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The study of fatty acid composition in *Cockscomb* (*Celosia cristata* (L.) Kuntze)

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ABSTRACT

Cockscomb (*Celosia cristata* (L.) Kuntze) is a well-known decorative plant belonging to the *Amaranthaceae* L. family. It is used in South East Asia folk medicine as an anesthetic, anti-microbial, hemostatic, tonic and anti-inflammatory drug. Some 11 to 12 fatty acids have been identified in *cockscomb* roots, leaves, stem, herb, flowers and seeds, and their quantitative content has been determined by gas chromatography. Accordingly, in all herb samples, unsaturated fatty acids dominated, forming 68.18 to 75.93% of total content. Mostly, these compounds were accumulated in the roots. Among the unsaturated fatty acids, linoleic acid prevailed in roots, stems, herb, flowers and seeds, whereas linolenic acid prevailed in the leaves. The highest content of linoleic acid ($45.57 \pm 1.14\%$) was found in the seeds, while that of linolenic acid ($53.63 \pm 0.53\%$) was found in the leaves. The obtained results will be applied in the development of drugs on the basis of *cockscomb* herb lipophilic fraction.

INTRODUCTION

Fatty acids are natural compounds having a hydrocarbon chain and a carboxylic group. The length of chain may substantially differ. These compounds usually contain from two to thirty carbon atoms. Depending on the number of unsaturated bonds, they are divided into saturated (without double bonds between carbon atoms), monounsaturated (having one double bond) and polyunsaturated (with more than one double bond). Depending on location of the first unsaturated bond in the chain from the final carbon atom (called n or ω), polyunsaturated fatty acids may be ω -3, ω -6 or ω -9 and may be deemed to belong to the essential compounds [1,2]. For instance, oleic (ω -9), linoleic (ω -6), linolenic (ω -3), eicosadienoic acids etc. belong to such acids [2].

Fatty acids, both free and within lipids, serve as the main energy source of the organism, taking part in heat regulation processes. These compounds are necessary for the organism to build cell membranes and to synthesize hormones and enzymes. They affect the metabolism, alleviate inflammatory manifestations, enhance the histological pattern of the intestine mucous membrane, reduce atherogenic index, as well as alleviate infarction and stroke risks [1,3]. Indeed, British researchers have found that 3 g daily consumption of the sum of unsaturated fatty acids within three months

alleviated articulation pain and relieved morning constraint under rheumatoid arthritis [1].

Cockscomb (*Celosia cristata* (L.) Kuntze) belongs to the *Amaranthaceae* family (*Amaranthaceae* L.) and is characterized by its high decorative value due to its unique bright inflorescence [4,6]. Some authors treat it as a subspecies of quail grass [6,7].

Cockscomb chemical composition, according to literary data, is represented by saponins, flavonoids, betalains and amino acids [6,8]. Chinese scientists identified in *cockscomb*, grass asparagine, hyaluronic acid, and such flavonoids as cristatein and tlatlancuayin [9,10]. Its seeds were found to accumulate phenolic (5-hydroxy-7-methoxyflavone, 5-methoxy-6,7-methylenedioxyflavone, 5-hydroxy-6,7-dimethoxyflavone, 5,7-dimethoxyflavone, cochliophyllin A and kaempferol) and steroid compounds (stigmasterol, β -sitosterol), including specific saponins such as celosins A, B, C, D, cristatain and semenoside A [9].

Literary data on fatty acid composition of some *Celosia* species are fragmentary and disjointed. However, Indian researchers have identified in *cockscomb* flowers, two saturated (palmitic and stearic) and four unsaturated (palmitoleic, oleic, linoleic, linolenic) fatty acids. This herb was found to contain 1.481 g/100 g saturated fatty acids, 0.903 and 2.195 g/100 g mono- and polyunsaturated fatty acids. Moreover, the prevailing identified fatty acids were linoleic (2027.9 g/100 g),

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palmitic (1302.5 g/100 g) and oleic (897.5 g/100 g) acids [11]. Other authors noted accumulation in *cockscomb* flowers of 0.0043% lauric and 0.0019 to 0.0079% palmitic acid [12], while Iraqi scientists found 7.2-7.9% content fatty oil in *cockscomb* seeds [13]. What is more, Chinese researchers identified arachic, arachidonic, palmitic, linoleic and linolenic acids in seeds of quail grass [14], and Indian scientists identified 8 fatty acids in this herb. The result of analysis showed almost three times prevalence of unsaturated fatty acids (74.4%) over the saturated ones (20.9%). Wheat celosia leaves were found to contain much oleic and linoleic acid – 22.3 and 51.5%, respectively [15], and researchers from Nigeria identified 10 fatty acids in wheat celosia leaves. Total content of polyunsaturated fatty acids was 49.6%. Dominating compounds were linoleic (23.29%) and linolenic (26.36%) acids [16]. Nevertheless, we do not know of any comprehensive studies of *cockscomb* herb.

