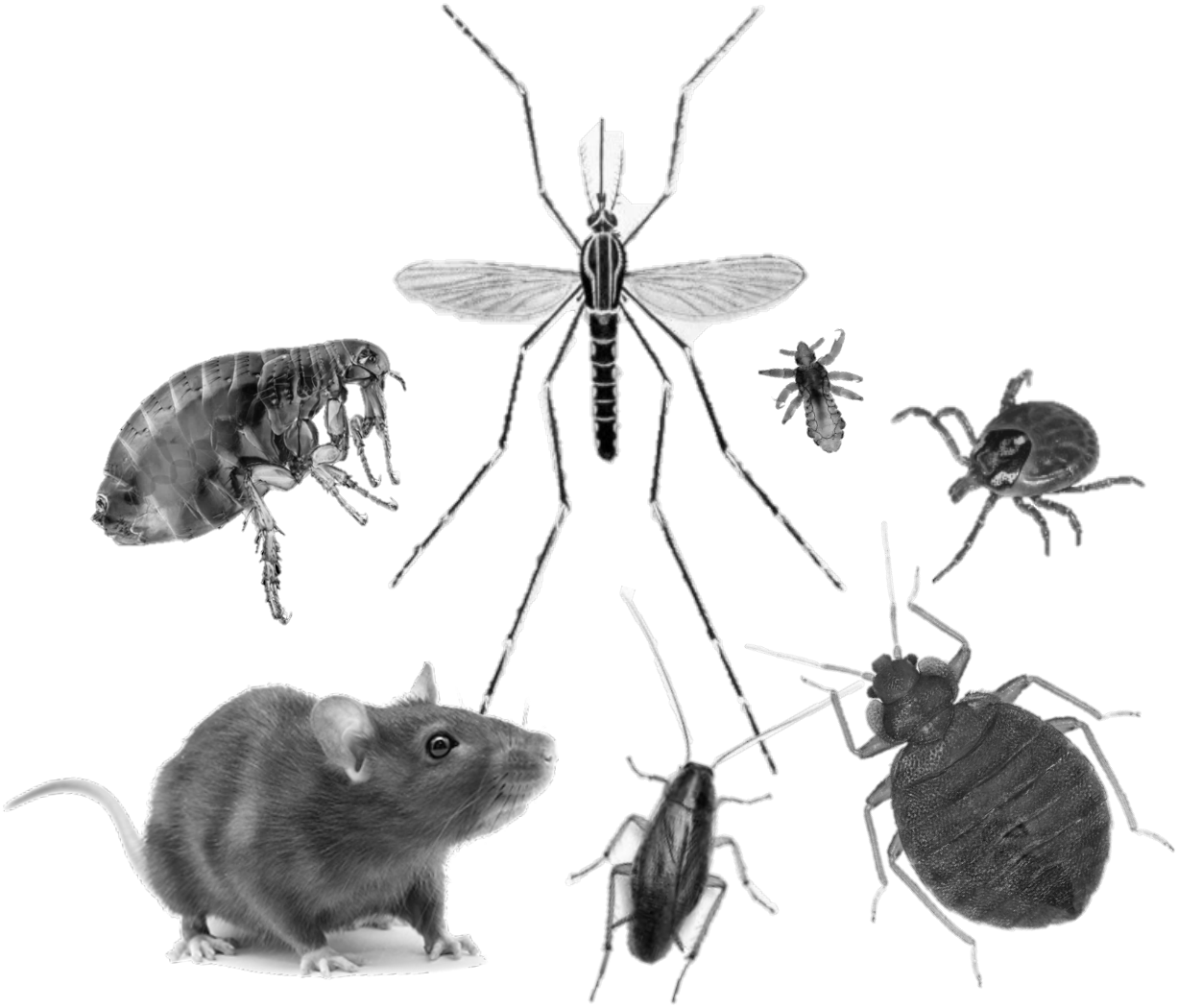
MEDICAL VECTORS

- An Education Guide for Border Health and Integrated Pest Management -



2019

Prepared by NZ BioSecure – Southern Monitoring Services

Purpose

Medical Vectors are an ongoing Public Health concern because of the role they play in the transmission of communicable diseases. New Zealand Public Health Services place a high priority on the strategy to exclude unwanted organisms, in particular exotic mosquitoes of human health importance. The risk of exotic mosquitoes becoming established in New Zealand is mitigated by:

- Monitoring New Zealand's Points of Entry in order to detect unwanted organisms
- Sanctioning the programs to disinsect inbound aircraft and to fumigate all imports of used tyres
- Responding rapidly to notifications of interceptions of unwanted organisms so as to contain control and eradicate them

The duties of Public Health staff are to implement mosquito surveillance at Points of Entry and to respond to interceptions of exotic mosquitoes as and when they occur.

The Ministry of Health has endorsed the use of the Guide for Public Health staff who are employed to deliver the operational outputs of mosquito surveillance and responses to exotic mosquito interceptions. The Guide provides the technical advice that staff will use to successfully carry out surveillance for and response to detections of exotic mosquitoes. It is to be read in conjunction with Reference A, the Environmental Health Protection Manual. The legislative envelope that sanctions this activity are references B, C and D.
It is envisaged that this document will be used by operational staff as a "Field Guide" and the operator will use it to access technical data that will ensure that their processes are robust and follow scientific standards.

References:

A. EHPO Manual Border Health Protection
B. Health Act 1956
C. Biosecurity Act 1993
D. International Health Regulations 2005

Disclaimer and Acknowledgement

This document has been put together as a guideline for Health Protection officers of the Ministry of Health New Zealand only and is not designed for widespread distribution.
Several of the images in this document were sourced from the internet e.g. the ICPMR website (http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/mosq it/mosquitoes.htm). Thank you to Richard Russell and Stephen Doggett who kindly gave permission for these images to be used here. All other images and text sources are cited where necessary.

1. Introduction to Vectors

1.1 What is a vector?

Disease-causing organisms can be viruses, bacteria, protozoan parasites or microworms.
Many of these organisms are species specific while others can be spread between different species and they can live as well as in humans as in other animals.
Actually the majority of vector-borne diseases survive in nature, utilizing animals as their hosts. In this case we speak of ***Zoonosis***, meaning that an infectious disease is able to be transmitted from wild or domestic animals to humans (or from humans to other animals called reverse zoonosis).

There are different methods of transmission for different diseases:
In some cases, zoonotic diseases are transferred by direct contact with infected animals.
Other diseases are spread by drinking water that contains the eggs of parasites or by eating the flesh of infected animals.
Other diseases are spread by ***vectors.*** That means in epidemiological terms, vectors are transmitters of disease-causing organisms, i.e. they carry the pathogens from one host to another. Arthropods account for over 85 percent of all known animal species and are the most important disease vectors globally.

There are two types of vector that convey infectious organisms to a host, mechanical and biological.
Mechanical vectors physically transport microorganisms from host to host; microorganisms do not multiply within them.
In contrast, microorganisms must develop or multiply within a ***biological vector*** before they become infective for the recipient.

The most significant mode of vector-borne disease transmission is biological transmission by blood-feeding arthropods. Several groups of arthropod play a key role in human and animal disease transmission, with mosquitoes and ticks being the most notable disease vectors.

- A **vector** is actually a secondary or intermediate host of a disease-causing organism with no ill effect. It is a host that harbours the parasite only for a short transition period, during which the pathogens change.
- A **reservoir host**, on the other hand, can harbour a pathogen indefinitely and is essential for the maintenance of the infection during times when active transmission is not occurring.
- A dead-end or **incidental host** is an intermediate host that does generally not allow transmission to the definitive host, thereby preventing the parasite from completing its development.
- However, the **primary** or definitive **host**, e.g. humans but also other animals, is the host that can become seriously ill. Parasites reach maturity and, if possible, reproduces sexually inside this host.

The key components that determine the occurrence of vector-borne diseases include:
- the prevalence of disease-causing pathogens suitably adapted to the vectors and the human or animal hosts;
- the abundance of vectors and reservoir hosts;
- the local environmental conditions, especially temperature and humidity; and
- the resilience behaviour and immune status of the host population.

1.2 Public health disease threat

Vector-borne diseases are prevalent in the tropics and subtropics and are relatively rare in temperate zones, although climate change could create conditions suitable for outbreaks of diseases such as Lyme disease, Rocky Mountain spotted fever, malaria, dengue fever and viral encephalitis in temperate regions.

There are different patterns of vector-borne disease occurrence:
Parasitic and ***bacterial diseases***, such as malaria and Lyme disease, tend to produce a high disease incidence but do not cause major epidemics. An exception to this rule is plague, a bacterial disease that does cause outbreaks. In contrast, many vector-borne ***viral diseases***, such as Yellow fever, dengue, and Japanese encephalitis, commonly cause major epidemics.

There has been a worldwide resurgence of vector-borne diseases since the 1970s including malaria, dengue, Yellow fever, louse-borne typhus, plague, leishmaniasis, sleeping sickness, West Nile encephalitis, Lyme disease, Japanese encephalitis, Rift Valley fever and Crimean-Congo hemorrhagic fever. Reasons for the emergence or resurgence of vector-borne diseases include:

- the development of insecticide and drug resistance;
- decreased resources for surveillance, prevention and control of vector-borne diseases;
- deterioration of the public health infrastructure required to deal with these diseases;
- unprecedented population growth;
- uncontrolled urbanisation;
- changes in agricultural practices;
- deforestation; and
- increased travel.

Changes have been documented in the distribution of important arthropod disease vectors. For example, the Yellow fever mosquito, *Aedes aegypti* has re-established in parts of the Americas where it had been presumed to have been eradicated; the Asian tiger mosquito, *Aedes albopictus* was introduced into the Americas in the 1980s and has spread to Central and South America; and the blacklegged tick, *Ixodes scapularis*, an important transmitter of Lyme disease and other pathogens, has gradually expanded its range in parts of eastern and central North America.

It is clear that people will always have to live with vector-borne diseases, but maintenance of a strong public health infrastructure and undertaking research activities directed at improved means of control (possibly utilising biological and genetic-based strategies), combined with the development of new or improved vaccines for diseases such as malaria, dengue and Lyme disease should lessen the threat to human health.

1.3 Vector and disease combat

Time line for key discoveries.

Arboviruses and protozoan parasites were not known to exist until the rise of modern medicine, with the germ theory and an understanding that viruses were distinct from bacteria.

1546. Girolamo Fracastoro proposed that epidemic diseases are caused by transferable tiny particles that could transmit infection by direct or indirect contact or even without contact over long distances.

1861. Louis Pasteur's experiments demonstrate the fact that the spoilage of liquid was caused by particles in the air rather than the air itself. These experiments were important pieces of evidence supporting the germ theory of disease.

1892. Dmitri Ivanovsky published an article describing for the first time a virus, a non-bacterial pathogen infecting tobacco plants.

1897. Ronald Ross an Indian-born British medical doctor discovered a parasite in the gastrointestinal tract of an *Anopheles* mosquito, which led to the realisation that malaria was transmitted by mosquitoes, and laid the foundation for combating the disease.

1898. Major Walter Reed, an U.S. Army physician, found that contact with faecal matter and food or drink contaminated by flies was the cause of typhoid fever in large U.S. Army camps in Cuba while fighting the Spanish-American War.

1901. Major Walter Reed confirmed Carlos Finlay theory that yellow fever is transmitted by a particular *Aedes* mosquito species.

1927. After Thomas Milton Rivers published the first clear description of a virus as distinct from a bacteria, in the same year, Adrian Stokes induced yellow fever in Rhesus monkeys from India and identified the disease as a virus.

1928. Max Theiler, a South African-American virologist and physician showed that the African and South American yellow fiver-causing viruses are immunologically identical.

1937. Max Theiler developed a vaccine against yellow fever.

1937. The West Nile virus was discovered and has since then been found in *Culex* populations causing epidemics throughout Africa, the Middle East, and Europe and since 1999 in the Western Hemisphere.

First measures against vector-born diseases

Yellow fever, alongside malaria, was a major obstacle in the construction of the Panama Canal. French supervision of the project in the 1880s was unsuccessful because of these diseases, forcing the abandonment of the project in 1889.

During the U.S.A. effort to construct the canal in the early 1900s, William C. Gorgas, the Chief Sanitary Officer of Havana, was tasked with overseeing the health of the workers. He had past success in eradicating the disease in Florida and Havana by reducing mosquito populations through draining nearby pools of water, cutting grass, applying oil to the edges of ponds and swamps to kill larvae, and capturing adult mosquitoes that remained indoors during the daytime.

Joseph Augustin LePrince, the Chief Sanitary Inspector of the Canal Zone, invented the first commercial larvicide, a mixture of carbolic acid, resin, and caustic soda, to be used throughout the Canal Zone.

The combined implementation of these sanitation measures led to a dramatic decline in the number of workers dying and the eventual eradication of Yellow fever in the Canal Zone as well as the containment of malaria during the 10-year construction period. Because of the success of these methods at preventing disease, they were adopted and improved upon in other regions of the world.

Paul Mueller, a Swiss chemist in 1939 discovered the insecticidal qualities and use of DDT in the control of vector-borne diseases such as malaria and yellow fever.

