Therapy of Nerve Agent Poisoning – Up-date and New Approaches

H. Thiermann and F. Worek

Bundeswehr Institute of Pharmacology and Toxicology,
80937 Munich, Germany

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Nerve Agents and other Organophosphorus Compounds

G - Agents

Tabun (GA), Sarin (GB), Cyclosarin (GF), Soman (GD) (and Analogs)

V - Agents

VX, Russian VX (VR), Chinese VX (CVX) (and Analogs)

OP- Pesticides

Diethyl-type, Dimethyl-type, hundreds of other compounds!

Characteristics of Poisoning by Nerve Agents

Generally assumed: patients exposed up to 5x LD_{50}

Inhalative G-Type Nerve Agent Poisoning

- Immediate onset of cholinergic crisis
- Short persistence of nerve agent in the body

Percutaneous V-type Nerve Agent Poisoning

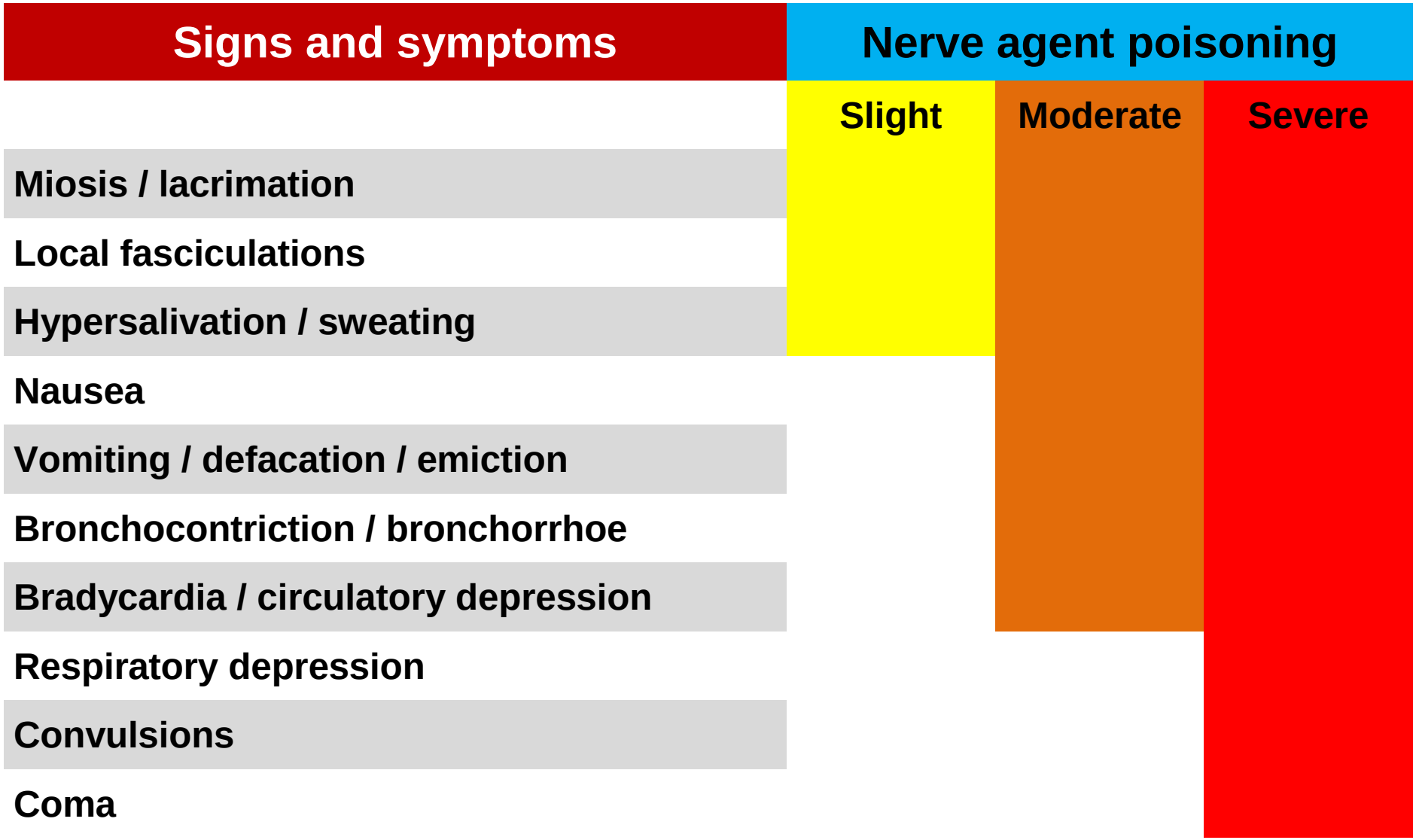
- Prolonged onset of cholinergic crisis
- Persistence of nerve agent over several days in the body

Effects of Organophosphorus Compounds

Mechanism:

- inhibition of AChE
- accumulation of ACh
- disturbance of cholinergic functions

Clinical Challenge of Nerve Agent Poisoning



Worek et al. Arch Toxicol 2016

Effects of Organophosphorus Compounds

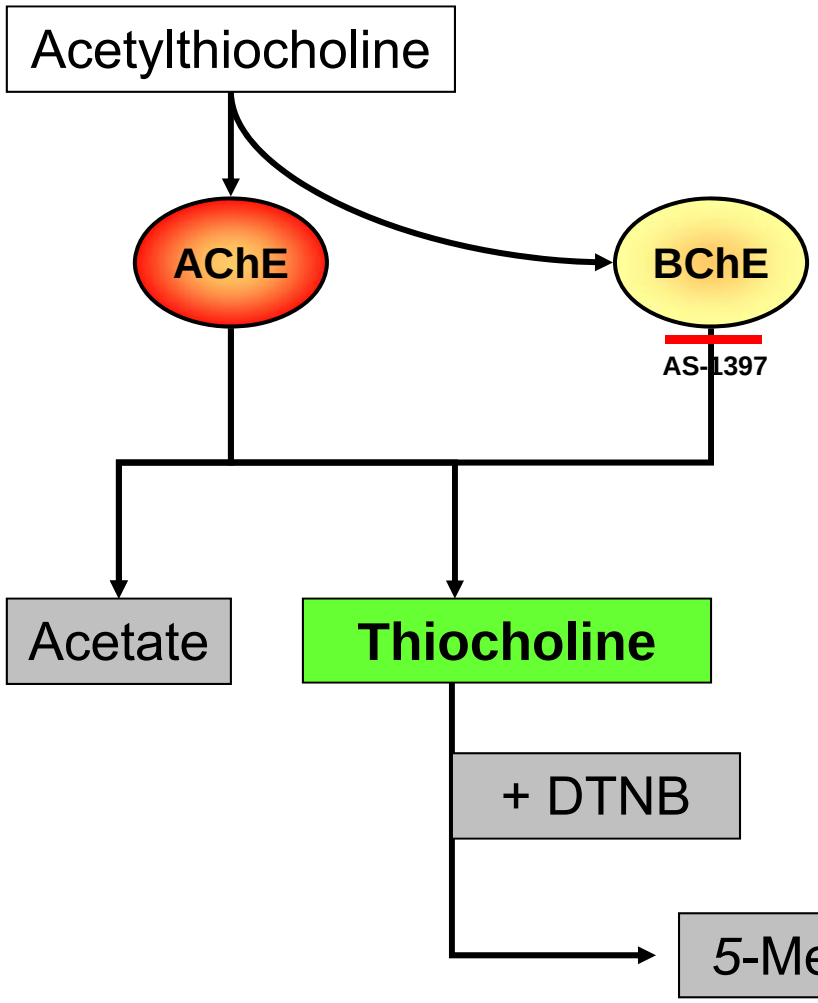
Mechanism:

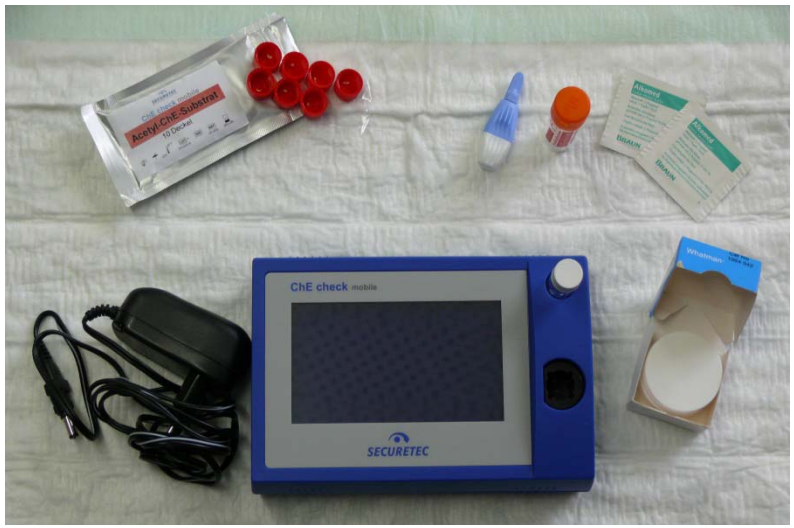
- inhibition of AChE
- accumulation of ACh
- disturbance of cholinergic functions

Life threatening effects:

- bronchoconstriction/ bronchorrhoea (muscarine receptors)
- central respiratory arrest (muscarine and nicotine receptors)
- peripheral respiratory arrest (nicotine receptors)

ChE Check Mobile for on-site Confirmation of the Clinical Diagnosis





Ellman et al. Biochem Pharmacol 1961

Med C- Special Diagnostics/ Reconnaissance / Verification and Analysis

Biomedical verification of exposure to chemical agents

- chemical warfare agents
- degradation products
- metabolites and adducts

Peculiarities in Treatment of Chemical Agent Poisoning

Self – Protection; Working under aggravated conditions;
Protection of medical units; Latency period.
Decontamination of victims with signs and symptoms

ouvry.com

Compare: Thiermann et al. Chem Biol Interact 2013;
Rosman et al. Annal Intern Med 2014

