

DEFENCE



DÉFENSE



## **HI-6: Oxime Research in Canada**

Development of an Intravenous Formulation of HI-6

Defence R&D Canada

Nov 4, 2009



Defence Research and  
Development Canada

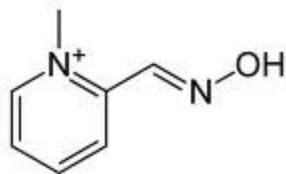
Recherche et développement  
pour la défense Canada

Canada



# Oxime Development Programs

- Before 1950 the only treatment for nerve agent or pesticide poisoning was atropine
- During the 1950's a group of chemicals known as "oximes" were developed that could reactivate cholinesterase inhibited by nerve agents
- The first clinically tested oxime was pralidoxime chloride (2-PAM)

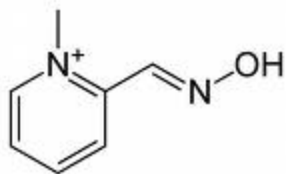


Pralidoxime

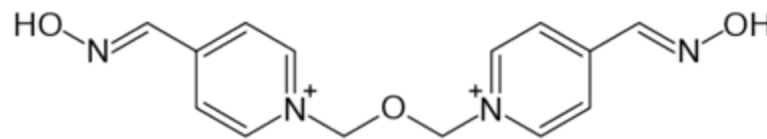


## Oxime Development Programs

- During the 1960's obidoxime which was effective against GA was also licensed in several countries
- Both 2-PAM and obidoxime lacked efficacy against all nerve agents;
  - Soman (GD), cyclosarin (GF), Russian VX (RVX)
- This led most nations to initiate programs to develop improved oximes



Pralidoxime

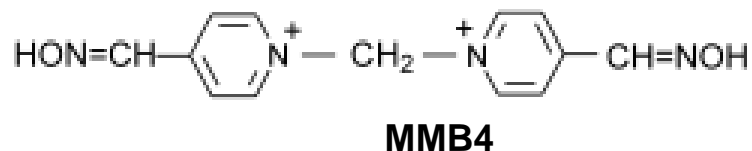
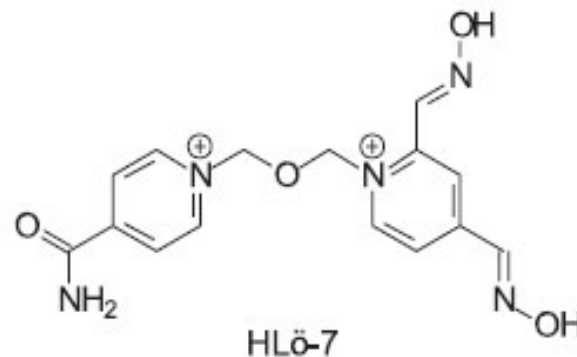
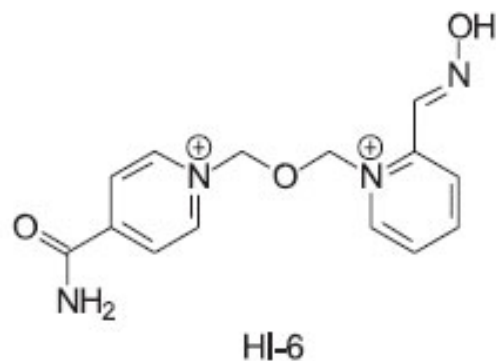


Obidoxime



# Oxime Development Programs

- Effort to develop new oximes led to the synthesis of several oximes including;
  - HI-6, HLö7 and MMB4





## HI-6

- HI-6
  - dichloride salt (2-Cl)
  - synthesized (1966) by Ilse Hagedorn and Irmo Stark (GER)
- HI-6 identified by several NATO nations as the primary future oxime based on;
  - improved efficacy against all nerve agents except GA
  - least toxic of all next generation oximes  
(HI-6, HLö7 and MMB4)
  - has been fielded by Canada, Sweden and Czech Republic



## DRDC Oxime Program (2003)

- HI-6 Development
  - autoinjector dose limited by solubility of 2-Cl
    - dimethanesulphonate salt (DMS) developed to increase solubility
  - stability in solution
    - wet/dry autoinjector
    - use of suspensions or emulsions in single chamber
  - BCME synthesis route for HI-6 2-Cl
    - novel synthesis route for HI-6 DMS



