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Original paper

Characterization of some vegetable oils in order to use them in the treatment of the varicose veins

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Abstract The aim of the paper was to conduct a study regarding the composition of saturated and unsaturated fatty acids for a series of vegetable oils in order to correlate it with their antioxidant activity. The oils were then used in the development of hydrogels based on nanostructured lipid carriers (NLC) with therapeutic effect on varicose veins. Several vegetable oils (pumpkin seed oil, wheat germ oil, evening primrose seed oil, castor seed oil, grape seed and jojoba oil) were analyzed by gas chromatography coupled with mass spectrometry (GC-MS), establishing the profile of saturated and unsaturated fatty acids. Antioxidant activities were determined by the FRAP (Ferric Reducing Antioxidant Power) method and were correlated with the composition ratio of saturated and unsaturated fatty acids. It has been established that pumpkin seed oil and wheat germ oil are most indicated in the development of NLC-based hydrogels.

Keywords Vegetable oils, NLC, GC-MS chromatography, antioxidant activity.

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Introduction

In recent years, the importance of vegetable oils obtained by cold pressing has increased not only for their fatty acid content but especially for their frequent use in the pharmaceutical and cosmetic industries [1]. Although medicinal plants have been used for millennia as the main source of treatment, their potential in skin therapy associated with the efficiency of nanostructured distribution systems has yet to be fully realized [2].

Varicose veins, a source of a number of serious health complications, are manifestations of chronic venous disease affecting over 20% of the adult population in the United States, 25% in the UK and 25% in Romania [3, 4].

Due to the disadvantages of conventional therapy, there is an obvious requirement for the development of new alternatives based on nanostructured formulations for topical use that contain natural active ingredients and lead to a coupled action – high therapeutic performance and improved safety profile – for the treatment of varicose veins [5, 6].

The fatty acids found in vegetable oils are contained in cell membranes and neuronal membranes and are useful for supplementing and normalizing unbalanced diets in adults and children and are often used in pharmaceuticals and cosmetics as a vehicle for transporting various active substances [7, 8, 9]. Several vegetable oils (pumpkin seed oil, wheat germ oil, evening primrose seed oil, castor seed oil, grape seed oil and jojoba oil) have been selected for cosmetic use based on their composition and its correlation with their antioxidant activity [7].

Beneficial for skin and hair's health, pumpkin seed oil is rich in Vitamin E, zinc, Omega-3 fatty acids, vitamin K and iron. It also has a rich content of phytosterols which plays an important role in lowering inflammation in the body and helps reduce cholesterol levels [5]. The large amount of Vitamin E improves the skin tonus and helps the skin heal and regenerate faster [10]. Pumpkin oil removes the feeling of heaviness and fatigue of the legs, reduces swelling and inflammation and gives firmness and elasticity to the blood vessel walls.

Wheat germ oil normalizes blood vessel volume and blood flow, beneficially influences microcirculation in the body and strengthens blood vessel walls [10].

Recent studies point out that evening primrose seed oil is beneficial for a variety of conditions characterized by inflammation, pain and swelling or decreased blood flow thus being indicated in the treatment of varicose veins [3, 9].

Castor oil has been intensely studied for its antibacterial, antiviral and antifungal action due to the rich levels of antioxidants. Used as a compress along with baking soda, castor oil improves blood circulation and the "spider web" appearance of the veins and varicose veins [11].

Containing over 70% of polyunsaturated acids (especially conjugated linoleic acid and alpha linolenic acid), flavonoids, vitamin E, complex antioxidants, grape seed oil strengthens the structure of blood vessels, reduces inflammation, improves blood circulation, being widely used in the treatment of varicose veins [12].

Jojoba oil is not considered an oil itself, but a wax in liquid form. It is obtained by cold pressing of the crushed seeds and it's made from the ester that is formed from fatty alcohols. The myristic acid present in the jojoba oil gives its anti-inflammatory properties, with a deep moisturizing effect, balances the skin's processes and regenerates the skin tissue, offering the skin's elasticity and softness [13].

The purpose of this paper is to conduct a study regarding the composition of saturated and unsaturated fatty acids for the above mentioned oils and its correlation with antioxidant activity in order to use them as a liquid lipid matrix in new nanostructured lipid carriers (NLC) [14, 15, 16].