Atropine for Nerve Agent Poisoning

Military doctrine: autoinjectors for self and buddy aid

Enhanced first aid and clinical care by medical personnel)

Aggressive atropine dosing

Recommended German regimen

2 mg, i.m. followed by consecutive doubling the dose (4 – 8 – 16 - 32 mg) and finally infusion at field hospital

Criteria

Clear chest on auscultation; heart rate >80 beats/min; pupils no longer pinpoint; dry skin (axilla); systolic blood pressure >80 mmHg

Thiermann et al. Chem Biol Interact, 2013; according to Eddleston et al. Crit Care 2004 and J Toxicol ClinToxicol 2004.
For review see: Thiermann et al. Treatment of Nerve Agent poisoning In:Chemical Warfare Toxicology, Volume 2: Management of Poisoning,

Development of Oximes

Synthesis of thousands of oximes since the early 1950ies

- Monopyridinium oximes
- Bispyridinium oximes
- Asymmetric oximes
- Substituted pyridinium oximes
- Uncharged oximes

2017

Oximes in use or in advanced development

2-PAM	**1955**
TMB-4	**1958**
MMB-4	**1959**
Obidoxime	**1959**
HI-6	**1968**

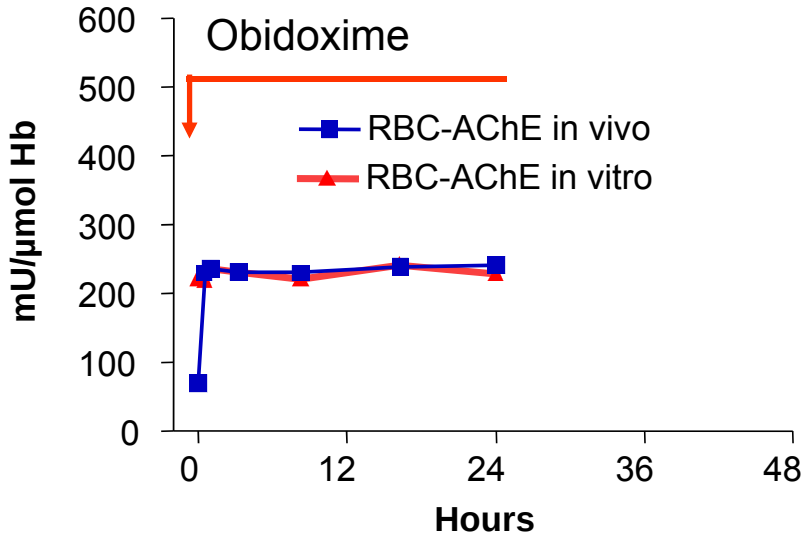
Effects of Obidoxime in a Patient with Parathion Poisoning

Patient: 56-year old, female

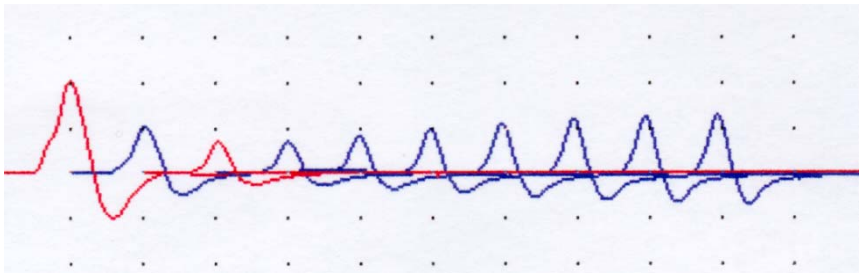
Emergency situation: Unconscious, cardiovascular resuscitation

Clinical course: As the clinical course situation remained critical for about 4 days without obidoxime, the regimen was started

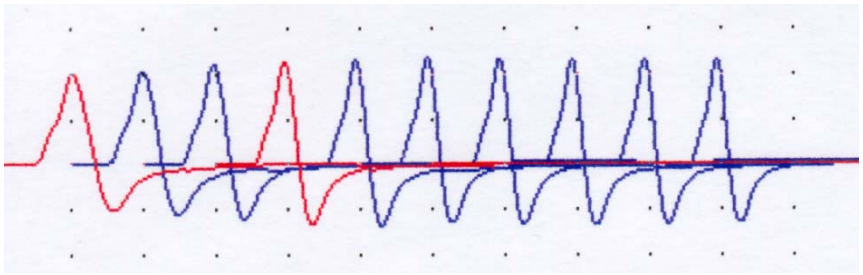
AChE activity of red blood cells



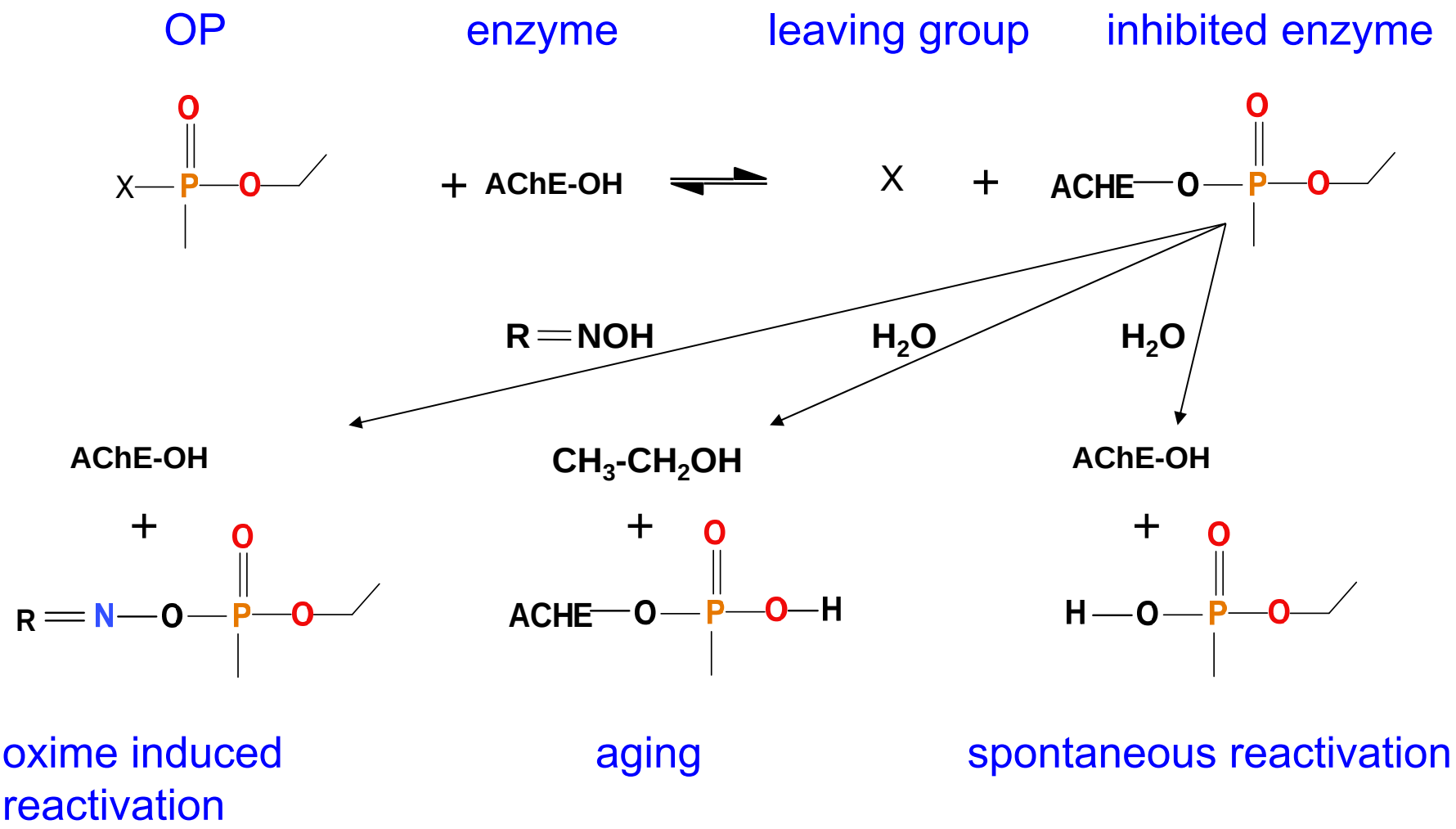
Neuromuscular transmission
prior to Obidoxime



after Obidoxime



Reactions Occurring at AChE with an OP and an Oxime



Kinetic-Based Computer Model

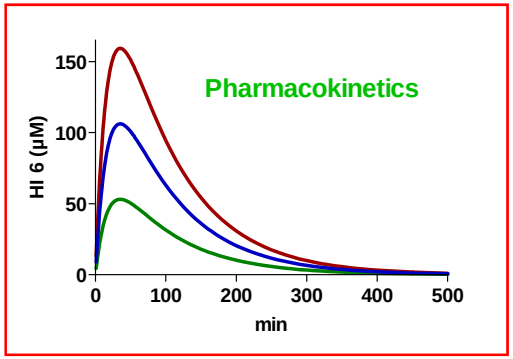
(1) $[E] + [OP] \xrightarrow{k_i} [EP]$

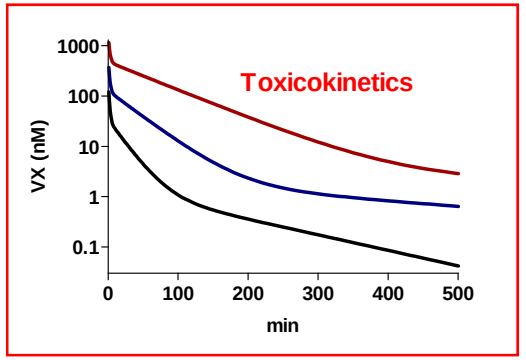
(2) $[EP] + H_2O \xrightarrow{k_a} [EP']$

