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# **OBTAINING STABLE FOOD EMULSIONS BETWEEN MILK AND CORN OIL**

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# Abstract

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The possibilities of preparing stable food emulsions by replacing the milk fat with corn oil in various amounts were studied. For emulsification of corn oil in the milk were used emulsifiers – glycerolmonostearate, soy lecithin and sunflower lecithin, in concentrations 1g.dm<sup>-3</sup>. It has been found that the emulsions with lycerolmonostearate by concentration of vegetable oil - 80 cm<sup>3</sup>.dm<sup>-3</sup> and in combination with milkfat - 40 cm<sup>3</sup>.dm<sup>-3</sup> and corn oil - 40 cm<sup>3</sup>.dm<sup>-3</sup> are most stable.

Key words: food emulsion, corn oil

*Abbreviations:* glycerolmonostearat (GMS); soy lecithin (SoyL); sunflower lecithin (SL); control sample (C); European union (EU); lowest standard deviation (LSD); standard deviation – (SD); oil-in-water emulsion - O/W

# Introduction

Milk fat is naturally a stable emulsion, which destabilizes under certain conditions. The emulsion has to be stabilized in cases where the milk fat is replaced by vegetable oil or when the fat content is increased by addition of cream. Therefore, very often in the dairy industry, homogenization by pressure is applied, to stabilize the milk fat, with or without the addition of vegetable oils (Vignola, 2002). The possibilities of using different emulsifiers are insufficiently explored (Lobato-Calleros et al., 2001, 2007; Vial et al., 2006; 2006a).

Milk fat, as a typical animal fat, is one of the most important constituents of the milk, and in the same time it is the most variable in terms of quantity and composition (Karleskind, 1992). It is known that the milk fat is characterized by a number of negatives with regard of human health. They are related to the fact that certain fatty acids, which are contained therein, as well as the cholesterol content, can lead to cardiovascular diseases, as a result of an increase in plasma cholesterol and low density lipoproteins. It has been shown that certain fatty acids - lauric (C<sub>12:0</sub>), myristic (C<sub>14:0</sub>) and palmitic acid (C<sub>16:0</sub>), can be atherogenic factors (Jensen, 1992). More recent studies show that palmitic acid cannot be atherogenic factor when consumed with adequate amounts of linoleic acid (C<sub>18:3</sub>) (Clandinin, 2000; Mensink et al., 2003). Therefore, more recent studies focus on finding new opportunities for full or partial replacement of milk fat with oils rich in omega fatty acids, such as sunflower, corn, soy, etc. (Bockisch, 1998). It has been found (Ivanova et al., 2012), that corn oil is suitable for use in O/W emulsions because of its high content of polyunsaturated fatty acids, tocopherols and appropriate sensory characteristics (Warner and Nelsen, 1996).

The purpose of this work is to investigate the stability of milk fat emulsions, to establish the ratio of corn oil and milk fat and suitable emulsifier.

# **Material and Methods**

## Material

For the oil phase butterfat and corn oil are used. The most widely used emulsifiers in the dairy industry are glycerolmonostearat (GMS), soy lecithin (SoyL) and sunflower lecithin (SL), in an amount of 1g.dm<sup>-1</sup> according to the literature data (Hasenhuettl and Hartel, 2008). For each variety is prepared a corresponding control (C), without emulsifier. The dispersion medium is skim milk obtained from raw cow's milk complying with the EU Regulation 853/2004.

#### Equipment

For the mixing of the two phases a laboratory homogenizer Polytron®PT45-80 Company Kinematika (Switzerland) with technical features 220 V; 50 Hz; 1600 W; max 15000 rpm is used. To determine the stability of the emulsions, according to the respective methods, centrifuge and microscope system (microscope CarlZeiss, Germany, equipped with a USBcamera connected to a personal computer) are used.

## Preparation of emulsion

Preparation of model emulsions is carried out by mixing the oil phase in the dispersion medium under constant stirring (speed - 9000 rpm), for 5 min.

The used concentrations are 80 cm<sup>3</sup>.dm<sup>-3</sup> corn oil (Variant 1), 30 cm<sup>3</sup>.dm<sup>-3</sup> milk fat and 50 cm<sup>3</sup>.dm<sup>-3</sup> corn oil (Variant 2), 40 cm<sup>3</sup>.dm<sup>-3</sup> corn oil and 40 cm<sup>3</sup>.dm<sup>-3</sup> milk fat (Variant 3) and 80 cm<sup>3</sup>.dm<sup>-3</sup> milk fat (Variant 4). The mixtures are heated to 55-60°C in order to dissolve completely the emulsifier (Dłużewska et al., 2004; Kuncheva et al., 2007; McClements, 2005).

