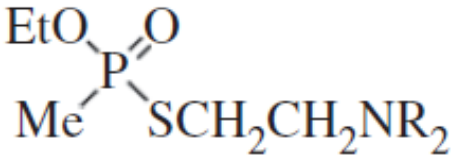
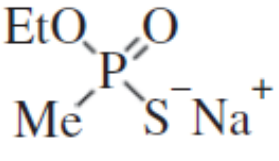
Toxicity and medical countermeasure studies on nerve agents VX and VM

Helen Rice, Chris Dalton, Matt Price, Stuart Graham, Christopher Green, John Jenner, Helen Groombridge and Christopher Timperley

Nerve agents of Syrian Arab Republic



VM R = Et or **VX** R = *i* – Pr



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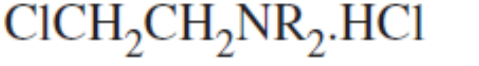
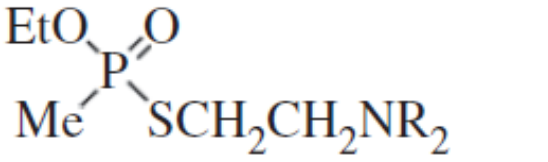


2 R = Et or **3** R = *i* – Pr

Nerve agents of Syrian Arab Republic



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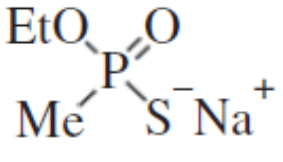

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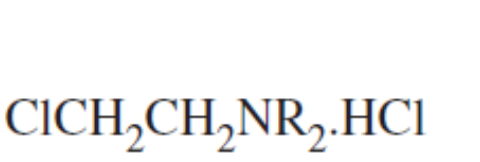

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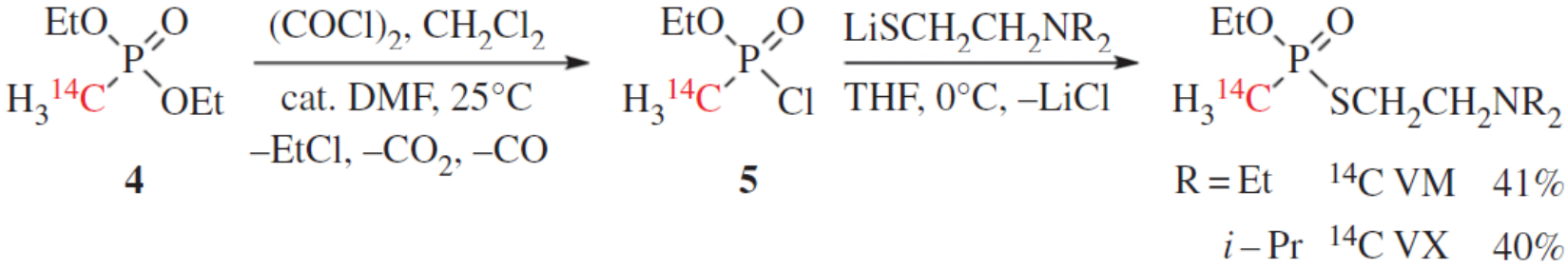
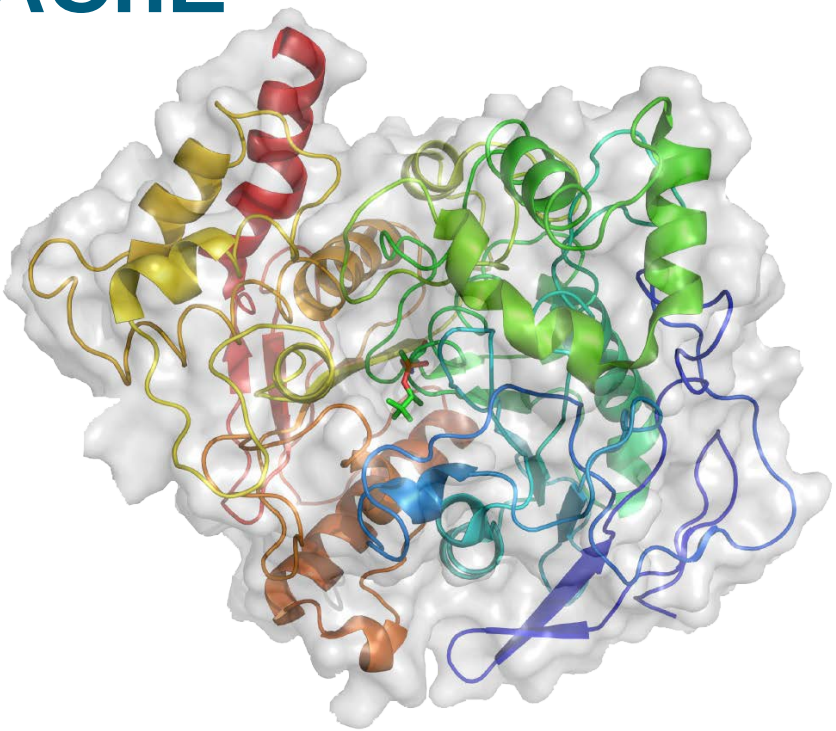
Synthesis of ^{14}C-labelled VM and VX



