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# Fatty Acids Composition by Gas Chromatography – Mass Spectrometry (GC-MS) and most important physical-chemicals parameters of Tomato Seed Oil

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***Abstract***

The objective of the study was to evaluate the fatty acids composition of tomato seed oil by Gas Chromatography combined with Mass Spectrometry and the most important physical-chemicals parameters. Tomato seed oil that was used for analysis has been obtained by cold pressed extraction method. It is known that individual fatty acids can be identified by GC because of their different retention times, the samples of tomato seeds oil were esterified to bring them into a vaporous phase, transforming the fatty acid from tomato seed oil into fatty acids methyl esters. The results showed that the major component of tomato seed oil was linoleic acid (48,2%), followed by palmitic acid (17.18%) and oleic acid (9.2%), all the fatty acids were expressed in methyl esters. It can be concluded that tomato seed oil is an excellent source of essential fatty acids omega-6 (linoleic acid) and omega-9 (oleic acid).

***Keywords***: fatty acids, tomato seed oil, gas chromatography, retention time, parameters

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## 1. Introduction

Tomato (*Lycopersicon Lycopersicum*) is a very nutritive vegetable that is cultivated throught the whole world for its fruit. Has a high nutritive value and because of the many uses in culinary domain it is one of the most important vegetable. Tomatoes are processed in manufactories to obtain products like puree, juices, ketchup, sauce, paste and tomato powder. The solid waste that remains after the juice/pulp extraction process, consists of skin, seeds, fibrous matter and cull tomato [1], represents an ecologically problem.

The industry development of tomato processing has presented problems regarding the economical area, but also technical and environmental problems, because of the amounts of resulting waste from this industry the idea of reevaluation of this wastes is very debated in our days.

Many researchers have been studying the major component of the resulting wastes, the seeds [2-9], with averages of data ranging: 2.1% crude fat; 28.3% crude fibre; 5.8% ash; 4.1 % carbohydrates and 29.8% crude protein. The tomato seeds are a source of vegetable oil, and was related that tomato seeds contains 20% oil [10]. A very important parameter of the vegetable oils is the degree of unsaturation of the fatty acids that are present in the chemically structure of the oil [7].

Some chemical compounds such as fatty acids can be separated and quantified by using gas chromatography, where the capillary system involves in cleavage the sample to prevent sample overloading on the GC. Nitrogen is usually mixed

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with the column effluent previous to the flame ionization detector (FID) to improve the characteristics of the responses [11].

Vegetable oils are known to be natural products with vegetable origin that contained mixtures of esters derived from glycerol that have chains of fatty acid with 14 to 20 carbon atoms that have different degrees of unsaturation [12].

Vegetable oils have an important functional and sensory role in food products, because of their fatty acids composition and the fat-soluble vitamins (A, D, E, and K). They are also sources of energy and essential fatty acids like linoleic and linolenic that are responsable for growth and the health of organisms [13].

Physical-chemical properties of triglyceride and its applications depend on fatty acid constituents in molecule. However, the differences appears due the chain length, unsaturation degree and position of unsaturation.

An oil that contains fatty acids with short chain have lower melting point and are more soluble in water. Whereas, the oils that contain fatty acids with longer chain have higher melting points. Unsaturated acids will have a lower melting point compared to saturated fatty acids of similar chain length [14]. Tomato processing products wins the consumers attention due to functional and nutritive benefits that tomatoes offer. Seeds of tomatoes have in composition essential fatty acids, vitamins (A,D,E,K), phytosterols and other components with an important role in nutrients that play an important role in the human health and diet. Therefore the objective of this work was to determine fatty acids concentrations of tomato seed oil (TSO) and its major physical-chemical indicators.

## 2. Material and methods

***2.1. Materials*** -Tomato seed oil obtained by cold pressed extraction method, standards of fatty acids: palmitic acid, oleic acid, arachidic acid, linoleic acid, stearic acid, behenic acid, with >99%, reagent grade, that were purchased from Merck&Co. n-hexan and Methanol·BF3 reagent from Fluka, ethanol 96%, KOH solution 0,1 and 0.5 N, Wijs reagent, acetone, HCl solution, petroleum ether from Merck&Co.

