

## BINDING AND OXIDATION OF HEME IN A LIPOCALIN SCAFFOLD

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The bilin-binding proteins (BBP) from lepidopteran insects are members of the lipocalin family of proteins and play special role in pigmentation through the binding of biliverdin IX $\gamma$ . While biosynthesis of biliverdin IX $\alpha$  from heme in mammalian cells by the action of heme oxygenase has been well characterized, the origin of biliverdin IX $\gamma$  ligand in lepidopteran BBPs remains unclear. Here we show that a BBP from the toxic caterpillar *Lonomia obliqua* (BBP<sub>Lo</sub>) binds heme and supports its conversion to biliverdin IX $\gamma$  under conditions used to support heme oxygenase catalysis or coupled oxidation. Using ascorbate or a NADPH<sup>+</sup>-ferredoxin reductase-ferredoxin system as a source of reducing equivalents, spectral changes are seen that suggest an initial reduction of heme to the Fe(II) state and formation of a oxyferrous complex. The complex then disappears and biliverdin is formed. Reversed phase HPLC and mass spectrometry indicate that the major biliverdin product is the  $\gamma$  isomer. Our findings suggest that BBP<sub>Lo</sub> may bind heme *in vivo* and facilitate its conversion to biliverdin IX $\gamma$  *in situ*.