In Chinese traditional medicine, *cockscomb* is applied for treatment of thrombophlebitis, hypertension, headache, palsy, cataract and diabetes. *Cockscomb* flowers are also administered to alleviate stomach pain, nosebleed, hematuria and locomotorium diseases. The leaves were found to be useful for cuts, wounds, edemas [8,17], and they are used to treat dysentery, bleeding and helminthiasis [6]. In Bangladeshi traditional medicine, *cockscomb* seeds are applied in treating inflammatory and bacterial diseases of oral cavity and the eyes [17]. Its grass and roots are also used under leucorrhea [18]. Traditional medicine of India advises *cockscomb* as a tonic under overstrain, as well as for treating atherosclerosis, leucorrhea and osteoporosis [6].

The US pharmaceutical industry (Hawaii Pharm LLC) manufactures a dietary supplement on the basis of a *cockscomb* seed ethanol extract that is to be used under hemorrhoidal and uterine bleeding, and for leucorrhea [7]. In Ukraine, *cockscomb*-based drugs are absent, and this herb requires standardization. For deep phytochemical study of this plant, we analyzed its fatty acid composition.

MATERIALS AND METHODS

Plant materials

For our experiments, we used air-dried crushed roots, stems, leaves, grass, flowers and seeds of *cockscomb*. *Cockscomb* leaves were harvested in the budding phase. The herb and flowers of this plant were harvested in the flowering phase, while the roots and seeds of *cockscomb* were harvested in the fruiting phase. Herbs were collected in 2019/2020 in the Kharkiv Region, Ukraine.

Identification of plants was carried out by the professor of the Department of Botany of National University of Pharmacy, A. G. Serbin, in comparison with voucher herbarium samples. The voucher herbarium samples are kept at the Department of Chemistry of Natural Compounds and Nutritionology of the National University of Pharmacy.

Determination of fatty acids

Qualitative composition and quantitative content of fatty acids after methylation were studied by applying the gas chromatography method, using a Selmichrom-1 (Selmi, Ukraine) gas chromatograph with a flame-ionization

detector. Fatty acid methyl esters were identified by retention time as compared to Sigma Aldrich standard samples of saturated and unsaturated fatty acid methyl esters [9].

For analysis, we used the hexane fraction from tested samples of *cockscomb* herb, which were methylated after the modified Peisker method [9].

Chromatographic analysis was performed using a chromatographic column 2.5 m long, with internal diameter of 4 mm. Static phase was Inerton-treated with 10% diethylene glycol succinate. Parameters of the unit were: column thermostat temperature – 180°C, vaporizer temperature – 230°C, detector temperature – 220°C, carrier gas (nitrogen) flow rate – 30 cm³/min, sample volume – 2 mm³ hexane solution of fatty acid methyl esters. Quantitative content of particular fatty acids was calculated via the normalization method [19,20].

RESULTS

The results of experiment identified 12 fatty acids in all tested samples, except for *cockscomb* seeds – where only 11 fatty acids were found. The chromatographic fatty acid profiles of *cockscomb* raw material are revealed in Figure 1-6, respectively. The results of qualitative composition study and quantitative content determination of fatty acids of *cockscomb* raw material are shown in Table 1.

Table 1. The qualitative composition and quantitative content of fatty acids of *cockscomb* raw material

