Physiol. Res. 41: 399 - 402, 1992

SHORT COMMUNICATION

Acute Toxicities of 2-Dialkylaminoalkyl-(Dialkylamido)-Fluoro-phosphates

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Received March 27, 1992
Accepted June 2, 1992

Summary

Toxicities expressed as LD_{50} values of 2-dialkylaminoalkyl-(dialkylamido)-fluorophosphates for rats and mice (i.m. administration) were determined. Rats were more sensitive to these compounds than mice: LD_{50} values varied from 17 (rats) to 1222 (mice) μg/kg. LD_{50} values at different routes of administration (i.v., i.m., s.c., p.o. and p.c.) for one derivative of this group, 2-dimethylaminoethyl-(dimethylamido)-fluorophosphate, were determined. Depending on the route of administration, LD_{50} values varied from 11 (i.v.) to 190 (p.o.) μg/kg for rats and from 27.6 (i.v.) to 222 (p.o.) μg/kg for mice, respectively. Percutaneous toxicity in rats only (LD_{50} = 1366 μg/kg) was determined.

Key words

LD_{50} values – 2-dialkylaminoalkyl-(dialkylamido)-fluorophosphates

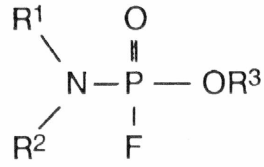
There exist many compounds comprising high toxicity and therefore exhibiting military significance for their possible use as chemical weapons (CW) (Document 1992). However, organophosphorus compounds like sarin, soman and VX are the main part of these chemicals. The existence of other classes of organophosphorus agents cannot be excluded. Therefore any kind of information dealing with highly toxic compounds is of importance for the risk assessement of their possible conversion into CW. Moreover, these types of chemicals could be used as model compounds in pharmacology, toxicology, neurochemistry etc.

The aim of this article is to describe toxicities of some organophosphorus compounds with different chemical structure than that observed for the most obvious representatives of CW like sarin, soman and VX. This new class of organophosphates can be described in general as 2-dialkylaminoalkyl-(dialkylamido)-fluorophosphates. In their chemical formulae (Table 1), the structural similarities with the group of so called G-compounds (i.e. sarin, soman) and V-compounds (i.e. VX and others) can be observed. These chemicals were designated as GV compounds. Their toxicities are not known at present.

Female rats (VELAZ Praha) weighing 180–220 g or mice (VELAZ Praha), weighing 21–27 g, were used. They were divided into groups with 6 animals in each. The animals in different groups received an i.m. injection of different doses of compounds tested (Table 1) and LD_{50} values were determined 2 hours after the injection of the organophosphate. In some cases, other routes of administration (i.v., s.c., p.o. and p.c.) were used. Organophosphates used were obtained from The Military Research Institute 070, Brno, (Czechoslovakia). The LD_{50} values were calculated according to the probit-logarithmic method. The calculations were done on a Hewlett-Packard calculator 9830 A.

The toxicities of the compounds studied are given in Table 1. It is clear from this Table that rats are more sensitive to these compounds than mice. It can also be observed that prolongation of the radical on the phosphorus head decreases the toxicity. A similar situation can be demonstrated for these dialkyl substituents on nitrogen. With the prolongation of alkyle chains on nitrogen, the toxicity also decreased (Table 1). For one compound (GV), the toxicities for different routes of administration were examined and

Table 1
Chemical formulae of the compounds examined



Designation	R^1	R^2	R^3	LD_{50} (μg/kg) with their 95% confidence limits	
				Mice	Rats
GV	Me	Me	$CH_2CH_2NMe_2$	30.5 (28-55)	17 (15.5-23.6)
GV 1	Et	Et	$CH_2CH_2NMe_2$	191 (180-203)	35.5 (33-38)
GV 2	Me	Me	$CH_2CH_2NEt_2$	162 (150-175)	94 (87-101)
GV 3	Et	Et	$CH_2CH_2NEt_2$	409 (378-441)	261 (238-286)
GV 4	Me	Me	$CH_2CH_2NMe_2$	105 (94-118)	59 (52-67)
GV 5	Et	Et	$CH_2CH_2NMe_2$	1222 (1118-1336)	261 (238-286)

are given in Table 2. It can be demonstrated that all routes of administration are very effective, including percutaneous administration.

If we compare the toxicities of well known compounds like G compounds (Clement *et al.* 1981, Bajgar 1991, Kovačevič and Maksimovic 1991) or V compounds (Bajgar 1991, Bajgar *et al.* 1982, Kovačevič and Maksimovic 1991), it can be concluded that the group of compounds examined is more toxic than G compounds and less toxic than V compounds.

Value of LD_{50} is a basic toxicological characteristic for toxic compounds. However, a comparison of this parameter is difficult because of many factors influencing this value, i.e. temperature (Kaliste-Korhonen *et al.* 1989), age (Shih *et al.* 1990), species (Maxwell and Brecht 1991), time of observation of animals (Bajgar 1991, Kaliste-Korhonen *et al.* 1989, Shih *et al.* 1990) etc.