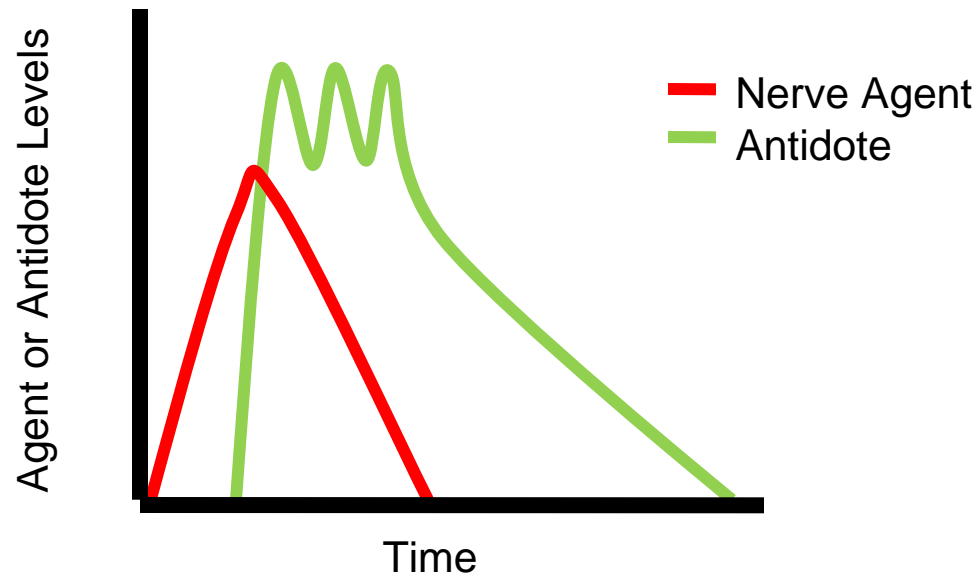
## CA-NL-UK Development (2005)

- Trilateral agreement to license autoinjector
- Based on 3-in-1 autoinjector containing HI-6, atropine and avizafone (*water soluble diazepam*)
- Pre-clinical and clinical studies to support regulatory approval
  - Non-BCME method
  - Small-scale (3kg) GMP batches (late-2009)
  - Scale-up (15kg) GMP batches (2010-2011)
  - Clinical toxicology studies (2010-11)
  - Ph1 clinical study (2011-12)



# Parenteral Formulation Program

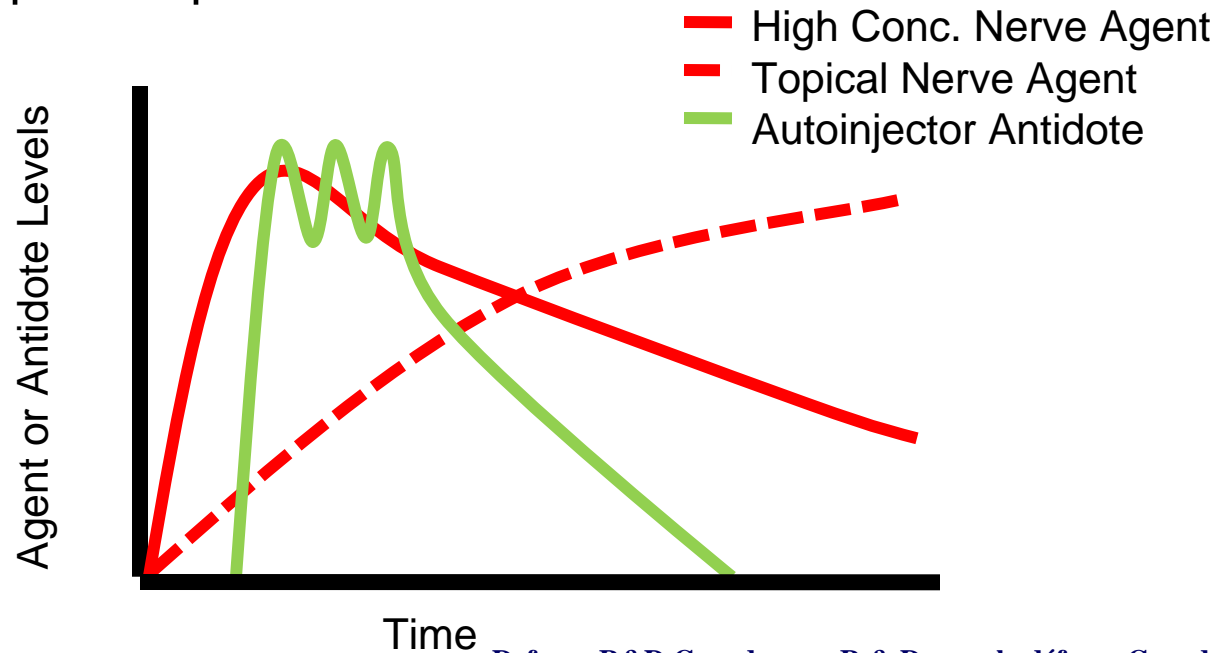
- Current nerve agent antidote treatment regimens are designed for field treatment of nerve agent exposure.
  - 3 autoinjectors, one at 1, 15 and 30 min





