Advances in chemical control of mosquito-borne disease: Where do we stand in using densovirus as synergistic agent?

Pr. Bruno Lapied,

Laboratoire SiFCIR UPRES EA 2647, USC INRAE 1330
Université d'Angers, Angers, France

bruno.lapied@univ-angers.fr

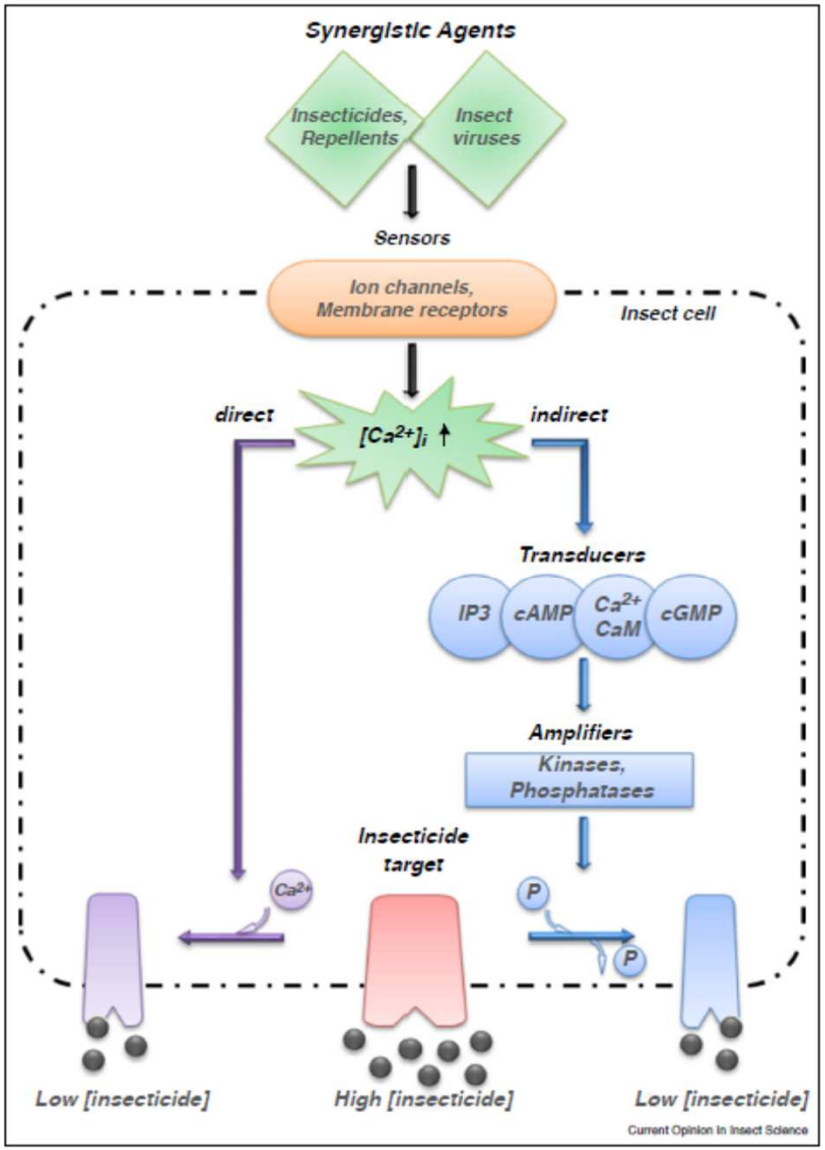
Use of synergistic agents in mosquito control

1

A synergistic agent is a chemical that **indirectly enhances the effectiveness of several class of insecticides**, although they usually have no effect or only a limited effect themselves.

2

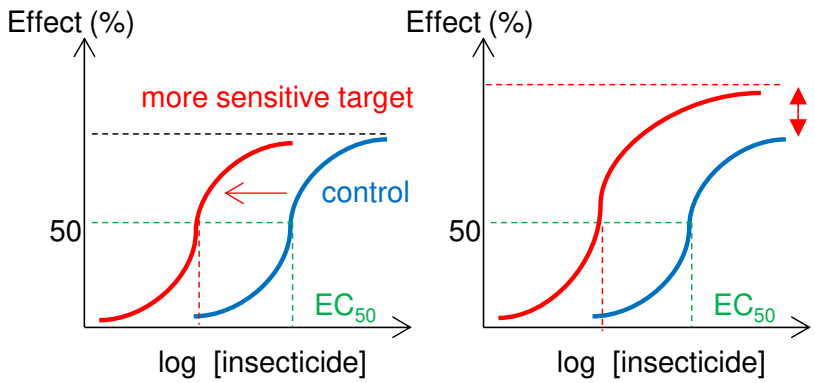
At non-toxic concentrations, a synergistic agent acts by **activating certain calcium-dependent intracellular signalling pathways**



Deshayes C., Moreau E., Pitti-Caballero J., Froger JA., Apaire-Marchais V. & Lapied B. Curr. Opinion Insect Sci., 2018, 30.