4 **5**

AChE



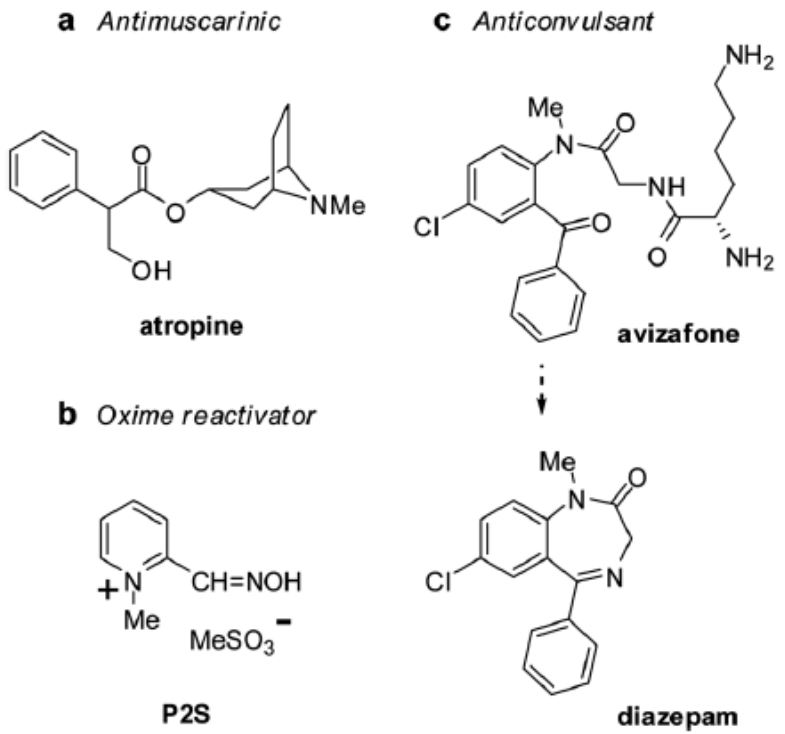
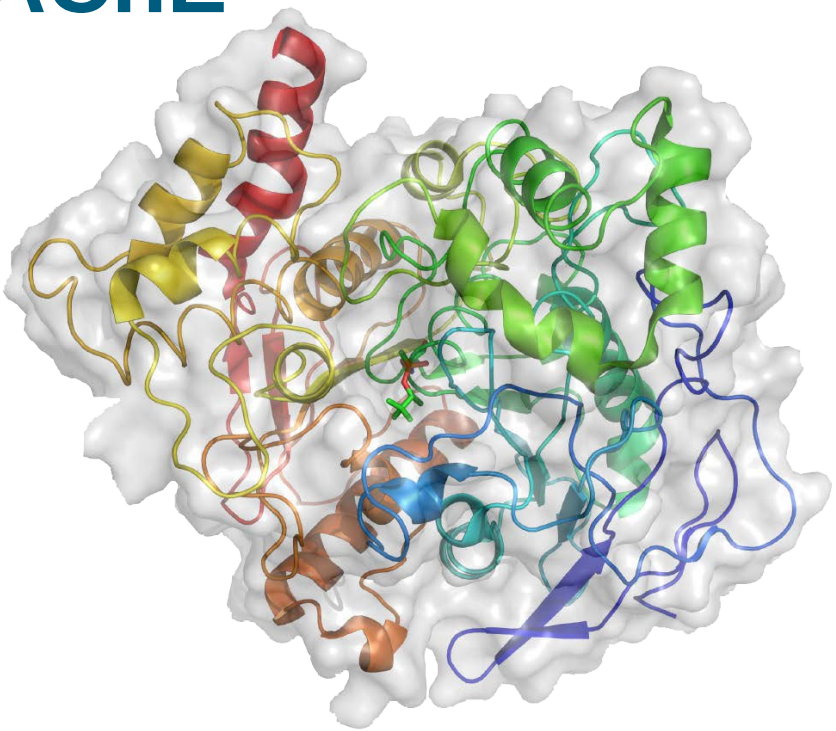
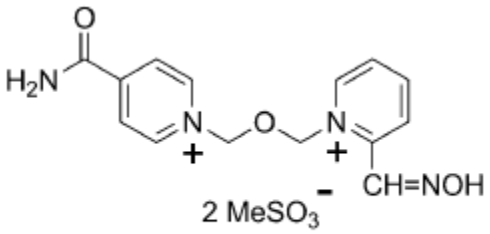


Fig. 1 Components of the British ComboPen® used to treat nerve agent poisoning. The triple therapy comprises (**a**) an antimuscarinic, (**b**) an oxime reactivator, and (**c**) a water-soluble pro-drug, avizafone, which is converted by enzymes in the blood to the anticonvulsant drug diazepam.

