Development and evaluation of transgenic insects for use in the control of vector-borne disease

Luke Alphey
vector-borne disease – a global unmet health challenge

Dengue: estimated 390m infections pa, increasing $5 Bn burden of cost
No specific medication or vaccine yet
Chikungunya, Yellow Fever – same vector
*Aedes aegypti*: alien invasive species in most countries

Avian malaria and avian pox in Hawaii
Multiple unique bird species threatened with extinction
Single vector species (*Culex quinquefasciatus*)

“Today, dengue ranks as the most important mosquito-borne viral disease in the world. Everywhere the human and economic costs are staggering”
*Dr Margaret Chan, 2012*  
*Director General, WHO*
vector-borne disease and conservation

Avian malaria and avian pox in Hawaii
Multiple unique bird species threatened with extinction
Single vector species (*Culex quinquefasciatus*)

Worthwhile target?

- **Major conservation issue**
  - avian malaria (*Plasmodium relictum*) threatens multiple native species
- **Human biting nuisance**
  - but not necessarily in the same locations
- **Potential vector of other diseases**
  - e.g. fowl pox, West Nile virus
- **Introduced species**
  - no mosquito species are native to Hawaii
- **Few alternative control measures available**

Amakihi with debilitating pox lesion on foot (Photo: USGS)
dengue control
mosquito reproduction
RIDL “genetically sterile” males
RIDL – “genetic sterility”

RIDL insects are genetically sterile
Repressible
Release homozygous males

Bi-sex lethal

Female-specific lethal

Time of death

introgression of genes through male line
[resistance management]

Thomas et al. 2000 Science 287: 2474-6
Gong et al. 2010 Nat Biotech 23: 453-6

Alphey et al. 2007 J. Econ. Ent. 100: 1642-9
Alphey et al. 2009 J. Econ. Ent. 102: 717-32
classifying genetic control strategies

Population suppression
  Goal: reduce numerical size of pest population

“Population replacement” or
  “Refractory insect strategy”
  Goal: change pest population to less harmful form

Self-limiting
  Modification will be eliminated from population unless maintained by periodic releases
  – Sterile insect methods

Self-sustaining (invasive)
  Modification will persist and spread in population (and potentially beyond)
  – Gene drive systems
classifying genetic control strategies

RIDL – “genetic sterility”

RIDL insects are genetically sterile
Repressible
Release homozygous males

Bi-sex lethal
Female-specific lethal

Time of death

introgression of genes through male line
[resistance management]

Thomas et al. 2000 Science 287: 2474-6
Gong et al. 2010 Nat Biotech 23: 453-6
Alphey et al. 2007 J. Econ. Ent. 100: 1642-9
Alphey et al. 2009 J. Econ. Ent. 102: 717-32
combinatorial control

Sex

Conditional

Tissue, time

RIDL

No effect

dead

ATG

Females

effector

Males

Stop

death

tTA

promoter

Antidote (Tc)

tetO

effector

No effect

dead

tTA

promoter

www.pirbright.ac.uk
Act4-tTA + tetO-DsRed

- tTA
- effector (DsRed)

Act4 promoter

tetO + promoter

OX3545F

Flight muscles on
OX3604C RIDL mosquitoes

Males

Flightless mosquitoes cannot survive in wild (or find hosts).
Unable to mate even in laboratory.

Females

Males have normal flight ability, as have females given antidote as larvae.
phased testing

- Molecular characterisation
- Genetic and phenotypic stability
- Bionomic characteristics
- Mating competitiveness
- Mating compatibility
- Insecticide resistance
- Suppression trials

- Mating competitiveness in semi-natural conditions
- Cage suppression trials

- Validation in field conditions

- Contained
- Semi-field
- Open field population suppression
bringing new technology to the field
presentations
TV and radio
local festivals
house visits

Houses Visited in Door to Door communication campaigns + Routine monitoring

- Visited houses
- Closed/uninhabited houses
- Uninhabited areas
- Treatment area boundary

100 m
operations
According to the results, a majority (61%) supports [the mosquito control agency] using GE technology in Key West to control mosquitoes that spread dengue. More important, perhaps, is that just 18% say they are opposed, while another 21% are neutral. Most respondents also consider GE mosquito technology safe (81%), which is a slightly higher percentage saying it is safe than said that about using chemicals and insecticides (73%).

Correlates of support: “(1) rating GE as safer than using chemical and insecticides, greater trust in (2) scientists at private businesses and (3) [the control agency] to manage any risks of GE, (4) more worrying about dengue fever, (5) thinking mosquitoes are bigger nuisance, and (6) having more formal education”
bringing new technology to the field
Cayman field trial 2010

OVITRAP INDEX treated (A) & non-treated (C)

- Trial was complete success; all endpoints met
  - Clear suppression from early August
- Sustained release of RIDL OX513A males can suppress a field population of *Aedes aegypti* mosquitoes
  - Maximum degree of suppression limited by immigration
- GM mosquitoes can perform successfully in the field

Harris et al. 2012
Nat. Biotech. 30:828-30
RIDL is effective in multiple settings

<table>
<thead>
<tr>
<th>Treated Site</th>
<th>Control Site</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>East End (A)</td>
<td>East End (C)</td>
<td>Cayman Islands</td>
</tr>
<tr>
<td>Itaberaba (T2)</td>
<td>Itaberaba (U)</td>
<td>Brazil</td>
</tr>
<tr>
<td>Mandacaru</td>
<td>Carnaiba</td>
<td>Brazil</td>
</tr>
<tr>
<td>Mandacaru</td>
<td>Maniçoba</td>
<td>Brazil</td>
</tr>
</tbody>
</table>

Relative Eggs/trap (Treated site/Control site)

- Before Treatment
- After Treatment

96%, 94%, 99%, 95%
summary

Widespread use against dengue depends on efficacy, and public and regulatory approval

- **Efficacy**: periodic release of RIDL males suppressed wild target population
  - No conventional control for *Aedes aegypti*
  - Full suppression in 2-3 months
  - Target population recovered only slowly post-release

- **Public approval**: initial indications positive at trial sites
  - Intended goal of dengue control recognised as desirable
  - May vary by country (culture, prior experience of GMOs, etc)

- **Regulatory approval**: independent regulatory authorities have approved use
  - Field use in Cayman Islands, Malaysia, Brazil (& USA for pink bollworm)
  - Import permits in many additional countries
where to use?

- **Species-specific:** released insects mate only with own species
  - Aggressive gene drive systems may penetrate species complex with incomplete reproductive isolation
  - Environmentally friendly
  - How many vector species?

- **Technical** (lab rearing, mating, single species)

- **Case-by-case environmental analysis**
  - Native or invasive species?

- **Self-dispersing and target-seeking:**
  - Based on mating behaviour and mobility of insect
  - Need to consider reproductive behaviours and ecology

- **Sterile males – simple precise suppression**
- **Gene drive systems – population level genetic modification**
Thank you