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STRUCTURE AND REGULATION OF THE *fds* OPERON ENCODING THE SOLUBLE FORMATE DEHYDROGENASE IN *RALSTONIA EUTROPHA*

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The soluble formate dehydrogenase(S-FDH) is encoded by the *fdsGBACD* operon composed of four structural genes(*fdsA, fdsB, fdsG* and *fdsD*) and one nonstructural gene (*fdsC*). An additional gene, designated *fdsR*, is located 150 bp upstream of *fdsG* and transcribed divergently to the *fds* operon. Analyses of the sequence homologies indicated that the α-subunit (FdsA) of S-FDH is the catalytic subunit performing the formate oxidation. Expression of the *fds* operon is regulated by the FdsR protein which functions either as activator in the presence of formate to induce transcription or as repressor in the absence of formate to repress transcription. FdsR also represses its own gene, indicating that it is subject to negative autoregulation. Gel retardation analyses showed that formate acts as effector of FdsR, and two operator regions were identified in the 150-bp intergenic region between *fdsR* and *fdsG*.

SIII-2-1

STUDIES ON PHYCOBILIN BIOSYNTHESIS: HEME OXYGENASE FROM CYANIDIUM CALDARIUM AND PORPHOBILINOGEN SYNTHASE FROM CHLOROBIUM VIBRIOFORME

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Algal heme oxygenase from *Cyanidium caldarium*, which catalyzes the first committed step of phycobilin biosynthesis, was partially purified and several mechanistic questions were investigated. The first mechanistic question deals with the role of NADPH in the heme oxygenase and phycobilin forming reactions. Using a system derived from partially purified photosystem-I of spinach leaves, it was found that the role of NADPH in the heme oxygenase reaction is to reduce ferredoxin via ferredoxin-$NADP^+$ reductase, and that reduced ferredoxin is sufficient to provide the electrons needed to drive the heme oxygenase reaction and phycobilin forming reactions. Based on this fact, heme oxygenase was partially purified using ferredoxin-Sepharose as an affinity column. The second question deals with the role of ascorbate, which is required as a second reductant in the reaction. Using desferrioxamine, it was found that the product of the algal heme oxygenase reaction is Fe(III)-biliverdin IX_α, and that ascorbate is not involved in the reduction of Fe(III)-biliverdin IX_α to release free biliverdin IX_α. Ascorbate also did not participate in the reduction of Fe(III)-protoheme to Fe(II)-protoheme. The gene (*hemB*) for porphobilinogen synthase which converts δ-aminolevulinic acid to porphobilinogen was cloned and sequenced in a strictly anaerobic green phototrophic bacterium *Chlorobium vibrioforme* and some catalytic properties of the enzyme were investigated. Also the positional relationship of *hemB* gene to and expressional independence of *hemB* gene from the *hemACD* genes were described.