

# Background Reading Module 3: Honey Bee diseases, pests and poisoning

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# HONEY BEE DISEASES, PESTS AND POISONING

## Introduction

These notes are intended to cover the syllabus for Module 3, the subsections do not follow those within the syllabus and include extra topics.

The notes are personal to the author and contain inaccuracies, typos and grammatical errors for which I apologise.

I have tried to credit all sources and in particular artwork, if I have missed any credits please let me know and I will sort.

References used include:

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Around and About, Celia Davis

Dave Cushman website: <http://www.dave-cushman.net/bee/disease.html>

Randy Oliver: <http://scientificbeekeeping.com>

BBKA Healthy Hive Guide

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Healthy Bees are Happy Bees, Pam Gregory

Infectious Diseases of the Honey Bee, L. Bailey

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## 1. the statutory requirements relating to pests and diseases of honey bees in the United Kingdom, and their implementation.

Taken from the Bee Diseases and Pests Control Order Explanatory Notes:

The Bees Act 1980 makes provision for the control of diseases and pests affecting bees. Current legislation made under the Act - the Bee Diseases Control Order 1982 and the Importation of Bees Order 1997 – needs modifying to take account of recent developments in domestic disease control and in the EU law that regulates the importation of bees from third countries.

The Bee Diseases and Pests Control (England) Order 2006 requires beekeepers (and others) to notify the Secretary of State of the suspicion of the presence of the notifiable diseases, American and European foul brood, which are already present in England, and of two new bee pests, the small hive beetle and tropilaelaps mites. Both pests are exotic to the EU but are considered serious threats to the economic sustainability of the apiculture sector. The European Commission has strengthened EU bee health biosecurity by harmonising import controls across the Community so as to reduce the risk of the introduction of the exotic pests through imports. The Order no longer provides a notification requirement for Varroasis, since varroa, the parasitic mite that spreads the disease, is now endemic.

In response to a notification of a suspected notifiable disease or pest, restrictions will be imposed on the movement of anything that might spread the disease or pest until an authorised bee inspector has visited the affected premises to confirm the identification and a decision has been made on action to eradicate or control the outbreak. The Secretary of State may also declare an infected area and implement control measures within it, if the small hive beetle or tropilaelaps has been found present in that area.

The Order also implements the part of Commission Decision 2003/881/EC (as amended by Commission Decision 2005/60/EC) that applies to consignments of queen bees imported from third countries once they have reached their designated destination. In particular, importers are required to send attendant worker bees, queen cages and any other material that accompanied the queen bees from their third country of origin to a laboratory for examination for the presence of the small hive beetle and tropilaelaps mites. Alternative less onerous conditions apply to imports of single bumble bee colonies bred under environmentally controlled conditions in the third country of origin. A Transposition Note is attached to this Memorandum. Commission Decision 2003/881/EC was not subject to scrutiny by the EU Scrutiny Committees.

As a consequence of the harmonised rules established by Commission Decision 2003/881/EC, beekeepers/ importers are no longer required to obtain a licence from Defra to import bees from third countries.

The Order will be enforced by bee inspectors from the Central Science Laboratory's.

[https://www.legislation.gov.uk/uksi/2006/342/pdfs/ukxi\\_20060342\\_en.pdf](https://www.legislation.gov.uk/uksi/2006/342/pdfs/ukxi_20060342_en.pdf)

<http://www.nationalbeeunit.com/downloadDocument.cfm?id=1281>

## 2. the virus diseases of the honey bee, and methods for their laboratory and field diagnosis.

A virus is a small piece of genetic material, usually ribonucleic acid (RNA) enclosed in a protein shell. They are organised in arrays of RNA and proteins and can be detected by an electron microscope.

To identify a virus enzyme-linked immunosorbent assay (ELISA) technique needs to be applied.

Enzyme-linked immunosorbent assay (ELISA), also known as an enzyme immunoassay (EIA), is a biochemical technique used mainly in immunology to detect the presence of an antibody or an antigen in a sample. The ELISA has been used as a diagnostic tool in medicine and plant pathology, as well as a quality-control check in various industries. In simple terms, in ELISA, an unknown amount of antigen is affixed to a surface, and then a specific antibody is applied over the surface so that it can bind to the antigen. This antibody is linked to an enzyme, and in the final step a substance that the enzyme can convert to some detectable signal is added, most commonly a colour change in a chemical substrate.

### Transmission routes for honey bee viruses

Vertical, (parent offspring):

- male and female eggs, queen ovaries
- drone sperm

Horizontal:

- Feeding through ingested into gut:
  - o virus in honey, pollen, cleaning infected cells
- Varroa mite injection into haemolymph: (parasitic feeding activity)

Virus cycle

- Can't live independently (survive only as part of host's cells)
- Can't reproduce independently (only inside cells of living things)
- Use's cellular mechanisms of host to reproduce

Budded agglomerations of genetic particle's:

- Composed of different proteins
  - o thus, help to genetically adapt to changing conditions.
- Continue without obvious damage to host, as long as cell is alive.
- When cell dies, releases millions of virus particles to infect other living cells.
- Bees present to be healthy and no visible effect of virus.

Factor's added to trigger problem of pathology: poor weather conditions, varroa or poor nutrition

Diagnose: use ELISA in form of lateral flow device.

Practical control methods:

- Reduce varroa mite
- Some queens (genetic) more resistant to viruses
- Nutrition
- Hygienic behaviour (honey bees & beekeeper)

Field diagnosis of the symptoms of various viruses are detailed below:

**Chronic Bee Paralysis**

**Associated: Probably Acarine**

<b>Type 1</b>	<b>Type 2</b>
Trembling wings and body	Trembling, not flying
Not flying – crawling on ground and up plant stems	Refused entry to hive
Huddle on top bars – do not react to smoke	Broad abdomen
Bloated abdomen (full honey sac)	Nibbled by other bees – black, shiny, hairless (appear smaller)
Dysentery	Deaths
Dislocated wings (K-wing)	
Deaths	

**Acute Bee Paralysis Virus**

**Associated: Varroa**

- Weakening of the colony without signs of brood diseases and mites
- Increasing numbers of dead or dying bees on the inner cover or front of the hive. Dying bees may be trembling and display uncoordinated movement.
- Affected Bees are partly or completely hairless where the upper surface of the Thorax is especially dark
- Older Adult Bees have a greasy or oily appearance while recently emerged Bees may appear opaque as if pigmentation of the tissue had not been completed prior to emergence
- Rapid decline within a few days

**Black Queen Cell Virus**

**Associated: Nosema**

- Turns queen cell black
- Prepupa or pupa is yellow
- Tough skin slightly resembles sacbrood

**Sac Brood**

**Associated: Varroa**

- The moult at prepupa to pupa goes wrong and the space fills with ectodysial (fluid)
- Mould skin resembles Chinese Slipper
- Changes from yellow to dark brown

- Pupa dies. Can give a short rope – can be confused with AFB
- Adult Bees can be infected when cleaning cell
- Life shortened
- Become foragers earlier
- Stop feeding larvae
- Rarely collect pollen
- Behaviour of adult bees can cause the disease to die out in a colony

### Deformed Wing Virus

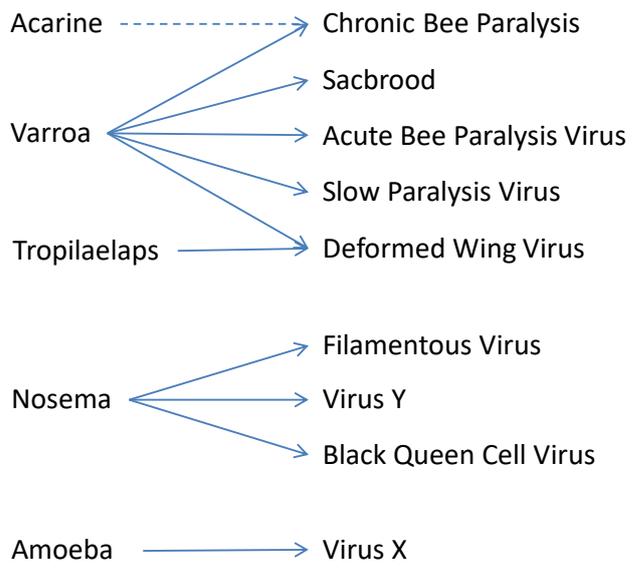
**Associated: Varroa and Tropilaelaps**

- Damaged appendages, particularly stubby, useless wings
- Shortened, rounded abdomens
- Micolouring
- Paralysis
- Severely reduced life-span (less than 48 hours)
- Typically expelled from the hive

### Other Viruses

Virus	Association	Symptoms
Slow paralysis virus	Varroa	Collapse late in the year
Filamentous virus	Nosema	Haemolymph goes milky
Virus Y	Nosema	No reported symptoms
Virus X	Amoeba	Shortens life; Colonies die in spring
Cloudy Wing Virus		Wings go cloudy. Bee dies

## Summary of Associations



### 3. Foulbroods

<http://www.nationalbeeunit.com/downloadDocument.cfm?id=7>

#### 3.1 *the symptoms of American Foul Brood (AFB) and European Foul Brood (EFB).*

<b>American Foul Brood</b>	<b>European Foul Brood</b>
AFB generally affects only sealed brood. (A good way of remembering is that AFB = A for After sealing of the cell.) When infected larvae die in the sealed cell, the appearance of the cell cappings changes.	EFB affects mainly unsealed brood, killing larvae before they are sealed in their cells. An easy way to remember is that EFB = E for Early infection before sealing of the cell.
At first only very few cells may show signs of disease, and the colony will appear normal in other respects.	A larva infected with EFB moves inside its cell instead of remaining in the normal coiled position characteristic of a healthy larva of the same age.
Wax cappings become sunken and perforated when adult bees nibble holes in them to try to remove the infected larva within. These perforations tend to be jagged and irregular in shape.	A larva dies in an unnatural attitude – twisted spirally around the walls, stretched out lengthways from the mouth to the base of the cell or across the mouth.
Eventually much of the sealed brood will become affected by the disease, causing a patchy or ‘pepper pot’ brood pattern.	When EFB kills a high proportion of the larvae, the brood pattern often appears patchy and erratic as the bees remove dead brood and the queen lays in the vacant cells.
Some cappings may become moist or greasy looking and slightly darker in colour than other cells.	The gut of an infected larva may be visible through its translucent body wall. The mass of bacteria living within it give it a creamy white colour.
At the sunken capping stage, the dead larval remains are light to dark brown in colour, and have a slimy consistency.	A very unpleasant odour may sometimes accompany severe EFB infection, depending on the presence of certain other species of bacteria in the remains of dead larvae.
There may then be an unpleasant smell associated with decomposition.	As it collapses, a dead larva often looks as though it has melted, turning yellowish-brown and eventually drying up to form a loosely attached brown scale.
Further drying leads to the final stage, which is a very dark brown, rather rough scale lying on the lower side of the cell and extending from just behind the mouth of the cell right back to the base.	