Control measures for vector-borne diseases will always be important as most zoonoses that are maintained in nature in cycles involving wild animals, are not amenable to eradication. Therefore, control methods targeting the arthropod vector help reduce the prevalence of these diseases and the risk to public health. Such methods should include:

- undertaking **personal protective measures** by establishing physical barriers such as house screens and bed nets; wearing appropriate clothing (boots, apparel that overlap the upper garments, head nets, etc.) and using insect repellents;
- areas such as ports and airports should be regularly **monitored**, with control measures utilized to prevent important arthropod disease vectors from entering the country;
- environmental modification to **eliminate specific breeding areas**, or chemical and biological control measures to kill arthropod vector larvae or adults may also be undertaken;
- some efforts to control vector-borne diseases **focus on the pathogen** - for example, vaccines are available for diseases such as Yellow fever, tick-borne encephalitis, Japanese encephalitis, tularemia, and plague;
- the **vertebrate host may be the target** - for example, vaccination of foxes against rabies in Europe and Canada is an effective means to reduce the threat of rabies;
- **reduction of host reservoirs**, such as rodents and birds, from areas of human habitation may lessen the risk for contracting certain vector-borne diseases such as plague and St. Louis encephalitis.

Please see chapter 4. for a thorough description and guidelines for Medical Vector Surveillance and Integrated Pest Management.

2. Vector-Biology

Before any measurements against vectors is conducted there is always one step before planning and action: You have to know your target. Knowledge about the lifecycle and the behaviour of the target vector is essential to find a way to detect and control it.

2.1. Mosquitoes

2.1.1 Species and genera

The word “mosquito” is derived from Spanish and means “little male fly”. Mosquitoes are in fact a type of fly, belonging to the order Diptera (“true flies” or “two-winged flies”) of the class Insecta and further separated from other flies in the family Culicidae.

There are about 40 genera (species groups, Figure 2.1.1) of mosquitoes worldwide, including approx. 3000 different described species.

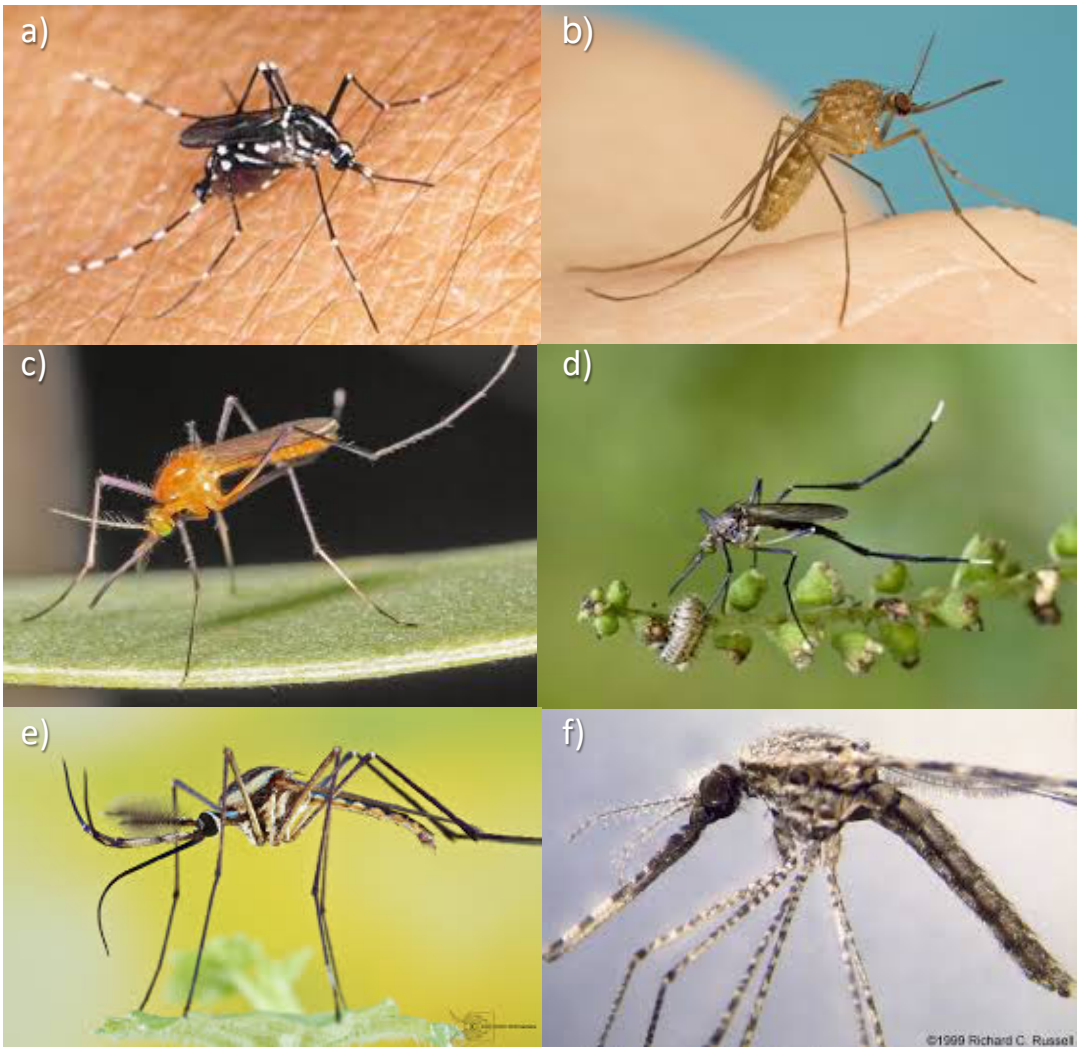


Figure 2.1.1. Mosquitoes from six different genera. a) *Aedes*, b) *Culex*, c) *Coquillettidia*, d) *Psorophora*, e) *Toxorhynchites*, and f) *Anopheles*

The family Culicidae is divided into 2 big groups, the subfamilies Anophelinae and Culicinae (Figure 2.1.1).

Table 2.1.1. List of mosquito genera in the two Subfamilies in Culicidae. Genus colour code: red, include unwanted species for New Zealand; green, present in New Zealand: blue, considered a big genus.

ANOPHELINAE	CULICINAE	
ANOPHELES	*AEDEOMYIA*	*MANSONIA*
BIRONELLA	*AEDES*	*MAORIGOELDIA*
CHAGASIA	*ARMIGERES*	*MIMOMYIA*
	COQUILLETTIDIA	*ONIRION*
	CULEX	*OPIFEX*
	CULISETA	*ORTHOPODOMYIA*
	DEINOCERITES	*PSOROPHORA*
	ERETMAPODITES	*RUNCHOMYIA*
	FICALBIA	*SABETHES*
	GALINDOMYIA	*SHANNONIANA*
	HAEMAGOGUS	*TOPOMYIA*
	HEIZMANNIA	*TOXORHYNCHITES*
	HODGESIA	*TRICHOPROSOPON*
	ISOSTOMYIA	*TRIPTEROIDES*
	JOHNBELKINIA	*UDAYA*
	KIMIA	*URANOTAENIA*
	LIMATUS	*VERRALLINA*
	LUTZIA	*WYEOMYIA*
	MALAYA	*ZEUGNOMYIA*

The Subfamilies are again divided into genera (Table 2.1.1), of which the most important and/or biggest ones are highlighted in colour. A big genus includes many species and is highlighted in blue.

Green means this genus is present in New Zealand whereas red means this genus includes species that are unwanted organisms to New Zealand (Figure 2.1.2)

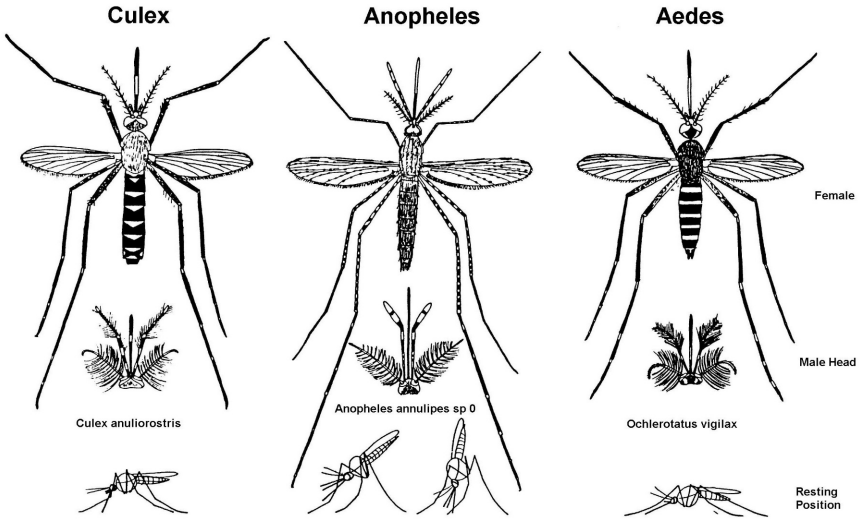


Figure 2.1.2. Scheme for three important mosquito genera showing females, males and resting position.

One genus can occur in many countries (e.g. *Aedes*) with species that are only abundant in one or few countries (e.g. *Aedes polyniensis*). Other species, such as *Culex quinquefasciatus,* although believed to be originated in the Americas, are introduced to many countries worldwide. *Maorigoeldia* instead is a genus that only occur in New Zealand (endemic genus). New Zealand contains very few mosquito species: 16 species established (12 endemic, 3 introduced plus 1 undescribed species found on the Chatham Islands).

NZ's native mosquitoes are primarily bird-biting species although some have adapted well to biting people. The introduced species tend to be nuisance biters of humans and known vectors of mosquito-borne disease. The table 2.1.2 shows a full list of NZ's described mosquitoes.

Table 2.1.2. List of mosquitoes species found in New Zealand. Introduced species are orange in colour.

- *Aedes antipodeus*
- *Aedes australis*
- *Aedes notoscriptus*
- *Aedes subalbirostris*
- *Coquillettidia iracunda*
- *Coquillettidia tenuipalpis*
- *Culiseta novaezealandiae*
- *Culiseta tonnoiri*
- *Culex asteliae*
- *Culex pervigilans*
- *Culex quinquefasciatus*
- *Culex rotoruae*
- *Maorigoeldia argyropus*
- *Opifex chathamicus*
- *Opifex fuscus*

2.1.2 Life cycle

The mosquito life cycle begins with an adult female laying eggs. Aquatic immature stages called larvae emerge and develop through four moults (instars), increasing in size until the final moult when it reaches the non-feeding pupal stage (Figure 2.1.3b and c). Inside the pupa the adult mosquito develops (either a male or female) and the terrestrial/aerial adult stage emerges from a split in the back of the pupal skin (Figure 2.1.3a and d). The adult mosquitoes feed, mate and the female develops eggs to complete the cycle and begin the next generation.

Some species of mosquito have only one generation per year. Others have several generations during a single season of favourable climatic conditions. Some continue to breed throughout the year, but may be more abundant in warmer seasons - this depends on the local environment, particularly temperature and rainfall.

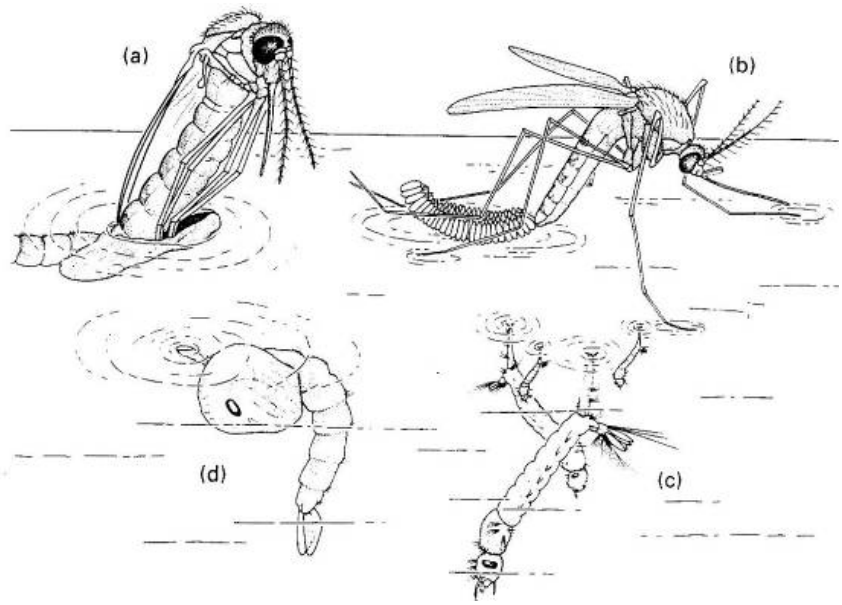


Figure 2.1.3. Life cycle of a *Culex pipiens* mosquito. a) emerging adult. b) female adult ovipositing egg raft on water surface. c) representative of each larval instar using siphon to breathe at water surface. d) comma-shaped pupa breathing using trumpet at water surface. Diagram ex Gullan, P.J. & Cranston, P.S. 2005. The Insects. 3rd Edition. Blackwell Publishing. 505pp.

2.1.2.1 Eggs

The female adult mosquito selects an appropriate habitat to lay her eggs. They are able to discern physical and chemical properties of different collections of water and choose between sites available. Factors including shade, temperature, salinity, water quality and the texture of the substrate may influence the female in her search for an appropriate oviposition site (see section 2.1.3.), which is dependent on species type as well as environmental factors.

The eggs are almost transparent when first laid, but gradually darken to brown or black as they mature. Eggs of different mosquito species have different morphology related to different oviposition strategies. Eggs of Culicine mosquitoes (e.g. *Culex* spp. and *Aedes* spp., Figure 2.1.4b and a respectively) are usually elongate-oval in shape with the anterior end rounded and the posterior bluntly pointed. Anopheline eggs (e.g. *Anopheles* spp., Figure 2.1.4d) are more cigar-shaped with flotation structures on each side.

The eggs are laid singly or in clusters and this can vary depending on the genus. *Aedes* species lay their eggs as single units (Figure 2.1.4b) and deposit them on moist substrate such as rock surfaces, moist earth and the inside wall of tree holes or containers above the receding water level. They also lay eggs under debris and in crevices in soil and dry mud, where they will be subsequently flooded. These eggs are able to withstand desiccation, and can survive long periods until they are submerged by water, at which time they begin to hatch.

