Food and Chemical Toxicology 121 (2018) 343–350

Contents lists available at ScienceDirect

Food and Chemical Toxicology

journal homepage: www.elsevier.com/locate/foodchemtox

Review

Chemical warfare agent NOVICHOK - mini-review of available data

Eugenie Nepovimova[a], Kamil Kuca[b,*]

[a] Department of Chemistry, Faculty of Science, University of Hradec Kralove, Rokitanskeho 62, 500 03, Hradec Kralove, Czech Republic
[b] Philosophical Faculty, University of Hradec Kralove, Rokitanskeho 62, 500 03, Hradec Kralove, Czech Republic

ARTICLE INFO

Keywords:
NOVICHOK
FOLIANT
Nerve agent
Chemical warfare agent

ABSTRACT

The Cold War period is characterized by the infighting between the Western countries and the USSR in diverse areas. One of such fields was development of the weapons of mass destruction. Within various programs on both sides, a wide scale of different agents have been developed. However, information about some of them are still protected under the designation "top secret". Notwithstanding, in history several cases are known when such information beheld the daylight. One of such cases was the program FOLIANT and NOVICHOK. Both programs were developed by the USSR as a reaction to English/American invention of VX agent. If at least a part of available information is truthful, we can allege that these compounds belong among the most toxic synthetic agents ever. Within this contribution, we have reviewed available Eastern and Western data about the A-agents and their precursors, so-called NOVICHOKs, including their history, synthesis, physical-chemical properties, pharmacological characteristics and clinical manifestation.

1. Historical overview

Immediately after the World War I, nobody doubted that the next war would run without chemical warfare agents (CWAs). The more the conflict seemed to be realistic, the more intensive were the preparations of potential participants. Not only the experience rising from the previous usage of CWAs was thoroughly evaluated, but also searching for novel more effective substances gradually escalated(Halámek and Kobliha, 2011; Klement, 2011).

In 1934, a project on synthetic insecticides was launched at industrial corporation I. G. Farben (Germany) by Otto Bayer who assigned this research branch to the chemist Gerhard Schräder. In 1936, Schräder's interest turned to organophosphorus compounds. His systematic work on organophosphate (OP) insecticides led to the synthesis of more than 2000 compounds, including highly toxic ethyl dimethylphosphoramidocyanidate (tabun, GA, Trilon 83, Fig. 1)(Szinicz, 2005). Subsequently, Schräder and co-workers discovered a more lethal OP compound similar to tabun – propan-2-yl methylphosphonofluoridate (GB, Trilon 46, Fig. 1). They named this compound sarin in honour of the team members: Schräder, Ambros, Ritter and van der Linde (Coleman, 2005). Since 1935, an official decree required for all inventions possessing potential military application to be reported to the German Ministry of War. In 1937, the samples of tabun and sarin were sent to the German Army Weapons Office (Wa Prüf 9) where their value for military purposes had been immediately recognized and hence all patent applications concerning these agents were declared secret (Szinicz, 2005). Work on these compounds was carefully guarded and was realized under the code name Trilon. The discovery of tabun and sarin was further followed by the revelation of a pinacolyl analogue of sarin – soman (3,3-dimethylbutan-2-yl methylphosphonofluoridate, GD, Fig. 1) in 1944 by the Nobel laureate Richard Kuhn and Konrad Henkel(Tucker, 2006). Tabun, sarin and soman belong to the class of nerve agents (NAs) that are collectively termed "*G-agents*"; the *G* stands for *German* since German researchers discovered this group of compounds(Ledgard, 2006).

Until April 1945, almost 9000 tonnes of tabun, 1300 tonnes of sarin and 20 tonnes of soman were mass-produced by the Nazi regime, however, they were never used (Halámek and Kobliha, 2011). There has been a considerable debate about why the Germans did not use their chemical arsenal in the World War II. Several explanations include: i) personal negative experience of Adolf Hitler who was seriously affected by mustard gas during the World War I; ii) fear of retaliation, and iii) underestimation of German superiority in chemical weapons (Paxman and Harris, 2011; Pitschmann, 2014; Tucker, 2006).

After World War II, another chapter in the history of development and production of CWAs began. As Nazi resistance collapsed, the Soviet and Allied forces captured the NA production facilities and technologies of the Germans' including scientific archives and experts in this field. Both groups quickly reorganized the military potential of these agents and set to work at developing and stockpiling their own supplies

* Corresponding author.
E-mail address: kamil.kuca@uhk.cz (K. Kuca).

https://doi.org/10.1016/j.fct.2018.09.015
Received 13 August 2018; Received in revised form 4 September 2018; Accepted 8 September 2018
Available online 11 September 2018

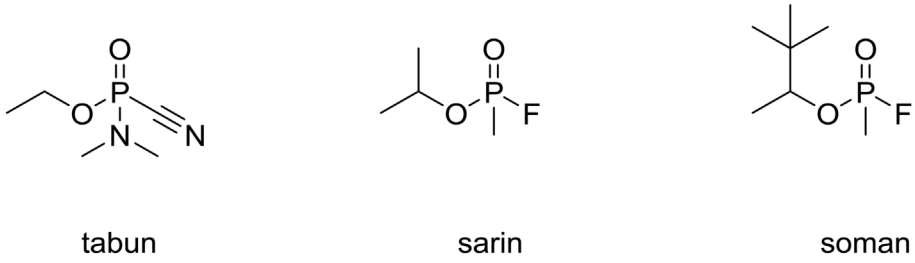

Fig. 1. Chemical structures of G-agents.

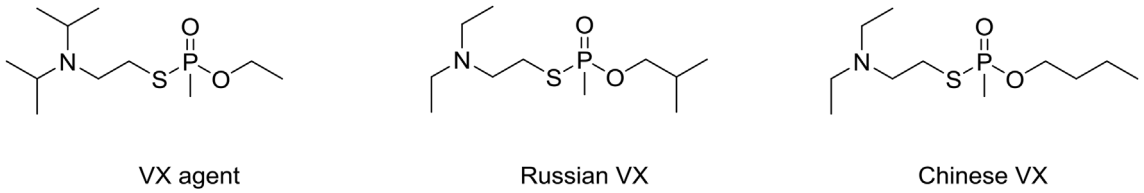

Fig. 2. Chemical structures of V-agents.