<b>American Foul Brood</b>	<b>European Foul Brood</b>
To detect scales, hold the comb facing the light: their rough surfaces reflect the light, making them easy to see even when they are almost the same colour as the comb itself.	
Conduct the 'ropiness' test: insert a matchstick and slowly withdraw it; a brown, mucus-like thread or 'rope' 10-30mm long a reliable indicator for AFB.	
The ropy condition is followed by a tacky stage as the larval remains in the cell gradually dry up and the colour changes to dark brown.	
The proboscis of dead pupae may sometimes remain intact, protruding upwards from the bottom edge of the cell	

## Sketches of Typical Foul Brood Bacteria

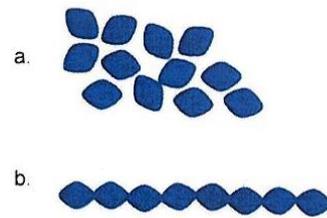
### *Brevibacillus laterosporus*

This bacterium is sometimes isolated from EFB-infected larvae, although it is not very common. Spores of *B. laterosporus* are highly distinctive under microscopy, almost looking like telephone handles, with a clear area in the centre (a). Vegetative cells are typical rods and it is really the spores that are of use for characterisation. Fortunately, these are formed readily in infected larvae and also in culture, once it is a few days old.



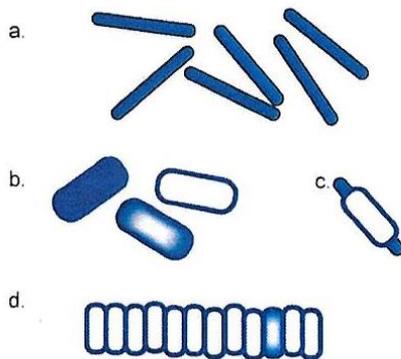
### *Melissococcus plutonius*

The bacterium is cited as being the cause of European foulbrood and its presence is used as a diagnostic confirmation of the disease. When taken directly from infected larvae and examined microscopically, it is recognisable as lanceolate cocci, often in groups (a) or chains (b). However, when grown in culture it sometimes loses these characteristics and care should be taken not to confuse it with other cocci. This is usually quite simple as it will not grow aerobically, and the other bacterium that it is sometimes mistaken for, *Enterococcus faecalis*, grows profusely under aerobic conditions.



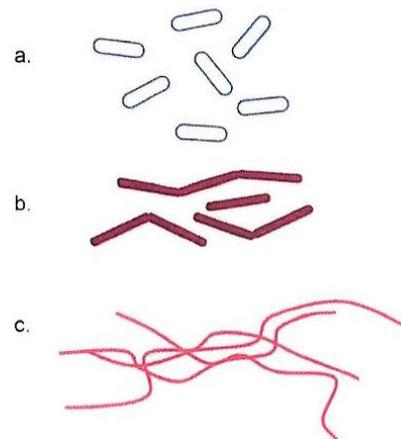
### *Paenibacillus alvei*

*P. alvei* is often found associated with EFB, and some researchers think it has an active part to play in the disease as a pathogenic organism. It is very easy to grow, and is straightforward to identify both macro- and microscopically. Upon microscopic examination, at early growth stages regular rods (a) are visible, but these very quickly develop into spores (b); sporulating cells (c) are sometimes evident. Spores generally do not retain stains, so usually just the outline is visible. An interesting characteristic is that the spores often line up side by side, to give the arrangement shown in (d).



### *Paenibacillus larvae* subsp. *larvae*

This is the causative agent of American foulbrood, and is only usually seen in monoculture. In larval samples, the spores are usually visible, and are narrow rod shapes, often described as like fly eggs (a). In culture, spores do not form as readily, and vegetative cells are usually seen. These can be two types, either discrete rods (b) or, usually seen in older cultures, long tangles hair-like threads (c), both of which may appear Gram negative.

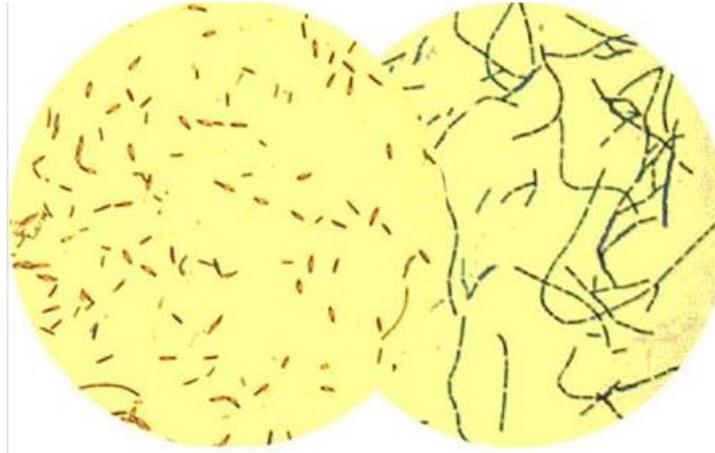


### 3.2 *the life cycle of Paenibacillus larvae and Melissococcus pluton, the causative organisms of AFB and EFB, and their development within the larva.*

#### **AFB**

American foulbrood (AFB) is considered to be the most fatal of honeybee brood diseases. The disease attacks only the very young larvae; larvae older than 48 hours and adult bees are not susceptible to it.

AFB is caused by the spore-forming bacterium known as Paenibacillus larvae. The bacterium exists in two forms: the spore stage and the vegetative stage, which consists of slender rod-shaped bacterial cells. Only the spore stage is contagious to bees.



LEFT: Paenibacillus larvae in the spore stage, without appendages.

RIGHT: Paenibacillus larvae in the vegetative stage.

Image credit: Baylor College of Medicine [www.hgsc.bcm.tmc.edu](http://www.hgsc.bcm.tmc.edu)



#### **Pathogenesis**

Bee larvae become infected when they ingest Paenibacillus Larvae spores in brood food given to them by nurse bees. A day after ingestion the spores germinate in the larval mid-gut into the vegetative form (rod stage), becoming bacteria. The rods penetrate the gut wall, entering the tissues where they proliferate rapidly and at an enormous rate, feeding at the expense of the tissues, until the larva dies. The larva dies after its cell has been sealed; sealing the cell stops the supply of nourishment to the bacteria; they cease to grow and proliferate and revert to the spore stage.

After death, the normally white larvae turn dark brown and decay into a glue-like mass, which will form a rope. The decaying mass has a foul smell - hence the name, foulbrood. At the final stage, within a month or so, a dead larva or pupa dries to a dark brown scale that adheres tightly to the lower side of the cell too tightly for the bees to remove. Each scale contains millions of infective spores. Once they are inside the larval gut again, the cycle repeats itself.

#### **EFB**

The bacterium responsible for causing the symptoms of European Foulbrood (EFB) is probably Melissococcus plutonius. When it infects a larva, other bacteria move in, causing secondary infections:

- Bacillus alveri and laterosporus
- Bacterium eurydice
- Streptococcus faecalis

The bacteria enter a larva in brood food and multiply in the ventriculus (stomach), feeding on the larval food. The bacteria lodge between the peritrophic membrane and the food in the ventriculus. The bacteria act essentially as a parasite competing for food, and the larva dies of starvation about 3 or 4 days before the cell is due to be sealed. During this period, the larva contorts itself into unusual positions, twisted spirally or flattened out lengthways in the cell. Its colour changes from pearly white to cream and then to a yellowy green. The bacterial mass in the larval stomach causes much of this early colour change. The supply of food to larvae affects the course of the disease. Because the bacteria compete with the larvae for food, increasing the supply of food can enable larvae to survive infection.

At the onset of nectar flow in early spring, the number of house bees recruited to forage may increase rapidly leaving fewer in the hive to feed larvae. Under these conditions, M. plutonius may be able to starve larvae to death and give rise to symptoms of the disease. When the ratio of nurse bees to larvae stabilises and larvae receive enough food to survive to pupation, symptoms disappear.

However, EFB can occur throughout a season and will sometimes not abate of its own accord. In severe cases, it can cause a colony to die. Also, contaminated combs and equipment can cause EFB to recur. The bacterium that causes EFB does not produce spores, but combs contaminated with it can still re-infect bees in subsequent years.

### *3.3 the development of AFB and EFB within the colony.*

#### **AFB**

Infection of the larva is by ingestion of the spores in contaminated brood food. The bactericidal effect of 10-hydroxydecenoic acid (10-HDA) from the worker bee's mandibular glands prevents germination of the spores in the adult bee.

The conditions in the larval gut are ideal for germination and the bacterial population doubles about every 8 hours. Sporulation begins when the larva voids the contents of its gut before metamorphosis, and the cell contents become a source of further infection. Bacteria continue to multiply in the haemolymph and eventually cause the larva to die. Once the larva dies the bacteria again sporulate within its body. Adult bees become infected as they clean away the dead remains in the hive.

AFB bacteria gradually destroy larval tissue.

House-cleaning bees come along and try cleaning up both the messy (pre)pupae and the scales, so becoming contaminated with the spores. The spores can get into every part of the hive including the honey. House-cleaning bees soon become nurse bees, feeding young larvae, and the spores will be passed to the larvae in this way. The disease may be quite slow to get going in the beginning; the bees can keep the spread under control for a time by the removal of diseased larvae in early stages. As the number of young bees declines the disease takes control and quickly destroys the colony.

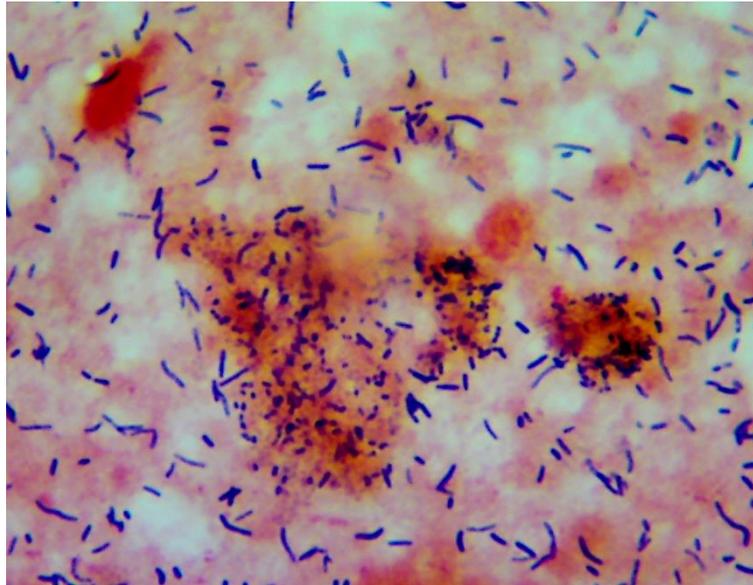
#### **EFB**

There are three important facts involved in the spread of Melissococcus plutonius in a colony:

- M. plutonius never forms spores. The normal vegetative cells are infective and reproduce in huge numbers in the infected larva.