Egg rafts of *Culex* and *Coquillettidia* spp. float on top of the water surface (Figure 2.1.4 b), while *Mansonia* spp. egg rafts can be found attached to the underside of leaves or twigs, just below the water surface. *Mansonia* species eggs differ from other mosquito species in that they have one end extended into a spike (Figure 2.1.4 c). Egg rafts cannot withstand desiccation and are usually associated with permanent or semi-permanent water bodies. They will hatch after about two days on the water and without constant water, they desiccate and die.

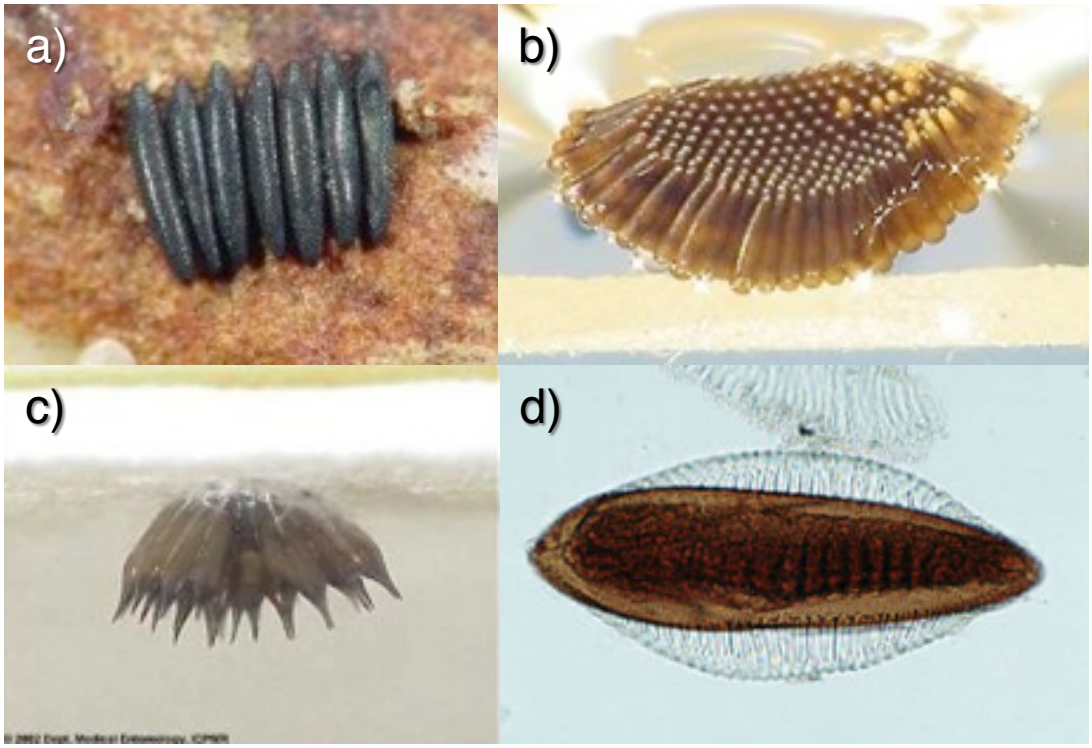


Figure 2.1.4. Four different egg morphologies and oviposition strategies in mosquitoes. a) *Aedes* spp. elongated-oval single eggs, b) *Culex* spp and *Coquillettidia* spp. elongated-oval eggs in a raft, c) *Mansonia* spp. spiked egg in a raft attached under debris and d) *Anopheles* spp. single eggs with floats on either side.

2.1.2.2 Larvae

The larval stage must have an aquatic habitat in which to complete its development to the pupal stage.

Figure 2.1.5. Relative size of larval instars from 1st to 4th.

The larvae hatch from the eggs and grow through **four** instars before developing into a pupa (Figure 2.1.5). Between each stage they moult their rigid outer skin so they can increase in size. The discarded skin is termed an exuvium/exuvia (singular) or exuviae (plural). The larval instar level is determined by the size of the head capsule, not the body length.

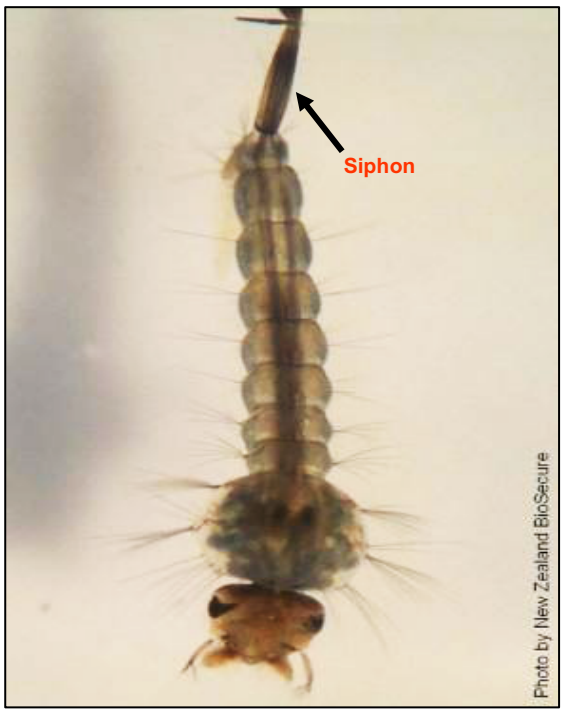


Figure 2.1.6. *Culex* sp. larva.

Larvae breathe air from openings (spiracles) at the tail end of the body, generally through a structure called siphon (Figure 2.1.6). They hang below the water surface with only the tip of the siphon exposed to the air. They can remain motionless on the bottom for some time, but need to return to the surface for air to prevent suffocation.

One of the two main exceptions to breathing behaviour occurs within larvae of the genera *Mansonia* and *Coquillettidia*. They attach to plants below the surface of the water after hatching, using a specially adapted piercing siphon and obtain their oxygen directly from the plant tissues (Figure 2.1.7a). Larvae of these two genera do not visit the water surface during their development and usually feed by filtering food particles from the surrounding water with their mouth brushes.

The second main exception in breathing behaviour occurs within the genus *Anopheles*, whose larvae do not have a siphon or breathing tube. Species of this genus lie alongside the surface of the water to breathe (Figure 2.1.7b).

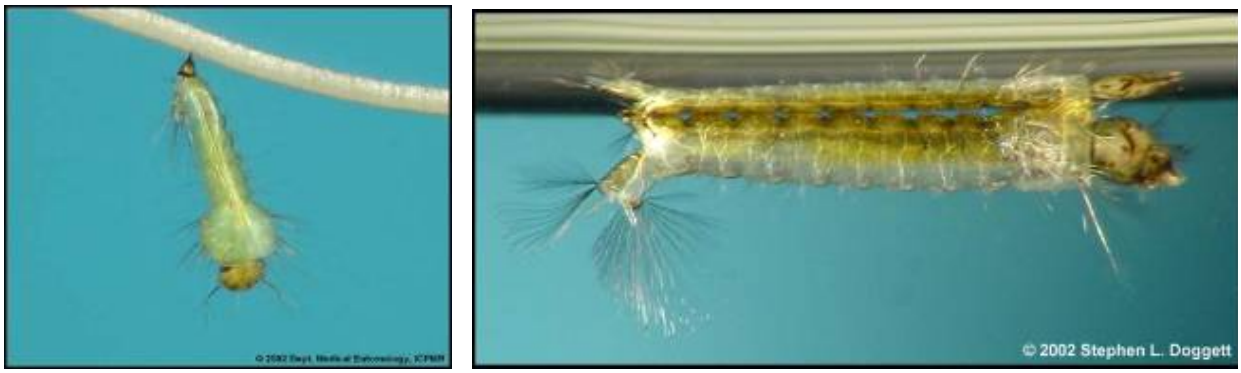


Figure 2.1.7. a) *Coquillettidia linealis* larvae breathing through a plant stem, b) *Anopheles* sp. larvae lying alongside the water surface.

Most larvae feed on microscopic organisms in the water and bottom detritus, either by filtering water through their mouth brushes or by grazing with specially adapted mouth appendages (Figure 2.1.8). Some larvae are predatory (*Aedes alternans* and *Toxorhynchites* spp.) and their mouth brushes are modified so they are strong enough to grasp prey. Some species feed habitually at the surface (*Anopheles*), some in the middle range below the surface (*Culex*) and others typically feed on the bottom of the habitat (*Aedes*).

Figure 2.1.8. Head of 4th instar *Aedes australis* larva with mouth brushes for filtering water

The time taken for development through the larval stages is dependent on a number of environmental factors, the most important of which is temperature. Availability of food and the extent of larval crowding within the habitat are also important.

During favourable summer conditions, *Anopheles* species may complete larval development in 7-10 days; *Aedes* species may complete larval development in as little as 4-5 days; and *Culex* species may require at least 7-10 days. Low temperatures usually delay development and may cause cessation of growth and induce an over-wintering of larvae in some species.

Identification of larvae is most easily accomplished with mature larvae, i.e. the fourth instar and microscopic examination is usually required. However, there are some genus characteristics that enable partial identification in the field.

For example, *Anopheles* species' lack of a siphon and larvae lying flat at the surface of the habitat when breathing or resting (Figure 2.7b,and 2.9a), distinguishes them from *Culex* (Figure 2.1.6 and 2.1.9b) and *Aedes* species which have siphons and hang suspended from the surface (Figure 2.1.9c).

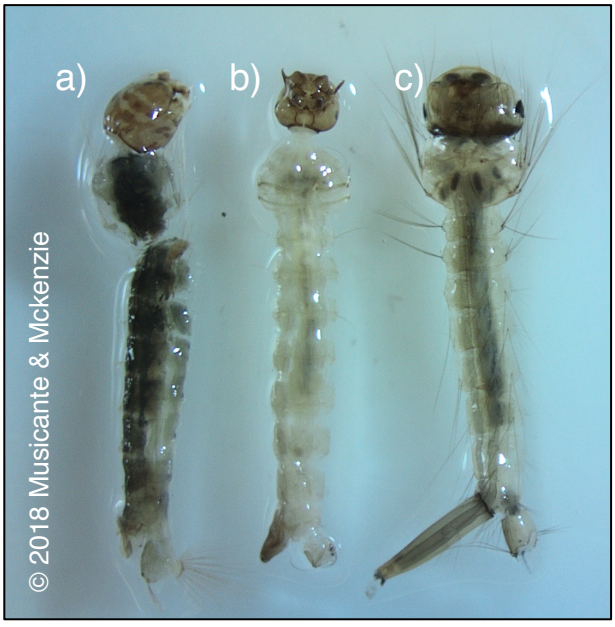


Figure 2.1.9. Larvae fom three different genera a) *Anopheles*, b) *Aedes* and c) *Culex*.

Culex species typically have longer siphons than *Aedes* species, which also can help assist in recognising the different genera in the field, however this can really only be achieved with experience and should always be checked under the microscope.

The larvae of *Mansonia* and *Coquillettidia*, although not commonly collected because of their attachment to submerged aquatic vegetation, can be identified as being from one of these two genera either by their attachment to a plant or if separated from the vegetation, by their modified siphon.

2.1.2.3 Pupae

After the 4th larval instar completes its development, it moults into a non-feeding but highly mobile stage called the pupa. Within the body casing of the pupa, the immature tissues are breaking down and adult tissues are forming.

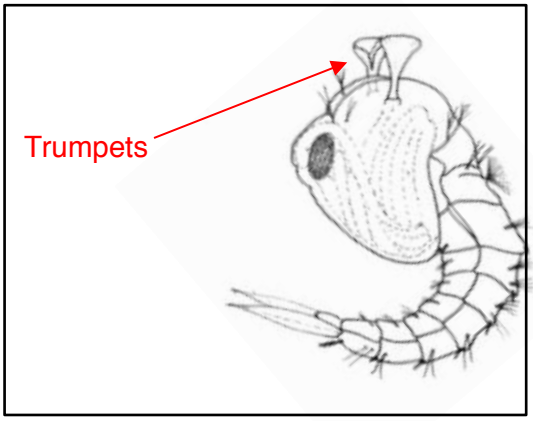


Figure 2.1.10. Mosquito pupa

The pupa breathes through a pair of tube-like organs (trumpet) situated at the 'head' end of the comma-shaped body (Figure 2.1.10).

Identification of pupae is only possible using microscopy. However, as with the larvae, some groups can be distinguished by their behaviour. *Mansonia* and *Coquillettidia* (Figure 2.1.11b) species for example, are different from other mosquitoes in that their pupae (like their larvae) obtain oxygen from plant tissues below the water surface, using modified trumpets.

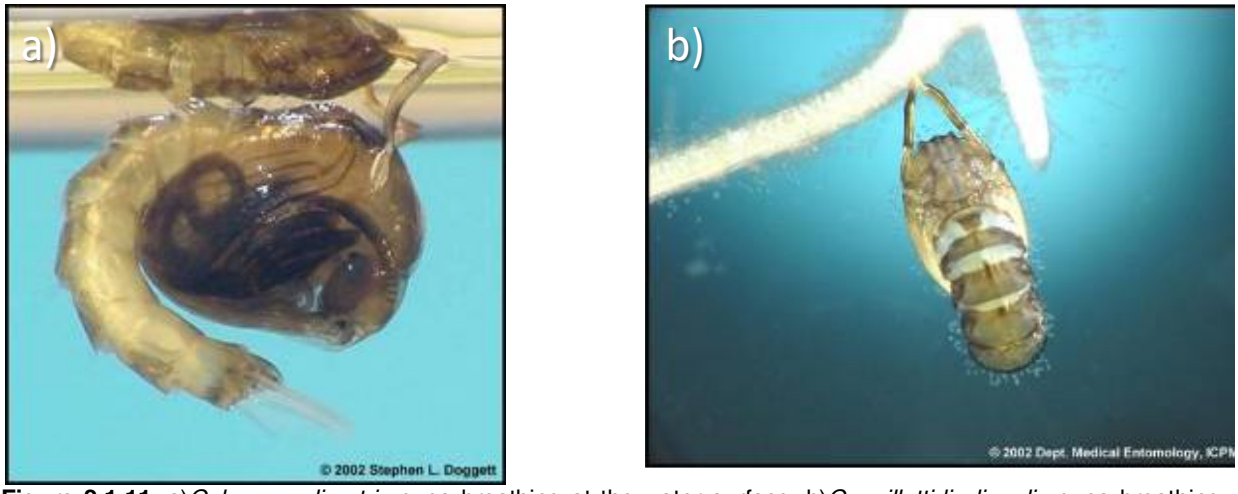


Figure 2.1.11. a) *Culex annulirostris* pupa breathing at the water surface, b) *Coquillettidia linealis* pupa breathing through plant stem.

