Charcaterization of Phus as a Heme Chaperone to Heme Oxygenase in Pseudomonas Aeruginosa

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The uptake of heme as an iron source is a common mechanism by which pathogenic bacteria obtain the iron necessary for their survival. Bacterial pathogens utilize heme directly from the host's hemeproteins or via a hemophore that sequesters and transports heme inside the cell, where it is degraded by heme oxygenases (HO) to give free iron, carbon monoxide and biliverdin. The mechanism of heme degradation is fairly well understood. However, very little is known about the acquisition of heme by HO. Herein, we report the characterization of Phus, a cytoplasmic heme-binding protein in *Pseudomonas aeruginosa*, as a heme chaperone to *pa*-HO. Spectroscopic as well as chromatographic analyses show a complete transfer of heme from Phus to *pa*-HO, without reversibility or back transfer. However, mutation of the proximal ligand H26A in *pa*-HO, which has similar heme affinity to the native *pa*-HO, gave only partial heme transfer from Phus and the process was reversible. Also, the lack of formation and/or detection of any biliverdin isomers from the oxidative heme degradation of Phus suggest that it does not have heme oxygenase activity. Although, coupled oxidation carried out with Phus, and *pa*-HO under similar experimental conditions gave primarily δ-biliverdin, as expected. These results suggest that Phus is not a HO, and it serve primarily as a specific chaperone to *pa*-HO.

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