Fatty acid methyl esters	Content of fatty acids of methyl esters, % of total					
	Roots	Stem	Leaves	Herb	Flowers	Seeds
Saturated fatty acids						
Myristic	0.58 ±0.01	0.20 ±0.01	2.48 ±0.16	0.50 ±0.01	0.33 ±0.01	0.10 ±0.01
Palmitic	25.68 ±0.64	21.27 ±0.53	21.80 ±0.57	21.70 ±0.54	21.15 ±0.53	21.12 ±0.53
Stearic	3.63 ±0.09	0.87 ±0.02	0.85 ±0.01	2.38 ±0.06	2.15 ±0.05	2.78 ±0.07
Behenic	0.64 ±0.02	0.57 ±0.01	0.68 ±0.02	0.30 ±0.01	1.78 ±0.04	0.69 ±0.02
Lignoceric	1.04 ±0.03	0.78 ±0.02	0.50 ±0.01	0.75 ±0.02	1.08 ±0.03	0.20 ±0.01
Total content of saturated fatty acids	31.57 ±0.79	23.69 ±0.59	26.31 ±0.77	25.63 ±0.64	26.49 ±0.66	24.89 ±0.62
Unsaturated fatty acids						
Myristoleic	0.21 ±0.01	0.28 ±0.01	0.10 ±0.01	0.12 ±0.01	0.08 ±0.01	0.08 ±0.01
Palmitoleic	0.86 ±0.02	0.12 ±0.01	1.35 ±0.07	0.38 ±0.01	0.87 ±0.02	0.20 ±0.01
Oleic	13.65 ±0.34	14.50 ±0.36	2.89 ±0.06	18.64 ±0.47	13.30 ±0.33	28.33 ±0.72
Linoleic	45.38 ±1.13	45.02 ±1.14	15.05 ±1.13	44.55 ±1.11	38.02 ±0.98	45.57 ±1.14
Linolenic	7.43 ±0.21	15.78 ±0.39	53.63 ±0.53	5.98 ±0.15	14.70 ±0.37	0.75 ±0.02
Gondoic	0.31 ±0.01	0.13 ±0.01	0.22 ±0.01	0.14 ±0.01	1.18 ±0.03	0.18 ±0.01
Erucic	0.34 ±0.01	0.10 ±0.01	0.10 ±0.01	1.15 ±0.03	0.70 ±0.02	trace quantity
Total content of unsaturated fatty acids	68.18 ±1.70	75.93 ±1.90	73.34 ±1.72	70.96 ±1.77	68.85 ±1.72	75.11 ±1.88
Total content of unidentified fatty acids	0.25 ±0.01	0.38 ±0.01	0.35 ±0.01	3.41 ±0.09	4.66 ±0.12	-