The duration of the pupal stage again is dependent on temperature but is generally of the order of 2-3 days for *Anopheles*, *Aedes* and *Culex* species. Once the adult tissues have developed and it is time for emergence, the pupa swims to the water surface and stretches itself out to full length and the pupal skins splits along the back and the teneral (soft and pale) adult mosquito emerges above the water surface (Figure 2.1.12).

2.1.1.4 Adults

After emerging from the pupal casing, the adult mosquito rests on the water surface for a short time allowing its wings and body to dry, before flying off in search of a mate. Typically, males and females emerge in equal numbers in a single generation, the males of a species usually develop marginally more quickly than the females, and males are usually first to emerge from the larval habitat. This is not always noticeable in the field where generations may overlap. Male mosquitoes do not normally travel far from the breeding site and feed on plant juices, nectar from flowers and fruit exudates.

The adult female also seeks out a sugar meal of nectar or similar plant juices to replenish expended energy reserves and then mates with a male, usually near a breeding site at dusk.

Female mosquitoes mate only once, as the sperm packet introduced by a male during the mating act is sufficient for the female to fertilise all batches of eggs she subsequently produces.

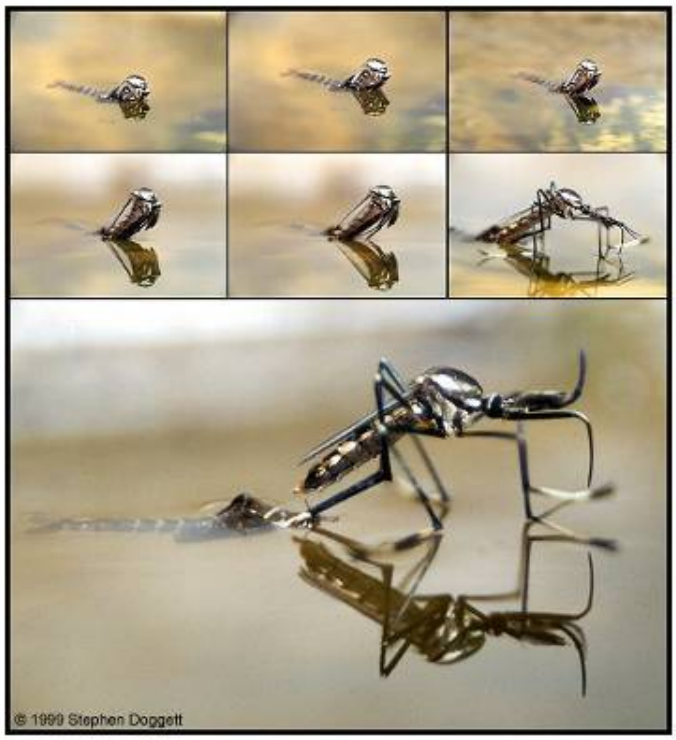


Figure 2.1.12. Adult *Toxorhynchites* mosquito emerging from the pupal case.

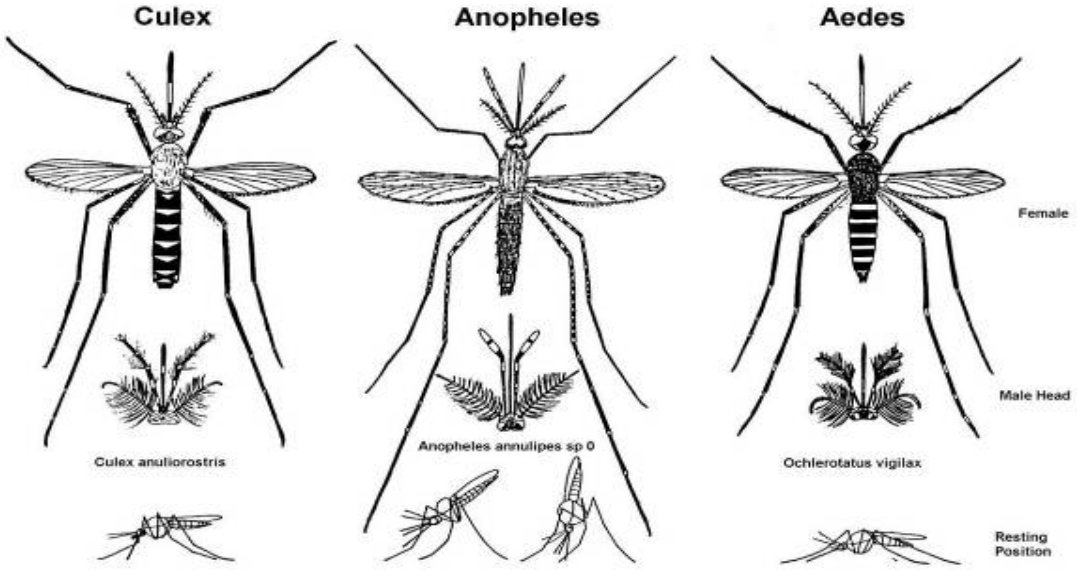


Figure 2.1.13. Summary diagram of the main mosquito genera. Ex: Carpenter & LaCasse (1955). Mosquitoes of North America (North of Mexico). University of California Press, Berkeley. 360pp.

Identification of adult mosquitoes is very complex, even to genus level and microscopes are required. However, the sex of mosquitoes caught in the field can often be determined by eye – if they stop flying around for a second! Adult males differ from females in that they have long palps protruding from their head next to their proboscis, and very bushy antennae compared to those of the females. An exception to this occurs in the *Anopheles* genus, which has both sexes with long palps, but the males still have the bushier antennae. Resting positions also vary between these genera (Figure 2.1.13).

The life span of adult mosquitoes is not well known. Some species apparently live one or two months during the summer, although under unfavourable conditions this period may be greatly reduced. Adults that hibernate during winter may live for six months or more. In laboratory conditions, *Aedes aegypti* adults have lasted as much as 240 days (about eight months).

All stages in the life cycle of a mosquito are dependent upon a number of environmental factors for their survival and development. Some common and measurable environmental factors, such as wind, light, temperature, rainfall and humidity have a known relationship to the survival of mosquitoes and can be used as the basis of an index for surveillance and control.

2.1.3 Habitats

The range of habitats utilised by mosquitoes is extremely diverse. With over 3000 species worldwide, mosquitoes have evolved to utilise almost any aquatic system in most parts of the world.An internationally accepted mosquito breeding habitat classification lists the following 11 larval habitats (Flowing stream, Ponded stream, Lake edge, Intermittent ephemeral puddle, Temporary pond, Permanent pond, Swamp/marsh, Natural container, Artificial container, Subterranean habitats-natural, Subterranean habitats-artificial, Figure 2.1.14).

The majority of mosquito species can be classified as either Container breeders or Groundwater breeders. Container breeders generally utilise smaller habitats such as tree holes, leaf axils and coconut shells. However, they have adapted well to artificial habitats, often found in discarded rubbish, tyres, tin cans, plastic sheeting as well as items that are in use, oil drums, buckets and guttering. Some containers may provide more permanent habitat, such as drain sumps and rock pools, but classification of those habitats may be debatable. Container breeders are often more commonly associated with populated areas as these generally provide a much greater opportunity for breeding.

Groundwater breeders utilise more expansive habitats: swamps, marshes, lake edges, field drains and mangroves etc. Groundwater breeders may be found in and around urban areas, although often their habitat will not occur within cities. An example of this involves saltmarsh habitats which often occur adjacent to urban areas. One of the key characteristics of saltmarsh species is often a long flight range, so the habitat existing outside of urban environments does not necessarily provide protection for the hosts within the city.

There are also some species whose behaviour allows for breeding in both container and groundwater e.g. *Culex gelidus* an important vector of Japanese Encephalitis.

Temperature plays a vital role in larval mosquito population dynamics. In tropical regions where there is no significant cold season, the seasonal pattern of mosquito population changes is related to the supply of water and rainfall. A slight rise in the level of water may cause an increase in mosquito production by re-establishing the less frequently inundated oviposition sites and increasing the number of temporary bodies of water. Excessively heavy rainfall and runoff during flood conditions may have a flushing effect and reduce the numbers of mosquitoes in the area. Such a reduction in the larval mosquito population is normally of a relatively short duration.

Adult mosquitoes will utilise different habitat for different purposes. In general, however, males will remain near the breeding habitat and only travel short distances to some source of a sugar feed (nectar, fruit etc.). Females will generally seek shelter from the environment, somewhere with little air movement, often dark, with sugar feeds nearby but also within distance of blood sources and breeding habitat. This will be significantly affected by the flight range of the mosquito - species with a short flight range will be found near to the breeding habitat, while mosquitoes with a longer flight range may be found sheltering several kilometres from the nearest breeding habitat.

Many mosquitoes prefer vegetation to rest in but domestic mosquitoes such as *Aedes aegypti* will be found in dwellings, resting in closets and under tables etc.


Intermittent ephemeral puddle


Saltmarsh


Permanent pond


Rock pool


Swamp


Natural container


Subterranean habitats-artificial


Artificial container

Figure 2.1.14. Eight examples of mosquito breeding habitat.

2.1.4 Hosts

For egg production, female mosquitoes require protein via a blood meal. A few species can develop the first batch of eggs using nutritional reserves carried over from the larval stage, this is called autogeny. They usually require a blood source to produce the second and subsequent batches.

The preferred source of a blood meal can vary widely between mosquito species. In general terms, mosquitoes are attracted to a warm-blooded host by a combination of factors; carbon dioxide, a product of respiration is an important attractant, as are various body odours, volatized chemicals, such as lactic acid.

These seem to be the longer range attractants. At closer distances, temperature can be a factor, as can visual perception at very close proximities.

Some mosquito species may take several blood meals to acquire sufficient protein for egg production. The female searches for secluded refuge where she can rest undisturbed, digest the blood meal and develop a batch of eggs. She will then fly off in search of additional blood meals to repeat this process. Subsequent blood meals may be taken the night of oviposition if a host is nearby, otherwise a day or more may elapse before the next feed.

Mosquitoes will utilise almost any land-based animal large enough to provide it with a blood feed. Some species are adaptable while others are quite host specific. Hosts include: Birds, Mammals, Reptiles and Amphibians.

The choice of host species for blood feeding is an important factor in disease transmission. *Aedes aegypti* is an urban mosquito with a preference for biting humans. Its ability to transmit dengue, combined with a close association with human populations, make it the most significant vector in dengue outbreaks. However, with diseases like Japanese encephalitis, where two host species are required for the disease cycle, a less specific mosquito species such as *Culex annulirostris* is a better vector.

2.1.5 Behaviour

Male and female adult mosquitoes are usually present in about equal numbers following emergence. Typically, the male mosquitoes reside near the breeding sites and have a shorter lifespan than females. Females may travel some distance to find a blood source. Only the female mosquitoes blood feed in order to obtain protein to produce fertile eggs.

Flight habits vary considerably: *Aedes aegypti*, arguably the most highly domesticated mosquito, typically flies very short distances (usually less than 500 metres). In studies some individuals have flown less than 35 metres from the water body they emerged from in their entire lifetime, while *Aedes vigilax* will comfortably travel 5-10 kilometres for sugar and blood feeds and may travel upwards of 300 kilometres in jet stream wind-assisted migrations. Although a coastal species, *Ae. vigilax* has been found as far inland in Australia as Alice Springs following a migration dispersal.

The possible flight range of Anophelinae mosquitoes varies considerably, depending on the species and circumstances in search of food and shelter. Generally, they will fly less than 3 kilometres, but they have been known to fly 30 kilometres in temperate climates with wind assistance.

Times of activity vary from species to species. Some species are active during the day (diurnal or day-biting) and others only at night (nocturnal or night-biting) with many more active at dawn and dusk (crepuscular).

2.2. Sand Flies

Sand flies are true flies belonging to the order Diptera, family Psychodidae, sub-family Phlebotominae (Figure 2.2.1). 'Sand fly' is the common name used to describe the small, hairy, biting flies of the subfamily Phlebotominae. The name 'sand fly' is often mistakenly used to describe other small biting flies such as biting midges of family Ceratopogonidae and blackflies of the family Simuliidae. Members of the family Psychodidae are characterised by their densely hairy wings, which may give them a moth-like appearance. The phlebotomines may be distinguished from other members of the family by the vertical 'v' position at which they hold their wings.

Figure 2.2.1. Adult sand fly (Psichodidae: Phlebotominae).

There are 6 genera and around 700 species of sand fly worldwide, with about 70 of these considered to be vectors of disease. Only the females are ectoparasites, feeding on the blood of a wide range of vertebrate hosts including lizards, birds, humans and other mammals.

