

#### **84. CHOLINESTERASES, OXIMES, AND ORGANOPHOSPHORUS HYDROLASES IN TANDEM CAN HYDROLYZE ORGANOPHOSPHATES**

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Reactivation of organophosphate (OP)-inhibited acetylcholinesterase (AChE) is a key objective in the treatment of OP poisoning. Reactivation is accelerated by ligands such as edrophonium and decamethonium which may be due to the protection of regenerated enzyme from inhibition by the putative product, the phosphoryloxime. Using recombinant wild-type and D74N mouse AChEs, each inhibited with MEPQ (7-(methylethoxyphosphinyloxy)-1-methylquinolinium iodide) and DEPQ (7-(O, O-diethyl-phosphinyloxy)-1-methyl-quinolinium methylsulfate), a relationship was established between ligand-induced acceleration of reactivation and phosphoryloxime inhibition of the reactivated enzyme. Results showed that phosphoryloxime inhibition occurs during reactivation by pyridinium oximes LüH6 and TMB4, and not with HI-6. This is due to the extreme lability of the phosphoryloximes formed by HI-6. Phosphoryloximes formed during reactivation of ethoxy methylphosphonyl-AChE conjugate by LüH6 and TMB4 were isolated and their structures confirmed by <sup>1</sup>H NMR. The purified phosphoryloximes formed with LüH6 and TMB4 are 10- to 22-fold more potent than MEPQ as inhibitors of AChE and stable for several hours at pH 7.2 in HEPES buffer. However, phosphoryloximes formed during the reactivation of diethylphosphoryl-AChE conjugate were not sufficiently stable to be detected by <sup>31</sup>P NMR. Reactivation of both ethoxy methylphosphonyl- and diethylphosphoryl-AChE by LüH6 and TMB4 was accelerated in the presence of rabbit serum paraoxonase, suggesting that organophosphorus hydrolase (OPH) can hydrolyze phosphoryloximes formed during the reactivation reaction. These results suggest that ChEs, oximes, and OPHs can work in tandem to hydrolyze or inactivate OPs *in vivo* and *in vitro*.

**Keywords:** phosphoryloxime, cholinesterase, organophosphorus hydrolase, oxime, reactivation