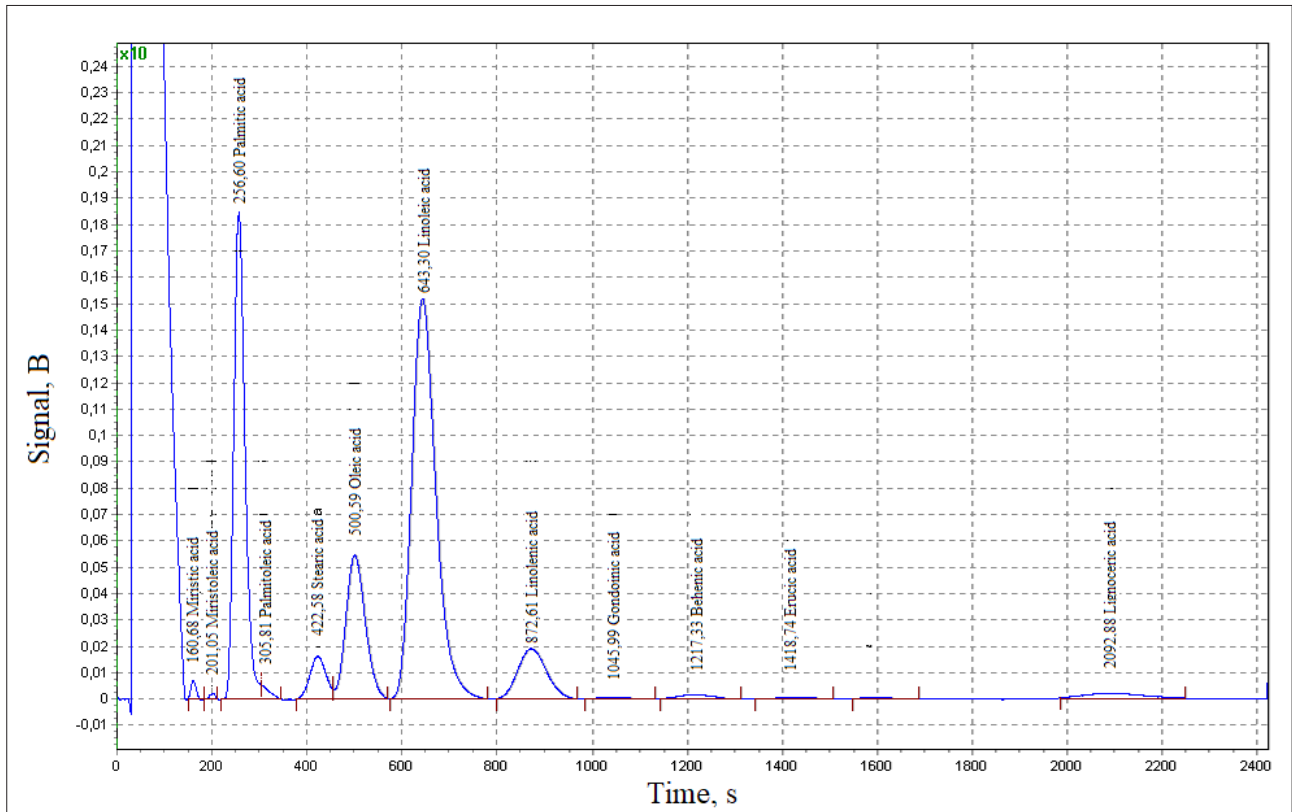


Figure 1. Chromatographic profile of fatty acid methyl esters in cockscomb roots

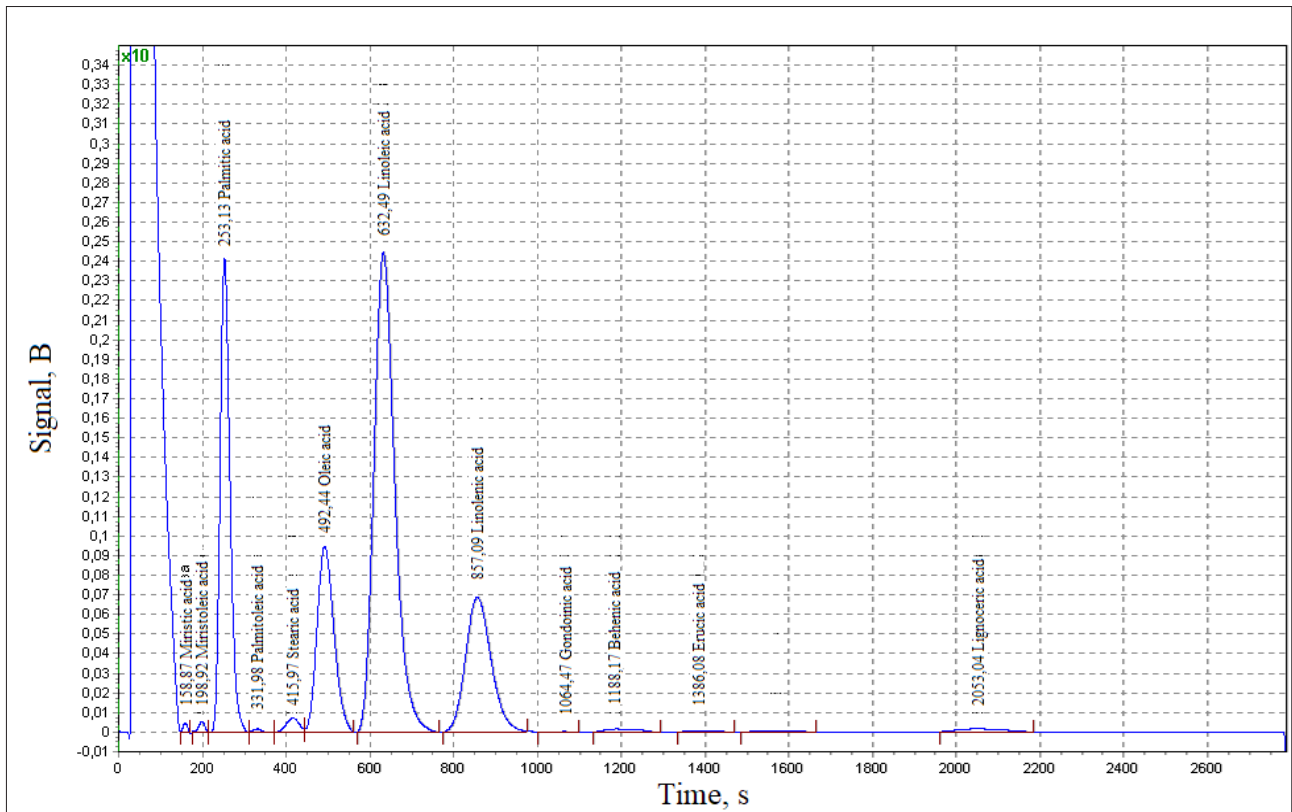