The 6 sand fly genera are grouped into 'Old World' and 'New World' species. There are 2 genera of sand flies implicated in transmission of disease to humans - the genus *Lutzomyia* is the only genus of phlebotomine flies that feed on human blood in the New World and the genus *Phlebotomus* is the main genus to feed on human blood in the Old World. Although *Sergentomyia* has been known to rarely bite humans, this genus does not transmit disease to humans.

2.2.1 Life cycle

The sand fly life cycle begins with an adult female laying eggs onto organically rich soil. Eggs hatch into tiny terrestrial larvae, which develop through four instars (moults) until they reach their final moult and develop into pupae. Inside a pupa an adult male or female sand fly develops before hatching and flying away. The adults then feed and mate, with the female taking a blood meal to complete development of the eggs before laying them 3-8 days later and so starting off the next cycle.

Unfortunately, there have been no in-depth studies of sand fly development in nature and most studies have been focused on the easier to find adults.

2.2.1.1 Eggs

Unlike others in the family Psychodidae, female sand flies oviposit their eggs onto organically rich soil rather than water. The eggs must be laid in soil in relatively cool and humid areas such as piles of rubble, cracks in rocks and buildings, or animal burrows. Eggs are elongated and oval in shape and are very pale when first laid but darken with exposure to air and develop a single black 'eye spot'.

Females may produce up to 100 eggs from one blood meal and will deposit these eggs scattered around the breeding site. The eggs and their subsequent larvae are exceedingly hard to find as they are not laid in well-defined habitats, though different species may have very specific requirements for the habitats in which they lay their eggs.

2.2.1.2 Larvae

Under favourable temperature conditions, the eggs will hatch after a few days to two weeks, with the first instar larvae emerging from the egg through a j-shaped fracture. The larvae are elongated and worm-like but possess a sclerotised head capsule with chewing mouthparts and have thick bristles on the body, with two pairs of long caudal hairs on the posterior end. The first instar larvae differ in that they only have one pair of the caudal hairs. The larvae develop in the soil and may burrow up to 30 centimetres into the ground to feed on decomposing organic material such as dead insects, faeces of small vertebrates, rotting plant material and other detritus.

Larval development is temperature dependent so varies greatly from just a few weeks to a few months. In temperate regions the fourth instar larvae may diapause over the cooler winter before moving closer to the surface of the substrate and forming a pupa.

2.2.1.3 Pupae

The final immature stage of the sand fly is a non-feeding, inactive stage - the pupa. Development usually takes between 5-10 days during which time the golden brown pupae must remain in humid conditions as they are very sensitive to desiccation. The fourth instar larval skin and caudal bristles remain attached to the pupae at the posterior end, helping to fix it to the substrate on which it is developing.

2.2.1.4 Adults

Adults emerge from the pupal skin in darkness, usually just before dawn. Shortly before the adult sand fly is ready to emerge the wings and eyes turn black. Males emerge around 24 hours before the females, which gives their genitalia time to rotate 180° to the correct position for mating.

Figure 2.2.2. Feamle sand fly feading on a human host.

Sand fly adults are small with a body size of about 3 millimetres in length. The adult flies possess long slender legs and many long slender scales on the body and wings, giving a hairy appearance. The head is elongated, possessing well-developed eyes and antennae (Figure 2.2.2).

The antennae are long, may appear beaded, and protrude near a large set of black compound eyes. The mouth parts are at least as long as the head and consist of labrum and mandibles (all building the food channel), laciniae and hypopharynx, the latter containing the salivary channel. The five-segmented maxillary palps are well developed. The pointed wings contain numerous parallel veins.
Males and females cannot be separated according to their antennae like some other flies, but by the weaker developed mouth parts of males (which do not feed on blood), in which mandibles are absent, and – more easily – by the long external genitalia of males. The wings are upwardly pointing at rest, and are a distinctive feature of Phlebotominae sand flies as they are never closed or laid flat across the body. This feature explains another vernacular name in Brazil, “cangalinha” or “little yoke”.

All sand fly adults are brownish in daylight but their bodies are densely covered in oily hairs which give the insects a whitish appearance when illuminated. This explains some of their common

names, e.g. “manta blanca” (white mantle) in Ecuador, “palomilla” (little dove) in Colombia and “asa branca” (white wing) in Brazil.

2.2.2 Habitats

Breed in humid, terrestrial habitats, within a great variety of sites where organic matter is present. Specific habitats of sand flies are poorly known or understood. Most research comes from studying sand fly adults at their resting sites. The eggs and larvae are exceptionally hard to find as eggs are not laid in well-defined habitats. Different species may have very specific requirements for the habitats in which they lay their eggs. Eggs and larvae are usually found in places such as cracks in walls, piles of rubble and animal burrows.

Sand flies are found mainly in the tropics and sub-tropics, with fewer species found in temperate regions. There are no Phlebotominae sand flies in New Zealand.

2.2.3 Hosts

It is only the adult female sand flies, which require a host for blood feeding before developing her eggs. Most species have broad host ranges. Known hosts include humans, dogs, equines, sheep and birds.

2.2.4 Behaviour

Adults are mainly active in calm conditions in the early morning, evening and at night (although they can bite during the day if disturbed). When inactive, adult sand flies have habitat-specific resting sites that are characteristic of particular species. Resting sites are often similar or near to the larval breeding sites and are usually places that are cool, humid and dark. Sand flies are able to survive in dry environments by withdrawing to cool, humid resting sites during the day and then becoming active at night when temperatures are cooler

Mating takes place at or near hosts. The males congregate in leks (a gathering of males for the purposes of competitive mating display) on or near the host and produce sex pheromones. Females are able to sense the hosts using both host odour and the odour produced by the males. Vibration of the wings by males can be important in encouraging females to mate.

Biting activity is crepuscular or nocturnal and adults will rest in dark humid areas during the day such as cracks and tree trunks. Their bite is sometimes not felt and leaves a small round, reddish bump that starts itching hours or days later. However, more usually their bite is relatively painful because they are a pool feeder (telmophage) and they inject anticoagulants and antihistamines into a small wound they make in the skin of the host. It has been likened to a tiny drop of hot oil and has earned them the names “quemadores” (burners) or “pringadores” (stingers) in Colombia.

In most species, the females are gonotrophically concordant, meaning they require a blood meal to complete the development of each batch of eggs. A few species are autogenous and so do not require a blood meal. Males do not blood feed but feed on various sugary secretions of plants and some homopteran insects such as nectar and honeydew. Females will also feed from these sources but still require a blood meal for development of mature eggs. Only female sand flies bite. They use their mouthparts to create a pool of blood, which is then sucked up. They are annoying biting pests, which may probe several times before feeding producing a “pricking” sensation. They can cause trouble sleeping in unprotected areas (300+ bites per night). Eventual desensitisation may occur over time.

In a sand fly carrying *Leishmania* parasites the diameter of the foregut may be reduced or blocked by the masses of parasites. Presumably due to the masses of parasites in the foregut, blood ingestion is affected and infected phlebotomines probe much more often than uninfected

specimens. This increases the likelihood of *Leishmania* being transmitted. Phlebotomine sand flies have a weak flight and once on the host progress by a series of small hops. They do not hover around a host and as such are often not recognised as a biting nuisance. They also do not create any buzzing or whining noise audible to the human ear and so may not be noticed until after it has bitten.

2.3. Fleas

Figure 2.3.1. Adult flea.

Fleas are small, wingless and laterally flattened insects of the order Siphonaptera.

There are about 2100 species of flea worldwide, all living on a variety of warm-blooded hosts such as dogs, cats, rodents, birds and humans. Some well known flea species include:

- Cat flea (*Ctenocephalides felis*)
- Dog flea (*Ctenocephalides canis*)
- Human flea (*Pulex irritans*)
- Northern rat flea (*Nosopsyllus fasciatus*)
- Oriental rat flea (*Xenopsylla cheopis*)

Fleas are small (1 to 10 millimetres, though usually not exceeding 5 millimetres), agile, usually dark coloured (for example, the reddish-brown of the cat flea), wingless insects with tube-like mouthparts adapted to feeding on the blood of their hosts. Their bodies are laterally compressed (flattened side to side), allowing easy movement through the hairs or feathers on the host's body. Their legs are long - the hind pair well adapted for jumping - around 200 times their own body length. The flea body is hard, shiny, and covered with many hairs and short spines directed backward, also allowing the flea a smooth passage through the hairs of its host while preventing it from falling off or being dislodged (Figure 2.3.1). Their tough body is able to withstand great pressure and can survive the host's scratching.

Adult fleas and their feeding may cause irritation to the host and in some cases cause the host to develop an allergic reaction to flea saliva, resulting in rashes. The bites often appear in clusters or lines, and can remain itchy and inflamed for up to several weeks afterwards. Fleas can also lead to hair loss as a result of frequent scratching and biting by the animal, and can cause anaemia in extreme cases.

Fleas are also vectors of a number of diseases, most notably the Plague (bubonic plague (*Yersinia pestis* bacteria)), murine typhus (endemic typhus) and Hymenolepiasis (infestation by one of two species of tapeworm). They have a formidable reputation of claiming more victims than all the wars ever fought, as a result of the "bubonic" (Black Death) plague they spread throughout the world in the 14th century causing the deaths of over 200 million people. Now, these insects are better known for their irritation and pest status worldwide.

2.3.1 Life cycle

Fleas are holometabolous insects, going through 4 life stages of egg, larva, pupa and imago (adult, Figure 2.3.2). The flea life cycle begins when the female lays eggs after feeding on a host. A female flea can lay well over 500 eggs during its lifetime, allowing for massive population explosions. Adult fleas must feed on blood before they are able to reproduce. It may take as little as 2 weeks for a flea to complete its life cycle (Figure 2.3.2).

The life stages of a flea population are unevenly distributed with around 34% of the population being eggs, 57% larvae, 8% pupae and 1% adults.

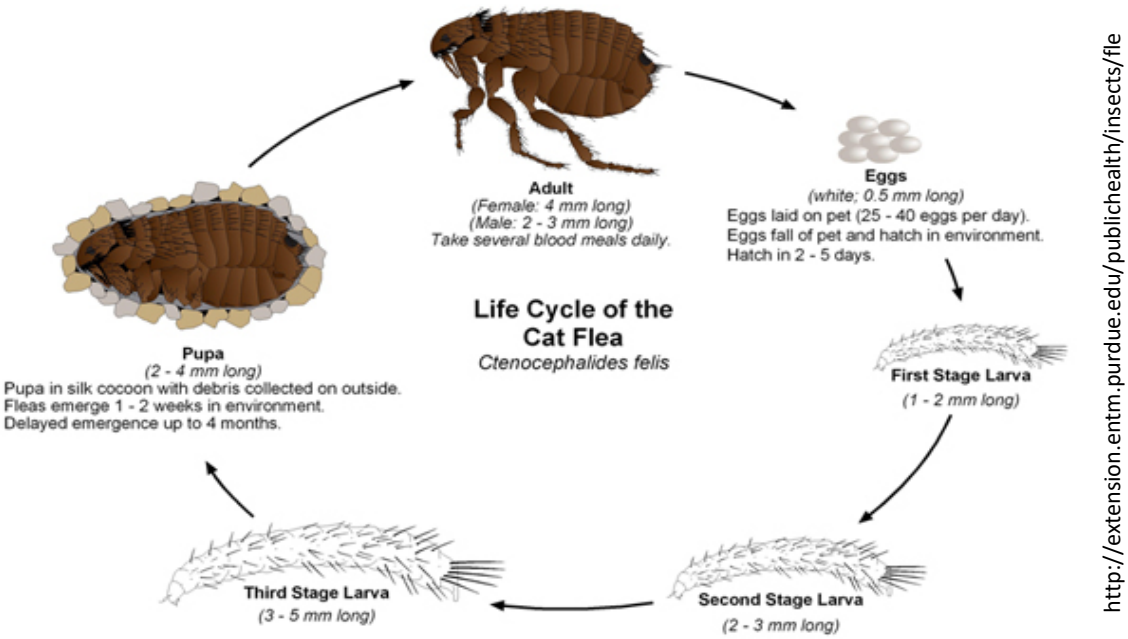


Figure 2.3.2. Life cycle of a cat flea.
Scott Charlesworth, Purdue University, based in part on Elbel, R.E., 1991, IN: Immature Insects, Volume 2

2.3.1.1 Eggs

Fleas lay tiny, white oval shaped eggs in batches of up to 20 (Figure 2.3.3). They are smooth, oval, pearly white and approximately 0.5 millimetres in size. Depending on the temperature and humidity, the eggs will begin hatching one and a half days to a week after being laid.

Because the eggs are smooth, and not laid attached to hair or skin, they easily fall off the host. This means that most eggs end up in areas where the host spends a lot of time such as in bedding, carpets and rest areas.

Figure 2.3.3. Flea eggs.

2.3.1.2 Larvae

Figure 2.3.4. Cat flea larvae.

Flea larvae emerge from the eggs to feed on any available organic material such as dead insects, faeces and vegetable matter. They are unable to see and are negatively phototropic, moving away from light and keeping to dark places like cracks, crevices and bedding. Larvae undergo 3 moults before pupating. With an adequate supply of food this may happen within 1 or 2 weeks (Figure 2.3.4).

2.3.1.3 Pupae

Figure 2.3.5. Flea cocoons.

After going through 3 larval stages, a sticky substance is secreted to spin a silken cocoon and incorporate debris from the surroundings. The cocoon provides a protection barrier resistant to chemicals and pesticides (Figure 2.3.5).

One or two weeks later, under optimal conditions, the adult flea is fully developed inside the cocoon and is ready to emerge.

The pupal stage can vary greatly in length between individuals as emergence may be delayed with the pupa lying dormant until cues alerting a potential host are sensed. Vibrations, including sound, heat and carbon dioxide are all stimuli which may indicate the presence of a host and trigger emergence.

