

Heme environment in HmuY, the heme-binding protein of *Porphyromonas gingivalis*.

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Porphyromonas gingivalis, a Gram-negative anaerobic bacterium implicated in the development and progression of chronic periodontitis, acquires heme for growth by a novel mechanism composed of HmuY and HmuR proteins. The aim of this study was to characterize the nature of heme binding to HmuY. The protein was expressed, purified and detailed investigations using UV-vis absorption, CD, MCD, and (1)H NMR spectroscopy were carried out. Ferric heme bound to HmuY may be reduced by sodium dithionite and re-oxidized by potassium ferricyanide. Heme complexed to HmuY, with a midpoint potential of 136mV, is in a low-spin Fe(III) hexa-coordinate environment. Analysis of heme binding to several single and double HmuY mutants with the methionine, histidine, cysteine, or tyrosine residues replaced by an alanine residue identified histidines 134 and 166 as potential heme ligands.

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