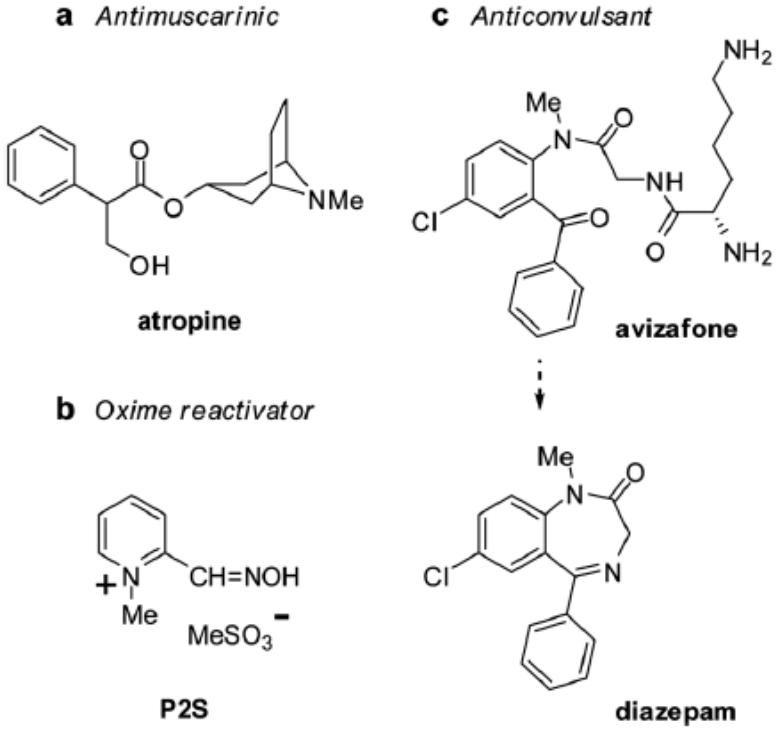
C M Timperley et al., Med. Chem. Commun. 2012, 3, 352-356

AChE





HI-6 di(methanesulfonate)



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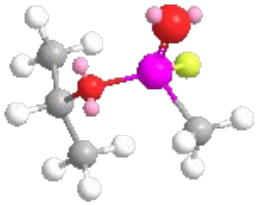
Nerve agent poisoning