- The contents of the ventriculus of a larva, and so the bacteria, are “sealed in” until the larva pupates and the connection between the ventriculus and hindgut opens, when all the waste and bacteria that have been stored in the larva’s gut pass out into the cell
- Young adults clean the cells out and later produce food that they feed to larvae.

Taken together these phenomena explain how the disease spreads through the colony. Infected larvae that survive to pupation discharge the contents of their guts into the cell. House bees pick the bacteria up when they clean the cell and subsequently feed them to the young larvae in brood food. When a larva spins an inadequate cocoon, the bacteria are more accessible to the house bees.



### *3.4 the development of EFB within the apiary.*

Robbing weakened colonies

Beekeeper moving equipment between hives

### *3.5 the ways in which AFB and EFB are spread between colonies.*

Natural methods of spread:

- drifting, where a worker bee may go into the wrong hive, taking spores with it
- swarm from an infected hive
- robbing; probably the most important bee-based method of spread. Bees from other colonies loot the stores of colonies weakened or killed by foulbrood and carry spores back to their own colonies.

Beekeeper methods of spread:

- moving infected combs from one colony to a healthy colony
- uniting a weak (diseased) colony with a stronger colony
- feeding honey from a dubious source to bees
- trapping pollen from infected colony and feeding to healthy colony
- inspecting hives on remote site with dirty gloves and suit after inspecting own infected colony

- hiving unknown swarms near healthy colonies
- buying old equipment without cleansing before use
- moving bees to area with large numbers of colonies close by, *e.g.* pollinating sites
- purchasing infected stock of bees

### 3.6 *the methods of laboratory and field diagnosis of AFB and EFB.*

EFB cannot be reliably identified visually, as the signs of disease can easily be confused with other brood abnormalities. FERA Bee Inspectors confirm suspect infections in the field by using Lateral Flow Devices (LFDs).

There is also a LFD for AFB.

Occasionally sample brood combs (or suspect larvae in plastic tubes) are sent to the NBU laboratory where larval gut contents and scale in cells are examined for the presence of the causative bacteria using microscopy tools.

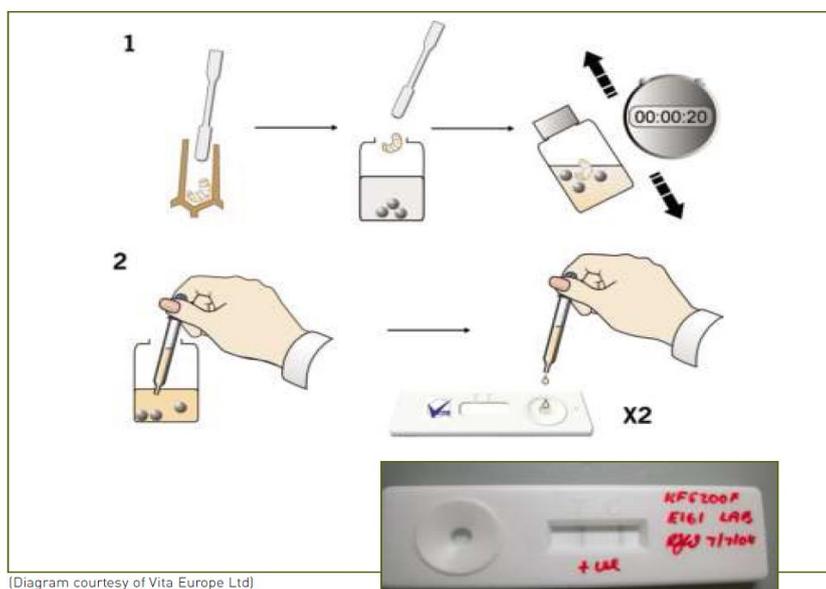
To test for foulbrood using an LFD, put a sample of suspect infected larval material into the buffer bottle and shake it for about 20 seconds. Then put 2-3 drops of the resulting suspension onto the LFD. The blue lines at the C (Control) and T (Test) lines indicate a positive result.

#### Advantages of LFD:

- Available for both AFB and EFB
- Can be used in the field
- Established, accepted mature technology
- Stable – shelf-lives of 12–24 months often without refrigeration
- Ease of use: minimal operator-dependent steps and interpretation
- Accuracy: LFDs detected *Melissococcus plutonius* in 96–100% (n = 137) of EFB-infected samples in laboratory trials. Field validation was equally robust: LFD-testing on site gave correct diagnoses for 96% (n = 184) of samples; false positives were rare (~1%).”

#### Disadvantages of LFD:

- Indicates only the presence of the disease, not its level
- Results must be recorded manually
- Based on a specific antibody; test might become ineffective if new strains emerge.



### *3.7 the treatment of colonies with AFB and EFB, methods of destruction of diseased colonies and the sterilization of equipment.*

#### **AFB**

AFB is a notifiable disease under the Bee Diseases and Pests Control Order (for England and Wales) and is subject to official control by a programme of apiary inspections carried out by the NBU. Control of the disease is through compulsory destruction of infected colonies, which is a highly effective measure.

Methods of control of AFB using antibiotics that are used in some countries are not effective, as they serve only to suppress signs of the disease without eradicating it and, through frequent use, allow resistant bacterial strains to develop. The use of antibiotics to control AFB is not permitted in the UK.

A diseased colony is eradicated by burning the bees and combs in an open pit.

Sterilise hive boxes by scorching with burner and clean clothes, gloves, tools, etc. thoroughly in hot water and soda crystals.

#### **EFB**

EFB is a notifiable disease under the Bee Diseases and Pests Control Order (for England and Wales) and is subject to official control by the examination of colonies for signs of disease and compulsory treatment or destruction of diseased colonies.

There are three options available to the beekeeper in the UK who has colonies infected with EFB:

1. The colonies may be treated with the shook swarm husbandry method. Trials conducted by the National Bee Unit showed that Shook swarm is more successful than OTC for the control of EFB in England and Wales: "In the Spring following treatment, shaken colonies were three times less likely to test positive for M. plutonius. This finding appears logical since OTC treatment does not remove the etiological agent present in the hive. In contrast, the Shook swarm method provides the bees with material free of M. plutonius. In addition, colonies treated with OTC were five times more likely to show recurrence of EFB the following year than colonies treated by Shook swarm."
2. The colonies may be treated with the antibiotic oxytetracycline (OTC, as the formulation Terramycin®); The Bee Inspector administers Terramycin, mixing it with sugar syrup in a jar with holes in the lid, then shaking the jar over the bees on each frame. It is not put in a feeder on the hive.
3. The colonies may be destroyed, as for AFB. This will be carried out if the colony is too small for other treatment methods, is too heavily infected to respond to treatment, or at the beekeepers request.

However, the range of options available will also depend upon the time of year that the disease is diagnosed and other factors such as the strength of the colony or the level of infection.

Weak colonies and colonies with a high proportion of diseased brood are destroyed, as with AFB, but lightly diseased colonies may be treated with antibiotics. Under the Order only an Appointed Officer may carry the treatment out, using drugs officially dispensed following confirmation of EFB in a disease sample submitted for diagnosis at an approved laboratory or by LFD. The designated Veterinary Laboratories Agency (VLA) prescribes the treatment.

#### 4. Chalk Brood, its symptoms, cause, and treatment.

Cause	A fungus, <u>Ascosphaera apis</u>
	The spores are ingested by bees in the brood food and germinate in the gut Fungal hyphae penetrate through the gut wall and eventually grow out through the cuticle The young bee dies once the cell has been capped
Symp-toms	The larva appears as a white “plug” filling the cell. Sometimes a yellow shrunken “head” is visible on the top. At first it is soft and fluffy but hardens to a solid lump called a “chalk brood mummy”. The bees try to remove the mummies from the cells and they can be often seen on the hive floor or under hive if mesh floor Chalk brood mummies differ from stone brood mummies in that they are softer and crumble easily when handled Brood takes on a “pepperpot” appearance in heavy infestations
Occurrence	Very common; many hives have a few cells affected Chilling the brood makes the disease worse It has also been said that damp conditions favour the development of chalk brood
Treatment	Mild infection does not harm the colony and no treatment is necessary Avoid chilling the brood if inspecting on a cold day Frames containing a lot of chalk brood should be destroyed and replaced with new foundation If the infestation is severe, re-queening is sometimes recommended but not all authorities agree that this is effective Essential Oils including cinnamon oil and thymol have been found to inhibit the growth of chalk brood fungus

#### 5 Stone Brood, its symptoms, cause, treatment and effect on humans.

Like Chalk brood except:

- mummies yellow, turning green when spores form
- mummies hard (chalbrood mummies crumble between fingers)
- can attack larvae

Caused by up to 6 different Aspergillus fungi, mainly either Aspergillus flavus or Aspergillus fumigatus.

Treatment, no chemical treatment, well ventilated hive, if persist requeen with hygienic strain.

Can cause breathing issues with humans with compromised immune systems.

## 6 those fungi which attack pollen and comb.

One-hundred and forty-eight molds were isolated from the following samples of almond, *Prunus dulcis*, pollen; floral pollen collected by hand; corbicular pollen from pollen traps placed on colonies of honey bees, *Apis mellifera*, in the almond orchard; and bee bread stored in comb cells for one, three, and six weeks.

The majority of molds identified were *Penicillia* (32%), *Mucorales* (21%), and *Aspergilli* (17%). In general, the number of isolates decreased in pollen as it was collected and stored by the bees. Each type of pollen sample appeared to differ in regard to mold flora and dominant species.

*Aureobasidium pullulans*, *Penicillium corylophilum*, *Penicillium crustosum*, and *Rhizopus nigricans* were among the molds that may have been introduced by bees during collection and storage of pollen.

*Mucor* sp., the dominant mold in floral pollen, was not found in corbicular pollen and bee bread.

Tests for 19 enzymes revealed that most of the molds produced caprylate esterase-lipase, leucine aminopeptidase, acid phosphatase, phosphoamidase, B-glucosidase, and Mactetyl-B-glucosaminidase. Thus, enzymes involved in lipid, protein and carbohydrate metabolism were produced by pollen molds.

