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## COMPOSITION AND PROPERTIES OF GLYCERIDE OIL FROM MILK THE SEEDS EXPELLER WHITE THISTLE ( *SILYBUM MARIANUM* L. )

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**Abstract:** This study investigates the composition, total phenolic content and antioxidant properties of a glyceride oil extracted from milk thistle seeds (*Silybum marianum* L.) expeller. In the lipid fraction of the oil (0.9%) by GC analysis, 10 fatty acids were identified, the main ones being oleic acid (43.41%, linoleic acid (11.90%) and palmitic acid (10.29%). The content of total phenols is 0.48 mg GAE/g oil, and the antioxidant activity relative to DPPH is 35.54 mM TE / g oil. The results obtained show that the glyceride oil extracted from milk thistle seeds expeller can be used in making healthy food products, subject of the following studies, thus respecting the principles of the circular economy.

**Keywords:** *Silybum marianum* L, expeller, fatty acids, total phenolics, antioxidant activity

### INTRODUCTION

Food production leads to various impacts on the environment, some of which are pollution of water basins and increased carbon emissions. The goals and tasks that the circular economy (CE) sets itself is gaining more and more influence around the world as a way to overcome the current production and consumption model (Ghisellini, P., Cialani, C., & Ulgiati, S., 2016). Its application aims to use the materials first as raw materials for the production of food products, then to process them again as recycled materials and finally as a source of energy. This is also the basic principle of the circular economy - regeneration and circulation. The waste of food industry should not be considered like such, and as raw material which may be included in the production of others products. Despite the growing interest around the circular economy concept, there are currently very few food producers who close the entire production cycle by minimizing food waste.

During the production of vegetable oils by pressing (sunflower, canola, olive, palm, etc.), a waste expeller product is obtained, which is mainly used for the production of fodder.

Bulgaria is rich in herbs with medicinal properties used to treat a number of physiological diseases. Milk thistle (*Silybum marianum* L.) of *Asteraceae* family is among them. The plant is found in the Mediterranean part of Europe, including in our country up to 1000 m above sea level.

In some countries of Europe, Africa, Asia and South America, milk thistle is grown commercially as a medicinal plant because the silymarin contained in it has been shown to stimulate liver regeneration (Polyak, S., Morishima, J., Lohmann, C., Pal, V., Lee, S., Graf, T., & Oberlies, H. 2010; Veres, T., & Tyr, S. 2012). Milk thistle glyceride oil is a valuable dietary product used in the treatment of viral hepatitis and cirrhosis (Elwekeel, A., Elfishawy, A., & AbouZid, S. 2013). Its beneficial properties are due to the contained tocopherols, carotenoids, polyunsaturated fatty acids.

Inserting oils of non-traditional oil-producing raw materials (linseed, pomegranate and milk thistle seeds, saffron, pumpkin and grape seeds) into food products has been increasing in recent years (Shaker, E., Mahmoud, H., & Mnaa, S. 2010; Dabbour, R., Al-Ismael, K., Takruri, R., & Azzeh, F. 2014). These oils contain mono and polyunsaturated fatty acids, tocopherols, carotenoids, phenolic and other compounds with proven health benefits (Garjani, A., Fathiazad, F., Zakheri, A., Akbari, N., Azmarmie, A., Andalib, S., & Maleki-Dizaji, N. 2009).

The waste (expeller) obtained after pressing milk thistle seeds to obtain glyceride oil is most often included in the composition of feed mixtures. Therefore, the aim of this study is to establish the composition of the lipid fraction of the secondary glyceride oil extracted from milk thistle expeller and determine its antioxidant properties.

### MATERIALS AND METHODS

An expeller, a waste product obtained during the cold pressing of milk thistle seeds, was used (*Silybum marianum* L.)

*n*-hexane was used for extracting secondary glyceride oil by Soxhlet extraction apparatus. The oil is stored in dark coloured vials and at 4° C until analysis. The yield of extracted oil was calculated relative to dry mass.

