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An evaluation of therapeutic and reactivating effects of newly developed oximes (K156, K203) and commonly used oximes (obidoxime, trimedoxime, HI-6) in tabun-poisoned rats and mice

Jiri Kassa*, Jana Karasova, Kamil Musilek, Kamil Kuca

Department of Toxicology, Faculty of Military Health Sciences, Trebesska 1575, 500 01 Hradec Kralove, Czech Republic

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Abstract

The potency of newly developed monoxime bispyridinium compounds (K156, K203) in reactivating tabun-inhibited acetylcholinesterase and reducing tabun-induced lethal toxic effects was compared with commonly used oximes (obidoxime, trimedoxime, the oxime HI-6) using *in vivo* methods. Studies determining percentage of reactivation of tabun-inhibited blood and tissue acetylcholinesterase in poisoned rats showed that the reactivating efficacy of newly developed oxime K203 is comparable with obidoxime and trimedoxime in blood and higher than the reactivating potency of trimedoxime and obidoxime in diaphragm and brain, where the difference in reactivating efficacy of obidoxime, trimedoxime and K203 is significant. On the other hand, the potency of newly developed K156 to reactivate tabun-inhibited acetylcholinesterase is comparable with obidoxime or trimedoxime in diaphragm and brain. It is significantly lower than the reactivating efficacy of trimedoxime and obidoxime in blood. Moreover, both newly developed oximes were found to be relatively efficacious in the reduction of lethal toxic effects in tabun-poisoned mice. Especially, the oxime K203 is able to decrease the acute toxicity of tabun nearly two times. The therapeutic efficacy of K156 and K203 corresponds to their potency to reactivate tabun-inhibited acetylcholinesterase, especially in diaphragm and brain. In contrast to obidoxime and trimedoxime, the oxime HI-6 is not effective in reactivation of tabun-inhibited acetycholinesterase and in reducing tabun lethality. While the oxime K156 does not improve the reactivating and therapeutic effectiveness of currently available obidoxime and trimedoxime, the newly developed oxime K203 is markedly more effective in reactivation of tabun-inhibited acetylcholinesterase in rats, especially in brain, and in reducing lethal toxic effects of tabun in mice and, therefore, it is suitable for the replacement of commonly used oximes for the antidotal treatment of acute tabun poisoning.

Keywords: Tabun; Acetylcholinesterase; K156; K203; Obidoxime; Trimedoxime; HI-6; Rats; Mice

1. Introduction

Tabun (GA; *O*-ethyl-*N,N*-dimethylphosphoramidocyanidate) belongs to a highly toxic group of organophosphorus compounds misused as chemical warfare agents for military as well as for terroristic purposes. Tabun differs from other highly toxic organophosphates in its chemical structure and by the fact that commonly used antidotes are not able to sufficiently reactivate tabun-inhibited acetylcholinesterase (AChE, EC 3.1.1.7). Deleterious effects of tabun are difficult to antagonize because the tabun-AChE conjugate is extraordinarily resistant towards most

* Corresponding author. Tel.: +420 973 251500; fax: +420 495518094.
E-mail address: kassa@pmfhk.cz (J. Kassa).

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reactivators (Cabal and Bajgar, 1999; Ekström et al., 2006).

