Investigation and characterization of O/W emulsions on stability with different methods

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Abstract
The properties and efficiency of O/W emulsions were investigated. Influence by oil phase in the emulsification processes was determined. After centrifugation measurements in emulsions with high percent oil four phases were observed. pH values in all emulsions was measured and found from 5.8 to 6.3 in connection with application of foods. During the storage period of fourteen days the spectroscopic measurements were perform in continuous phase and middle radius of particle sizes were calculated.

1. Introduction
In foods the protein soluble in water and oil phase are the main phases that form the emulsions. Different concentration and addition of protein determined stability against flocculation and coalescence. Sunflower oil and soybean protein isolate (SI) is appropriate compound to prepared emulsions for food industries. Some foods are: milk, cream, butter, margarine, soups, mayonnaise, desserts, ice cream and etc. [1-3]. The influence of protein concentration as addition relative to homogenization and extent of droplet flocculation in heated oil-in-water emulsions were examined. Influence of pH was observed from authors [4].

Emulsions with type water-in-oil-in-water were prepared. Skim milk was used as protein and investigation was provided by 17 20 kHz ultrasound. The fat displacement and shelf stability of the emulsions were found [5]. The effect of bovine serum albumin (BSA) and another proteinson physical stability of internal or external aqueous phase of water-oil-water (W/O/W) emulsions were evaluated [6]. Investigation of the emulsification process of the mixtures from sunflower seeds, obtained by separation steps and links their properties with their molecular composition. The investigated molecular mixtures were the cold-pressed sunflower cake, a protein-based and a fiber-based mixture [7]. A simple methodology to unabsorbed emulsifiers in the aqueous phase of O/W emulsions was developed. Unabsorbed emulsifiers affect the physical and chemical behavior of oil-in-water (O/W) emulsions [8]. The influence of complexation with curcumin at pH 3.0 and 7.0 on the emulsifying
properties of soy protein isolate (SPI) was providing. At both test pH values, most of proteins were present in the nanoparticle form, with larger sizes, higher surface hydrophobicity and better emulsifying properties of these nanoparticles observed at pH 3.0 [9]. Study of effect of pH on the stability of oil-in-water emulsions stabilized by commercial splittable surfactants was providing. The emulsion stability was explored by measuring the volume of oil phase separated and the size of the dispersed droplets and found that the addition of inorganic acids [10]. A method that combines centrifugation and conductivity measurements was used for emulsion stability. The method was compared with a creaming measurement method based on fat content [11].

The aims of this work are to investigate experimentally emulsion stability with different oil percent, water and protein using experimental and calculated methods.

2. Materials and methods
Twelve emulsions were prepared after mixing compound. Sunflower oil from 10 to 40 percent and soybean protein isolate (SI) were used. The emulsion preparation was performed with different concentrations of protein which were added in water and solubilise in it. The oil phase was added slowly. A homogeniser was used for mixing for 1 min and 30 s duration [12].

pH in samples was determined by pH meter Hanna HI 98127 with Replaceable Electrode. The pH values of the 12 emulsions were determined immediately after their preparation.

A centrifugation measurement (mechanical stress) of emulsions was providing. After preparation of emulsions they heat at 45°C for 20 min. After that they leave in centrifuge at 6000 rpm for the duration of 5 min.

Determination of emulsion turbidity was providing and used to calculate the size of emulsions particles. The investigations were performed using a Camspec M107 UV/VIS spectrophotometer as described in [4, 5]. The change in the emulsion turbidity was measured at wavelength 450 nm [14].

Determination of turbidity:

\[ \tau = \frac{A}{l} \]  

where: \( A \) – absorption, \( l \) – cuvet length, 1cm.

Determination of particle size:

\[ z = \frac{2\pi r}{\lambda} \]  

where: \( z \) – constant, connected with turbidity; \( \pi \) – constant; \( \lambda \) – wavelength, nm; \( r \) – radius of particles, µm.

3. Results and discussion
A numbers of emulsions were prepared and investigated with different methods. They were prepared with oil from 10% to 40%, water from 57% to 89% and soybean protein isolate (SI) 1, 2 and 3%. Chemical composition of different emulsions was presented in Figure 1. On the figure presented connection between different percents oil and water and after that with addition of protein percents.
Figure 1. Primary composition of 12 O/W emulsions prepared with different percents oil, water and 1, 2 and 3% soybean protein isolate.

Influence by oil phase in the emulsification processes were determined. pH values in all emulsions was measured and found between 5.8 and 6.3 in connection with application of foods. Figure 2 presented variation of pH values as dependence of oil percents. In the figure observed, that the emulsions with less pH (approximately 5.9) connected with larger oil percents and with high stability of emulsions.

Figure 2. Variation of pH values as dependence of oil percents.

To determine the emulsion stability centrifugation test was performed. These observations were connected with mechanical properties of emulsions known as mechanical stress. At the end of the cycle each emulsion was checked. It is found that after centrifugation measurements in emulsions with high percent oil four phases were observed. Descriptions of different phases and quantity of emulsions are presence in Table 1.
Table 1. Different phases observed in investigation emulsions after centrifugation test.

| em № | O/W | W/O | Protein (SI) | Oil | m, g |
|------|-----|-----|-------------|-----|------|
| 1    | 80  | 12  | 8           | -   | 15   |
| 2    | 74  | 18  | 8           | -   | 15   |
| 3    | 40  | 47  | 9           | 4   | 15   |
| 4    | 60  | 23  | 8           | 9   | 15   |
| 5    | 79  | 10  | 11          | -   | 15   |
| 6    | 74  | 14  | 12          | -   | 15   |
| 7    | 43  | 43  | 9           | 5   | 15   |
| 8    | 25  | 52  | 6           | 8   | 15   |
| 9    | 69  | 20  | 13          | -   | 12   |
| 10   | 59  | 29  | 12          | -   | 13   |
| 11   | 44  | 38  | 10          | 8   | 13   |
| 12   | 32  | 49  | 10          | 9   | 11   |

First appeared two main phases O/W and W/O. They presented the generally compound of emulsions and they had a high percent from common quantity. In all emulsions a deposition of protein was observed as described of [13]. In some emulsions prepared with high percent oil phase from 30 and 40% oil a small pure oil phase between 4 to 9% was observed. After centrifugation test on Figure 3 some part of emulsions with their different phases can be seen. On the figure observed high cream phase in emulsion 8 prepared with 30% oil and small protein deposition according the prime composition of preparation.

Figure 3. Emulsions 5 – 8 prepared with 2% protein after provide centrifugation measurements.

During the storage period of fourteen days the spectroscopic measurements were perform in continuous phase and middle radius of particle sizes were calculated. At the presented period in the beginning was observed emulsions with small size. With the end of the period the particles increased their sizes. The graphical view of obtained results is presence in Figure 4.
Figure 4. Graphical presentation of particle size in the emulsions during the storage period.

After providing the experiment, the high turbidity observed at emulsions prepared with 30 and 40% oil. The high turbidity connected with small particle sizes and high oil phase too. Some good results were observed in samples with high oil percent and 1% soybean protein isolate.

4. Conclusion
The twelve emulsions were investigated prepared with different oil and water percent and with different methods. Influences of pH show that the emulsions with pH around 5.9 are more stable. The centrifugation connected with mechanical properties was used and found that some appropriate at foods are emulsions with high oil percent. After storage period the size of the emulsion droplets were determined after spectroscopic measurements. Emulsions prepared with high oil percent and 1% soybean protein isolate exhibit good qualities and stability.

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