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Surface activity of hexane extract of egg yolk. A preliminary study.

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### **INTRODUCTION AND OBJECTIVE**

Liposomes and nanoliposomes made of pure phospholipids are already used in pharmaceutics to increase the bioavailability and the bioefficiency of drugs (Arab Tehrany et al 2012). In food technology this procedure is developed in aim to preserve different bioactive substances (Maherani et al. 2012). The most popular method for manufacturing order to bioactive liposomes in substances preservation in food products is thin film hydration method. The advantage of this method is simplicity, however it makes impossible to control the size and structure of obtained vesicles. Although there are a lot of methods used for the examination of the structure and stability of liposomes, such as high resolution microscopy, transmission electron microscopy (TEM), high resolution microscopy, immunofluorescence confocal laser scanning microscopy (CLSM), particle size and zeta ( $\zeta$ ) potential measurement, none of them refers to the properties of raw material used for manufacturing of phospholipid bilayer. To create that structure it is necessary to use surface active agents of relative low HLB value appropriate to formation water in oil w/o, but not oil in water o/w ones. However, surprisingly few works regarding surface activity of lecithin or its fractions, derivatives or analogues were done (Jendrasiak et al 1996, Li et al 1996a, b, Wu et al. 2000). They concern mainly water-chloroform system and application of the drop volume method (Li et al 1996a, Wu et al. 2000).

To develop understanding the mechanism of the formation of phospholipid bilayer the surface activity of the hydrophobic fraction of egg yolk, obtained by extraction with hexane, was considered in this study.

## MATERIALS AND METHODS

Fresh egg yolk was lyophilised using LMC-1 apparatus (Christ, Germany). The following parameters of the process were applied for this process: freezing -80°C; primary drying -18°C, 0.12 mbar, 20 h; finally drying 22 °C, 0.025 mbar, 4h. Then the dried product was subjected to extraction with boiling hexane using Soxhlet extractor. Obtained product was used to further analyses.

The equilibrium surface tension and interface tension values were determined with the use of the du Noüy ring technique (KSV tensiometer). Dilutions from  $10^{-6}$  to  $10^{-1}$  mol/dm<sup>3</sup> of the extract in hexane solutions were investigated. The ring was rinsed with acetone



and burnt in a flame before each measurement. Experiment was carried out in a temperature controlled cell, at  $21.0\pm0.1$  °C. The reproducibility of the surface/interfacial tension measurements was in approximately 0.05 N/m.

### **RESULTS AND DISCUSSION**

The product of lyophilisation process of egg yolk was yellow/intensely yellow material. Extraction with hexane gave light orange and orange liquid of relatively high density with 0,54% of moisture.

The surface activity of hexane extract of egg yolk was studied by means of equilibrium surface/interfacial tension experiments. The interfacial tension isotherm for lecithin is shown in Fig. 1.

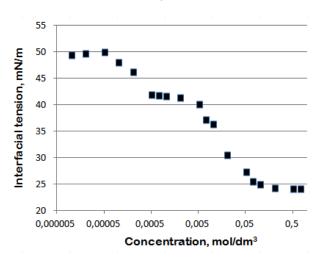


Figure 1 : Interfacial tension isotherm for lecithin in the hexane/water system

The investigated lecithin reveals the high ability of reducing the surface/interfacial tension. The effective lowering of hexane/water tension by approximately 25 mN/m is observed. Taking into account the run of curve shown in Fig.1 one can conclude that two ranges of critical micelle concentration can be observe. Thus, one can assume the lecithin considered is not a molecular entity and some additional components are present in the sample investigated. It pointed that even hexane, hydrophobic solvent of low ability to dissolve separates a mixture of substances was not sufficient for highly exclusive lecithin separation process from egg yolk.

The surface/interface tension data, obtained experimentally, can be fitted by various adsorption equations. From a physicochemical point of view it is

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suitable to use the Szyszkowski and Frumkin equations (Chattoraj at al. 1984). The adsorption coefficients of the Szyszkowski isotherm (calculated by the least square method matching Szyszkowski equation to the experimental data) can be use to estimate the surface excess at the saturated interface  $(\Gamma^{\infty})$  and the Gibbs free energy of adsorption ( $\Delta G_{ads}$ ). These two adsorption parameters allow for quantitative characterization of adsorption in both systems considered.

The value of  $\Gamma$ - i.e. adsorption density of a saturated interface can be used as the measure of the adsorption effectiveness at the fluid/fluid interface. On the other hand, the adsorption effectiveness can be also defined by the lowest surface/interfacial tension  $\gamma_{min}$  which can be obtained in the considered system or by the highest surface/interfacial concentration attainable by a compound.

The value of Gibbs free energy of adsorption estimated in the hexane/water system equal to 18.8 kJ/mol confirmed the high tendency of lecithin for adsorption at the considered interface. On the other hand the estimated value of  $\Gamma^{\infty}$  equal to  $1.754*10^{-6}$  indicates the formation of densely populated adsorption monolayer by lecithin molecules at the saturated hexane/water interface.

## CONCLUSIONS

For all the above mentioned results, it can be assumed, that extraction of egg yolk by hexane is not a highly exclusive process and it does not give chemically pure product. Hexane extract of egg yolk the high ability of reducing reveals the surface/interfacial tension. The effective lowering of hexane/water tension by approximately 25 mN/m is observed. Moreover two ranges of critical micelle concentration can be observed, so investigated material is a mixture of substances but not molecular entity. The surface/interface tension data, obtained experimentally, can be fitted by various adsorption equations. Using Szyszkowski equation the value of Gibbs free energy of adsorption estimated in the hexane/water system equal to 18.8 kJ/mol confirmed the high tendency of lecithin for adsorption at the considered interface. the estimated value of  $\Gamma^{\infty}$  equal to  $1.754*10^{-6}$  indicates the formation of densely populated adsorption monolayer by lecithin molecules at the saturated hexane/water interface. Even though more precise physicochemical description of surface activity of different fractions of egg yolk needs further investigations, it can be concluded, that examined lecithin is a potentially valuable raw material of natural origin for liposomes preparation.

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