Food Chemistry 68 (2000) 347–351

Food Chemistry

www.elsevier.com/locate/foodchem

Mineral composition of two walnut cultivars originating in France and California

F. Lavedrine[a], A. Ravel[a,*], A. Villet[a], V. Ducros[b], J. Alary[a]

[a]*Laboratoire de Chimie Analytique, Université J. Fourier, Domaine de la Merci, F 38706 La Tronche-Cedex, France*
[b]*Laboratoire de Biochimie C, Hopital Michallon, BP 217, F 38043 La Tronche Cedex 9, France*

Received 1 March 1999; received in revised form; accepted 1 August 1999

Abstract

Mineral composition of two walnut cultivars (Franquette and Hartley) originating both in France and California was determined. Microwave assisted mineralisation was followed by atomic absorption spectrometry for all minerals except selenium which was quantified by gas chromatography–mass spectrometry (GC–MS), and phosphorus by a colorimetric method. Three elements presented major level differences linked to variety or origin. For origin, potassium levels of the French walnuts were higher (Franquette: 487 and Hartley: 466 mg 100 g^{-1}) than those of the Californian walnuts (Franquette: 358 and Hartley: 372 mg 100 g^{-1}). Inversely, sodium level was higher in the Californian Hartley than in the French Hartley (6.7 vs 0.3 mg 100 g^{-1}). For variety, high levels of magnesium were observed for the Franquette cultivars (French: 191 and Californian: 202 mg 100 g^{-1}) while the Hartley levels were lower (French: 129 and Californian: 134 mg 100 g^{-1}). The same observation could be made for zinc, but the differences were weaker. Nevertheless, these data could not differentiate the walnuts according to origin or variety. © 1999 Elsevier Science Ltd. All rights reserved.

1. Introduction

This work is part of a larger study whose purpose was to determine the composition of two varieties of walnuts originating both in France and California, the Franquette cultivar as it represents 80% of all walnut production in Dauphiné (France) and the Hartley cultivar which is the most common in California.

In a previous study, we reported the fat-soluble vitamin composition of the French and the Californian walnuts (Lavedrine, Ravel, Poupard & Alary, 1997). Both contained high levels of gamma tocopherol. Some differences were shown according to variety or origin. Nevertheless, these differences were not discriminant either for variety or origin.

As referred to food composition tables (Randoin, Legallic, Dupuis & Bernardin, 1985; Feinberg, Favier & Ireland-Ripert, 1987; Klepping, Guilland, Fuchs, Marcer & Houard-Malval, 1989; Souci, Fachmann & Kraut, 1994), walnuts contain high levels of potassium (390–700 mg 100 g^{-1}), phosphorus (310–510 mg/100 g) and magnesium (90–140 mg 100 g^{-1}). On the other hand their content in sodium is quite low (1–15 mg 100 g^{-1}). However, data from food tables do not refer to variety and geographic origin. Moreover, trace element levels are often missing.

So our purpose was to carry on with the comparison between the French and the Californian walnuts with mineral and more especially trace elements composition analysis in order to establish the influence of variety and origin.

The first step of mineral analysis usually consists of a sample mineralisation. In the past several years, microwave assisted mineralisation methods have been developed (Kingston & Jassie, 1988; Dunemann & Meinerling, 1992; Sun, Waters & Mawhinney, 1997; Lamble & Hill, 1998). The advantages of microwave digestion are numerous: a strict control over the heating power and the time heating, a short digestion time, a reduced blank level (when Teflon flasks were used), and a little risk of contamination. Moreover, these methods are often automated, allowing good reliability and efficiency (Feinberg, 1991).

The aim of our study was to develop and validate the determination of selected minerals in four walnut cultivars after an open microwave digestion method, using atomic absorption spectrometry (AAS). However, because

---

* Corresponding author. Tel.: +33-76-63-71-45; fax: +33-76-51-86-67.

