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STRUCTURAL INVESTIGATIONS OF ENTEROSORBENTS BASED ON NATURAL SILK

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Features of an enterosorbent based on the fibroin of natural silk hydrolyzed in acid and alkaline solutions have been investigated by x-radiographic, electron-microscopic, and sorption methods. It has been shown that, regardless of the pH of the solution, a longitudinal-transverse cleavage of the fibroin of natural silk takes placed on hydrolysis, although in the presence of alkali this process is more intensive than in an acid solution of the same normality.

In recent years, great attention has been devoted to the elimination of toxic and ballast metabolites from the gastrointestinal tract with the aid of sorbents. This process has acquired the name of enterosorption, and the sorbents used in it the name of enterosorbents [1, 2]. In an investigation of the hydrolysis of the fibroin of natural silk (FNS) in dilute solutions of acids and alkalis we have obtained enterosorbents the detoxication properties of which are superior to those of the carbon sorbent SKN-2K. In the present paper we consider the results of structural investigations of FNS hydrolyzed in solutions of HCl and of KOH.

The powders obtained were studied by x-radiographic, electron-microscopic, and optical means and also by sorption methods. On a diffractogram of the initial FNS (Fig. 1) a maximum was observed at $2\theta = 20.5°$, which is characteristic for rather disordered silk; the degree of crystallinity (DC) of this specimen was 55%.

After hydrolysis in 2 N hydrochloric acid (Table 1), the DC of the FNS had risen to 62%, obviously through removal of the amorphous fraction (sericin residues). On the diffractograms a second maximum has appeared at $2\theta = 20.9°$, showing a more ordered structure of the specimen, which consisted mainly of fibroin. However, a further increase in the acidity of the medium caused degradation of the fibroin that was the greater the higher the concentration of acid and led to the disappearance of the second maximum and to a decrease in the DC.

The same extremal relationship of the change in the DC of the FNS was observed on hydrolysis in an alkaline medium. Although after treatment with 1 N KOH two maxima appeared at the same angles (Fig. 1) and the DC rose to 59%, a further increase in the concentration of alkali (2 N KOH) led to a fall in the DC to 52% and the retention of only one maximum. Thus, it has been shown that, regardless of the pH of the solution, the hydrolysis of FNS first leads to a rise in the DC as the result of the removal of the amorphous fraction, and then to its fall through the degradation of the fibroin, while in an alkaline medium this takes place more intensively than in an acid medium of the same normality.

We determined the dimensions of samples under the optical microscope with the aid of an ocular micrometer (Table 1). A decrease was observed in the dimensions of the FNS, the values of which were the smaller the more severe the conditions of hydrolysis: this decrease was more effective in an alkaline medium (17.9 μm at 2 N KOH, and 18.0 μm at 6 N HCl).

The microscope investigations showed that the initial sample of FNS (Fig. 2) consisted of long thin fibers with bright fluorescence in polarized light, showing their anisotropy.

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TABLE 1. Degree of Crystallinity (DC) and Particle Size of Samples of FNS after Hydrolysis

Expt. No.	Conditions of hydrolysis	DC, %	Particle size, μm		
			min.	max.	mean
1	initial FNS	55			
2	2N. HCl, 90°, 1h	62	30.0	385	108.8
3	4N. HCl, 90°, 1h	55	7.7	54	27.0
4	6N. HCl, 90°, 1h	44	7.7	38	18.0
5	1N. KOH, 90°, 1h	59	14.4	230	66.9
6	2N. KOH, 90°, 1h	52	7.3	38	17.9

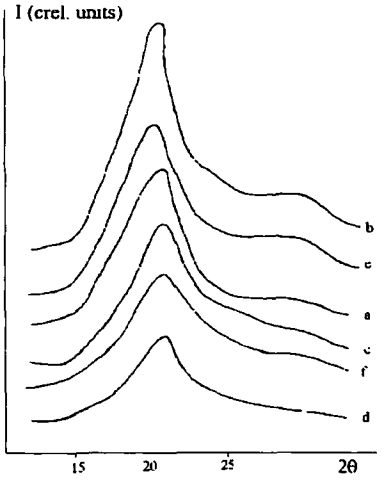


Fig. 1. X-Ray diffractograms of FNS after hydrolysis at 90°C for 1 h in various media: *a*) initial; *b*) 2 N HCl; *c*) 4 N HCl; *d*) 6 N HCl; *e*) 1 N KOH; *f*) 2 N KOH.