Minutes

SIGNS

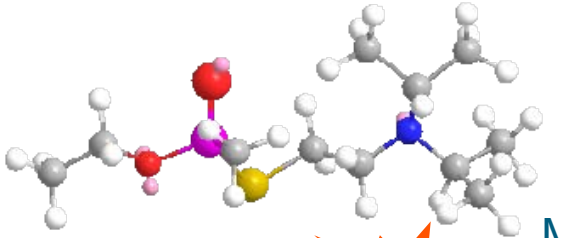
Inhaled volatile agent e.g. sarin

All Agent Absorbed

Therapy

VX Human LD_{50} No treatment

Photo: Ed Clarkson, USAMRICD



Minutes

Hours

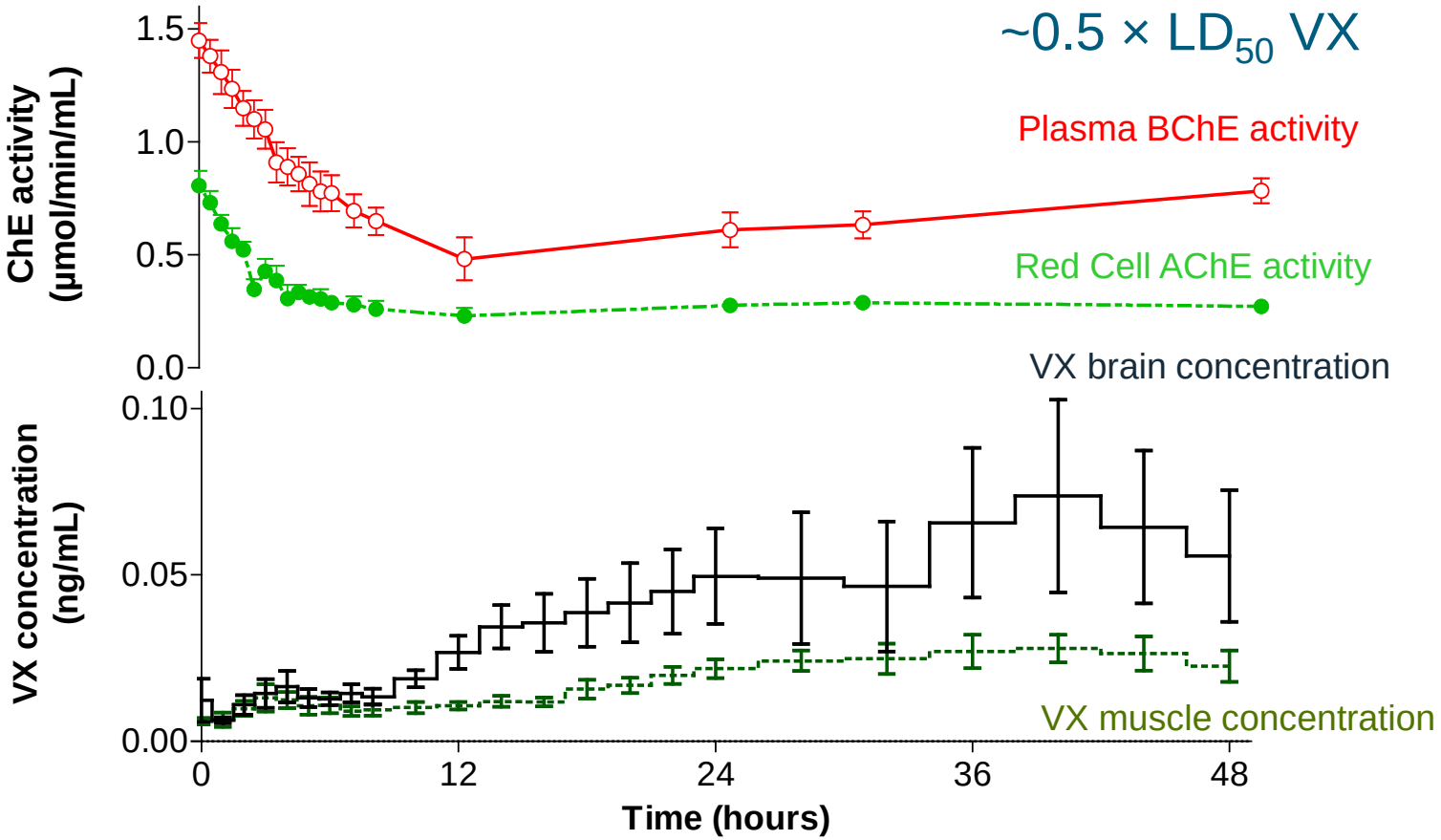
SIGNS

Persistent liquid agent on skin e.g. VX

Therapy

Agent continues to be absorbed

PK & PD for percutaneous VX

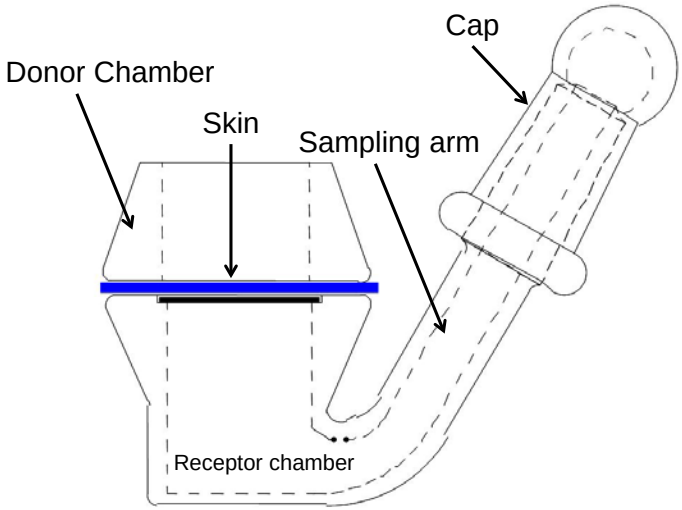


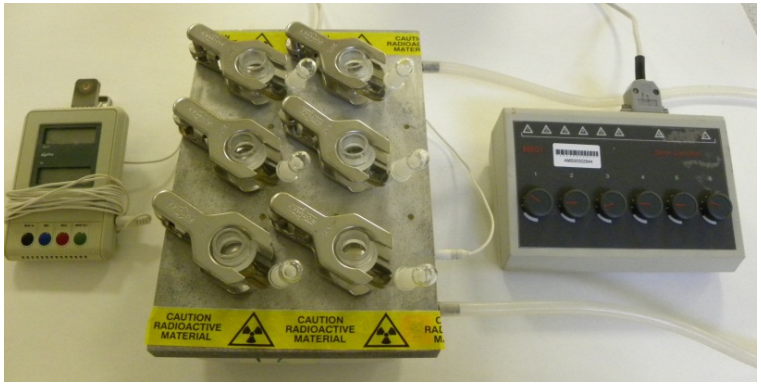
n = 7

