The Vulnerability of Aotearoa to Introduced Mosquito Species
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1. Focus species

Endemic: Culex pervigilans
- Endemic to and wide spread within New Zealand
- Bird-biter
- Low vector competence

Exotic: Culex quinquefasciatus
- Most widespread species in the world
- Introduced in 1830's but only recently established in the South Island
- Mammal-biter
- High vector competence

2. Background

Cx. quinquefasciatus (Cx.quinx) is known to transmit harmful human diseases such as Ross River Virus, West Nile Virus and Filariasis. It is also a known vector of avian malaria and multiple parasitic worms which could threaten our bird and lizard species. Such diseases have yet to become established in New Zealand however it is clear that if an outbreak were to occur, establishment is highly likely.

With increased international travel, trade and climate changes, New Zealand’s border biosecurity is more important now than ever. Our current mosquito surveillance doesn’t target established species like Cx.quinx which is dangerous as we cannot quantify how many Cx.quinx interceptions had originated in NZ or from overseas.

Our aim is to assess the population changes of both species over the past 13 years looking particularly at larval morphological differences between species and genetic mixing of samples from different regions. Our big questions include:

1. Is there a statistical relationship between exotic and native Culex population numbers over the last 13 years?
2. Is there preliminary evidence of interbreeding or hybridisation between the two Culex species?
3. Is there genetic evidence of unnoticed, international introductions of Cx.quinx over the past 10 years?

3. Methods

Data was pooled from 12 district health boards around New Zealand spanning the last 13 years. ANOVA and multiple regression analysis will be performed using SAS statistical software and Excel.

Genetic analysis has been done with 5 microsatellite primers developed in previous studies. PCR protocols were trialled and altered before beginning to successfully test 3 samples of each of the 2 species over 3 time periods (2007-08, 2013-14 & 2017-18). Gel electrophoresis helped to indicate suitable samples to be sent to Massey University Genome Services and Geneious software was used for visualising results. Genalex software will be used for microsatellite interpretation.

For the larval morphometric analysis, a minimum of 7 individuals all in the last stage of development (instar 4) of each of the 2 species from a minimum of 3 localities were measured. Photos were obtained using a compound microscope with a Leica ICC50W camera attachment and LASEZ computer program.

4. Preliminary Results

The microsatellite results have indicated a widening genetic pool across the last 10 years however this will not be conclusive until a larger data set has been analysed.

For most primers, Cx.quinx has a larger pool of allele frequencies particularly than Cx.pervigilans. There was also significant overlap of alleles between species which indicates relatability.

<table>
<thead>
<tr>
<th></th>
<th>CeqQ115</th>
<th>CeqQ118</th>
<th>CeqQT10</th>
<th>CeqQ112</th>
<th>CeqQT14</th>
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<tbody>
<tr>
<td>Total pop.</td>
<td>21</td>
<td>9</td>
<td>28</td>
<td>13</td>
<td>3</td>
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<tr>
<td>Cx.pervigilans</td>
<td>10</td>
<td>7</td>
<td>15</td>
<td>6</td>
<td>1</td>
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<tr>
<td>% Alleles present</td>
<td>47.6</td>
<td>77.8</td>
<td>53.6</td>
<td>46.2</td>
<td>33.3</td>
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<tr>
<td>Cx.quinx</td>
<td>16</td>
<td>5</td>
<td>19</td>
<td>7</td>
<td>2</td>
</tr>
<tr>
<td>% Alleles present</td>
<td>76.2</td>
<td>55.6</td>
<td>67.9</td>
<td>53.8</td>
<td>66.7</td>
</tr>
</tbody>
</table>

Figure 1: The above table shows the results of the 79 analysed microsatellites (we predict 216 will be saved for accurate final analysis). Percentage of alleles refers to the number of alleles found in each species relative to the total number of alleles observed per primer.

5. Discussion & Further...

Preliminary results suggest that there is a diverse group of alleles present particularly in Cx. quinquefasciatus populations however more samples are required to draw any further conclusions regarding locational differences.

Significant overlap of alleles and mixing of physical characteristics is suggestive of interbreeding and possible hybridisation. The continuation of the project will be able to isolate key movements and introductions hopefully showing the flow and transfer of alleles throughout New Zealand.

The Hardy Weinberg equilibrium will be used to determine the extent of mixing between populations and will also show relative amount of international introductions which have gone unnoticed. We hope that further results from the project will confirm doubts regarding New Zealand’s biosecurity efforts, promoting reform and improvement for the continued well-being of Aotearoa.

6. References:

- Amy Gault 300375637
- Supervised by Stephen Hartley and Julia Kasper
- The Vulnerability of Aotearoa to Introduced Mosquito Species
- Cx.quinx
- Cx.pervigilans
- Microsatellites: simple sequences with complex evolution.
- Nature Reviews Genetics, 5:435-445
- Parasites and Vectors, 7:468