Open Peptide Chains in α-Chymotrypsin

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After oxidation of either α-chymotrypsin or di-*iso*propyl - phosphat - α - chymotrypsin with performic acid it is in both cases possible to separate the reaction products in three fractions: The reaction mixture is dissolved in alkali, and pH brought down to 6, whereby a precipitate (fraction B) is formed. The supernatant is placed on a small Dowex 50 column (H^+ form). By elution with water fraction A is collected and by subsequent elution with NH_4OH fraction C is obtained. Fraction A is a peptide of 13 amino acids having N-terminal cysteic acid and C-terminal leu. Preliminary experiments seem to indicate that fraction C mainly consists of a peptide of 22 amino acids having N-terminal ala, but no C-terminal residue susceptible to attack by carboxypeptidase. Fraction B seems to be a long peptide chain containing N-terminal leu or ileu, and C-terminal tyr. In the case of DIP-chymotrypsin this fraction contains all the phosphorus. It seems to be possible to correlate the results with the structural model for α-chymotrypsin which has been proposed by Neurath and by Desnuelle.

Dielectric Increment and Dispersion of Tropomyosin — a Highly Polar Protein

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A specimen of pure tropomyosin from rabbit skeletal muscle [1] (kindly supplied by Drs Bailey and Tsao, Cambridge) has been studied dielectrically in pure aqueous solution. With concentrations between 0.01 and 0.1 g/100 ml (pH about 6.7) dielectric increments between 1.6 and 7.0 dielectric constant units were obtained. From these values reduced dielectric increments per g per liter, $\Delta\varepsilon/g$, between 4.5 and 7.7 were calculated. There was a slight tendency for the $\Delta\varepsilon/g$ values to increase at low concentrations. As the solutions of tropomyosin showed marked time effects (mainly a decrease of the increment with age of solution) this tendency is not considered significant, however. $\Delta\varepsilon/g$ is thus essentially independent of concentration as has also been found with other simple proteins studied. Such proteins give $\Delta\varepsilon/g$ values between 0 and 2. The $\Delta\varepsilon/g$ obtained for tropomyosin is thus about 3 times higher than with the most polar protein studied so far (β-lactoglobulin).

Dielectric dispersions were obtained for tropomyosin which were markedly influenced by concentration, the critical frequency being displaced towards lower frequencies with decreasing concentration. This is contrary to the case with other simple proteins for which the critical frequency is independent of concentration or displaced towards higher frequencies with decreasing concentration (β-lactoglobulin). The dispersion curve corresponds approximately to a single relaxation time in each case. No further dispersion was obtained down to very low frequencies. The relaxation times calculated varied between 0.75×10^{-7} sec. at 0.10 % concentration and 1.5×10^{-7} sec. at 0.027 %.

The type of dielectric dispersion and the order of the dielectric increments obtained for tropomyosin (and for a nucleotropomyosin previously reported to the Society in 1953) approach those of a number of polyelectrolytes studied dielectrically, but differ markedly from those of other simple proteins. This is probably due to the unusually high content of free ionizable groups [1] (from arginine, lysine and especially glutamic acid residues) and the linear shape [2] (double chain, asymmetry *ca.* 30) of tropomyosin. Tropomyosin shows polyelectrolyte properties also in other respects. The marked viscosity and streaming birefringence of aqueous tropomyosin solutions is sharply reduced by small amounts of simple electrolytes [2]. This has been ascribed to a desaggregation [2], but a contraction of the molecules due to electrostatic screening may also contribute. There is thus a suggestive parallelism between this contractile muscle protein and synthetic mechano-chemical systems obtained by crosslinking acid polyelectrolytes to 3-dimensional networks [3].

1. Bailey, K. *Biochem. J. London* **43** (1948) 271; **49** (1951) 23.
2. Tsao, T.-C. and Bailey, K. *Discussions Faraday Soc.* **13** (1953) 145.
3. Katchalsky, A. *J. Polymer Sci.* **12** (1954) 159.