Short note

GC/MS ANALYSIS OF FATTY ACIDS IN SOME CORN INBRED LINES[#]

STĂNCUȚA SCROB, SEVASTIȚA MUSTE*, MONICA CULEA***

*Faculty of Food Science and Technology, University of Agricultural Sciences and Veterinary Medicine, 3–5, Mănăştur St., Cluj-Napoca, Romania, e-mail: stancuta_ale@yahoo.com
**Faculty of Physics, "Babeş-Bolyai" University, 1, M. Kogălniceanu St., Cluj-Napoca, Romania

Abstract. A GC/MS method was developed for the determination of the fatty acid composition of 25 corn samples of 5 corn inbred lines. Significant differences between saturated, monounsaturated and polyunsaturated fatty acid of corn inbred lines flour was observed. The PUFA concentration in the different inbred lines flour samples was three times higher than SFA. In selection, choosing inbred lines with good quality traits can improve the nutritional and functional quality of the corn flour.

Key words: GC/MS, fatty acids, corn, inbred lines.

INTRODUCTION

Gas chromatography coupled with mass spectrometry (GC/MS) is an excellent technique for fatty acids identification and quantitation [2, 4, 5]. The fatty acids composition is a characteristic of plant species and has nutritional, biochemical and technological importance. Corn is an excellent source of polyunsaturated fatty acid (PUFA), especially linoleic acid, an essential PUFA. Among commercial vegetable oils, only safflower oil (77.7%) and sunflower oil (72.6%) have higher percentages of PUFA than corn oil. Although it is highly polyunsaturated, corn oil is considered fairly stable to oxidation, due to minor level of linolenic acid [6].

A gas chromatography-mass spectrometry (GC/MS) technique was used for qualitative and quantitative analysis of total fatty acids in corn seeds [1–6]. The method involves extraction procedure following Folch's technique [1, 3], derivatization and GC/MS analysis. Fatty acids were derivatized as methyl ester derivatives, separated by GC and identified by MS techniques. The method was

[#]Presented at The 12th National Conference on Biophysics, Iași, June 13–16, 2013.
Received: May 2013;
in final form May 2013.

applied to analyze the total fatty acids in corn inbred lines seeds samples. Maize occupies an important position in the world economy and trade as a food, feed and as industrial grain crop. Development of high yielding hybrids is the most important objective to enhance productivity. For developing hybrids, better inbred lines with high mean performance are required. Analysis of genetic diversity and of relationship among the elite breeding materials can significantly aid in crop improvement [7].

The aim of the paper was to study the differences in fatty acids composition of 25 corn samples of 5 corn inbred lines by using GC/MS analysis.

MATERIALS AND METHODS

The experimental material comprised 25 corn seed samples of 5 corn inbred lines, new varieties of seeds created at SCDA Turda, Romania. The 5 new inbred lines corn seeds studied for fatty acid composition noted: 1.2–1.5; 2.2–2.5; 3.2–3.5; 4.2–4.5; 5.2–5.5, represent the corn seeds nucleus TC 209(1.1), TC 316(2.1), TC 243(3.1), TB 367(4.1), D 105(5.1) experimented with the cytoplasm types: (cit T 248), (cit TB 329), (cit TC 177) and (cit TC 221). The ground seeds were extracted for analysis following Folch's extraction method [1, 3]. 0.3 g of flour, 0.5 mL of chloroform: methanol 2:1 (v:v) were used by mixing 2 minutes, then the lower layer was collected and solvents were removed in a nitrogen flow. The extraction was repeated twice. Before the fatty acids can be separated by gas chromatography, they must be converted to low molecular weight, volatile, nonpolar derivatives (e.g., fatty acid methyl esters). The fatty acids were derivatized into fatty acids methyl esters (FAME) at 80^{o}C for 20 minutes. The samples were injected into gas chromatograph on a Rtx-5MS capillary column, 30 m × 0.25 mm, 0.25 μm film thickness, using a temperature program from 50^{o}C, 1 min, 8 ^{o}C/min at 300^{o}C, the flow rate 1ml/min, with helium 5.5 as carrier gas. The separated compounds were identified into the mass spectrometer. A Trace DSQ Thermo Finnigan quadrupole mass spectrometer coupled with a Trace GC was used. The following conditions were followed: transfer line temperature 250^{o}C, injector temperature 200^{o}C; ion source temperature 250^{o}C; Splitter: 10:1. Electron energy was 70 eV and emission current, 100 μA. Undecenoic acid (C11:1) was used as internal standard for quantitative determination.