Molds could also contribute organic acids, antibiotics and other metabolites.

## 7 the laboratory diagnosis of *Nosema*, an in depth account of the life histories of *Nosema apis* and *Nosema ceranae*, their effect upon individual bees and upon the colony, and their treatment.

*Nosema* is the most common disease and is found in seemingly healthy colonies.

- *Infectious Diseases of the Honey Bee* (Dr. Bailey & Brenda Ball) states that 79 of 80 apparently healthy colonies contained *Nosema* spores.

Two *Nosema* species have been identified in honey bees in England and Wales: *Nosema apis* and, more recently, the Asian species *Nosema ceranae*.

Both are parasitic microsporidian fungal pathogens.

*N. ceranae* is a more "generic" parasite than *N. apis*, and can infect various hosts. It is more closely related to *N. vespula* (from yellowjacket wasps) than it is to *N. apis*.

Different "strains" (haplotypes) of *N. ceranae* exhibit different degrees of virulence.

### Life Cycle

*Nosema spp.* infect the epithelial cells lining the mid-gut of the bee, where they multiply rapidly.

Within a few days the cells are packed with spores, the resting stage of the parasite.

The protozoa multiply in the ventriculus (30-50 million spores) and impair the digestion of pollen thereby shortening the life of the bee.

*N. ceranae* goes on to infect the basal cells, and then spreads throughout the entire alimentary tract, including the hypopharyngeal and salivary glands, but it infects only 20% of fat bodies and no muscle tissue.

When the host cells rupture, they shed spores into the gut where they are later excreted by the bees.

The spores in excreta can germinate and become active once more, when ingested by another bee

### Pathology

N. ceranae is a more virulent parasite than N. apis. It is more adapted to heat than N. apis; it can survive a broader temperature range:

Temp.	Context	Activity of <u>N. apis</u>	Activity of <u>N. ceranae</u>
25°C	a bit cool for a bee	Can multiply	Infects bees more quickly than <u>N. apis</u> does.
33°C	low brood nest temperature	Thrives	
37°C	typical of warmed bee flight muscles, or hot brood nest	Dies	Survives

The pathology of N. apis reflects its response to temperature:

- A few bees go into winter infected; they spread spores to neighbouring bees in the winter cluster (forming 'pockets of infection' within the cluster).
- These pockets get larger toward the end of winter until they are completely eliminated in spring when the infected bees fly out and die.
- Generally, levels of Nosema stay low over summer, until autumn when there is a small peak, and again this is mostly temperature driven.

The seasonality of N. ceranae is different.

- Instead of spiking only in November and March as N. apis does, N. ceranae is present throughout the year, and thrives in summer.
- The warmth of summer (or induced fever in the bees) does not kill N. ceranae off; colonies may struggle or collapse even during a spring or summer bloom.
- The spore is resistant to temperature extremes and dehydration, and cannot be killed by freezing the contaminated comb.

### Effects on Queens

N. apis often causes early supersedure of queens.

- Chilling and stress of shipment or holding at room temperature promotes transmission from attendants to queen.
- Attendant bees lick up her infected faeces

N. ceranae is not readily transmitted to queens

### Symptoms and Effects

There are no obvious signs of Nosema, although **Dysentery** (q.v.), excreta on combs and hive, frequently accompanies heavy infections.

- Bees normally defecate away from the hive – sometimes the bees defecate in and about the hive because of the excessive build-up of waste matter in their guts.

- House bees become infected by cleaning up the excreta containing spores.

Nosema inhibits the ability of infected bees to digest food.

Bees infected by N. ceranae simply starve to death in the midst of plenty as a result of lack of digestive function.

Bees infected with N. ceranae are hungry, and so attempt to feed more, indulging in risky foraging behaviour, and depopulating their colonies

They tend to forage at cooler temperatures, or even simply fly off to die.

Foragers infected with N. Ceranae die prematurely, and so inhibit the build-up of the colony.

Infected colonies fail to build up normally in the spring. Dead bees may be seen outside the hive after cleansing flights.

N. ceranae also appears to suppress the bees' immune functions.

Bees ramp up their immune systems in response to N. apis, but N. ceranae suppresses that system.

In addition, infection by N. ceranae depresses the level of the bee "fountain of youth," vitellogenin, suggesting that infection may decrease their lifespan by this effect.

Nosema stresses the bees nutritionally and immunologically leaving them prone to viruses.

Nosema breaches a bee's main barrier to virus infection—the intact gut epithelium.

### **Diagnosis and Treatment**

Confirm Nosema is by microscopic examination (400x): crush 30 bees in water and examine a droplet for white, rice-shaped bodies.

- Send a sample to a microscopist in a paper container (not plastic).

Crushing bees can release millions of spores; avoid doing it.

Replace and sterilize combs with 80% acetic acid (100 ml./brood box for one week – air before use).

Treatment with the antibiotic Fumidil B (prepared from *Aspergillus fumigatus*, the causative agent of Stone Brood!) is no longer permitted in Europe and the UK. (Fumidil B inhibited the reproduction of spores in the ventriculus, but does not kill them. It also tainted the honey.)

## **8 the laboratory diagnosis of *Malpighamoeba mellifica*, an in depth account of its life history its effect upon individual bees and upon the colony, and its treatment.**

Amoeba is caused by a protozoan amoeba-like parasite Malpighamoeba mellifica.

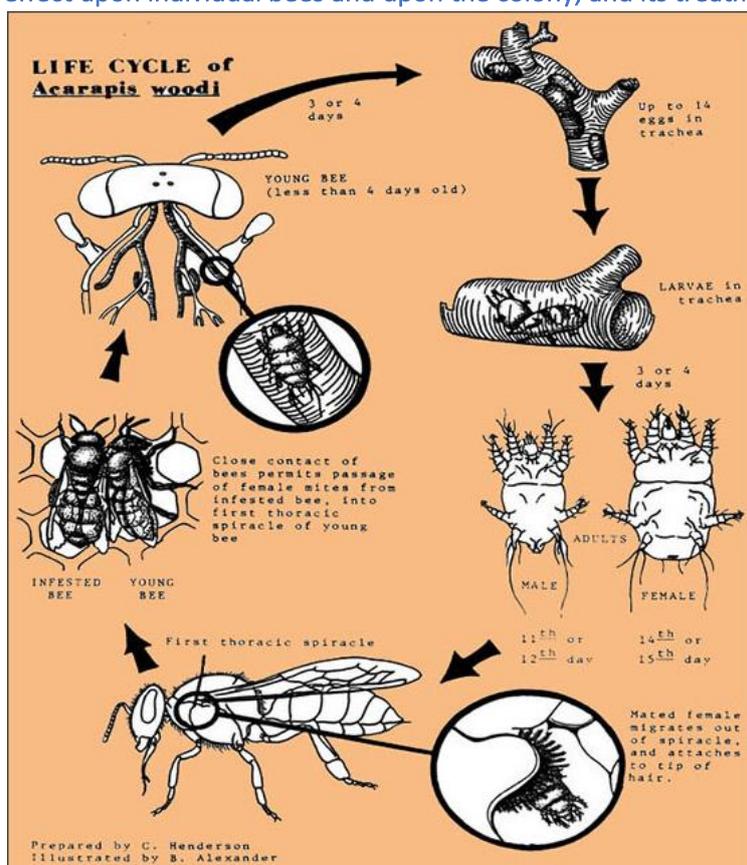
Cysts are ingested with food and germinate in the rectum. They migrate to the Malpighian tubules (the 'kidneys') to create more cysts that then accumulate in the rectum and are excreted.

The infection seems to have no effect on the colony; there are no specific symptoms and no treatment.

Often seen under a microscope when examining a sample for Nosema - grainy circular cysts, larger than the rice-shaped Nosema spores.

Acetic acid destroys the spores.

- 9 the laboratory diagnosis of the tracheal mite *Acarapis woodi*, an in depth account of its life history, its effect upon individual bees and upon the colony, and its treatment.



Acarine is an infestation by the mite Acarapis woodi. The Isle of Wight disease in 1904 – 1920s was probably acarine.

There are no visible external signs – the signs that beekeeping books usually give - crawling bees, dislocated wings, etc. - are those of Chronic Bee Paralysis associated with Acarine (although not proved as a vector).

The mites infest the trachea. Dissection and microscopic examination (20x) of the first thoracic trachea can confirm diagnosis. Send a sample to a microscopist (in a paper container not plastic).

There has been no approved medicament in the UK since FolbexVA was withdrawn in early 1990 and the Frow Mixture was banned.

Folbex VA (Bromopropylate impregnated paper strips). The strips were set alight and allowed to smoulder in the hive, distributing the active ingredient as fumes.

The '**Frow**' remedy (named after Richard Watson Frow MBE) contained nitrobenzene, as well as Safrol oil, Ligroin (petroleum ether), Petrol or Oil of Wintergreen (methyl salicylate). It was highly inflammable and poisonous to both bees and humans. (Nitrobenzene is highly toxic and possibly carcinogenic.)

Both treatments had a poor therapeutic ratio – i.e. the amount required to kill the mite was too close to the amount that would harm or even kill the bees.

Even creosote has been used as a treatment

There is some cumulative evidence that essential oils are effective as treatments:

Oil of Wintergreen (Methyl Salicylate) and menthol have been used as treatments.

Grease patties (containing sugar and essential oils such as Oil of Wintergreen) are used in the USA

Frow's mixture contained an essential oil (Oil of Wintergreen)

"Some beekeepers believe that using thymol for several years has reduced acarine considerably."

The potential basis of the efficacy of essential oil is that their smell might mask the smell of the young bees that the female acarine mite uses to identify them as suitable hosts.

Hence, the use of Apiguard or similar anti-varroa treatments containing thymol might help treat acarine.

Acarine shortens the life of an infected bee, but this usually has little effect in the active season. The mite is spread from old bees to very young bees. A severe winter may cause an infected colony to dwindle in the spring.

Some strains of bees are more susceptible than others – the 'tracheal mite' is a huge problem in the USA where Italian/NZ crosses are used.

There are external acarine mites: A. exturnus, A. dorsalis and A. vagans – little is known about them

## Diagnosis

The disease can easily be diagnosed only by carrying out a dissection and microscopic examination (using a dissecting microscope with up to x40 magnification) of the primary trachea.