Materials and Methods

Materials

A series of vegetable oils were obtained by cold pressing of pumpkin seeds, wheat germ, evening primrose, castor and grape seeds harvested from our own organic crops and jojoba oil was purchased from the store.

Methods

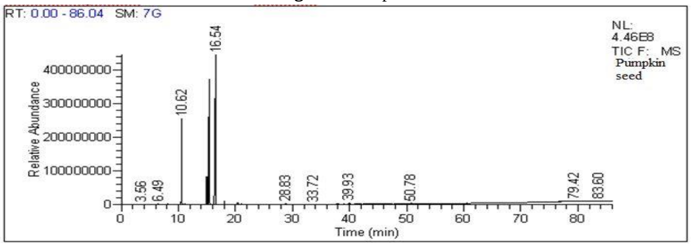
The determination of fatty acids was performed by gas chromatography coupled with mass spectrometry (GC-MS) after derivatization to methyl esters by transesterification of the triglycerides present in the studied oils with methanol in an acidic environment, using a gas chromatograph Termo-GC having a mass spectrometer detector DSQ P 5000. A Macrogol 2000 column was used, with the following characteristics: $\Phi = 0.25$ mm, $l = 30$ m, helium carrier gas, flow rate = 1 mL/min, injection temperature = 25°C, column temperature = 250°C; in order to identify the peaks corresponding to the analyzed compounds, the NIST spectrum library was available.

The antioxidant activity was determined by the FRAP method (Ferric Reducing Antioxidant Power) [12]. The method follows the principle of reducing the complex of phenanthroline or tripyridyltriazine in the presence of oxidants to colored ferrous complexes, with readable absorbents at $\lambda = 510$ nm or 593 nm. Acetone solutions of oils consistent with literature data were used.

Results and Discussion

The chromatograms presented in Figure 1 and Figure 2 highlight that the studied vegetable oils are rich in saturated and unsaturated fatty acids; the graphs in Figure 3 and Figure 4 also illustrate the percentage content of the main saturated and unsaturated fatty acids: oleic acid (Omega-9), linoleic acid (Omega-6), linolenic (Omega-3) acid, ricinoleic acid, 11-eicosenoic (C20:1) acid in each type of oil. The graph in Figure 5 shows the antioxidant activity for all types of oils studied.

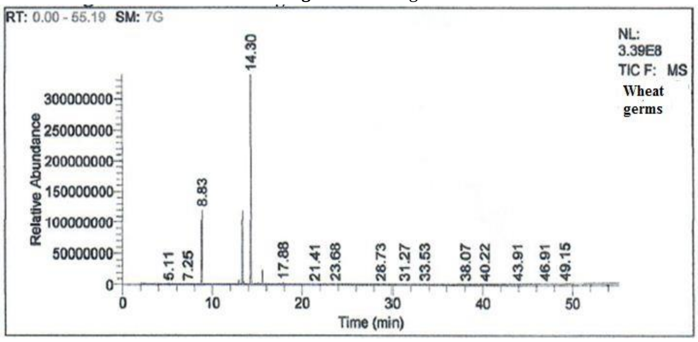
Chromatogram: Pumpkin seed oil



Pumpkin seed oil

Compound name	RT	Area %
Palmitic C16:0	10.62	14.00
Stearic C18:0	14.96	6.45
Oleic C18:1	15.41	30.73
Cis Vacc	15.49	0.67
Linoleic C18:2	16.54	46.03
Linolenic C18:3	18.04	0.52
Arachic C20:0	20.40	0.48
Ricinoleic	37.76	0.38
Squalene	39.93	0.53
----	50.78	0.22