#### Determination of emulsion stability

The stability of the emulsions is evaluated by the following methods:

• Centrifugal Test - The stability of the emulsion is carried out by centrifugation of 5 cm<sup>3</sup> of the emulsion for 10 min at 2500 rpm. The degree of creaming is evaluated by measurement of the layers after centrifugation «buttery» (upper), "emulsified" in the middle, and "water - skimmed milk" (bottom) and in some cases, "precipitate". The emulsion stability (S) is determined by the formula:

$$S = [(V_0 - V)/V_0] x 100,$$
(1)

where: S – emulsion stability, %,  $V_0$  – volume of emulsion, cm<sup>3</sup>, V – volume of separated layer, cm<sup>3</sup>.

The obtained data shows that the higher the value of S (stability), the more stable is the emulsion (Dłużewska et al., 2004; McClements, 2007; Reineccius, 1994).

• Microscopic Test - After taking a picture of the microscopic slide preparation, the number and size of fat globules are calculated. For processing the received data, they are divided into 4 classes with predetermined dimensions: first class  $-0 - 4 \mu m$ ; second class  $-4 - 8 \mu m$ ; third class  $-8 - 12 \mu m$  and fourth class above  $12 \mu m$ . The total number of analyzed dispersed particles is in the range of  $10^2 \div 10^3$  (special attention is paid to avoid measuring the same field twice). The stability of the emulsion is determined by the parameters characterizing the dispersion (Vasileva, 2011; Dłużewska et al., 2004; McClements, 2007).

- Determination of the mean diameter of the fat globules for a certain module, in  $\mu$ m, is obtained according to the formula:

$$d_{mean} = (X_{max} + X_{min})/2,$$
 (2)

where:  $d_{mean}$  - average diameter of fat globules,  $\mu m$ ;  $X_{min}$  – the lower limit of the previously selected module;  $X_{max}$  – upper limit of the previously selected module.

The mean arithmetic diameter "longueur-nombre" dln of fat globules in emulsion is given by:

$$dln = (\sum Mn^* d_{mean}) / \sum n,$$
(3)

where: dln - mean arithmetic diameter,  $\mu$ m; Mn - number of fat globules in a group (module); d<sub>mean</sub> - average diameter of fat globules in a module,  $\mu$ m;  $\sum n$  - amount of total fat globules.

Homogeneity F (frequency of the size of fat globules) is calculated using the formula:

$$F = (n * 100) / \sum n,$$
 (4)

where: F – frequency, %; n – number of fat globules in a group;  $\sum n$  – amount of total fat globules;

• Temperature Test - 10 cm<sup>3</sup> of the emulsion is placed in tubes, which are stored at three different temperatures – 4°C (refrigerator temperature) – 23°C (room temperature) and 45°C (water bath or thermostat) for 24 h (Tan and WuHolmes, 1988). The visible appearance of the oil ring shows that the emulsion is unstable, and the degree of creaming is evaluated by: "+" if there is an oil ring, "+/-" suspected creaming (not well-formed oil ring) and "-" absence of an oil ring. In order to visualize better the oil ring a fat soluble dye Soudan III is used in the experiments.

#### Statistical analysis

Computer processing of the results is performed using the program Microsoft Excel 2010 (ANOVA). Multiple comparisons are made by LSD method. The results are presented as mean value  $\pm$  SD (n=4).

## **Results and Discussion**

One of the most common optical methods for determination of the emulsion stability is the centrifugal test.

Table 1 represents the results of the emulsion stability (S) after centrifugal test.

It is evident that the emulsions remain stable with GMS and SL in option 1 and 2.

By increasing the amount of milk fat to 40 cm<sup>3</sup>.dm<sup>-3</sup> and the reduction of the oil content to 40 cm<sup>3</sup>.dm<sup>-3</sup> emulsion with emulsifier GMS is more stable in comparison with an emulsifier SoyL. This emulsion is unstable and in the case of the emulsifier SL. Similar behavior is reported for milk fat concentration of 80 cm<sup>3</sup>.dm<sup>-3</sup>.

The statistical analysis found significant influence of the factor concentration of oil phase and type of used emulsifier (p<0,05). The analysis shows that both factors have a statistically significant impact on the stability of the studied emulsions.

Criteria for their stability, in the application of the microscopic test, are dispersing characteristics of the system - dln (the average diameter) and frequency (size distribution). It is evident that the emulsions remain stable with GMS and SL in option 1 and 2.

