

Oligomerization of the proteolytic enzyme THIMET-oligopeptidase following the oxidation by high valence states of Myoglobin and hydroxyl radical formed during the reaction of the heme protein with H<sub>2</sub>O<sub>2</sub>

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A probable influence of THIMET-oligopeptidase (TOP) in the reduced glutathione/oxidized glutathione ratio(GSH/GSSG) was investigated in a model system formed by myoglobin (Mb) and hydrogen peroxide. In this study, Mb high valence states (oxoferryl p cation and oxoferryl) were formed by assaying the heme protein with hydrogen peroxide and the catalytic cycle accompanied in the presence and in the absence of TOP. In the absence of TOP, Mb is converted to the oxoferryl form (Compound II) and exhibits progressive Soret bleaching. The presence of TOP, led to a more intense without significant recycling of Mb to the native form. The addition of the spin trapping DMPO in the system led to partial recycling of Mb to the native form. The corresponding Western-blot analysis of TOP revealed that significant dimerization in the presence of Mb and hydrogen peroxide that was impaired by DMPO, probably by trapping hydroxyl and thiyl radicals. The corresponding SH dosage revealed oxidation of 6, and 12 cysteines (total 15), in the presence of hydrogen peroxide, in the presence of hydrogen peroxide and Mb and in the latter plus DMPO, respectively. Thus, the oligomerization of TOP that controls the enzyme digestion and generation of signaling peptides can occur due to SH oxidation by free radicals and by peroxidase high valence states and could be linked to cell signaling.

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