(3) $[EP] + H_2O \xrightarrow{k_s} [E] + [P]$

(4) $[EP] + [OX] \underset{}{\overset{K_D}{\rightleftharpoons}} [EPOX] \xrightarrow{k_r} [E] + [POX]$

$$k_{obs} = \frac{k_r * [OX]}{K_D + [OX]}$$





$$\frac{d[E]}{dt} = -k_i * [OP] * [E] + k_s * [EP] + k_{obs} * [EP + EPOX]$$

$$\frac{d[EP]}{dt} = k_i * [OP] * [E] - k_s * [EP] - k_{obs} * [EP + EPOX] - k_a * [EP]$$

Worek et al. TAP 2005

The Use of Kinetic Constants, Pharmacokinetic and Toxicokinetic Data for Prediction of AChE-Activity in Human Poisoning

Patient:
A 45-year old, male

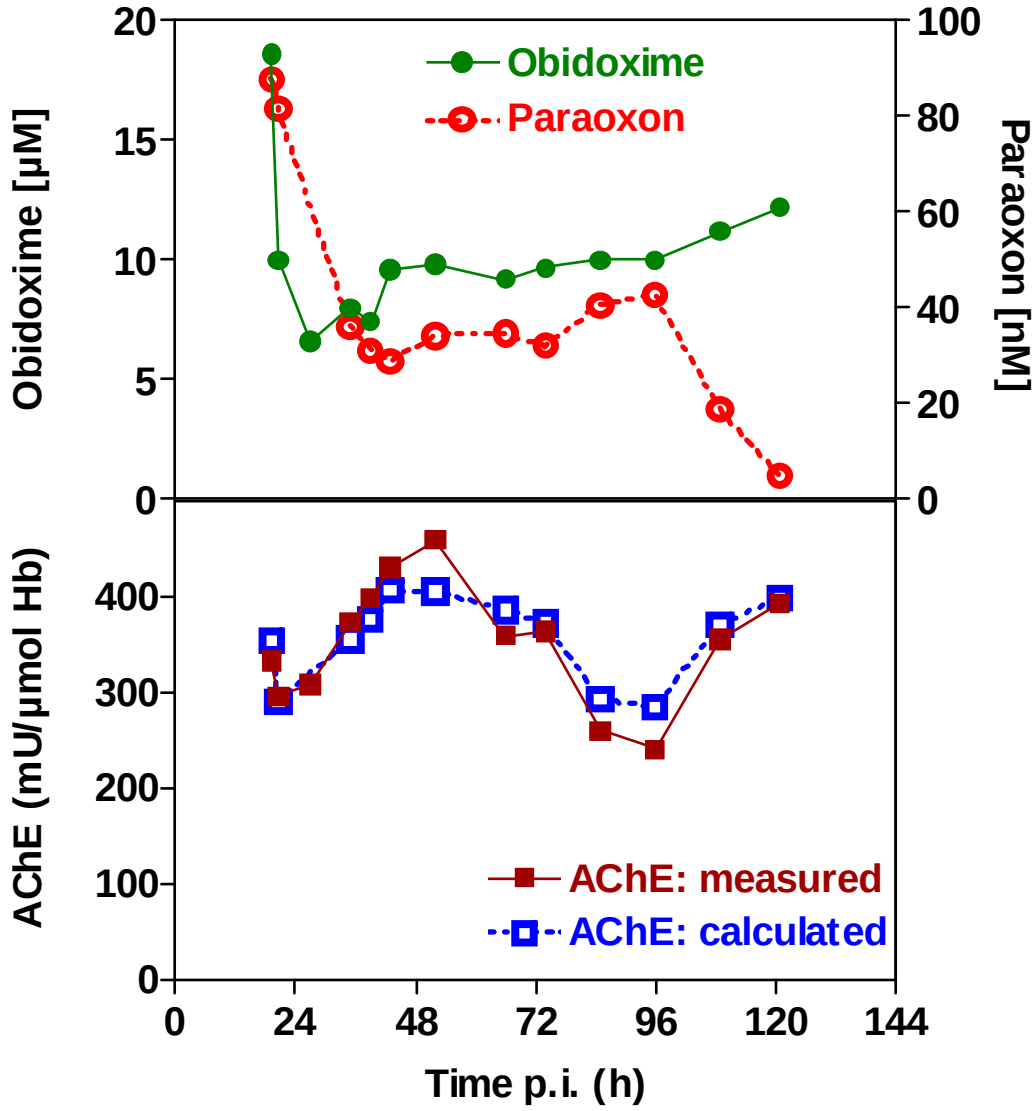
Emergency situation:

Unconscious, severe signs and symptoms of cholinergic crisis.
1.5 mg of atropine, intubation and initiation of artificial ventilation.

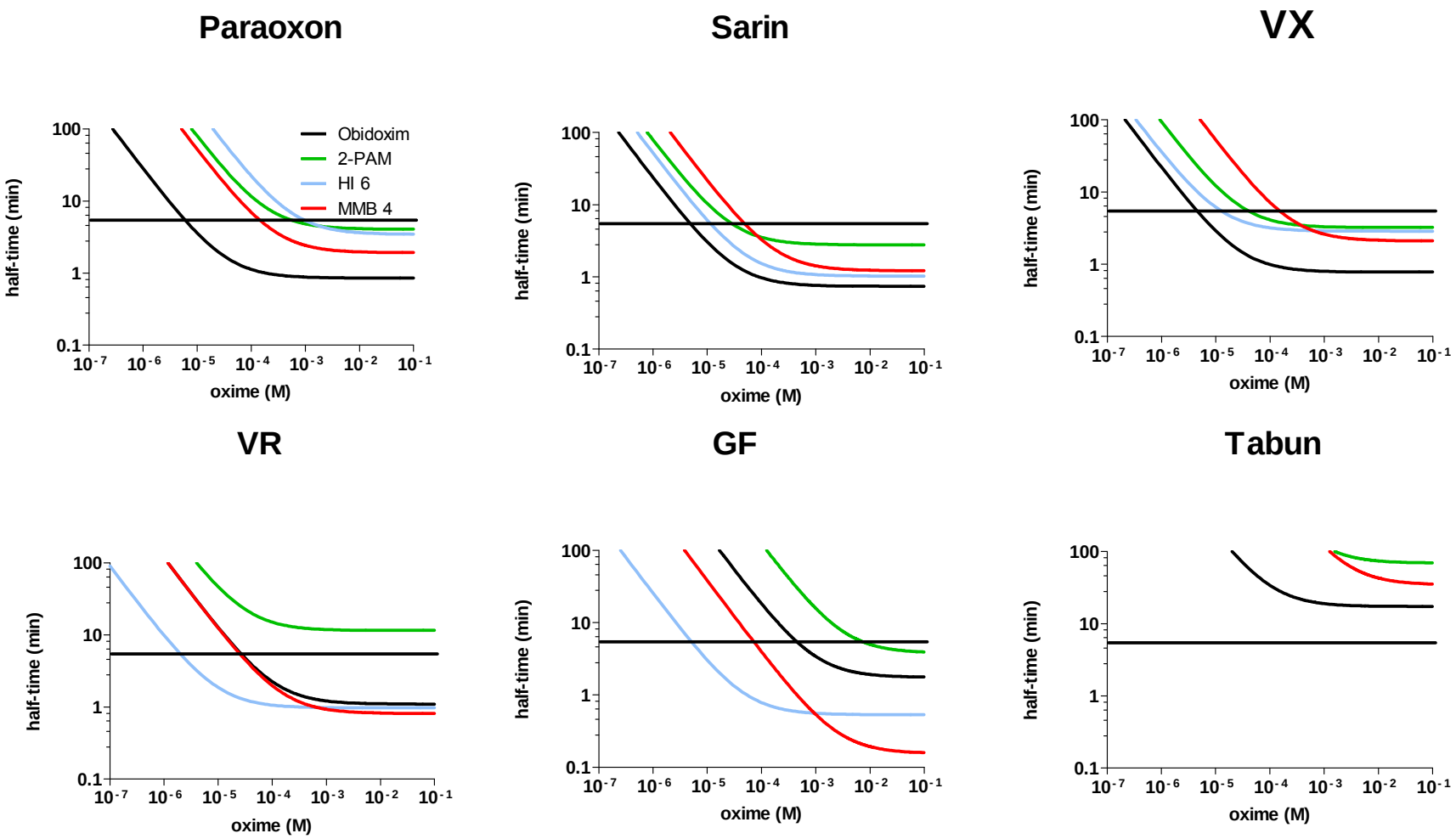
Clinical course:

2 bolus doses of obidoxime together with an atropine infusion at the local hospital. Transfer to the ICU of Technical University, Munich.
The patient recovered uneventfully.

