

HYGROSCOPIC PROPERTIES OF ENOXIL-SILICA COMPOSITES

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Abstract. Enoxil-silica composites with various Enoxil-to-silica ratios were prepared by mechanical mixing of the biologically active Enoxil and fumed silica powders. The hygroscopic properties of the composites were studied by the gravimetric method. It has been shown that the use of Enoxil in composites with silica may significantly reduce the Enoxil ability to absorb water from the gas phase and, therefore, improve its storage stability. The strongest hygroscopicity reduction is observed for the composites with Enoxil-to-silica ratio of (0.15÷0.35):1, which corresponds to an approximate monolayer distribution of the Enoxil biomolecules on the silica surface.

Keywords: Enoxil, biomolecule, fumed silica, composite, hygroscopicity.

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Introduction

Enoxil is a biologically active powder with high antioxidant and antimicrobial properties [1,2]. Enoxil production procedure includes extraction of biologically active substances from grape seeds and subsequent oxidation of the extracted compounds (mainly proanthocyanidins, the representatives of flavonoids) with hydrogen peroxide [3]. Enoxil is a mixture of monomeric catechin/epicatechin molecules and their derivatives; mass-spectrometric studies revealed that the main components of Enoxil are gallic acid, catechin, epicatechin and catechingallate [2]. All these compounds have three or more reactive hydroxyl groups in the phenolic rings of the molecules. These are the groups that are mainly responsible for chemical and biological activity of flavonoids [4,5] and that provide Enoxil with high antioxidant/antimicrobial properties.

Due to a large amount of hydrophilic groups, Enoxil possesses enhanced hygroscopicity and requires to be kept under special conditions. The hygroscopicity of medicinal powders, dry extracts and other pharmaceutical medications may be reduced using auxiliary agents, in particular, silica [6,7]. The results of our previous study [8] show that adsorption of Enoxil on the silica surface or even simple mechanical mixing of Enoxil and silica powders leads to a marked decrease in the integral hygroscopicity of the composite, while bioactive molecules in such composites were found to retain their antioxidant

properties during prolonged storage at ambient conditions. The decrease in hygroscopicity seems to be due to the involvement of hydroxyl groups of Enoxil molecules into an interaction with silanol groups of silica surface and should depend on the contents of Enoxil and silica in the mixture.

The aim of this work was to prepare Enoxil-silica composites with various Enoxil-to-silica ratios and to examine the effect of this ratio on the composites water absorption.

Experimental

To prepare Enoxil-silica composites, fumed silica with specific surface area of 280 m²/g (A-300, Kalush, Ukraine) was mechanically mixed with Enoxil powder (produced at the Institute of Chemistry, Academy of Sciences of Moldova [3]) in a ball mill, the processing time was 5 min. The Enoxil-to-silica ratio in the composites was within the range (0.08÷0.90):1.

The *hygroscopicity* of the Enoxil and Enoxil-silica composites was determined gravimetrically as the sample mass (*m*) change after water absorption from the gas phase. Samples of certain mass were placed into weighing bottles, which then were placed into a desiccator with distilled water and held there at a temperature of 20÷22°C for 1÷365 days. The presented results are the average of three independent experiments, the data scattering corresponds to a 95% confidence interval. In order to evaluate the effect of the Enoxil-to-silica

ratio on the hygroscopicity of the composite, the data on the water absorption were presented in the following manner. For each composite and each period of time that the sample was held in the desiccator, we calculated the amount of water, $m(\text{H}_2\text{O})_{\text{cal}}/m(\text{composite})$ that would be absorbed by a sample consisting of appropriate amounts of silica and Enoxil, provided that the components of the composite do not effect the water absorption of each other. To do this, we used the determined in the same experiment values of water absorption for pure Enoxil, $m(\text{H}_2\text{O})_{\text{abs}}/m(\text{Enoxil})$, and pure silica, $m(\text{H}_2\text{O})_{\text{abs}}/m(\text{silica})$, as well as the data on the amount of the components in the composite. The obtained $m(\text{H}_2\text{O})_{\text{cal}}/m(\text{composite})$ value was compared with the actual water absorption value, $m(\text{H}_2\text{O})_{\text{abs}}/m(\text{composite})$, measured for the given composite. The difference was divided by the calculated water absorption value, $m(\text{H}_2\text{O})_{\text{cal}}/m(\text{composite})$, and expressed as a percentage. The obtained value Δ (Eq.(1)) characterizes the degree of reduction (when negative values) or the increase (when positive values) of the hygroscopicity of the composite in comparison with the sum of its components. It is equal to zero when the components do not affect the hygroscopicity of each other.