Pupae may remain dormant for years if they are not stimulated to hatch. This explains why some people report coming home to a flea plague after being away for some time. Their vibrations when they re-enter the house can trigger a wave of flea emergence in dormant pupae.

2.3.1.4 Adults

Both the male and female adult fleas are ectoparasites and require a host to survive. Both feed on the blood of the host and the females require this not only as a source of food but it is essential to the development of her eggs.

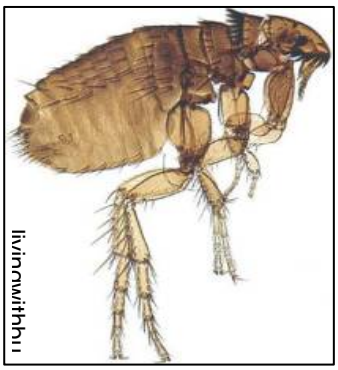


Figure 2.3.6. Adult cat flea.

Adult fleas (Figure 2.3.6) must take their first blood meal within about a week of emergence, but after this first meal they may survive for a number of months with no food.

Adult fleas may survive in the environment without a host for days, although they usually live on the host they are feeding on.

The life span of an adult flea ranges between as little as 12 days to over 100 days. If the host should die before the flea, then the flea will vacate to find a new host.

2.3.2 Habitats

Fleas parasitise hosts in nearly all habitats where their hosts live and are found not only on their bodies but also in their burrows and nests. Bird fleas only parasitise species that reuse their nests year after year, including swallows, seabirds, some ground-dwelling species, and those living in tree holes and cavities. A few flea species that live in coastal, warm and moist, and tropical regions are free-living. Cat, dog, and human fleas all regularly spend time away from their hosts and are commonly found on the floors of homes, foot paths, animal pens, and pet beds. Most larvae are free-living and do not make their home on the body of a bird or mammal. They are usually found in pet beds and nests.

2.3.3 Hosts

About 5% of all flea species occur on birds, while the remaining 95% parasitise, or live off of, mammals. They usually do not parasitise amphibians and reptiles.

Some fleas can attack a range of hosts and their ability to move from one host to another allows for the possible transfer of pathogens including viral, bacterial and parasitic diseases. For example, cat fleas are the intermediate host for the dog and cat tapeworm (*Dipylidium caninum*) which is also easily transmitted to humans.

The main flea species that attack humans include the cat flea *Ctenocephalides felis* (Figure 2.3.6), the dog flea *C. canis,* and the human flea *Pulex irritans*. The latter two species are relatively rare. The common cat flea is found on both cats and dogs. It is this species which is often identified in attacks on humans and usually responsible for flea plagues.

2.3.4 Behaviour

Newly emerged fleas which have not taken a blood meal are almost black in colour and very flat. As soon as the adult flea has hatched out of the pupa it will keep jumping until it finds a suitable host. After they have found a host and taken a blood meal the engorged flea is less flat and turn a red/brown colour.

Adult fleas locate their hosts with visual, chemical and physical cues. Carbon dioxide will cause a random jumping response, though visual and heat stimuli are their primary means of finding a host. Once fleas have found a suitable host both the male and female will blood feed, then reproduce and the females will lay eggs.

Adults feed every 4-6 hours for around 5 minutes at a time.

Egg production begins 2 days after the first blood meal is taken by the female. The largest number of eggs is produced 6-7 days after the first blood meal. The average female flea will lay between 20-50 eggs per day and up to 800 eggs over a lifetime.

Eggs are laid loosely on the host and quickly fall off into the surrounding environment, usually in a place frequented by the host such as a den or bedding. Once hatched the larvae remain in this area but seek out the darkest areas, moving away from sources of light.

2.4. Lice

Lice (plural), louse (singular) are small (0.5-8 millimetres), wingless, dorsoventrally flattened, ectoparasitic insects of vertebrates. They are found on all continents, including Antarctica.

There are two types of lice, chewing lice (Figure 2.4.1a.) and sucking lice (Figure 2.4.1b.). Only species of sucking lice cause problems for humans.

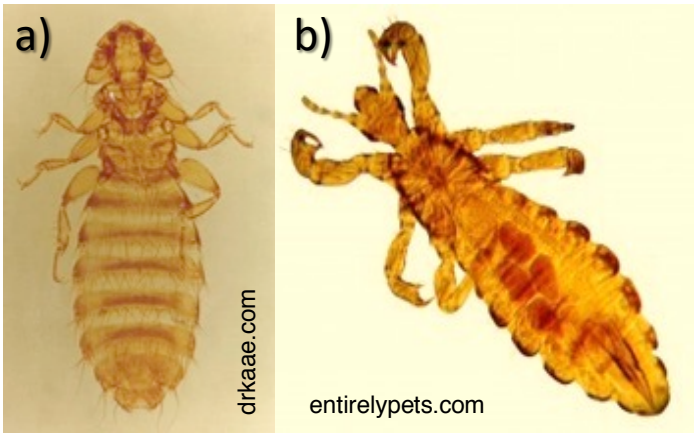


Figure 2.4.1. a) Chewing louse, b) sucking louse

Many species are pale whitish or yellowish, while other species are brown or black. If feeding on blood, a louse's colour may become considerably darker. Some species have colour patterns that help them to blend in with the fur or feathers of the animal on which they live.

The extinction of a bird or mammal species leads directly to the extinction of many of their parasites. Nearly 370 species of birds and mammals are listed by the IUCN as Extinct in the Wild or Critically Endangered. At least 50 species of lice share their fate. By 1990 at least 8 species of lice had already followed their host birds and mammals to extinction.

2.4.1 Life cycle

Lice are hemimetabolous so undergo incomplete metamorphosis whereby the egg hatches out into a nymph (tundergoes hree nymphal stages), which is already similar in appearance to the final adult stage (Figure 2.4.2.). Lice have no pupal stage.

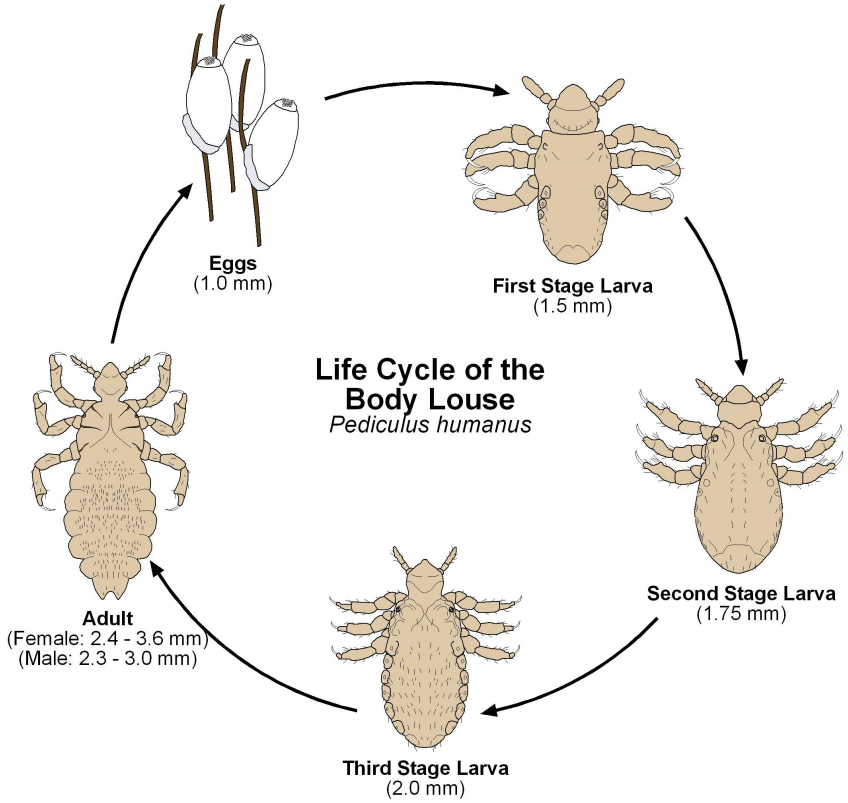


Figure 2.4.2. Life cycle of the body louse Pediculus humanus.

2.4.1.1 Eggs

Louse eggs or “nits” are sub-cylindrical in shape and are glued to the base of a host’s hair, feathers or clothing (Figure 2.4.3). The exception to this is the body louse, which tends to oviposit in the hosts clothing, particularly along seams. They have an anterior operculum which is pushed off by the emerging first instar nymph. Louse eggs hatch in 4-15 days.

Figure 2.4.3. Head louse egg.

A complete head louse egg consists of a tube which encircles the hair shaft with the egg attached to the end furthest from the scalp. Note the operculum forming a lid on the top of the egg (Figure 2.4.3). The sides of the egg are rounded when egg is alive, and collapse in when it’s dead. A hatched egg has lost the operculum and has a flat top in profile. Living louse eggs tend to be pale white, while dead lice eggs appear orange.

2.4.1.2 Nymphs

There are three nymphal stages/instars which closely resemble the adult louse, but are smaller, lack external genital openings and have progressively more setae, i.e. first instars have less setae than second instars and so on. Each nymphal instar typically lasts 3-8 days before moulting to the next stage.

2.4.1.3 Adults

Adult lice live for up to 35 days. Mated females glue 2-10 eggs per day, one egg at a time onto a hair, feather or clothing, depending on the species. Females are typically larger than males.

For most species of lice, it is known that there are both males and females and they reproduce primarily by mating. A few species reproduce by parthenogenesis, a process where the young develop from unfertilized eggs.

2.4.2 Habitats

Chewing and sucking lice are ectoparasites, organisms that live on the outside of their host. All species spend their entire lives on the body of the host animal. They require the constant temperature and moisture of this habitat to feed and reproduce. Most species of lice are found only on a single kind of host or on small groups of closely related species.

Although the host body would seem to be a uniform habitat, it is actually a series of smaller habitats that differ slightly in terms of temperature and moisture. For example, the different parts of a bird's body, such as the head, back, wings, and rump, are completely different habitats from the viewpoint of a louse. These different habitats might allow several species of lice with slightly different temperature and humidity requirements to live on the same host animal without having to compete with one another directly for food and space. Some species occupy more than one part of the body at different times in their lives. For example, a species of lice lives inside the throat pouches of pelicans and cormorants where they feed on blood. However, they must return to the head feathers to lay their eggs.

2.4.3 Hosts

Many species are host specific and feed on a single host species. Some are further specialised, in that they predominantly occur only on certain body regions of their hosts. Chewing lice feed mainly on feathers, fur, skin debris, or (rarely) blood of birds or mammals (Figure 2.4.1a). Sucking lice feed exclusively on the blood of placental mammals. Because of their blood-feeding habits, sucking lice are much more important as vectors pathogens, especially with respect to human diseases (Figure 2.4.1b).

The geographic distribution of lice is roughly similar to that of the birds and mammals on which they live. However, their distribution within the host population is not uniform. Direct physical contact between hosts is usually the best way for lice to disperse within a host species population. Host animals also pick up new lice by sharing nests and nest materials with other infested animals.

2.4.4 Behaviour

The flattened bodies of lice are perfect for moving in the narrow spaces between feathers and fur. Most louse species remain attached to their host for their entire lives. Their populations vary greatly in size and are strongly influenced by the condition and health of their hosts. For example, birds with damaged bills or feet may have more lice because they are unable to preen or clean themselves efficiently. Some lice escape preening by wedging themselves between feather barbs or by living at the bases of fluffy feathers on the bird's abdomen. They will bite into the feathers with their mouthparts and lock their jaws in place.

Some species go to the extreme of actually living inside the quills of wing feathers to escape preening by their shorebird hosts. The dead, dried bodies of lice are found firmly attached to bird and mammal skins in museum collections, sometimes hundreds of years after the collection and death of their host.

Figure 2.4.4. Chewing louse attached to the abdomen of a hippoboscid fly.

One of the most unusual and rare methods of louse dispersal is by means of phoresy, or hitchhiking. These lice attach themselves to the abdomens of certain flies and hitch a ride to the next host (Figure 2.4.4).

Lice can vary in the number of times they feed each day, for example, head lice feed regularly every few hours, while body lice feed only once or twice per day when the host is resting.

Chewing lice generally feed by chewing the skin, fur or feather on their host. The few hematophagous species typically chew the skin until it bleeds and then imbibe the blood from the wound site.

Sucking lice use three sharp stylets to penetrate the host to initiate blood feeding. Before feeding begins these stylets are withdrawn into a stylet sac inside the head. Externally the labrum is modified into a broad partially flattened tube-like structure termed a haustellum with tiny teeth which latch onto the skin. Once in place, the stylet bundle is pushed through the skin until a host blood capillary is penetrated. A cocktail of enzymes, anticoagulants and other compounds is secreted in the saliva. The blood is sucked up through the haustellum.

Excerpts in this section from Marquardt, W.C. *et al*. 2005. Biology of Disease Vectors, Elsevier Academic Press. 785pp, and http://animals.jrank.org/pages/2416/Chewing-Sucking-Lice-Phthiraptera.html.

2.5. Bed Bugs

Bed bugs (*Cimex* spp.) are insects (True bugs, order Hemiptera) that are wingless and dorsoventrally flattened. Adults are a reddish brown, 5-6 millimetres when unfed to almost 10 millimetres when fully blood engorged (Figure 2.5.1).

Figure 2.5.1. Side view of adult bed bug showing how flat it is even when partially engorged

The two common species are *Cimex hemipterus* (the Tropical bed bug) and *Cimex lectularius* (the Common bed bug).

2.5.1 Life cycle

Bed bugs develop from egg to adult via simple metamorphosis, with the last nymphal stage developing into an adult without passing through a non-feeding pupal stage (Figure 2.5.2).

