THE INFLUENCE OF ADDING OF FLAXSEED OIL TO SUNFLOWER OIL ON THE CONTENT OF TOCOPHEROLS AND CAROTENOIDS IN BLENDED EDIBLE OILS

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ABSTRACT

Blending vegetable oils of different composition and properties is one of the simplest methods for creating new specific products with the desired properties, which increases their commercial application and improves their nutritional quality.
The effect of blending vegetable oils on tocopherols and carotenoids content was examined. Refined sunflower seed oil (S) and cold pressed flaxseed oil (F) were used in the experiment. These oils are blended in three different content of mass: sample 70S:30F (70% S and 30% F), sample 50S:50F (50% S and 50% F) and sample 30S:70F (30% S and 70% F).
The results showed significant differences in the content of total tocopherols and total carotenoids between the two oils used for the preparation of three blended oils. Refined sunflower seed oil contains higher amounts of tocopherols and fewer amounts of carotenoids compared to cold pressed flaxseed oil in which the content of tocopherols is lower and the content of carotenoids is higher. In the obtained blends of edible vegetable oils, the content of total tocopherols ranged from 387.96 to 447.83 mg/kg while the determined total carotenoids content (as β-caroten) ranged from 3.11 to 5.63 mg/kg.
By blending refined oil of sunflower seed and cold pressed oil of flax-seed, the balance of the parameters studied is contributed. The research in the work showed that the blending of vegetable oils provides the possibility of modulating their composition, and therefore of nutritive quality.
***Keywords**: sunflower oil, flaxseed oil, blending, tocopherols, carotenoids*

INTRODUCTION

In many food production industries, finished products are blended to achieve certain new characteristics of the food, physical or chemical properties or a certain health effect. Blending vegetable fats and oils of various compositions and properties is one of the simplest methods for creating new specific products with the desired texture, oxidative and nutritional properties, which leads to improvements in industrial application. Blending has no known negative effects on health (Hashempour-Baltork et al., 2016). The results of previous oil blending studies show that oil blending is widely used in the food industry to produce blended oils with improved stability and nutritional characteristics at an affordable price (Serjouie et al., 2010). There are many economic and health reasons for the production of various oil blends in the future (Hashempour-Baltork et al., 2016).
Flaxseed oil is characterized by the high content of unsaturated fatty acids, which make up to 90% of the total fatty acid content, of which 70% are polyunsaturated fatty acids (Čolović et al., 2016). The fatty acid composition of flaxseed oil differs from other commercial oils due to the very high content of alpha-linolenic fatty acid (ALA), usually above 50%, resulting in the high nutritive value of this oil (Shukla et al., 2002; Przybylski, 2005). Omega 3 fatty acids are essential for growth and development and encourage proper functioning of the cardiovascular system, contribute to the reduction of the risk of heart disease, they act anti-inflammatory and contribute to lowering blood cholesterol and triacylglycerol levels (Verghese et al., 2010). Flaxseed oil contains low content of saturated fatty acids and even less content of tocopherols and sterols (phytosterols) compared to sunflower and many other vegetable oils (Shukla et al., 2002; Przybylski, 2005).

The aim of this study was to examine the effect of adding cold pressed flaxseed oil in refined sunflower seed oil on nutritional quality, i.e., to the content of tocopherols and carotenoids in the blends. Based on obtained results, MLR models were designed. MLR models serve to predict the content of tocopherols and carotenoids in tested blends with different ratio of used oils.