$$\Delta = \frac{m(\text{H}_2\text{O})_{\text{abs}}/m(\text{comp.}) - m(\text{H}_2\text{O})_{\text{cal}}/m(\text{comp.})}{m(\text{H}_2\text{O})_{\text{cal}}/m(\text{comp.})} \cdot 100\% \quad (1)$$

Quantum chemical calculations were carried out using semiempirical PM3 method (GAMESS, current version [9]) to find the optimal configuration for the adsorbed biomolecules on the surface of silica particle modelled by clusters of 36 $\text{SiO}_{4/2}$ tetrahedrons with 24 silanol groups.

Results and discussion

Figure 1 shows the water absorption data for the pure Enoxil, pure silica and for the composites with the Enoxil-to-silica ratio of (0.08÷0.90):1. As one can see from Figure 1, pure Enoxil rapidly absorbs water from the gas phase; after 12 months in the desiccator, the increase in the weight of the initial Enoxil, $m(\text{H}_2\text{O})_{\text{abs}}/m(\text{Enoxil})$, reached more than 1000%, while Enoxil became liquid, *i.e.*, it was in a dissolved state. The initial silica is also characterized by considerable hygroscopicity, the water absorption reached the value of $m(\text{H}_2\text{O})_{\text{abs}}/m(\text{silica})= 260\%$. All composites have less water absorption per unit mass, $m(\text{H}_2\text{O})_{\text{abs}}/m(\text{composite})$, than the initial Enoxil. Upon this, the hygroscopicity of the composites does not change monotonically with the change in

the Enoxil-to-silica ratio, and for the samples with the ratio of 0.15:1 and 0.23:1 (Figure 1, curves 4 and 5), the water absorption per unit mass is less than that for both pure Enoxil and pure silica (Figure 1, curves 1 and 2). Figures 2 and 3 give the Δ values *versus* Enoxil-to-silica ratio for the holding time in the desiccator of 1, 14, 30, 92, 141, and 365 days.

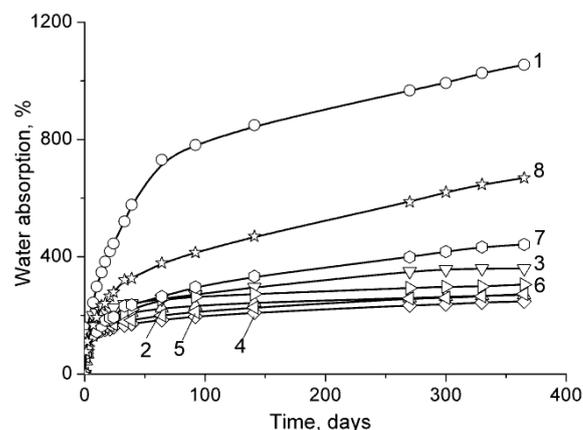


Figure 1. Absorption of water by Enoxil (1), silica (2) and composites with the Enoxil-to-silica ratio of 0.08:1 (3), 0.15:1 (4), 0.23:1 (5), 0.35:1 (6), 0.55:1 (7), 0.90:1 (8).