E-mail address: anne.ravel@ujf-grenoble.fr (A. Ravel).

PII: S0308-8146(99)00204-6

of selenium volatility, its determination implies the use of a closed microwave digestion. As it was not possible to operate with such an apparatus, a wet washing of walnuts was carried out. Then selenium quantification was achieved by gas chromatography–mass spectrometry (GC–MS), a method allowing the determination of ultra trace levels.

2. Materials and methods

2.1. Sample description

Commercial (5 kg) packs of Franquette and Hartley both harvested at two locations Dauphiné (France) and California (USA) were supplied by the CING (Comité interprofessionnel de la noix de Grenoble). Unshelled walnuts were stored at +4°C until analysed. For each cultivar, a sample (200 g) was removed randomly from the 5 kg package and rapidly ground into a fine powder with a plastic robot food processor (Bühler H04). They were rehomogenised just before mineralisation.

2.2. Reagents

All reagents were of analytical grade. Mineralisation and AAS: HNO_3 65%, H_2SO_4 95%, $CsCl_2$ and $LaCl_3$, were purchased from Merck (Germany). Mineral laboratory standards [$CaCO_3$; $Cu(CH_3COO)_2$; $Fe(NO_3)_3$; $MgCl_2$; $MnSO_4$; KCl; NaCl; $Zn(CH_3COO)_2$] (Merck) were diluted before use to cover the specified range (Fe 0.1–0.5; Na 0.2–1; Mg 0.02–0.1; Cu 0.25–1.25; K, Zn 0.5–2.5; Ca 1–5; Mn 2–10) mg l^{-1}.

Selenium determination: HCl 30%, chloroform (Prolabo France), HNO_3 65%, $HClO_4$ 70% (Merck) were used. Standards were prepared from a solution of SeO_2 tritisol (Merck) at 1 g l^{-1} and diluted to obtain 20, 50, 100, 150 and 200 μg l^{-1} solutions. 4 Nitro-*o*-phenylenediamine was purchased from Janssen Chimica (Belgium). ^{76}Se (atomic abundance) in elemental form was obtained from CEA (France).

Phosphorus determination: HNO_3 65%; Na_2HPO_4; NH_4VO_3 were obtained from Merck and $(NH_4)_6 Mo_7O_{24}$, $10H_2O$; MgO from Prolabo.

2.3. Instrumentation

Flame AAS determinations were effected using a UNICAM 929 model spectrophotometer using the manufacturer's specifications.

Selenium was determined by GC–MS with a HRGC mega 2 series gas chromatograph linked to a TRIO 1000 quadrupole mass spectrometer (ThermoQuest, France) (Ducros & Favier 1992).

The microwave digestion was performed with a Microdigest TM301 (Prolabo, France) apparatus equipped with a 200 W magnetron and operating at atmospheric pressure, a three-way pump (Pump PS11, Prolabo, France) and a TX32 programmer (Prolabo, France).

A muffle furnace (Furnace Thermolyne 62700, USA) was used for phosphorus determination and a classical heating-block (Grant, Jouan, France) for selenium.

Phosphorus was quantified by colorimetry using a Perkin–Elmer 550SE spectrophotometer.

2.4. Mineralisation

Microwave mineralisation was carried out as follows. One gram of ground walnuts was accurately weighed into a Teflon flask. Ten ml H_2SO_4 were added. Five min later 5 ml HNO_3 were added and the mixture was heated for 5 min at power 60W. It was heated 10 min more at power 100 W. At last, 5 ml HNO_3 were poured into the flask. The mixture was heated 5 min at power 100 W. After cooling to room temperature for 20 min, the digest was transferred into a 50-ml vial. The Teflon flask was rinsed three times with 5 ml ultrapure water and the volume was made up to 50 ml. Iron, copper and zinc were analysed on this digest while magnesium and potassium were determined in the digest 1/50 diluted and calcium in the digest 1/2 diluted.

