Letters to the Editor.

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Examination of Protein Films.

THE simplest types of molecular architecture are to be found in materials in the form of films. The examination of films of protein materials is attended with many difficulties, absent in the case of simpler organic substances like the fatty acids.

Whilst the long chain organic compounds containing polar groups can readily be converted into the state of uniform unimolecular films on liquid surfaces on account of their solubility in volatile non-polar solvents, this method is not available for proteins. The method of dispersing the protein in alkaline or acid solutions and spreading the dispersion on a water surface was adopted by Gorter and Grendel, who with the aid of a Langmuir trough obtained a series of interesting measurements on the force-area curves of various proteins. Optical ultra-microscopic examination of such films by Zocher and Stiebel revealed that, except in a few cases, for example, hæmoglobin and casein on strongly acid solutions, protein films spread in this manner were not uniform, whilst the magnitude of the mean limiting thickness obtained, 9-10 A., indicates either that films obtained in this manner even when uniform must be in a relatively close packed state or that part of the protein must have disappeared into the bulk phase. Denatured protein films alone appear to be formed from disperse solutions by the operation of the forces of surface tension ; a somewhat lengthy procedure.

We have found it possible to spread a number of proteins such as egg albumen, gliadin, and glutenin in the form of highly disperse uniform films on the surface of water in the following manner : a thin quartz fibre which may for convenience serve as the arm of a microbalance is coated with pure paraffin wax of relatively low melting point. The surface of the wax is lightly dusted with the powdered protein. The protein is then rendered slightly moist by holding the fibre over steam until the increase in weight is some ten per cent of the weight of protein present. On touching a water surface with the fibre tip the protein spreads relatively slowly and apparently quite uniformly over the surface. The microbalance is readily calibrated by spreading myristic acid from the fibre, for both the force-area curves as well as the potential-area curves are well established for this material.

Some proteins, for example, keratin and the xerogel of gelatine, cannot be spread in this manner, and, indeed, unimolecular films of this latter protein do not appear attainable on water surfaces owing to the readiness with which the gelatine undergoes dispersion in the bulk phase. We have examined a number of such films with the aid of a Langmuir trough and, as was to be anticipated, their properties are distinct from the thicker films examined by Gorter and Grendel. In the case of gliadin, for example, a homogeneous film exerting a perceptible surface pressure is obtained with a surface concentration of $0{\cdot}36 \times 10^{-7}$ gm./cm.^{2}. On compression a linear force-area curve is obtained up to a surface concentration of about $0{\cdot}7 \times 10^{-7}$ gm./cm.^{2}. The form of the curve is altered by alteration of the acidity of the substrate. At higher compressions the film, originally fluid, undergoes transition to a gel-like film. Over a limited range of pressure at least, this transformation appears to be reversible. The ' gel ' form is curiously elastic, and the protein in this form may be related to that which, in the bulk phase, is generally described as denatured.

Examination of aqueous protein systems by the method of surface potentials gives us information even more complete than that obtainable from data on the force-area characteristics. It is found, for example, that no change takes place in the magnitude of the vertical component of the electric moments of the polar groups comprising the film material until the two-dimensional sol gel transition sets in ; an indication that this latter form is associated with some type of interaction of the polar groups in adjacent polypeptide chains. A study of the three-dimensional gels of gelatine by this method reveals the fact that the surface phase has a different setting point from that of the bulk phase and is essentially lyophobic in character, but the spreading of fatty acids over sols can readily be followed. The change in the air-liquid potential occurring on the tryptic digestion of an albumen film on a water surface by means of trypsin in the substrate is relatively large, about 75 mv., and permits of closer analysis of such chemical reactions occurring at interfaces.

The dependence of the rate of tryptic digestion in a bulk phase on the acidity of the environment is likewise to be observed in the surface reaction, but with phosphate buffer solutions there appears to be a shift towards the acid side of about 1 *p*H in the optimum acidity. The characteristic curve of the reaction of trypsin and egg albumen shows that the digestion of the albumen apparently takes place in stages and that an inhibition of the reaction can occur by means of an addition of small quantities of already digested albumen solutions. The digestion of egg albumen by pepsin reveals a simpler mechanism and no final solution of the egg albumen film takes place.

Preliminary investigation holds out some promise of obtaining more precise information both as to the constitution and reactions—general and specific—of protein films by these methods.

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Absolute Values of X-Ray Wave-lengths and the Fundamental Atomic Constants.

THE wave-lengths of the X-ray spectra are now known with a considerable degree of accuracy (in favourable cases to at least 1 in 100,000). But this means only that the ratio between the wave-lengths and a certain distance within a crystal lattice (for example, the distance between the atomic layers parallel to the cleavage faces of calcite) is measurable with this accuracy. The computation of the dimensions of the crystal lattice from the data involved (electronic charge, density of the crystal, and so on) give the absolute value of the atomic distance with only moderate accuracy (say 3 in 1000). It has therefore been necessary to fix an arbitrary value for the atomic distance in question and thereby for the scale of the X-ray wave-lengths, the *X units*. The agreement between this scale and the C.G.S. units is for this reason ascertained only within the last-mentioned limit. On the other hand, if we were able to fix the scale of the *X units* in *centimetres*, this gives us a possibility of determining in absolute values the atomic distances and, indirectly, the other fundamental constants (for example, the electronic charge).