PRESENTATION DU PROJET ERC REVOLINC
Jeremy BOUYER
Thierry BALDETT
UMR ASTRE Cirad/INRA

Journées EMBA, 13 juin 2018 UM

thierry.baldet@cirad.fr
Toxicity and ecotoxicity of insecticides

Reduction of authorized insecticides (by 75% in Europe)

Impact of Vector Control?

Insecticide resistance

Biological invasions Vectors/Pests Pathogens

Environmental changes (local/global)

Diseases outbreaks Nuisance Health impact Economical loses

Strong Need for INNOVATION

THE CHALLENGE
The range of insecticides available for vector control

... worse in Europe!

<table>
<thead>
<tr>
<th>Vector Control Intervention type</th>
<th>Number of WHO recommended insecticide families</th>
<th>Insecticide families</th>
<th>Number of molecules recommended in each family</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indoor Residual Spraying</td>
<td>4</td>
<td>Pyrethroids</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Carbamates</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Organophosphates</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>DDT</td>
<td>1</td>
</tr>
<tr>
<td>Long-lasting insecticide treated nets</td>
<td>1 (+1)</td>
<td>Pyrethroids</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(Piperonyl butoxide)</td>
<td></td>
</tr>
<tr>
<td>Space Sprays</td>
<td>2 (+1)</td>
<td>Pyrethroids</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Organophosphates</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(Piperonyl butoxide)</td>
<td></td>
</tr>
<tr>
<td>Larvicides</td>
<td>5</td>
<td>Bacterial larvicide</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Benzoylureas</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Juvenile Hormone mimics</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Organophosphates</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Spinosyn</td>
<td>1</td>
</tr>
</tbody>
</table>

Resistance to pyrethroids and DDT is now widespread in many mosquito populations

Source: Justin McBeath ISNTD Bites – March 19th 2015
THE PRINCIPLE

SIT

Boosted SIT

Bouyer & Lefrançois
Trends Parasitol 2014
INSECT MODELS
PUBLIC HEALTH

Mosquitoes
*Aedes albopictus*
An invasive species in Europe

Nuisance in Late Summer & Autumn
Risk of Outbreaks of Aedes-borne viral diseases

2010

2015
Moustique tigre: « Appel à la vigilance » dans 42 départements métropolitains il y a 4 jours par la Direction Général de la Santé
Situation au 1er Janvier 2018

« surveillance renforcée »
Other Targets

**AGRICULTURE**

Fruit flies
*Ceratitis capitata*

**PUBLIC & ANIMAL HEALTH**

Tsetse flies
*Glossina palpalis gambiensis*
Research Partners

VectopoleSud

LISIS

UP EM

ISEM

Inserm

University of Manitoba

University of Manitoba

Tragsa

REvolutionLINC

FAO/IAEA Division of Nuclear Techniques in Food and Agriculture

Astre

IRD

Cirad

Intertryp
WHAT IS NEEDED TO RUN BOOSTED SIT?
WHAT WILL WE ADDRESS?
Pipeline of R & D for vector control tools/strategies

Transfer of biocides during mating and impact on female fertility (PP & Densovirus)

Contamination technique of males

Resilience on males & transfer to females during mating

Impact on female fertility (+ transfer to breeding sites)
Laboratory Evaluation I
Survival of sterile males
of *Glossina palpalis gambiensis*
treated with a new formulation of PP (F15)

No impact of PP (F15) on male survival

Master2 Lison Laroche
Laboratory Evaluation II
Persistency of PP (F15) on treated males of *Glossina palpalis gambiensis*

Fast initial decline of PP followed by a level off around 100ng during 10 days
Laboratory Evaluation III
Transfer of PP from males* to females of *Glossina palpalis gambiensis*

*3h after treatment by PP (F15)

To be followed: Impact of PP transferred on female fertility...

Amount of pyriproxyfen per female (ng)

<table>
<thead>
<tr>
<th>Nb of contacts without mating</th>
<th>Amount (ng)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.35</td>
</tr>
<tr>
<td>1</td>
<td>15.35</td>
</tr>
<tr>
<td>2</td>
<td>17.82</td>
</tr>
<tr>
<td>3</td>
<td>17.65</td>
</tr>
<tr>
<td>11</td>
<td>18.26</td>
</tr>
<tr>
<td>Accouplement 30 min</td>
<td>42.78</td>
</tr>
<tr>
<td>Accouplement 40 min</td>
<td>71.45</td>
</tr>
</tbody>
</table>

Master2 Lison Laroche
Laboratory Evaluation I
Survival of males of *Aedes albopictus* treated with a new formulation of PP (F15)