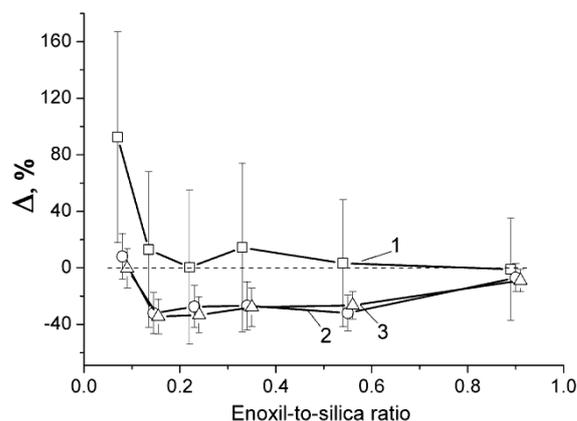


Figure 2. Dependence of Δ values on Enoxil-to-silica ratio for the holding time in the desiccator of 1 (1), 14 (2) and 30 (3) days.

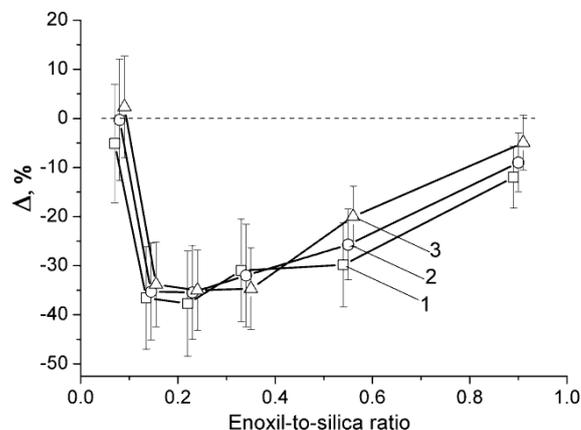


Figure 3. Dependence of Δ values on Enoxil-to-silica ratio for the holding time in the desiccator of 92 (1), 141 (2) and 365 (3) days.

As it was mentioned above, the Δ values characterize the changes in hygroscopicity of the composites in comparison with the sum of their components. Figures 2-3 show that for the samples with the ratio of (0.15÷0.90):1 and a holding time in the desiccator of 14 days or more, the Δ values are negative. It means that the composites absorb less water than individual components of the composite. As we noted earlier [8], this effect can be related to the interaction of hydroxyl groups of Enoxil and silanol groups of silica with each other and, therefore, to their exclusion from the interaction with water molecules. The interaction of surface groups of silica with hydroxyl groups of the biomolecules was confirmed by IR spectroscopic studies. As it was shown earlier [8], the intensity of the band at 3750 cm^{-1} (corresponding to isolated silanol groups of silica) in the Enoxil-silica composites spectra decreased in comparison with the initial silica spectrum, which indicates the involvement of silanol groups in an interaction with Enoxil. The decrease in the intensity of the band was observed not only in the case of Enoxil adsorption on the silica surface, but also for the mechanical mixture of Enoxil and silica powders prepared by grinding the components in a ball mill.

Apparently, in such a mixture Enoxil is in the form of small crystals or associates, bound, nevertheless, to the silica surface. The growth of powder dispersity during the grinding process should lead, on the one hand, to an increase in the hygroscopicity due to the increase of the surface area of the crystallites/associates and, on the other hand, to a reduction in water absorption due to an increase in the number of bonds between the crystals/associates and the silica surface groups. As it was noted in previous work [8], the first

factor appears to play a major role in the initial stage of powders/composites exposure to water, which is reflected in the enhancement of the composites hygroscopicity (Figure 2, curve 1). Over time, the hygroscopicity of the composites decreases in comparison with the sum of hygroscopicities of the composite components. It can be assumed that after the absorption of certain amounts of water, crystallites are further dispersed and dissolved in the absorbed water, followed by the redistribution of biomolecules on the silica surface to form an adsorbed layer. Indeed, as can be inferred from the data in Figure 1, after 14 days of maintaining the samples in the desiccator, the amount of water absorbed by the composites is 1.5-2.5 times greater than the total mass of the composites and 5-15 times greater than the mass of Enoxil in the composites. According to our previous studies on Enoxil solubility, such an amount of water is several times higher than the amount needed to completely dissolve Enoxil and, therefore, it is sufficient to induce Enoxil dissolution and adsorption of the dissolved biomolecules on the silica surface.