Fatty acid composition of the lipid fraction was determined after pre-esterification of the sample with 2% methanolic KOH at 50°C (ISO 12966-2:2011). The fatty acids methyl esters are purified by 32 thin layer plate chromatography with Silica gel 60 G Merck (20 cm x 20 cm, layer thickness 0.2 mm) with mobile phase *n* - hexane:acetone = 100:8 (ISO 12966-2:2011).). For GC analysis was used Hewlett Packard 5890 A apparatus with EC 30-Wax capillary column and flame ionization detector under the following conditions: column temperature from 130°C (4 min) with 15°C/min to 240°C (5 min); temperature of the injector 250°C, carrier gas - hydrogen, speed 0.8 cm<sup>3</sup>/min; split 50:1. The fatty acids are identified by comparing retention time of a standard mixture of fatty acids esters acc. to the conditions of the standard (ISO 12966-2:2014).

For the determination of total phenols in the oil is done sample preparation acc. to the method of (Parry, J., Hao, Z., Luther, M., Su, L. , Zhou, K., & Yu, L. 2006).

The general phenolic content of the secondary oil obtained from the expeller was determined by Folin-Ciocalteu reagent.

Determination of antioxidant activity was acc. to a method described by Parry, W., Cheng, Z., Moore, J., & Lucy, L. 2008. The obtained results are presented as Trolox equivalents mMTE / g oil.

## RESULTS AND DISCUSSIONS

The yield of extracted glyceride oil from milk thistle expeller is 0.9% (v/w).

The chemical composition of the glyceride oil is presented in Table 1. The data show that 17 compounds were identified, which is 79.22% of the total composition. The main fatty acids in the lipid fraction of the oil (over 2%) are: oleic acid (43.41%), linoleic acid (11.90%), palmitic acid (10.29%), stearic acid (6.19%) and eicosanoic acid (3.69%). The amounts of fatty acids identified differ from literature data (Dogan, G., Kara, N., Gur, S., & Eyup Bagci, (2022) and prove that the content of oleic acid in the oil is 30.2%, and that of linoleic acid is 50.5%.

Table 1. Chemical composition of glyceride oil of milk thistle seed expeller

| No  | Compounds                    |                    | RT <sup>a</sup> , min | RI <sup>b</sup> | Composition, g/100 g |
|-----|------------------------------|--------------------|-----------------------|-----------------|----------------------|
| 1.  | 14:0                         | Myristic acid      | 22.78                 | 1726            | 0.24 ± 0.0           |
| 2.  | 16:1                         | Palmitoleic acid   | 26.49                 | 1904            | 0.18 ± 0.0           |
| 3.  | 16:0                         | Palmitic acid      | 27.01                 | 1923            | 10.29 ± 0.09         |
| 4.  | 17:0                         | Margaric acid      | 28.87                 | 2025            | 0.17 ± 0.0 1         |
| 5.  | 18:2                         | Linoleic acid      | 30.35                 | 2096            | 11.90 ± 0.10         |
| 6.  | 18:1                         | Oleic acid         | 30.62                 | 2101            | 43.41 ± 0.42         |
| 7.  | 18:0                         | Stearic acid       | 30.88                 | 2123            | 6.19 ± 0.06          |
| 8.  | 20:0                         | Eicosanoic acid    | 34.25                 | 2330            | 3.69 ± 0.03          |
| 9.  | 22:0                         | Docosanoic acid    | 37.42                 | 2531            | 1.17 ± 0.01          |
| 10. | 24:0                         | Tetracosanoic acid | 40.40                 | 27.36           | 0.45 ± 0.0           |
| 11. | 1-Monasterin                 |                    | 41.82                 | 2840            | 0.12 ± 0.0           |
| 12. | $\alpha$ -Glyceryl linoleate |                    | 46.0 0                | 3086            | 0.91 ± 0.0           |
| 13. | $\beta$ -Sitosterol          |                    | 48.10                 | 3292            | 0.50 ± 0.0           |