# Parenteral Formulation Program

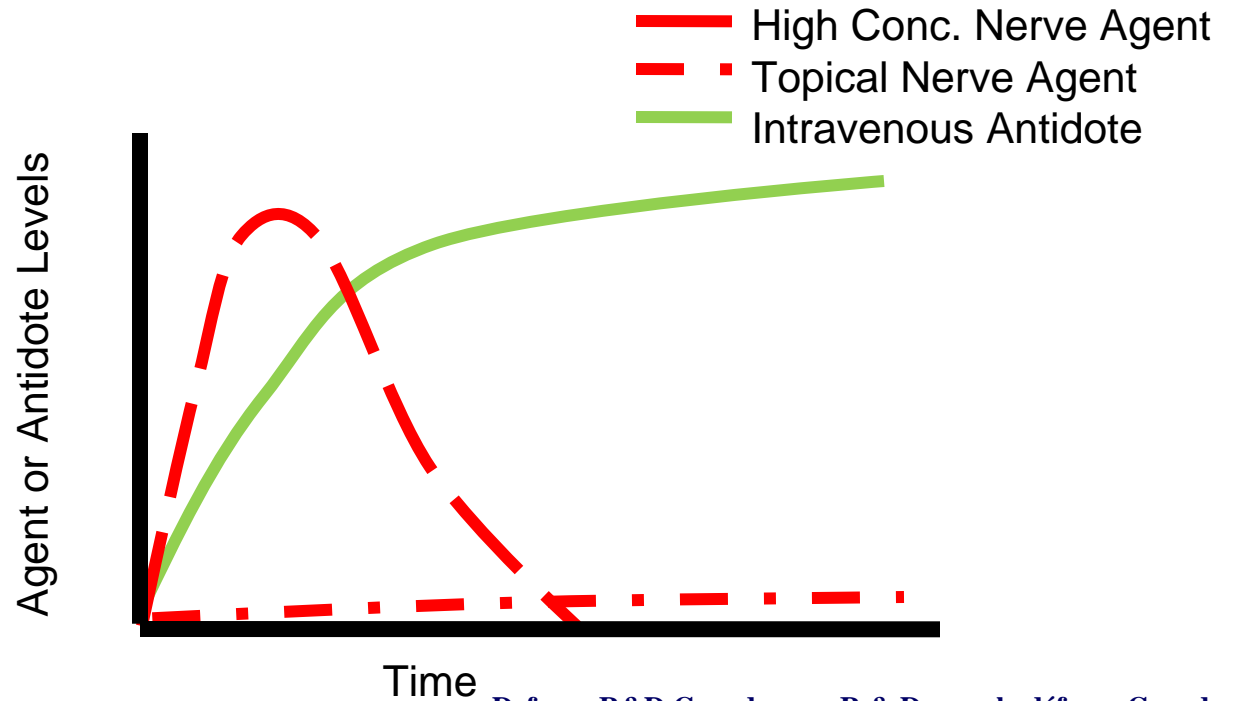
- How do we continue required treatment following extraction from the area or in a medical facility?
  - Exposure to high concentrations
  - Topical exposures