(Wiener and Hoffman, 2004). In 1949, British chemist Ranajit Ghosh discovered novel, military important group of OP esters derived from variously substituted 2-aminoethanethiole. Shortly after this discovery, the existence of these compounds had been reported to the British Chemical Warfare Establishment in Porton Down. VX agent (*S*-{2-[di (propan-2-yl)amino]ethyl} *O*-ethyl methylphosphonothioate, Fig. 2) has been selected as the most promising substance of the series(Szinicz, 2005). In a deal brokered between the British and the US governments, the British traded the VX technology for the thermonuclear weapons technology of the United States. Thereafter, full scale production of VX commenced in 1961 in the USA (Tucker, 2006). During the same period, the Soviet scientists developed independently of the UK and the USA an isomer of VX agent – the so-called Russian VX (VR, RVX, Substance 33, *S*-[2-(diethylamino)ethyl] *O*-(2-methylpropyl) methylphosphonothioate, Fig. 2) which later became a prototype for the series of NOVICHOK agents. Another structural analogue of VX known as Chinese VX (CVX, *O*-butyl *S*-[2-(diethylamino)ethyl] methylphosphonothioate, Fig. 2) was also developed and studied(Romano et al., 2007).

VX and its analogues belong to the class NAs that are collectively termed "*V-agents*"; the *V* stands for *venemous* because they are very toxic and the symptoms of their intoxication resemble the manifestation of snake venom poisoning(Tucker, 2006). These phosphonylated and phosphorylated thiocholine derivatives, lacking electron-withdrawing groups such as halogen or cyano group, are very reluctant to nucleophilic substitution and so to hydrolysis. This fact together with extremely low volatility ensure the resorption of these substances through unprotected skin into the bloodstream(Ellison, 2007; Wiener and Hoffman, 2004).

Until the 1950′s, the CWAs were unitary, i.e. the toxic agent was filled in the ammunition and then stored until its use. The problems resulting from the production, stockpiling of unitary CWAs as well as from requirement for expensive disposal of defective or expired chemical ammunition, disagreement of local population, activities of various ecological organisations and last, but not least from the absence of novel CWAs with desirable stability within the process of storage launched in the USA the project denoted Binary Lethal Weapons System. For the purpose of this project two NAs were selected – sarin and VX (Fig. 3) (Gupta, 2015; Halámek and Kobliha, 2011). Such technology involves two or more non-toxic chemical precursors physically separated from each other. The final step of synthesis of the toxic agent from above-mentioned precursors is performed immediately before or in the process of firing of the ammunition(Wiener and Hoffman, 2004). Therefore, binary weapons (BWs) prevent unintentional toxicity to those handling, transporting or disposing the weapons. The complications associated with BWs involve complicated construction of ammunition, smaller cartridges where the precursors are imposed and lower yields of the final step.(Halámek and Kobliha, 2011).

Initially, the Soviets had very reserved attitude to BWs. They were convinced that this type of ammunition is generally less efficient and due to complicated construction more expensive comparing to weapons filled with unitary agents. The Soviets perceived the BW program as a way of the USA how to circumvent the upcoming Chemical Weapons Convention (CWC) (Halámek, 2008). Such negative attitude of the USSR was probably determined by huge amounts of stockpiled unitary CWAs but also by relatively low age of chemical arsenal. Not negligible was also the fact that the USSR did not puzzle over the disposal of expired or defective ammunition. However, with the progress of the time all these arguments lost importance since signing of the CWC was drawing nearer as well as the Soviet chemical arsenal was gradually expiring.(Halámek, 2008).

At the time, when in the US binary weapon program was at its peak, several long-term research projects took place also in the USSR. Among them, the most important in the field of chemical sciences were FLUORINE (in Russian "*FTOR*") and PHOSPHORUS (in Russian "*FOSFOR*"). These projects were of high priority not only from the point of view of national economy but also of the military sector. Particular attention was paid to compounds with strong biocidal effect(Halámek, 2008; Vásárhelyi and Földi, 2007).

Moreover, it was found that within the storage process Russian VX was very sensitive to moisture. The necessity of RVX stabilization together with development of its thickened version or even conversion to the binary form led to initiation and escalation of the top secret Soviet project FOLIANT between 1973 and 1976. The knowledge obtained within FLUORINE and PHOSPHORUS projects was incorporated into the FOLIANT program as well. The main aim of this project was synthesis of the third generation of NAs with higher toxicity compared to V-agents that will be undetectable using NATO standard chemical detection equipment(Halámek, 2008; Halámek and Kobliha, 2011; Vásárhelyi and Földi, 2007). More than 200 chemists and engineers were involved in the FOLIANT program. According to available sources, at least three unitary chemical weapons were synthesized (A 230, A 232 and A234, Fig. 4). Structures of so-called A-agents have been never published. Over the last few years, the information that these compounds are derivatives of dihaloformaldoxime started to appear. This assumption was based on published works of Soviet chemists who probably participated on the FOLIANT program(Kruglyak et al., 1972a, 1972b; Malekin et al., 1972; Martynov et al., 1969; Petrov et al., 1967;