Worek et al. TAP 2005



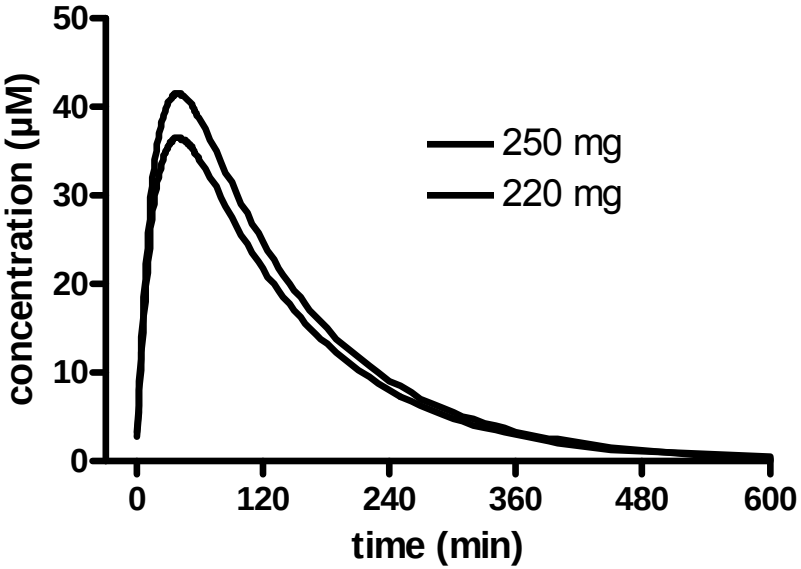
Reactivation of OP Inhibited AChE with Oximes



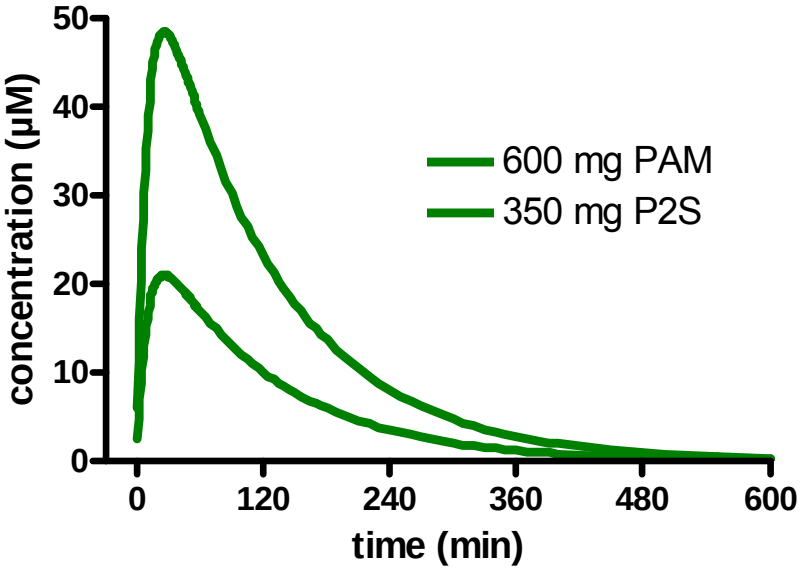
Compare: Thiermann et al. Chem Biol Interact 2013 and Tox Lett 1999; modified by using data from Aurbek et al. Toxicolgy 2006; Bartling et al. Toxicolgy 2007 Worek et al. Tox Lett 2011

Calculated Plasma Concentration of Oximes after i.m. Injection

Obidoxime



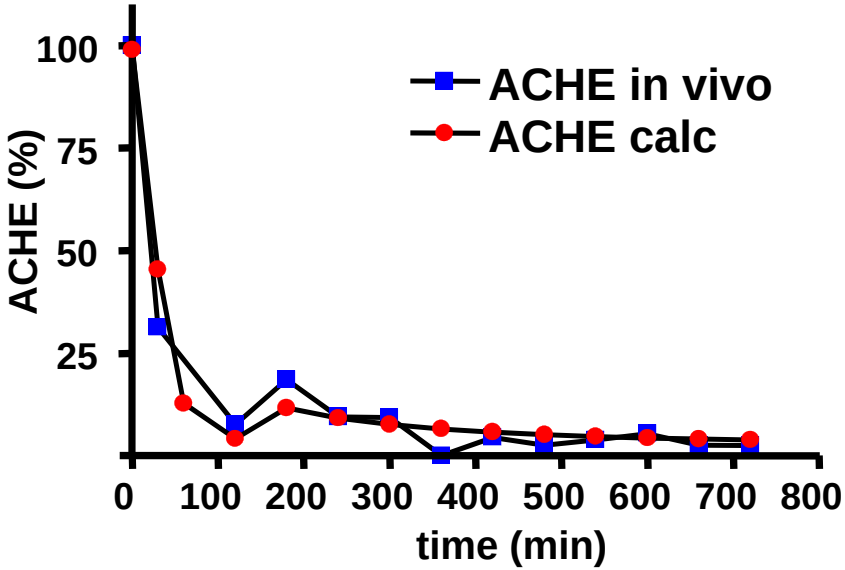
Pralidoxime

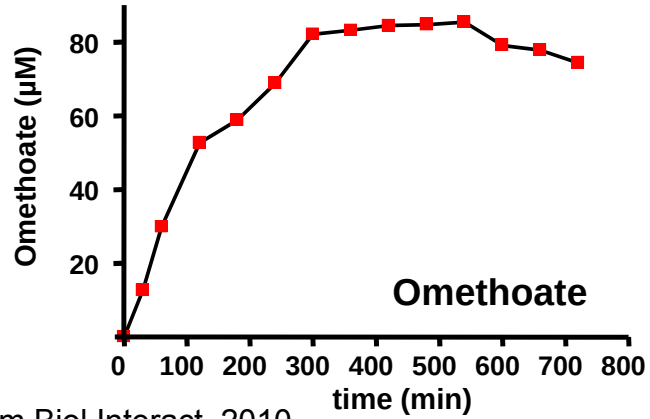


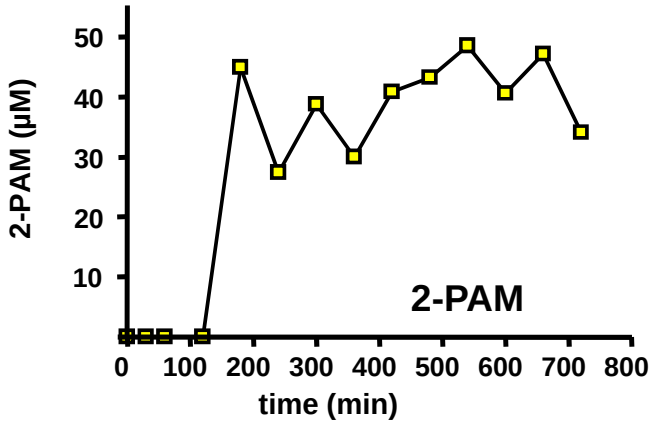
Calculated according to Erdmann et al. Dtsch Med Wschr 1965 and Sidell et al. J Pharm Sci 1972;

Translation of Clinical Findings from Human Poisoning to an Animal Model

Poisoning by dimethoate (oral) and treatment with 2-PAM





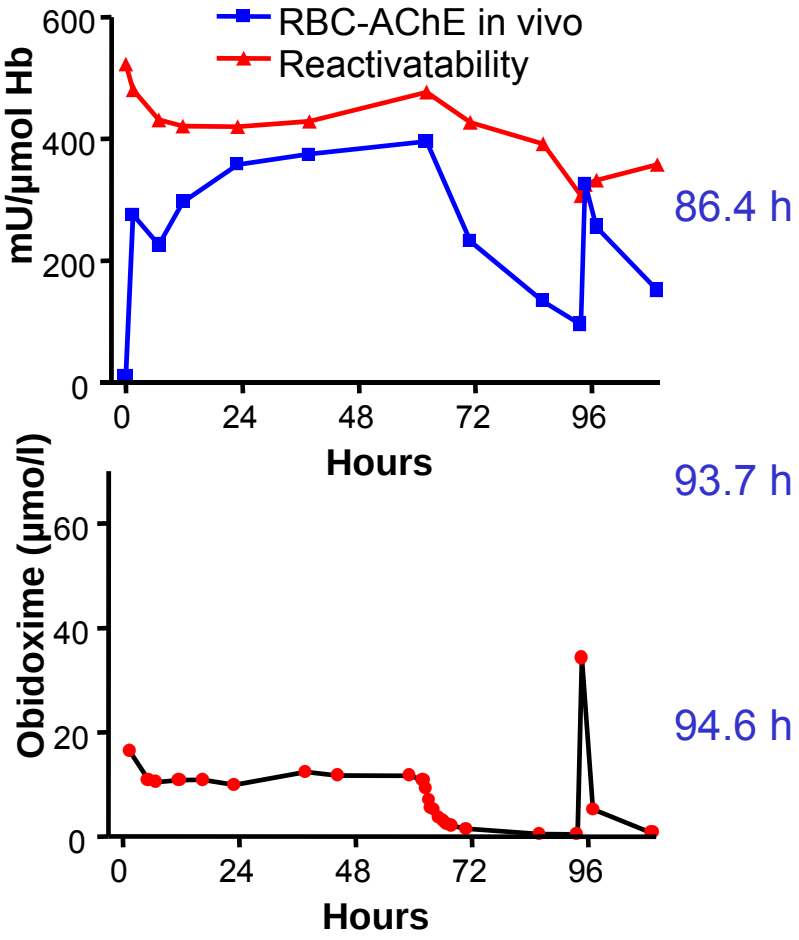


Worek et al. Chem Biol Interact. 2010

Is there a Correlation between RBC-AChE-Activity and Clinical Status?

A parathion poisoned patient was treated with obidoxime.