H Mumford et al., Chem. Bio. Interact., 2013, 203, 160

Measurement of agent diffusion *in vitro*

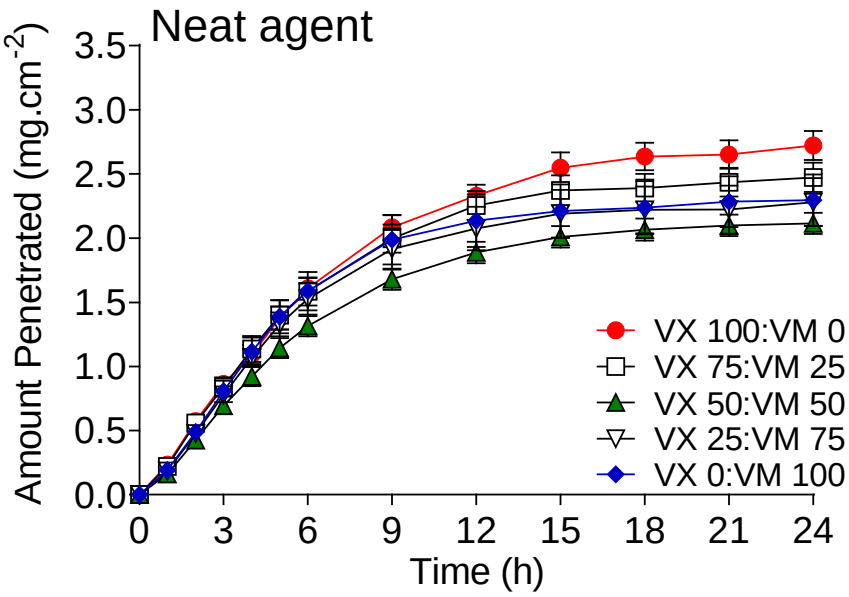
- Franz type diffusion cell
- application volume 10 µL
- area 2.54 cm²
- temperature 32 °C
- receptor volume ~5 mL
- stirred continuously

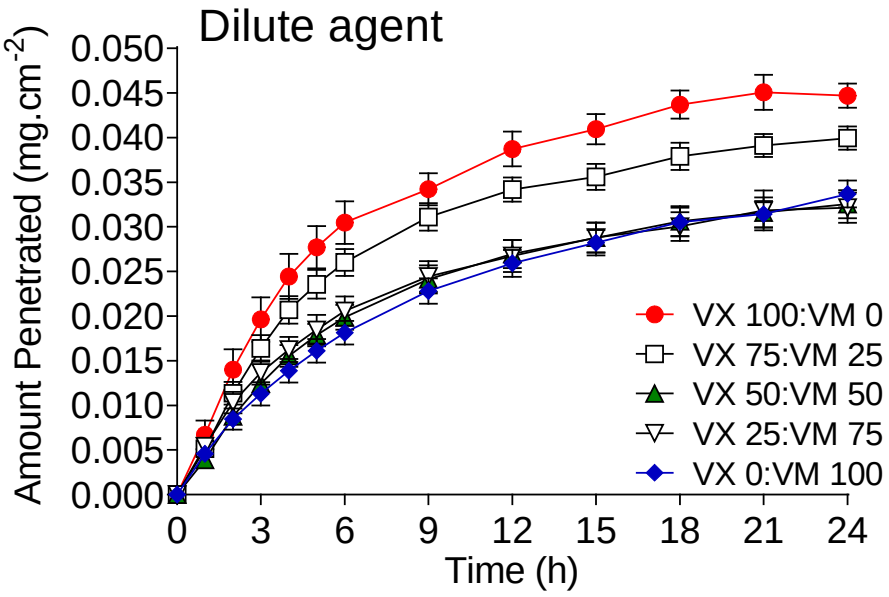




Guinea-pig skin

No significant difference between combined penetration rates of either neat or dilute agent mixtures was observed