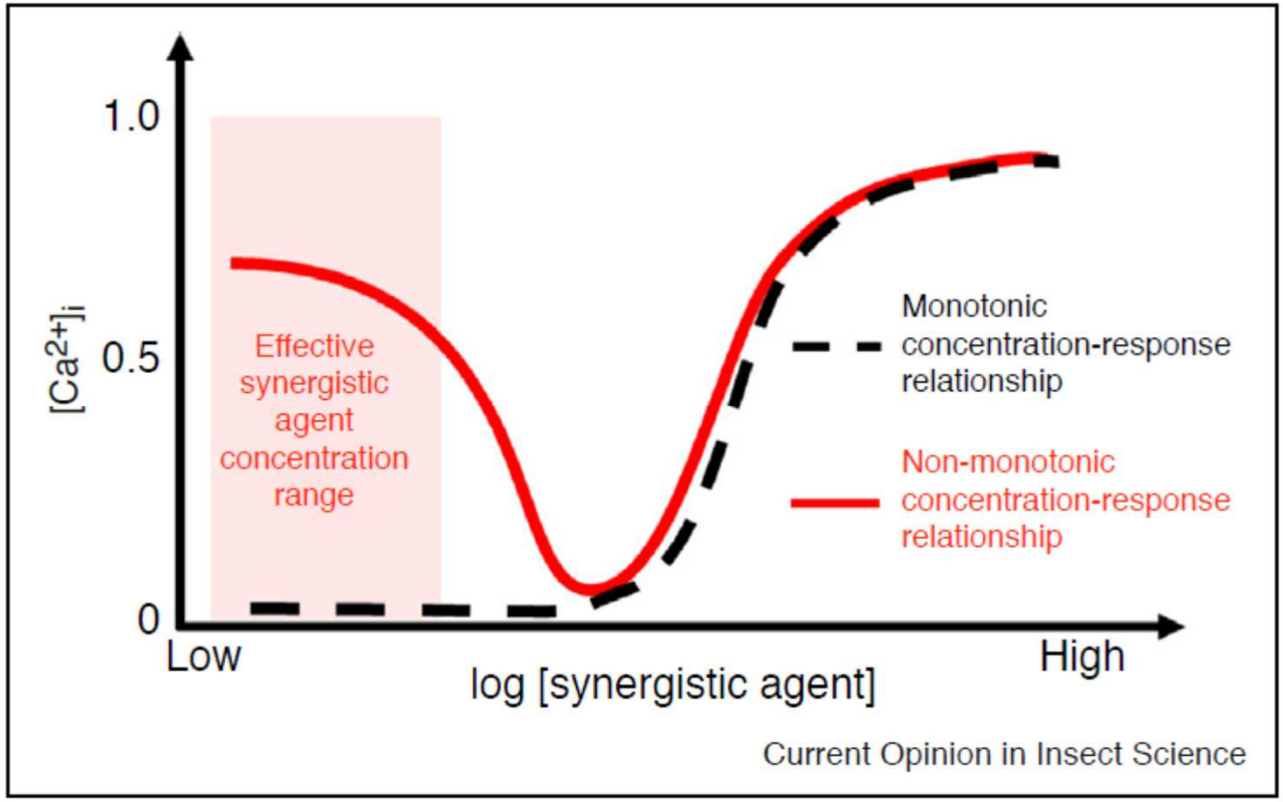
3

This leads to sensitization of the insect thus **increasing the efficacy of the treatment.**



Use of synergistic agents in mosquito control

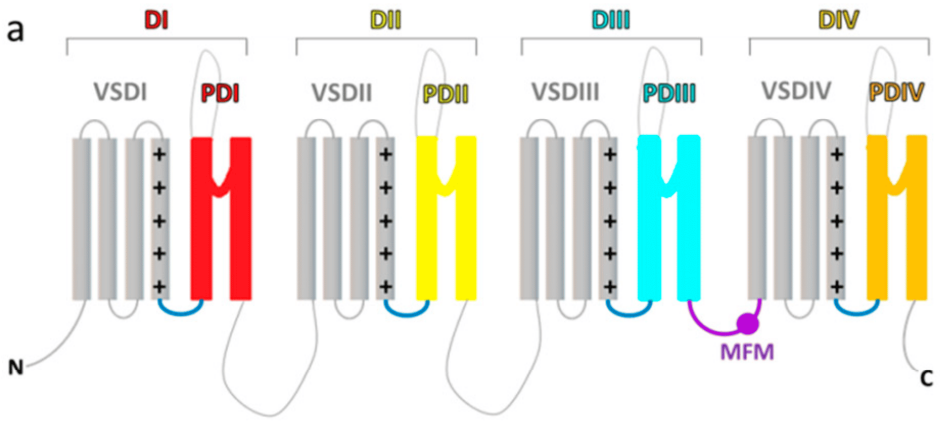
Monotonic and **non-monotonic concentration-response curves**.



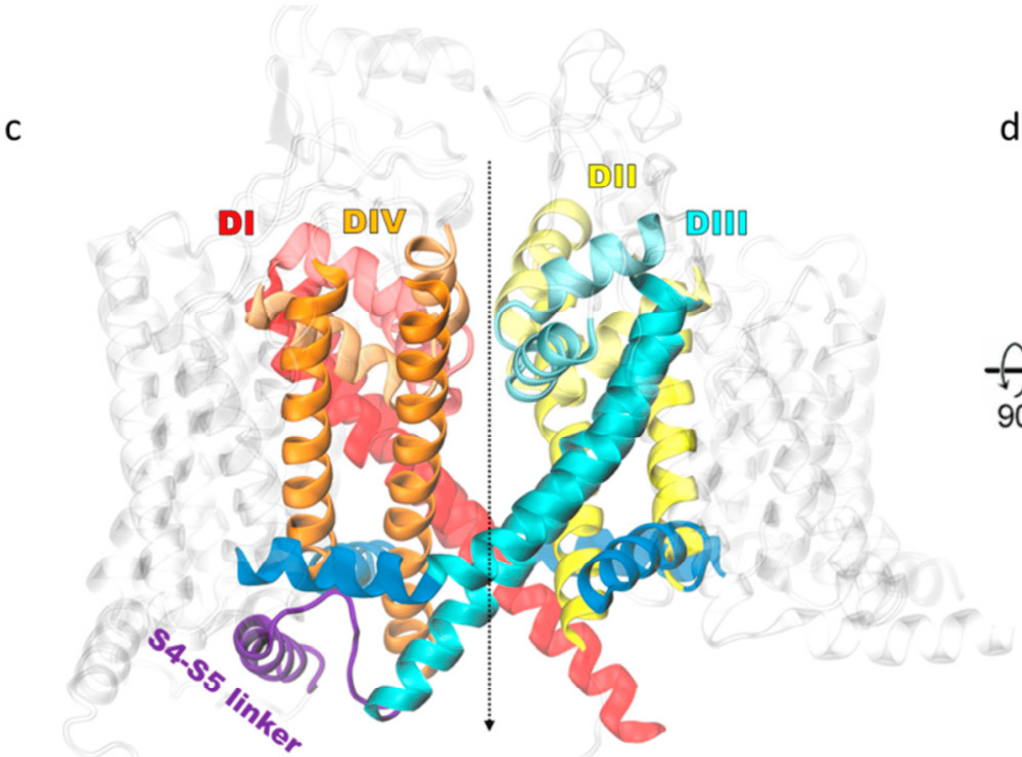
Deshayes C., Moreau E., Pitti-Caballero J., Froger JA., Apaire-Marchais V. & Lapied B. Curr. Opinion Insect Sci., 2018, 30.