MATERIAL AND METHODS

For the purposes of this study, samples of blended vegetable oils and one additional sample were used, according to the list shown in Table 1. Blended vegetable oils were obtained by blending refined sunflower seed oil (S) and cold pressed flaxseed oil (F), in appropriate proportions. The required mass of the oil is carefully measured into a 1000-ml glass beaker, using a glass stick. The oil which was present in the mixture in lower proportion was added into the beaker first, following the oil present in the mixture in higher proportion. The oils were blended with a magnetic stirrer and immediately transferred to PET bottles of 500 ml volume which were completely filled with oil (without empty space), sealed with an original seal, and the oil was stored at 4°C before testing.
The obtained results are compared with the results obtained by the analysis of the sample OMEGOL which represents a commercially available blend of three refined vegetable oils: rapeseed, sunflower and corn oils, in decreasing order. The ratio of these oils in the blend is not known.
The total carotenoids content (expressed as β-carotene), in mg per kg of oil, was determined by the standard spectrophotometric method (British Standard, 1977). 0.2-1 g of oil was measured to a dosing volume of 10 ml (the expected determined value should be in the range of 0.1-0.8) and supplemented with cyclohexane to the mark. The absorbance of the solution was measured using UV/VIS spectrophotometer T80+ ("PG Instruments", United Kingdom) in glass cell at wavelength 445 nm compared to pure cyclohexane.
The tocopherols analysis of the samples was done in according to the SRPS EN ISO 9936, 2017 standard on HPLC ("Sykam", Germany) equipped with FLD detector. The column was a Nucleosil 100-5 NH2 (5 µm). The peak integration and the quantitative calculations were performed with clarity chromatography software (Data Apex) and calibration curve was obtained by injecting standard solutions of tocopherols at different concentrations. The HPLC analyses were performed using a mobile phase composed of n-hexane and ethyl acetate (70:30,v/v). The fluorescence detector was set as follows: λex= 280 nm, λem=340 nm.

Table 1. Labels and identification of samples

Sample	S (%)	F (%)
100S:0F	100	0
70S:30F	70	30
50S:50F	50	50
30S:70F	30	70
0S:100F	0	100
OMEGOL	-	-

S - refined sunflower seed oil
F - cold pressed flaxseed oil
OMEGOL - blended refined vegetable oil, the only blended oil available on the market (Serbia)

All determinations were made in three replications, and the results were shown as mean value ± standard deviation. Tabelar and graphical representations, as well as statistical processing of obtained results, were done using the Microsoft Excel 2013 software (Microsoft, Washington, USA) and Statistica 13.0 (StatSoft, Tulsa, USA).

RESULTS AND DISCUSSION

The influence of the different fraction of refined sunflower seed oil (S) and cold pressed flaxseed oil (F) in blended vegetable oils on nutritional quality - the content of tocopherols and carotenoids of blended vegetable oils, was examined on the basis of the obtained results of the study: content of total tocopherols and total carotenoids content.
The results of the determined content of total tocopherols and carotenoids of blended vegetable oils are shown in Table 2.

Table 2. The total tocopherols and total carotenoids content in blended vegetable oils

Sample	Total tocopherols content (mg/kg)	Total carotenoids content (mg/kg)
100S:0F	543.58±3.52	0.42±0.01
70S:30F	447.83±5.13	3.11±0.00
50S:50F	383.15±14.39	4.74±0.01
30S:70F	387.96±4.53	5.63±0.01
0S:100F	371.35±9.43	6.90±0.01
OMEGOL	506.23±9.51	0.25±0.01

Tocopherols present in vegetable oils are very attractive because of their physiological importance in the process of health promotion. From the results obtained, it can be seen that the content of tocopherols is in the range of 371.35±9.43 - 543.58±3.52 mg/kg. The largest content of these compounds was found in the sample 100S:0F, while the smallest content of these compounds was found in a sample that does not contain sunflower oil. OMEGOL - blended refined vegetable oil, as the only blended oil available on the Serbian market contains 506.23±9.51 mg/kg of tocopherols. Carotenoids are highly desirable ingredients of vegetable oils. In addition to affecting the color of the oil, they are also desirable from the aspect of antioxidant activity. The carotenoid content is in the range of 0.42±0.01 - 6.90±0.01 mg/kg. The highest content of carotenoids was found in the sample 0S:100F, while the smallest content of these compounds was found in a sample that does not contain flaxseed oil. OMEGOL contains 0.25±0.01 mg/kg of carotenoids.
The blending of refined sunflower seed oil and cold pressed flaxseed oil contributes to the balance of the parameters studied. It means that the blending of vegetable oils provides the possibility of variation of their composition, and therefore of nutritive quality. These variations can be described in both cases by second degree polynomials with high determination coefficients (polynomial model 1 and polynomial model 2):