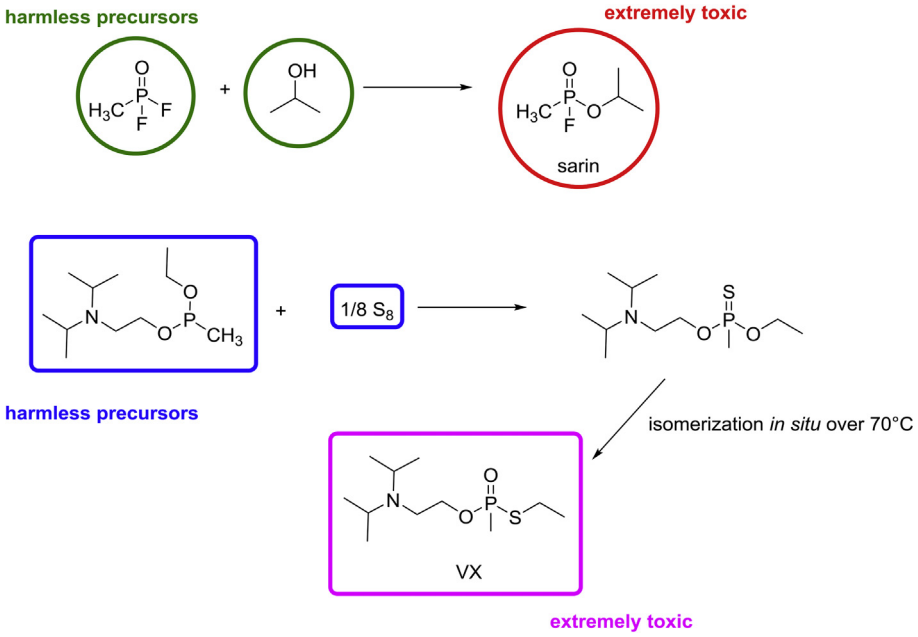

Fig. 3. The principle of binary weapons depicted on sarin and VX.

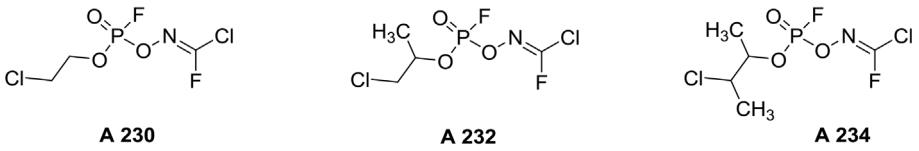

Fig. 4. Plausible chemical structures of A-agents.

Razumova et al., 1968). Interestingly, almost at the same time the fact that acetylcholinesterase (AChE, E. C. 3.1.1.7) reactivators are able to restore the function of OP-inhibited AChE was published(Hobbiger, 1957). From the chemical point of view, such agents are oximes. Later, however, it was revealed that phosphorylated/phosphonylated oximes are formed immediately after the process of enzyme recovery. Such intermediates, instead of reducing the inhibitory effect of parent OP towards AChE, inhibit it more strongly(Andersen, 1978; Fossier et al., 1983).

A significant drawback of A-agents was their low stability in the environment(Karev, 2009). However, this shortcoming seems to be negligible when a binary form of afore-mentioned agents is developed passing the requirement of stability on their precursors. Thus, at least 5 types of binary A-agents were prepared. This program bore the code name NOVICHOK. Compounds, referred to as NOVICHOKs, were commonly used intermediates within chemical industry and were not included in proposed CWC. Therefore, particularly these compounds were selected as precursors of binary form of A-agents(Karev, 2009). According to S. L. Hönig, the structural moiety of so-called NOVICHOKs could be 2-fluoro-1,3,2-dioxophospholane (Fig. 5) (Hoenig, 2007). Though much of the knowledge about the synthesis and various properties of phosphorylated and/or phosponylated oximes, amidates and related compounds was published in Russian literature in the 1960′s and 1970′s, (Kruglyak et al., 1972b, 1972a; Malekin et al., 1972; Martynov et al., 1969; Petrov et al., 1967; Razumova et al., 1968), the first exact information on A-agents and NOVICHOKs was posted only after the end of the FOLIANT project (after 1992) from the defectors (Mirzayanov, 2009).

In September 1992, an article describing that the USSR violates the CWC continuing to produce and test the NAs of the third generation, was published in the newspaper the Moscow News(Mirzayanov, 1992). The author of this publication was Vil Mirzayanov, a researcher of the State Research Institute of Organic Chemistry and Technology. Promptly after the article publication, he was arrested for high treason but due to a huge public echo a few months later he was released and emigrated into the USA (Halámek, 2008; Karev, 2009). In this way, the West first learned about the existence of the novel class of CWAs with a much stronger toxic effect than the synthetic chemical warfare available at that time. Today, the information on synthesis, physical-chemical properties, toxicity and military characteristics of A-agents and NOVICHOKs is still guarded under the designation “top secret”. A substantial part of revealed data is derived only from the interviews and publications of Mirzayanov, Uglev and Zheleznyakov, as well as from the chairman of the Union for Chemical Security Lev Fyodorov(Karev, 2009). The latter environmental activist was intensively involved in declassification of the Russian chemical weapons program, in its disposal, environmental burden elimination as well as health protection of the population in affected areas. The ending of the project NOVICHOK was influenced by the coincidence of the CWC adoption (Table 1) (Halámek, 2008).

In March 2018, the former Russian spy Sergey Skripal and his daughter Yuliya were found unconscious in Salisbury, UK. Later, the British authorities classified this incident as poisoning with a nerve agent from the Novichok group. On March 12, 2018, British Prime Minister Theresa May stated in the parliament: “Either this was a direct action by the Russian state against our country, or the Russian government lost control over the potentially catastrophically destructive substance and allowed it to fall into hands of others. Russia refused this blame. Inasmuch as the investigation of this incident is still under way, it is currently not possible to certainly identify which country is right and which is deceiving(Nepovimova and Kuca, 2018).

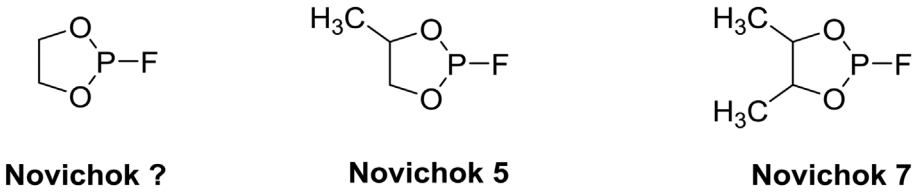

Fig. 5. Plausible chemical structures of precursors – NOVICHOKs.