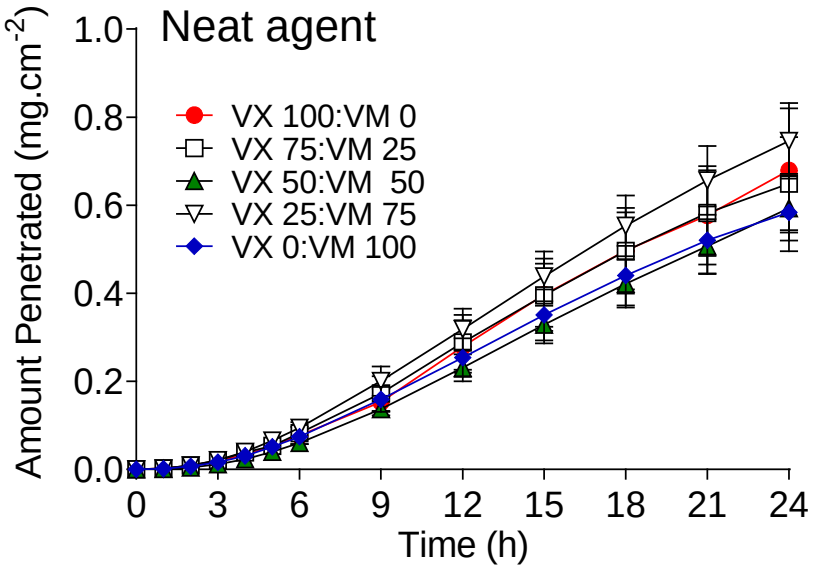


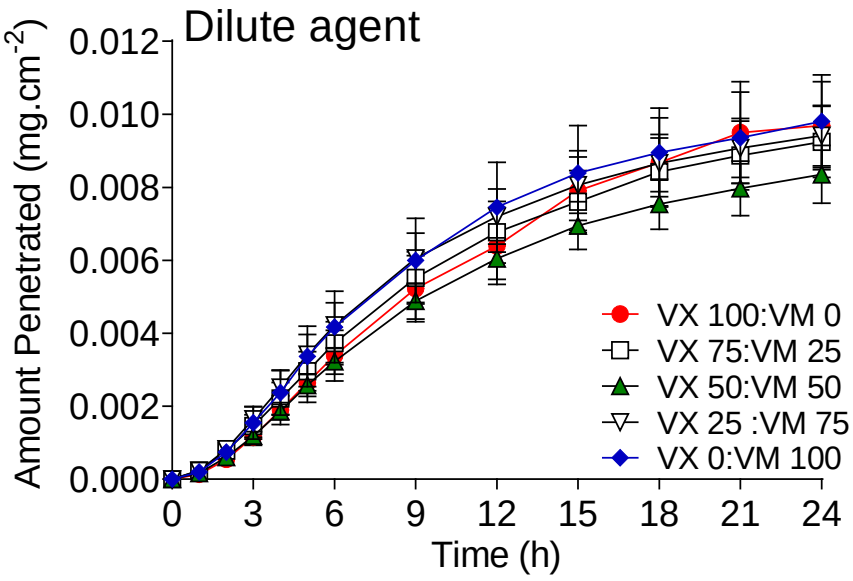


*H Rice et al., Proc Roy Soc A. 2015, **471**, 20140891*

Pig skin

No significant difference between combined penetration rates of either neat or dilute agent mixtures was observed





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Historical results comparison

	Penetration of ^{14}C VX $\mu g.cm^{-2}.h^{-1}$ (mean ± SD)		
	Guinea-pig skin	Pig skin	Human skin
Previous study *	3.69 ± 0.72	0.73 ± 0.35	1.01 ± 0.21
Current study	5.23 ± 0.95	0.74 ± 0.43	n.d.

** C Dalton et al., Toxicology in vitro 2006, 20, 1532*

n.d. = not determined

Experimental design: *in vivo*

Dunkin-Hartley guinea-pigs: male, conscious

Toxicity - 24 h percutaneous LD_{50}

- VM alone and VM-VX mixtures (3 different proportions)
- agent diluted in isopropanol and applied to clipped skin
- left unoccluded and no decontamination