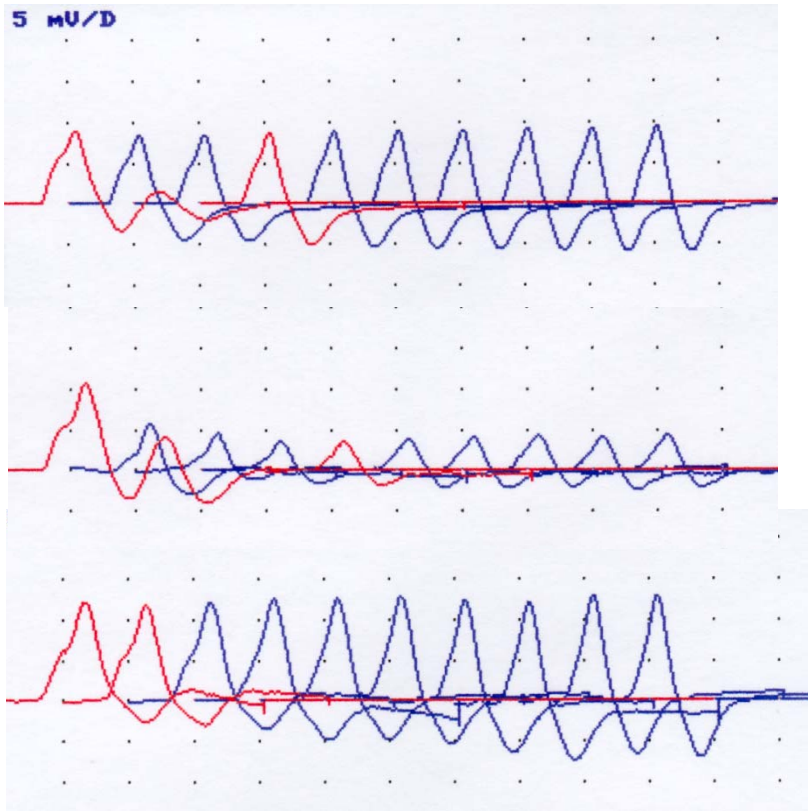
The cholinesterase status and NMT were monitored during treatment at the ICU.



86.4 h

93.7 h

94.6 h



Eyer et al. Clincal Toxicology 2009

Patient-Oriented Therapy by Using the Cholinesterase Status

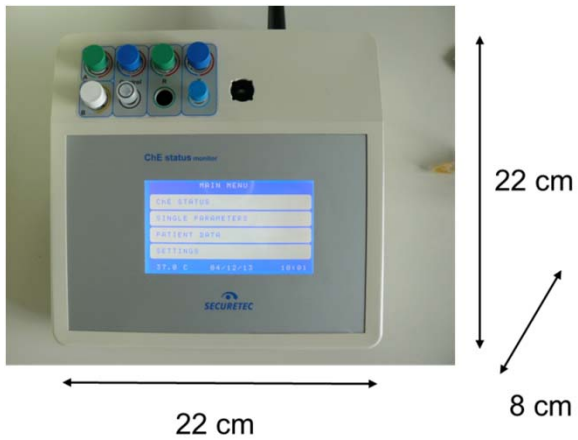
Cholinesterase Status:

- AChE and BChE activity
- Reactivatability of AChE
  - Reactivatability at all
  - Aging
- Persistence of poison load

Treatment:

- Appropriate oxime
- Oxime as long as substantial reactivation may be expected
  - Given reactivatability
  - Persistence of active poison
- Oxime stop:
  - Reactivation achieved and no poison load
  - Complete aging
  - Increase of BChE activity

ChE Status Monitor



Translation of Findings from OP-Pesticide-Poisoning to Nerve Agent-Poisoning

Male York-Landrace cross swine (about 20 kg)

Poisoning:

3 x LD_{50} VX, p.c.

Treatment:

HI-6 (12.7 mg/kg) / Atropine sulfate (0.05 mg/kg) i.m., according to signs and symptoms

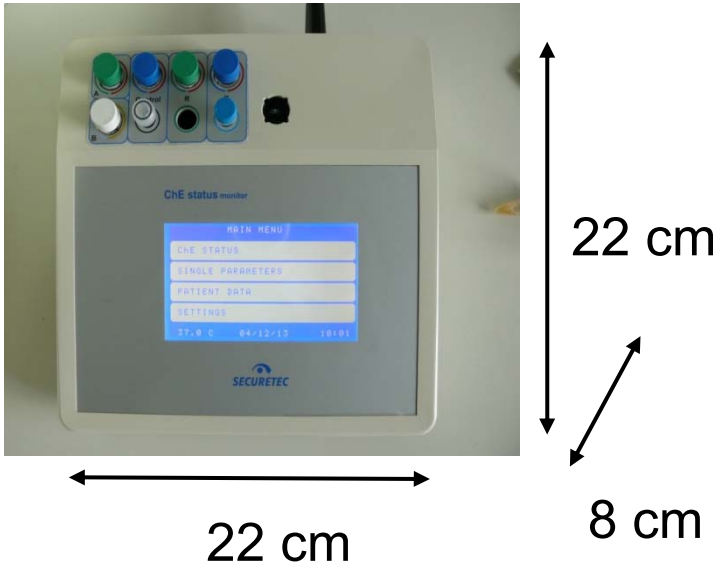
Field laboratory diagnosis:

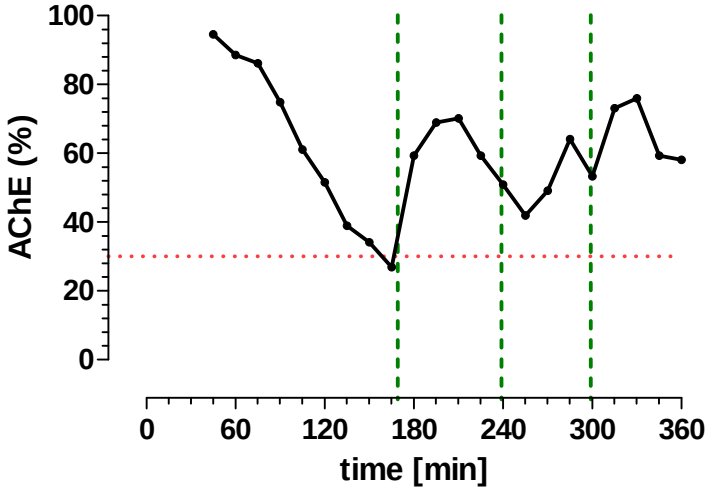
AChE activity on-site

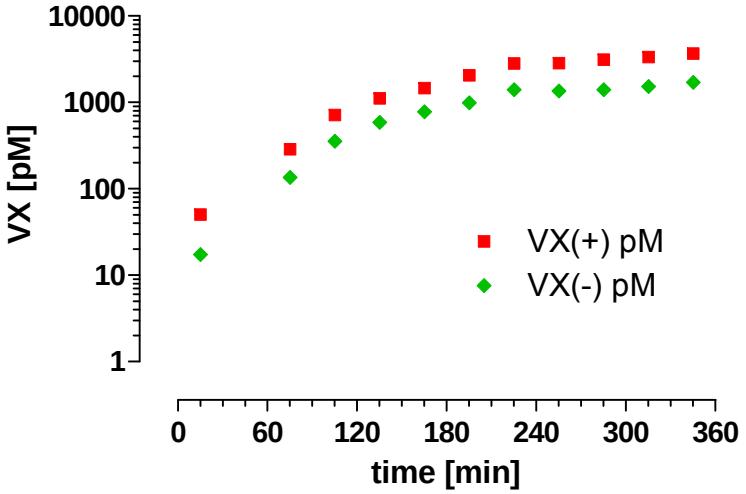
Plasma sampling for laboratory analysis:

HI-6, VX

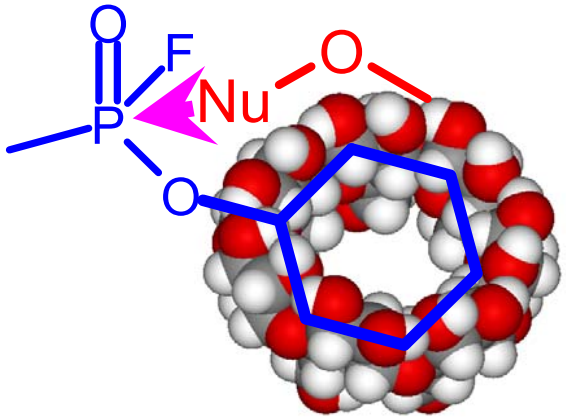
Treatment of a Percutaneously VX Poisoned Pig







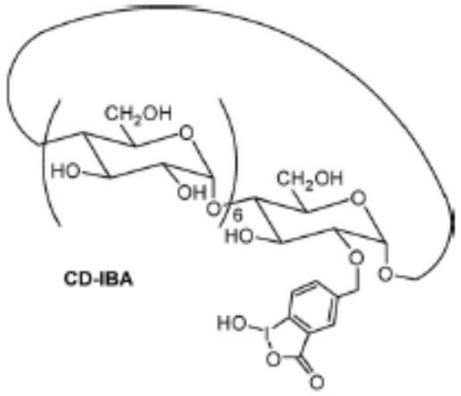
Alternative Approaches for Therapy of Nerve Agent Poisoning