Table 1
The results of FOLIANT and NOVICHOK projects.

	RVX	RVX	A 230	A 232	A 232	A 234
Form	unitary	binary	unitary	unitary	binary	binary
Precursor	–	n.a.	–	–	Novichok 5	Novichok 7
Amount	15 000 tonnes	tens of tonnes	tens of tonnes	several tonnes	several tonnes	tens of tonnes
Years of Testing	n.a.	1988–89	1988–89	n.a.	1989–90	1993
Weaponization	n.a.	weaponized in 1990	weaponized in 1990	was not weaponized	approved in 1989	n.a.

n.a. data not available.

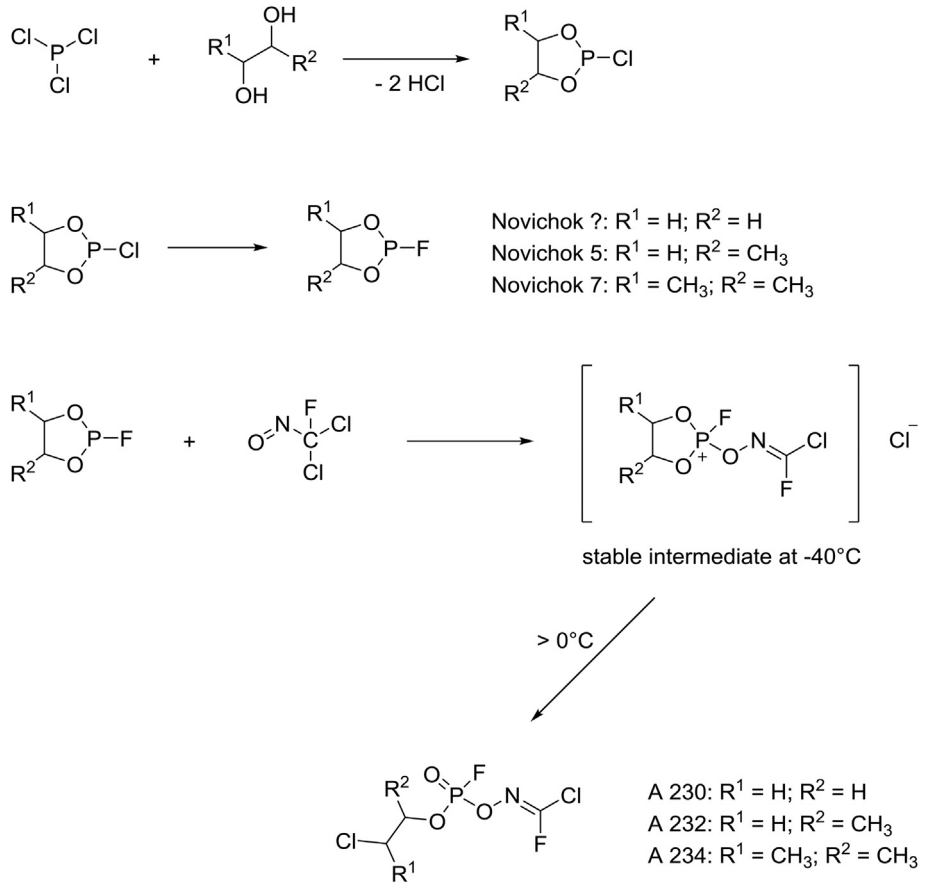

Fig. 6. Possible synthetic approach leading to phosphorylated oximes.

2. Synthesis

Since there are discrepancies in exact chemical structure of A-agents, this section is divided into two parts describing plausible synthetic routes leading to single structural type.

If A-agents belong to the group of phosphorylated oximes (Fig. 4), their synthesis would probably involve three steps(Ellison, 2016; Halámek and Kobliha, 2011) (Fig. 6). The first two steps would lead to preparation of A-agents precursors, so-called NOVICHOKs (Novichok ?, Novichok 5, Novichok 7) (Fig. 5) by the reaction of phosphorus trichloride with appropriate diol and subsequent nucleophilic substitution of chlorine atom by fluorine. Resulting 2-fluoro-1,3,2-dioxaphospholanes readily react with dichloro(fluoro)nitrosomethane, a compound structurally similar to choking agents such as chloropicrin (trichloronitromethane), fluoropicrin (trifluoronitromethane) or trifluoronitrosomethane. The mechanism of such reaction between phosphites and α-trihalonitrosoalkanes to form oxime esters of phosphates was formerly described by Allen(Allen et al., 1956; Allen and Johnson,

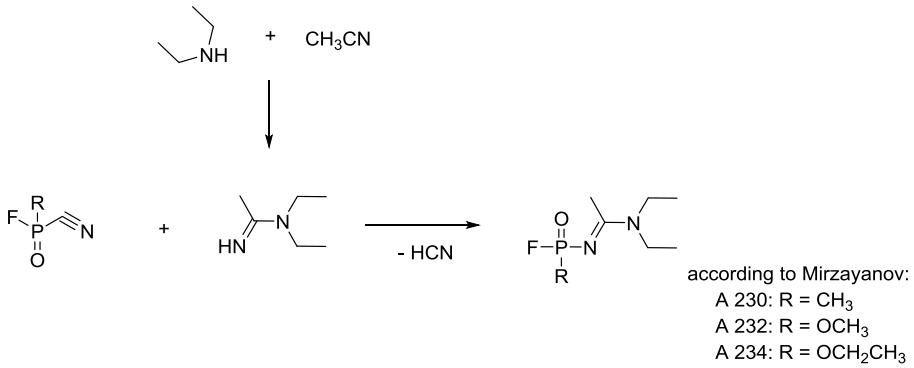

Fig. 7. Synthetic route leading to phosphoroamidates described by V. Mirzayanov.

