest values of iodine number (82.09) and peroxide value (11.46); while hazelnuts had the highest Acid number and Acidity % (0.8309 and 0.4177%); peanuts had the lowest Acid number and Acidity % (0.2458 and 0.1235%). Such results are not in agreement with Mexis *et al.* (2009) for peroxide value, who recorded lower values. Such variation could be attributed to the environmental, agricultural conditions of nuts planting.

Table (1): Physical and chemical properties of nuts oil*:

Nuts kinds	Refractive index (25°C)	Acid number mg KOH/g.	Acidity %	Peroxide value Meq/ kg	Iodine number	Saponification number
Almonds	1.4632	0.3566	0.1793	8.33	68.59	250.45
Hazalnuts	1.4612	0.8309	0.4177	8.75	62.75	244.01
Walnuts	1.4685	0.6713	0.3375	11.46	82.09	244.24
Peanuts	1.4626	0.2458	0.1235	10.75	57.11	244.30

*: Mean of three replicates.

3.2 Total lipids fractionations of nuts oil

The qualitative and quantitative of the fractionation of total lipids fractions of nuts oils are shown in Table (2) and Fig (1). The results revealed that total lipids consisted of eight fractions in the four nuts kinds with some variation in their percentages. The triglycerides constituted the major percentage of total lipids and accounted for 85.17, 80.44, 71.85 and 70.11% of almonds, hazalnuts, walnuts and peanuts; respectively.

The perentage of total lipid fractions as percent of total lipids indicated that triglycerides were the predominant fraction followed by 1,2 &2,3 diglycerides, monoglycerides, sterols, 1,3 diglycerides, polar lipids. sterolester & hydrocarbons and free fatty acids in almonds oil. The data given in Table (2) revealed that there was adverse relationship between the triglyceride compounds and free fatty acids content in the four studied nuts kinds. While the second fraction percentage of total lipids was free fatty acids and followed by 1,2 &2,3 diglycerides, sterolester & hydrocarbons, monoglycerides, sterols, 1,3 diglyceride and polar lipids in the hazelnuts, but in the walnuts the second fraction percentage of total lipids was monoglycerides, and followed by free fatty acids, 1,2 &2,3 diglycerides, 1,3 diglycerides, sterols, sterolester & hydrocarbons and polar lipids.

Finally, in the peanuts the second fraction percentage of total lipids was sterolester & hydrocarbons, and followed by free fatty acids, 1, 3 diglycerides, sterols, monoglycerides, 1, 2 & 2, 3 diglyceride and polar lipids.

The data also showed that the lowest component of total lipids was polar lipids, which accounted for 0.70, 1.73 and 1.50% of hazalnuts, walnuts and peanuts; respectively, while free fatty acids recorded the lowest component of almonds (0.51%).

Table (2): The total lipid fractions of the four studied nut oils (% of total lipids):

No.	Total lipids fractions	Nuts kind			
		Almonds	Hazalnuts	Walnuts	Peanuts
1	Polar lipids	1.18	0.70	1.73	1.50
2	Monoglycerides	3.30	2.69	8.56	3.49
3	1,2 &2,3 diglycerides	3.85	3.90	3.97	2.89
4	Sterols	2.82	2.16	3.11	3.50
5	1,3 diglycerides	2.26	2.15	3.25	4.33
6	Free fatty acids	0.51	4.96	5.02	6.22
7	Triglycerides	85.17	80.44	71.85	70.11
8	Sterolester & Hydrocarbons	0.91	3.00	2.51	7.96

	Total lipids fractions	Nuts kinds			
		Almonds	Hazenuts	Walnuts	Peanuts
1	Sterolester & Hydrocarbons				
2	Triglycerides				
3	Free fatty acids				
4	1,3 diglycerides				
5	Sterols				
6	1,2 &2,3 diglycerides				
7	Monoglycerides				
8					
	Polar lipids				

Fig (1): Thin layer chromatograms of the total lipid fractions of nut oils.

3.3 Unsaponifiable matters of nuts oil

Unsaponifiable matter content was 0.48, 0.55, 0.48 and 0.48 for almonds, hazelnuts, walnuts and peanuts; respectively. **Kornsteiner *et al.*, (2006)** found similar results of unsaponifiable matter content of the investigated nuts of almonds that represented 0.35- 0.53, hazelnuts 0.20- 0.30, walnuts 0.25- 0.40 and peanuts 0.35- 0.59 (g/100 g extracted oil).

Table (3): Unsaponifiable matter of nuts oil:

Item	Nuts kind			
	Almonds	Hazelnuts	Walnuts	Peanuts
Unsaponifiable matter	0.48	0.55	0.48	0.48

3.4 Fatty acids composition of nuts oil

The data presented in Table (4) illustrated the fatty acids composition of nuts oil extracted from dried nuts. The tabulated data showed that the predominant unsaturated fatty acid in the extracted oils of studied nuts kinds was Oleic acid ($C_{18:1}$) followed by linoleic acid ($C_{18:2}$) of almonds, hazelnuts and walnuts, while in peanut was linolenic ($C_{18:3}$). While the predominant saturated fatty acid was palmitic acid in all studied nut samples.

