Samples for bee disease diagnosis

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Bee diseases and their characteristic signs can be very confusing to the average beekeeper. Wrong diagnosis can be a major problem, particularly when serious brood diseases are involved. Beekeepers can obtain accurate diagnoses by sending samples of suspect material to the NSW DPI Veterinary Laboratory.

What to send

Three types of samples can be submitted — larvae (smears and ‘mummies’), adult bees or comb sample containing diseased brood, according to the disease suspected:

<table>
<thead>
<tr>
<th>Suspected disease</th>
<th>Sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>American foulbrood (AFB)</td>
<td>Larval smear (preferred) or comb sample containing diseased brood</td>
</tr>
<tr>
<td>European foulbrood (EFB)</td>
<td>Larval smear (preferred) or comb sample containing diseased brood</td>
</tr>
<tr>
<td>Chalkbrood</td>
<td>‘Mummies’ in or from comb</td>
</tr>
<tr>
<td>Sacbrood</td>
<td>Comb sample containing diseased brood</td>
</tr>
<tr>
<td>Nosema</td>
<td>Adult bees</td>
</tr>
<tr>
<td>Unknown disease</td>
<td>Contact an apiary officer immediately</td>
</tr>
</tbody>
</table>

Smears

Smears are preferred for the diagnosis of the bacterial diseases AFB and EFB. Smears are prepared from larvae, which are showing signs of disease. The technique is simple and the smears are ready for laboratory examination.

To prepare smears, follow these steps:

- Start with a clean glass microscope slide and some matches. Clearly label the slide with a pencil, felt pen or permanent marker. Some microscope slides have a frosted end which will accept pencil. Number each sample and mark the hive it came from. Select three or four suspect larvae and place them on the slide. A match can be used to remove the larvae from the cells (Figure 1).

Figure 1. Larvae on a slide

- With a match, pulp the larvae together. Spread the pulped larvae over the slide (Figure 2).

Figure 2. Spreading pulped larvae

- Then, using the side of the match, push the excess material off one end, leaving a thin smear of milky liquid on the slide (Figure 3).

Figure 3. Wiping off the excess

Use a fresh match for each sample and leave the match in the hive. This will minimise disease spread.
Allow the smear to dry in the open air, but not in direct sunlight (Figure 4). Do not cover with a cover slip or another slide. If slide carriers are not available, wrap the slides individually in paper and protect with stiff cardboard or plywood before wrapping.

Figure 4. Allow the smear to dry

In disease outbreaks in commercial apiaries, a number of smears should be taken – three or four smears should be sufficient. It is also possible to have more than one disease in a hive, thus the need to take multiple samples. This is suitable for AFB and EFB diagnosis.

**Comb samples**

Cut out a piece of brood comb about 5 x 10 cm, containing the suspected diseased brood. Place the sample in a strong cardboard box, and wrap in paper. Avoid honey in the sample, and don’t put the sample in a plastic or airtight container.

An old knife, heated in the smoker, will facilitate the removal of the brood comb sample. This is suitable for AFB, EFB, chalkbrood and sacbrood diagnosis (see figure 5).

**Adult bees**

For nosema diagnosis collect a sample of 30 adult bees at random from the entrances of the hives in the apiary. Place the bees in a queen cage or other suitable container with a small supply of queen candy. The bees must be alive when they reach their destination.

Bee samples for nosema can also be collected and stored in alcohol or metho. Samples stored in this manner cannot be sent by post. They need to be delivered by courier or in person to the laboratory.

For other adult disease diagnosis collect 30 bees showing signs of the disease, or recently dead bees. Place in a ventilated container, such as a plastic jar with holes in the lid, and dispatch to the laboratory by the fastest available means.

**Sending samples**

Contact the laboratory to confirm packing arrangements and to obtain current laboratory fees before submitting samples.

In a covering letter include information on the nature of the sample, and what the problem is, as well as the following information.

- The age of bees affected (sealed or unsealed brood, young or old workers).
- Anything unusual about the shape, position or colour of the dead bee larvae (for example a ‘C’ shape or stretched out).
- Anything unusual about the adult bees.
- Did dead larvae rope or string out when touched with a match?
- Any unusual odour, either from the dead brood or the match used to rope the larvae.
- Date you took the samples.
- Number of hives infected.
- Your address and contact details.

Send samples to the laboratory:

**NSW DPI Laboratory Services**

Courier: Woodbridge Road, Menangle NSW 2568
Postal: Private Bag 4008, Narellan NSW 2567
Ph: 1800 675 623
E: laboratory.services@dpi.nsw.gov.au

Figure 5. Sample of diseased comb cut and ready for packing in a cardboard box.

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