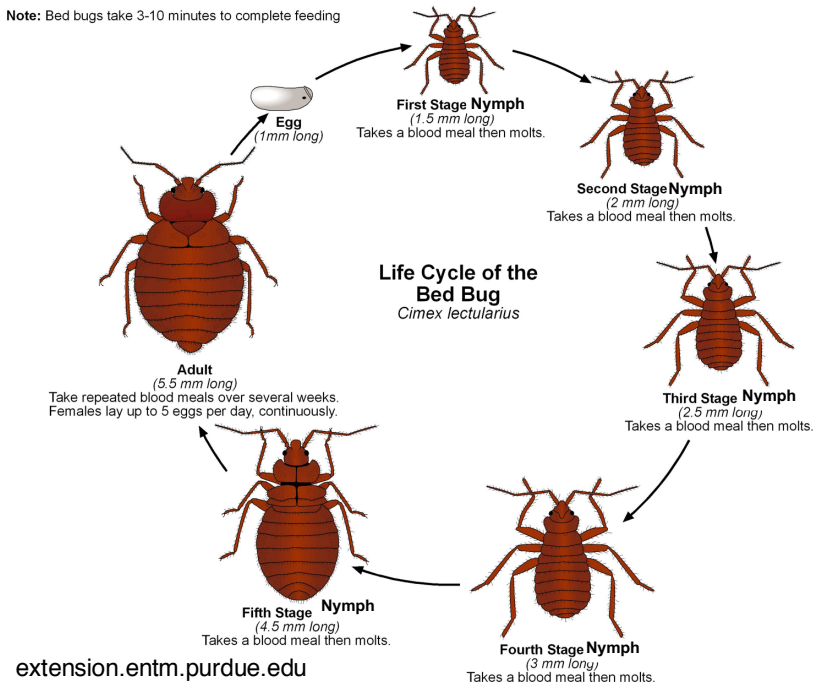


Figure 2.5.2. Bed bug life cycle.

2.5.1.1 Eggs

Eggs are approximately 1 millimetre, cream in colour with a slight bend. They are laid individually, almost anywhere but tend to be around harbourage sites, and laid in crevices in dark areas, preferably onto textured materials (fabrics, wood, behind pictures, in furniture, along edges of baseboards, under floor boards, etc.).

Figure 2.5.3. bed bug egg cases on the fabric of a mattress.

They will be cemented firmly onto the surface and not easily removed. Females sometimes randomly lay single eggs while walking, making detection of all eggs virtually impossible.

2.5.1.2 Nymphs

Bed bugs have 5 numphay stages. The first instar nymph emerges from the egg approximately 7-10 days after it has been laid.

The nymphal stages have a similar body shape to the adults but start out translucent and cream in colour in the first instar, becoming darker in the later instars. The size of the juveniles varies between 1-4 millimetres depending on growth stage (Figure 2.5.4).

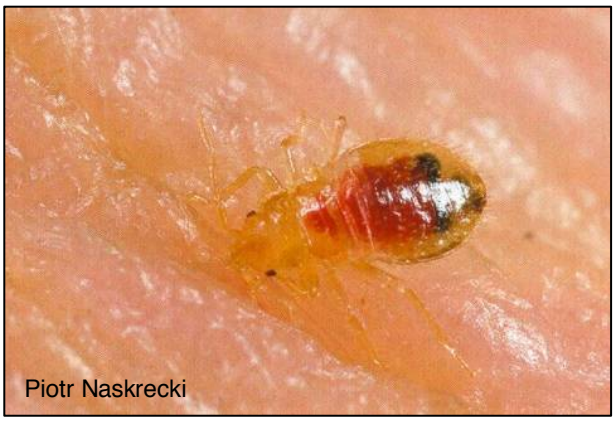


Figure 2.5.4. Bed bug nymph feeding on the arm of a human host.

The bed bug moults into each consecutive life stage by shedding its exoskeleton and requires a blood meal to do so. They can remain dormant for several months without a blood meal but they do not moult without one. Under optimum conditions all 5 nimphal stages can be completed in about a month.

2.5.1.3 Adults

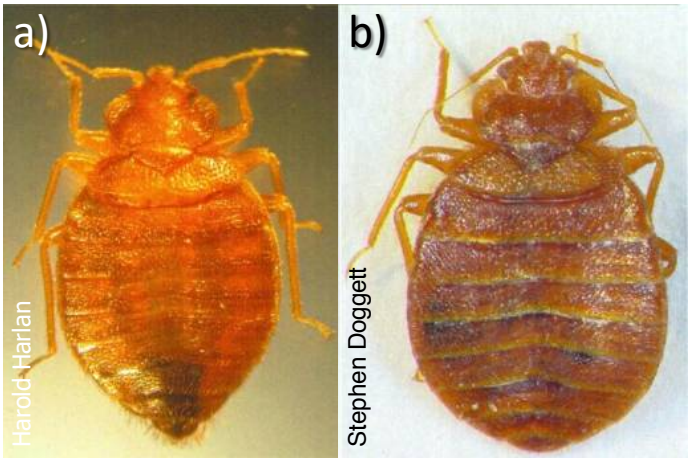


Figure 2.5.6. Adult bed bug, a) male with pointed abdomen and b) female with rounded abdomen

Under favourable conditions the newly emerged female will feed and mate and then start laying eggs 3-6 days later. In perfect conditions 3-6 eggs are laid a day, more commonly 5-7 per week. Females can last 6 months to 2 years, during which time they may lay 200-500 viable eggs.

Both adult male and female bed bugs take repeated blood meals during their lives (Figure 2.5.6). Females require blood for the development of eggs.

2.5.2 Habitats

Bed bug adults and larvae are found in the same environments. They are typically active at night and hide during the daytime. Human dwellings provide ideal habitat (harbourage sites, temperature, humidity) as well as a blood source. As they are very flat they can squeeze into almost any cavity, including mattress seams, beneath loose flooring, behind loose wallpaper, inside box springs, behind pictures and headboards, upholstered furniture, within electrical appliances and behind light switches (Figure 2.5.7a and b). They can be transported from place to place on clothing or in suitcases but do not typically venture too far once they have established in a new suitable habitat.

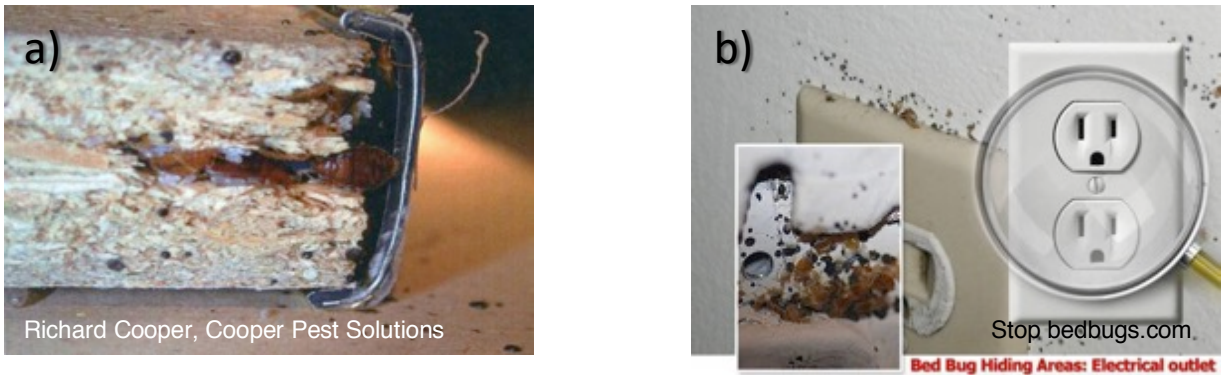


Figure 2.5.7. a) Adult and egg bed bugs packed into a crack in a hotel head board. b) Bed bugs around electrical outlets.

Bed bugs usually live within 2-3 metres of where people sleep. However, they can travel up to 30 metres for a feed at night up walls, across ceilings, through air conditioning ducts, along wiring, behind walls and even out one window and into another.

Bed bugs are often associated with dirty conditions, but can live in very clean new homes, as there are still plenty of harbourage sites and hosts for feeds. However, in very cluttered homes obviously more habitat is provided such as behind peeling wallpaper, cracks around doors, windows, floorboards etc. where they can shelter from insecticides.

Humans are the preferred host for a blood meal (Figure 2.5.8). Bed bugs don't tend to live on people like lice. Generally, the only real contact is during feeding, which may take 5-10 minutes. In the absence of humans, bed bugs will feed on other warm blooded animals including dogs, cats, birds and rodents.

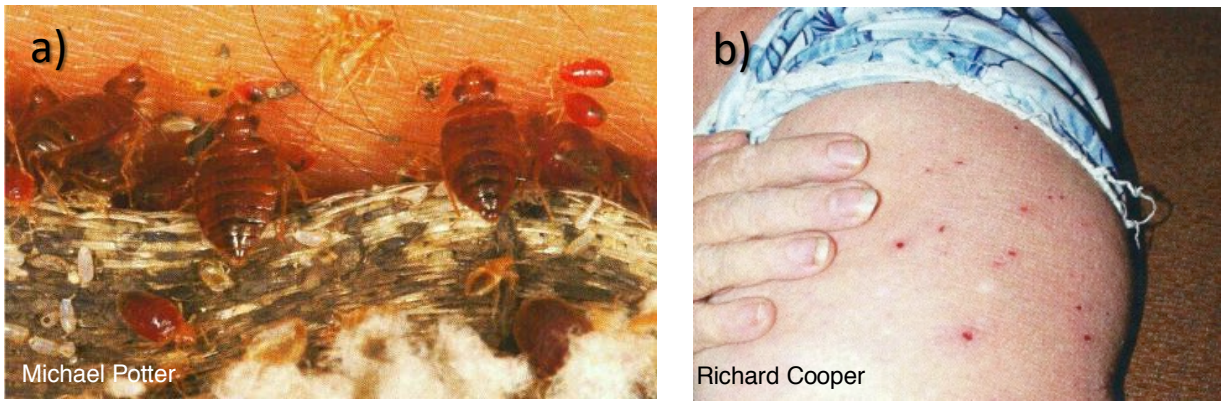


Figure 2.5.8. a) Bued bugs linded up while feding on a human host. Note the precence of adults, nymphs and eggs. b) Bed bug bites on a woman's arm.

2.5.4 Behaviour

Bed bugs are very resilient. Nymphs and adults can persist months without feeding. The ability to survive without a blood meal is longer at cooler temperatures - potentially a year or longer at 12°C or less. In temperature-controlled buildings, a more typical duration is about 2-6 months. When infested dwellings such as apartments are vacated, bed bugs often disperse to nearby units, or reduce their activity until the unit is reoccupied.

Bed bugs are active mainly at night. During the daytime, they prefer to hide close to where people sleep. Their flattened bodies enable them to fit into tiny crevices - especially those associated with mattresses, box springs, bed frames and headboards. Bed bugs do not have nests like ants or bees, but do tend to congregate in habitual hiding places. Characteristically, these areas are marked by dark spotting and staining, which is the dried excrement of the bugs. Also present will be hatched and un-hatched eggs, the tannish shed skins of maturing nymphs, and the bugs themselves (Figure 2.5.8a). Another possible sign are rusty or reddish smears on bed sheets or mattresses from crushed engorged bed bugs. Although it's often stated that bed bugs have a tell-tale "buggy" odour, the smell is seldom evident except in extreme infestations and should not be relied upon for detection.

2.6. Cockroaches

2.6.1 Life cycle

Cockroaches are hemimetabolous so undergo incomplete metamorphosis whereby the egg hatches out into a nymph, which is already similar in appearance to the final adult stage. This nymph undergoes a number of moults before finally developing into the fully reproductive adult stage. Adult females lay clusters of eggs in a case called an ootheca, which may be dropped or attached to a surface. It can take anything between just a few weeks to over a year for a cockroach to complete its growth cycle, depending on species and environmental conditions.

2.6.1.1 Eggs

Figure 2.6.1. Cockroaches eggs and nymphs emerging from an ootheca.

Eggs of cockroaches are laid in a bean or purse shaped casing called an ootheca. The cases are formed in a special chamber of the abdomen behind the egg pore which can be closed off by flaps. Glands lining this chamber secrete a white fluid that coats the egg. This gradually hardens and as the eggs are laid the flaps are relaxed and the egg can protrude from the abdomen. This process is repeated several times and eventually a ridged casing containing the eggs can be seen attached to the abdomen of the female cockroach. The casing makes the eggs water and pesticide resistant.

Once the case of eggs is completed, the female may either carry the eggs around until they hatch, shortly before they hatch or deposit them in a suitable location to develop and hatch.

2.6.1.2 Nymphs

Figure 2.6.2. Cockroraches nymphs leaving the ootheca.

Nymphs hatch out from their eggs resembling small adults, though they have undeveloped wings and may also be a different colour. Nymphs undergo their first moult at the same time as they hatch out from the egg case, and are able to move about and feed upon hatching (Figure 2.6.2). Nymphs develop quite slowly and in successive stages called instars. Each stage is completed with a moulting of their exoskeleton which enables them to increase in size, as well as revealing newly developed structures.

As the nymphs develop they undergo several moults at the end of each development stage and gradually develop wings, increase number of joints in things such as antennae, and increase in size. The exact number of moults undergone before adulthood depends on the species of cockroach.

2.6.1.3 Adults

Once the cockroach has reached its final adult form it will not moult again. Cockroaches do not have a pupal form as they undergo incomplete metamorphosis, developing from a small wingless nymph to the winged adult.

Figure 2.6.3. Adult male German cockroach, *Blattella germanica*.

Cockroach adults may survive without food for an extended period, in some cases up to a month, but cannot survive without moisture for more than a few days as they will desiccate.