### Dissection to expose Acarine infestation (images from Dave Cushman website)

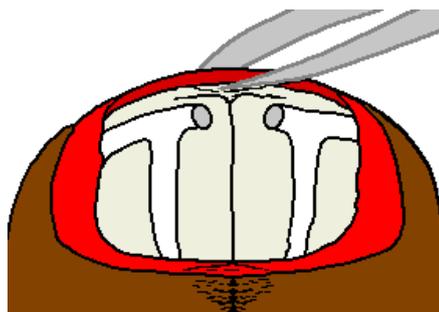
Collect a sample of 50 bees from the suspect colony. Choose bees crawling and unable to fly, found within about 3 metres of the front of the hive, rather than random collection from within the colony. The bees may be living, dying, or dead. Kill live bees with ethyl alcohol or by putting them in a deep freeze at -20°C.

Impale each using a double needle placed at an angle away from the head through the thorax between the second and third pairs of legs (as shown). The bee should ventral side up on an angled cork base, the angle is not critical, but is usually between 45° and 60°.

Using a single edged razor blade, cut the head and first pair of legs off; make the cut from behind the first pair of legs to the back of the bee's head, indicated by the red line on the drawing. Remove the severed head and front pair of legs with tweezers.



Use fine-tipped tweezers to peel the collar away (shown red at right) and expose the tracheae more fully. Pull upwards with a circular motion, following the ring of the collar. It will peel off easily, usually in one piece. Save the collar itself for later preparation as a microscope slide specimen, if required, by immersing in 70% isopropyl alcohol.



In a healthy or uninfested bee the tracheae have a uniform, creamy-white appearance. In infested bees the tracheae show patchy discolouration or dark staining, (melanisation, caused by mites feeding). In addition the eggs, nymphs and adult stages of the mite may also be seen in the trachea.



**Acarine infested trachea**



**Acarine infested trachea X40**

As mites enter through the spiracle, check the outer end of the trachea first. Light infestations may be difficult to see, heavy infestations are easily visible as shadows or lumpy dark objects in tracheae that can be clear to dark brown. Old and/or heavy infestations will render the trachea orange, brown or black.

## 10 the life cycle and natural history of *Varroa destructor*, its role in vectoring viruses and treatment methods.

### What is Varroa?

The varroa mite, *Varroa destructor*, formerly known as *Varroa jacobsoni*, is an external parasite of honey bees. Originally confined to the Asian honey bee, *Apis cerana*, it has spread in recent decades to the Western honey bee, *Apis mellifera*.

### Development within the colony and spread between colonies

#### Reproduction

The success rate of reproduction (new mature female mites) in worker brood is about 1.7 to 2 but the longer development period of drone brood increases it to 2-3.

The development and status of a colony affects mite population growth, and depending on circumstances, mite numbers will increase between 12 and 800 fold.

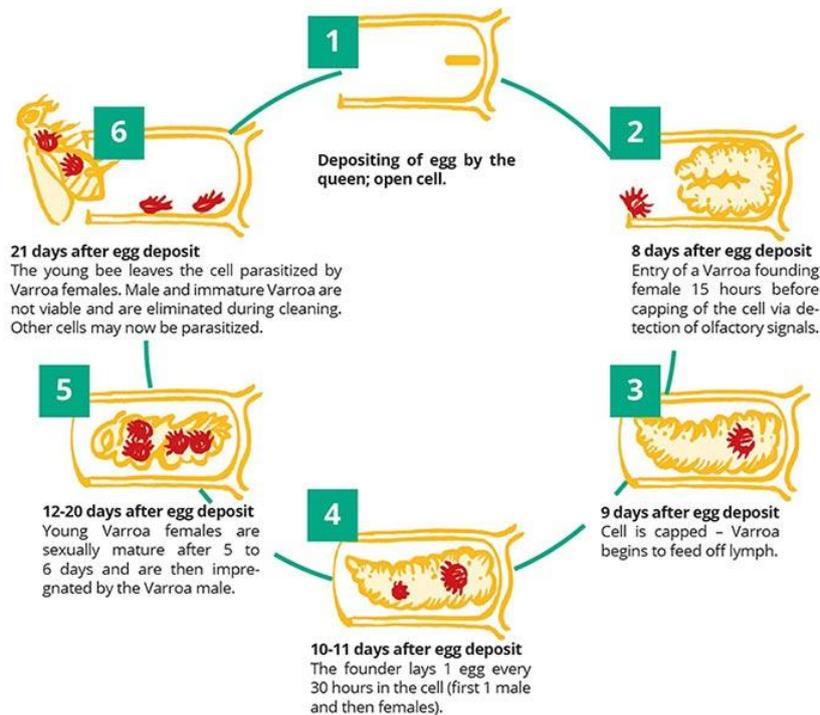


Figure 1: About Varroa destructor mites, Vêto-Pharma

### Life Span

The life expectancy of varroa mites depends on the presence of brood and will vary from 27 days to about 5 months.

During the summer varroa mites live for about 2-3 months during which time they can complete 3-4 breeding cycles, providing brood is available.

In winter, when brood-rearing is restricted, mites over-winter solely on the bodies of the adult bees within the cluster, until brood-rearing commences the following spring.

### How Varroa Spreads

Varroa mites are mobile and can readily move between bees and within the hive. However, to travel between colonies they depend upon adult bees for transport – through the natural processes of drifting, robbing, and swarming. Varroa can spread slowly over long distances in this way.

However, the movement of infested colonies by beekeepers is the principle means of spread over long distances.

### Effects of Varroa

Unlike *Apis cerana*, *Apis mellifera* has few natural defences against varroa. The mites feed on both adult bees and brood, weakening them and spreading harmful pathogens such as bee viruses.

Infested colonies eventually die out unless control measures are regularly applied.

#### References and Further Reading:

[About Varroa destructor mites, Vêto-Pharma](#)

<b>Signs and Detection</b>	
Recognition	Mature females are reddish brown, with flat bodies and six legs at the front Immature females and males are pale and smaller (exist in cells only)
On Frames	Low infestation – no visible signs Poor brood build up Poor brood pattern Varroa found in drone brood on uncapping
On bees	In severe infestation young emerge poorly developed with stunted growth and deformed wings Mites may be seen on adults
In hives	On solid floors – dead mites Open mesh floors – dead mites found on varroa tray
In cells	Mites may be seen on larvae, particularly drones, removed from sealed brood cells Bald brood and poor brood pattern Dead brood often discoloured brown and partly removed by bees Drone uncapping – mites may be found in unsealed drone brood at pink eyed stage

### **Monitoring**

Mite drop count using debris found on trays under varroa floor

- Mix debris from floor with methylated spirits. Varroa mites float to top, wax and other debris sink to bottom of container
- To calculate daily mite drop – count number of varroa mites and divide by the number of days since last count
  - Frequency – 4 times per year –
    - early spring,
    - after spring flow,
    - at time of honey harvest,
    - late autumn
  - All colonies if possible
  - Issues – Varroa trays may harbour wax moths if trays are not emptied

Drone Uncapping

- Test about 100 drone larvae
- Count trapped mites:
  - 5% infestation is light

- 25% infestation is severe
- May be carried out at every hive inspection

Production of drone brood may be encouraged by:

- Adding drone foundation to brood frames
- Leaving an empty frame for bees to produce comb
- Add super frame to brood chamber

**References and Further Reading:**

<http://www.nationalbeeunit.com/downloadDocument.cfm?id=1389>

**Methods of Control**

Current control methods used by beekeepers against varroa can be divided into two main categories:

‘Varroacides’ The use of chemicals to kill mites (or otherwise reduce their numbers). These are applied in feed, directly on adult bees, as fumigants, contact strips or by evaporation. These may include authorised proprietary veterinary medicines and unauthorised generic substances.

‘Biotechnical Methods’ The use of methods based on bee husbandry to reduce the mite population through physical means alone. Many of the most popular and effective methods involve trapping the mites in combs of brood which are then removed and destroyed.

	Advantages	Disadvantages
<b>Biotechnical methods</b>	<ul style="list-style-type: none"> <li>✓ Do not require the use of chemical varroacides</li> <li>✓ Can be combined with summer management</li> <li>✓ Inexpensive or free</li> </ul>	<ul style="list-style-type: none"> <li>✗ Can be time-consuming</li> <li>✗ Some need a high level of beekeeping skill</li> <li>✗ Generally not sufficient if used alone</li> <li>✗ Misuse can harm colonies</li> </ul>
<b>Authorised varroacides</b>	<ul style="list-style-type: none"> <li>✓ Proven efficacy</li> <li>✓ Proven safety</li> <li>✓ Convenient to use</li> </ul>	<ul style="list-style-type: none"> <li>✗ Mites likely to develop resistance</li> <li>✗ Residue problems in bee products if misused</li> <li>✗ Can be expensive</li> </ul>
<b>Unauthorised varroacides (generic substances)</b>	<ul style="list-style-type: none"> <li>✓ Some can be very effective</li> <li>✓ Usually relatively cheap</li> <li>✓ Usually natural substances</li> <li>✓ Many present low residue risk</li> <li>✓ Some offer control options not currently provided by biotechnical or authorised varroacides</li> </ul>	<ul style="list-style-type: none"> <li>✗ Use not approved by law in most situations</li> <li>✗ Efficacy may be low/variable</li> <li>✗ Safety typically not proven: some present serious risks to bees and beekeeper</li> <li>✗ Some present risk of residues in bee products</li> </ul>