1955). So-called Allen's reaction is a subtype of Michaelis – Arbuzov reaction of trialkyl phosphite and alkyl halide to form phosphonates (Burton and Flynn, 1977). The stability of formed intermediates is probably temperature-dependent. Under subzero temperatures (- 40 °C) they are stable but upon being warmed the nucleophilic attack by chloride anion is facilitated resulting in phospholane ring opening (Halámek and Kobliha, 2011; Hoenig, 2007).

Not only phosphorus chlorides or phosphorus oxychlorides could be used as starting materials, but also many other intermediates of phosphorus chemistry used in pesticide, plasticizer or detergent industry.

With regard to the structures of A-agents, several general conclusions should be mentioned:

1) There is no C-P bond in the molecule. Therefore, these compounds are related to fluorophosphates which are NOT within the scope of the CWC.
2) The secondary alkoxy side chains, not being methyl, ethyl, propyl or isopropyl, are also OUTSIDE the CWC scope.
3) P-O-N linkage is NOT mentioned in the CWC.
4) The precursor fluorodichloronitrosomethane is NOT regulated by the CWC, although chloropicrin is.("Organisation for the Prohibition of Chemical Weapons," n.d.)

According to Mirzayanov, a defector devoted to Soviet chemical weapons, A-agents belong to the group of phosphoroamidates (Fig. 7). In his book *State Secrets. An Insider´s Chronicle of the Russian Chemical Weapons Program*, he alleges that for the preparation of A 232 a reaction of methyl phosphorocyanidofluoridate with *N,N*-diethylethanimidamide had been approved. The latter intermediate is a product of the reaction of diethylamine and acetonitrile(Halámek and Kobliha, 2011; Mirzayanov, 2009). In context of above-mentioned synthesis and character of reactants, a question arises – which of the starting compounds act as NOVICHOK precursor?

3. Physical-chemical properties

Only very little information is available about physical-chemical properties of A-agents and NOVICHOKs(Halámek and Kobliha, 2011; Hoenig, 2007; Karev, 2009; Mirzayanov, 2009; Pitschmann, 2014). Table 2 represents the best known physical-chemical properties of A-agents, sarin and VX.

A significant drawback of currently available NAs is an imbalance between their persistency and volatility. On one side, there are G-agents for which high volatility and low persistency is characteristic. On the other hand, there are V-agents that at the expense of high persistency display decreased efficacy in live power elimination by a direct action of vapours(Newmark, 2007; Wiener and Hoffman, 2004). The aim of military chemists working on the third generation of NAs was to develop compounds with balanced physical-chemical properties such as volatility, density and stability towards light and moisture. Based on the data listed above, one can assume that several shortcomings typical for the first two generations of NAs had been removed.

4. Mode of action

The mode of action of A-agents is irreversible inhibition of the AChE (Hoenig, 2007). Under physiological conditions, Ser-His-Glu triad, located in the active site of AChE, hydrolyzes neurotransmitter acetylcholine (ACh) reducing thus its concentration at neuronal cholinergic synapses and neuromuscular junctions(Dvir et al., 2010). Once the A-agent reaches the bottom of the active site gorge, the nucleophilic attack of phosphorus atom by the hydroxyl group of serine occurs. This attack is accompanied by a simultaneous departure of the fluoride ion and formation of phosphorylated enzyme (Fig. 8). Due to formation of the covalent bond between the phosphorus atom of OP and AChE's serine, the spontaneous hydrolysis of phosphorylated enzyme is extremely slow varying from hours to days(Korabecny et al., 2014).

From the literature it is well-known that phosphorylated oximes are relatively unstable (Leader et al., 1999; Stenzel et al., 2007). Therefore, an analogy between phosphorylated oxime and AChE-inhibited by A-agent could be assumed (Fig. 9). Rapid hydrolysis of = N-O- bond within the A-agent-AChE adduct may result in so-called aged form of the enzyme where the phosphonic oxyanion forms a salt bridge with the protonated histidine that strongly stabilizes the conjugate (Fig. 8). Once aging occurs, the enzyme is permanently inactivated and no therapy can restore its activity(Kuca and Pohanka, 2010; Sharma et al., 2015). Such hypothesis is at least partly supported by the well-known fact that AChE inhibited by the A-agent ages rapidly. In addition, according to available literature, the aging half-time of A 230 is similar to that observed in soman, i.e. 2–4 min(Karev, 2009; Sirin et al., 2012).

From the therapeutic point of view, we should probably part with the idea of restoring the function of inhibited AChE by A-agents. Not only because of the rapid aging process, but also due to debilitated partial positive charge on phosphorus atom. Forasmuch as, the nucleophile represented by currently available reactivators (pralidoxime, obidoxime, HI-6) would attack such phosphorus very hardly (Fig. 8). Therefore, the only effective treatment approaches would be symptomatic therapy (combination of anticholinergic agent and anticonvulsant) or administration of so-called bioscavengers, such as butyrylcholinesterase (BChE, E. C. 3.1.1.8). Such exogenously administered molecule/enzyme would bind and detoxify free NA entering a patient's circulation so that it would not be able to reach the tissue AChE and produce clinical symptoms of intoxication(Bajgar et al., 2009). Generally, bioscavengers belong among the prophylactic approaches, however, in case of A-agents intoxication when causal therapy is ineffective, they can be used *ex post*. Additionally, BChE is able to detoxify all types of NAs representing thus the universal approach since it takes quite long time to determine which NA has been used and select appropriate therapy(Bajgar et al., 2009).

Table 2
Available data about physical-chemical properties of A-agents, sarin and VX.

	A 230	A 232	A 234	sarin	VX
Molecular mass	241.95	255.97	270.00	140.09	211.2
Boiling point	61–62 °C	70–71 °C	73–74 °C	147 °C	256 °C
Density	1.612 g/mL	1.515 g/mL	1.414 g/mL	1.102 g/mL	1.062 g/mL
State	liquid	n.a.	n.a.	liquid	liquid
Behaviour at low temperature	solidifies at low temperature	does not solidify at low temperature	n.a.	does not solidify at low temperature	does not solidify at low temperature
Volatility	volatile	more volatile than A 230 or RVX	n.a.	volatile	not volatile
Moisture stability	resistant to moisture	less stable against moisture than A 230 or RVX	n.a.	resistant to moisture	resistant to moisture

n.a. data not available.

