Can we immunise honey bees against virulent viruses?

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Factors affecting honey bee health

Pathogens & parasites

Management

Globalization

Xenobiotics

Climate change

What can we do about bee disease?
Parasites and pathogens are a major cause of colony loss and suboptimal honey bee health

Are there genetic mechanisms that could reduce the impact of viruses in honey bees?

Honey bee viruses and how to stop them
Viruses

**GENOME**
Genetic material

**PROTEIN**
Capsid/Shell

Always changing and mutating
Eg. Influenza, new vaccine every year

Viruses

Virus
100nm

Bacteria
2000nm

Animal Cell
10,000nm
VIRUSES

...like unwanted house-guests...
Viruses

**GENOME**
RNA or DNA

**RNA:**
Ribonucleic Acid

**DNA:**
Deoxyribonucleic Acid

- Single-stranded negative-sense RNA: eg. Influenza, Ebola
- Single-stranded positive-sense RNA: eg. Dengue fever, Common cold
- Double-stranded DNA: eg. Herpes, Smallpox

Honey bee viruses

- Historically: around 24 viruses
- **Australia** has 5 common viruses:
  - Black queen cell
  - Sacbrood
  - Lake Sinai I and II
  - Israeli Acute paralysis
- **New viruses** discovered all the time

Dr John Roberts, CSIRO
Session 8B (later today)
Honey bee viruses: Picornavirales

- Deformed Wing Virus
- Slow Bee Paralysis Virus
- Sacbrood Virus
- Black queen cell virus
- Acute Bee Paralysis Virus

Virus Classification

<table>
<thead>
<tr>
<th>Genome type</th>
<th>Honey bees</th>
<th>Insects</th>
</tr>
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<tbody>
<tr>
<td>Single-stranded negative-sense RNA</td>
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<tr>
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<td>Double-stranded DNA</td>
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</tbody>
</table>
Who’s really in control?

Us?

Parasites

Bacteria

Viruses

The Microbiome

We are FULL of microorganisms

that impact how our body works
The Microbiome

What’s living inside our bees?

The good, the bad and the ugly
The bee gut

- 8-10 core species of bacteria in the gut, with key roles in health and development

Dr Julia Jones, Uppsala University
Session 3A (yesterday)

Honey bee diseases

**VIRUSES**
- Deformed wing
- Kakugo
- Black Queen Cell
- Sacbrood
- Cloudy wing
- Israeli Acute Paralysis
- Kashmir Bee
- Acute Paralysis
- Chronic Paralysis
- Slow paralysis
- Lake Sinai 1 & 2
- Apis Rhabdo 1 & 2
- etc......

**FUNGI**
- Nosema
- Chalkbrood

**BACTERIA**
- American foulbrood
- European foulbrood
Arrival of a new pest

*Varroa destructor*: Parasitic mite

Jumped from *Apis cerana* → *Apis mellifera*

Prof Madeleine Beekman, University of Sydney, Session 5A
Impact of Varroa

• Wounding and weakening
  • Damage to the cuticle
  • Feeds on haemolymph

• Vector for viruses
  • Spreads viruses
  • Viruses replicate in mites

Gilles San Martin

Prof Madeleine Beekman,
University of Sydney, Session 7B
Bees brought to their knees

Viruses have changed since the arrival of Varroa

The main culprit: **Deformed wing virus (DWV)**

- Varroa arrives: DWV levels rapidly increase
- Left untreated for mites, hives die in 2-3 years

Deformed wing virus: Global pandemic

Global Honey Bee Viral Landscape Altered by a Parasitic Mite

Deformed wing virus is a recent global epidemic in honeybees driven by Varroa mites
So how do we stop viruses?  
…get rid of mites…?

Ways to get rid of mites

- Chemical treatments
  - In hive residues bad for bees
  - Mites can become resistant

- Natural selection of Varroa-tolerance or resistance
  - Over time, bees learn to live with, or get rid of, mites

Dr Jody Wu-Smart, University of Nebraska-Lincoln, Session 4A
Dr Medhat Nasr, Ministry of Alberta Agriculture and Forestry, Session 5A
Prof Madeleine Beekman, University of Sydney, Session 5A
Worldwide evolution of Varroa-resistance/tolerance

Colonies survive mite infestations without chemical intervention

1. Breeding programs
2. “Live and let die”

But... are there viruses in Varroa-resistant bees?