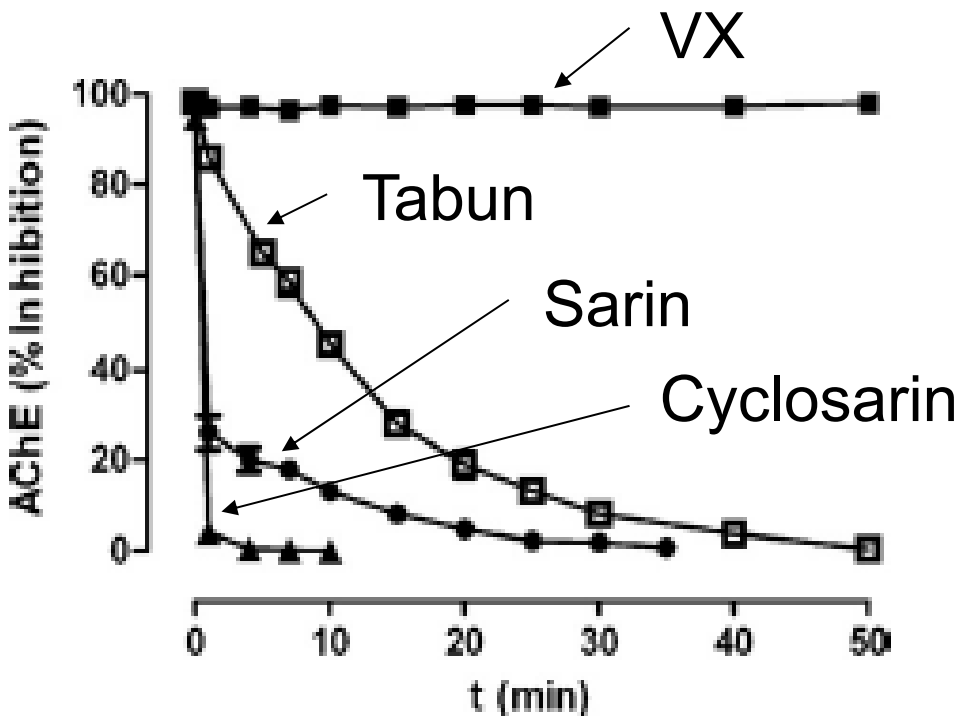
Enhanced elimination by scavengers

Cyclodextrins

Cyclodextrines as Small Molecular Scavengers in Nerve Agent-Poisoning



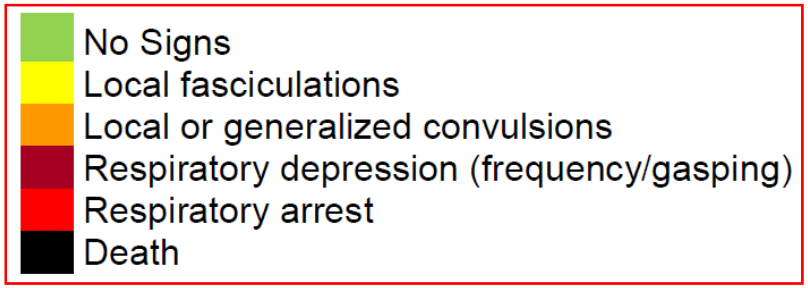
2-O-(3Carboxyl-4-iodosobenzyl)-ß-cyclodextrin



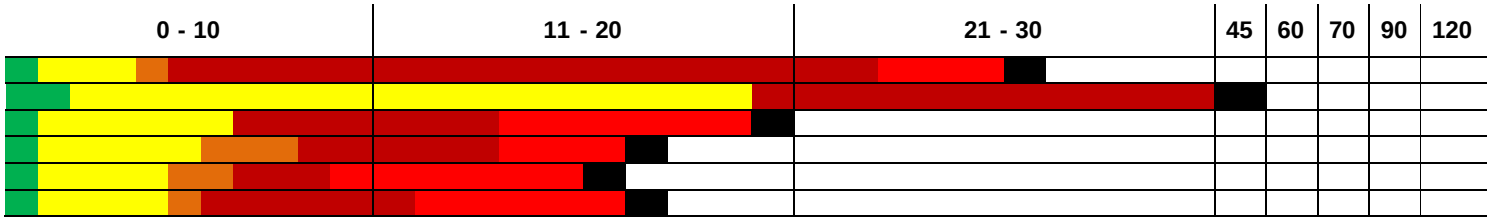
Wille et al. Toxicology 2009, Zengerle et al. J Org Chem 2011, Müller et al. Toxicol Lett 2011; 2013 a,b

Treatment of a GF Poisoned Guinea Pig with with 6-OxP-CD

Anesthetized (Medetomidine – Fentanyl – Midazolam)
Cannulated A. carotis and V. jugularis
ß-CD (6-OxP-CD; 100 mg/kg i.v. at -5 min)
Cyclosarin (100 µg/kg s.c.; ~2LD50 at 0 min)
No post-exposure treatment



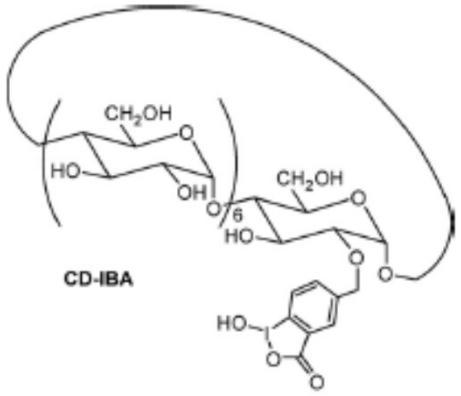
GF 100 µg/kg s.c.



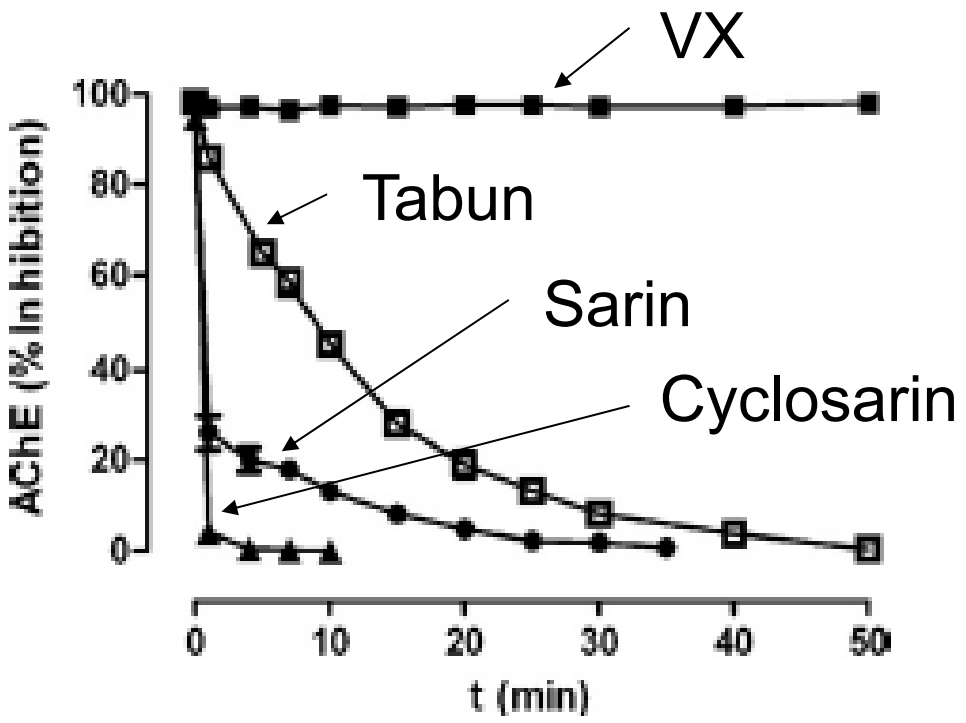
6-OxP-CD 100 mg/kg i.v. 5 min prior to GF 100 µg/kg s.c.

Worek et al. Toxicol Lett 2014

Cyclodextrines as Small Molecular Scavengers in Nerve Agent-Poisoning

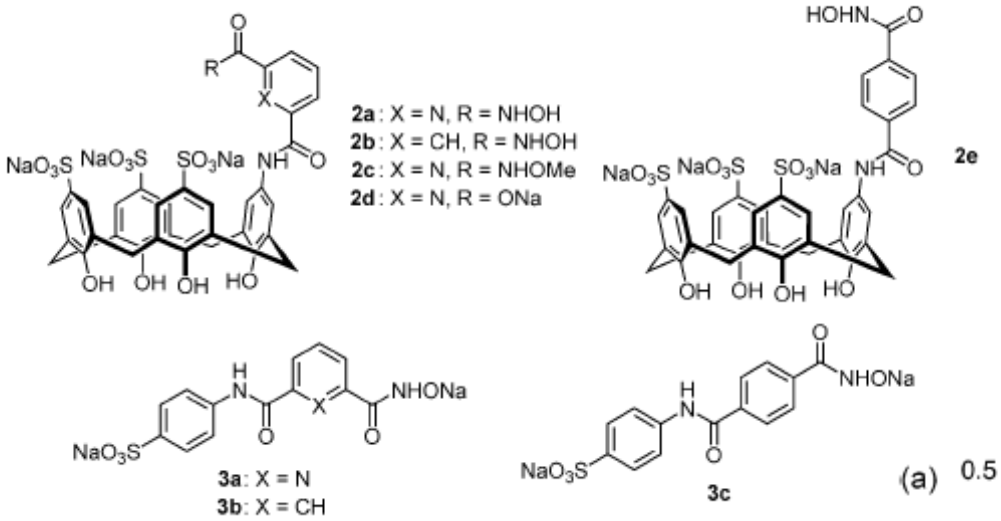


2-O-(3Carboxyl-4-iodosobenzyl)-ß-cyclodextrin



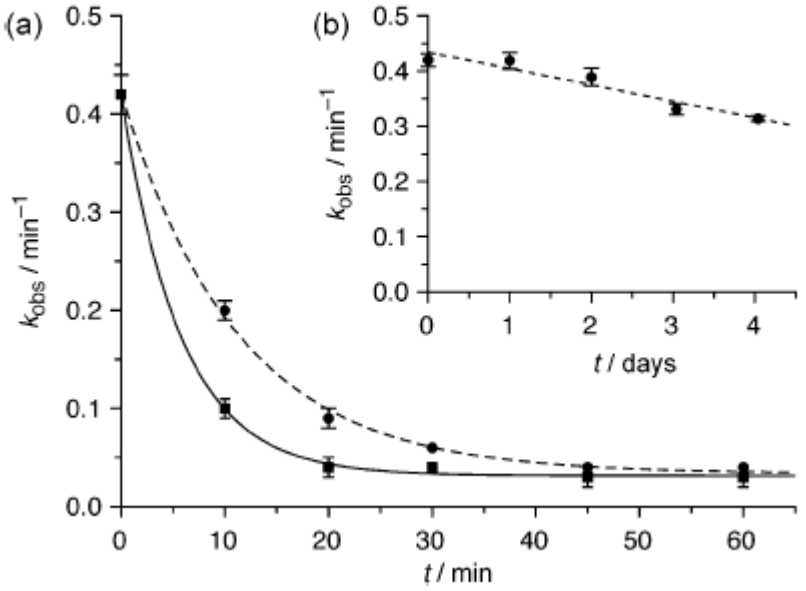
Wille et al. Toxicology 2009, Zengerle et al. J Org Chem 2011, Müller et al. Toxicol Lett 2011; 2013 a,b