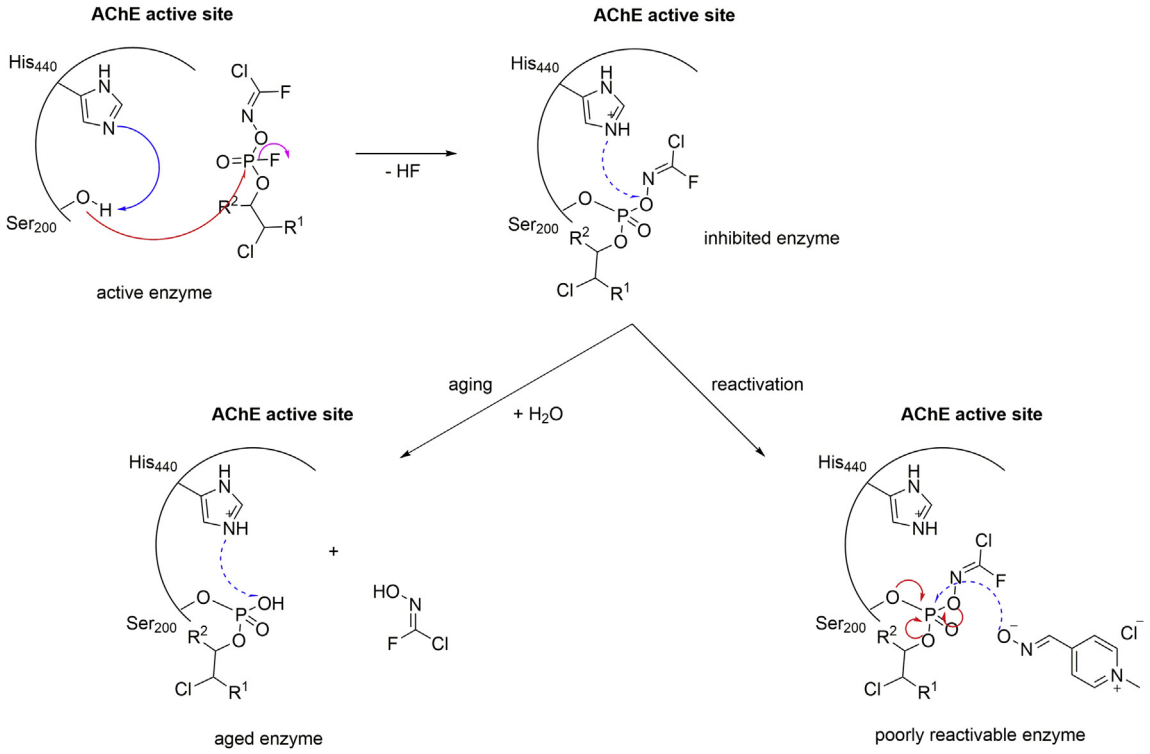

Fig. 8. Mechanism of AChE inhibition by A-agent, aging and reactivation by oximes.

5. Toxicity

According to unauthorized sources, so-called A-agents exert higher or at least similar toxicity as VX. Within seminar work of Karev elaborated at Penza State University, Russia, the exact toxicological data of A-agents could be found(Karev, 2009). These parameters are listed in Table 3.

As NAs, also A-agents belong to the class of irreversible inhibitors of AChE. These compounds block the action of AChE preventing thus physiological breakdown of the neurotransmitter ACh. Therefore, ACh accumulates in the synaptic cleft where it causes cholinergic receptors overstimulation. The severity of the symptoms depends mainly on the amount of the agent, which entered the body. Generally, the symptoms could be divided into three groups: muscarinic, nicotinic and central. Overstimulation of muscarinic cholinergic receptors causes pupils constriction, glandular hypersecretion, urination, defecation, diaphoresis and gastric emesis. Among nicotinic symptoms belong initial defasciculation followed by weakness and flaccid paralysis. Finally, within central nervous system, NA poisoning manifests as irritability, giddiness, fatigue, lethargy, seizures, coma and mostly fatal respiratory depression(Korabecny et al., 2014; Kuca et al., 2013).

Nevertheless, there are also several differences. Within unauthorized reports it was mentioned that the symptoms of intoxication were virtually incurable as well as that people who were exposed to such kind of NA remained disabled invalids(Karev, 2009). Probably these reports are describing the so-called delayed neurotoxicity characterized by severe disruption of the nerve system manifesting as paralysis occurring within 1–3 weeks after the intoxication(Gupta, 2015). The above-mentioned scientist Andrey Zheleznyakov was probably one of the persons who was exposed to A 232. Five years after the intoxication, he died suffering from cirrhosis, trigeminal neuritis and epilepsy(Karev, 2009).

6. Treatment management

General treatment protocol of nerve agent intoxication includes pharmacological as well as non-pharmacological approaches. Among the non-pharmacological methods belong oxygen supply, resuscitation and decontamination(Sharma et al., 2015). For the decontamination purposes, different countries are using diverse decontamination means as well as different decontamination approaches based on the national standards. Among them, probably the most preferred seems to be

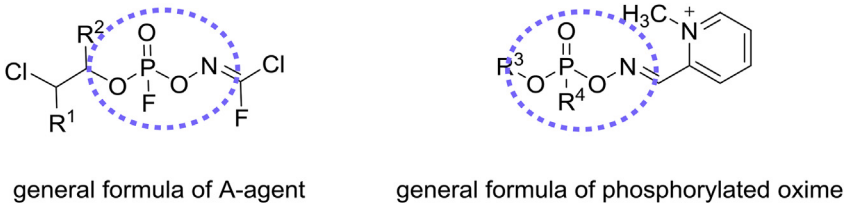

Fig. 9. Analogy between the A-agent and phosphorylated oxime.