Figure 2. Chromatographic profile of fatty acid methyl esters in cockscomb stem

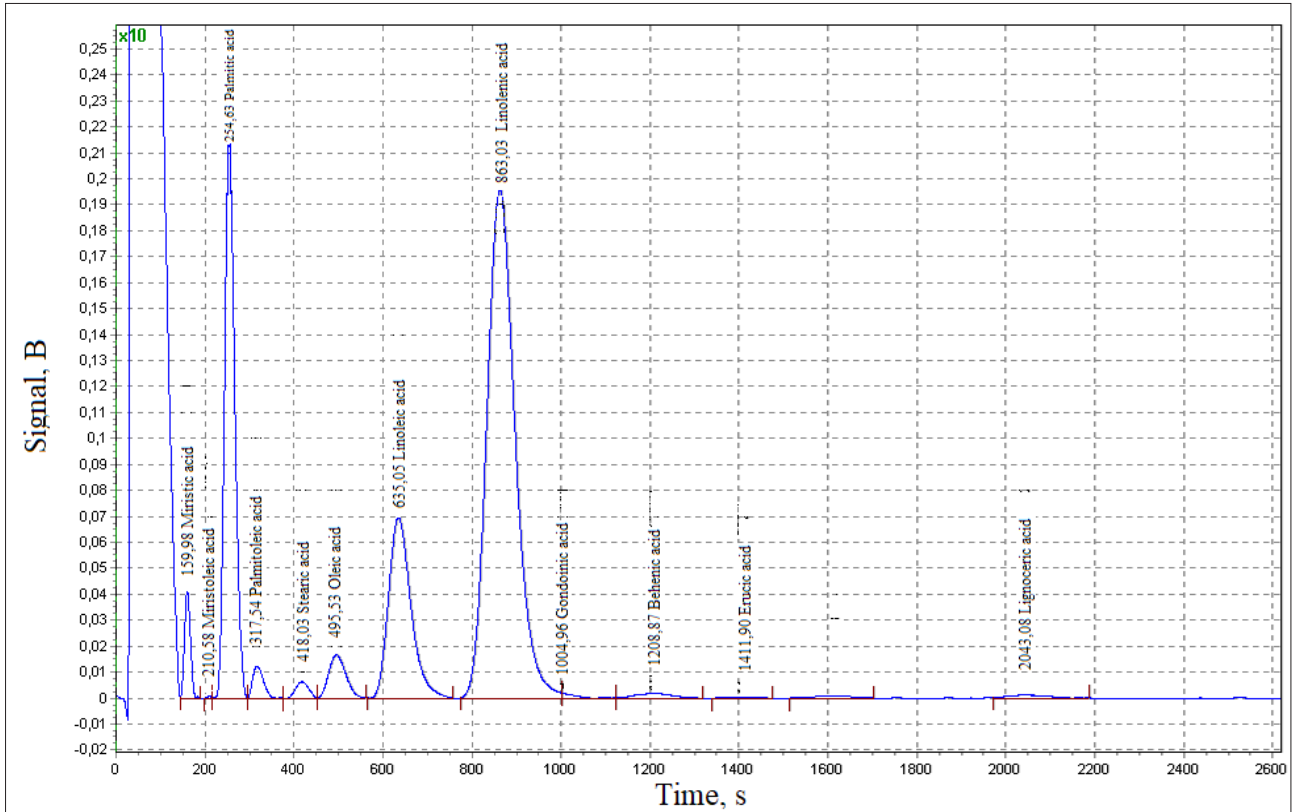


Figure 3. Chromatographic profile of fatty acid methyl esters in cockscomb leaves

