Research Highlight #151

Mitochondria-targeted cyclic nitrone spin traps: Detection of mitochondrial superoxide

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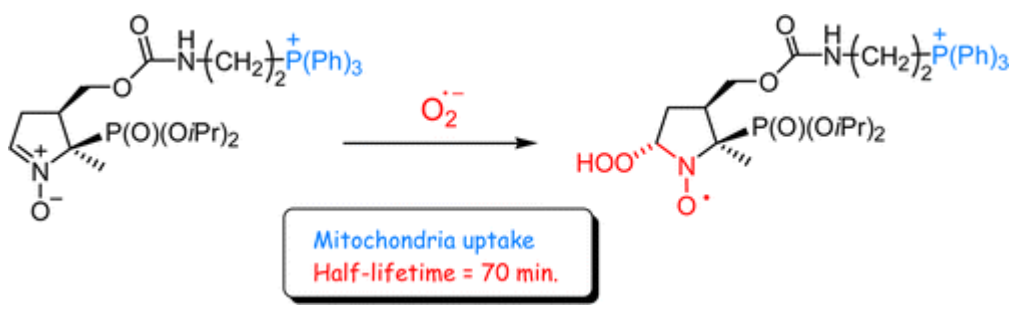
Introduction and Methods: One of the major intracellular sources of superoxide is mitochondria. Abnormal generation of mitochondrial superoxide and hydrogen peroxide is linked to the onset and progression of several diseases, including cardiovascular and neurodegenerative diseases. Progress in this field is hampered because of the lack of specific probes and analytical techniques to directly detect mitochondrial superoxide. Studies using the mitochondria-targeted fluorescent probe (e.g., Mito-SOX) are flawed due to nonspecific oxidation of this probe. Thus, there is a critical unmet need for developing alternate probes and assays for detecting mitochondrial superoxide.

Results: Nitrone spin trapping technique has long been used to detect superoxide in biological systems. In this project, we synthesized novel cyclic nitrone spin traps (Mito-DEPMPO and Mito-DIPPMPO) by conjugating a long-chain triphenylphosphonium (TPP^+) moiety to DEPMPO or DIPPMPO spin trap. The uptake of Mito-DEPMPO into mitochondria is facilitated by the negative mitochondrial membrane potential. The superoxide spin adduct formed from trapping of $O_2^{\bullet -}$ by Mito-DIPPMPO was relatively persistent (t=73 min).



EPR parameters:
Diastereoisomer I (88%)
a_P = 53.4 G
a_N = 12.9 G
$a_{H\beta}$ = 12.1 G
Diastereoisomer II (12%)
a_P = 4.81 G
a_N = 12.8 G
$a_{H\beta}$ = 7.4 G

Implications: Studies indicate that cyclic nitrone trap linked to TPP+ via a long carbon chain ($Mito_{10}$-DIPPMPO) exhibits the highest mitochondrial uptake.

Discussion: We conclude that Mito-DEPMPO and Mito-DIPPMPO are suitable candidate spin traps for detecting mitochondrial superoxide.

Reference:

Hardy M, Poulhés F, Rizzato E, Rockenbauer A, Banaszak K, Karoui H, Lopez M, Zielonka J, Vasquez-Vivar J, Sethumadhavan S, Kalyanaraman B, Tordo P, Ouari O. Mitochondria-targeted spin traps: synthesis, superoxide spin trapping, and mitochondrial uptake. *Chem. Res. Toxicol*. 27:1155–65, 2014.