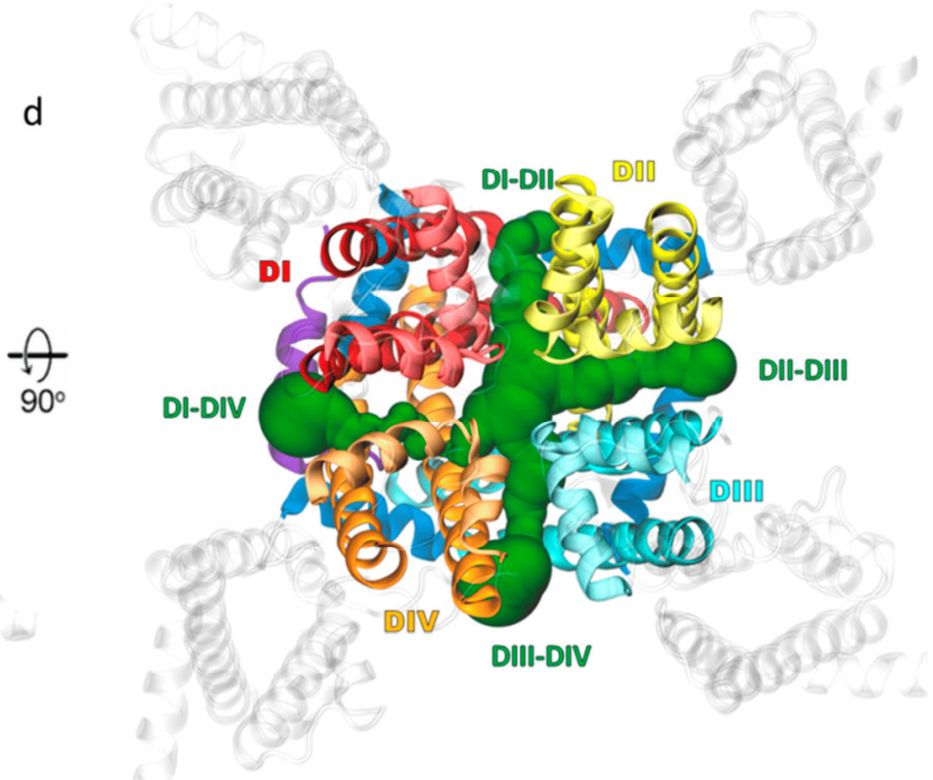
With **synergistic agent**, the illustrated U-shaped curve is considered as non-monotonic concentration-response relationship because the shape of the curve changes sign one or more times within the range of the concentrations examined.

Use of synergistic agents in mosquito control



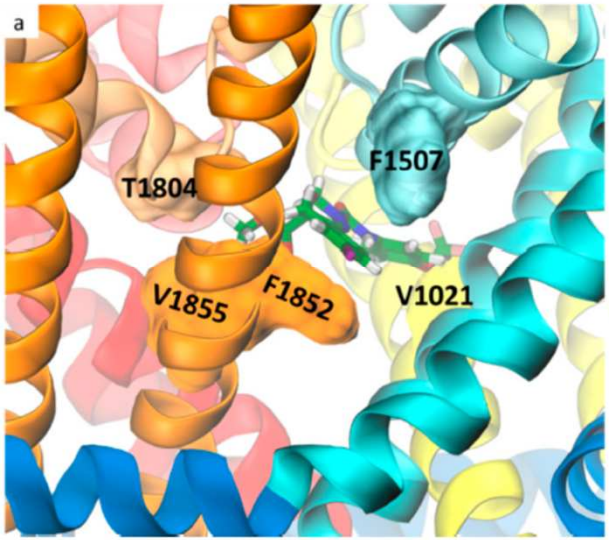
Topology of the *Anopheles gambiae* pseudotetrameric voltage-gated sodium channel alpha subunit (molecular docking)



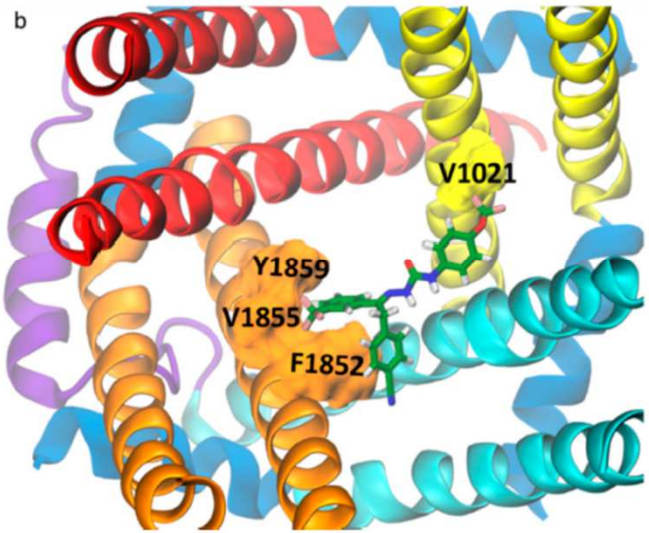


Niklas B., Rydzewski J., Lapied B. & Nowak N., Int. J. Mol. Sci., 2023, 24.

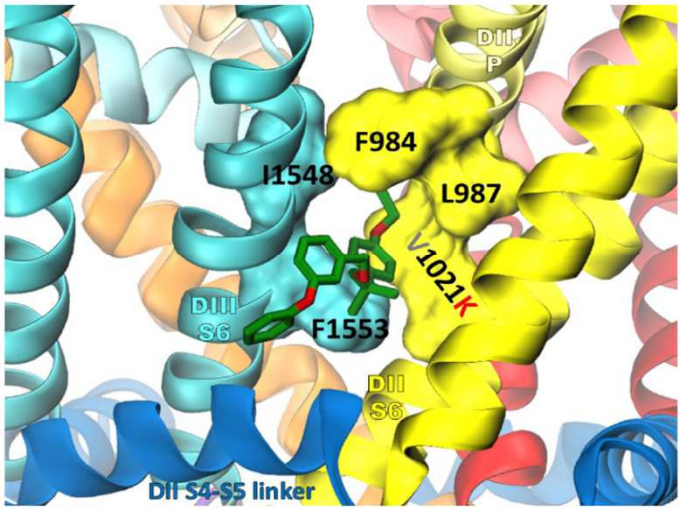
Use of synergistic agents in mosquito control



DCJW (indoxacarb)



metaflumizone



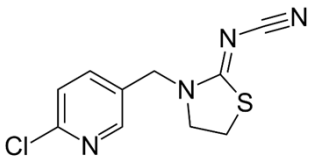
etofenprox

Niklas B., Rydzewski J., Lapied B. & Nowak N., Int. J. Mol. Sci., 2023, 24.