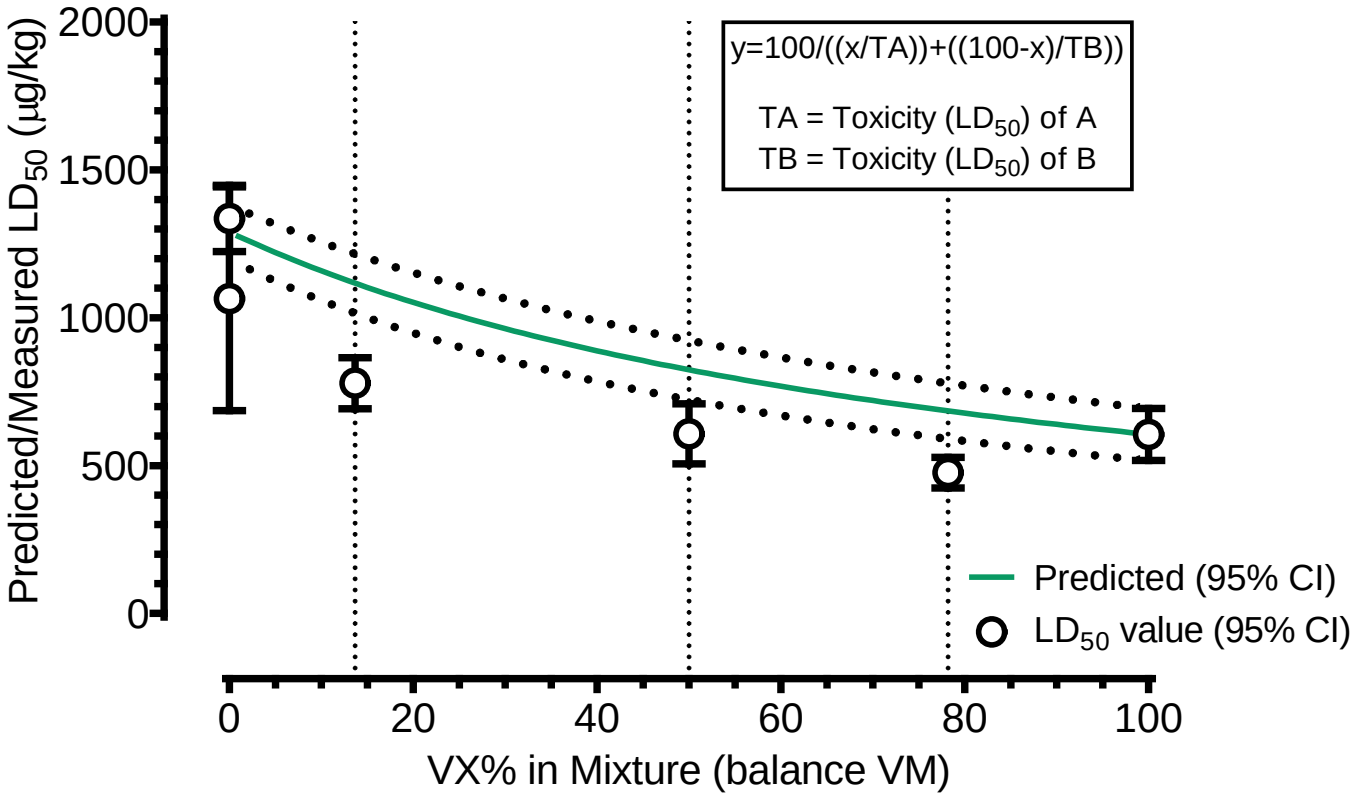
MedCM study - 2 × LD_{50} challenge

- VM 2.388 mg·kg^{-1} or VX 1.226 mg·kg^{-1},
- Therapy (i.m.) on signs of cholinergic poisoning and worsening signs of poisoning (max. 3 doses over 9 h)

Results: 24 h LD_{50}

LD_{50} VX alone 0.613 $mg.kg^{-1}$

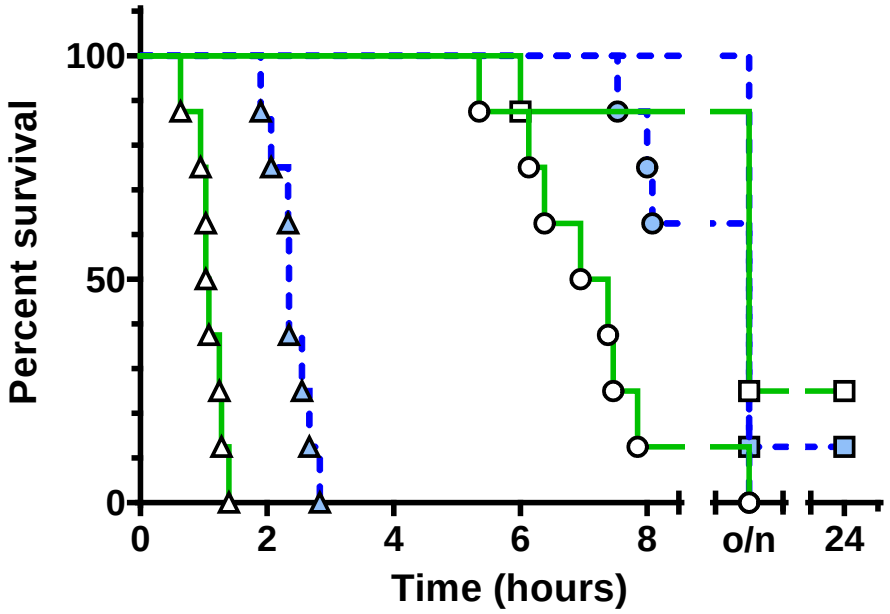
LD_{50} VM alone 1.290 $mg.kg^{-1}$



H Rice et al., Proc Roy Soc A. 2015, 471, 20140891

Therapy (3 ×) of atropine, avizafone and either P2S or HI-6 did not protect guinea pigs from 2 × LD_{50} of either VM or VX

- Therapy on signs of poisoning and worsening signs of poisoning prolonged the times to death compared to saline-treated controls
- At 6 h, survival was significantly higher in treated groups than saline controls, for both nerve agents and either treatment