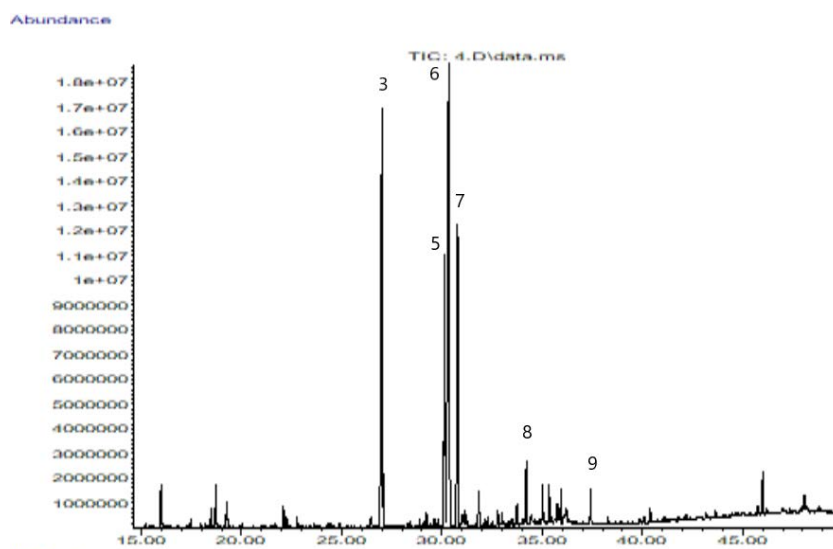


Fig.1. Chromatogram of glyceride oil from the milk thistle seeds expeller.  
(peak numbers correspond to those in Table 1)

This shows that in the expeller remain a significant amount of these acids. Therefore, it can successfully be used in obtaining food products with functional properties, respecting circular principle economy - regeneration and circulation. The use of waste raw materials from food industry for biological active substances extraction prolongs the life cycle of waste products and limits the negative human impact on the environment.

The differences in the amounts of identified acids in milk thistle oil with those in the literature can be explained by the influence of soil and climatic conditions, the method of obtaining the oil and analysis methods (Parry, W., Cheng, Z., Moore, J., & Lucy, L. 2008); Kosinska, A., & Karamac, M. 2006).

A distribution of chemical identified compounds (% of composition) is presented in Fig. 2. The data show that polyunsaturated fatty acids predominate in the lipid fraction (70.05%) followed by saturated (28.02 %).

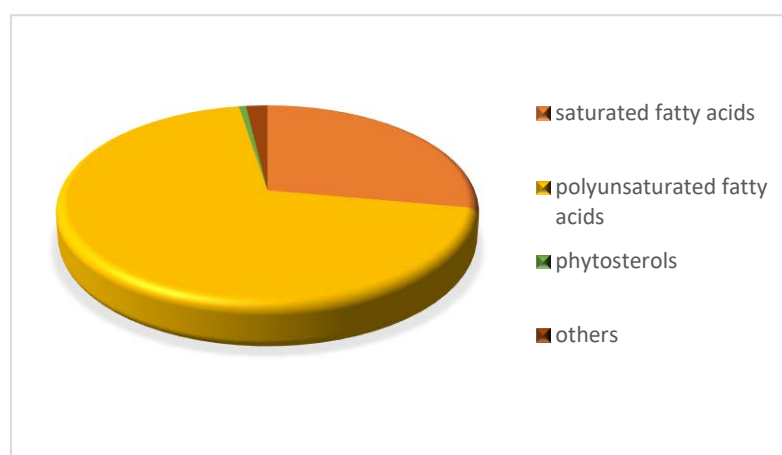


Fig. 2. Distribution of components by compound groups in glyceride oil obtained from milk thistle seed expeller.

The content of total phenolics and antioxidant activity of milk thistle seed expeller oils was determined. The value of total phenols in the oil is 0.48 mg GAE/g.

In comparison, the values of total phenolics of glyceride oil of milk thistle seeds is 1.16 mg GAE/g (Dabbour, R., Al-Ismael, K., Takruri, R., & Azzeh, F. (2014). This gives a reason to propose the use of milk thistle seed expeller oil as an additive in the development of healthy foods.

Antioxidant activity determined by DPPH method of the milk thistle seed expeller oil is 35.54 mM TE/g. This value is lower than the data of Samah, M., Ghzaïel, I., Mahmoudi, M., Mighri, H., Pires, V., Vejux, A., Martine, L., Pais de Barros, J., Prost-Camus, E., Lizard, G., & Abdellaoui, R. (2024) for milk thistle seed oil (237.79 mg TE/g). It is known that conditions such as soil and temperature, interactions between genotype and growing conditions, mechanical milling during seed oil processing as well as storage can significantly change the chemical composition.

## CONCLUSION

The glyceride oil extracted from the milk thistle seeds expeller contains polyunsaturated fatty acids and has antioxidant properties, which is why it can be successfully used as a raw material in the production of foods with functional properties.

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