### Some commonly used biotechnical methods

Method	How to use	Main features
<b>Drone brood removal</b>	<ol style="list-style-type: none"> <li>Place two shallow combs in the brood chamber in spring, and allow the bees to build natural drone comb beneath them. A good time to put these in the colony is when the queen first begins to lay up drone brood. Place the combs in the colony one at a time and alternate at 9 day intervals [a run of alternating pair of frames]. Another option is to use an empty deep frame fitted with a starter strip of foundation to avoid possible misshapen comb.</li> <li>When a drone comb is full of sealed drone brood (infested with varroa), cut it from the frame <b>before it emerges</b> and destroy it. Failure to do this will breed more mites. The frame can be re-used immediately.</li> <li>Repeat the process several times in the season for maximum effectiveness.</li> </ol>	<ul style="list-style-type: none"> <li>✓ easy to use</li> <li>✓ no special apparatus required</li> <li>✓ no varroacide used</li> <li>✓ well tolerated by colony</li> <li>✗ time-consuming</li> <li>✗ useful, but limited efficacy</li> </ul>
<b>Comb trapping</b>	<ol style="list-style-type: none"> <li>Confine the queen to a worker comb 'A' using a purpose-made comb-cage [available commercially].</li> <li>After 9 days confine her to a new, empty comb 'B' and leave comb 'A' in the brood chamber to become infested with mites.</li> <li>After a further 9 days remove comb 'A' [now sealed]. Confine the queen to a new comb 'C', leaving comb 'B' in the brood chamber.</li> <li>After 9 more days remove comb 'B'. Release the queen [or re-queen by introducing another queen] leaving comb 'C' in the brood chamber.</li> <li>After 9 more days, remove comb 'C'.</li> </ol>	<ul style="list-style-type: none"> <li>✓ can be very effective</li> <li>✓ no varroacide used</li> <li>✓ more bees recruited to foraging</li> <li>✗ time-consuming</li> <li>✗ requires good beekeeping skill</li> <li>✗ can harm/weaken the colony if used without regard to time of season (e.g. late summer)</li> </ul>
<b>Artificial swarm</b>	<ol style="list-style-type: none"> <li>Move parent colony to one side of the original site, at least 4 metres away.</li> <li>Place a second hive containing newly drawn combs and the queen (alone) on the original site to house the artificial swarm. Foragers will return to this hive creating the artificial swarm.</li> <li>After 9 days remove all but one queen cell from the parent colony. The cell can be protected in a queen cell nursery cage which prevents the virgin queen from leaving the hive to mate, but allows worker bees access to care for her.</li> <li>After 3 weeks all brood in the parent colony will have hatched. Transfer two bait combs of unsealed brood from the artificial swarm to the parent colony, and when they are capped, remove and destroy them. At this stage, cull the virgin and introduce a new queen to the parent colony.</li> <li>The old queen in the swarm can later be removed and the two colonies reunited.</li> </ol>	<ul style="list-style-type: none"> <li>✓ combines swarm control with varroa control</li> <li>✓ removes a high proportion of varroa mites present</li> <li>✓ new queen introduced</li> <li>✗ only suitable for use in the swarming season</li> <li>✗ it may be necessary to take precautions to prevent absconding in the artificial swarm – such as placing a queen excluder below the brood chamber for a few days.</li> </ul>
<b>Open mesh Floors</b>	<ol style="list-style-type: none"> <li>Fit a mesh varroa-monitoring floor [without a collection tray] to the hive.</li> <li>Many of the mites falling from the colony are alive. The mesh floor allows these to drop out of the hive rather than returning to the colony.</li> <li>A lower proportion of mites is considered to enter the brood to reproduce.</li> <li>Used in conjunction with other control methods this method helps keep mite numbers down.</li> </ol>	<ul style="list-style-type: none"> <li>✓ open mesh floor removes some live varroa</li> <li>✓ no debris on floor to encourage wax moths</li> <li>✓ improves hive ventilation</li> <li>✓ can use collection tray to measure mite drop when required</li> <li>✗ not sufficient if used alone</li> </ul>

Tables taken from NBU Varroa leaflet

The cycles of both comb trapping and the artificial swarm cycle are based on the 9 days from egg-laying to egg-capping, during which larvae are prone to infestation by the varroa mite.

## Misuse of agrochemicals

The active ingredients of many proprietary varroacides were originally developed to control pests of crops or livestock. When marketed as varroacides, they are specifically formulated for safe and effective use with bees. Under the authorisation process the specific formulation, along with the container and packaging (which may affect chemical stability) and the labelling are assessed for use in accordance with the manufacturer's instructions.

Home-made concoctions made with the active ingredients of these (often available as agrochemicals) should never be used. These pose serious risks to the user and to bees, and can leave harmful residues in bee products. Furthermore, misuse of this sort has been attributed to rapid development of resistance in countries overseas.

## Chemical residues in bee-products

Any chemical substance applied to bee colonies has the potential to leave residues in bee products. Following the following rules minimises the risk of harming bees with chemicals.

- Use authorised products with a proven track record in preference to alternatives that may lack reliable residue data
- Always follow the label directions supplied with all authorised products
- Never treat immediately before or during a honey-flow, or while supers are on the hive, unless the label directions of an authorised product specifically permit this

## Approved Varroacides and their Recording

This document from the National Bee Unit identifies the approved varroacides for use in the UK:

<https://secure.fera.defra.gov.uk/beebase/downloadDocument.cfm?id=1059>

All chemicals applied to a colony must be recorded as described here:

<http://www.nationalbeeunit.com/index.cfm?sectionid=110>

Extract from the above guidelines:

Name of Treatment	Active Ingredients	For the control of
Apiguard (Gel)	<i>Thymol</i>	<i>Varroa</i>
Apilife-VAR (Strips)	<i>Thymol, and essential oils (Camphor, Eucalyptus and Menthol)</i>	<i>Varroa</i>
Apistan * (Strips)	<i>Tau F</i>	<i>Varroa</i>
Api-Bioxal (Powder or Solution)	<i>Oxalic acid dihydrate</i>	<i>Varroa</i>
Apitraz (Strips)	<i>Amitraz</i>	<i>Varroa</i>
Apivar (Strips)	<i>Amitraz</i>	<i>Varroa</i>
Bayvarol * (Strips)	<i>Flumethrin</i>	<i>Varroa</i>
Dany's BienenWohl (Powder and Solution)	<i>Oxalic acid dihydrate</i>	<i>Varroa</i>
Mite Away Quick Strips (MAQS)	<i>Formic acid</i>	<i>Varroa</i>
Oxuvor (Powder and Solution)	<i>Oxalic acid dihydrate</i>	<i>Varroa</i>
Oxybee (Powder and Solution)	<i>Oxalic acid dihydrate</i>	<i>Varroa</i>
PolyVar Yellow (Strips)	<i>Flumethrin</i>	<i>Varroa</i>
Thymovar (Strips)	<i>Thymol</i>	<i>Varroa</i>
VarroMed (Solution)	<i>Formic acid, Oxalic acid dihydrate</i>	<i>Varroa</i>

## 11 the life cycle and effect upon colonies of the exotic pest *Tropilaelaps clareae*.



There are currently four species of *Tropilaelaps* mites, but only two of these (*Tropilaelaps clareae*, *Tropilaelaps mercedesae*) are considered to be serious threats to the Western honey bee, *Apis mellifera*.

The females of *Tropilaelaps clareae* are light-reddish brown and about 1.0 mm long x 0.6 mm wide, and the males are almost as large as the females (about one-third the size of a *Varroa* mite). The life cycle and parasitism of *Tropilaelaps* is similar to that of *Varroa destructor*.

*Tropilaelaps clareae* readily infests colonies of *Apis mellifera* in Asia, particularly those that produce brood

Adult mites enter and reproduce within particularly those of female lays three to larvae 48 hours about one day

The eggs hatch then the larva goes

(protonymph, deutonymph) before reaching the adult stage. Once hatched, all stages of both female and male mites feed on the haemolymph (blood) of the developing bee, causing damage through feeding by depriving the developing bee of essential nourishment required for growth.

Development from egg to adult takes about 6 days, and the adults (including the mother mite) emerge with the hatching adult bee and then search for new hosts.

Up to 14 adult mites and 10 nymphal stages of mite have been recorded in a single cell.

Mites move rapidly across the brood combs and are therefore easier to spot than *Varroa*, although they are much smaller.

Unlike the *Varroa* mite, *Tropilaelaps* cannot feed on adult bees because its mouthparts are unable to pierce the body wall membrane of the bees. The mites depend on the developing brood for food, and move from the adult bees to feed on the larvae as quickly as possible after emergence, so the phoretic stage is much shorter than that of *Varroa*, and may be only between 1-2 days. Gravid female mites (carrying eggs) will die within two days unless they deposit their eggs.

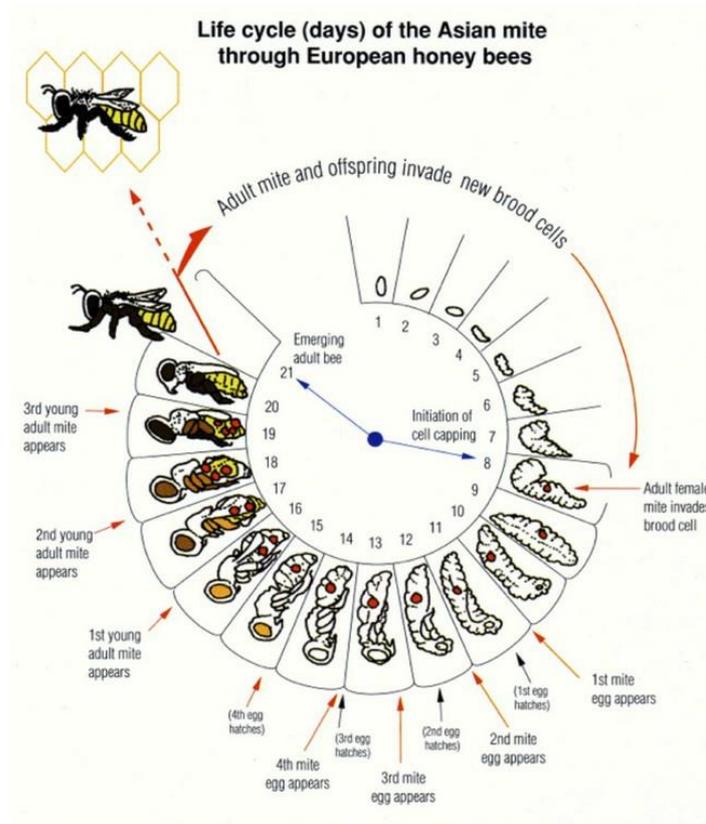
*Tropilaelaps* mites 'hide' in brood cells rather than on adult bees. Adult female mites may be seen walking rapidly out of cells and along the faces of the comb; immature mites are pale and remain motionless when feeding on their hosts in the brood cells.

*Tropilaelaps* infestation causes damage similar to *Varroa*: irregular brood patterns; stunted adults with deformed wings and shrunken abdomens. Effects may cause absconding or colony loss.



cells containing larvae and sealed brood cells, drones. Typically, the four eggs on mature bee after the cell is capped, apart.

after around twelve hours, through nymphal stages



*Figure 2: Life cycle of Tropilaelaps mite on European honey bee  
Denis Anderson, [www.beesdownunder.com.au](http://www.beesdownunder.com.au)*

12 pollen mites and their effects on stored pollen.



*Chaetodactylus furunculus*

Smaller than varroa, managed by varoacides, found in stored brood/pollen comb, leave dusty remains of pollen that is easily cleaned out by honey bee.