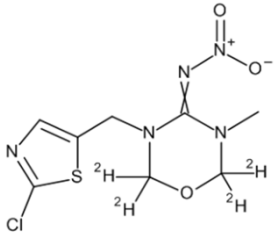
Use of synergistic agents in mosquito control

First example: IR3535

In vitro evaluation

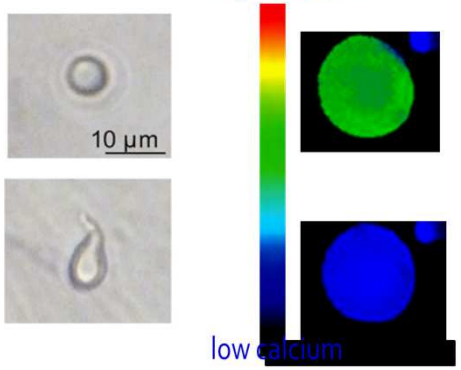


Thiacloprid
Cyanoimine (NCN, IRAC 4A)



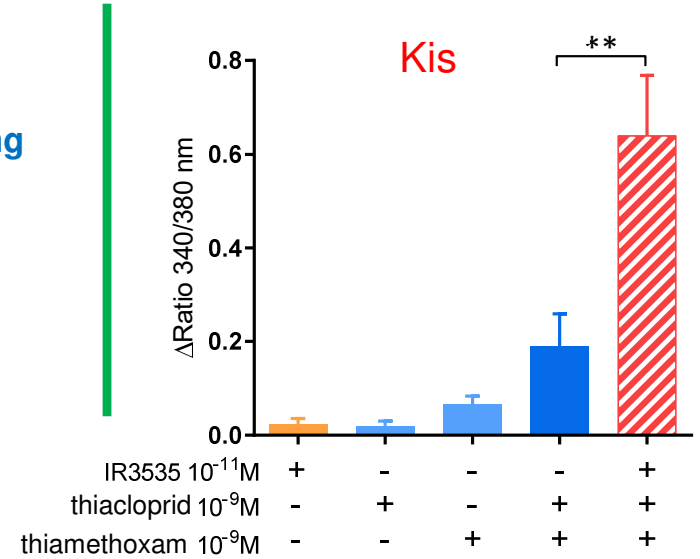
Thiamethoxam
Nitroimine (NNO2, IRAC 4A)

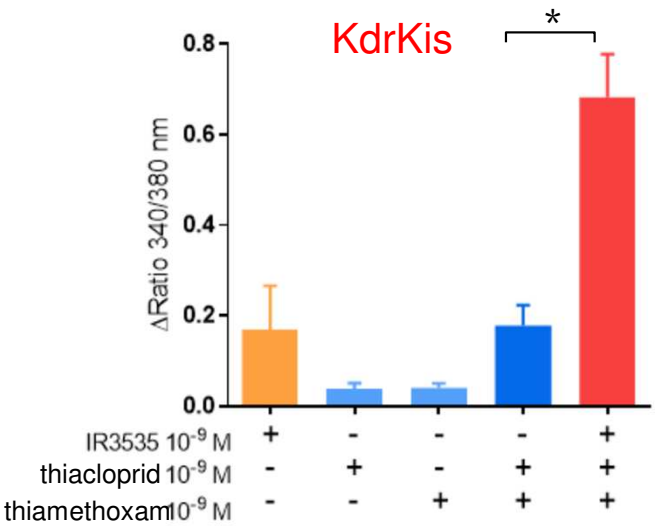
Two types of neonicotinoid insecticides acting on distinct nicotinic receptor sites producing synergistic effects

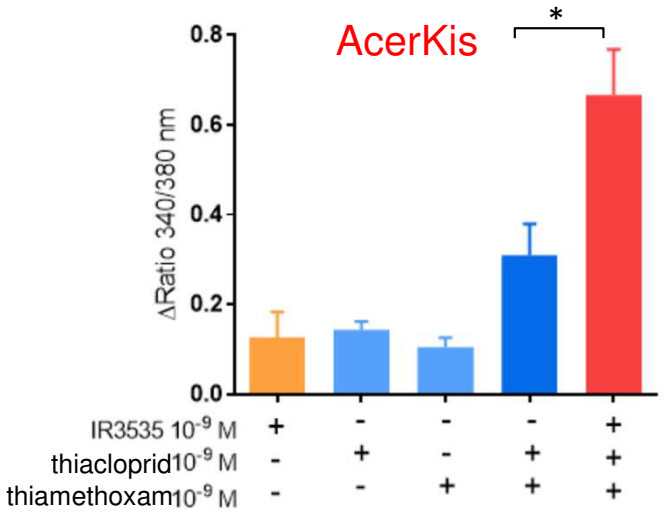


Calcium imaging
Isolated neuron from *Anopheles gambiae* (n=6-9)

Lavialle-Defaix C., Apaire-Marchais V., Legros C., Pennetier C., Mohamed A., Licznar P., Corbel V. & Lapied B. J. Neurosci. Meth., 2011, 200







IR3535 – insect mAChR *Moreau E., Mikulska-Ruminska K.,Goulu M. Perrier S., Deshayes D., Stankiewicz M., Apaire-Marchais V., Nowak W. & Lapied B. Sci. Rep., 2020, 10*

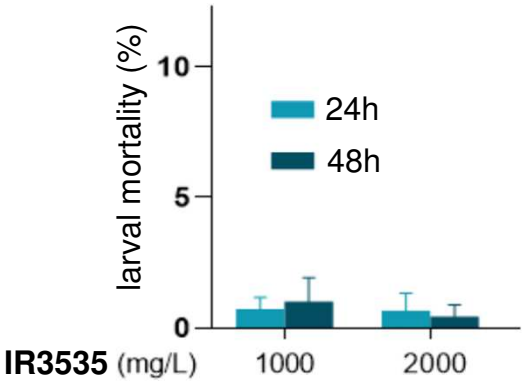
Use of synergistic agents in mosquito control

First example: IR3535

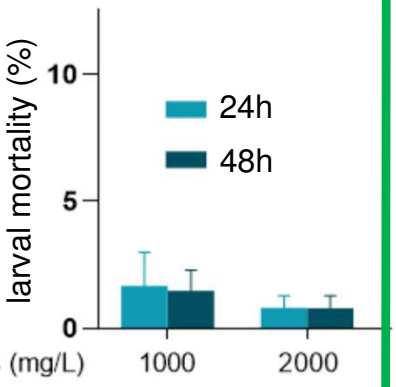
In vivo evaluation

Evaluation of the toxicity of IR3535 alone at relatively high concentration

AcerKis



KdrKis



Increasing insecticide effect while reducing the concentration

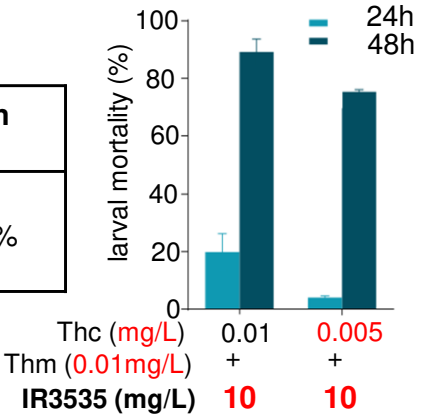
Collaborations: Marine El Adouzi & Fabrice Chandre