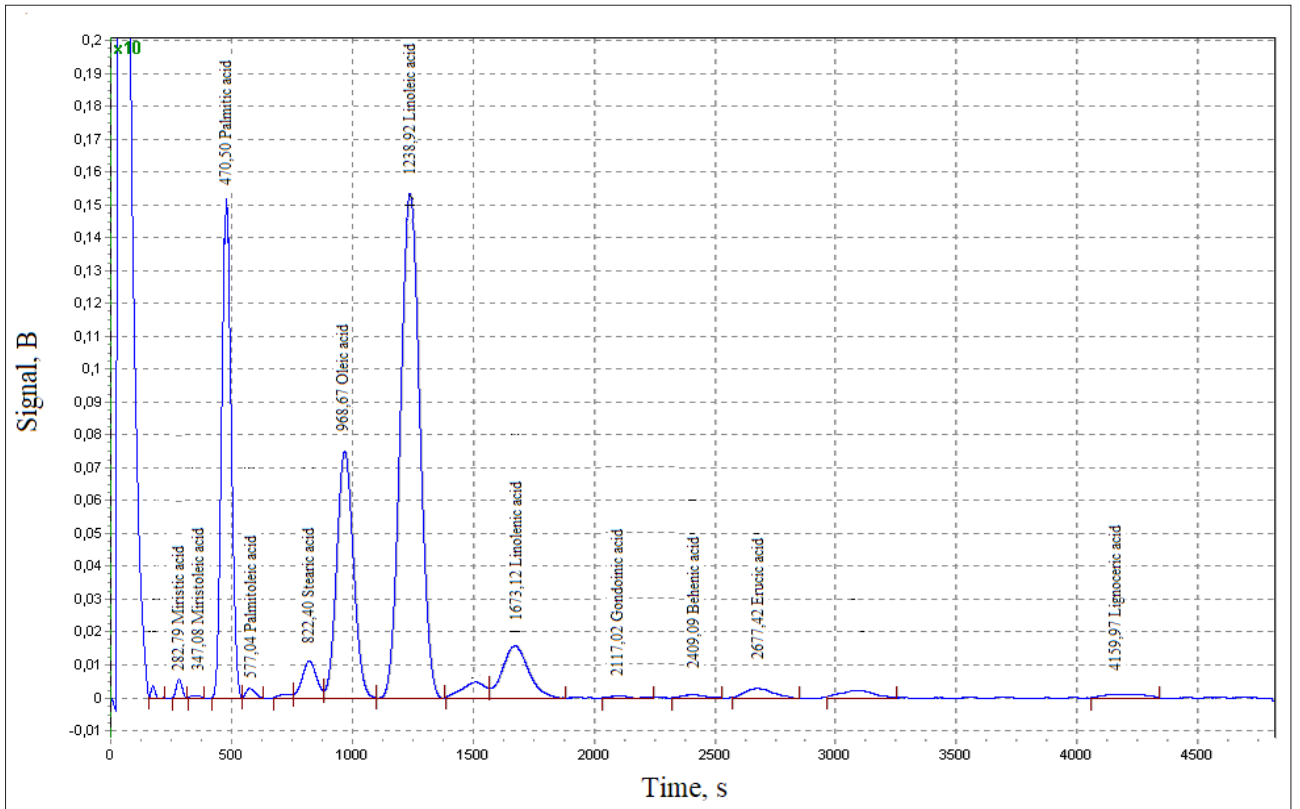


Figure 4. Chromatographic profile of fatty acid methyl esters in cockscomb herb

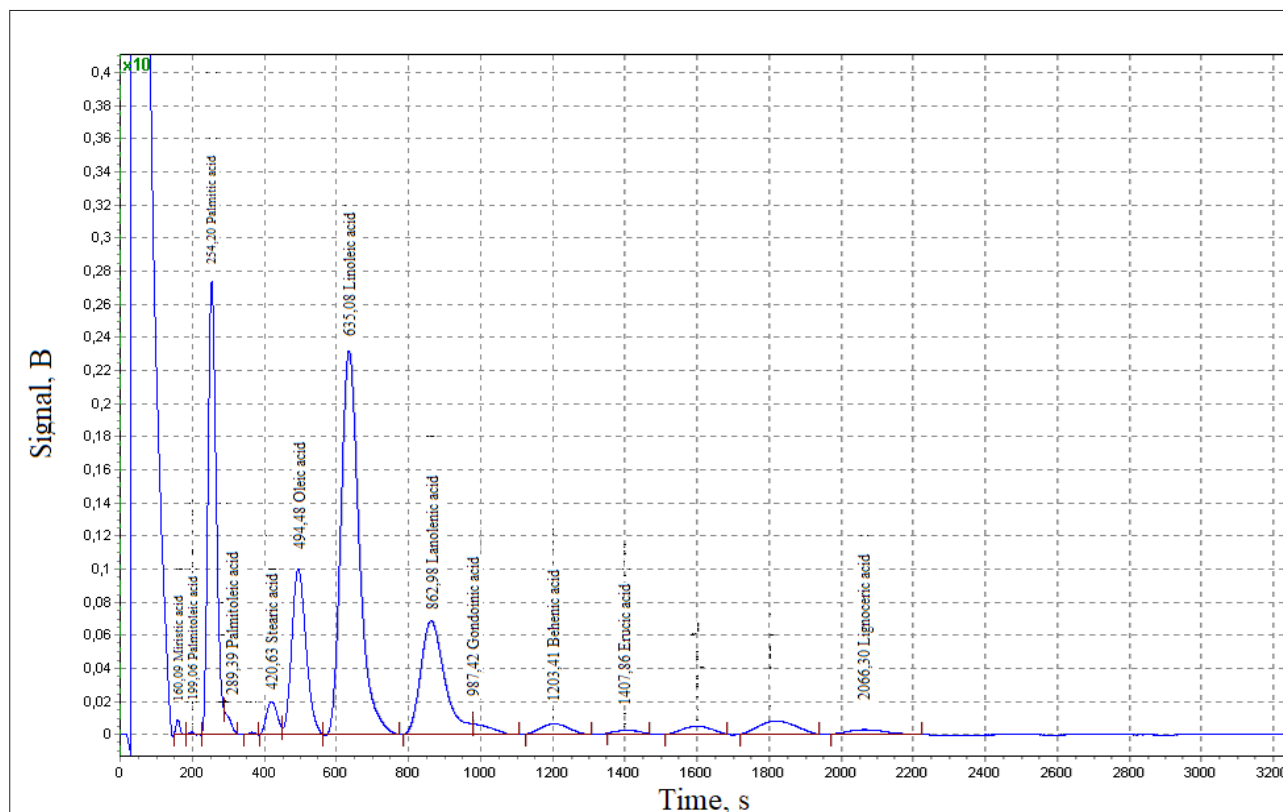


Figure 5. Chromatographic profile of fatty acid methyl esters in cockscomb flowers

