portional to the acidity, it ceases to increase proportionally to the acid when enough acid has been introduced to ionize nearly the whole of the base. No evidence has yet been found that chloride ion becomes involved in the rate-determining process, even when chlorine appears in the product. No sign has been discovered of quadratic dependence on acid, as in the benzidine change.

We think it would be very difficult to accommodate all these facts on any other view of the reaction than that advanced. But the matter is still being tested. As to products, Dr. C. A. Bunton is checking oxygen identities among factor, product and solvent, by the use of heavy-oxygen water; and as to kinetics, evidence of an S_N2' form of the reaction is being sought.

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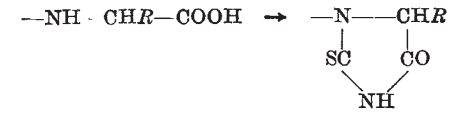
Identification of the Free Carboxyl Groups in Peptides

The Schlack and Kumpf procedure[1] for identifying the amino-acids which provide terminal carboxyl groups in peptides has been made the basis of a micro method which works well with simple peptides and may be of use in studying the free α-carboxyl groups in proteins. In the case of glycylleucine and glycylvaline, the method has been used successfully, although preliminary experiments with insulin and wool keratin have indicated that some modification of the procedure may be necessary before it can be used with proteins.

Dry ammonium thiocyanate (0·1 gm.) and acetic anhydride containing 10 per cent acetic acid (1·0 ml.) are heated with the benzoylated dipeptide (0·1 gm.) for 30 min. on a boiling water-bath. After cooling the orange solution, 50 ml. of water is added, the mixture left overnight, and the light brown precipitate filtered off. It is then washed well with water and dried; yield 0·1 gm.



The thiohydantoin derivative of the benzoylated dipeptide (20 mgm.) is refluxed with 20 per cent hydrochloric acid (10 ml.) for 8 hr. and the hydrolysate extracted twice with nitromethane. After washing with water, the extract is taken to dryness *in vacuo* and hydrolysed in a sealed tube with 10 ml. of 0·25 *N* barium hydroxide for 48 hr. at 140° C.[2]. The barium hydroxide is removed as carbonate and the hydrolysate evaporated down and examined for amino-acids by paper chromatography. Ninhydrin development of the chromatogram shows a clear spot corresponding to the acid carrying the free carboxyl group. No other spot could be observed.

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Influence of Salt on the Size and Shape of a Protein – Detergent Complex

Studies of the interactions between detergent and protein solutions by a wide variety of techniques have provided useful information on the types of binding involved in these systems[1] and on the denaturation and re-orientation of protein molecules[2]. In particular, the system bovine serum albumin – sodium dodecyl sulphate has been extensively investigated by electrophoresis, sedimentation and viscosity methods by Putnam and co-workers[3], and the heats of interaction for this system have recently been measured in this Laboratory by a direct calorimetric method[4]. The present communication deals with the effect of salt on the turbidity of bovine serum albumin – sodium dodecyl sulphate solutions.

The light-scattering apparatus used was similar in principle to that already described[5], with the improvement that variations in the intensity of the incident light were corrected by reflecting a constant fraction of the latter on to a second photomultiplier tube. Moreover, since intensities of the scattered light were measured at 50° and 130° as well as at 90° to the incident beam, a large circular glass water-bath was placed around the optical cell containing the scattering solution in order to minimize reflexions and water–air refractive index corrections. The apparatus was calibrated by measurement of the turbidity of solutions of bovine serum albumin, a molecular weight of 70,000 being assumed for this protein. Since the protein solutions showed no dissymmetry of scattering, the turbidity (τ) could be calculated from the equation[6]:

$$1/M = \frac{32\pi^3 n_0^2 (dn/dc)^2}{3N_0 \lambda_0^4} \cdot \frac{c}{\tau} + 2Bc = \frac{Hc}{\tau} + 2Bc;$$

where M is the molecular weight of the protein, c its concentration in gm./c.c., dn/dc its refractive increment in solvent of refractive index n_0, N_0 is Avogadro's number, B and H are constants.

The refractive index increments were determined with a Rayleigh–Haber interferometer (Hilger). Solutions were optically clarified by gravity filtration through sintered glass, Grade 4 (clarification was not complete if suction were used), or, in the case of solutions containing sodium hydroxide, by centrifugation.

Curve A in the accompanying graph shows the value Hc/τ as a function of the detergent/protein ratio for the system sodium dodecyl sulphate – bovine serum albumin in salt-free solutions. The concentration of protein was maintained constant throughout at 1 per cent (w/v). Since the addition of detergent has little effect on the refractive increment, dn/dc, of the protein, the linear increase in Hc/τ with detergent/protein ratio is due almost entirely to a decrease in turbidity. Curve B shows that this