Polynomial model 1: $y_{tocopherols} = 16.271\,x^2 - 138.06\,x + 661{,}97; \quad R^2=0.975$
Polynomial model 2: $y_{carotenoids} = -0.2557\,x^2 + 3.0823\,x - 2.274; \quad R^2=0.9914$

Multiple linear regression analysis, MLR is a mathematical method that allows for taking into account several factors - independently variable (x_1, x_2 ...), which have an effect on one dependent variable (y). The content of tocopherols ($\omega_{tocopherols}$) and the content of carotenoids ($\omega_{carotenoids}$) were taken as the dependent variable (y), while the independent variables were the fraction S in blended vegetable oils (S,%/100), (ω_S) and the oil fraction F in blended vegetable oil (F/S), ($\omega_{F/S}$). Based on experimental data (Table 2), mathematical models were formed. The following MLR models have been designed to predict the selected variables:

MLR model 1: $\omega_{tocopherols} = 442.3732\,\omega_S + 64.4790\,\omega_{F/S} + 103.5043$
MLR model 2: $\omega_{carotenoids} = -10.3526\,\omega_S - 0.8661\,\omega_{F/S} + 10.7591$

The formed MLR equations (MLR model 1 and MLR model 2) well describe the dependence of the content of tocopherols and carotenoids and the content of S (ω_S) and F/S ($\omega_{F/S}$) in blended vegetable oils. The negative values of the coefficients with independent variables indicate their negative impact on the indicators tested.

Verification of the validity of the set MLR models was done by cross-validation with the basic statistical parameters shown in Table 3.

Table 3. The total tocopherols and carotenoids content in blended vegetable oils

Parameter	Model	
	MLR 1	**MLR 2**
R	0.9972	0.9999
R^2	0.9945	0.9998
R^2_{adj}	0.9834	0.9996
F	89.8149	4337.0652
p	0.0744	0.0107
SD	9.6239	0.0426

Based on the correlation and determination coefficients, it can be concluded that the set MLR models describe very strong correlations between the variables and that predicting the contents of total tocopherol and total carotenoids content can be achieved with an acceptable predictive error. According to R and R^2 values obtained MLR models indicate a very high correlation between the variables. Also, it is an indicator that estimation error of the total tocopherol and total carotenoids prediction is in allowed range. Very high values of R^2_{adj} (0.9834 and 0.9996) indicate the high predictive ability of obtained MLR models. High values of F-parameter confirm good fit of data.

For the established MLR mathematical dependencies between the indicators of the content of tocopherol and carotenoids and fraction S and fraction F/S, the correlation between the experimentally obtained values ($y_{exp.}$) and the values obtained using the set MLR mathematical models (x_{MLR}) was investigated:

Tocopherols: $y_{exp.} = 0.9945\ x_{MLR} + 2.4393$; $R^2=0.9945$

Carotenoids: $y_{exp.} = 0.9999\ x_{MLR} + 0.0004$: $R^2=0.9999$

Based on the relations shown, it is obvious that there is good accordance between the experimental and predicted values, the dependence sections for the determined models tend to zero, while the slopes are close to one.

The cluster method is based on the content of tocopherols and carotenoids in the test samples. This method found that the samples 70S:30F, 50S:50, 30S:70F and 0S:100F constitute one subgroup, or from the aspect of the content of tocopherols and carotenoids, these samples are similar, in contrast to the sample 100S:0F which is separated from this group. The largest distance was recorded between the initial sample 100S:0F and 0S:100F, 172 (expressed as Euclid's distance), a somewhat lower distance was recorded between the samples 100S:0F and 50S:50F (160) and the samples 100S:0F and 30S:70F (156), then the distance between the sample 70S:30F with the samples 50S:50F (65), 30S:70F (60) and 0S:100F (77) is quite equal, while its distance with the sample 100S:0F is somewhat higher and is 96, a small distance was found between the sample 0S:100F with samples 30S:70F (17) and 50S:50F (12), while the smallest distance was between the samples 50S:50F and 30S:70F (5), as is clearly seen in Fig. 1.