The total unsaturated fatty acids were 75.66, 59.75, 86.60

Table (4): Fatty acids composition of nuts oil

Fatty acids	Carbon chain	Almonds	Hazelnuts	Walnuts	Peanuts
Myristic	$C_{14:0}$	---	1.08	0.40	0.79
Palmitic	$C_{16:0}$	19.74	27.15	7.67	19.96
Stearic	$C_{18:0}$	4.60	11.16	3.39	6.99
Arachidic	$C_{20:0}$	---	0.86	1.94	3.11
Palmitoleic	$C_{16:1}$	1.64	0.82	0.21	2.10
Oleic	$C_{18:1}$	70.60	50.27	70.11	60.02
Eicosenoic	$C_{20:1}$	---	1.06	---	---
Linoleic	$C_{18:2}$	2.80	7.60	16.28	2.58
Linolenic	$C_{18:3}$	0.62	---	1.60	4.45
total saturated		**24.34**	**40.25**	**13.19**	**30.85**
Monounsaturated		72.24	52.15	69.20	62.12
Diunsaturated		2.80	7.60	16.02	2.58
Polyunsaturated		0.62	---	1.60	4.45
Total Unsaturated		**75.66**	**59.75**	**86.60**	**69.15**
Saturated/ Unsaturated		**0.32**	**0.67**	**0.15**	**0.45**

and 69.15%, while the total saturated fatty acid were 24.34, 40.25, 13.40 and 30.85% in almonds, hazelnuts, walnuts and penuts; respectively. Likewise, the obtained herein data are in agreement with Kris-Etherton *et al.* (1999), Higgs (2003) and Xu and Hanna (2009). Solà-Alberich *et al.* (2002), who found similar values. While, the present data are in disagreement with Parcerisa et al. (1998), Spiller et al. (1998), McKay and Sibley (2009), Banel and Hu (2009), Sabaté and Wien (2010) and Yücesan et al. (2010).

Besides, Table (4) revealed that the Saturated / Unsaturated fatty acids ratio accounted to 0.32, 0.67, 0.15 and 0.45 for almonds, hazelnuts, walnuts and penuts; respectively.

In conclusion the data indicated that the four studied nuts recorded significant levels of crude oil, which contained some important components as well as unsaturated fatty acids. It is noteworthy that such studied four nut kinds are highly recommended for caring diabetic, hypercholestermic and obese patients.

References:

Agunbiade, S. O. & Olanlokun, J. O. (2006): Evaluation of some nutritional characteristics of Indian almond (*Prunus amygdalus*) nut. *Pakistan J of Nutrition* 5 (4), 316-318.

AOAC (1995): Official Methods of Analysis, 16thedn. Association of Official Analytical Chemists, Washington, DC.

Banel, D. K. & Hu, F. B. (2009): Effects of walnut consumption on blood lipids and other cardiovascular risk factors: a meta-analysis and systematic review. *Amr. J. Clin. Nutr.* 90, 56–63.

Christian, A. & Ukhun, M. E. (2006): Nutritional potential of the nut of tropical almond (*Terminalia Catappia* L.). *Pakistan J. of Nutrition* 5 (4), 334-336.

Dreher, M. L., Maher, C. V. & Kearney, P. (1996): The traditional and emerging role of nuts in healthful diets. *Nutr. Rev.* 54(8), 241–245.

Eaton, S. B. & Konner, M. (1985): Paleolithic nutrition. A consideration of its nature and current implications. *N. Engl. J. Med.* 312, 283–289.

Folch, J., Lee, M. & Sloane-Stanleg, G. H. (1957): A simple method for the isolation and purification of total lipids from animal tissues. *J. Biological Chem.* 226, 497-510.

Higgs, J. (2003): The beneficial role of peanuts in the diet - part 2. *Nutr. & food sci.* 33(2), 56-64.

Kates, M. (1972): Techniques of lipidology Isolation. Analysis and Identification of Lipids. North Holland Publishing Co. Amsterdam, London.

Kornsteiner, M., Wagner, K. H. & Elmadfa, I. (2006): Analytical, Nutritional and Clinical Methods: Tocopherols and total phenolics in 10 different nut types. *Food Chemistry* 98, 381–387.

Kris-Etherton, P. M., Yu-Poth, S., Sabaté, J., Ratcliffe, H. E., Zhao, G. & Etherton, T. D. (1999): Nuts and their bioactive constituents: effects on serum lipids and other factors that affect disease risk. *Amr. J. Clin. Nutr.* 70, 504S-511S.

McKay, D. L. & Sibley, D. (2009): Omega-3 fatty acids from walnuts. *Nutrition Dimension*, 1-14.

Mexis, S. F., Badeka, A. V., Chouliara, E., Riganakos, K. A. & Kontominas, M. G. (2009): Effect of γ-irradiation on the physico-chemical and sensory properties of raw unpeeled almond kernels (Prunus dulcis). Innovative Food Science and Emerging Technologies 10(1), 87-92.

