NON-SPECIFIC EFFECTS OF ORGANOPHOSPHORUS INHIBITORS OF CHOLINESTERASES

Jiří KASSA
Department of Toxicology, Purkyně Military Medical Academy, Hradec Králové

Summary

This paper proposes the main aspects of the pathogenesis of acute poisoning with organophosphorus inhibitors of cholinesterases and describes non-specific effects of these toxic substantes such as the activation of multiple non-cholinergic neurotransmitter systems in the central nervous system, the alteration of energetic metabolism, stressogenic effects, mutagenic effects, immune effects, the alteration of water and mineral metabolism, hepatotoxic effects, membrane effects as well as haematological effects. One of the most important non-specific effects produced by organophosphorus agents is the activation of multiple non-cholinergic neurotransmitter system in the central nervous system that can lead to the neuronal damage following the exposure to organophosphates.

KEY WORDS: Organophosphates; Cholinergic effects; Non-specific effects; Neuronal damage; Antidotes; Anticonvulsive drugs.

Some highly toxic organophosphates (OP) belong to the most important and the most dangerous chemical warfare agents (CWA). They have been produced in a large amount for potential chemical war and they can be also misused for the terroristic goals (1, 2). Other OP are used as insecticides. Organophosphorus insecticides (OPI) have become the most widely used class of insecticides in the world. They are used in large quantities because of their high potential for insect knockdown capacity (3, 4). In spite of relatively low toxicity in comparison with highly toxic OP, they have passed occupational hazards to workers employed in the application of these insecticides. Careless handling of OPI and their voluntary exposure with suicidal intent are the main reasons for intoxication (5, 6).

OP are usually characterized as irreversible inhibitors of acetylcholinesterase (AChE, EC 3.1.1.7),

nevertheless, their toxic effects are much more complex (7, 8, 9). It is very important and useful to know about the effects of OP as much as possible, because only the knowledge of pathogenesis of OP poisoning makes the effective treatment of OP--induced acute toxic effects possible.

The effects of OP are usually divided into two parts - cholinergic and non-specific (non-cholinergic) effects. The basic mechanism of OP action is well--known. It involves irreversible inhibition of cholinesterases, in particular AChE, and the subsequent increase in the amount of the neurotransmitter acetylcholine (ACh) at central and peripheral neuronal synapses and the overstimulation of cholinergic system. These effects are called **cholinergic effects** (1, 7).

Moreover, OP have got many other effects that have an influence on various organs and systems of organs. They are called an **non-specific (non--cholinergic) effects**. It is really difficult to decide which non-specific effects not depend on the overstimulation of cholinergic nervous system (primary effects) and which non-specific effects are caused by OP-induced cholinergic effects (secondary effects) (8, 9).

Cholinergic and non-specific effects of OP compounds differ from each other by the time of their onset following their exposure. While the cholinergic effects of OP compounds are detectable immediately following the OP poisoning (the earliest biochemical and neurochemical events detectable in the central nervous system are the inhibition of AChE and subsequent increase in ACh level), the non-specific effects of OP compounds are usually registered significantly later (10, 11).

The acute OP poisoning can be divided into three phases (11):

- **cholinergic phase** that is characterized by cholinergic effects (so called acute cholinergic crisis),
- **transitional phase** that is characterized by mixed cholinergic and non-specific effects,
- **non-cholinergic phase** that is characterized by the predominance of non-specific effects.

Till now, many kinds of non-specific effects of OP compounds have been demonstrated with the help of animal experiments. They involve activation of multiple non-cholinergic neurotransmitter systems in the central nervous system (CNS) with subsequent accumulation of the neurotransmitters such as catecholamines, excitatory and inhibitory amino acids, mutagenic effects, stressogenic effects, immune effects, hepatotoxic effects, membrane effects, haematological effects and the influence on energetic metabolism as well as on water and mineral metabolisms (8, 9, 10) (Fig. 1).

