

INVESTIGATION OF INFRARED SPECTRA OF NANOSTRUCTURED COMPOSITES BASED ON THE COLLAGEN PROTEINS WITH IMMOBILIZED SELENIUM PRODUCTS

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A promising trend in nanobiotechnology is the development of sorption materials and their further use as carriers for biologically active substances [1]. The promising biosorbents are modified collagen proteins [2-3].

Collagen substances were obtained from a mixture of veins, tendons, fascia of cattle. At the first stage peroxidic- and alkaline hydrolysis was carried out under the following conditions: $c(\text{NaOH}) = 10\%$; $c(\text{H}_2\text{O}_2) = 3\%$; $\tau = 6$ ч; $t = 20^\circ\text{C}$). After solid residue to $\text{pH} = 8.5$ neutralizing and its gyroscope grinding with a hole diameter of 2-3 mm enzymatic hydrolysis was carried out under the following conditions: enzyme product dosage 0.02 % to the mass of the raw material, hydronic module 1:2-2,5; $\tau = 2,5-3,0$ h; $t = 37-40^\circ\text{C}$. Commercial enzyme product "Food Collagenase" from the hepatopancreas of the Kamchatka crab (the king crab) was used (the manufacturer – CJSC "Bioprogress", Shelkovo, Moscow region).

Sodium selenite (PhAE 42-0250-1024-01, manufacturer – "MCD", Moscow) and 4.4 di[3 (5-methylpyrazole] selenite (DMDPS) with the content of 0.657 g DMDPS in 100 cm³ of product (manufacturer – "Saffron" Ltd., Moscow, sanitary and epidemiological certificate №77.99.13.003.T.000518.03.06) were used as a source of selenium in the immobilization on collagen proteins.

For the IR spectroscopic study samples of the obtained collagenous substances were dried in the vacuum- sublimation unit at a temperature ranged from -40 to $+36^\circ\text{C}$ for 24 hours. After that they were thoroughly ground in an agate mortar grinder to obtain a uniform fine powder, and then tablets were made with the previously dried and ground powder of the optically pure monocrystalline KBr at the following ratio : 0.1 mg of Sample is 100 mg of KBr.

The infrared spectra of collagen substances were obtained by the spectrometer with Fourier transform «Vertex-70." GRAMS 4/32 program was used for further processing.

It was proved [4], that the two-staged modification of collagen substrates with the use of sequential peroxygen-alkaline and enzymatic hydrolysis resulted in the formation of peptides having a molecular mass in the range of 50-100 kDa, which allowed their use as a matrix for the

immobilization of selenium compounds, dimethyldipirosolilselenite (DMDPS) in particular.

To obtain comparable data the technique of spectra processing by the baseline method was used by us. [5]. Maxima in the longwave length of 1453 cm^{-1} , and the shortwave length of 2923 cm^{-1} were selected as the standard. Selection of standard maxima is due to the fact that the baseline method gives the most adequate data when related to the analyzed frequency lines are taken as the standard ones.

Calculation of h/h_{st} on a maximum 3308 cm^{-1} indicated that these values in the sequence: starting collagen - sorbed Na_2SeO_3 at $\text{pH} = 5$ - sorbed Na_2SeO_3 at $\text{pH} = 10$ - sorbed DMDPS varies as $0.25 - 2.36 - 0.62 - 0.76$. This indicates the most high water content in the samples of collagen having immobilized sodium selenite at $\text{pH} = 5$. The reason is the increase of water content in the hydration membranes of carboxylate ions and a quaternary nitrogen of collagen.

It is found out that the immobilization of selenium products on collagen goes by means of their chemical interaction with the carboxyl and the amino groups of side chains of the protein molecules to form oppositely charged ions. The degree of interaction of selenium products with collagen varies as follows Na_2SeO_3 ($\text{pH} = 5$) > DMDPS > Na_2SeO_3 ($\text{pH} = 10$). In this case, the immobilization of selenium products have little effect on the conformation of the collagen molecules.

References

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