Sulfonatocalix[4]arenes as Small Molecular Scavengers in VX-Poisoning



Detoxification of VX with Calixarenes containing hydroxamic acid was about 3500 times faster when compared with spontaneous hydrolysis

Calixarene-derivates were decorated with substituents in order to achieve fast detoxification in water

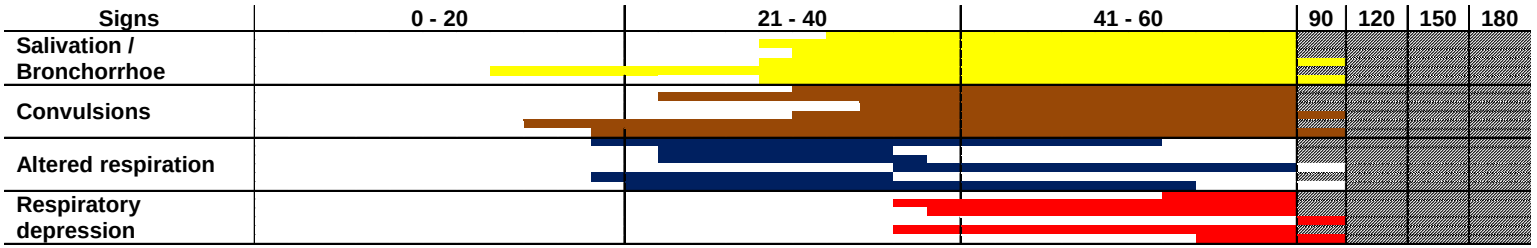


Schneider et al. Angew Chem Int Ed Engl 2016

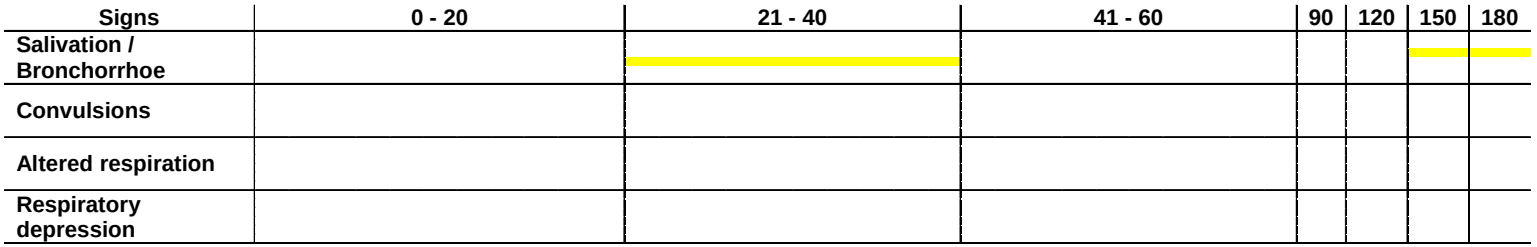
Treatment of VX Poisoned Guinea Pigs with PTE C23

Anesthetized (Medetomidine – Fentanyl – Midazolam)
Cannulated A. carotis and V. jugularis
VX (18 µg/kg s.c.; ~2LD50 at t = 0 min)
PTE C23 (5 mg/kg at t = 5 min)
No post-exposure treatment

18 µg/kg VX s.c. (~2LD_{50})

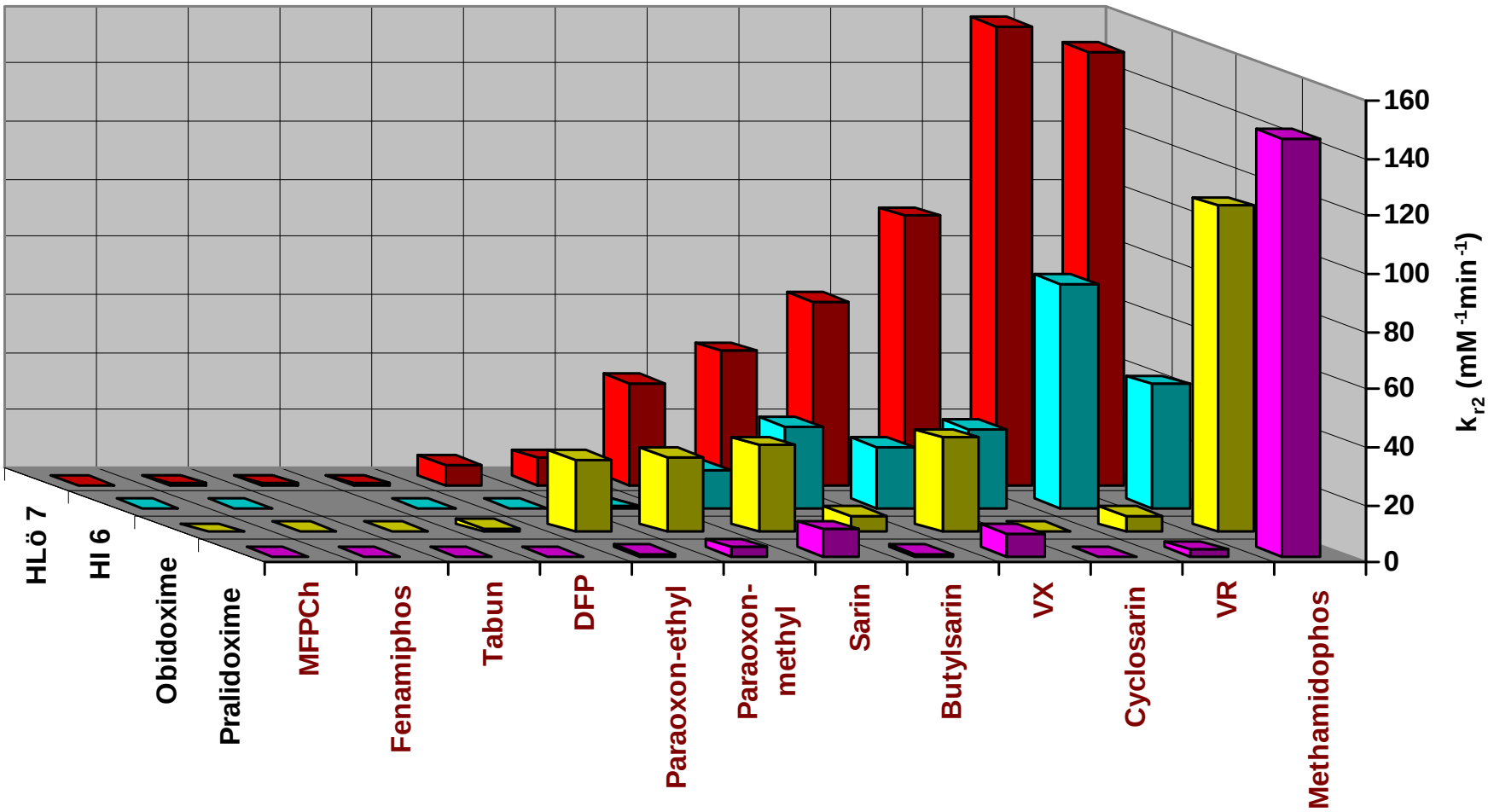


18 µg/kg VX s.c. (~2LD_{50}) followed by 5 mg/kg PTE i.v. after 5 min



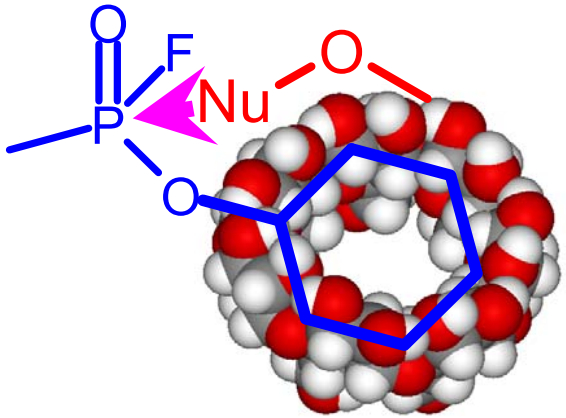
Worek et al. Toxicol Lett 2014

Oxime in OP-poisoning – Hope and Despair



Worek et al. Biochem Pharmacol, 2004; Worek et al. Toxicology, 2016, Worek et al. Chem Biol Interact, 2016