International application PCT/EP2021/067833

Larval susceptibility of *Anopheles gambiae* AcerKis and KdrKis to IR3535 combined with thiacloprid and thiamethoxam

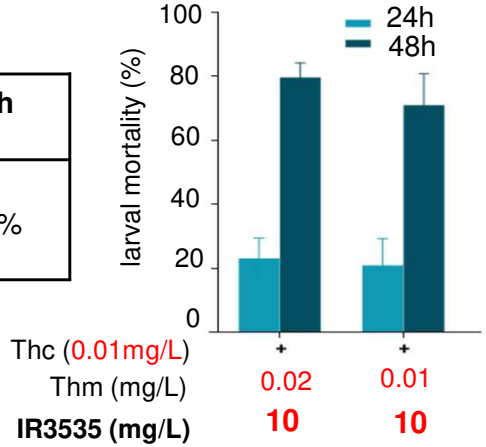
AcerKis

	24h	48h
Thc 0.01mg/L + Thm 0.01mg/L	10%	41%



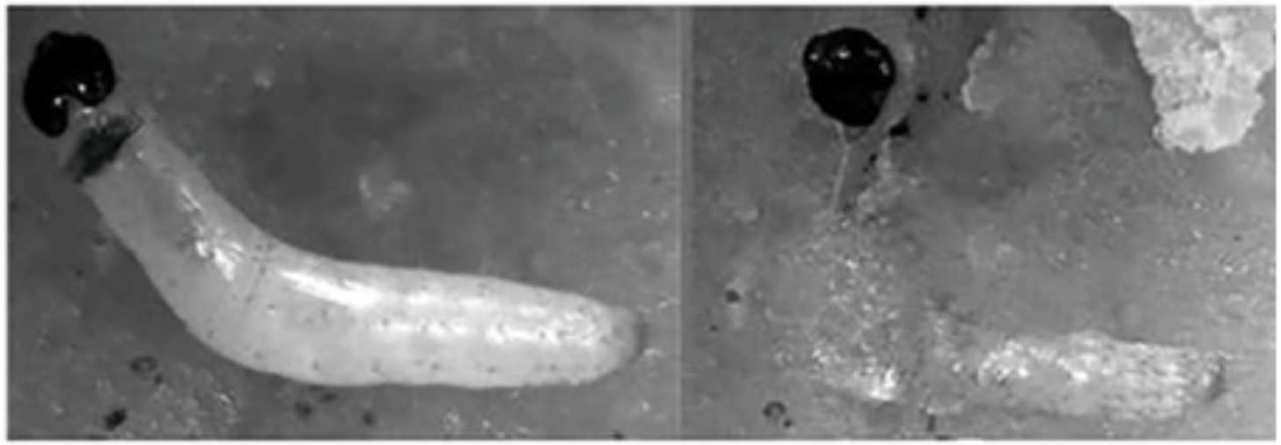
KdrKis

	24h	48h
Thc 0.01mg/L + Thm 0.02mg/L	8%	38%



From chemical to biological synergistic agents in mosquito control

Highlighted facts: insect viruses used as microbial pest control agent



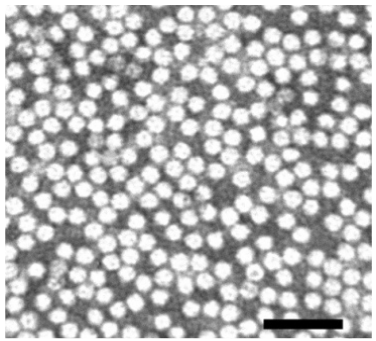
Codling moth *Cydia pomonella* infection by the granulovirus *Cp*GV at larval stage

Deshayes C, Siegwart M, Pauron D, Froger JA, Lapied B, Apaire-Marchais V. Curr Med chem. 2017;24(27):Review.

But…GV as bioinsecticides could be limited particularly by their slow speed in killing the targeted insects. It can take days to weeks from virus application to insect death

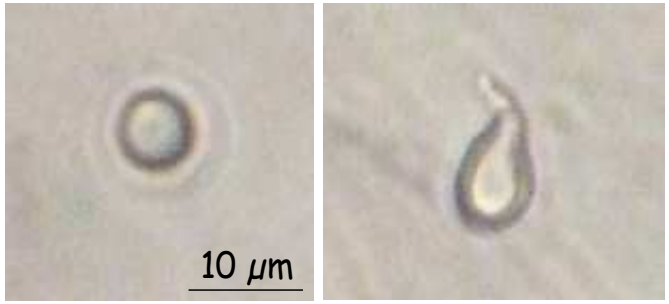
From chemical to biological synergistic agents in mosquito control

Densovirus
(*Junonia coenia* (*Jc*DV)

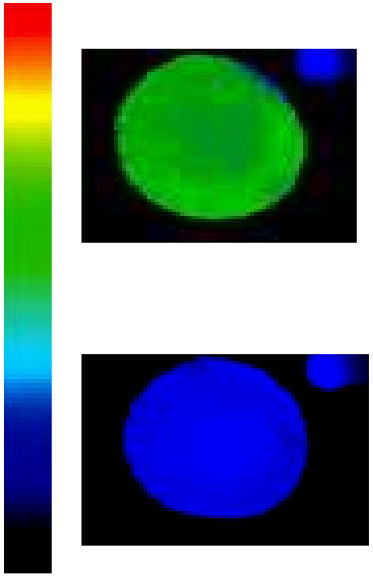


Anopheles gambiae

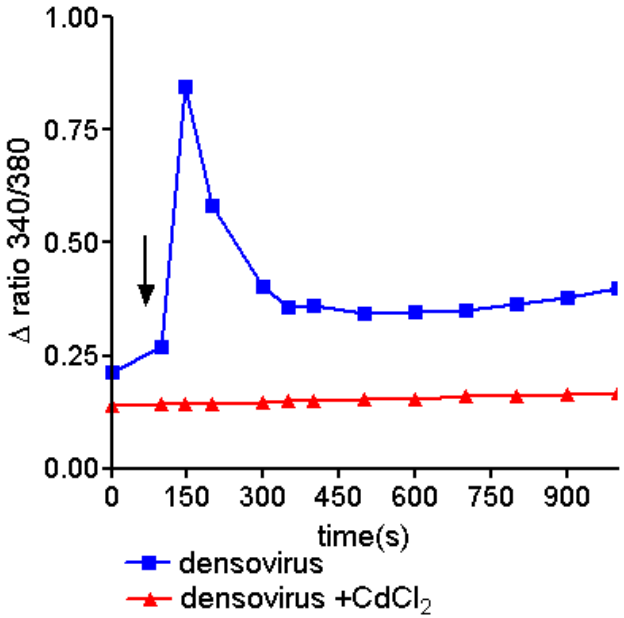
Isolated mosquito neurons
(*Anopheles gambiae*)