2.5. Analysis

AAS: the mineral elements were quantified by atomic absorption using appropriate conditions (AFNOR 1995).

Selenium: because of its very low level in walnuts, selenium was determined as 2,1,3 benzoselenadiazole complex by a GC–MS method as previously described (Ducros & Favier, 1992).

Phosphorus: it was determined on 0.5 grams of ground walnuts according to the vanadomolybdic method (AFNOR, 1978).

2.6. Method validation

Selenium and phosphorus were quantified with specific methods which have already been validated (AFNOR, 1978; Ducros, Ruffieux, Belin & Favier, 1994). For the other minerals, both microwave mineralisation and AAS method had to be validated. So, two minerals (magnesium and copper) were selected for the validation as they represented the highest (Mg) and the lowest (Cu) element in walnuts. Linearity was assessed by two ways on three replicates:

- (method 1) spiking increasing amounts of minerals (Mg: 1; 2; 3 mg and Cu: 10; 20; 30 μg) to one gram of walnut immediately before digestion;
- (method 2) analysing increased amounts of walnuts (0.5; 1; 1.5; 2; 2.5 grams).

Recovery percentages were performed by spiking appropriate amounts of Mg (1 mg) and Cu (10 μg) into the 1 g walnut sample before mineralisation (six replicates). Repeatability and reproducibility were evaluated both with standard solution and walnut digests (six replicates).

2.7. Statistical analysis

Two-way analysis of variance was used to determine the influence of variety and of geographical origin on mineral composition.

3. Results and discussion

3.1. Method validation

Linear relationships were obtained. Regression factors were 0.999 for copper and 0.999 for magnesium (method 1). They were 0.987 for copper and 0.996 for magnesium (method 2).

Flame AAS was sensitive enough to determine the main elements in walnut. The overall precision of the method was evaluated. Repeatability rSD for standards ranged 0.95% for copper and 0.5% for magnesium. They were 3.86% (copper) and 1.17% (magnesium) for walnut digests. Reproducibility RSD for standard were 2.12% (copper) and 1.09% (magnesium) while they were a little higher for walnut digests 5.03% (copper) and 3.24% (magnesium).

3.2. Analysis methods

Food composition data are often obtained from AAS measurements. Recently Miller-Ihli (1996) compared flame AAS and inductively coupled plasma atomic emission spectrometry (ICP–AES) methods focusing on their precision, accuracy and detection capability. She concluded that both methods were similarly accurate and precise for food composition analysis. However, because of the high temperature of the plasma and the completeness of atomisation, ICP–AES was less prone to chemical matrix interference due to molecule formation. It also covered a wider concentration range.

Nevertheless, AAS is still widely used since it is a rapid method, easy to perform.

Miller-Ihli (1996) also compared two mineralisation procedures, a wet ashing and a dry ashing. There were no apparent differences between the results. The same observations were shown by Oles and Graham (1991) who compared hot plate digestion and microwave assisted digestion of different food matrices.

Optimal conditions for microwave digestion depend not only on the sample composition but also on the nature and volume of reagent, reaction procedure and digestion time. The method we described is convenient for fatty foods.

3.3. Walnut mineral composition

All the data of elemental composition for walnuts are listed in Table 1. They represent the mean ± standard deviation of six analyses of the same batch of each walnut cultivar.

In the whole, mineral composition of the four cultivar showed high levels of potassium, phosphorus and magnesium, and at the opposite very low levels of sodium. Trace elements were also quantified. Selenium levels were about 1 μg 100 g^{-1}.