Alternative Approaches for Therapy of Nerve Agent Poisoning



Enhanced elimination by scavengers

Cyclodextrins

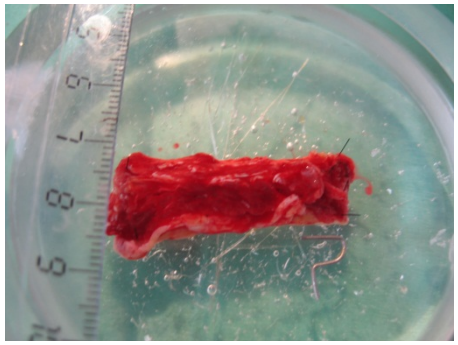
Enzymes,
e.g. PON1, PTE

Modulation of nicotinic ACh-receptors

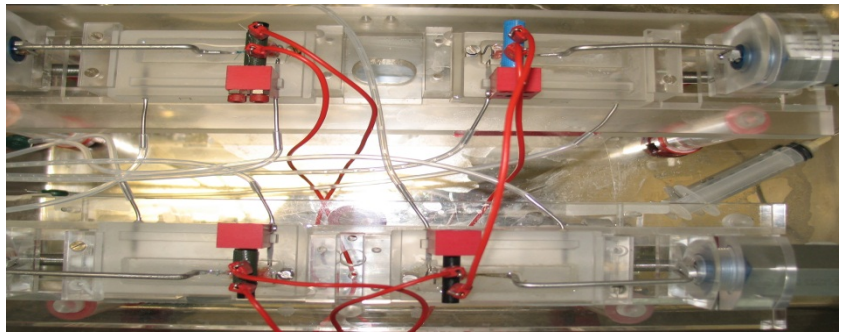
Goldsmith et al. Arch Toxicol ,2016
Wille et al. Tox Lett, 2016
Ashani et al. Chem Biol Interact, 2016

Restoration of Nerve Agent Blocked Muscle Force

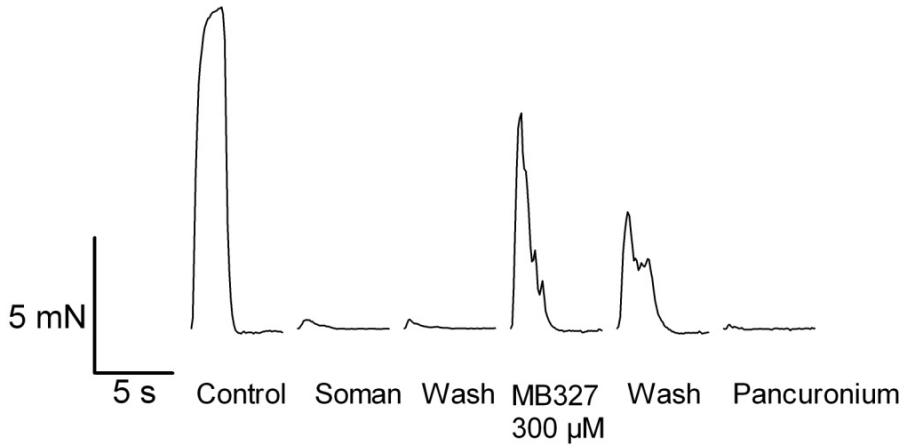
Human intercostal muscle



Horizontal 4-chamber-organ bath with stimulation-electrodes



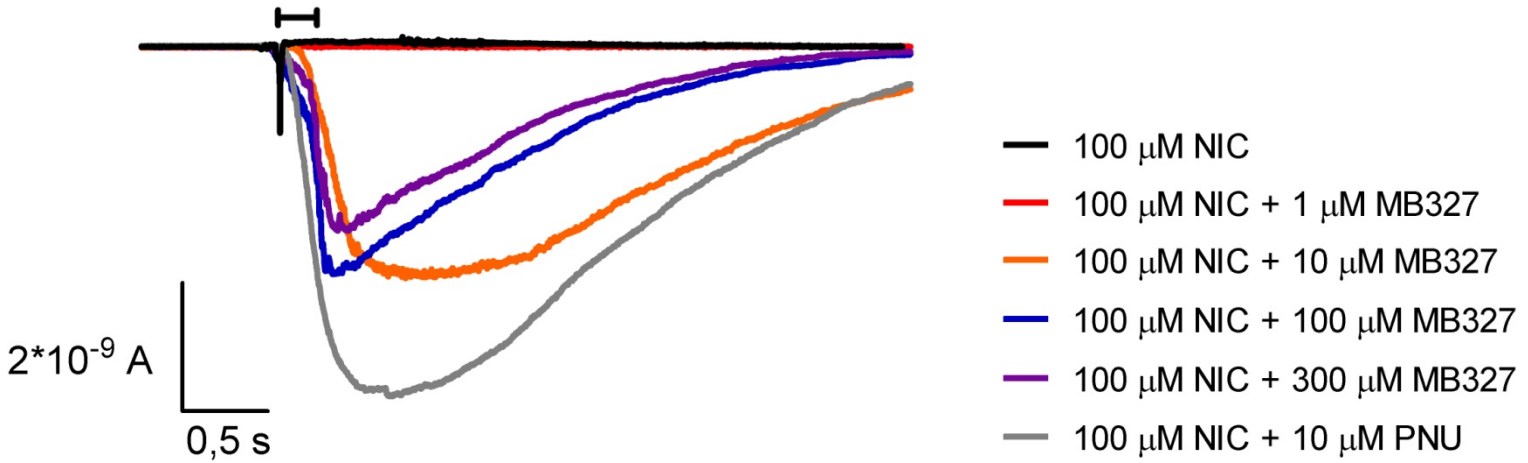
Muscle-force after electrical field stimulation (25 Hz)



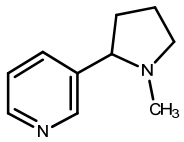
Seeger et al. Toxicology 2012

Interaction of MB 327 with Human Nicotinic Acetylcholine Receptors (α7)

Functionality: Whole cell recording by automated patch clamp platform

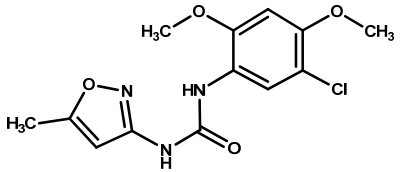


Agonist

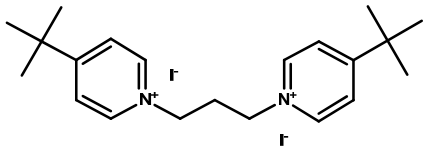


Nicotine

Modulators



PNU 120596

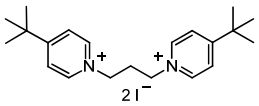


MB327

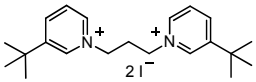
Scheffel et al. 2017 to be published

Interaction of MB 327 with Nicotinic Acetylcholine Receptors (Muscle-type)

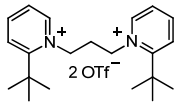
Membrane preparation of *Torpedo californica* electric organ (muscle-type nAChR)



MB327

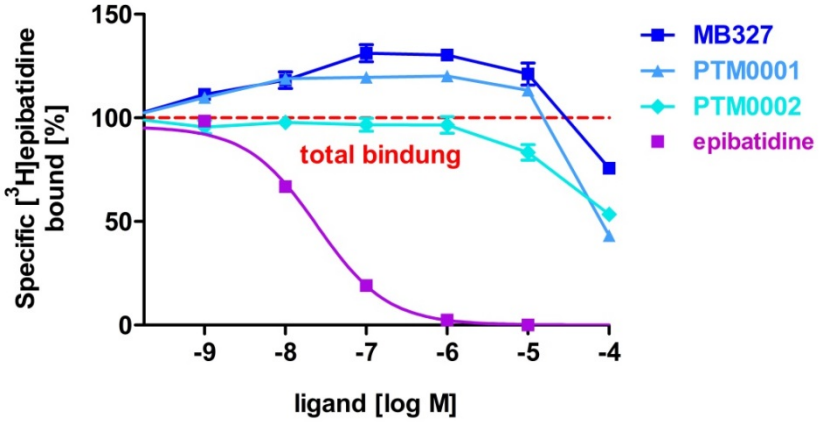


PTM0001

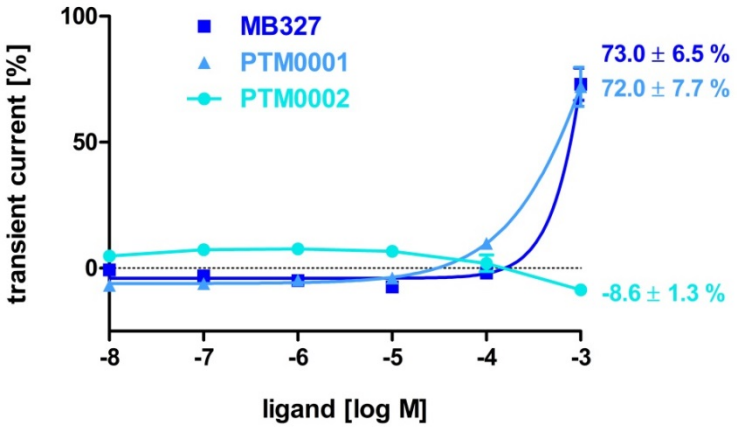


PTM0002

- Affinity on [³H]epitatidine binding sites



- Functionality based on solid supported membranes (SSM)



Niessen et al. Chem Biol Interact 2013, Niessen et al. Tox Lett 2016, Niessen at al. 2017 to be submitted

Therapeutic Approach in Nerve Agent-Poisoning

Self protection

Prompt reactivation of inhibited AChE

- even in the absence of severe signs and symptoms

- prolonged oxime treatment

Atropine for muscarinic signs and symptoms

Benzodiazepines for treatment and/or prevention of seizures

Supportive therapy

artificial ventilation, sedation, cardiovascular stabilisation

Outlook: Alternative Therapeutic Approach in Nerve-Agent Poisoning

Binding and enhanced elimination by scavengers:

small molecular scavengers
human BChE / & oxime
human AChE / & oxime
PON1; PTE

Receptor active compounds

Summary and Recommendations

Don’t wait for symptoms to develop!

Determine AChE activity in case of possible exposure as soon as possible!

Administer atropine according to signs and symptoms!

Administer an effective reactivating oxime when AChE activity has dropped even in the absence of clinical signs and symptoms!

Maintain oxime therapy as long as the organophosphorus compound is persisting in the body!

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Thank you for your attention