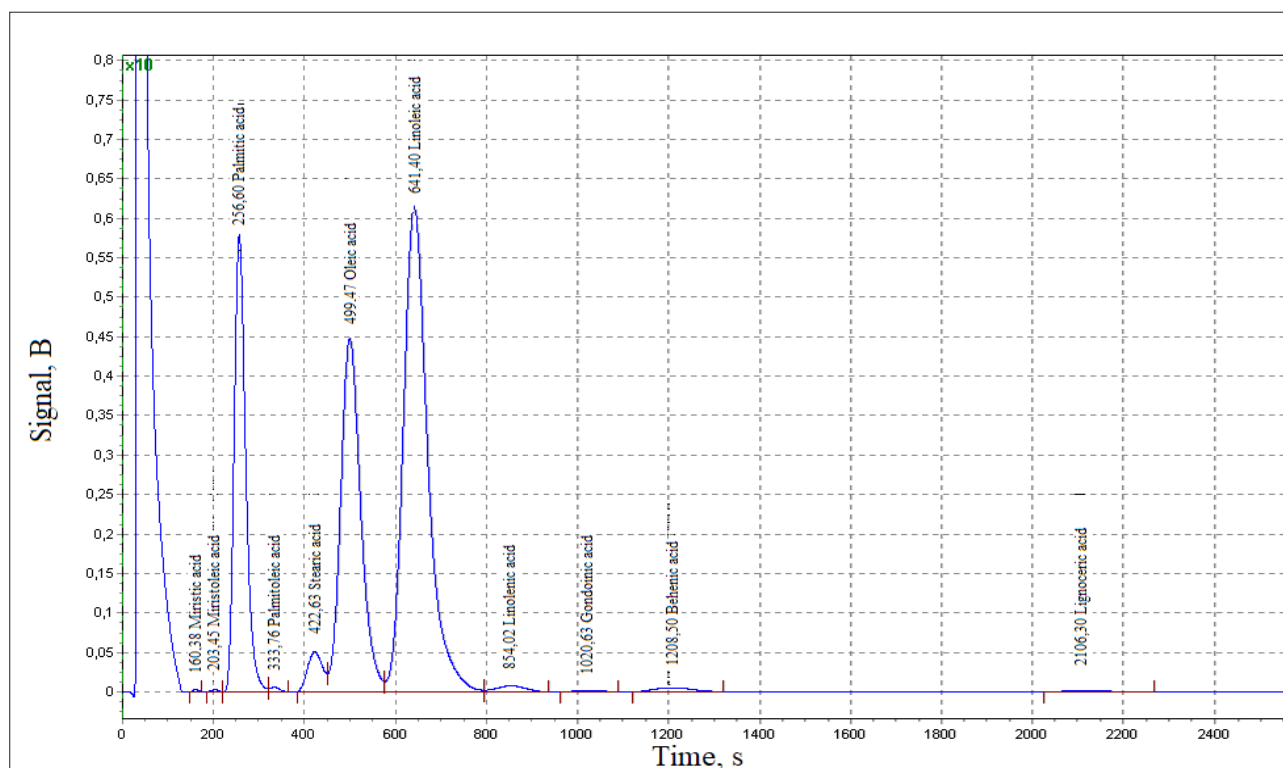


Figure 6. Chromatographic profile of fatty acid methyl esters in cockscomb seeds

The analysis results revealed that unsaturated fatty acids dominated in all tested *cockscomb* herb samples. In contrast, saturated fatty acids made about 1/4 of total identified fatty acid content in *cockscomb* stems and seeds, whereas in roots, leaves, herb and flowers saturated fatty acids make about one third of total identified fatty acids. The highest

saturated fatty acids content – $31.57 \pm 0.79\%$ – was found in *cockscomb* roots.

Among saturated fatty acids, palmitic acid dominated. Its content in tested samples of the herb was within the range 21.12 to 25.68%. This compound especially accumulated in *cockscomb* roots. The highest content of stearic acid,

3.63±0.09%, was also in *cockscomb* roots, while the content of stearic acid was almost identical in the plant flowers, herb and seeds – 2.15±0.05, 2.38±0.06 and 2.78±0.07%, respectively. We noted in the course of the experiment that *cockscomb* roots and flowers accumulated about 1% lignoceric acid.

Cockscomb showed identically high content of unsaturated fatty acids in its stems and seeds – 75.93±1.90 and 75.11±1.88%, respectively. The content of unsaturated fatty acids in roots, leaves, herb and flowers was somewhat less, being 68.18±1.70, 73.34±1.72, 70.96±1.77 and 68.85±1.72%, respectively.

The prevailing unsaturated fatty acid in roots, stems, herb, flowers and seeds was linoleic acid, whereas linolenic acid prevailed in the leaves. The content of this compound in roots, stems, flowers and seeds was nearly the same. The highest content of linoleic acid was in tested plant seeds at 45.57±1.14%. Its content in leaves was almost three times less, being 15.05±1.13%.

Linolenic acid content was the highest in *cockscomb* leaves, at 53.63±0.53%, whereas in the stems and flowers, the content of linolenic acid was similar and 3.5 times less than in the leaves. The content of linolenic and oleic acids in the stem was 15.78±0.39%, while in the flowers it was 14.70±0.37%. The seeds of *cockscomb* held the least accumulation of this compound (0.75±0.02%).