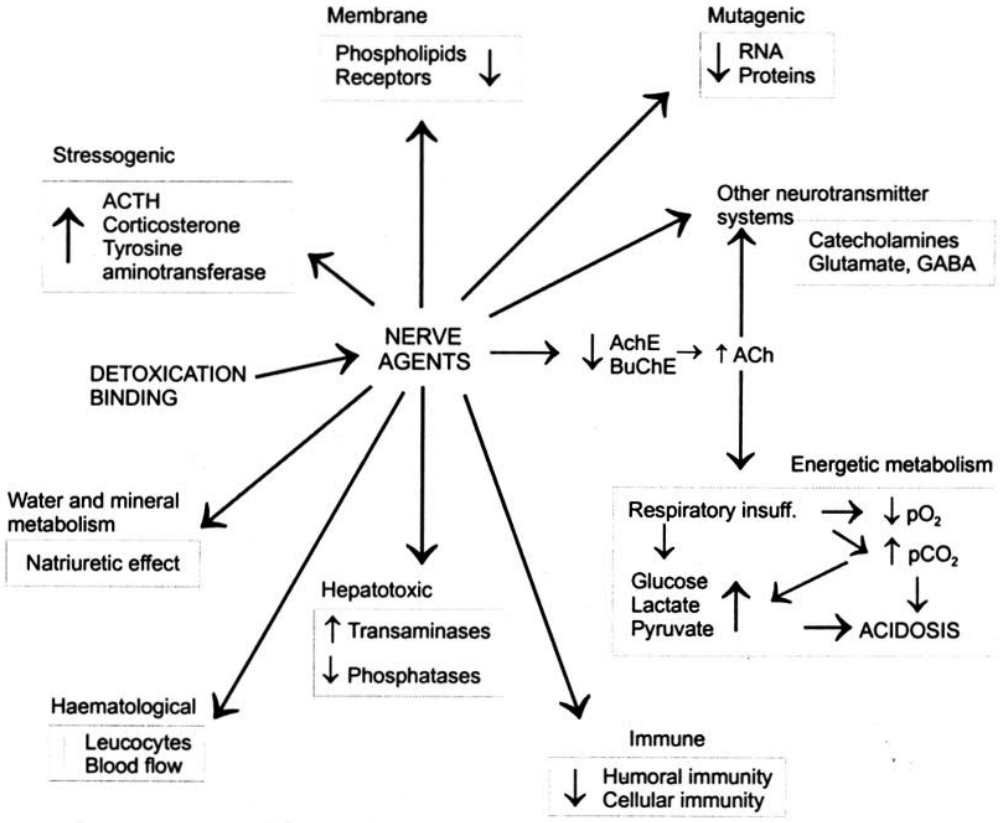

Figure 1 A schematic illustration of complex OP-induced toxic effects

One of the most important OP-induced non--specific effects is the activation of multiple non--cholinergic neurotransmitter systems in the CNS that can lead to the **neuronal damage** following the exposure to OP. The neuronal damage is probably caused by non-specific neurochemical changes such as activation of non-cholinergic neurotransmitter systems, nevertheless, these changes are secondary. They originate as a result of the seizure activity caused by OP-induced cholinergic effects - namely by the activation of central muscarinic receptors. Therefore, the seizure activity in the CNS, caused by the activation of central muscarinic receptors, is necessary for the rise of the neuronal damage. The overstimulation of central cholinergic nervous system is followed by the activation of other neurotransmitter systems. It is postulated that once seizure activity begins, the excitatory activity spreads rapidly and perturbs other neurotransmitter systems including the activation of glutamate receptors. Following increase in extracellular levels of the excitatory and potentially neurotoxic amino acid glutamate, the extracellular calcium mobilisation is enhanced. The accumulation of excessive levels of intracellular calcium can cause neuronal death through the cellular swelling and disruption of cellular membranes (11, 12, 13).

The sequence of events resulting in brain damage after OP poisoning may be summarized as follows:

1. The OP compound transverses the blood-brain barrier and inhibits brain AChE leading to a rapid increase in ACh level in the CNS.
2. The excess of ACh triggers seizure activity in susceptible brain areas.
3. Once seizures are initiated, non-cholinergic cell systems are progressively recruited and the seizures become refractory to muscarinic receptor antagonists.
4. The seizures cause the release of excessive amount of glutamate from affected glutamatergic neurones that contain NMDA receptors.
5. The released glutamate damages neighbouring neurones and eventually leads to their death. Glutamate is therefore a major excitotoxin mediating central neurotoxicity of OP compounds (14) (Fig. 2).

Another serious OP-induced non-specific effect is their **influence on energetic metabolism.** This effect is classified as non-specific effect, nevertheless, the overstimulation of cholinergic nervous system plays also important role in changes in energetic metabolism. The accumulation of ACh causes bronchoconstriction, convulsions of respiratory muscles and the depression of respiratory centers in the CNS and, thus, leads to acute respiratory insufficiency. The shortage of oxygen causes changes in metabolism, among others non-oxidative catabolism of glucose leading to the increase in lactate and pyruvate level. Respiratory