# Parenteral Formulation Program

- Intravenous injection or infusion
  - optimize therapeutic concentrations





## Parenteral Study Outline

- GLP Implementation at DRDC Suffield 2007
- Determine pharmacokinetics (PK) of HI-6 2-Cl and DMS salts in guinea pigs (GLP) and domestic swine
- Develop an infusion protocol to maintain a target plasma concentration of 100  $\mu\text{Mol/L}$ 
  - 8 hour infusion period
  - with and without atropine sulfate
- Efficacy against percutaneous nerve agent exposure (3Q 2010)



# GLP-Validated HPLC Method for Quantification of HI-6 in Plasma

- Mobile Phase
  - Component A
    - PIC-B7 (acetic acid, methanol, alkane sulfonate salts), water
    - triethylamine
    - Water
  - Component B
    - 1 part Component A
    - 1 part Methanol
- Linear Gradient Elution (A:B - 60:40 to 0:100)
- UV detection (302 nm)



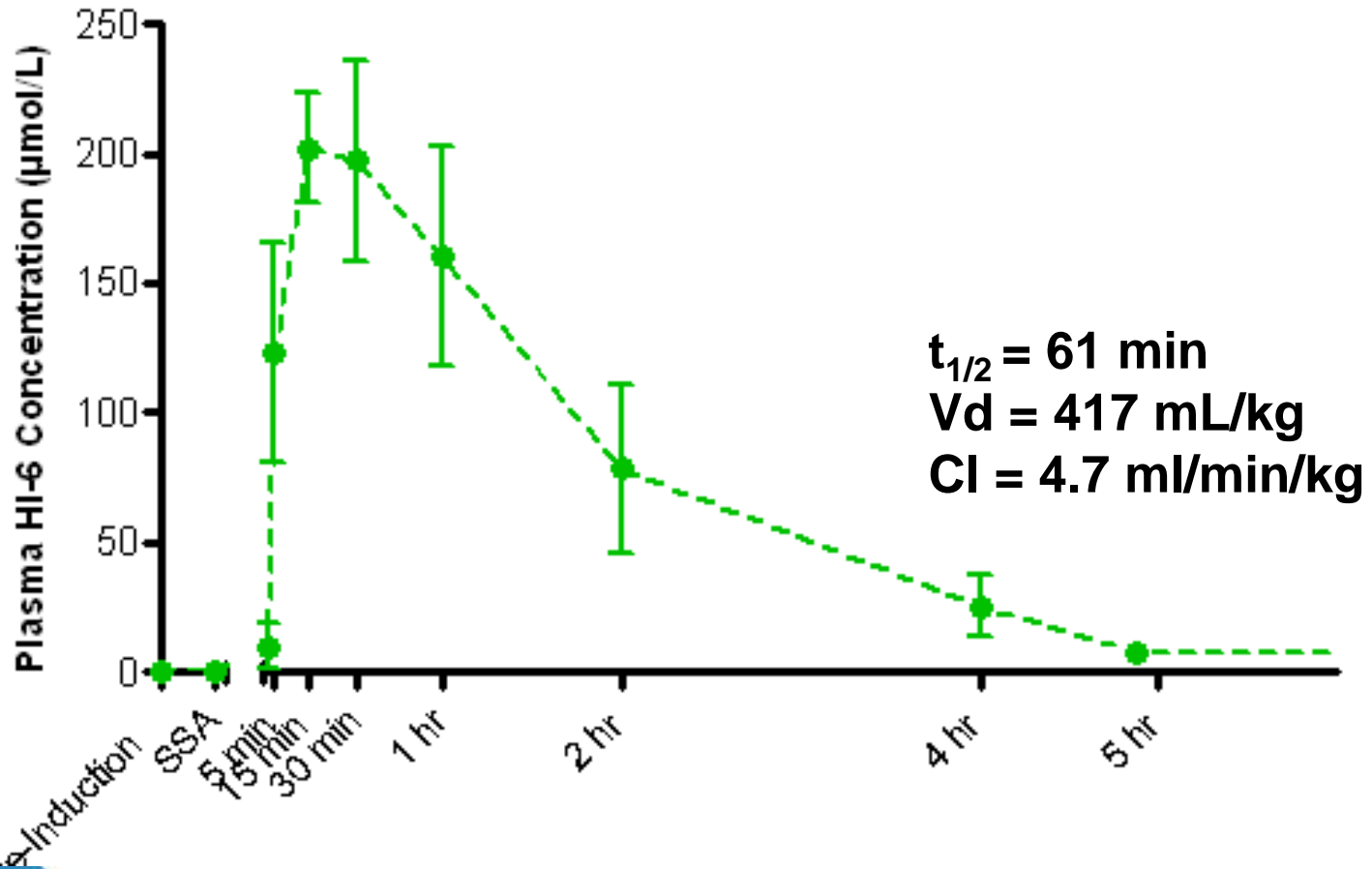
# Plasma Sample Preparation

- ~ 100  $\mu\text{L}$  of whole blood collected via an indwelling catheter located in the jugular vein (guinea pig)
- Plasma is collected and then stored at  $-80^{\circ}\text{C}$
- 35  $\mu\text{L}$  of plasma mixed with 35  $\mu\text{L}$  0.10 mg/mL 2-PAM (internal standard)
- Precipitate proteins with 10% TCA, centrifuge, remove supernatant
- Neutralize with 0.2 M NaOH and filter
- Aliquot filtrate into glass insert
- 2-PAM used as an internal standard