We found substantial content of oleic acid in the seeds, this being 28.33±0.72%. The quantitative content of oleic acid in *cockscomb* roots, stems, herb and flowers was within the range 13.30±0.33 to 18.64±0.47%, and was almost twice less than in leaves.

The tested *cockscomb* raw material samples contained no more than 0.87% myristic, myristoleic, behenic, gondoic and erucic acid.

DISCUSSION

When comparing the obtained results with the literature data, it was found that unsaturated fatty acids predominated in *cockscomb* flowers of Ukrainian origin, and saturated fatty acids in *cockscomb* flowers of Indian origin. In both samples of raw materials, among the unsaturated fatty acids, linoleic dominated, and among the unsaturated – palmitic acids dominated [11,12]. Lauric acid was identified only in the *cockscomb* flowers of Indian origin [12]. In the *cockscomb* flowers grown in Ukraine, this compound is absent. The *cockscomb* seeds of Ukrainian origin contained more fatty acids than the seeds of this plant of Iraqi origin [13]. No data were found on the fatty acid composition of roots, leaves, stems and herb of *cockscomb*.

CONCLUSION

The performed study in fatty acid composition of *cockscomb* herb did not reveal any substantial differences in qualitative composition and quantitative content of fatty acids. The results have shown that unsaturated fatty acids prevailed in *cockscomb* roots, leaves, stems, herb, flowers and seeds. Their content was 3-4 times, depending on herb type, higher than that of saturated fatty acids.

We noted that the prevailing fatty acid in roots, stems, herb, flowers and seeds of the tested plant was linoleic acid, with its content ranging from 38.02 to 45.57%. The major fatty acid in the leaves was linolenic acid. At the same time, a substantial content of oleic acid was found in almost all types of tested herb, which is not contradictory to information specified in literature sources.

The obtained results will be of use in the development of drugs based on *cockscomb* herb.

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