Spin trapping studies of the reaction mechanism involved in enhanced peroxidase function of Cu,Zn-SOD

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In presence of higher concentrations of hydrogen peroxide, one of the products during the dismutation of superoxide anion by SOD, copper-zinc superoxide dismutase (Cu,Zn-SOD or SOD1) acts as a peroxidase. In the presence of bicarbonate anion the peroxidase function of Cu,Zn-SOD is enhanced to a great extent. We have hypothesized that bicarbonate bound to Arg-141 supports the incoming neutral H_2O_2 molecule allowing it to anchor at the active site of copper and enabling its redox cleavage. The peroxidase function was studied using the spin trap, 5,5-Dimethyl-1-pyrroline N-oxide (DMPO). During the peroxidase function DMPO is oxidized into EPR active DMPO-OH adduct by the Cu^{2+} bound hydroxyl in SOD1. To support our hypothesis we carried out additional experiments and observed that the iso-structural and iso-electronic anion, carbamate, enhances the peroxidase function of Cu,Zn-SOD through its binding with Arg-141. The magnitude of the enhancement of peroxidase function of Cu,Zn-SOD due to carbamate is higher than that of the bicarbonate anion at the initial stage. Enhanced peroxidase function by the carbamate anion may be due to the increased hydrogen bonding between arginine bound carbamate and H_2O_2. In contrast, the iso-structural and iso-electronic acetate molecule doesn’t favor the enhanced peroxidase function. The present study demonstrates that isostructure, negative charge and size of the anions are vital for the enhanced peroxidase function of Cu,Zn-SOD.