Reduced survival of treated males (+50% mortality in 14 days)
Laboratory Evaluation II
Persistency of PP (F15) on treated males of *Aedes albopictus*

Amount of PP per individual (ng)

<table>
<thead>
<tr>
<th>Time after treatment</th>
<th>Amount of PP per individual (ng)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T0 (0 heure)</td>
<td>183.48</td>
</tr>
<tr>
<td>T1 (3 heures)</td>
<td>132.01</td>
</tr>
<tr>
<td>T2 (6 heures)</td>
<td>107.67</td>
</tr>
<tr>
<td>T3 (24 heures)</td>
<td>62.01</td>
</tr>
<tr>
<td>T4 (48 heures)</td>
<td>42.03</td>
</tr>
<tr>
<td>T5 (4 jours)</td>
<td>16.76</td>
</tr>
</tbody>
</table>
What about associating SIT to DENSOVIRUSES?
A Release of sterile males coated with a densovirus

B Adult horizontal transmission to the breeding site

C Multiplication of the densovirus plus larval horizontal transmission

D Adult vertical transmission to new breeding sites
### Mosquito Densovirus

High specificity

<table>
<thead>
<tr>
<th>Animal species</th>
<th>Number of individuals</th>
<th>Developmental stage</th>
<th>Route of infection</th>
<th>Pathological effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>Insects</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ae. aegypti</td>
<td>1140</td>
<td>Instar I–IV larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. albopictus</td>
<td>550</td>
<td>Instar I larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. togoi</td>
<td>450</td>
<td>Instar I larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. vexans</td>
<td>419</td>
<td>Instar I, II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. geniculatus</td>
<td>233</td>
<td>Instar I larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. caspius dorsalis</td>
<td>905</td>
<td>Instar I, II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. cantans</td>
<td>440</td>
<td>Instar II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Ae. caspius caspius</td>
<td>90</td>
<td>Instar II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>C. pipiens pipiens</td>
<td>915</td>
<td>Instar I, II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>C. p. molestus</td>
<td>641</td>
<td>Instar I larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>C. annulata</td>
<td>315</td>
<td>Instar I, II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>An. maculipennis</td>
<td>548</td>
<td>Instar I, II larvae</td>
<td>PO</td>
<td>+</td>
</tr>
<tr>
<td>Chironomus sp.</td>
<td>142</td>
<td>Larvae</td>
<td>PO</td>
<td>–</td>
</tr>
<tr>
<td>M. domestica</td>
<td>335</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>P. regina</td>
<td>210</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>A. mellifera</td>
<td>200</td>
<td>Adult</td>
<td>PO</td>
<td>–</td>
</tr>
<tr>
<td>G. mellonella</td>
<td>450</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>B. mori</td>
<td>115</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>A. crataegi</td>
<td>184</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>M. neustria</td>
<td>270</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>P. dispar</td>
<td>225</td>
<td>Instar III, IV larvae</td>
<td>PO, IL</td>
<td>–</td>
</tr>
<tr>
<td>Crustaceans</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Daphnia sp.</td>
<td></td>
<td>Adults and youth</td>
<td>PO</td>
<td>–</td>
</tr>
<tr>
<td>Cyclops sp.</td>
<td></td>
<td>Adults and youth</td>
<td>PO</td>
<td>–</td>
</tr>
<tr>
<td>Worms</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lumbricus sp.</td>
<td>50</td>
<td>Adults</td>
<td>SC</td>
<td>–</td>
</tr>
</tbody>
</table>

Carlson et al. 2006
Advances in virus research
<table>
<thead>
<tr>
<th>DVs moustiques</th>
<th>Origine des DVs</th>
<th>Production en cellules (virus équivalent génome – veg/µl) sécrétion dans le surnageant</th>
<th>Test d’infection en larves</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Aedes albopictus</em></td>
<td><em>C6/36P chroniquement infectées</em> Jousset <em>et al.</em>, 1993</td>
<td>2,4 x 10^8</td>
<td>En cours</td>
</tr>
<tr>
<td><em>AalDV2</em></td>
<td>Récolte des surnageants de culture</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Aedes aegypti</em></td>
<td><strong>Clone viral</strong> origine: synthèse de genes</td>
<td>C6/36</td>
<td>3,4x10^6</td>
</tr>
<tr>
<td><em>AaeDV1</em></td>
<td>Production de virus par transfection de culture de cellules de moustiques</td>
<td>AAG2</td>
<td>1,2x10^3</td>
</tr>
<tr>
<td></td>
<td><strong>Clone viral</strong> origine: clonage génome viral</td>
<td>C6/36</td>
<td>1,3 x 10^1</td>
</tr>
<tr>
<td></td>
<td>Production de virus par transfection de cultures de cellules de moustiques</td>
<td>AAG2</td>
<td>3,8x10^6</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>En cours</td>
</tr>
</tbody>
</table>
**Aedes albopictus** Densovirus 2 produced from infectious plasmid
Infection in *Aedes albopictus* and *Aedes aegypti* larvae