It can be concluded from the results presented that the class of chemicals studied can be considered as extremaly toxic, comparable with other known chemical warfare agents. It is also clear that there exist differences of species sensitivities: in general, rats are more sensitive than mice. This is in agreement with literature data for soman (Clement *et al.* 1981, Kovačevič and Maksimovic 1991, Shih *et al.* 1990, Maxwell and Brecht 1991, Clement 1989, Doctor *et al.* 1991, Shapira *et al.* 1990), sarin (Kovačevič and Maksimovic 1991, Kaliste-Korhonen *et al.* 1989) and VX (Bajgar *et al.* 1982, Kovačevič and Maksimovic 1991, Clement 1989, Doctor *et al.* 1991) toxicities. Our toxicity data on mice (Bajgar 1991, Bajgar *et al.* 1982, Clement *et al.* 1981, Maxwell and Brecht 1991, Clement 1989, Doctor *et al.* 1991) or rats (Bajgar 1991, Bajgar *et al.* 1982, Kovačevič and Maksimovic 1991, Shih *et al.* 1990, Doctor *et al.* 1991) correspond to similar values described in the literature. Comparison of toxicities at various routes of administration [i.v.(Shapira *et al.* 1990), i.p. (Kaliste-Korhonen *et al.* 1989), i.m. (Bajgar1991, Bajgar *et al.* 1982, Shih *et al.* 1990,Doctor *et al.* 1991), s.c. (Clement *et al.* 1981, Kovačevič andMaksimovic 1991, Maxwell and Brecht 1991)] showed the highest toxicity for VX followed by soman and sarin, respectively. There is no information dealing with toxicity of GV and its derivatives.

Table 2
LD_{50} values of GV in mice and rats with various routes of administration

	LD_{50} (μg/kg) with their 95 % confidence limits	
Route of administration	Mice	Rats
i.v.	27.6 (25.6-29.4)	11 (8.5-17.6)
i.m.	30.5 (28-55)	17 (15.5-23.6)
s.c.	32 (29-53)	21 (18-26)
p.o.	222 (194-255)	190 (881-272)
p.c.	not tested	1366 (881-3138)

Though this class of compounds is not stable – one compound (GV) is spontaneously isomerized to less toxic aziridine derivative (Halámek *et al.* 1988) – its possible use as CW cannot be excluded especially in binary form. Preliminary determination of anticholinesterase activity (lying in the range of 0.2-0.5 $\mu g.ml^{-1}$) (Halámek *et al.* 1988) corresponds well to its toxicity. It can be pointed out that this class of chemicals is not included in the present lists of chemicals (Schedules) to be controlled within the scope of the future Chemical Weapons Convention (Document 1992). Our observations support the requirement for further toxicological study of this group, especially in the light of inhibition of cholinesterase activity *in vitro* and *in vivo* and therapeutic possibilities: with regard to the chemical structure, it cannot be excluded that the therapeutic efficacy of the usual antidotes (atropine and reactivator) will be very limited. This could be caused by the character of cholinesterase inhibition by GV as was demonstrated by our previous results (Bajgar 1992).

Acknowledgements

The authors wish to thank to Mrs. E. Vodáková for skilled technical assistance and Mrs. H. Roobová for preparation of the manuscript.

References

BAJGAR J.: The influence of inhibitors and other factors on cholinesterases. Sborník věd. prací LF UK Hradec Králové **34:** 1–77, 1991.

BAJGAR J., HRDINA V., FUSEK J., PATOČKA J.: Correlating the effects of highly toxic organophosphates on animals and humans (in Czech). *Čas. Lék. čes.* **121:** 1095–1098, 1982.

BAJGAR J.: Biological monitoring of exposure to nerve agents. Accepted by *Brit. J. Ind. Med.* **49:** in press, 1992.

CLEMENT J.G.: Survivors of soman poisoning: recovery of the soman LD50 to control value in the presence of extensive acetylcholinesterase inhibition. *Arch. Toxicol.* **63:** 150–154, 1989.

CLEMENT J.G., HAND B.T., SHILHOFF J.D.: Differences in the toxicity of soman in various strains of mice. *Fundam. Appl. Toxicol.* **1:** 419–420, 1981.

DOCTOR B.P., RAVEH L., WOLFE A.D., MAXWELL D.M., ASHANI Y.: Enzymes as pretreatment drugs for organophosphate toxicity. *Neurosci. Behav. Rev.* **15:** 123–128, 1991.

DOCUMENT of the Conference on Disarmament, CD/1116, Geneva, 1992.

HALÁMEK E., TUŠAROVÁ I., KOBLIHA Z., SOUČEK J., FÖLDEŠI V.: Identification of the product of spontaneous decomposition of model compound with intermediate volatility (in Czech). Sborník VVŠ PV Vyškov **13:** 2–16, 1988.

KALISTE-KORHONEN E., RYHÄNEN R., YLITALO P., HÄNNINE O.: Cold exposure decreases the effectiveness of atropine-oxime treatment in organophosphate intoxication in rats and mice. *Gen. Pharmacol.* **20:** 805–809, 1989.

KOVAČEVIČ V., MAKSIMOVIC M.: Protective effects of mixture of oximes in poisoning by nerve chemical warfare agents. *Acta Pharm. Yugosl.* **41:** 75–78, 1991.

MAXWELL D.M., BRECHT K.M.: The role of carboxylesterase in species variation of oxime protection against soman. *Neurosci. Behav. Res.* **15:** 135–139, 1991.

SHAPIRA S., KADAR T., COHEN G., CHAPMAN S., RAVEH L.: Effects of CBDF and MEPQ on the toxicity and distribution of [3H] - soman in mice. *Arch. Toxicol.* **64:** 663–668, 1990.

SHIH T.M., PENETAR D.M., MC DONOUGH J.H., ROMANO J.A., KING J.M.: Age-related differences in soman toxicity and in blood and brain regional cholinesterase activity. *Brain Res. Bull.* **24:** 429–436, 1990.

Reprint Requests

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