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**Identification of protein W, the elusive sixth subunit of the *Rhodopseudomonas palustris* reaction center-light harvesting 1 core complex**

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**Abstract** The X-ray crystal structure of the *Rhodopseudomonas (Rps.) palustris* reaction center-light harvesting 1 (RC-LH1) core complex revealed the presence of a sixth protein component, variably referred to in the literature as helix W, subunit W or protein W. The position of this protein prevents closure of the LH1 ring, possibly to allow diffusion of ubiquinone/ubiquinol between the RC and the cytochrome *bc*<sub>1</sub> complex in analogous fashion to the well-studied PufX protein from *Rhodobacter sphaeroides*. The identity and function of helix W have remained unknown for over 13 years; here we use a combination of biochemistry, mass spectrometry, molecular genetics and electron microscopy to identify this protein as RPA4402 in *Rps. palustris* CGA009. Protein W shares key conserved sequence features with PufX homologs, and although a deletion mutant was able to grow under photosynthetic conditions with no discernible phenotype, we show that a tagged version of protein W pulls down the RC-LH1 complex. Protein W is not encoded in the photosynthesis gene cluster and our data indicate that only approximately 10% of wild-type *Rps. palustris* core complexes contain this non-essential subunit; functional and evolutionary consequences of this observation are discussed. The ability to purify uniform RC-LH1 and RC-LH1-protein

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