# Magnetic circular dichroism study of the heme scavenging Isd proteins of *Staphylococcus aureus*

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We describe a new chemistry for the Gram-positive human pathogen *Staphylococcus aureus*. Iron, which is an essential nutrient, is limiting to extracellular pathogens and, therefore, to be successful in surviving the host environment, the pathogen must adopt specialized mechanisms to acquire this metal from the host. Several different mechanisms are used by bacteria to acquire limiting quantities of extracellular iron from the host, including the production of low-molecular-weight iron-binding molecules termed siderophores, but as well non-siderophore mediated methods, including the direct binding of host iron sources such as transferrin, hemoglobin, or heme at the bacterial cell surface.

We report the characterization of a number of components of a newly discovered heme scavenging system involving iron-regulated surface determinant (Isd) proteins. These proteins are present within the cell envelope when expressed heterologously in *Escherichia coli*, efficiently scavenging intracellular heme and resulting in *de novo* heme synthesis in excess of 100-fold above background. Magnetic circular dichroism analyses showed that the heme binding properties of each of the Isd proteins differ significantly. In addition to binding heme, IsdC bound almost exclusively free-base protoporphyrin IX (PPIX), whereas the IsdE protein was associated with low spin Fe(III) and Fe(II) heme. Absorption, CD, emission and MCD spectroscopic data are used to characterise these proteins. These properties provide important insight into the possible mechanisms of iron scavenging from bound heme by Isd proteins.