### 13 the life cycle of *Braula coeca* and its relationship with the honey bee.

- Egg laid in cells containing honey just under the cappings in cells
- After hatching larvae tunnel through the cappings feeding on honey and pollen
- Pupate inside tunnels
- Adult fly emerges 21 days after egg is laid and climbs onto body of a bee
- Feeds from the mouthparts of the bee, does not harm bee

#### Effects on colony

- It is an inquiline in bee nests – lives with bees without harm to either self or bees
- Eats food from mouthparts of bees, particularly the queen
- May act as irritant to queen if she is overloaded with Braula mites thus rendering her less effective
- Tunnels in cappings containing larvae make cut comb unattractive. Freezing kills mites.
- Varroacides have reduced numbers

#### Differences between Braula and Varroa

Braula	Varroa
Six legs	Eight legs
Harmless to colony	Harmful to adult bees in colony
Coexists in colony	Can overwhelm colony causing collapse
Does not pierce bees	Pierces bees to feed on haemolymph
Does not vector other diseases	Vector for viruses and disease
Feeds on mouthparts of bee	Feeds on larval food and larvae in cells and haemolymph in adults



Braula is a six legged insect or more precisely a wingless fly . It is remarkably similar to the Varroa mite (an arachnid with eight legs) at first glance due to its colouration, size and form, but on closer inspection the similarities disappear.

Compare the two images on the left. The Braula coeca in the left image looks more spider-like than the flatter crab-like form of the Varroa mite on the right.

*Taken from NBU Varroa Booklet*

14 the life cycles of the two wax moths *Galleria mellonella* and *Achroia grisella*, the damage they do to colonies and to combs, the effect of wax moth faeces on honey bee brood.

	<b>Greater Wax Moth</b> ( <i>Galleria Mellonella</i> )		<b>Lesser Wax Moth</b> ( <i>Achroia grisella</i> )
<b>Lifecycle</b>	Total cycle 4 weeks in good conditions 35 °C and food		Slightly shorter
<b>Stage</b>	<b>Duration</b>	<b>Greater Wax Moth</b> ( <i>Galleria Mellonella</i> )	<b>Lesser Wax Moth</b> ( <i>Achroia grisella</i> )
Egg	5-8 days, depending on temperature	Female moth enters the hive to lay eggs in crevices (where they are out of reach of nurse bees).	
Larva	6-7 weeks at 29°-32° C and high humidity; longer if weather cool and food short	Once the larvae hatch, they immediately search for comb on which to feed. Larvae feed on brood comb, so stored comb or colony weakened by Varroa or other diseases most at risk	
		Larvae tunnel through the comb, surrounding themselves with silken tunnels to which their faeces and bits of wax become attached. (If the hive is infested the frames become unusable.)	
		Sometimes larvae tunnelling through brood comb cause bald brood.	
		Sometimes larvae tunnel through comb honey; their tunnels under the cappings damage its appearance.	
		Freeze cut comb and sections for a few days to destroy any larvae.	
		Larva moults 7 times, reaching a length of around 20 mm. Its body turns grey with a brown prothoracic shield having a broad band across it.	

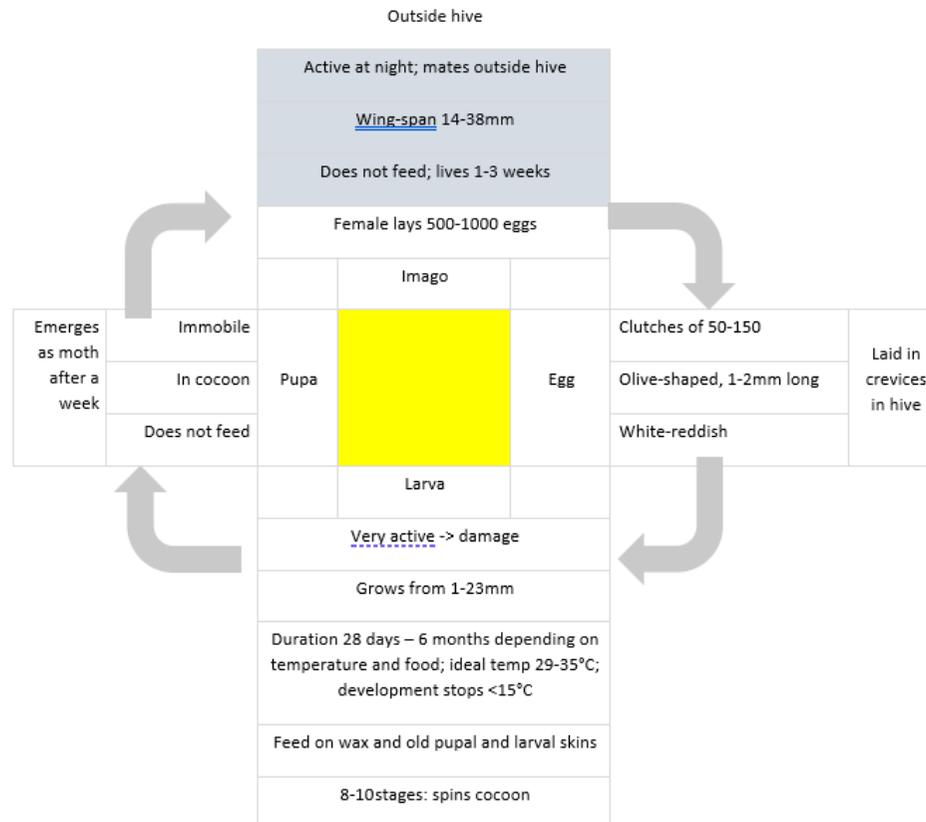
Pupa	6 to 55 days, depending on factors such as temperature	Excavates boat-shaped hollows in woodwork; can make holes in frames	
		Spins a silk thread cocoon; cocoons in rows. Pupate within cocoon	
		Rows of cocoons attached to the excavated indentations.	Rows of cocoons on the comb
Adult	Leave hive to mate, shortly after emergence. Males attract females with ultrasonic signals; females respond by fanning wings.		
	This moth flies from May to October in the temperate parts of its range, such as Belgium and the Netherlands.	Typically resides in milder climates.	
	Wingspan is 30–41 mm.	Average wingspan is 31 mm, length 15 mm.	
	Mouthparts are atrophied; adult does not feed		
	The wings are grey but the hind third, normally hidden, is bronze.		
			

Wax moth larvae feed on bee larval and pupal skins and pollen. In doing so they can destroy abandoned nests of feral bee colonies and thereby sanitise them (to some extent).

The Greater Wax Moth (*Galleria mellonella*) is more destructive and prevalent than the Lesser Wax Moth (*Achroia grisella*).

Poor management practices cause wax moth infestations; moths are attracted to scraps of burr comb lying around the apiary and drawn comb in empty and exposed supers or brood boxes. Drawn comb can be eaten away, making in unworkable for colonies of honey bees.

Some beekeepers store their supers “wet” because wet supers are less attractive to wax moth than dry ones.



## 15 the prevention of wax moth damage.

Store come in cool dry place.

Treat comb as appropriate:

### A. Biological methods e.g. Certan

Certan is a suspension of the bacterium *Bacillus thuringiensis*, which is specific against Lepidoptera larvae. Apply by spraying both sides of the frame. Lasts one season in the comb. Does not taint wax or honey.

### B. Flaming

Apply flame torch to used 'woodwork' (floors, roofs, boxes, crown board), concentrating on cracks and joins in the woodwork. The eggs and larvae of the wax moth are tiny and can easily get into these gaps, where they will hide and grow.

For **Chemicals**, **Temperature** and **Barrier** methods, stack equipment so that it is proof against the adult wax moth and mice. This means:

- use a floor or crown board as a base, raised on bricks off the ground; use entrance blocks and cover holes to make it moth-tight
- stack supers/brood boxes and frames on this. Use parcel tape to make the joints airtight over winter
  - metal grille or queen excluder and empty super if using sulphur (see below)
  - well-fitting roof.

### C. Chemicals

In all cases, ventilate combs well before re-using them in the hive.

#### Acetic acid

Effective against wax moth. Applied by fumigation. (See Section **Error! Reference source not found.** on page **Error! Bookmark not defined.**)

#### Sulphur:

Burn paper strips coated with yellow sulphur at the top of the stack; sulphur dioxide gas is heavier than air and sinks through the stack, killing every life form it encounters. Use a small tin can with holes or a smoker on its side with the top open as a burner, resting on the queen excluder.

Make sure you don't set the whole thing alight!

DO NOT BREATHE THE FUMES IN! Light upwind and stand well away.

Repeat in three to four weeks.

#### PDB (Paradichlorobenzene)

PDB now illegal. PDB does NOT kill wax moth at any stage in its lifecycle, but merely deters the adult from approaching the wax comb and laying the next cycle of eggs. It also taints wax and honey.

### D. Temperature

Freezing (to -15°C for at least two hours) is effective against moths and larvae. Safe and non-intrusive. After treatment protect frames, as below.

- A hard frost over several days will kill all stages of the moth in a stack made outdoors.
- Otherwise place in a deep-freeze for 48 hours. Stack as above afterwards.

#### E. Barrier

As wax moth enters hive through cracks, ensure that hive components fit together well.

Seal equipment for storage, making joints air-tight with packing tape.

Larvae love rolls of corrugated cardboard etc., so do not allow these to accumulate near stored wax.

## 16 the life cycle and effect upon colonies of the exotic pest *Aethina tumida*



The small hive beetle is a member of the family of scavengers or sap beetles, native in Africa. The adult beetle is dark brown to black in colour and about 5mm in length. Its antennae have a distinctive club shape. Adult beetles can be observed almost anywhere in a hive, although they are most often found at the rear of the bottom board.

Females lay irregular masses of eggs in cracks or crevices in a hive. The eggs hatch in 2–3 days into white larvae that grow to 10–11mm in length. The larvae feed on pollen and honey, tunnelling through comb with stored honey or pollen, damaging or destroying cappings and comb. They defecate in honey and thereby discolour it. The activity of the larvae causes the honey to ferment; it becomes frothy and develops a characteristic odour of decaying oranges. Damage and fermentation cause honey to run out of combs.

Larvae mature in about 10–16 days. When they are ready to pupate, they leave the hive and burrow into the soil near it. Pupation may last 3–4 weeks. Adults start to look for honey bee colonies as soon as they emerge, and females generally mate and begin laying eggs about a week after emergence. The adults may live for up to 6 months. Hive beetles may produce 4–5 generations a year during the warmer seasons. Heavy infestations cause bees to abscond.