Our data are in agreement with those of earlier reports shown in Table 2 except for selenium. This element is generally missing in food composition tables. It appears only in the Souci, Fachmann and Kraut (1994) table, but at a higher level than ours. However, walnut variety and origin were not stated. These differences could result from walnut origin. In the same way, Chang, Gutenmann, Reid and Lisk (1995) compared the selenium levels of another variety of walnut originating in Brazil. They showed variations from 0.3 to 3.6 mg 100 g^{-1} depending on their origin. It is well known that elemental composition and also pH of soil greatly influence mineral absorption by plants (Charlot & Germain, 1994). For instance, acidic soils enhance Cu and Mn absorption. Inversely, chalky soils have been shown to lower iron absorption. So the main composition of the four walnut cultivars was rather similar although some differences appeared. The influence of variety most notably affected magnesium concentrations. The Hartley walnuts contained the lowest levels [129 mg 100 g^{-1} (France) and 134 mg 100 g^{-1} (California)] while the Franquette had the highest levels [191 mg 100 g^{-1} (France) and 202 mg 100 g^{-1} (California)]. Although similar results were observed for calcium, the differences

Table 1
Mineral concentrations of the four walnut cultivars[a]

	Franquette France	Hartley France	Franquette California	Hartley California
Ca	91 ± 1.8	63 ± 2.4	67 ± 3.8	58 ± 2.4
Cu	1.2 ± 0.1	1.5 ± 0.2	1.1 ± 0.1	1.4 ± 0.1
Fe	1.8 ± 0.1	2.4 ± 0.4	2.9 ± 0.1	2.3 ± 0.1
K	487 ± 12	466 ± 23	358 ± 12	372 ± 9
Mg	191 ± 19	129 ± 3	202 ± 17	134 ± 2
Mn	4.3 ± 0.2	2.4 ± 0.1	1.1 ± 0.2	3.3 ± 0.1
Na	0.4 ± 0.1	0.3 ± 0.1	0.6 ± 0.1	6.7 ± 0.7
P	385 ± 5	308 ± 12	363 ± 3	353 ± 11
*Se	1.0 ± 0.1	0.7 ± 0.1	1.1 ± 0.1	1.0 ± 0.1
Zn	1.9 ± 0.2	1.8 ± 0.1	1.2 ± 0.1	1.6 ± 0.3

[a] Mean values ± SD (mg 100 g^{-1} except *selenium as μg 100 g^{-1}) ($n = 6$).

Table 2
Mineral average levels reported in the literature for walnut cultivars[a]

Author	Payne 1985	Randoin et al. 1985	Feinberg et al. 1987	Klepping et al. 1989	Ravai 1992	Souci et al. 1994
Origin	California	California	–	–	California	–
Ca	90	80	61	87	89	60–100
Cu	1	1	–	–	1.3	0.3–1.4
Fe	2.6	2.1	–	2.5	2.4	2–3.1
K	423	600	690	544	391	440–700
Mg	137	132	130	129	113	92–144
Mn	1.7	–	–	–	2.1	0.75–3.21
Na	15.4	3	3	2.4	10	1–4
P	346	400	510	409	348	310–510
*Se	–	–	–	–	–	5.5
Zn	2.7	2	–	–	2.9	2–3.2

[a] Values expressed as mg 100 g^{-1} except *selenium as μg 100 g^{-1}.

were weaker. Also, the Hartley walnuts contained a slightly higher content of copper than the Franquette.

The influence of geographical origin was also studied. The French walnuts had a somewhat higher content of zinc. However, the most important differences concerned the potassium content. The Californian walnuts had the lowest levels (Franquette 358, Hartley 372 mg 100 g^{-1}) opposed to the French walnuts (Franquette 487, Hartley 466 mg 100 g^{-1}). Conversely, the Californian Hartley had the highest levels of sodium (6.7 vs 0.6 mg 100 g^{-1} for the French Hartley).

On the other hand, copper, iron and magnesium showed no differences according to the geographical origin of walnuts.

All the differences we observed were statistically significant ($p < 0.0001$, ANOVA test).

With regard to minerals, the nutritional interest of walnuts is mainly due to trace elements and magnesium content. For that reason the higher content of magnesium in Franquette walnuts is of particular interest as is the higher level of zinc in French Franquette.

