

interact with four groups of the enolase molecule, the overall dissociation constant being about 2×10^{-5} mole/l., a value of the same order of magnitude as that calculated from the activity measurements.

If it is assumed that the four binding groups are identical and do not interact, the logarithm of the first association constant can be calculated and is 5.3. Thus, the interaction of manganous ions with enolase is stronger than that demonstrated with any amino-acid or simple peptide⁶⁻⁷. It is of interest to note that the enolase molecule contains eight sulphur atoms⁸, since Cohn *et al.*⁸ have recently suggested that cystine may furnish the complex-forming group in metal-activated enzymes.

The work described in the present communication is being extended and a detailed report will be published elsewhere.

I wish to thank Prof. Arne Tiselius for his interest in this investigation.

Note added in proof. Experiments on the activation of enolase by zinc ions, carried out since the above communication was submitted for publication, demonstrated a fall in activity at high metal ion concentrations. This led to a re-investigation of the activation by manganous ions, which showed that the data are more correctly interpreted by assuming two types of interaction between manganous ions and enolase, one giving an active complex and the other resulting in inhibition. If this inhibitory effect is taken into account, the hypothetical dissociation constant of the active complex is found to be 4.4×10^{-5} mole/l. (as compared with 2.5×10^{-5} mole/l. above), while that of the inactive complex is 2×10^{-3} mole/l.

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A Biophysical Test for Aldohexoses

I HAVE observed that some sugars inhibit the flocculation of bottom-fermentation brewer's yeast in tap water, whereas others do not.

Sugars which inhibit flocculation	Sugars not inhibiting flocculation
Glucose	Fructose
Mannose	Galactose
Maltose	Lactose
Sucrose (after interaction for some time)	Xylose
	Arabinose
	Dextran, etc.

The flocculation is thus inhibited by aldohexoses. This fact can be used for a quick test for glucose in the presence of fructose. About 30 mgm. of freeze-dried brewer's yeast were suspended in 5 ml. of tap water. When held at rest, the yeast flocculates, and is precipitated within one minute. On addition of an equal volume of the solution under examination, the presence of the above aldohexoses is indicated by the fact that the yeast suspension remains stable for a longer time (some hours). The influence is distinct up to a sugar concentration of 0.1 per cent.

The pH of the solution should be kept in the range 4-7, and the temperature above 25° C. As a condition for flocculation is the presence of calcium or magnesium salts in the medium, anions giving undissociated salts with these metals, such as oxalate, fluoride, phosphate and carbonate, should be avoided.

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Dynamic Potentials of Freshly Formed Liquid Surfaces

As a basis for the study of the adsorption of a surface-active solute to a fresh liquid surface, it is necessary to establish with accuracy the changes in the physical properties of the surface which occur immediately after its formation, and throughout the adsorption process. Two important properties, which undergo considerable change as adsorption proceeds, are the surface tension and the surface potential. We have recently observed that the surface potential of aqueous solutions of many long-chain compounds, at the instant when adsorption is complete, differs widely from the ultimate equilibrium value. The changes in potential following establishment of concentration equilibrium between surface and bulk solution are comparable in magnitude with those occurring during adsorption, and erroneous deductions may result from the assumption that dynamic surface tensions and potentials change in a similar manner with age of surface.

Measurements of the rate of change of surface tension in aqueous solutions of straight and branched-chain alcohols¹ have shown that the surface tension of such solutions is determined primarily by surface concentration of solute, and is little influenced by molecular orientation in the surface. Changes in dynamic surface tension therefore provide a satisfactory means of following the development of an adsorbed film; when concentration equilibrium is established, no further appreciable change in tension occurs.

Curves A and B in the graph compare the change in surface tension and potential respectively for a 0.0045 per cent aqueous solution of decyl alcohol at 15°. Whereas tension is constant after one minute, the potential shows a considerable rise after adsorption is complete. In interpreting these effects, we consider that adsorbing molecules of solute arrive at the surface in a disorientated state, and that the continual arrival of solute molecules maintains this disorientation throughout the adsorption process. Unlike tension, the surface potential is influenced greatly by surface orientation as well as by surface concentration; the potential of the surface when adsorption is complete is therefore characteristic of the disorientated film. The general use of the well-known expression $\Delta V = 4\pi n\mu$ in surface potential studies implies that the decrease in the air-water contact potential (ΔV) is brought about by the solute only. However, it seems probable that the potential changes which follow adsorption are greater than can be attributed entirely to variations in the effective dipole moment μ ; it is therefore considered that water as well as solute molecules are disorientated by adsorption to fresh surfaces.

When the disturbing effect of adsorption is no longer operative, the film can become rearranged to