Physicochemical Studies on Chloroplast Thylakoid Protein from *Phaseolus vulgaris*

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'Structural' or 'organizer' protein forms a large proportion of the total protein of the chloroplast and is believed to form the backbone of the thylakoid membranes (Criddle, 1966). The protein can be solubilized with the aid of detergents such as sodium dodecyl sulphate, and this has enabled sedimentation-velocity studies to be carried out (Biggins & Park, 1965; Criddle, 1966; Mani & Zalik, 1970). Such studies have yielded conflicting and sometimes confusing results (see Keylock, Kirk & Rogers, 1971).

The present communication reports studies on chloroplast thylakoid protein isolated from *Phaseolus vulgaris* (French-bean) seedlings by a method similar to that used for isolation of thylakoid protein from *Euglena gracilis* (Keylock *et al.* 1971). The results we report, based on some 50 sedimentation-velocity experiments, have yielded results clarifying and extending the observations previously published.

Solubilized in the presence of high concentrations of sodium dodecyl sulphate the protein migrated as a single symmetrical boundary at all protein concentrations. At protein concentrations above 3.5 mg/ml the sedimentation coefficient was 1.8–2.0S; however, at lower protein concentrations the sedimentation coefficient progressively decreased to an extrapolated $s_{20,w}$ value of approx. 0.7S at infinite dilution. No boundary characteristic of sodium dodecyl sulphate micelles alone appeared in addition to the protein boundary, suggesting that the decreasing $s_{20,w}$ values may reflect protein-sodium dodecyl sulphate complexes of changing ratio rather than dissociation of the 2S species to smaller units.

Solubilization in lower sodium dodecyl sulphate concentrations enabled identification of species of thylakoid protein sedimenting at about 5.2S. Below about 2 mg/ml the 5.2S species dissociated to smaller subunits, the sedimentation coefficient of the single symmetrical boundary observed progressively decreasing to an $s_{20,w}$ value of approx. 3.6S at infinite dilution. Treatment with 6M-urea caused the 5.2S aggregates to dissociate to a species sedimenting at 3.6S over a wide range of protein

concentrations. There is evidence to suggest that under some conditions this dissociation is reversible.

The amino acid composition of the protein was in good agreement with that obtained for *Euglena* chloroplast structural protein, with some minor differences that may be especially significant in view of the different aggregation behaviour of the two proteins.

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Inhibition of the Hill Reaction and Photophosphorylation by 1,1,1-Trichloro-2,2-bis-(p-chlorophenyl)ethane (DDT)

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It is known (Hayes, 1959) that certain barley varieties are susceptible to spraying with DDT [1,1,1-trichloro-2,2-bis-(p-chlorophenyl)ethane] and develop a severe chlorosis some 4–10 days after treatment. The reaction to DDT is controlled by a single major gene on chromosome 7 and susceptibility is the dominant character (Hayes, 1959; Wiebe & Hayes, 1960).

Previous studies (Lawler & Rogers, 1967, 1968, 1969) have shown that DDT affects the photosynthesis of susceptible barley, as evidenced by a decrease in rate of O_2 evolution of DDT-treated leaves and diminished Hill activity of the chloroplasts isolated from treated plants. In chloroplasts isolated by a procedure adapted from Kleese (1966) 2,6-dichlorophenol-indophenol photoreduction and phenazine methosulphate-catalysed cyclic photophosphorylation were inhibited, whereas there appeared to be little effect on ferricyanide photoreduction or concomitant non-cyclic photophosphorylation.

Recently we have studied the effect of DDT on these biochemical functions in some 30 varieties of barley from widely differing parts of the world [e.g., Fimbul (Finland), Sinop 248 (Turkey), Lyallapur (India), Tang May (China), Mosdoksky H. V.

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