***2.2. Methods*** - GC-MS analysis. For the analysis of the methyl esters of fatty acids a Hewlett Packard HP 6890 Series gas chromatograph coupled with a Hewlett Packard 5973 mass spectroscopy detector (GC-MS) system was used. A HP-5 MS capillary column (30 m length, 0.25 mm i.d., 0.25 µm film thickness) was used for the GC system. The temperature program was set up from 50°C to 250°C with 4°C/min, both the injector and detector temperatures were 280°C and He was used as carrier gas. The injection volume was 2µL.Ionization energy EI of 70 eV was used for mass spectroscopy detector, with a source temperature of 150°C, scan range 50-300 amu, scan rate 1s-1. The mass spectra were compared with the NIST/EPA/NIH Mass Spectral Library 2.0. [15]

Physical and chemical indicators: Melting point – Boethius lamels, Unsaponified matter – extraction with petroleum ether, Refraction index – ABBE refractometer, density – pycnometer method, relative density – Mohr-Westhal balance, Acid value – KOH, saponification value – KOH, Iodine value (counted by I) – Wijs method, hydroxyl, acetyl, Hehner, Reichert -Meissl and Polenske values - , Water content – Karl Fischer titration, Total fat content – Soxhlet extraction method, Ash content – calcination method, Viscosity – capillary method with Ubbelohde Viscometer.

*Preparation of fatty acid methyl ester.* For the GC-MS analysis, the samples containing fatty acids were esterified to the more volatile methyl esters by methanol·BF3 method. 100 mg TSO or 5mL hexane solution obtained from the extraction of complexes were treated with 5mL methanol·BF3 solution and refluxed for 2 min on a waterbath, and then 5mL hexane was added; after another one minute of reflux, the solution was treated with 15mL saturated NaCl solution under vigorous stirring. The organic layer was separated and dried over anhydrous $CaCl_2$. [15]

## 3. Results and Discussion

The MS identification of compounds from derivatised TSO are presented in the **Table 1**. We can observe that the fatty acid that were identified by their time of retention from derivatised TSO are in order of their retention time: myristic acid, palmitic acid, oleic acid, linoleic acid and linolenic acid. We can also observe that the higher percentage area is registred for the linoleic acid (54.91%), which is the most important fatty acid from TSO. Were detected also unimportant compounds with very low area percentage like: decane, tetralin, hexanal dimethyl acetal.

In the chromatogram from the **Figure 1.** it is shown that if we superpose the GC-MS chromatograms for derivatised Standards of fatty acids (palmitic, oleic, linoleic) and derivatised TSO we can observe that the retention time are almost similar.

We used for GC-MS comparision of derivatised TSO three Standards of derivatised fatty acids (palmitic acid, oleic acid and linoleic acid); their retention time are presented in the **Table 2** and also the retention time of the major compounds of derivatised TSO.

***Table 1***. MS identification of compounds from derivatised TSO

| No | RT (min) | Area (Abund.min) | Area (%) | Name |
|---|---|---|---|---|
| 1 | 6.303 | 15863 | 0.147833 | Decane |
| 2 | 6.908 | 15743 | 0.146715 | Hexanal dimethyl acetal |
| 3 | 12.977 | 45003 | 0.419399 | Tetralin |
| 4 | 23.265 | 13812 | 0.128719 | Myristic acid, methyl ester |
| 5 | 26.685 | 1450646 | 13.5191 | Palmitic acid, methyl ester |
| 6 | 29.705 | 2022748 | 18.85073 | Oleic acid, methyl ester |
| 7 | 29.869 | 5892298 | 54.91247 | Linoleic acid, methyl ester |
| 8 | 30.175 | 315516 | 2.940409 | Linolenic acid, methyl ester |
| 9 | 30.58 | 116262 | 1.083488 | Oleic acid, ethyl ester |
| 10 | 30.733 | 379397 | 3.535739 | 9,12-Octadecadienoic acid, ethyl ester |
| 11 | 32.096 | 145501 | 1.355977 | 9,12-Octadecadienoic acid, ethyl ester |