The similarity obtained by the clustering method between blended vegetable oils (70S:30F, 50S:50F and 30S:70F) is explained by the possibility of variation of their composition by blending the initial samples, i.e. by blending refined oil of sunflower seeds and cold pressed flaxseed oil, it contributes to the balance of the parameters studied compared to the initial sample 100S:0F.

By comparison of the tested samples with different contents of tocopherols and carotenoids, blended vegetable oils 70S:30F, 50S:50F, 30S:70F have a more balanced ratio of tocopherol and carotenoid content (447.83±5.13 : 3.11±0.00 mg/kg, 383.15±14.39 : 4.74 ± 0.01 mg/kg, 387.96±4.53 : 5.63±0.01 mg/kg, respectively) compared to the initial sample 100S:0F (543.58±3.52 : 0.42 ± 0.01 mg/kg).

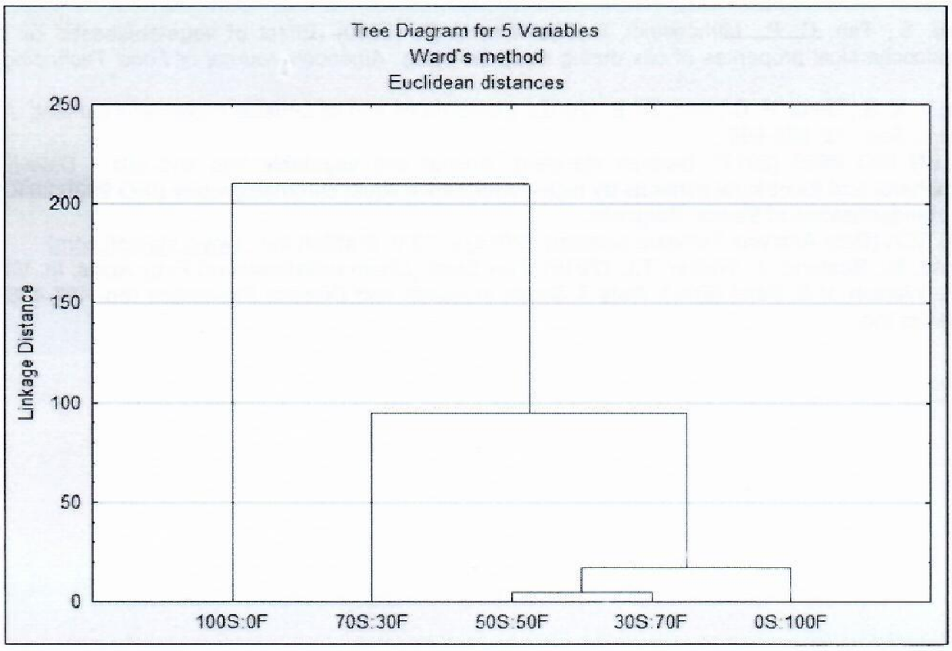

Figure 1. Dendrogram of the hierarchical cluster analysis of the tested samples based on the content of tocopherols and carotenoids

CONCLUSIONS

Blended vegetable oils are characterized by the presence of certain contents of tocopherols and carotenoids. From the aspect of the total tocopherols content, the most preferable blend is with the ratio 70S:30F, while the most preferable blend from the aspect of the total carotenoids content is the blend with the ratio of 30S:70F. Determined MLR models have high statistical quality and can serve to predict a favorable share of flaxseed oil in blended oil, from the aspect of total tocopherols and total carotenoids content.

On the basis of the obtained results it can be concluded that the blending contributes to the balance of the studied parameters. The search for new vegetable oil for blending and the proper blending ratio should be a research indicator depending on the target and the application in which the blended vegetable oil will be used.

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REFERENCES

British standard (1977). British standard method of analysis of fats and fatty oils - Determination of carotene in vegetable oils, British Standard Ilustration, London.

