Inducing female lethality and male sterility in Queensland Fruit Fly
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RNAi-mediated Sterile Insect Technology
- A non-radiation method of producing sterile male only flies
- A simpler method of sex-sorting than mechanical methods
- A non-GM approach to the new SIT methods
- Adaptable to many species
- Can be used with field-caught strains – minimize assortative mating issues

Inducing QFF male sterility
(larvae fed purified dsRNA)

<table>
<thead>
<tr>
<th>dsRNA in diet (tissue gene)</th>
<th>males emerged</th>
<th>% transcript knockdown</th>
<th>% sterile males</th>
<th>% viable progeny for non-sterile males</th>
<th>% impact on fertility on non-sterile males</th>
</tr>
</thead>
<tbody>
<tr>
<td>gus (control)</td>
<td>21</td>
<td>-</td>
<td>2</td>
<td>9.5</td>
<td>16 ± 6</td>
</tr>
<tr>
<td>dsxF</td>
<td>19</td>
<td>46 ± 11</td>
<td>6</td>
<td>32</td>
<td>14 ± 5</td>
</tr>
<tr>
<td>gaul</td>
<td>22</td>
<td>42 ± 16</td>
<td>7</td>
<td>32</td>
<td>13 ± 5</td>
</tr>
<tr>
<td>dynain light chain</td>
<td>17</td>
<td>39 ± 13</td>
<td>6</td>
<td>35</td>
<td>13 ± 6</td>
</tr>
</tbody>
</table>

Preventing QFF female development
(larvae fed purified dsRNA)

Gene expression profiling

RNAi target identification using QFF transcriptome analyses
- Males and females separately considered for all life stages (genetic sorting test developed)
- 48 genetic libraries covering 7 major life stages (tissues include ovaries and testes)
- Strand-specific library construction

RNAi production options
- Express and purify dsRNA from bacteria
- Clone target sequence in E. coli
- Induce expression with IPTG
- ~200 mg dsRNA/L culture (enough for 300 QFF)
- Killed bacteria/yeast expressing dsRNA
- Can add directly to the diet without purification
- Synthetic dsRNA

Project AI13001 Highlights
- Establishment of RNA protocols for creating and analysing QFF transcriptome data
- A genetic test to determine the sex of any QFF life stage in under 2 hrs
- Creation of the first developmental transcriptome map for QFF, covering all life stages of both sexes from egg to mated adult
- Demonstration that male sterility can be induced in QFF by including RNAi in the diet
- Demonstration that female development in QFF can be prevented by RNAi in the diet

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