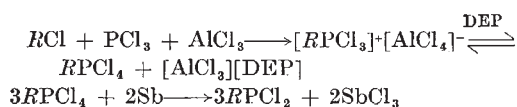


ized from similar solutions of the complex by reduction with finely powdered antimony. The reaction scheme is:



The reactions are exothermic and proceed smoothly in a single flask. Yields of $RPCl_2$ from 90 to 55 per cent have been obtained repeatedly for $R = CH_3$ to $R = C_6H_{11}$. Full details of the method and its application in high-activity synthesis of organo-phosphorus compounds labelled with radioactive phosphorus will be published elsewhere.

J. L. FERRON
B. J. PERRY
J. B. REESOR

Chemistry Section,
Suffield Experimental Station,
Defence Research Board of Canada,
Ralston, Alberta.

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Use of Spectrophotometric Evidence for Protein-Dye Binding

SHIFTS in the ultra-violet absorption spectra of aqueous organic solutions of some amino-azo dyes on the addition of buffered protein solutions have frequently been interpreted as evidence of complex formation between protein and dye¹⁻⁴. The effect of the buffer itself on the spectrum of the dye has apparently not always been checked, and the fact that this may influence the results is shown by the following experiments.

The powerful carcinogen dimethyl-*p*-aminoazobenzene and its biologically inactive isomer monoethyl-*p*-aminoazobenzene, purified chromatographically on an activated alumina column, were investigated in approximately 10⁻⁴ molar solutions in 20 per cent dioxane-water mixtures, between 350 and 550 m μ , using a Unicam SP500 spectrophotometer. Solutions were thermostatted at 25° C. for 24 hr. to attain equilibrium before measuring the spectra at the same temperature.

The spectra of the azo-dye solutions were determined under the following conditions: (1) With the addition of hydrochloric acid (analytical reagent) to give a range of pH 0-5. (2) At pH approximately 6 maintained by the use of acetate, phosphate and phthalate buffers, immediately after preparation of the solutions and at intervals of time up to 24 hr. thereafter. (3) In acetate buffered solution at pH 6, in the presence of 0.1 per cent gelatin, high grade, edible (Imperial Chemical Industries, Ltd.), and of 0.1 per cent bovine plasma albumin fraction V, and on the addition of 5 ml. of concentrated solution of bovine fibrinogen fraction I, pepsin and trypsin (Armour Biochemicals, Ltd.) per 95 ml. of dye solution. (4) In unbuffered solution in the presence of 0.1 per cent bovine plasma albumin. (5) Before and immediately after shaking for 1 hr. with solid bovine plasma albumin and centrifuging off the solid.

The results of these measurements may be summarized as follows: (1) Plots of extinction coefficient against wave-length for solutions of different pH show an isobestic point at 475 m μ for dimethyl-*p*-amino-

azobenzene and at 465 m μ for monoethyl-*p*-aminoazobenzene. The peak in the curves for the latter is shifted towards shorter wave-lengths compared with that for the former, which, according to Kubo², indicates a lesser ability to release electrons. (2) The spectra of the dye solutions with acetate buffer were unchanged after 24 hr., and those of the dye solutions alone were unchanged after several days, showing that dioxane had not affected the spectra. In the case of the phosphate and phthalate buffers there was a marked and progressive shift with time, up to 24 hr. at least, of the peaks of the curves for both dyes, the changes being most pronounced for solutions of dimethyl-*p*-aminoazobenzene. The shifts are similar to those expected from complex formation, but the nature of any such complex and its slow formation in the present systems seem difficult to explain. The phosphate buffer in addition caused some flattening and lowering of the peaks. This normally indicates the removal of the absorbing compound, although it is impossible to state from the present data the way in which this is occurring here. (3-5) In no case was there any change in the dye spectrum on the addition of protein whether to acetate buffered solutions or to the unbuffered solutions.

The pH values of all solutions remained unaltered with time, and observed spectral shifts cannot, therefore, be ascribed to changes in pH.

In view of the observed effects of certain buffers on the spectra of the azo-dyes used here, a re-examination of some of the earlier findings seems desirable, together with a more extensive investigation of the effect of these and other buffer solutions on the spectra of such systems.

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C. M. FRENCH
R. PRITCHARD

Queen Mary College,
Mile End Road,
London, E.1.

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BIOCHEMISTRY

Enzymatic Degradation of Deoxyribonucleic Acid into Sub-units

THE generally accepted value for the molecular weight of deoxyribonucleic acid is 6-8 \times 10⁶ (refs. 1 and 2). In the course of a study of methods of preparation involving extraction with solutions of low ionic strength, it has been found that, under a given set of experimental conditions, preparations from chicken erythrocytes could be obtained which showed reproducibly lower molecular weights (Champagne, M., unpublished work). This finding is reminiscent of some results scattered in the literature³ and suggestive of some enzymatic activity involving depolymerization of deoxyribonucleic acid. An investigation was therefore undertaken of the action on deoxyribonucleic acid of preparations from chicken erythrocytes.

Two preparations of deoxyribonucleic acid were mainly used in this work: B1s from calf thymus