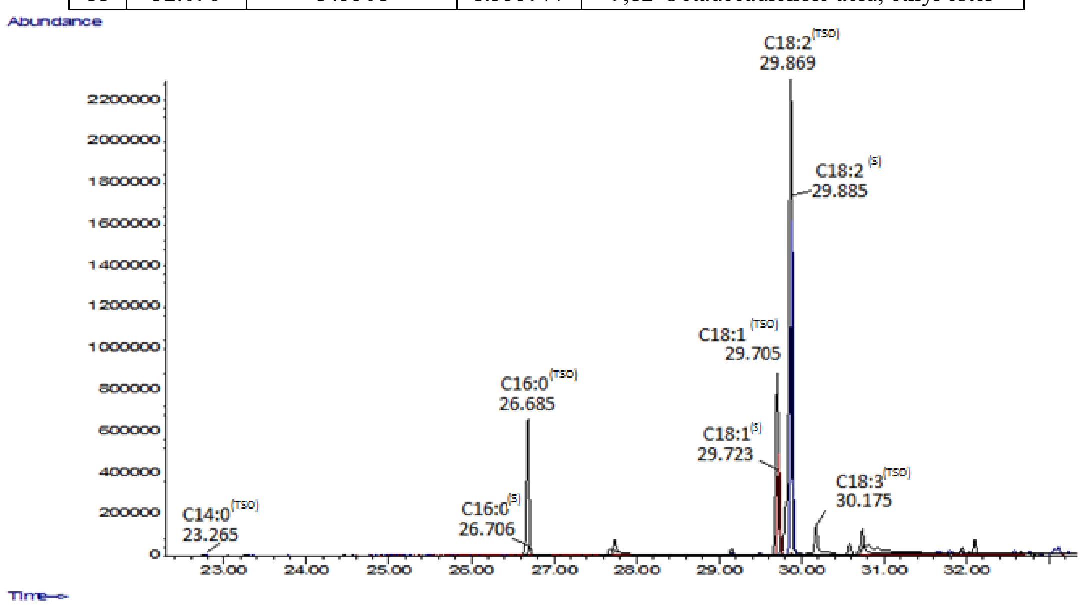


***Figure 1.*** Superposition GC-MS chromatograms for derivatised standards of fatty acids (palmitic, oleic, linoleic) and derivatised TSO

***Table 3.*** Fatty acid composition of TSO

| FAME | Concentration FAME[1] (mg/mL) | Mass of oil[1] (g) | Volume of oil[1] (mL) | Mass of FAME[1] (mg) | Concentration of FAME in TSO[1] (%) |
|---|---|---|---|---|---|
| PAME | 0.05 | 0.11 | 3.80 | 0.18 | 17.18 |
| OAME | 0.07 | | | 0.27 | 9.20 |
| LAME | 1.34 | | | 2.96 | 48.22 |

[1] – The data are presented as average of duplicates

***Table 2.*** Retention time of fatty acids methyl ester from derivatised TSO and from standards of derivatised fatty acid methyl esters

| No. | FAME | Retention time for derivatised TSO (min) | Retention time for Standards derivatised fatty acids (min) |
|---|---|---|---|
| 1. | MAME | 23.265 | - |
| 2. | PAME | 26.685 | 26.706 |
| 3. | OAME | 29.705 | 29.723 |
| 4. | LAME | 29.869 | 29.885 |
| 5. | LnAME | 30.175 | - |

After the analysis of TSO samples (derivatisated) with GC-MS, we can observe that linoleic acid, expressed in methyl esters was found out in a concentration of 48,2% compare to other bibliographic sources that admits values between 35-60%. Regarding palmitic acid in the TSO derivatised samples, we can see that the concentration of it expressed in methyl esters was 17.18%, and the literature has shown values between 9-15%. In the case of oleic acid expressed in meyhyl esters, the concentration obtained was 9.2% compared with other bibliographic sources that have values between 15-46% [2,3, 16-18].

All the derivatised TSO samples were made in duplicates, the standard deviation had small values.Also in the derivatised TSO samples was determinated myristic acid (expreseed in methyl ester), but this had a small concentration, hasn't been calibrated but was expressed like percentage of area to the other components. In the same mode was also expressed linolenic acid.

All major fatty acids have been identified by many authors [2,3,5,7,16-18]; the fatty acid composition of TSO is presented in **Table 3**. The differences in individual contents of fatty acids when compared to the bibliographic references, may be due to the cultivars used and to the cultivation or environmental factors [9].