RESULTS AND DISCUSSION

The method was validated by using fatty acid standard samples. The GC/MS method developed gave good validation parameters. Precision gave R.S.D. values lower than 10% for the important fatty acids determined. The recovery gave 82.6%. The method was applied to study the variation of the total fatty acids in the

corn inbred lines seeds. The main fatty acids obtained by GC/MS analysis in investigated corn seeds were saturated palmitic (C16:0) and stearic acids (C18:0), and unsaturated oleic (C18:1) and linoleic acid (C18:2). The compounds were identified in the corn extracts by using the NIST library and are presented in Table 1.

Table 1

The compounds identified in the inbred lines of corn seed extracts

No.	Compound	Retention time (min)
1	C11:1 (undecenoic acid = SI)	16.09
2	C16:1 (palmitoleic acid)	23.13
3	C16:0 (palmitic acid)	23.40
4	C18:2(9,12) (linoleic acid)	25.55
5	C18:1(9) (oleic acid)	25.60
6	C18:0 (stearic acid)	25.79
7	C20:1(11-eicosenoic acid)	27.75
8	C20:0 (eicosanoic acid, arachidic acid)	27.99
9	C22:0 (behenic acid) M = 354	30.04
10	C24:0 (lignoceric acid) M = 382	31.87
11	beta-tocopherol M = 416	34.93
12	Vitamin E (gamma-tocopherol) M = 430	35.65
13	campesterol M = 400	37.06
14	stigmasterol M = 412	37.33
15	gamma-sitosterol M = 412	38.14
16	stigmastanol M = 416	38.33

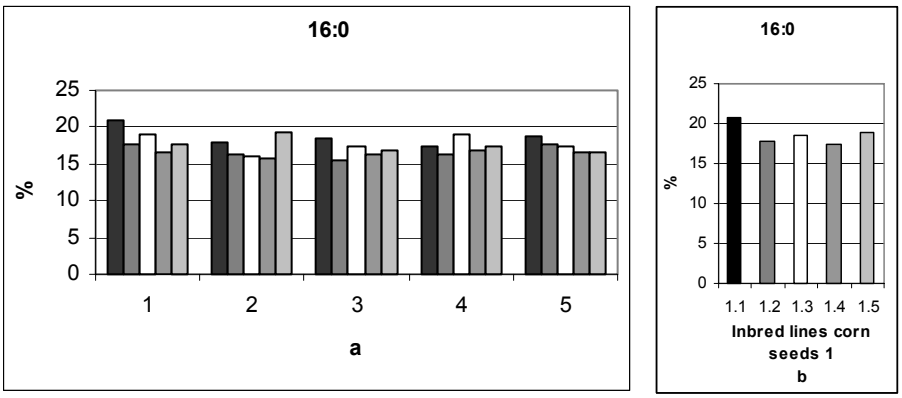

Fig. 1. Saturated fatty acids from inbred lines corn seeds 1–5.

The results (%) for fatty acids noted as 16:0, 18:2 and 18:1 (Table 1) for the five inbred lines corn seeds (1–5) are presented in Fig. 1 and Fig. 2. The black bars represent the results for corn seeds nucleus TC 209(1.1), TC 316(2.1), TC 243(3.1), TB 367(4.1), D 105(5.1) and the other 4 colored bars are the results for inbred lines

corn seeds 1.2, 1.3, 1.4, 1.5 obtained in the experiments of TC 209 (1.1) with the cytoplasm types: 2(cit T 248), 3(cit TB 329), 4(cit TC 177) and 5(cit TC 221) (Fig.1.b) and so on; 1.1 is compared with 1.2, 1.3, 1.4 and 1.5 seeds; 2.1 with 2.2–2.5; 3.1 with 3.2–3.5; 4.1 with 4.2–4.5; 5.1 is compared with 5.2–5.5 seeds (Fig. 1.a).

The results indicate that the dominant saturated fatty acids were palmitic acid, 15.83–20.81% (Fig. 1) and stearic acid 3.2–3.7%. The most present fatty acid in corn seeds was linoleic acid, belonging to polyunsaturated fatty acids, with amounts of 45.06%–62.34%. Oleic acid was the second most abundant unsaturated fatty acid, monounsaturated (MUFA), in corn seeds, with amounts ranging from 15.03% to 23.59% (Fig. 2). Oleic acid concentration was strongly negatively correlated with that of linoleic acid, suggesting that selection of genotype may be improved functional and nutritional qualities of this [6]. Very small amounts of linolenic (18:3), palmitoleic (C16:1), arachidic (C20:0), behenic (C22:0), erucic (C22:1) and lignoceric (C24:0) acids have been determined (<1%).