# Pharmacokinetics - Intramuscular

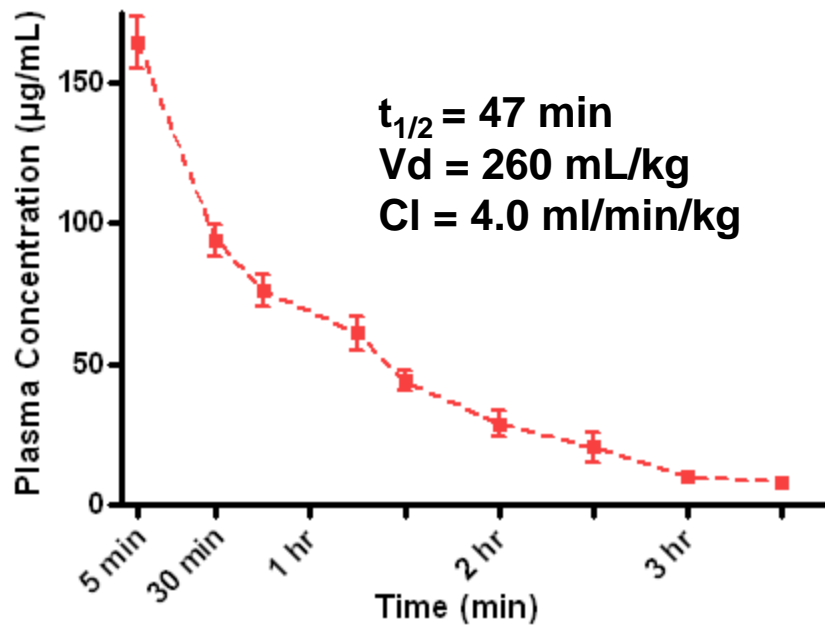
## HI-6 2CI IM



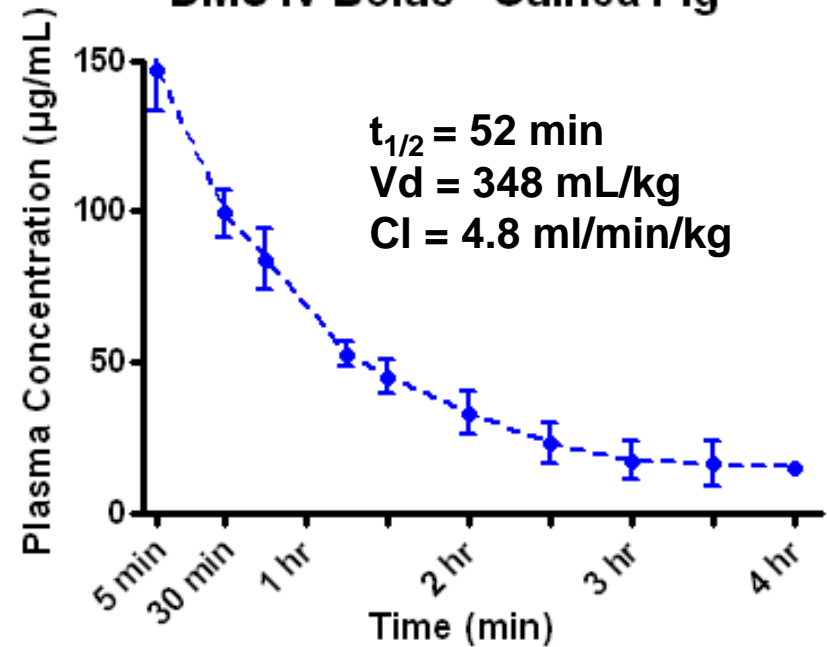


# Intravenous Pharmacokinetics – Guinea Pig

2Cl IV Bolus - Guinea Pig