Čolović, D., Berenji, J., Levart, A., Lević, J., Salobir, J., Pezo, L., Čolović R. (2016). Nutritional characteristics of seeds of eighteen linseed (Linum humille Mill.) cultivars from Serbia, *Zemdirbyste-Agriculture, 103(2),* 175-182.

Hashempour-Baltork F., Torbati, M.A., Azadmard-Damirchi, S., Savage, G.P. (2016). Vegetable oil blending: A review of physicochemical, nutritional and health effects. *Trends in Food Science & Technology, 57,* 52-58.

Microsoft Excel 2013, Microsoft. Microsoft Excel. Computer Software. Redmond, Washington, USA, 2013. (www.microsoft.com).

Przybylski R. (2005). Flax Oil and High Linolenic Oils. In: F. Shahidi (Ed), *Bailey's Industrial Oil and Fat Products, Edible Oil and Fat Products: Edible Oils* (Vol. 2, pp. 281-302). J. Wiley & Sons, New Jersey, USA.

Serjouie, S., Tan, C. P., Mirhosseini, H.,Che Man Y. B. (2010). Effect of vegetablebased oil blends on physicochemical properties of oils during deep fat-frying. *American Journal of Food Technology*, *5*, 310-323.

Shukla, V. K. S., Dutta, P. C., Artz, W. E. (2002). Camelina oil and its unusual cholesterol content. *J. Amer. Oil Chem. Soc.*, *79*, 965-969.

SRPS EN ISO 9936 (2017). Serbian standard. Animal and vegetable fats and oils - Determination of tocopherol and tocotrienol contents by high-performance liquid chromatography (ISO 9936:2016). Institute for standardization of Serbia, Belgrade.

STATISTICA (Data Analysis Software System), (2015) v. 13.0, StatSoft Inc., (www.statsoft.com).

Verghese, M., Boateng, J., Walker, T.L. (2010). Flax Seed (*Linum usitatissimum*) Fatty Acids. In: V.R. Preedy, R.R. Watson, V.B. Patel (Eds.), *Nuts & Seeds in Health and Disease Prevention* (pp. 487-498). London: Elsevier Inc.

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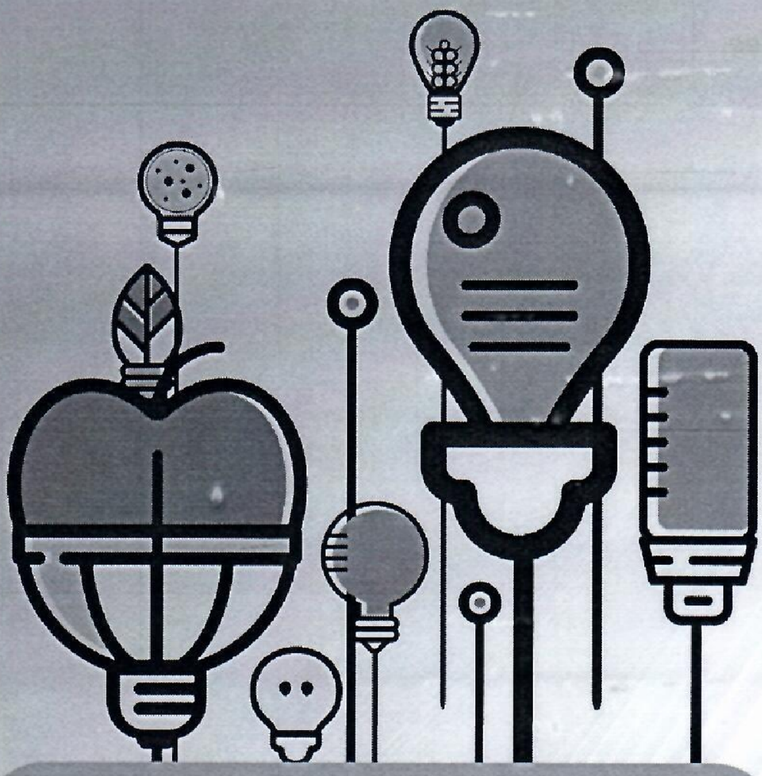

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