Wilfert et al. (2016) Deformed wing virus is a recent global epidemic in honeybees driven by Varroa mites. Science, 351(6273), pp. 594-7
Adapted from: Locke, B., 2015 Natural Varroa mite-surviving Apis mellifera honeybee populations. Apidologie; 47: 467-482.
Deformed wing virus is a recent global epidemic in honeybees driven by Varroa mites. Science 351(6273), pp. 594-7

Adapted from: Locke, B., 2015 Natural Varroa mite-surviving Apis mellifera honeybee populations. Apidologie: 47: 467-482.
Virus levels in bees can be pretty high!

![Bar chart showing virus levels in bees across different regions.](chart1.png)

- New Zealand
- South Africa
- Mainland Robben Island
- USA
- Arnot Forest
- Tongatapu & 'Eua
- Hawaii
- Africanised
- Brazil
- Varroa-absent
- Collapsing

DWV is absent in some populations surviving Varroa

![Bar chart showing DWV levels in bees across different regions.](chart2.png)

- New Zealand
- South Africa
- USA
- Mainland Robben Island
- Arnot Forest
- Tongatapu & 'Eua
- Hawaii
- Africanised
- Brazil
- Varroa-free

Note: The charts illustrate the percentage of reads aligned to DWV across various regions and population conditions.
High levels of DWV in other populations surviving Varroa

New Zealand Collapsing

Netherlands

Varroa UK USA Tonga Hawaii

Percent of reads aligned to DWV

0.00 25.00 50.00 75.00 100.00

1 2 3 4 1 2 3 4 5 6 1 2 3 4 1 2 3 4 5 1 2 3 4

Populations surviving Varroa

7 new RNA viruses
First negative sense RNA viruses in bees

<table>
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<tr>
<th>Virus Type</th>
<th>Netherlands</th>
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Apis Rhabdovirus 1

Geographically widespread, multi-host virus


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We don't know what the new viruses do
What can we do about it?

• Varroa has increased virus levels in bees
  • Long term effects on colony survival

• Impact of viruses remains high in most places
  • Varroa-resistance is not a complete solution
  • *Can we reduce virus levels in bees by other mechanisms?*

The Insect Immune System

• Insect immunity - less sophisticated than humans
  • No antibodies
  • Can’t give them the bee version of a flu shot

• Bees rely on general immune pathways
  • At the colony level: **Hygienic behaviour**
  • Immune genes and pathways

Dr Brock Harpur, *University of Toronto*, Session 3B
Social immunity

- Hygienic behaviour
  - Worker bees detect sick or dying brood and remove them from the hive
  - Worker bees groom mites from other adult worker bees

[Images of bees and honeycomb with text: Good hygiene, Bad hygiene]

[Source: https://beeinformed.org/2011/07/25/hygienic-behavior/]

The Insect Immune System

- Insect immunity- less sophisticated than humans
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- Bees rely on general immune pathways
  - At the colony level: Hygienic behaviour
  - Immune genes and pathways
  - The Microbiome is important
For example, in other insects...

What is Wolbachia?

- Bacteria that lives inside cells
- Infects about half of all insect species
- Manipulates insect hosts to increase transmission
- Provides resistance to viruses in flies and mosquitoes
‘Eliminate Dengue’

• Dengue fever: WHO #1 mosquito-borne disease
  • 30-fold increase in past 10 years
• Aedes aegypti mosquito:
  • vector of Dengue, Chikungunya, Zika virus
• Global team spearheaded by Australian labs
• Use a natural mechanism to prevent spread of Dengue fever: infect mosquitoes with Wolbachia

The Wolbachia method

Images: Perran Ross
The Wolbachia method

Wolbachia in bees?

- Present in African honey bee subspecies
- Little evidence for Wolbachia elsewhere in Apis
  - Antibiotic treatments will remove Wolbachia

Can Wolbachia provide virus resistance to honey bees?
Can we immunise honey bees with bacteria?

Wolbachia transinfection:
- Contained in our quarantine lab
- Microinjection of honey bee eggs
- Injection into queen pupae → ovaries
- Next steps: test viruses in positive pupae

WATCH THIS SPACE!

Prof Phil Lester, Victoria University of Wellington; Session 8B
Take home messages

1. Varroa + viruses = **Bad News**

2. We are in a unique position to prevent damage caused by viruses as seen elsewhere in the world, if Varroa mites arrive

3. Protective bacteria could provide a new way to reduce viruses

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