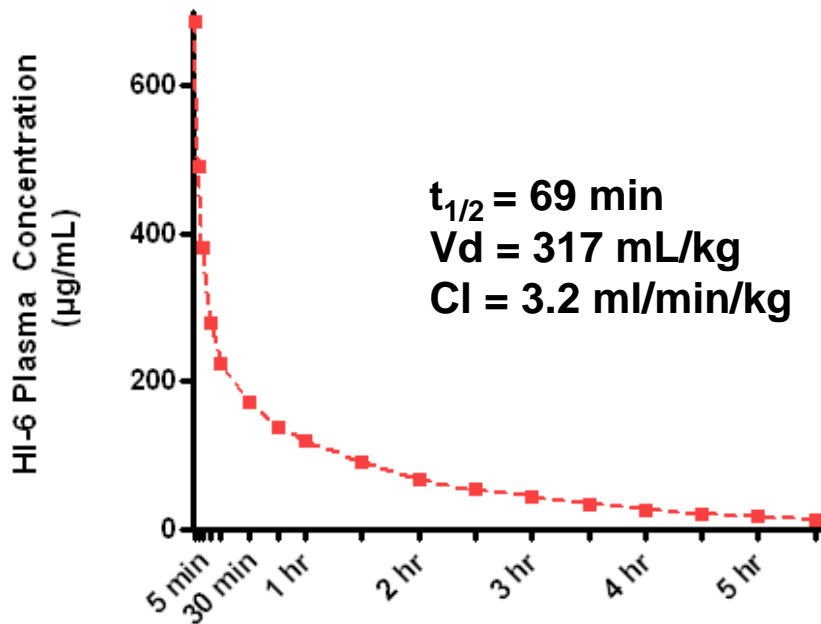
DMS IV Bolus - Guinea Pig



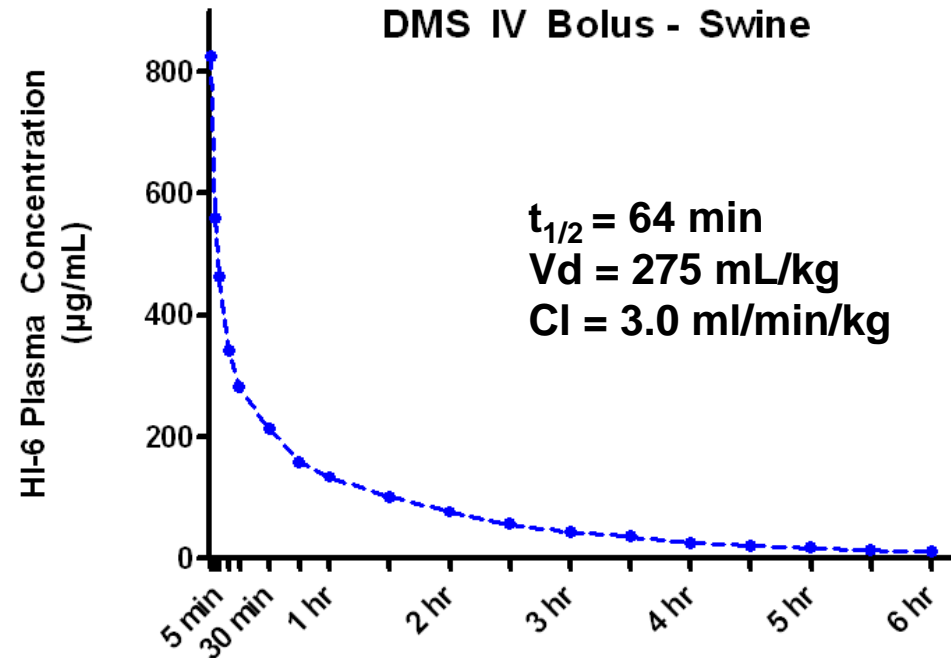


# Intravenous Pharmacokinetics - Swine

2CI IV Bolus - Swine



DMS IV Bolus - Swine





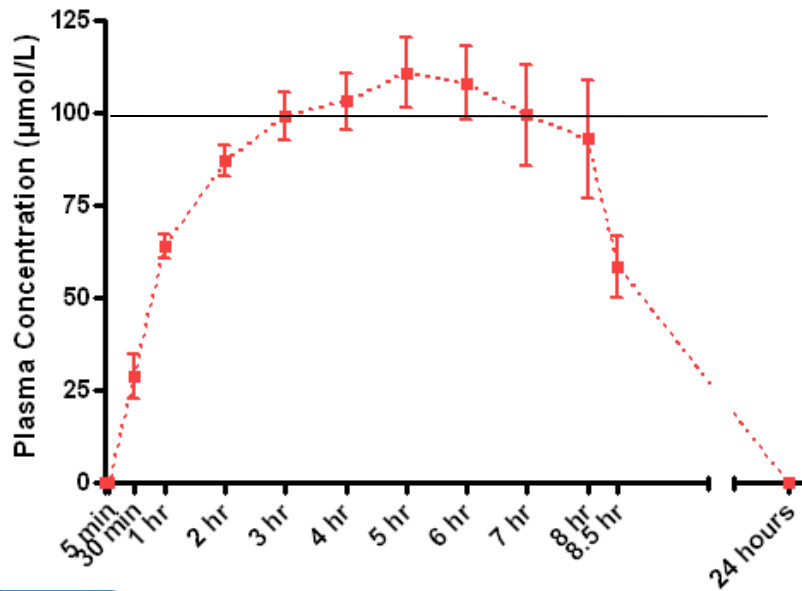
## Comparison of HI-6 PK

	Guinea Pig			Swine		Human
	IM (2-Cl)	IV (2-Cl)	IV (DMS)	IV (2-Cl)	IV (DMS)	IM (2-Cl)
$t_{1/2}$ (min)	61	47	52	69	64	67
Vd (mL/kg)	417	260	348	317	275	240
Cl (mL/min/kg)	4.73	3.96	4.8	3.20	2.96	2.47