Parcerisa, J., Richardson, D. G., Rafecas, M., Codony, R. & Boatella, a. J. (1998): Fatty acid, tocopherol and sterol content of some hazelnut varieties (*Corylus avellana* L.) harvested in Oregon (USA). *J. of Chromatography A* 805, 259–268.

Rossell, J. B., King, B. & Downes, M. J. (1983): Detection of adulteration. *J. Amr. Oil Chem. Soc.* 60, 333.

Sabaté, J. (1993): Does nut consumption protect against ischaemic heart disease? *Eur. J. Clin. Nutr.* 47, Suppl. 1, S71–S75.

Sabaté, J. (1999): Nut consumption, vegetarian diets, ischemic heart disease risk, and all-cause mortality: evidence from epidemiologic studies. *Amr. J. Clin. Nutr.* 70, 500S–503S.

Sabaté, J. & Wien, M. (2010): Nuts, blood lipids and cardiovascular disease. *Asia Pac. J. Clin. Nutr.* 19(1), 131-136.

Solà-Alberich, R., Godàs-Bonfill, G., Salas-Salvadó, J. & Masana-Marín, L. I. (2002): Effects of Tree Nuts on cholesterol and cardiovascular diseases. *Formación Continuada en Nutrición y Obesidad* 5, 194-202.

Spiller, G. A., Jenkins, D. A. J., Bosello, O., Gates, J. E., Cragen, L. N. & Bruce, B. (1998): Nuts and Plasma Lipids: An Almond-Based Diet Lowers LDL-C while Preserving HDL-C. *J. of the American College of Nutrition* 17(3), 285-290.

Xu, Y. X. & Hanna, M. A. (2009): Synthesis and characterization of hazelnut oil-based biodiesel. *Industrial Crops and Products* 29(2-3), 473-479.

Yücesan, F. B., Örem, A., Kural, B. V., Örem, C. & Turan, İ. (2010): Hazelnut consumption decreases the susceptibility of LDL to oxidation; plasma oxidized LDL level and increases the ratio of large/small LDL in normolipidemic healthy subjects. *Anadolu Kardiyol Derg* 10, 28-35.

تقدير التركيب الحامضي الدهني والتجزؤات الليبيدية وخواص الزيت لأربعة أنواع من النقل

مخلص احمد محمد محمد عبد الرحمن، محمد كمال السيد يوسف، محمد نجيب احمد الريفى، السيد عبد النبى رمضان

قسم علوم وتكنولوجيا الاغذية – كلية الزراعة – جامعة أسيوط

تم تصميم هذا البحث لاستخلاص وتعريف التجزءات الليبيدية لأربعة انواع من النقل وهى على وجه التحديد اللوز، والبندق، والجوز، والفول السودانى. وقد اشتملت الدراسة على دراسة التجزؤات الليبيدية والتركيب الحامضى الدهنى لها وذلك باستخدام طريقة التحليل الكروماتوجرافى الغازى. هذا الى انه تم تقدير نسبة الدهن الخام فى هذة الانواع الاربعة من النقل وقد كانت كالتالى55.18، 67.52، 71.40، و46.93% لكل من اللوز، والبندق، والجوز، والفول السودانى، على التوالى. فضلا عن انه تم تقدير كل من الثوابت الدهنية كمعامل الانكسار، ورقم الحموضة، ورقم البيروكسيد، والرقم اليودى، ورقم التصبن فى زيوت هذه الانواع الاربعة من النقل.

وقد تم استخدام طريقة التحليل الكروماتوجرافى على الطبقة الرقيقة لتفريد التجزءات الليبيدية للانواع الاربعة المدروسة من النقل وقد اظهرت النتائج وجود 8 تجزءات ليبيدية. سجلت الجلسريدات الثلاثية النسب التالية 85.17، و80.44، و71.85، و70.11% بالنسبة لليبيدات الكلية فى كل من اللوز، والبندق، والجوز، والفول السودانى، على التوالى، بينما بلغت قيم الليبيدات القطبية 1.18، و0.70، و1.73، و1.50% لنفس الانواع على التوالى. بينما كانت المواد غير القابلة للتصبن فى الانواع الاربعة المدروسة على النحو التالى 0.48، و0.55، و0.48، و0.48 على التوالى.

ومن جهة اخرى اظهرت نتائج تحليل التركيب الحامضى الدهنى وجود 9 احماض دهنية فى زيوت الانواع الاربعة المدروسة من النقل وهى على وجه التحديد الميرستك، والبالميتك، والبالميتوليك، والاوليك، والاستيريك، والايكوسينويك، واللينولينك، واللينوليك، والاراشيدك. كان حامض الاوليك هو الحامض غير المشبع السائد، بينما كان حامض البالميتك هو الحامض المشبع السائد في الأنواع الأربعة المدروسة من النقل. وقد استبان من نتائج البحث ان مجموع الأحماض الدهنية غير المشبعة بلغ 75.66، و59.75، و86.60، و69.15%، بينما بلغ مجموع الأحماض الدهنية المشبعة 24.34، و40.25، و13.40، و30.85% فى كل من اللوز، والبندق، والجوز، والفول السودانى، على التوالى.