Regarding the physical-chemical parameters of TSO, the determination results are presented in the **Table 4.** The physical and chemical parameters are very important because they are giving information about the composition of TSO, for example the melting point gives information about the degree of unsaturation of fatty acids from TSO, the refractive value is in correlation with molecular weight and degree of unsaturation of fatty acids from TSO, the density and viscosity are very important parameters, because TSO can be used as fuel after transesterification, that has the purpose to decrease viscosity of it to not damage the engine. The flash point it is also important if we want to use this oil as a fuel; water content show us the quality of TSO and ash content is important to determine the concentration of heavy metals and expresses the degree of impurification. Acid value is used to quantify the amount of acid present in TSO and shows the level of freshness for the oil, if the concentration is lower then the quality of TSO will be higher, saponification value shows the capacity of forming soaps of TSO, iodine value is important because it gives us information about the composition in unsaturated fatty acids of TSO, the value of it is high and that means that TSO has a high content of unsaturated fatty acids and this was also shown in the present study by determination of fatty acid composition of TSO using GC-MS method.

***Table 4.*** Physical-chemical parameters of TSO

| No. | Physical-chemical indicators | Value [1] |
|---|---|---|
| 1 | Melting point ($^0$C) | -3.85 ± 0.0707 |
| 2 | Unsaponified matter (%) | 1.747 ± 0.4540 |
| 3 | Refraction index $n^{20}$ | 1.4708 ± 0.0002 |
| 4 | Density $d^{20}$ | 0.877 ± 0.0007 |
| 5 | Relative density $d^{20}{}_{20}$ | 0.895 ± 0.0084 |
| 6 | Acid value (KOH) | 1.336 ± 0.281 |
| 7 | Saponification value (KOH) | 192.5 ± 0.2394 |
| 8 | Iodine value (Counted by I) | 125.83 ± 4.57 |
| 9 | Hydoxyl value | 40.33 ± 2.08 |
| 10 | Acetyl value | 8.41 ± 0.51 |
| 11 | Hehner value | 96.43 ± 1.87 |
| 12 | Reicher-Meissl value | 0.44 ± 0.14 |
| 13 | Polenske value | 0.06 ± 0.02 |
| 14 | Water content (%) | 0.14 ± 0.0173 |
| 15 | Total fat content (%) | 99.35 ± 0.0665 |
| 16 | Ash content (%) | 0.1342 ± 0.1043 |
| 17 | Flash point ($^0$C) | 307 ± 0.5773 |
| 18 | Viscosity (mPas·s) | 51.9933 ± 0.0045 |

[1] – The data are presented as average of triplicates, ± standard deviation.

The levels and values found for the most important physical-chemical indicators of TSO were in agreement with other authors [16-18].Total fat content is very high so the TSO has a small quantity of impurification products. The rest of parameters that weren't mentioned, repectively hydroxyl, acetyl, Hehner, Reichert -Meissl and Polenske parameters are used to differentiate the oils and fats.

## 4. Conclusion

TSO contains a healthy mixture of fatty acids saturated and unsaturated. The results showed that the major component of tomato seed oil was linoleic acid (48,2%), followed by palmitic acid (17.18%) and oleic acid (9.2%). Tomato seed oil an excellent source of essential fatty acids omega-6 (linoleic acid), omega-9 (oleic acid).

Physical-chemical properties of triglyceride and its applications depends upon fatty acid constituents in molecule and are very important in the determination of the composition of TSO, for example the melting point gives information about the level of unsaturation of fatty acid from TSO. The levels and values of the parameters that were determinate shown that TSO is rich in unsaturated fatty acids, has a high level of freshness cause it has a low acid value. Because of its relative high value of viscosity and low value of the flash point can be used as fuel only after transesterification. The high value of total fat content, low value of ash and water content shows that TSO is an oil with superior quality and a very low degree of impurification.

The fatty acid profile plays an important role to the chemical properties therefore this is useful knowledge for further researches. The study shows that TSO is an excellent source of essential fatty acids such as linoleic acid and oleic acid.

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***Abbreviations:*** GC-MS - Gas Chromatography – Mass Spectroscopy, TSO – tomato seed oil, PAME – palmitic acid methyl ester, OAME – oleic acid methyl ester, LAME – linoleic acid methyl ester, LnME – linolenic acid methyl ester, C14:0 – myristic acid, C16:0 – palmitic acid, C18:1 – oleic acid, C18:2 – linoleic acid, C18:3 – linolenic acid, (S) – Standards of fatty acids.