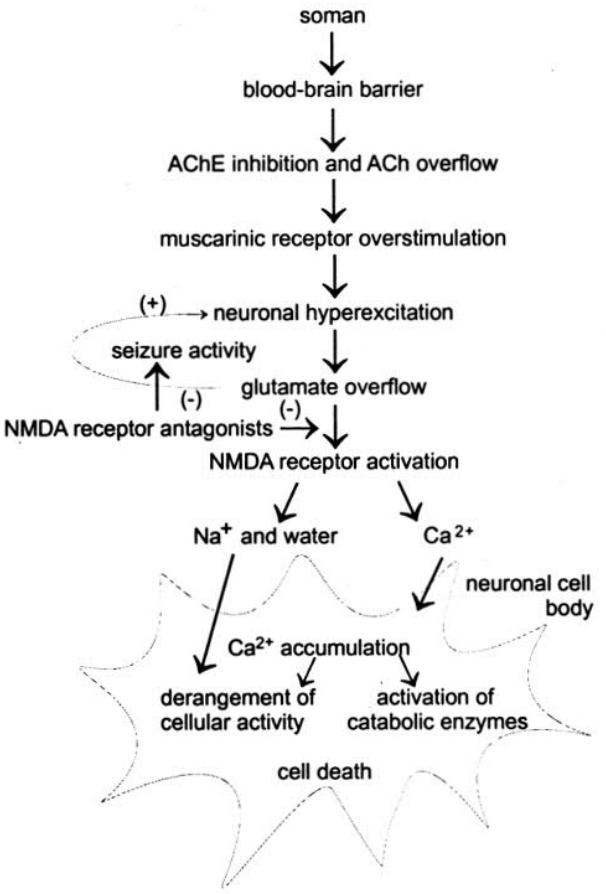

Figure 2 A schematic diagram showing glutamate mediated neuronal toxicity following the exposure to OP compound (soman)

and metabolic acidoses are the main changes in energetic metabolism following the exposure to OP compounds (8, 9, 15).

Stressogenic effects of OP compounds belong to the most important non-specific effects too. They involve general stressogenic reaction of poisoned organism characterized by the activation of sympathoadrenal and hypothalamic pituitary adrenocortical systems. We can follow the OP--induced stressogenic effects by the observation of stressogenic markers. Sympathoadrenal activation is characterized by the increase in the level of catecholamines (16), and hypothalamic pituitary adrenocortical activation is characterized by the increase in ACTH, glucocorticoids and some enzymes that realize the metabolic effects of glucocorticoids, among others tyrosine aminotransferase (TAT, EC 2.6.1.5) (17, 18).

Other OP-induced non-specific effects include **mutagenic effects** - it was found that OP compounds can influence RNA and protein metabolisms (19, 20, 21) and **immune effects** - the decrease in humoral as well as cellular immunity was demonstrated in OP-poisoned subjects (22, 23, 24). OP compounds can also cause **hepatotoxic effects** as it was demonstrated in OP-poisoned subjects by the observation of the changes in liver trans-

aminases and phosphatases in plasma (25, 26). The changes in some haematological parameters, especially the increase in the number of leucocytes, following the exposure to OP compounds were also described (8). OP compounds also **influence water and mineral metabolisms** such as natriuretic effect. The changes in water and mineral metabolisms can cause the **changes in renal functions.** A transient increase in diuresis and the decrease in glomerular filtration as well as renal tubular functions were observed following experimental OP intoxication (8, 9).

In conclusion, the pathogenesis of OP poisoning is very complicated. It is very difficult but very useful to find all relationships among all possible effects of OP compounds because the knowledge of complex pathogenesis of OP poisoning can help us to find effective antidotes against these toxic substances. Thus, the knowledge of complex pathogenesis of OP action leads to the improvement of antidotal treatment of acute OP poisoning. According to presented data, not only the careful choice of antidotes but also the timing of their administration following OP poisoning are very important (27). The complex treatment of acute OP poisoning should include not only the anticholinergic drugs to counteract the accumulation of the neurotransmitter ACh and oximes to reactivate OP-inhibited AChE but also anticonvulsive drugs to terminate the seizure activity as soon as possible and protect OP-poisoned subjects from neuronal damage. Till now, benzodiazepines have been used as anticonvulsive drugs in the treatment of OP poisoning. In the recent studies, centrally acting antimuscarinic drugs, NMDA receptor antagonists including glutamate antagonists or GABA agonists are preferred for this reason (11, 18, 28, 29). The antidotes must be administered as soon as possible not only because of the rapid onset of acute cholinergic crisis, but also because of the threat of neuronal damage of poisoned subjects following exposure to OP compounds. When the OP--induced brain excitatory activity is terminated by antidotes during the cholinergic phase of OP poisoning, no neuropathology originates because the poisoned subject is protected from the increase in extracellular glutamate level. On the contrary, when the seizure activity is terminated during the non-cholinergic phase of OP poisoning, the poisoned subject is threatened by neuronal damage because of potentially neurotoxic level of extracellular glutamate in the brain (11). Therefore, the knowledge of complex pathogenesis of OP poisoning leads to the improvement of its antidotal treatment.

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Correspondence: Plk. doc. MUDr. Jiří Kassa, CSc.
Vojenská lékařská akademie J. E. Purkyně
Třebešská 1575
500 01 Hradec Králové

Received: 6. 10. 1998