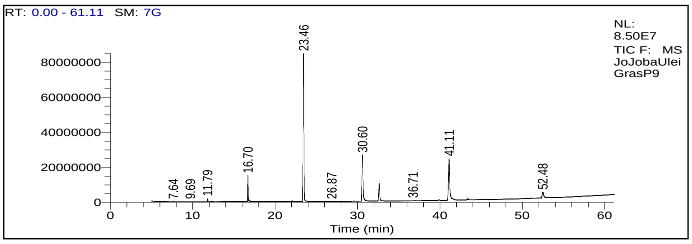
Chromatogram: Wheat germs oil



Wheat germs oil

Compound name	RT	Area %
Myristic C14:0	5.1	0.14
Pentadecanoic acid C15:0	6.86	0.08
Azelaic acid $C_9H_{16}O_4$	7.25	0.11
Palmitic C16:0	8.83	16.92
Palmitoleic C16:1	9.20	0.11
11-Hexadienoic C16:1	9.31	0.15
Heptadecanoic C17:0	10.88	0.06
Stearic C18	12.98	1.21
Oleic C18:1	13.34	18.06
Cis-Vaccenic C18:1	13.48	0.90
Linoleic C18:2	14.30	56.90
γLinolenic C18:3	15.57	3.71
αLinolenic C18:3	15.98	0.11
Arachic C20:0	17.43	0.18
11 Eicosanoic C20:1	17.88	0.59
Behenic C22:0	24.40	0.13
Lignoceric C24:0	33.52	0.12

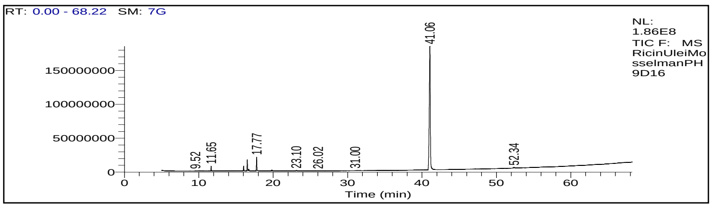
Chromatogram: Evening Primrose oil



Evening Primrose oil

Compound name	RT	Area %
Palmitic C16:0	11.80	6.64
Stearic C18	16.20	1.57
Oleic C18:1	16.71	6.30
Cis-Vaccenic C18:1	16.90	0.53
Linoleic C18:2	18.02	76.18
γLinolenic C18:3	18.94	8.10
αLinolenic C18:3	20.07	0.31
Arachic C20:0	22.64	0.15
11 Eicosanoic C20:1	23.44	0.14
Behenic C22:0	31.64	0.07

Chromatogram: Castor oil

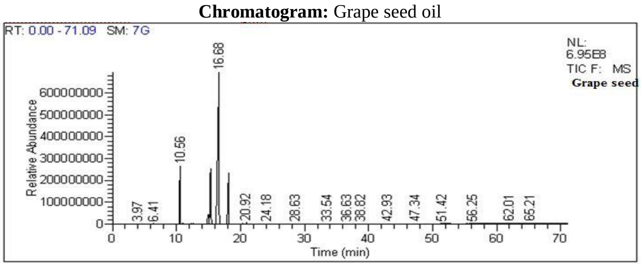


Castor oil

Compound name	RT	Area %
Palmitic C16:0	11.65	1.12
Stearic C18:0	16.03	1.32
Oleic C18:1	16.52	3.11
Linoleic C18:2	16.70	0.48
Linoleic C18:2	17.77	4.38
Linolenic C18:3	19.80	0.91
Eicosenoic C20:1	23.10	0.27
Ricinoleic acid	41.06	88.40

Figure 1. GC-MS chromatograms and fatty acids content for four (Pumpkin seed oil, Wheat germs oil, Evening Primrose oil, Castor oil) from six the studied oils.

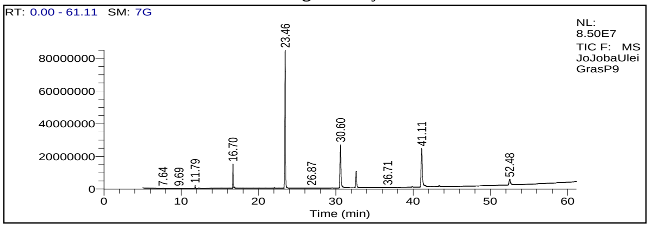
Chromatogram: Grape seed oil



Grape seed oil

Compound name	RT	Area %
Palmitic C16:0	10.56	8.37
Palmitoleic C16:1	10.86	0.08
11-Hexadienoic C16:1	10.99	0.10
Stearic C18:0	14.99	2.99
Oleic C18:1	15.36	17.07
Cis Vacc C18:1	15.48	0.50
Linoleic C18:2	16.66	59.22
Linolenic C18:3	18.13	11.33
Arachic C20:0	20.28	0.11
Eicosenic C20:1	20.92	0.22

Chromatogram: Jojoba oil



Jojoba oil

Compound name	RT	Area %
Palmitic C16:0	11.79	0.45
Oleic C18:1	16.70	4.75
Cis-Vaccenic C18:1	16.88	0.31
11Eicosenoic C20:1	23.45	45.42
11Eicosenoic-ol	30.60	18.10
Erucic C22:1	32.63	7.07
Erucic acetate	39.85	0.27
13 Docosen-ol	41.11	19.53
Nervonic C24:1	43.36	0.55
Oleyl alcool	52.48	3.55

Figure 2. GC-MS chromatograms and fatty acids content for two (Grape seed oil and Jojoba oil) from six the studied oils.

