SPECTROPHOTOMETRIC STUDY OF THE INTERACTION OF TRICHLOROACETIC ACID WITH HEMOGLOBIN IN UREA SOLUTION

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The structure of proteins [1] can be studied with the aid of denaturing agents, and such investigations may also detect fine structural changes.

We have used trichloroacetic acid (TCA) and urea, which are described as denaturing agents causing the dissociation of hemoglobin [2], while a combination of them is used for the isolation of its subunits [3].

A spectrometric study of the interaction of TCA with solutions of hemoglobin was performed in a Perkin-Elmer 402 spectrophotometer in the visible and ultraviolet regions of the spectrum. In the visible region, the spectra of 0.02% solutions of hemoglobin in water and 6 N urea were identical. The addition of TCA to aqueous solutions of hemoglobin led to the disappearance of characteristic peaks in the 540-580 nm region and to the appearance of a diffuse peak with a maximum at 380 nm (Fig. 1a, curve 1). The interaction of TCA with hemoglobin in 6 M urea solution caused the appearance of another narrow absorption band with a maximum at 400 nm and an isobestic point at 377 nm (Fig. 1, a, curve 3), the size of this peak depending on the amount of acid added (Fig. 1, b).

Similar investigations were carried out with aqueous solutions of heme-albumin (λ_{max} 404 nm). The addition of TCA to them did not lead to any spectral changes whatever. The influence of urea on these solutions could not be determined because of the complete denaturation of the protein, leading to the appearance of a precipitate.

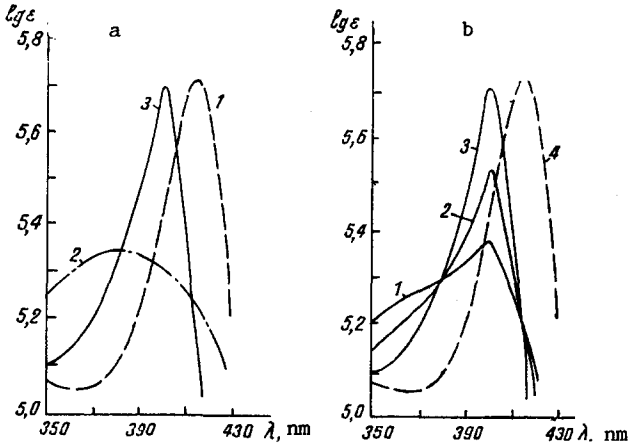

Fig. 1, a. Spectrograms of solutions of hemoglobin in the 350-440 region: 1) initial solution of 0.02% of hemoglobin in 6 M urea; 2) solution of 0.02% of hemoglobin in water with the addition of trichloroacetic acid; 3) solution of 0.02% of hemoglobin in 6 M urea with the addition of trichloroacetic acid.

Fig. 1, b. Spectrograms of solutions of 0.02% of hemoglobin in 6 M urea with various concentrations of trichloroacetic acid in them: 1) 0.60%; 2) 0.69%; 3) 0.75%; 4) initial solution.

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These results permit possible structural changes in the hemoglobin molecule taking place under the action of TCA in urea solution to be suggested, as is indicated by the appearance of characteristic absorption at 400 nm.

LITERATURE CITED

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ELECTROPHORETIC INVESTIGATION OF THE PROTEINS OF COTTON SEEDS

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The detection and study of protein markers responsible for valuable economic features is of great importance for the national economy [1].

An electrophoretic study of the protein composition of cotton seeds in the presence of sodium dodecyl sulfate and 2-mercaptoethanol has permitted the detection of the heterogeneity of the salt-soluble fraction [2] but in this process the quaternary structure of the proteins is disturbed, while the retention of the native nature of marker proteins is important for the investigation of their biological functions [3].

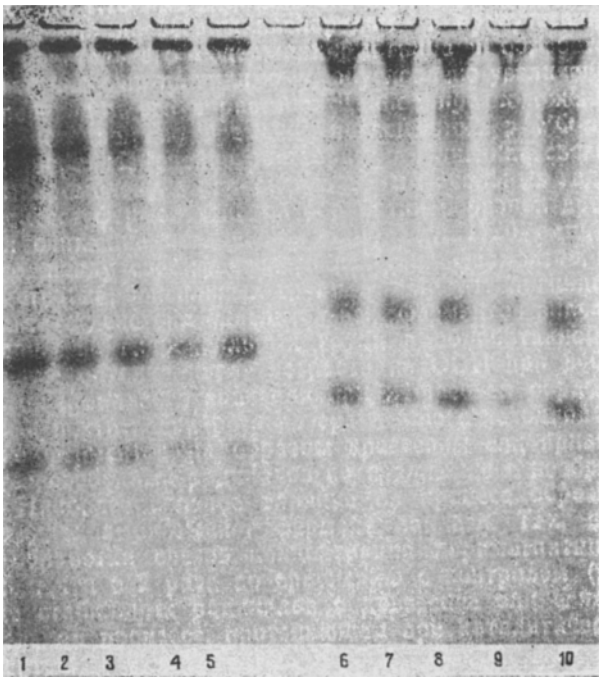

Fig. 1. Electrophoretogram of the proteins of individual seeds of the varieties Tashkent-1 (G. hirsutum L (1-5) and S-6030 (G. barbadense) (6-10).

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