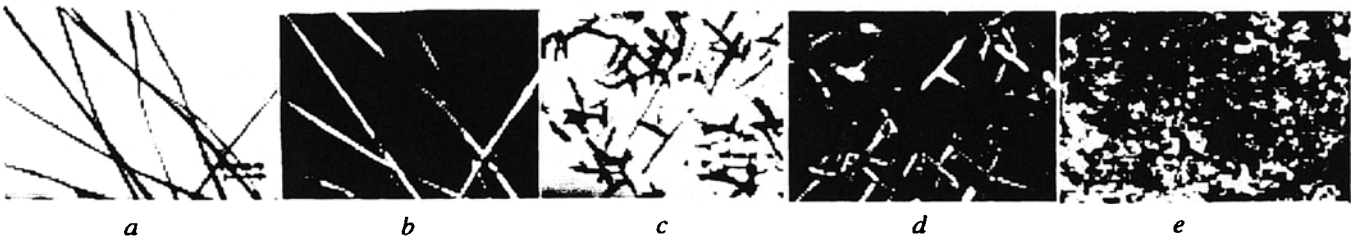
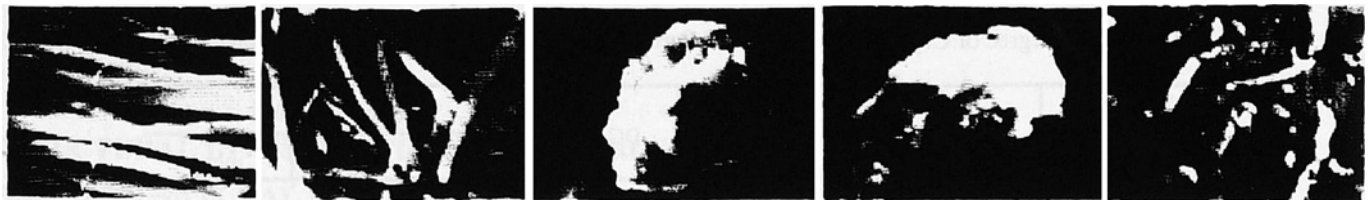


Fig. 2. Optical photographs in transmitted (*a*, *c*, *e*) and polarized (*b*, *d*) light after hydrolysis of the FNS at 90°C for 1 h in HCl of various concentrations: *a*, *b*) initial FNS; *c*, *d*) 2 N HCl; *e*) 4N HCl.

After hydrolysis in 2 N HCl, the fibers had become much shorter: the formation of rod-like particles of different thicknesses was observed, which suggests transverse cleavage of the FNS during hydrolysis. They fluoresced brightly in polarized light. The number of such particles decreased with a rise in the concentration of acid. At the same time, a large number of small spherical particles of crystalline fibroin with bright fluorescence and larger shapeless formations with weak fluorescence, obviously of amorphous nature, appeared. A further increase in the concentration of the acid (6 N HCl) led to the disappearance of the anisotropic particles; only small spherical particles and also shapeless formations appeared, which indicated further amorphization of the fibroin.



a *b* *c* *d* *e*

Fig. 3. SEM photographs of the FNS after hydrolysis at 90°C for 1 h in various media: *a*) initial; *b*) 2 N HCl; *c*) 4 N HCl; *d*) 6 N HCl; *e*) 1 N KOH.

On hydrolysis in an alkaline medium (1 N KOH), an association of anisotropic fibrils, small spherical particles with bright fluorescence, and shapeless weakly fluorescing formations appeared. A rise in the concentration of alkali (2 N KOH) led to more intensive hydrolysis as a result of which the elongated particles disappeared and the number of fluorescing spherical particles and of shapeless elements increased, which presupposes the same degradation mechanism as in acid hydrolysis and, in addition, showed an amorphization of the material, confirming the x-radiographic results.

The SEM investigations (Fig. 3) showed that, in addition to long smooth fibers fairly uniform in diameter, hydrolysis led first to short fibers the width of which either decreased or remained the same as for the initial fibers. A further increase in the concentration of the hydrolyzing reagent led to the disappearance of the elongated fibrils and to the appearance of shapeless formations the relief surface of which was coated with adherent small particles formed during hydrolysis. It must be mentioned that the nature of the structure of the surface after hydrolysis did not depend of the composition of the hydrolyzing medium.