The most significant decrease in the hygroscopicity is observed for the composites with the Enoxil-to-silica ratio of (0.15÷0.35):1 (Figures 2, 3). As it is shown below, this range of ratios corresponds to an approximate monolayer distribution of biomolecules on the silica surface. The results of quantum-chemical calculations make it possible to estimate the equilibrium configuration of the main Enoxil biomolecules in the adsorption layer on the silica surface (examples of such equilibrium configurations are given in Figure 4) as well as to evaluate the surface area, occupied by one molecule.

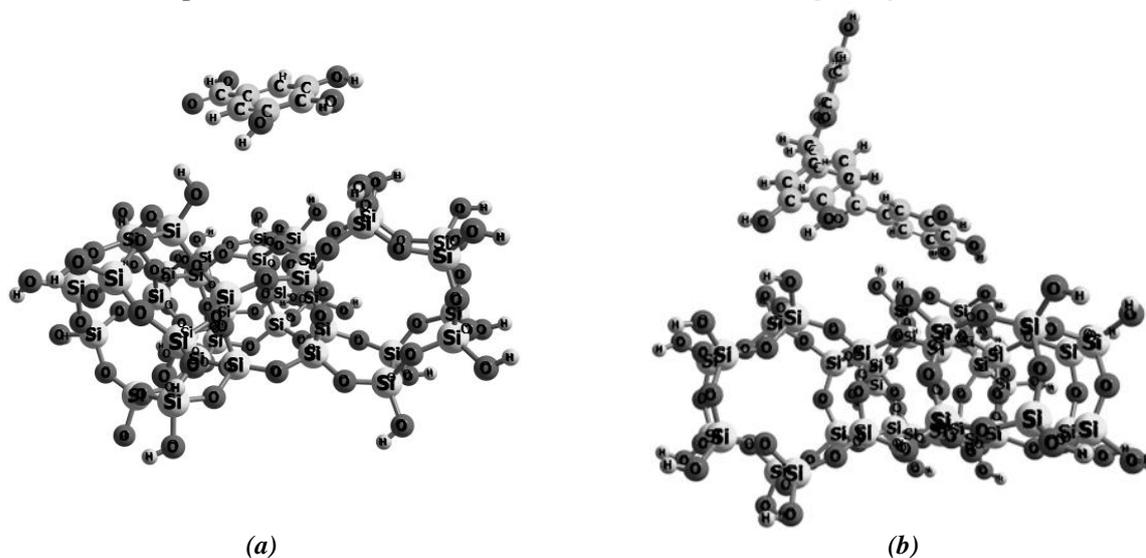


Figure 4. Equilibrium configuration for the smallest (gallic acid, (a)) and the largest (epicatechingallate, (b)) Enoxil biomolecules on the silica surface.

The obtained data show that this parameter for various molecules of Enoxil (gallic acid, catechin, epicatechin, catechingallate) ranges between 0.4 and 0.7 nm². Taking into account the silica surface area, 280 m²/g, the maximum amount of the smallest (gallic acid, molar mass 170 g/mole) and the largest (epicatechingallate, molar mass 442 g/mole) molecules, there should be around 0.20 and 0.31 g, respectively, on the surface of 1 g of silica.

With a monolayer distribution of Enoxil on the surface of silica, the largest number of hydroxyl groups of Enoxil biomolecules appears to be involved in interaction with surface silanol groups, as well as in lateral interactions with neighbouring Enoxil molecules. According to the results of quantum chemical calculations, bioactive molecules of Enoxil interact with the silica surface involving on the average 2 hydroxyl groups [8]. The molecules of gallic acid, catechin/epicatechin and catechingallate contain 3, 5 and 7 hydroxyl groups, respectively, *i.e.* from 1 to 5 hydroxyl groups remain free and can interact with water (or with neighbouring Enoxil molecules). The maximum reduction in the hygroscopicity of the composite compared to the sum of the components is about 40%, which approximately corresponds to excluding of about two-fifths of the groups of Enoxil molecules from the interaction with water.