**Protocol:**
- 1 larvae L1, P96 well, +/- virus
- Transfert 48hpi in P24 well plate + food

**Survival curve for *Ae. albopictus* larvae (Italian strain) infected with AalDV2 virus**

**Protocol:**
- 1 larvae L1, P96 well, +/- virus
- Virus +/- FB28 0,1%
- Transfert 48hpi in P24 well plate + food

**Survival curve for *Ae. aegypti* larvae (Bora strain) infected with AalDV2 virus**
Aedes albopictus Densovirus 2 produced from C6/36 chronically infected cell line

Infection in Aedes albopictus larvae

Protocol:
- 100 larvae L1, 5 ml cell culture
- Transfert 48hpi in 300 ml water + food
- Analysis 14 dpi

AalDV2 effects:
- ↗ mortality
- ↘ development
- ↗ cannibalism
Impact of boosted SIT in semi-field and field trials with the best biocide 
\((Ae. \ albopictus \ & \ C. \ capitata)\)
Relative impacts of SIT and boosted-SIT on population dynamics and resilience

\[ \kappa = \frac{1 - \Phi^l (1 - \xi \psi^T (1 - \eta \Phi^u))}{\xi \Phi^l \psi^T} \]

*Deterministic models*

*Individual-based models*
Preliminary models with Pyriproxyfen

Elimination with minimised $R_{\text{total}}$:
- $R_{\text{SIT opt}} = 2528$
- $R_{\text{BSIT opt}} = 1214$

David Pleydell
Preliminary models with Pyriproxyfen

David Pleydell
Population dynamics of *Ae. albopictus* in La Réunion (AlboRun)

Spatial model
Accounting for Climate
Validated on entomological data
Used by Agence Régionale de Santé (ARS) to target monitoring and control

PhD Marion Haramboure
First results of integrating SIT to the model

200,000 sterile males released weekly for 3 months
With a constant climate, return to equilibrium only 1.5 month after releases

Perspectives

Boosted SIT
Study various release scenarios
Impact of Climate on vector control

PhD Marion Haramboure
Mass rearing and irradiation procedures
Sex separation method (female elimination) and quality control

Sexing of mosquitoes and handling procedures

Quality control of produced strains

2 genetic strategies to be compared to Tragsa Laser sorter:
- Development of non-transgenic GSS
- RNAi sexing

PhD student (Start in July 2018)
Development of an automatic release machine

Regulatory issues and social acceptability of boosted SIT

Risk assessment → Regulatory issues → Social acceptability

Review of regulatory issues, technical constraints and social acceptability of genetic control in Europe
Ministère de la Transition Écologique et Solidaire
Direction générale de la prévention des risques

Ministère des Solidarités et de la Santé
Direction générale de la santé

Paris, le 10 OCT. 2017

Objet : Saisine du Haut Conseil de Santé Publique concernant l’élaboration de recommandations pour autoriser le lâcher de moustiques stériles à des fins de lutte antivectorielle.

Audition des experts
Saisines en phase finale de rédaction
Présentation des saisines pour décision auprès HCSP & AFB (fin juin 2018)
Revolinc & social sciences

• La Reunion island

• 29 Jan - 16 Feb. 2018

• Survey by observations

• Meeting of scientific actors from CIRAD, Revolinc, IRD, TIS-La Réunion, ARS and collaborators on CYROI platform of Saint-Denis in the context of phase 2 preparation meeting of TIS-project

PhD Kamil Ghouati
Revolinc & social sciences

• Vienna, Seibersdorf (Austria)

• 26 - 29 mar. 2018

• Survey by interviews & observations

• Meeting of members from Insect Pest Control Lab (FAO / IAEA Programme on Nuclear Techniques in Food and Agriculture)
Revolinc & social sciences

- Vienna, Seibersdorf (Austria)
- 23 - 30 April 2018
- Survey by interviews & observations
- Meeting with European stakeholders in charge of mosquito control during a meeting on stakeholder engagement in mosquito SIT projects organized by IAEA
1. Identification of **actors** in the different sites: project leaders, stakeholders, collaborators, funding agencies, civil society, public(s), beneficiaries...

2. Identification of actors **logics, categories** involved to account for the action & social dimension of SIT projects, on scientific & technical thematics in the society, on technical democracy and on research participation: **education, sensitization, communication, vulgarization**, etc.

3. Study of **social relationships** and their dynamics along with ongoing projects deployment
Thanks!