high calcium



low calcium



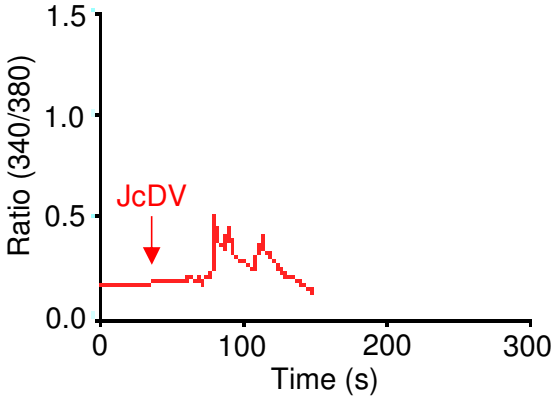
Calcium imaging

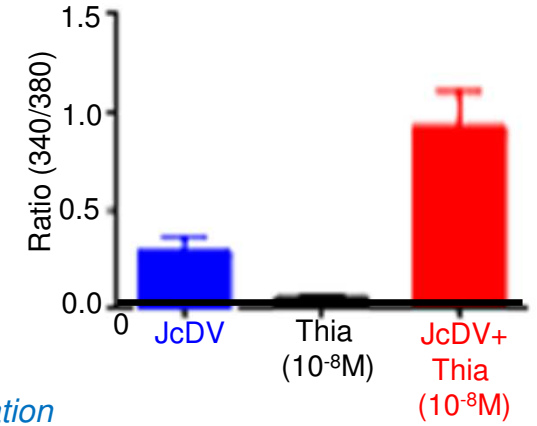
JcDV induces intracellular calcium rise in isolated mosquito neurons

V.Apaire-Marchais, M. Ogliastro, F. Chandre, C. Pennetier, V. Raymond & B. Lapied
Env. Microbiol. Reports 8(2), 2016

From chemical to biological synergistic agents in mosquito control

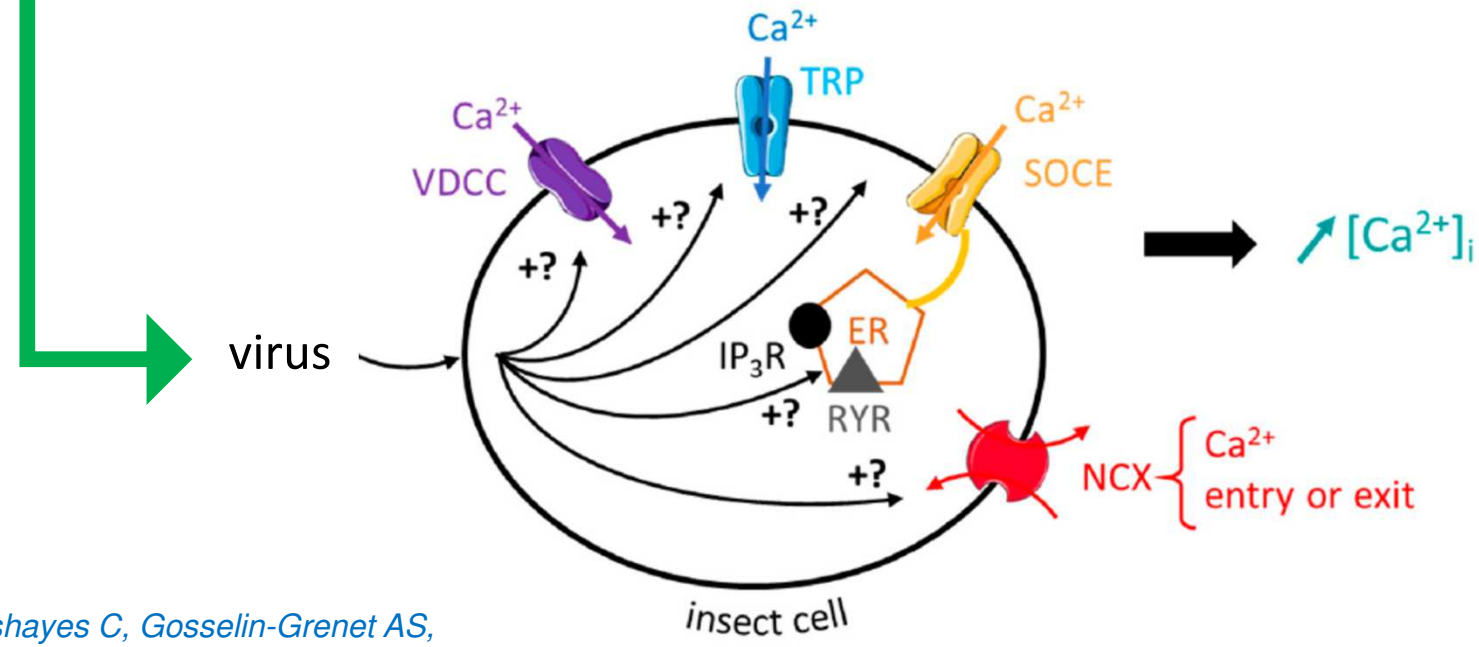
JcDV induces **multiphasic** intracellular calcium rise in isolated mosquito neurons