Table 3
Toxicological parameters of G-, V- and A-agents.

	LCt_{50} (mg*min/m^{-3})	LD_{50} percutaneous administration (mg/person)
sarin	100	1700
soman	70	350
VX	50	10
A 232	6–10	1–2
A 234	7	5

personal and scene decontamination. Immediate skin decontamination can preserve survival of casualties and prevent the progression of harmful effects associated with intoxication, while scene decontamination could prevent the intoxication of medical personnel and/or bystanders. Frequently used reactive skin decontamination lotion (RSDL) contains Dekon 139 and a small amount of 2,3-butandione monooxime (Worek et al., 2016). For the purpose of scene decontamination, it is assumed that nerve agents could be neutralized by basic solutions such as NaOH, Na_2CO_3 or undiluted household bleach. Pharmacological treatment of nerve agent poisoning involves: (1) application of anticholinergic drug (mostly atropine); (2) administration of anticonvulsant agent (usually diazepam or avizafon); and (3) use of reactivating agents represented by AChE reactivators (called oximes)(Kuca et al., 2013). However, as it has been mentioned previously, we assume that oximes would be hardly effective, since the intermediate formed between the active site of AChE and A-agent would make it harder to initiate the nucleophilic attack by the reactivators. For this reason, apart from the symptomatic treatment, bioscavangers should be also considered as an alternative way how to treat A-agent intoxication, since they non-selectively inactivate unbound A-agents in the blood stream.

7. Conclusion

Almost thirty years passed since the first information about the mysterious compounds, called NOVICHOKs, has been revealed. Although the design, synthesis, analysis and testing of these agents have passed through a relatively large number of people, the exact reliable data are still missing. Probably the ability to protect the secrets is much stronger than it seems to be. In such cases, there is nothing more than creating the assumptions. If at least a part of supposed information was/is true, these extremely lethal weapons could represent a huge security risk. To avoid any tragic scenario, it is necessary to strengthen international regulations and verifications. First of all, it is necessary to include the A-agents and their precursors on the CWC list. Subsequently, small amounts of these substances should be prepared to test the effectiveness of potential antidotal therapy and/or to develop effective detection. Hopefully, future advances in military chemistry/biology will further improve our protection against such agents, while juristic advances within CWC will decrease the likelihood of the attack.

Acknowledgement

This work was supported by the project Excellence of the Faculty of Science, University of Hradec Kralove.

Transparency document

Transparency document related to this article can be found online at https://doi.org/10.1016/j.fct.2018.09.015.

Abbreviations

ACh – acetylcholine; AChE – acetylcholinesterase; BChE – butyrylcholinesterase; BW – binary weapon; CVX – Chinese VX; CWA – chemical warfare agent; CWC – Chemical Weapon Convention; GA – tabun; GB – sarin; GD – soman; NA – nerve agent; OP – organophosphate; RSDL – reactive skin decontamination lotion; RVX – Russian VX; VR – Russian VX.

References

Allen, J.F., Johnson, O.H., 1955. The synthesis of monovinyl esters of phosphorus(V) acids. J. Am. Chem. Soc. 77, 2871–2875. https://doi.org/10.1021/ja01615a061.
Allen, J.F., Reed, S.K., Johnson, O.H., Brunsvold, N.J., 1956. Structure of the reaction products of fully esterified phosphorus(III) acids and α-halo carbonyl Compounds1. J. Am. Chem. Soc. 78, 3715–3718. https://doi.org/10.1021/ja01596a044.
Andersen, R.A., 1978. Affinity and phosphorylation constants for two phosphorylated oximes in the inhibition of chicken brain cholinesterase. Gen. Pharmacol. 9, 333–334.
Bajgar, J., Fusek, J., Kassa, J., Kuca, K., Jun, D., 2009. Chemical aspects of pharmacological prophylaxis against nerve agent poisoning. Curr. Med. Chem. 16, 2977–2986.
Burton, D.J., Flynn, R.M., 1977. Michaelis-arbuzov preparation of halo-F-methylphosphonates. J. Fluor. Chem. 10, 329–332. https://doi.org/10.1016/S0022-1139(00)83108-5.
Coleman, K., 2005. A History of Chemical Warfare. Palgrave Macmillan UK, London. https://doi.org/10.1057/9780230501836.
Dvir, H., Silman, I., Harel, M., Rosenberry, T.L., Sussman, J.L., 2010. Acetylcholinesterase: from 3D structure to function. Chem. Biol. Interact. 187, 10–22. https://doi.org/10.1016/j.cbi.2010.01.042.
Ellison, D.H., 2016. Emergency Action for Chemical and Biological Warfare Agents, second ed. CRC Press.
Ellison, H.D., 2007. Handbook of Chemical and Biological Warfare Agents. CRC Press WWW Document. https://www.crcpress.com/Handbook-of-Chemical-and-Biological-Warfare-Agents-Second-Edition/Ellison/p/book/9780849314346, Accessed date: 1 April 2018.
Fossier, P., Tauc, L., Baux, G., 1983. Side effects of phosphorylated acetylcholinesterase reactivators on neuronal membrane and synaptic transmission. Pflugers Arch 396, 8–14.
Gupta, R.C., 2015. Handbook of Toxicology of Chemical Warfare Agents. Academic Press.
Halámek, E., 2008. Poslední generace chemických zbraní. Czech Mil. Rev. 137–146.
Halámek, E., Kobliha, Z., 2011. Potential chemical warfare agents. Chem. Listy 105, 323–333.
Hobbiger, F., 1957. Protection against the lethal effects of organophosphates by pyridine-2-aldoxime methiodide. Br. J. Pharmacol. Chemother. 12, 438–446.
Hoenig, S.L., 2007. Compendium of Chemical Warfare Agents. Springer-Verlag, New York.
Karev, S.A., 2009. Проблемы Химического Разоружения И Пути Их Ре♯ения.
Klement, C., 2011. Mimoriadne udalosti vo verejnom zdravotnictve. PRO, Banska Bystrica, Slovakia.
Korabecny, J., Soukup, O., Dolezal, R., Spilovska, K., Nepovimova, E., Andrs, M., Nguyen, T.D., Jun, D., Musilek, K., Kucerova-Chlupacova, M., Kuca, K., 2014. From pyridinium-based to centrally active acetylcholinesterase reactivators. Mini Rev. Med. Chem. 14, 215–221.
Kruglyak, Y.L., Malekin, S.I., Leibovskaya, G.A., Khromova, Z.I., Sretenskaya, I.I., Martynov, I.V., 1972a. Reaction of a-chloronitrosoalkanes with phospholanes and iminophosphites. In: Khim. Primen. Fosfororg. Soedin. 1969. Presented at the Khim. Primen. Fosfororg. Soedin. 1969, Nauka, Moscow, USSR, pp. 307–312.
Kruglyak, Y.L., Malekin, S.I., Martynov, I.V., 1972b. Phosphorylated oximes. XII. Reactions of 2-halophospholanes with dichlorofluoronitrosomethane. Russ. J. Gen. Chem. 42, 811–814.
Kuca, K., Jun, D., Musilek, K., Pohanka, M., Karasova, J.Z., Soukup, O., 2013. Prophylaxis and post-exposure treatment of intoxications caused by nerve agents and organophosphorus pesticides. Mini Rev. Med. Chem. 13, 2102–2115.
Kuca, K., Pohanka, M., 2010. Chemical warfare agents. EXS 100, 543–558.
Leader, H., Vincze, A., Manisterski, B., Rothschild, N., Dosoretz, C., Ashani, Y., 1999.