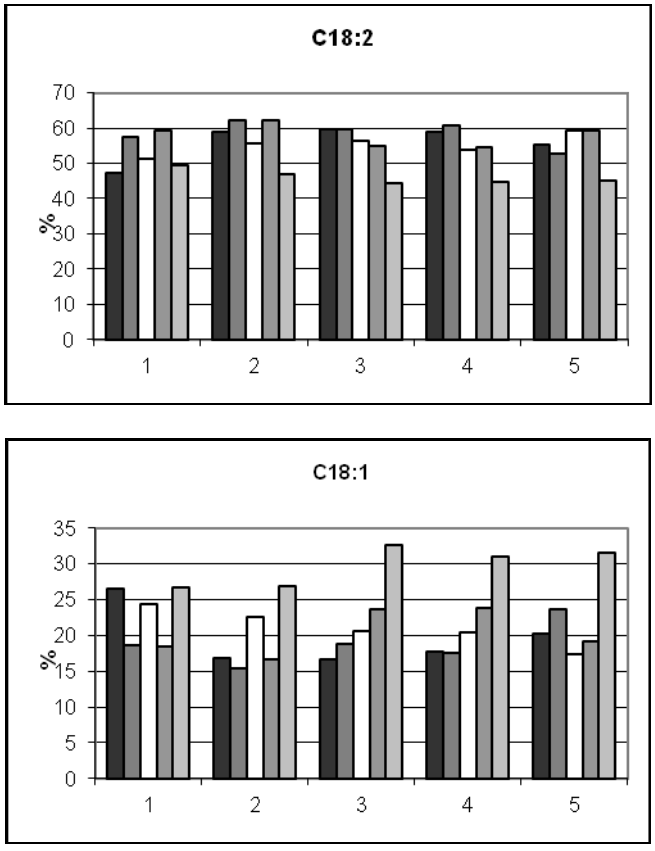

Fig. 2. Unsaturated fatty acids from inbred lines corn seeds 1–5.

CONCLUSIONS

The GC/MS method showed good precision in the analysis of fatty acids from grain seeds. The highest value of fatty acid was linoleic acid in the corn seeds, followed by palmitic acid. By choosing inbred lines with good quality traits can improve the nutritional and functional quality of the corn seeds. PUFA and MUFA composition of corn creates a beneficial effect for heart disease risk reduction. These effects may be due to the unique combinations of PUFA and MUFA found in the corn oil, but may also be the result of phytosterols from corn oil [6].

Acknowledgements. The authors thank Prof. dr. Ioan Haş for providing the corn inbred lines.

REFERENCES

1. CHRISTIE, W.W., Preparation of lipid extracts from tissues, In: *Advances in Lipid Methodology – Two*, Oily Press, Dundee, 1993, pp.195–213.
2. COCAN, D., ELENA HORJ, MONICA CULEA, VIOARA MIRESAN, ADELA PINTEA, Variation of free fatty acid in rainbow trout *(Oncorhynchus mykiss)* plasma during spring-summer seasons, *Bulletin UASVM Agriculture*, 2010, **67**, 137–141.
3. IVERSON, S.J., SHELLEY L.C. LANG, MARGARET H. COOPER, Comparison of the bligh and dyer and folch methods for total lipid determination in a broad range of marine tissue, *Lipids,* 2001*,* **36**, 1283–1287.
4. PINTEA, A., A. VARGA, P. STEPNOWSKI, CARMEN SOCACIU, MONICA CULEA, H.A. DIEHL, Chromatographic analysis of carotenol fatty acid esters in *Physalis alkekengi* and *Hippophae rhamnoides, Phytochem. Anal*., 2005, **16**, 188–195.
5. POP, C., L. BARA, ELENA HORJ, ANDREEA IORDACHE, C. LASLO, MONICA CULEA, Determination of free fatty acids in sausage meat, *Bulletin UASVM Agriculture*, 2010, **67**, 373–377.
6. SUDAR, R., I. BRKIĆ, ZORICA JURKOVIĆ, TATJANA LEDENČAN, VLATKA JURKOVIĆ, D. ŠIMIĆ, Fatty acids composition of oil from OS corn hybrids, *Proceedings. 47th Croatian and 7th International Symposium on Agriculture*, Opatija, Croatia, 2012, pp. 335–339.
7. YADAV, V.K., I.S. SINGH, Comparative evaluation of maize inbred lines (*Zea mays* L.) according to dus testing using morphological, physiological and molecular markers, *Agricultural Sciences,* 2010, **1**, 131–142.