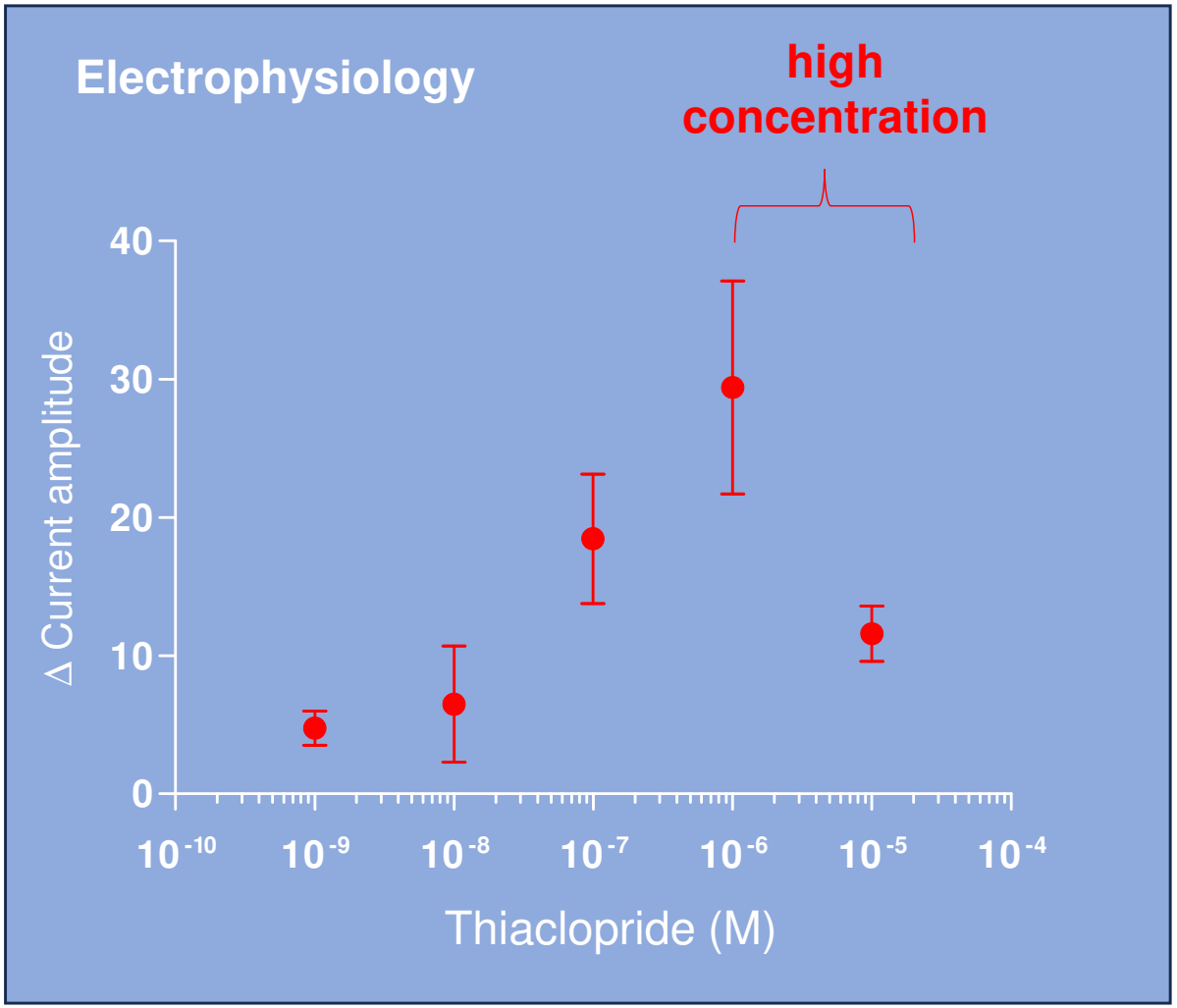
J. Froger SiFCIR, personnal communication

Complex calcium signaling pathways



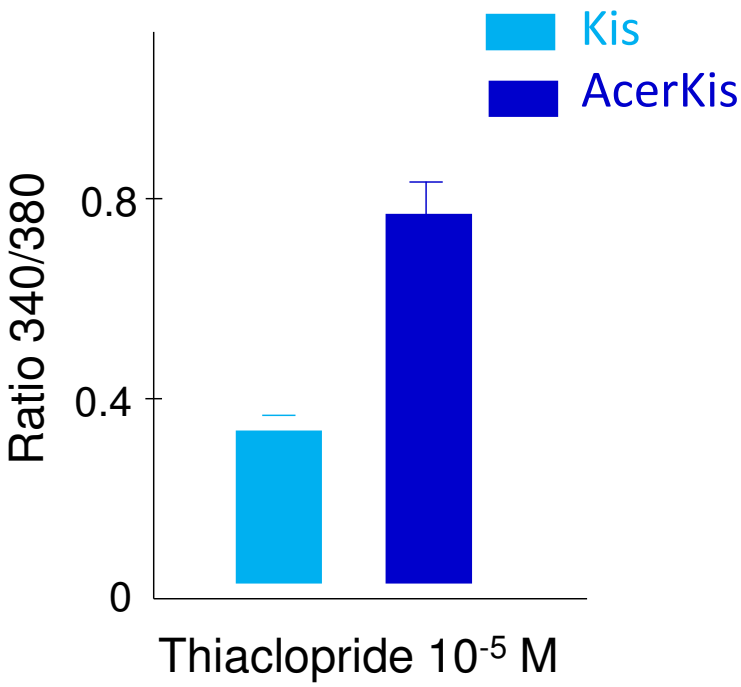
Modified from Deshayes C, Gosselin-Grenet AS, Ogliastro M, Lapied B, Apaire-Marchais V. Viruses. 2022;14(5)

Densovirus as synergistic agents in mosquito control



Biphasic concentration-response semi-logarithmic curve

Calcium imaging

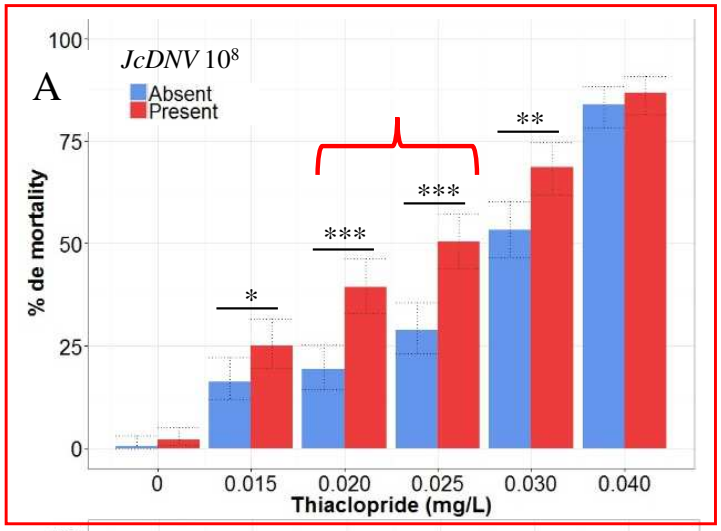


Thiaclopride increases intracellular calcium concentration

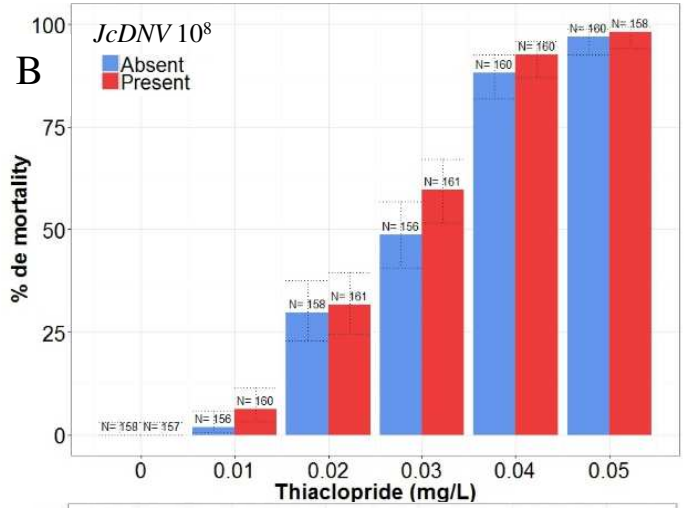
Intracellular calcium rise needs to be controled