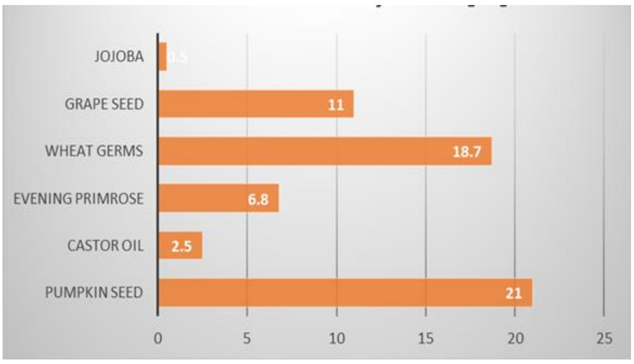


Figure 3. Total saturated fatty acids [%] content in the studied oils.

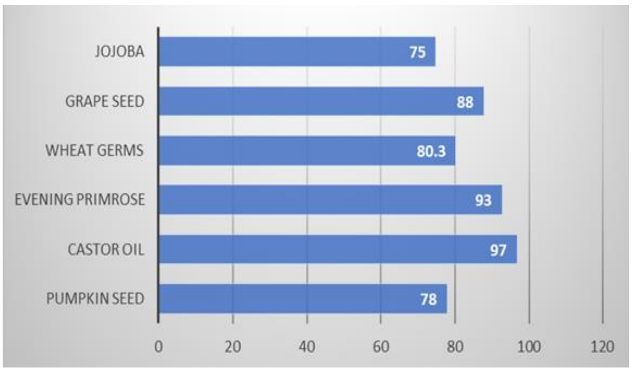


Figure 4. Total unsaturated fatty acids [%] content in the studied oils.

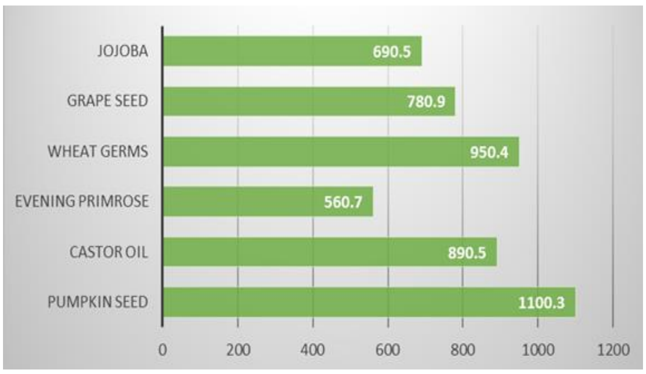


Figure 5. Antioxidant activity [mg Fe/g sample] in the studied oils.

From the presented graphs we can observe a content rich in saturated fatty acids for pumpkin seed oil (21.0%), followed by wheat germ oil (18.7%), the other studied oils having a much lower acid content of saturated fats.

Regarding the unsaturated fatty acids content, the richest is castor oil (97%) having a predominantly amount of oleic castor acid (88.4%), followed by evening primrose (93%), grape seeds (88%), germ wheat (80%), pumpkin seeds (78%) and Jojoba (75%).

The antioxidant activity determinations showed high values and a good correlation with the values of the saturated and unsaturated fatty acids content for pumpkin seed oil (1100.3 mg Fe/g) and wheat germ oil (950.4 mg Fe/g).

Conclusion

The composition study performed by gas chromatography analysis coupled with mass spectrometry (GC-MS) for pumpkin seed oil, wheat germ oil, evening primrose seed oil, castor seed oil, grape seed oil and jojoba oil showed the presence of a large variety of saturated and unsaturated fatty acids.

The antioxidant activity determination by the FRAP method showed high values for pumpkin seed oil and wheat germ oil. The above shown results for the two mentioned oils confirm their recommendation for their use in obtaining a hydrogel based on lipid nanostructures with therapeutic effect in the treatment of varicose veins.

Acknowledgments

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