Therapy

oxime	30.0 $mg.kg^{-1}$
atropine	17.4 $mg.kg^{-1}$
avizafone	3.14 $mg.kg^{-1}$

oxime = P2S or HI6 DMS

*H Rice et al., Proc. Roy. Soc. A. 2015, **471**, 20140891*

Conclusions

VM is approximately half as toxic as VX by the p.c. route

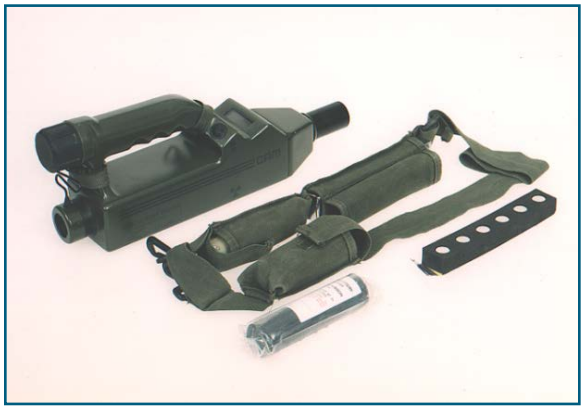
In-service (P2S) or future oxime (HI-6) MedCM did not fully protect guinea-pigs but extended time to death to > 5-6 h

Mixtures of VM + VX do not penetrate skin faster than the individual agent

Mixtures of VM + VX were not more toxic than the pure agent

There is no requirement to handle mixtures of VM + VX in demilitarisation operations differently from the pure materials

MedCM are part of a system of



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Toxicity and medical countermeasure studies on the organophosphorus nerve agents VM and VX

Helen Rice[1], Christopher H. Dalton[1], Matthew E. Price[1], Stuart J. Graham[1], A. Christopher Green[1], John Jenner[1], Helen J. Groombridge[2] and Christopher M. Timperley[2]

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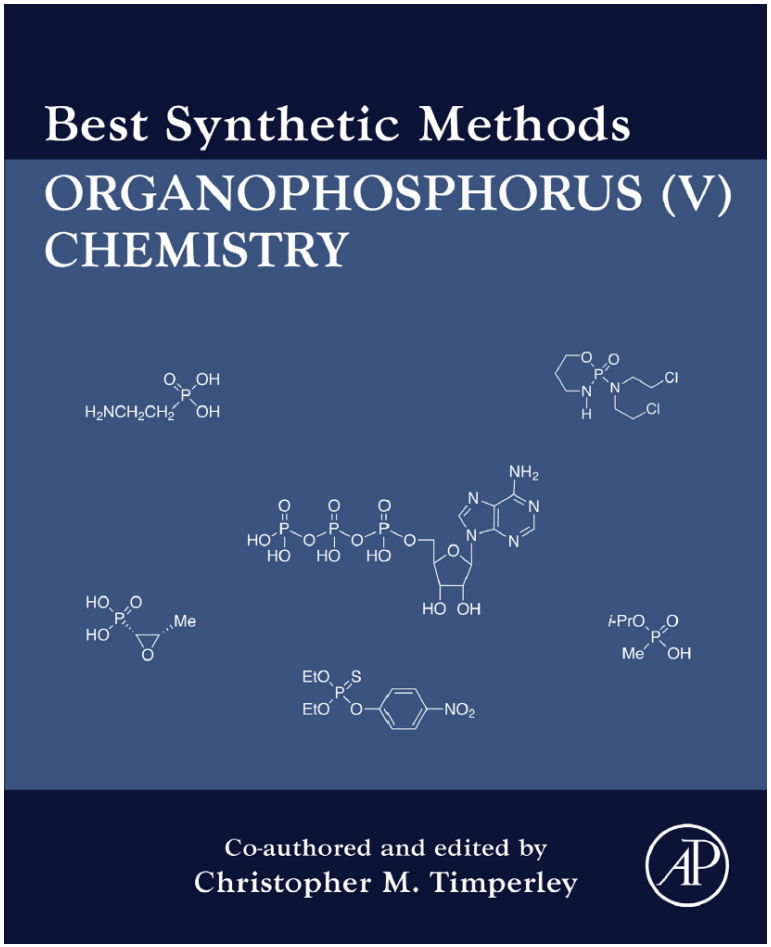
To support the effort to eliminate the Syrian Arab Republic chemical weapons stockpile safely, there was a requirement to provide scientific advice based on experimentally derived information on both toxicity and medical countermeasures (MedCM) in the event of exposure to VM, VX or VM–VX mixtures. Complementary *in vitro* and *in vivo* studies were undertaken to inform that advice. The penetration rate of neat VM was not significantly different from that of neat VX, through either guinea pig or pig skin *in vitro*. The presence of VX did not affect the penetration rate of VM in mixtures of various proportions. A lethal dose of VM was approximately twice that of VX in guinea pigs poisoned via the percutaneous route. There was no interaction in mixed agent solutions which altered the *in vivo* toxicity of the agents. Percutaneous poisoning by VM responded to treatment with standard MedCM, although complete protection was not achieved.

1. Introduction

In October 2013, the Organisation for the Prohibition of Chemical Weapons (OPCW) was awarded the Nobel Peace Prize for its 'extensive efforts to eliminate chemical weapons' [1]. This, combined with efforts to destroy



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- Animal care staff

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