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Adsorption hemolysis.

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This report concerns the mechanism of action of certain hemolytic agents which have the property of lowering surface tension, examples of which are castor oil soap and saponin. These belong to a general class of substances which reduce the surface tension of their solutions according to the formula

$$y = \frac{\gamma_1 - \gamma_2}{ekx} + \gamma_2$$

y = surface tension at any concentration.

x = concentration of (hemolytic) solute.

γ_1 = surface tension of liquid solvent.

γ_2 = surface tension of saturated surface.

k = constant, the value of which depends upon the relative distribution of the solute in the body and surface of the solvent.

In these preliminary investigations erythrocytes from sheep have been placed in normal salt solutions containing known concentrations of the hemolytic substance, the time of hemolysis being carefully measured by means of a stop watch and a well defined end point. The surface tensions of solutions of hemolytic substance in the concentrations used were also measured. It was found, as is evident from the formula, that as the concentrations of hemolytic substance increased, the surface tension of the solution dropped from that of the pure solvent to the value always obtained for a very concentrated solution of the substance.

It has been observed that as the concentration of the hemolytic substance increased, the time of hemolysis decreased until it reached a fairly constant value. The significant observation is that the time of hemolysis and the surface tension reach a fairly constant value at the same concentration of hemolytic substance. As the surface tension is dependent upon surface concentration we may say that the time of hemolysis decreases as long as the surface can adsorb more hemolytic agent, and that when the surface is relatively saturated the time of hemolysis is no longer

decreased by a further increase in solution concentration. Thus, it would appear that hemolysis by these agents is concerned with surface concentration rather than with solution concentration. If we assume that the time of hemolysis is inversely proportional to the surface adsorption the time of hemolysis can be expressed by the equation

$$t = amx^{-1/n} \text{ when}$$

t = time of hemolysis,

x = concentration of hemolytic substance,

m = constant dependent upon the extent of adsorbing surface.

a and n are empirical constants.

This equation, which is the reciprocal of the adsorption formula, is graphically similar to the plotted values from experimental data which further indicates that hemolysis by surface tension reducing substances is a matter of surface concentration or an adsorption phenomenon.

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The fragility of erythrocytes treated with soap and saponin.

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It was shown by preliminary experiments with hemolytic agents which were active in reducing surface tension that in very low concentration the time of hemolysis was greatly prolonged. The work here reported has been done to determine if a surface concentration of these substances on red blood cells could be demonstrated when the surface concentration was insufficient to cause hemolysis.

Having determined what concentration of castor oil soap and saponin in 0.9 per cent. NaCl would not hemolyze erythrocytes in a number of hours, red blood cells were placed in solutions of the determined concentration and allowed to stand for varying lengths of time. These cells were then removed and resuspended in 0.9 per cent. NaCl and time-fragility tests performed. We have found that the adsorption of castor oil soap on the sur-