A Toxidromic Approach for Chemical Medical Countermeasure Development

Translational Micro-CT Imaging as a Monitoring Tool to Detect Persistent Neurological Damage in Survivors of OP Intoxication

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Organophosphates (OPs) are highly lethal compounds that are used as both pesticides and chemical weapons. OPs induce excitotoxicity by inhibiting acetylcholinesterase (AChE), resulting in a buildup of acetylcholine in the synaptic cleft. As a result, survivors of high-dose OP intoxication present with immediate seizures and long-term neuropathology and cognitive dysfunction. Current medical countermeasures against OPs include atropine to dampen cholinergic signaling, oximes to reactivate AChE, and benzodiazepines to terminate seizures. While these therapies may terminate seizure activity completely, they remain insufficient in protecting against long-term neurological consequences that are known to occur in human survivors. Therefore, there is an urgent need to better monitor the chronic neurological health of OP-exposed humans to identify neurological outcomes not protected by benzodiazepine treatment. The goal of this study was to evaluate the utility of preclinical micro-computed tomography (CT) imaging, a common in vivo imaging modality, for longitudinally monitoring brain health in individuals intoxicated with the OP chemical threat agent diisopropylfluorophosphate (DFP). To do so, adult male Sprague-Dawley rats (6-8 weeks) were acutely intoxicated with DFP (4 mg/kg, s.c.) and post-treated with atropine sulfate (2 mg/kg, i.m.) and the oxime 2-PAM (25 mg/kg, i.m.) within 1 min to increase survival. Additional animals were also injected with either diazepam (DZP) or midazolam (MDZ) at 40 min post-DFP to evaluate therapeutic efficacy against long-term brain damage. All individuals were subject to micro-CT scans at 3 and 6 mo post-intoxication to evaluate mineralization in the brain, a phenotype known to be associated with both chemical exposures and cognitive deficits. Interestingly, a subset of animals that received DFP displayed partial or complete resistance to seizure activity. This population, described as DFP low responders, was not treated with benzodiazepines but underwent micro-CT imaging at 2 mo post-intoxication to determine whether micro-CT imaging can also detect neurological damage in the brains of non-seizing patients. Our results demonstrate that acute DFP intoxication induced chronic mineralization in the brain detectable up to 6 mo post-exposure. Our data also demonstrate that while benzodiazepine treatment caused a partial reduction in brain mineralization, neither DZP nor MDZ offered complete neuroprotection. Additionally, DFP low responders presented with significant levels of mineralization in the brain that was detectable by micro-CT, despite the absence of seizure activity. These findings highlight the ability of micro-CT imaging to effectively capture neurological damage following OP intoxication, including in individuals that were treated with benzodiazepines to terminate seizures. In addition, the presence of mineralization in DFP low responders strongly suggests that OPs are capable of inducing neurological damage independent of seizure activity, emphasizing the need to investigate alternative therapeutic targets when developing medical countermeasures against OPs. Given that neither benzodiazepine-treated nor seizure-resistant animals were protected against cerebral mineralization, mineral deposits in the brain may play a significant role in the long-term neurological deficits observed in human survivors of OP intoxication. In conclusion, this research supports the potential use of micro-CT imaging in humans to better predict patient outcomes following a chemical emergency.