Characterization of O,O-diethylphosphoryl oximes as inhibitors of cholinesterases and substrates of phosphotriesterases. Biochem. Pharmacol. 58, 503–515.
Ledgard, J., 2006. A Laboratory History of Chemical Warfare Agents. Jared Ledgard.
Malekin, S.I., Yakutin, V.I., Sokaĺskii, M.A., Kruglyak, Y.L., Martynov, I.V., 1972. Mechanism of the reaction of alpha-chloronitrosoalkanes with phosphorus(III) compounds. Russ. J. Gen. Chem. 42, 807–811.
Martynov, I.V., Kruglyak, Y.L., Malekin, S.I., 1969. Carbonyl Halide Oxime B-chloroalkyl Fluorophosphates. SU 241433 19690418.
Mirzayanov, V.S., 2009. State Secrets: an Insider's Chronicle of the Russian Chemical Weapons Program. Outskirts Press, Incorporated.
Mirzayanov, V.S., 1992. Poisoned Politics. The Moscow News.
Nepovimova, E., Kuca, K., 2018. The history of poisoning: from ancient times until modern ERA. Arch. Toxicol. https://doi.org/10.1007/s00204-018-2290-0.
Newmark, J., 2007. Nerve agents. The Neurologist 13, 20–32. https://doi.org/10.1097/01.nrl.0000252923.04894.53.
Organisation for the Prohibition of Chemical Weapons [WWW Document], n.d. URL https://www.opcw.org/(Accessed 25 March 2018).
Paxman, J., Harris, R., 2011. A Higher Form of Killing. Random House.
Petrov, A.A., Razumova, N.A., Evtikhov, Z.L., 1967. Condensation of acid fluorides of glycol phosphites with diene hydrocarbons. Russ. J. Gen. Chem. 37, 1410–1411.
Pitschmann, V., 2014. Overall view of chemical and biochemical weapons. Toxins 6, 1761–1784. https://doi.org/10.3390/toxins6061761.
Razumova, N.A., Evtikhov, Z.L., Petrov, A.A., 1968. Phosphorus-containing heterocycles. XV. Acid fluorides of glycolphosphorous acids. Russ. J. Gen. Chem. 38, 1117–1122.
Romano, J.A., Romano, J.A., Salem, H., Lukey, B.J., Lukey, B.J., 2007. Chemical Warfare Agents: Chemistry, Pharmacology, Toxicology, and Therapeutics, second ed. CRC Press.
Sharma, R., Gupta, B., Singh, N., Acharya, J.R., Musilek, K., Kuca, K., Ghosh, K.K., 2015. Development and structural modifications of cholinesterase reactivators against chemical warfare agents in last decade: a review. Mini Rev. Med. Chem. 15, 58–72.
Sirin, G.S., Zhou, Y., Lior-Hoffmann, L., Wang, S., Zhang, Y., 2012. Aging mechanism of soman inhibited acetylcholinesterase. J. Phys. Chem. B 116, 12199–12207. https://doi.org/10.1021/jp307790v.
Stenzel, J., Worek, F., Eyer, P., 2007. Preparation and characterization of dialkylphosphoryl-obidoxime conjugates, potent anticholinesterase derivatives that are quickly hydrolyzed by human paraoxonase (PON1192Q). Biochem. Pharmacol. 74, 1390–1400. https://doi.org/10.1016/j.bcp.2007.07.013.
Szinicz, L., 2005. History of chemical and biological warfare agents. Toxicology 214, 167–181. https://doi.org/10.1016/j.tox.2005.06.011.
Tucker, J.B., 2006. War of Nerves: Chemical Warfare from World War I to Al-qaeda. Pantheon Books.
Vásárhelyi, G., Földi, L., 2007. History of Russia's chemical weapons. AARMS 6, 135–146.
Wiener, S.W., Hoffman, R.S., 2004. Nerve agents: a comprehensive review. J. Intensive Care Med. 19, 22–37. https://doi.org/10.1177/0885066603258659.
Worek, F., Wille, T., Koller, M., Thiermann, H., 2016. Toxicology of organophosphorus compounds in view of an increasing terrorist threat. Arch. Toxicol. 90, 2131–2145. https://doi.org/10.1007/s00204-016-1772-1.