Tables 2 and 3 give the results of sorption investigations of the samples. For the initial FNS we observed a fairly high sorption capacity (8.20% at 65% relative humidity) due to the high values of the specific surface (144.05 m^2/g) and the pore volume (0.287 cm^3/g), which is connected with silk's high hydrophilicity. After hydrolysis, the samples possessed a lower sorption capacity (1.6—4.00% at 65% relative humidity) because of the increased crystallinity, the minimum capacity for absorbing water being possessed by FNS that had been hydrolyzed with 2 N HCl (1.60%), which had the maximum DC, resulting in low values of S_{sp} (21.19 m^2/g) and of W_0 (0.040 cm^3/g). However, with an increase in the concentration of the hydrolyzing medium (4 and 6 N HCl) the sorption capacity of the powders obtained rose somewhat through an increase in the total specific surface (72—74 m^2/g) and in the amorphization of the FNS.

Thus, the sorption properties of the preparations were determined by two oppositely directed processes from the point of view of their influence on sorption capacity.

Thus, it has been established that, as the result of transverse-longitudinal cleavage on the alkaline or acid hydrolysis of natural silk, the anisotropy of the fibers disappears and a large number of small particles fluorescing in polarized light arise the change in the degree of crystallinity and the sorption capacity of which bear an extremal nature, which is connected with an initial rise in the DC because of the elimination of amorphous sericin followed by amorphization through the destruction of the crystalline structure of the fibroin. This ensures a fairly high specific surface and large pore volume of the pulverulent material with a structure in high relief, which makes it possible to use the products of the hydrolysis of FNS as enterosorbents with good detoxication properties.

The sorbents synthesized are effective in the treatment of acute toxic hepatitis and hepatonephric insufficiency and lead to a fall in the level of creatinine, urea, bilirubin, and cholesterol in the blood [3, 4].

TABLE 2. Sorption of Water Vapor (%) by FNS Samples (1—4) after Hydrolysis

Rel. humidity, %	1 (init.)	2 (2N. HCl)	3 (4N. HCl)	4 (4N. HCl)
10	2.60	0.25	1.30	0.90
30	4.90	0.60	2.50	2.10
50	6.70	1.20	3.30	2.90
65	8.20	1.60	4.00	3.50
80	11.60	2.30	5.50	4.40
90	15.30	2.90	7.60	5.90
100	22.70	4.00	10.10	9.70

TABLE 3. Sorption Characteristics of Samples (1—4)

Index	1	2	3	4
X_m, g/g	0.0409	0.0060	0.0212	0.0205
S_{sp}, m^2/g	144.05	21.191	74.426	72.057
W_0, cm^3/g	0.227	0.040	0.101	0.097
r_g, Å	31	38	27	27

X_m - capacity of monolayer.

EXPERIMENTAL

The samples studied were obtained by hydrolyzing FNS with solutions of hydrochloric acid (2, 4, and 6 N) and of caustic potash (1 and 2 N). Hydrolysis was conducted at 90°C for 60 min. Liquor ratio 1:20.

The x-radiographic investigations were conducted on a DRON-3M diffractometer with monochromatized CuK_α radiation at a voltage of 25 kV and a current strength of 13 mA in the angular interval of 2θ = 10—35°. Degrees of crystallinity were calculated from the ratios of peak intensities:

$$CD = [(I_{cr} - I_a)/I_{cr}] \times 100\%.$$

The electron-microscopic investigations were made on a RÉM-200 instrument. The specimens were first shadowed with silver in a VUP-4k vacuum apparatus. The microscope studies were performed with a MBI-6 microscope in transmitted and polarized light. Particle sizes were measured with an ocular micrometer, after which the mean values of two parallel determinations of 30 measurements each were taken.

The sorption measurements were made on a McBain vacuum balance at 25°C in the interval of relative humidities from 0 to 100% with water as the sorbate. We used the BET equation to calculate from the sorption results the specific surface, S_{sp}, the pore volume, W_0, and the mean effective radius of the capillaries r_c.

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