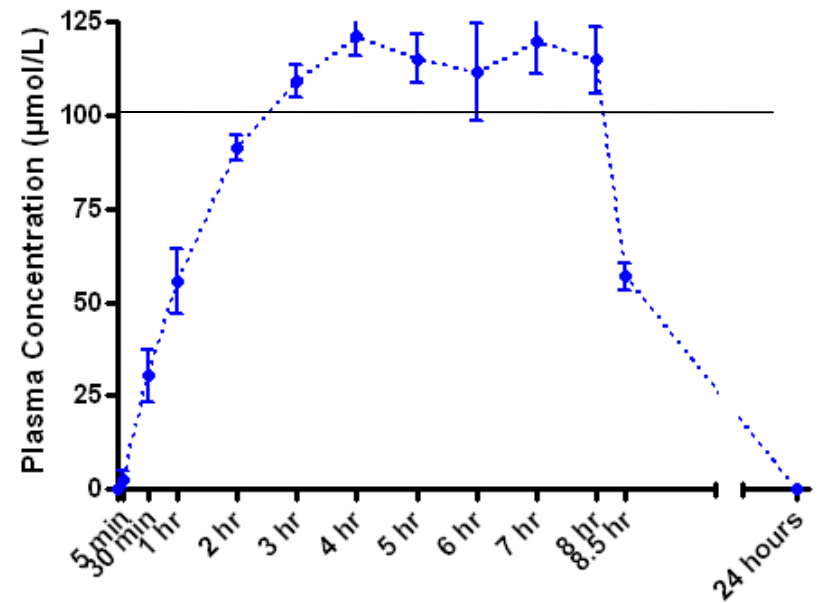


# HI-6 Infusion (8 hr) – Guinea Pig

### HI-6 2Cl Infusion



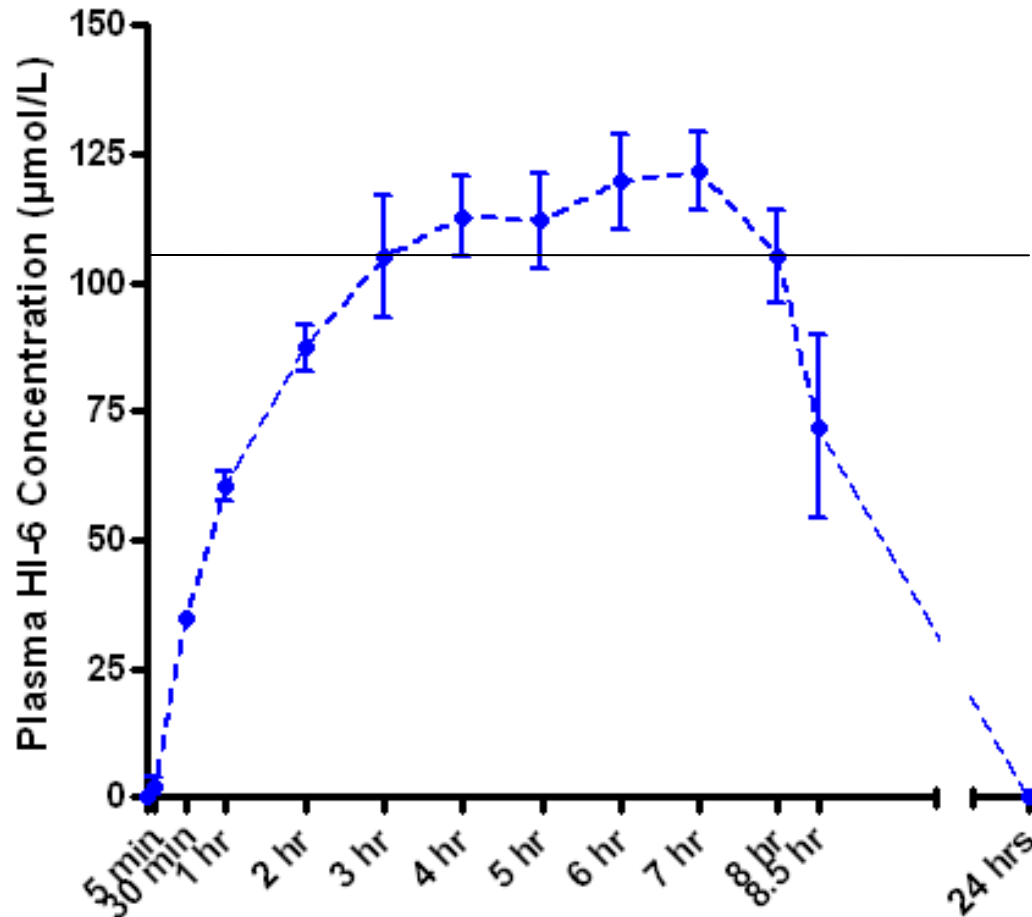
### HI-6 DMS Infusion





# HI-6 and Atropine Infusion – Guinea Pig

HI-6 DMS & AS Infusion





## Conclusions

- Paired-ion chromatography effectively resolves HI-6 and 2-PAM in small plasma samples
- HI-6 pharmacokinetics in anesthetized guinea pigs are similar to those in anesthetized swine
- These kinetics are sufficient to formulate infusion rates to sustain target plasma concentrations for up to 8 hours
- Efficacy experiments need to be conducted (3Q 2010)

DEFENCE



DÉFENSE