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What we do know about rice (*Oryza sativa*) hemoglobins

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Nonsymbiotic hemoglobins (nsHb) are proteins widely distributed in land plants, however their function in plant organs is still not known. Rice nsHbs have been characterized in detail and represent a model to study plant nsHbs. Rice recombinant Hb1 (rHb1) has a very high affinity for O₂ (1,800 μM⁻¹ s⁻¹), because the dissociation constant is extremely low (k_{off} = 0.038 s⁻¹). X-ray analysis and site-directed mutagenesis showed that the low dissociation constant of rHb1 is partially due to the stabilization of bound O₂ by distal His. This characteristic suggests that the function of rHb1 is other than transport of O₂. A family of *nshb* genes exists in rice, which codes for Hb1, Hb2, Hb3 and Hb4 proteins. Potential promoters have been detected upstream the rice *nshb* genes, and RT-PCR analysis showed that rice *hb1* and *hb2* are differentially expressed in embryonic and vegetative organs. These observations suggest that a number of nsHbs exist in rice plants. In normal rice, western blot analysis revealed that nsHbs are synthesized in embryonic and vegetative organs, and confocal microscopy showed that nsHbs are located at specific tissues, including seeds aleurone and scutellum, roots cap and leaf schlerenchyma. In stressed rice, levels of nsHbs increased in etiolated leaves and flooded roots, but not under oxidative, nitrosative, and hormonal stresses. Thus, the results from the above work suggest that rice nsHbs play more than one function in different tissues, and that their function varies with the metabolism of the plant cell.

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