Despite elemental composition differences between the four walnut cultivars, it is not possible to classify walnut according to origin based on average elemental composition. Other studies such as sensorial analyses should be carried out to differentiate walnut origin.

Acknowledgements

This work was supported by the CING (Chatte-Isère-France). The authors greatly thank A. Serrano and L. Sauze for technical assistance.

References

AFNOR. (1978). Recueil de normes françaises des Corps gras, graines oléagineuses, produits dérivés. Dosage colorimétrique du phosphore. Méthode vanadomolybdique. Norme AFNOR NF V 60-227, Association Française de Normalisation, Paris.

AFNOR. (1995). Contrôle de la qualité des produits alimentaires. Méthodes d'analyses officielles Tome 1. AFNOR-DGCCRF, Association Française de Normalisation, Paris.

Chang, J. C., Gutenmann, W. H., Reid, C. M., & Lisk, D. J. (1995). Selenium content of Brazil nuts from two geographic locations in Brazil. *Chemosphere, 30*, 801–802.

Charlot, G., & Germain, E. (1994). Le noyer: nouvelles techniques. Ed CTIFL, Paris.

Ducros, V., & Favier, A. (1992). Gas chromatographic–mass spectrometric method for the determination of selenium in biological samples. *J. Chromatogr, 583*, 35–44.

Ducros, V., Ruffieux, D., Belin, N., & Favier, A. (1994). Comparison of two digestion methods for the determination of selenium in biological samples. *Analyst, 119*, 1715–1717.

Dunemann, L., & Meinerling, M. (1992). Comparison of different microwave-based digestion techniques in view of their application to fat-rich foods. *Fresenius' J. Anal. Chem, 342*, 714–719.

Feinberg, M., Favier, J. C., & Ireland-Ripert, J. (1987). Repertoire général des aliments (INRA), Technique et documentation. Ed Lavoisier, Paris, p. 189.

Feinberg, M. H. (1991). Definition of reference procedures for focused microwave digestion. *Analyst, 19*, 47–55.

Kingston, H. M., & Jassie, L. B. (1988). Introduction to microwave sample preparation: theory and practice. American Chemical Society, Washington.

Klepping, J., Guilland, J. C., Fuchs, F., Marcer, I., & Houard-Malval, M. (1989). Recueil de données sur la composition des aliments, CEIV, Roche, Neuilly sur Seine, p. 128.

Lamble, K. J., & Hill, S. J. (1998). Microwave digestion procedures for environmental matrices. *Analyst, 123*, 103R–133R.

Lavedrine, F., Ravel, A., Poupard, A., & Alary, J. (1997). Effect of geographic origin, variety, and storage on tocopherol concentrations in walnuts using HPLC. *Food Chem, 58*, 135–140.

Miller-Ihli, N. (1996). Trace element determinations in foods and biological samples using inductively coupled plasma atomic emission spectrometry and flame atomic absorption spectrometry. *J. Agric. Food Chem., 44*, 2675–2679.

Oles, P. J., & Graham, W. M. (1991). Microwave acid digestion of various food matrices for nutrient determination by atomic absorption spectrometry. *JAOAC Int.*, 74, 812–814.

Payne, T. (1985). California walnuts and light foods. *Cereal Foods World, 30*, 215–218.

Randoin, L., Legallic, P., Dupuis, Y., & Bernardin, A. (1985). Table de composition des aliments. Institut scientifique d'hygiène alimentaire, Ed J. Lanore, Paris.

Ravai, M. (1992). Quality characteristics of California walnuts. *Cereal Foods World, 37*, 362–366.

Souci, S. W., Fachmann, W., & Kraut, H. (1994). Food composition and nutrition tables. Medpharm, CRC Press, Stuttgart, pp. 955–956.

Sun, D. H., Waters, J. K., & Mawhinney, T. P. (1997). Microwave digestion of aluminium, boron, and 13 other elements in plants by inductively coupled plasma atomic emission spectrometry. *JAOAC Int, 80*, 647–650.