Further Reading:

<http://www.nationalbeeunit.com/downloadDocument.cfm?id=17>

### 17 the damage caused by birds to the honey bee colony and methods of prevention.

Bird	Threat	Prevention
<b>Woodpeckers</b> esp. Green Woodpecker ( <i>Picus viridis</i> )	<p>Bores holes in side of hive in very cold weather when they cannot find forage on hard ground</p> <p>Can cause chilling and death</p> <p>Damages hive walls, frames and combs</p> <p>Loss of bees through eating, woodpecker has long barbed tongue to extract bee</p>	<p>Cover the hive with wired netting, leaving space between netting and hive but block gaps</p> <p>Cover with plastic bags but ensure ventilation is not affected</p>

### 18 the damage caused to colonies by mice and shrews and methods of prevention.

Pest	Behaviour	Prevention
<b>Mice</b> , esp. Wood Mouse ( <i>Apodemus sylvaticus</i> ) and House Mouse ( <i>Mus domesticus</i> )	<p>Enter hives in Autumn (October) looking for somewhere warm and dry to hibernate</p> <p>Mice have oval skulls and can squeeze through a 1 cm (3/8 inch) wide slot but cannot pass through the same diameter</p> <p>Feed on winters stores (pollen, honey) and possibly bees</p> <p>Smell, urine, faeces disrupt cluster, causing chilling and death of colony</p> <p>Nests destroy brood comb, cause mess</p> <p>Damage comb, frames and hive equipment</p>	<p>Fit mouse guards in September before ivy flow; remove in February or March</p> <p>Mouse guard should have a slot less than 8mm high or holes less than 10mm diameter</p>

Pigmy shrew holes in mouse guards need to be 6mm

19 the following abnormalities of the honey bee; "Addled Brood", white eyed drones, red eyed drones, "cyclops bees", undeveloped wings on honey bees, and bald brood.

Addled Brood	<p>Catchall phrase for unknown brood pattern</p> <p>Pepperpot</p> <p>Can be associated with sac brood</p> <p>Normally dead late stage pupae, with shrunken abdomens compared with developed head and thorax</p> <p>Related to genetic fault in queen brought about by colony stress</p>
White eyed drone	<p>A recessive gene gives drones white eyes. White eyed bees behave normally up until they are old enough to take their first flight. Once they fly, it's obvious that they are sightless, flying wildly in circles, they rarely return to the hive.</p>
Red eyed drones	<p>Think this is a mutation</p>
Cyclops	<p>Extreme mutation, where externally compound eyes merge into one. Internally still supported by two optic elements of the brain.</p>
Bald brood	<p>Genetic fault in queen</p>

20 the symptoms of poisoning by natural substances, pesticides, and other manmade chemicals.

A sudden reduction in the number of foraging bees, many dead or dying bees outside the hive, may indicate poisoning by bees alighting on sprayed crops. Legislation has reduced the number of incidents.

Apart from the evidence of dead bees, the colony may become bad tempered and shivering, staggering and crawling bees may be seen (like CBPV). Returning foragers spin around on the ground until they die. Dead bees usually have their proboscis ('tongue') extended.

## 21 the crops most likely to be sprayed causing damage to honey bee colonies.

Types of pesticide

Systemic – taken up by the plant through its roots and leaves, e.g. imidacloprid

Specific – attacks one particular species of insect

Crops most likely to be sprayed with chemicals toxic to bees

**Oil Seed Rape:** Cypermethrin, Deltamethrin, Fenvalerate, *Triazopho*; *imidacloprid*

**Field Beans:** Pyrethroid Hallmark Zeon

**Wheat:** imidacloprid. Note, bees will not forage on wheat, but might be caught by sprays applied to it.

**Oats:** imidacloprid

**Linseed:**

**Sugar Beet:** imidacloprid

**Fruit: apple, pear and cherry**

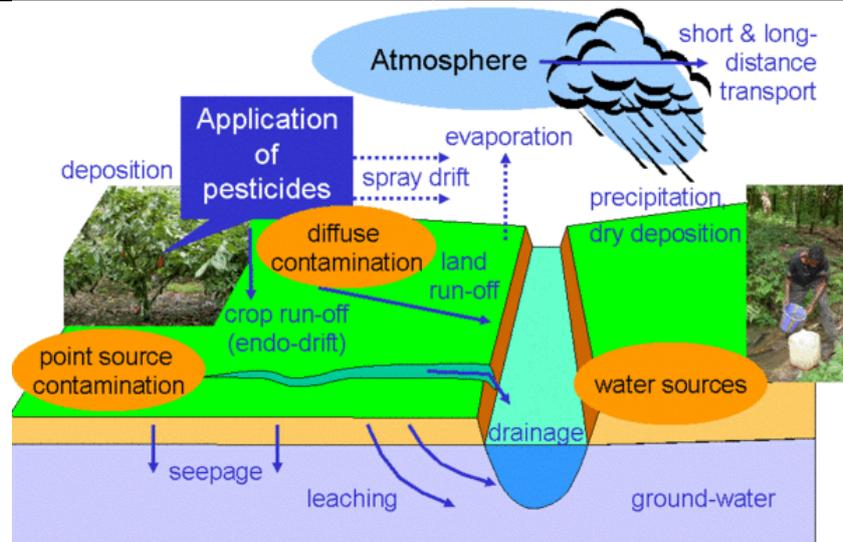
The bee can be caught by sprays:

- When the crop on which it is working is sprayed
- When spray is used on a crop not flowering but contains a lot of flowering weeds
- When a bee is flying over a crop which is being sprayed
- When wind drives spray to hive or bee forage (drift)

22 the methods of spraying and the sprays which are likely to be most detrimental to honey bee colonies and methods used to diminish the problem of spray poisoning.

All spraying follows the same theme of spraying droplets into air, so drift occurs, usually performed on mature crops with flowers.

Method	Diminish problem by:
Aerial spraying	Control of droplet size
Top dressing mature crop	Ultra-low and extremely low application rates
Foggers and air blast sprayers	No atomiser dispersal
	No drift application



## 23 Asian Hornet; life cycle, threat and actions if sighted



By Francis ITHURBURU - Own work, CC BY-SA 3.0,  
<https://commons.wikimedia.org/w/index.php?curid=25339575>

### **Extract from NBU website:**

*Vespa velutina*, the yellow legged hornet, commonly known as the Asian hornet, is native to Asia and was confirmed for the first time in Lot-et-Garonne in the South West of France in 2004. It was thought to have been imported in a consignment of pottery from China and it quickly established and spread to many regions of France. The hornet preys on honeybees, *Apis mellifera* and disrupts the ecological role which it provides and damages commercial beekeeping activities. It has also altered the biodiversity in regions of France where it is present and can be a health risk to those who have allergies to hornet or wasp stings.

In 2016, the Asian hornet was discovered in the UK for the first time, in Tetbury. After 10 days of intensive searching, the nest was found and later destroyed and on the same day, a single hornet was discovered in a bait trap in North Somerset. Genetic analysis has confirmed that the hornet nest found in Tetbury and the dead hornet found in North Somerset were of the same genetic population (*Vespa velutina nigrithorax*) as those which came from Eastern China to France. Although we cannot rule out the hornet arriving directly from the same area in China, we believe this is highly unlikely.

The following year, in 2017, another Asian hornet nest was discovered in Woolacombe by a vigilant beekeeper who reported seeing Asian hornets hawking and hunting in his apiary. Upon confirmation of the hornet, our contingency plan was again activated, and a nest discovered and destroyed. No other Asian hornets have been seen in the area.

### Appearance and biology of the Asian hornet



The Asian hornet is smaller than our native hornet, with adult workers measuring from 25mm in

length and queens measuring 30mm. It's abdomen is mostly black except for it's fourth abdominal segment which is a yellow band located towards the rear. It has characteristic yellow legs which accounts for why it is often called the yellow legged hornet and it's face is orange with two brownish red compound eyes.

### **Spring**

After hibernation in spring, the queen, usually measuring up to 3 cm, will emerge and seek out an appropriate sugary food source in order to build up energy to commence building a small embryonic nest. During construction of the nest, she is alone and vulnerable, but she will rapidly begin laying eggs to produce the future workforce. As the colony and nest size increases, a larger nest is either established around the embryonic nest or they relocate and build elsewhere.

### **Summer**

During the summer, a single colony, on average, produces 6000 individuals in one season. From July onwards, Asian hornet predation on honeybee colonies will begin and increase until the end of November and hornets can be seen hovering outside a hive entrance, waiting for returning foragers. This is the characteristic "hawking" behaviour. When they catch a returning bee, they will take it away and feed off of the protein rich thorax; the brood requires animal proteins which are transformed into flesh pellets and then offered to the larvae.

### **Autumn**

During autumn, the nest's priorities shift from foraging and nest expansion to producing on average 350 potential gynes (queens) and male hornets for mating, however, of these potential queens, only a small amount will successfully mate and make it through winter. After the mating period, the newly fertilised queens will leave the nest and find somewhere suitable to over-winter, while the old queen will die, leaving the nest to dwindle and die off. The following spring, the founding queen will begin building her new colony and the process begins again.

In light of the Asian hornet finding in the UK in September 2016, it is imperative that you make sure you know how to recognise and can distinguish them from our native hornet, *Vespa crabro* – a very helpful ID sheet and poster is available to help you:

[ID Sheet](#);

[ID Poster](#);

### **Monitoring for the Asian hornet**

Monitoring for arrival of the Asian hornet is strongly encouraged throughout the UK, but especially in areas where likelihood of arrival is considered to be highest (S & SE England). We strongly encourage that all beekeepers monitor for the Asian hornet. Should you wish to monitor for the hornet's arrival, some helpful tips and advice on how to make your own trap can be found in our fact sheet '[An Asian hornet monitoring trap](#)' and on our youtube video [How to make an Asian hornet monitoring trap](#).

Information from beekeepers in France shows that nest numbers can be reduced over time by > 90% in areas where traps are deployed in springtime coupled with IPM techniques and nest location and

destruction. Should the Asian hornets become established in the UK, springtime trapping will thus be a very useful management tool. When hanging out traps, please remember that it is important that damage to native wasps, hornets and any other insects is kept to an absolute minimum.

### Where to report sightings

If you think you have seen an Asian hornet, please notify the Great British Non Native Species Secretariat (NNSS) immediately. In the first instance sightings should be reported through the free Asian Hornet Watch App, available for [Android](#) and [Iphone](#).

Other methods of reporting the hornet also include using the NNSS [online notification form](#). Finally, you can send any suspect sightings to the Non Native Species email address [alertnonnative@ceh.ac.uk](mailto:alertnonnative@ceh.ac.uk) . Where possible, a photo, the location of the sighting and a description of the insect seen should be included.

If you would like to know more about the Asian hornet or any other Invasive Species, the [NNSS website](#) provides a great deal of information about the wide ranging work that is being done to tackle invasive species and tools to facilitate those working in this area.

It is also important that beekeepers sign up to BeeBase. In the event that the Asian hornet (or any other exotic threat to honeybee colonies) arrives here, efforts to contain it will be seriously jeopardised if we don't know where vulnerable apiaries are located.