A current standard treatment for poisoning with tabun usually consists of combined administration of an anticholinergic drug (preferably atropine) and an oxime (preferably pralidoxime or obidoxime). The anticholinergic drug blocks the effects of overstimulation by acetylcholine accumulated at the muscarinic receptor sites, while oxime (compound with nucleophilic oximate anion) repairs biochemical lesions by dephosphorylating tabun-inhibited AChE and restoring its activity (Kassa, 2002; Marrs, 1993). While the anticholinergic drugs such as atropine are able to counteract the effects of tabun at the muscarinic cholinergic receptors (Bajgar, 2004), commonly used reactivators of phosphorylated AChE based on monopyridinium (e.g. pralidoxime) and bispyridinium oximes (e.g. obidoxime, trimedoxime) are not able to counteract the acute toxic effects of tabun because of their low reactivating efficacy (Koplovitz and Stewart, 1994). In addition, the reactivating efficacy of the oxime HI-6, which is relatively efficacious against adverse effects of soman (Kassa, 1995), is not efficient for tabun-inhibited AChE (Puu et al., 1986; Worek et al., 1998). Therefore, the replacement of commonly used oximes (pralidoxime, obidoxime) as well as H-oximes (the oxime HI-6) with a more effective oxime has been a long-standing goal for the treatment of tabun poisoning. For this reason, the new bispyridinium oximes: K203: (*E*)-1-(4-carbamoylpyridinium)-4-(4-hydroxyiminomethyl-pyridinium)-but-2-ene dibromide and K156: 1-(4-hydroxyiminomethylpyridinium)-3-pyridiniumpropane dibromide (Fig. 1) were synthesized at our department (Kuca et al., 2007; Musilek et al., 2007a) to improve the efficacy of the antidotal treatment in reactivating tabun-inhibited AChE and reducing tabun-induced lethal toxicity.

The main aim of this study was to compare the reactivating and therapeutic efficacy of the newly developed oximes (K156, K203) with the currently available oximes (obidoxime, trimedoxime, the oxime HI-6) against tabun using *in vivo* methods.

2. Material and methods

2.1. Animals

Male albino Wistar rats weighing 180–200 g and NMRI male mice weighing between 20 and 24 g were purchased from Konarovice, Czech Republic. They were kept in an air-conditioned room with the light from 07:00 to 19:00 h and were allowed access to standard food and tap water *ad libitum*. The rats were divided into groups of eight animals. Handling of the experimental animals was done under the supervision of the Ethics Committee of the Faculty of Military Health Sciences, Czech Republic.

2.2. Chemicals

Tabun was obtained from the Technical Institute in Brno (Czech Republic) and was 95% pure. All oximes (obidoxime, trimedoxime, the oxime HI-6, K156, K203) were synthesized at the Department of Toxicology of the Faculty of Military Health Sciences (Czech Republic) and were more than 98% pure. Their purities were analyzed using a HPLC technique. All other drugs and chemicals of analytical grade were obtained commercially and used without further purification. All substances were administered intramuscularly (i.m.) at a volume of 1 ml/kg body weight (b.w.).

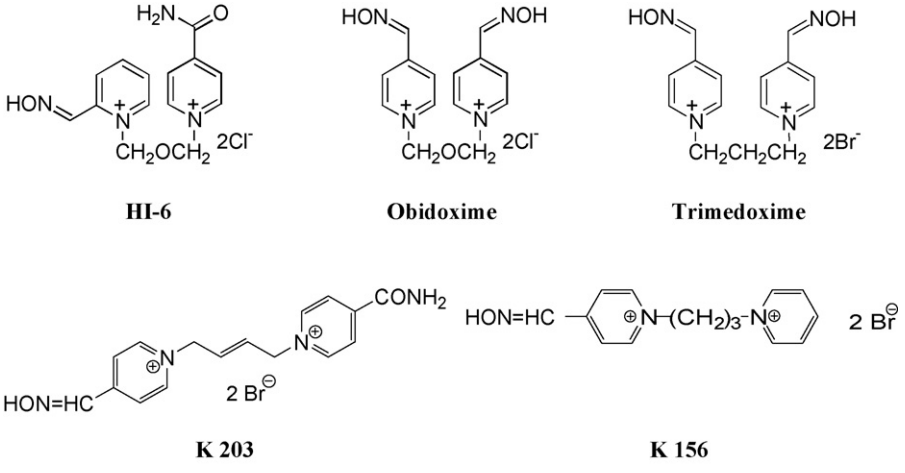

Fig. 1. Chemical structure of oximes.