Table 2 represents the data obtained for the indicator arithmetic mean diameter "longueur-nombre" dln of fat globules. It shows the emulsion stability - the smaller, the more stable.

The obtained data shows that in Variant 1, the smallest size of the fat globules is in the emulsions prepared with GMS. This gives us reason to believe that the probability of these emulsions to remain stable over time is the greatest. In Variant 2 it is found that the stability of emulsions with GMS and SoyL, is the same. The emulsions are characterized by equal arithmetic mean size of the fat globules, which identifies them as uniformly stable and more stable than the emulsion with sunflower lecithin.

In the other variants, stabilized by the emulsifier GMS, the fat globules are characterized by a reduction in the arithmetic average size and the highest stability of the emulsions.

In all four variants it is found that their respective controls remain unstable. From the statistical analysis it is apparent

Table 1Centrifugal test for determination of emulsion stability

	S, %	S, %	S, %	S, %	
Emulsifier	Variant 1	Variant 2	Variant 3	Variant 4	
GMS	92.32 ± 0.10 <sup>aw</sup>	90.91 ± 0.17 <sup>bw</sup>	90.33 ± 0.27 <sup>cw</sup>	90.69 ± 0.33 <sup>cw</sup>	
SoyL	$85.00 \pm 0.21^{ax}$	$\begin{array}{c} 82.50 \pm \\ 0.35^{bx} \end{array}$	$86.50 \pm 0.14^{cx}$	$83.21 \pm 0.22^{dx}$	
SL	$\begin{array}{c} 92.46 \pm \\ 0.22^{aw} \end{array}$	${}^{88.24\pm}_{0.25^{by}}$	$88.06 \pm 0.14^{cy}$	$87.99 \pm 0.14^{cy}$	
С	${}^{86.37\pm}_{0.40^{ay}}$	${}^{87.83\pm}_{0.17^{\rm bz}}$	${}^{84.40\pm}_{0.15^{cz}}$	$83.16 \pm 0.24^{dx}$	

a,b,c,d Means with different letters within a row are significantly different (p<0.05)

w,x,y,z Means with different letters within a column are significantly different (p<0.05)

that the influence of the factors used in the type of emulsifier, and the concentration of the oil phase, is significant. It is established a mutual influence of these factors (p<0.05) with respect to the stability of milk fat emulsions. These results are reported after the centrifugal test.

In Figures 1, 2, 3 and 4 is shown experimental data from micro photographic analysis, for the distribution of fat globules according to their size.

The index homogeneity of the emulsion demonstrates its stability. Dense distribution of fat globules in the emulsion shows a better homogeneity and greater stability of the emulsion.

Fairly uniform distribution of the fat globules is established in the range of  $0 - 4 \mu m$  in the emulsion with an emulsifier GMS. It stabilizes better the emulsion in Variant 1, compared to the soy and sunflower lecithin. The control sample is instable due to the large number of fat globules from 4 to 8  $\mu m$ .

The data presented in Figure 2 shows the distribution of the fat globules, depending on their size and frequency (distribution by percentage occupied by their size). It was found that in comparison to the results presented in Figure 1, the proportion of fat globules up to 4  $\mu$ m, is lower. This tendency is reported in all emulsions except contained sunflower lecithin as an emulsifier. There is an increase in the amount of fat globules with sizes between 4 and 6  $\mu$ m. Based on these data it can be concluded that the stability of the emulsions is lower in option 2. There is an increased stability in the emulsion with sunflower lecithin. This tendency is confirmed by the indicator arithmetic mean size of fat globules, independently of the type of emulsifier.

The data presented in Figure 3 shows that the highest stability of the emulsion is in the case of GMS - Variant 3. There

# Table 2Mean arithmetic diameter of fat globules

Emulsion	Mean arithmetic diameter dln, $\mu m$					
	GMS	SoyL	SL	С		
Variant 1	$2.24 \pm 0.17^{aw}$	$\begin{array}{c} 2.34 \pm \\ 0.23^{aw} \end{array}$	$2.42 \pm 0.18^{\rm aw}$	$2.85 \pm 0.17^{bw}$		
Variant 2	$2.77 \pm 0.11^{ax}$	$2.77 \pm 0.11^{ax}$	$2.82 \pm 0.23^{ax}$	$\begin{array}{c} 2.86 \pm \\ 0.16^{\rm aw} \end{array}$		
Variant 3	$2.22 \pm 0.11^{ay}$	${}^{2.36\pm}_{0.11^{bw}}$	$\begin{array}{c} 2.39 \pm \\ 0.10^{\rm bw} \end{array}$	${}^{2.43\pm}_{0.05^{bx}}$		
Variant 4	$2.04 \pm 0.02^{ax}$	$2.70 \pm 0.11^{bx}$	$2.76 \pm 0.04^{bx}$	$2.73 \pm 0.18^{\rm bw}$		