Conclusions

The obtained results indicate that the use of Enoxil in the composition with fumed silica can lead to a significant decrease in the Enoxil ability to absorb water from the gas phase and to improve its storage stability.

The hygroscopicity of the Enoxil-silica composites depends on the components ratio, the greatest effect of hygroscopicity reduction (35-40%) is achieved for the samples with the Enoxil-to-silica ratio of (0.15÷0.35):1, which corresponds to an approximate monolayer distribution of biomolecules on the silica surface.

References

1. Lupascu, L.; Rudic, V.; Cotos, V.; Lupascu, T. Antimicrobial activity of the autochthonous compound Enoxil. *Journal of Biomedical Science and Engineering*, 2010, 3(8), pp. 758-762. DOI: [10.4236/jbise.2010.38101](https://doi.org/10.4236/jbise.2010.38101)
2. Duca, Gh.; Lupascu, T.; Vlad, P.; Kulcički, V.; Nastas, R. Studies on the water solubilization processes of oenotannins and their physico-chemical properties. *Chemistry Journal of Moldova*, 2006, 1(1), pp. 60-64. DOI: [https://dx.doi.org/10.19261/cjm.2006.01\(1\).01](https://dx.doi.org/10.19261/cjm.2006.01(1).01)
3. Lupascu, T.; Lupascu, L. The obtaining procedure of the water soluble enotannins. MD Patent, 2006, No. 3125. (in Romanian).
4. Pandey, K.B.; Rizvi, S.I. Plant polyphenols as dietary antioxidants in human health and disease. *Oxidative Medicine and Cellular Longevity*, 2009, 2(5), pp. 270-278. DOI: <https://doi.org/10.4161/oxim.2.5.9498>
5. Men'shchikova, E.B.; Lankin, V.Z.; Kandalintseva, N.V. Phenolic Antioxidants in Biology and Medicine. Structure, properties, mechanisms of action. Saarbrücken: Lap Lambert Academic Publishing, 2012, 496 p. (in Russian). <https://www.twirpx.com/file/2051208/>
6. Gallo, L.; Llabot, J.M.; Allemandi, D.; Bucalá, V.; Piña, J. Influence of spray-drying operating conditions on *Rhamnus purshiana* (Cáscara sagrada) extract powder physical properties. *Powder Technology*, 2011, 208(1), pp. 205-214. DOI: <https://doi.org/10.1016/j.powtec.2010.12.021>
7. Gallo, L.; Ramírez-Rigo, M.V.; Piña, J.; Palma, S.; Allemandi, D.; Bucalá, V. *Valeriana officinalis* dry plant extract for direct compression: preparation and characterization. *Scientia Pharmaceutica*, 2012, 80(4), pp. 1013-1026. DOI: <https://doi.org/10.3797/scipharm.1206-05>
8. Kuzema, P.O.; Laguta, I.V.; Stavinskaya, O.N.; Kazakova, O.A.; Borysenko, M.V.; Lupascu, T. Preparation and characterization of silica-Enoxil nanobiocomposites. *Nanoscale Research Letters*, 2016, 11(68), 7 p. DOI: <https://doi.org/10.1186/s11671-016-1287-y>
9. Schmidt, M.W.; Baldrige, K.K.; Boatz, J.A.; Elbert, S.T.; Gordon, M.S.; Jensen, J.J.; Koseki, S.; Matsunaga, N.; Nguyen, K.A.; Su, S.; Windus, T.L.; Dupuis, M.; Montgomery, J.A. Jr. General atomic and molecular electronic structure system. *Journal of Computational Chemistry*, 1993, 14(11), pp. 1347-1363. DOI: <https://doi.org/10.1002/jcc.540141112>