**References**

1. Sogi D.S, Bhatia R, Garg S.K, Bawa A.S, Biological evaluation of tomato waste seed meals and protein concentrate, *Food Chemistry*, **2005**, 89(1), 53–56, http://dx.doi.org/10.1016/j.foodchem.2004.01.083
2. Gad, A.M.; El-Khalafy, H.M.; Hassan, M.M.; Shoeb, Z.E. Chemical investigations on Egyptian vegetable fats and oils. XIII. The chemical constitution of some *Rosaceae, Solanaceae* and *Oleaceae* seed oils. *Grasas y Aceites*, Sevilla, **1968**, *19*(4), 139-145
3. Tsatsaronis. G.C.; Boskou, D.G. Oils and fats. Fatty acid composition of tomato seed oil. *Journal of the Association of Official Agricultural Chemists*, Washington, **1972**, *55*(3), 645-647
4. Kramer, A.; Kwee, W.H. Functional and nutritional properties of tomato seed protein concentrate. *Journal of Food Science*, Chicago, **1977a**, *42*, 207-211
5. Canella, M.; Cardinalli, F.; Castriotta, G.; Nappucci, R. Chemical properties of the seeds of different tomato varietie s. *Rivista Italiana delle Sostanze Grasse*, Milano, **1979**, *56*(l), 8-11
6. Brodowski, D.; Geisman, J.R. Protein conten and amino acid composition of protein of seed from tomatoes at various stages of ripeness. *Journal of Food Science*, Chicago, **1980**, *45*(2), 228-229, doi: 10.1111/j.1365-2621.1980.tb02582.x
7. Lazos, E.S; Kalathenos, P. Composition of tomato processing wastes. *International Journal of Food Science and Technology*, Chicago, **1988**, *23*(6), 649-652, doi: 10.1111/j.1365-2621.1988.tb01052.x
8. Cantarelli; P.R.; Palma, E.R.; Caruso, J.G.B. Composition and amino acid profiles of tomato seeds from canning wastes. *Acta Alimentaria*, Budapeste, **1989**, *18*(l), 13-18
9. Cantarelli; P.R.; M.A.B. Regitano-d'Arce; E.R. Palma, Physicochemical characteristics and fatty acid composition of tomato seed oils from processing wastes, *Sci. agric*. (Piracicaba, Braz.), **1993**, *50*(1), 117-120
10. Rabak F., The utilization of waste tomato seeds and skins. *US Dept Agric Bull*, **1917**, 632:15
11. Nikolaos B. K. and Theophanis K.. Calculation of Iodine Value from Measurements of Fatty Acid Methyl Esters of Some Oils: Comparison with the Relevant American Oil Chemists Society Method. *JAOCS*. **2000**, *77*(12):1235-1238
12. Nesma E., Hany A. A., Mohammed H. A. E. and Mohammed M. Y.. Red Palm Olein: Characterization and Utilization in Formulating Novel Functional Biscuits. *J Am Oil Chem Soc*. **2010**, *87*(3), 295–304, doi: 10.1007/s11746-009-1497-x
13. Emmanuel O. A., and Mudiakeoghene O.. The use of antioxidants in vegetable oils – A review. *African Journal of Biotechnology*. **2008**, *7*(25): 4836-4842.

14. Fasina O.O., Hallman C. H. M., and Clementsa C. Predicting Temperature-Dependence Viscosity of Vegetable Oils from Fatty Acid Composition. *JAOCS*. **2006**, *83*(10): 899-903.
15. Daniel I. Hădărugă, Nicoleta G. Hădărugă, Anca Hermenean, Adrian Riviș, Vasile Pâslaru, Georgiana Codina Bionanomaterials: Thermal Stability of the Oleic Acid and cyclodextrin Complexes, Daniel I., *Revista chimie (București*), **2008**, *59*( 9), 994-998
16. Vigo, M.S.; Dasso, I.; Cattaneo, P. Studies on the seeds remaining after the processing of tomatoes. *Anales de la Academia de Ciências Exactas, Físicas y Naturales*, **1977**, *29*,193-203,
17. Amelotti, G.; Garcia, V.; Federico, L.G. La composizione acidica di alcuni oli vegetali. *Rivista Italiana delle Sostanze Grasse*, Milano, **1967**, *44*, 372-378,
18. El-Tamimi, A.H.; Morad, M.M.; Raof, M.S.; Rady, A.H. Tomato seed oil. I. Fatty acid composition, stability and hidrogenation of the oil. Fette Seifen Anstrichmittel, Berlim, **1979**, 81(7), 281-284