a,b,c,d Means with different letters within a row are significantly different (p<0.05)

w,x,y,z Means with different letters within a column are significantly different (p < 0.05)

is a tendency to increase the stability of milk fat mixtures when used emulsifier lecithin, compared to Variant 2. Values obtained for the arithmetic mean diameter of the fat globules, at the indicated concentrations, confirm this statement. It was found that the proportion of fat globules of the second group - between 4 and 6  $\mu$ m, is smaller.

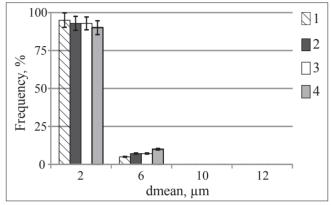


Fig. 1. Distribution and size of fat globules in Variant 1

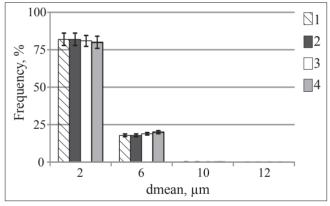


Fig. 2. Distribution and size of fat globules in Variant 2

The results presented in Figure 4 show that GMS is the most suitable emulsifier for Variant 4. It was found that in the emulsion with emulsifier lecithin and in control sample, increasing the amount of milk fat leads to increase in number of fat globules in the range of 4 - 6  $\mu$ m. There are single fat globules in the range 8 - 12  $\mu$ m.

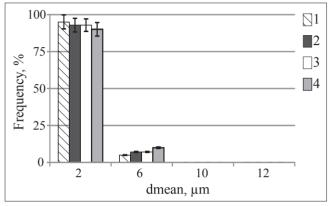


Fig. 3. Distribution and size of fat globules in Variant 3

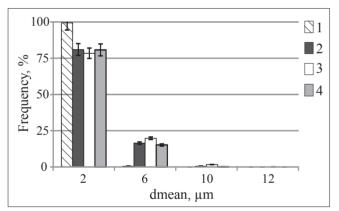


Fig. 4. Distribution and size of fat globules in Variant 4

## Table 3

Emulsion stability obtained from temperature test according to the applied concentrations of vegetable oil and milk fat and the type of emulsifier

Variant 1			+ °C	Variant 2				
t, °C 1	2	3	4	ι, τ	1	2	3	4
-	-	-	+	4	-	-	-	+/-
+	+	+	+	22	-	+/-	-	+/-
+	+	+	+	45	+/-	+	+	+
t, °C Variant 3			4 00	Variant 4				
1	2	3	4	t, C	1	2	3	4
-	-	+/-	+	4	-	+/-	+/-	+/-
+/-	+/-	+	+	22	-	+	+	+
+/-	+	+	+	45	+/-	+	+	+
	+/-	1         2           -         -           +         +           +         +           Varia         1           1         2	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$					

 $\overline{(1 - GMS; 2 - SoyL; 3 - SL; 4 - C)}$ 

The results from the storage at different temperatures are shown in Table 3.

The results in Table 3 show that the stability of all the emulsions at low temperatures is higher. There is a deviation from this tendency in Variant 3 - with sunflower lecithin as emulsifier and Variant 4 – when the emulsifiers soya and sunflower lecithin are used. All the emulsions with GMS remain stable at 4°C. This characterizes the emulsifier GMS as very good emulsifier for fats for these model emulsions at low temperature.

There is an established tendency to destabilize the emulsion systems with increasing temperature. This tendency begins at 22°C, with a temperature rise to 45°C, the emulsions remain permanently unstable.

Controls in low, medium and high temperatures, are relatively fragile.

## Conclusions

The stability of the fat-oil emulsions with corn oil is determinate. It has been found that in order to obtain a stable dairy based fatty emulsion, the optimal ratio between the vegetable oil and milk fat is variant 3 - 40 cm<sup>3</sup>.dm<sup>-3</sup> corn oil and 40 cm<sup>3</sup>. dm<sup>-3</sup> milk fat. Milk fat emulsions demonstrate the high stability in the application of an emulsifier glycerolmonostarat. The resulting stable food emulsion is applicable to the development of high fat fresh cheese.

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