

cule occupies a position down the centre of an amylose helix, in the same way that iodine does¹.

A complete account of this work will be published elsewhere.

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Influence of X-Rays on the Activity of Carbonic Anhydrase in Erythrocytes and on their Haemolytic Resistance

SINCE sickle cell anaemia is possibly related to the activity of carbonic anhydrase¹, it is of interest to find some way of inhibiting the enzyme without altering the resistance of the cell membrane. Carbonic anhydrase is very stable² but its activity can easily be changed experimentally by adjusting the pH and temperature, or by adding sulphonamides or certain inorganic ions. All these factors, however, have a marked influence on haemolysis, whereas erythrocytes seem to be rather resistant to X-rays³. We therefore decided to examine whether carbonic anhydrase could be inhibited by a dose of X-rays which would have little or no effect on haemolysis.

For the determination of the enzyme activity we designed an improved Warburg technique⁴. This method is based upon measuring the rate of evolution of carbon dioxide when a bicarbonate solution is treated with a buffer and the enzyme. We calculated the unimolecular velocity constant of the reaction, which is also used as an index of activity by Mitchell, Pozzani and Fessenden⁵. The standard error of our measurements was 12 per cent and for the non-catalysed reaction 4 per cent. As a measure of the haemolytic resistance we used the haemolytic index: this is, in conventional experimental conditions⁶, the highest dilution of lysin which produces 100 per cent haemolysis within 2 hr. The standard error of our haemolytic indices was 2.5 per cent. Suspensions of 10⁷ cells/cm.³ in physiological saline solution were irradiated in vessels of 2 cm.³; dose 100,000 r., instrument Philips 'Compactix', 210 kV., h.v.l. = 4 mm. of aluminium, dose-rate 6,700/min.

In non-irradiated blood we found the following carbonic anhydrase activities, calculated per cubic centimetre of full blood: ox blood, 1.85 Mitchell units; human blood, 1.4; chicken blood, 1.2. Irradiations of four samples of ox blood, two samples of human blood and two samples of chicken blood had no effect on the activity. Repeating this dose after 24 hr. yielded no inhibition. Solutions of purified enzyme (Schering's 'Cartase', 0.1 per cent), treated in the same way, showed an inactivation of 20 and 50 per cent respectively. The fact that these high doses of X-rays do affect erythrocytes in other respects, was shown by their haemolytic index:

in our controls this was 13,300 with saponinum album (Merck); after irradiation, the index was increased by 10 per cent and after the second dose of 100,000 r. the increase was 17 per cent.

Our experiments confirm the great stability of carbonic anhydrase, and indicate that this stability is still greater inside the red blood cell: we found no decrease in activity of the carbonic anhydrase of erythrocytes after irradiation with 200,000 r. The haemolytic resistance of the cells was clearly diminished by this dose, so that it is possible that irradiation by X-rays will permit us to modify the structure of the cell-membrane without altering its carbonic anhydrase activity.

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A Method for distinguishing between α - and β -Glycosides by the use of Plant Haemagglutinins (Lectins)

SEVERAL plant seeds contain proteins that agglutinate red blood cells^{1,2} as do many antibodies. For these proteins the name 'lectins' has been suggested³. They can be as specific as animal antibodies in that they react only with erythrocytes of certain blood groups. The ABO blood group specific lectins—as well as the ABO specific antibodies—are inhibited by some simple sugars, mainly components of blood group polysaccharides^{1,2,4}. Several 'un-specific' lectins are also inhibited by simple sugars, but these sugars are not necessarily components of blood group polysaccharides^{1,2}. The explanation of this phenomenon is that the sugars, the structure of which most closely resembles the specific (sugar) group of the red cell receptor, attach themselves to the active site(s) of the lectin molecule, thus blocking them.

A monosaccharide as a rule retains its inhibiting power even when linked to other sugars through the hydroxyl group of the first (in ketoses, the second) carbon atom^{5,6}. The type of link, whether α or β , seems to be significant. In order to throw light on this problem, I made plant agglutinin inhibition tests by a method described in detail elsewhere⁶.

The following sugars were used in all experiments: D-glucose, D-glucosamine, N-acetylglucosamine, D-