

# **EPR Spectroscopy Studies on the Structural Transition of Nitrosyl Hemoglobin in the Arterial-Venous Cycle of DEANO-Treated Rats**

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Hemoglobin exists in two alternative quaternary structures: low-affinity tense state (T) and high-affinity relaxed state (R). The three-line hyperfine structure observed in the EPR spectrum of  $\alpha$ -NO hemoglobin in the T state is associated with pentacoordinate heme. In the R state of HbNO hexacoordinate heme shows no three-line hyperfine structure. In the present in vivo studies a dynamic cycle is observed in which  $\alpha$ -nitrosylated hemoglobin is in the R state in arterial blood of rats treated with 25 $\mu$ M of 2-(N,N-diethylamino)-diazene-2-oxide (DEANO) and converts to the T state during arterial-venous transit. A detailed analysis shows that different EPR spectra recorded for  $\alpha$ -nitrosyl hemoglobin in arterial and venous blood at 77K originate only from a different ratio between 5- and 6-coordinated heme without any change in the concentration of HbNO. In venous blood the five- $\leftrightarrow$ six-coordination equilibrium of the  $\alpha$ -nitrosyl heme is shifted in favor of the 5-coordinate state (58% venous versus 20% arterial).