

Tomato hemoglobin – spectroscopic characterization  
of the heme environment

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Recently, a non-symbiotic hemoglobin, SolyGLB1, was identified in tomato. Combined UV/Vis, resonance-Raman and EPR measurements reveal that a bis-histidine ligation (F8His-Fe<sup>2+/3+</sup>-E7His) can occur for both the deoxy ferrous and ferric state of the protein. For the ferric form, this hexacoordination of the heme iron is dominating, whereby a considerable amount (up to almost 50 %) of the deoxy ferrous SolyGLB1 is in a pentacoordinated form. Pulsed-EPR techniques were used to study the low-spin ferric form of the protein. These experiments reveal that, in ferric SolyGLB1, one of the histidine planes is rotated 20°(±10°) away from a N<sub>heme</sub>-Fe-N<sub>heme</sub> axis. Based on the observed *g* values and the counterrotation principle, two sets of possible positions for the second histidine could be derived. From the HYSCORE measurements, the hyperfine and nuclear-quadrupole couplings of the heme and histidine nitrogens are identified and compared with known EPR/ENDOR data of vertebrate Hbs and cytochromes. In combination with the resonance Raman and the kinetics studies the EPR data reveal structural information that can be linked to possible functions of the tomato hemoglobin.