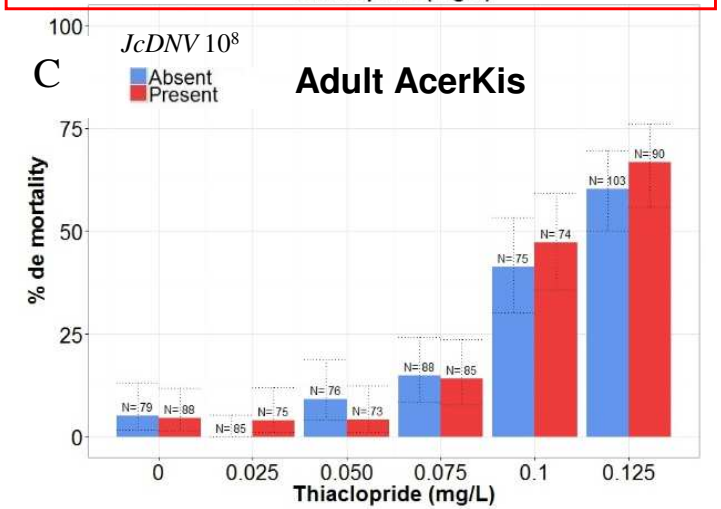
Densovirus as synergistic agents in mosquito control

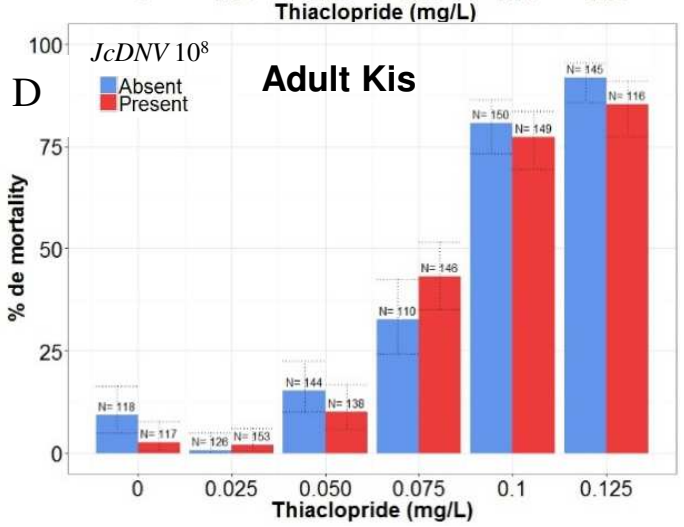
Larvae AcerKis



Larvae Kis







Ecotoxicology – Tests on aquatic organisms

Non observed synergistic effect, whatever the dose of thiacloprid tested, but also with combined densovirus/insecticide on daphnia.

Daphnia magna
copepodes

Nombre d'animaux: 15	24 h					48 h				
	replicate					replicate				
treatment	A	B	C	D	E	A	B	C	D	E
water control	0	0	0	0	0	0	0	0	0	0
ethanol control*	0	0	0	0	0	2	0	0	0	0
Thiacloprid 0.01 mg/L	0	0	0	0	0	0	0	0	0	0
Thiacloprid 0,02 mg/L	0	0	0	0	0	1	0	0	2	1
Thiacloprid 0,03 mg/L	0	0	0	0	0	0	1	0	1	1
Thiacloprid 0,04 mg/L	0	0	0	0	0	1	4	1	0	0
Virus1E8 équivalent génome/µl	0	0	0	1	0	0	0	0	1	0
Virus1E8 + Thiacloprid 0.01 mg/L	0	0	0	0	0	0	0	0	0	0
Virus1E8+ Thiacloprid 0,02 mg/L	1	0	0	0	0	1	0	0	0	1
Virus1E8 + Thiacloprid 0,03 mg/L	0	0	0	0	0	2	2	1	1	0
Virus1E8 + Thiacloprid 0,04 mg/L	0	0	1	0	0	3	0	3	2	0

on AcerKis larvae
0.02-0;03mg/L

«*Daphnia sp.*, immediate immobilisation larval bioassays (48h)»
DOI 10.1787/9789264069954-fr

OECD Guideline

Ecotoxicology – Tests on bee larvae

Document guide OCDE n° 239 :
Abeilles (*Apis mellifera*)
Tests de toxicité sur larves
Expositions répétées.

Etudes écotoxicologiques d'une exposition chronique à l'association densovirus / thiaclopride sur les larves d'abeilles domestiques au laboratoire

Trois colonies d'abeilles domestiques *Apis mellifera L.* de même race (Buckfast)

Les larves sont exposées oralement et de façon chronique

Les mortalités sont relevées de J+4 à J+8 et à J+15 durant la nymphose

Le taux de mortalité larvaire et nymphale est calculé en comparant le nombre d'abeilles mortes durant la période J+3 à J+8 (pour la période larvaire) et à J+15 (pour la période nymphale)

La combinaison densovirus/insecticide aux doses testées (sur larves AcerKis) n'a pas entrainé d'effets létaux accrus durant le développement larvaire et l'émergence adulte comparée aux abeilles exposées à l'insecticide seul.

Merci pour votre attention…!

OUEST VALORISATION
Ressources d'innovation

Vectopole Sud

anses
agence nationale de sécurité sanitaire
alimentation, environnement, travail

SFCIR
Signalisation Fonctionnelle des
Canaux Ioniques et Récepteurs

RÉSEAU SATT

MiVEGEC
Maladies Infectieuses et Vecteurs
Écologie, Génétique, Évolution et Contrôle

UMR IRD/CNRS/UM
Montpellier

UMT PrADE

UMT PrADE
Avignon

UNION EUROPÉENNE

Région PAYS de la LOIRE

CE PROJET EST COFINANCÉ PAR
LE FONDS EUROPÉEN DE DÉVELOPPEMENT RÉGIONAL

U3E Unité Expérimentale d'Ecologie et d'Ecotoxicologie aquatique

UMR UE3 INRAE
Rennes

DGIMI
Diversité, Génomes & Interactions
Microorganismes Insectes