inert medium most of the amino-azo-dye-bound protein is basic, just as it is in fresh soluble cytoplasm. It has been found that the heat treatment used partially denatures the purified basic protein, and this may account for the abnormal behaviour of amino-azo-dye-bound protein in the heat-treated extracts observed by Sugimoto & Terayama (1970).

The physicochemical properties of the purified basic protein have now received further study. Its molecular weight by meniscus-depletion approach-to-equilibrium sedimentation in aqueous buffer is 45900 and in 6M-guanidine hydrochloride in the presence of thiol-blocking agents is 23000. The same value of 23000 is obtained by polyacrylamide-gel electrophoresis in sodium dodecyl sulphate-8M-urea. The protein is thus composed of similar subunits and these are not held together by disulphide links (there are four cysteine residues/molecule of protein).

The amino-azo-dye derivative is bound to a cysteinyl residue (Ketterer & Christodoulides, 1969), but in two forms. Mild denaturing agents release one form as an amino-azo-dye-bound peptide (Glu, Cys, Gly), but most of the dye remains covalently bound to the protein.

The electrophoretic properties of the protein vary from batch to batch. This is probably due to its affinity for charged compounds. It has been shown to bind both bromosulphophthalein and bilirubin. Litwack & Morey (1970) have drawn parallels between it and a binding protein for charged cortisol metabolites. This may be related to its physiological role: it has also been suggested that it may be involved in bilirubin excretion (I. M. Arias & J. Levi, personal communication).

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Ketterer, B. & Christodoulides, L. (1969). *Chem.-Biol. Interactions*, **1**, 173.

Ketterer, B., Ross-Mansell, P. & Whitehead, J. K. (1967). *Biochem. J.* **103**, 316.

Litwack, G. & Morey, K. S. (1970). *Biochem. biophys. Res. Commun.* **38**, 1141.

Sorof, S., Young, E. M., McCue, M. M. & Fetterman, P. L. (1963). *Cancer Res.* **23**, 864.

Sugimoto, T. & Terayama, H. (1970). *Biochim. biophys. Acta*, **214**, 533.

Rotational Mobility of a Fluorescent Rhodopsin Derivative in the Rod Outer-Segment Membrane

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In the simultaneous presence of light and sodium borohydride, rhodopsin is converted into a fluorescent derivative known as *N*-retinyl-opsin (Bownds, 1967). From 'decay-of-fluorescence-anisotropy' (Stryer, 1968; Tao, 1969) measurements the rotational mobilities were obtained for the following materials: (1) *N*-retinyl-opsin solubilized by digitonin; (2) *N*-retinyl-opsin solubilized by cetyltrimethylammonium bromide; (3) membrane-bound *N*-retinyl-opsin. For cases (1) and (2) the measured rotational relaxation times (148 ns and 52 ns respectively) were consistent with the sizes of the corresponding rhodopsin-detergent micelles. For case (3) no anisotropy decay was observed within the limits of detection. A lower limit of 300 ns can be set for the rotational relaxation time of membrane-bound *N*-retinyl-opsin. Other reports suggest that membrane rhodopsin possesses both translational (Blasie & Worthington, 1969) and rotational (Hagins & Jennings, 1959) mobility on a millisecond time-scale. This work indicates that rhodopsin in the rod outer-segment membrane is rotationally immobile on a nanosecond time-scale.

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Blasie, J. K. & Worthington, C. R. (1969). *J. molec. Biol.* **39**, 417.

Bownds, D. (1967). *Nature, Lond.*, **216**, 1178.

Hagins, W. A. & Jennings, W. H. (1959). *Disc. Faraday Soc.* **27**, 180.

Stryer, L. (1968). *Science, N.Y.*, **162**, 526.

Tao, T. (1969). *Biopolymers*, **8**, 609.

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