Male and female cockroaches may be determined by comparing the number of appendages at the tip of their abdomen. Male cockroaches have two pairs of sensory appendages at the tip of their abdomen whereas females only have one. Males have pairs of both styli and cerci, while the females have only a pair of cerci.

Females may lay many hundreds of eggs in a lifetime. Some females mate once and are able to continually reproduce after this one insemination from a male. Males mate with females by attaching a spermatophore to her abdomen.

2.6.2 Habitats

The presence of cockroaches indicates inadequate sanitary practices or ineffective cockroach control measures.

Figure 2.6.4. Ideal hiding place for cockroaches.

Moist, damp, dark and narrow spaces are favoured by cockroach nymphs and adults alike. They can, and prefer, to hide in very small gaps. During the day both the adults and nymphs shelter inside walls, cluster together at backs of refrigerators, ovens, dishwashers, plumbing, inside crevices, in cupboards and behind mouldings and other fittings. Ideal areas include bathrooms and food preparation areas. The greater a site provides for the insect to conceal itself, the more ideal it becomes as a harbourage for cockroaches.

Cockroaches also need a fairly warm temperature and moisture. If the environment is too dry, then they will quickly dehydrate. However, cockroaches are in general notoriously hardy and many species can withstand higher or near freezing temperatures for a short period of time. There are over 4000 species of cockroach in the world, inhabiting a vast array of climates, but their basic needs are the same.

The German cockroach is one of the most commonly encountered pests aboard ships (Figure 2.6.3). The way that ships are constructed include numerous gaps, partitions, fittings and abundant moisture, food and warmth making an ideal environment for their survival and reproduction.

2.6.3 Hosts

Cockroaches are a serious sanitary concern for humans but may also play a role in transmission of some worms and diseases to other animals when they are ingested.

Although cockroaches can bite, diseases are almost exclusively passed on through mechanical transmission whereby their bodies are contaminated with bacteria which is then passed on to other surfaces they encounter as they move about.

Only a small number of the thousands of identified species play a significant role in transmission of disease to humans because they are well adapted to life inside buildings.

2.6.4 Behaviour

Pest species of cockroach live in close association with humans and are well adapted for life in buildings and constructed environments. They are active at night and during the day they will hide in cracks, crevices and narrow spaces such as behind fridges, or behind and underneath cupboards.

Their habits and body structure enable them to potentially transmit pathogens that cause dysentery and diarrhoea. Because cockroaches are omnivorous they will readily eat and move between food sources such as faecal matter and fresh food intended for immediate human consumption, and in doing so enable humans to become exposed to potentially dangerous pathogens through contaminated surfaces and food products. They also do not feed exclusively on one food source but will scavenge for a variety of foods.

Cockroaches impart a foul odour where infestations are well established. Glands on their bodies discharge a malodorous pheromone which signals safe harbourages to other cockroaches. Cockroaches tend to aggregate because of this.

Some species of adult cockroach, such as the German cockroach, are known to be able to bite humans but this event is rare. Diseases associated with cockroaches are linked with their feeding preferences and movement, rather than by an infective bite.

Cockroaches may disperse to new habitats by crawling or flying, though very often in the case of pest species they are transported around in food sources, in vehicles including ships, and in parts, appliances or fittings they have been sheltering in. They can survive months without food, and some species can survive up to 4 weeks without water. This can make infestations and the prevention of new infestations hard to control as they are a very hardy group of insects.

2.7. Ticks

Ticks are external parasites (ectoparasites) that feed off the blood of mammals, birds, reptiles and amphibians. They belong to the Class Arachnida, which also includes spiders, scorpions and mites, they have eight legs (the exception being the larval stage which has six) and are not insects.

Figure 2.7.1. *Ixodes scapularis* a hard tick.

A number of tick species are vectors of human and animal diseases, as they can carry and transmit a range of viruses, as well as haemoparasitic protozoans and bacteria.

There are 2 main groups of ticks: the ixodids or hard ticks (Ixodidae), and the argasids or soft ticks (Argasidae). The hard ticks have a hard dorsal shield (scutum). They are most commonly seen as they remain attached to their host for long periods of time. The soft ticks lack a hard scutum and feed only for a short time and are therefore seldom seen.

Globally ca. 825 tick species are currently described, with 10 native species occurring in New Zealand. Most of them are bird (mainly sea bird) parasites (*Carios carpensis, Ixodes kergulensis, I. amersoni, I. anatis* (kiwi), *I. eudyptidis, I. jacksoni, I. uriae*) but there is also the endemic tuatara tick (*Amblyomma sphenodonti*), as well as an undescribed bat tick, and *I. auritulus zealandicus* can also infest sea lions. Only one species, the cattle tick (*Haemaphysalis longicornis*) is introduced and established. It has a wide range of mammal hosts, including humans.

2.7.1 Life cycle

Figure 2.7.2. The four tick life stages.

Both hard ticks and soft ticks have 4 stages in their life cycle: egg, larva, nymph and adults (Figure 2.7.2). Individuals transition to each new life stage by moulting, following a blood feed.

In general, the life cycle of a hard tick is as follows: a newly hatched (six-legged) larva feeds on a host, drops off to the ground and moults to a nymph (Figure 2.7.3). A nymph seeks out and feeds on a second host, drops off to the ground and moults to an adult. Hard ticks have only one nymphal stage, unlike soft ticks.

Male and female adults seek out a third host, feed, mate and drop off to the ground. However, the number of host species utilised varies – there are also one-host ticks (e.g. *Rhipicephalus microplus*, Figure 2.7.4a) and two-host ticks (e.g. *Rhipicephalus evertsi*, Figure 2.7.4b).

Males die soon thereafter, while females eventually lay eggs on the soil for several days to a few weeks. Depending on the species, a single female may lay 3,000 - 8,000 eggs and then dies without reattaching to a host again.

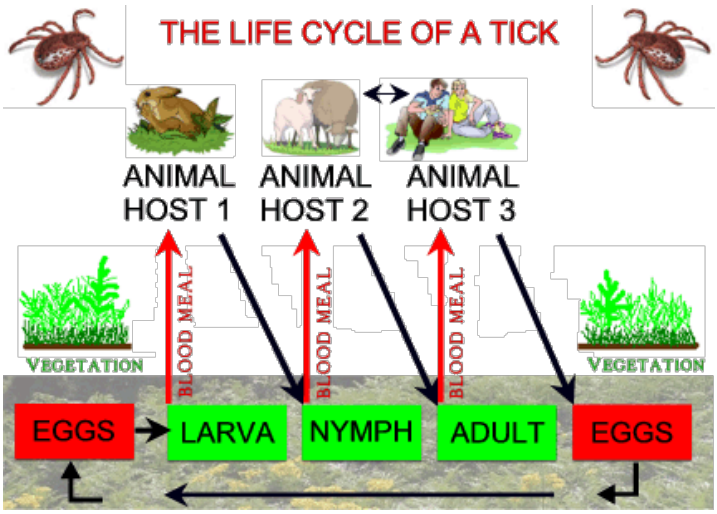


Figure 2.7.3. Hard tick life cycle.

The duration of the life cycles differs depending on several factors such as the type of hosts on which they feed, or which developmental stage survives winter. For example, depending on the species, hard ticks may spend winter either as larvae, nymphs or female adults. A few species may have 2 or 3 developmental stages that over-winter. Some species complete a life cycle in as few as 90 days, others take a year, and a few require 2 years to complete a life cycle.

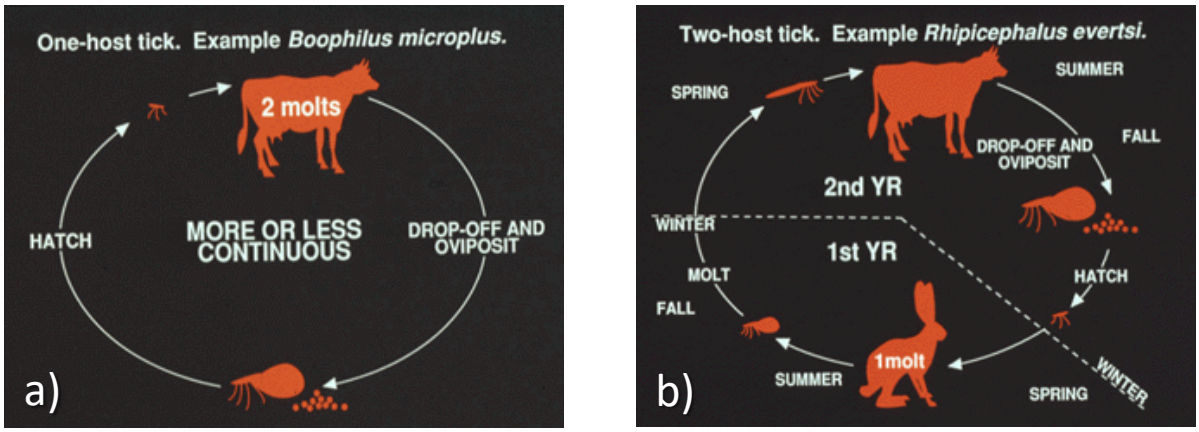


Figure 2.7.4. a) a one-host tick life-cycle. b) a two-host tickelife-cycle

2.7.2 Habitats

Hard ticks are found in habitats that support large numbers of vertebrate hosts, such as mammals, ground-dwelling birds and lizards. Some of the most productive habitats are moist woodlands and areas of vegetation around the edge of forests, along forest trails and in grassy fields. Additional habitats include areas surrounding power line routes made through forests, in and around campgrounds, and in abandoned grassy yards in urban areas.

They use vegetation for host seeking and are often found in tall grass and shrubby areas. They crawl up the vegetation and sit on the ends of leaves or blades of grass to seek out their host.

Hard ticks are more sensitive to desiccation than soft ticks and are usually found in environments which are protected from high temperatures, low humidity and constant winds.

2.7.3 Hosts

Hard ticks are less host specific than soft ticks and will feed on a variety of hosts, including mammals, reptiles and birds. They usually come into contact with people through pasture animals like cattle, deer or horses, or on smaller animals like dogs, cats and rodents who have “collected” questing ticks.
There are also some species commonly found on reptiles including the New Zealand species *Aponomma sphenodonti*, a parasite of the tuatara.

The type of hosts utilised varies between life stages of hard ticks. The most common hosts of larvae are small mammals, especially mice, and ground-dwelling birds, but larvae of some species feed on humans. Nymphs tend to feed on small to medium-sized mammals, but nymphs of a few species are attracted to larger hosts, including pets and humans (Figure 2.7.3). Adult ticks tend to feed on larger mammalian hosts such as deer, livestock, dogs, and humans.

2.7.4 Behaviour

Host Seeking

Figure 2.7.5. Questing tick.

Being flightless, hard ticks "wait" for passing vertebrate hosts (Figure 2.7.5). Larvae, nymphs and adults detect carbon dioxide, host odours, vibrations and warm, moist air currents. Tick larvae usually remain on the ground where they encounter potential hosts. Nymphs remain on the ground or climb grassy vegetation from which they are able to grasp a passing host.

Adult ticks may remain on the ground, but more commonly climb up vegetation, from which they grasp a passing host. They often can be seen at the tops of grass blades or on lower leaves of bushes engaging in "questing". Questing is a behaviour in which the tick reaches upward with waving front legs ahead of an approaching host (Figure 2.7.5).

Feeding

Hard ticks typically take one blood meal in each of the 3 developmental stages - larval, nymphal and adult. Both sexes are blood feeders but only the female becomes greatly distended during engorgement (Figure 2.7.6c). Most species feed on a different host during each stage but there are some one-host and two-host species as described in the life cycle section above (Figure 2.7.4a and b).

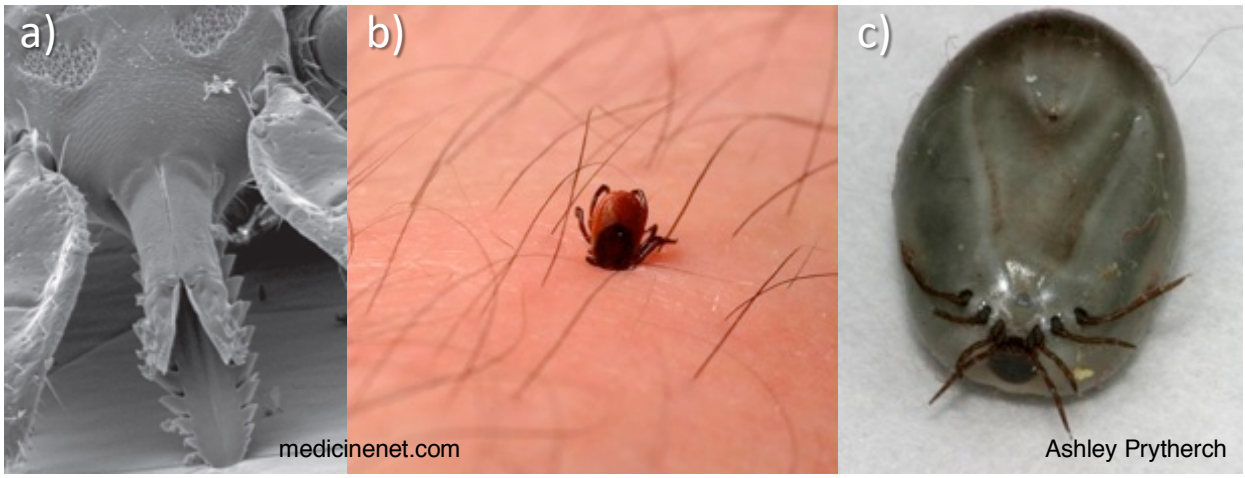


Figure 2.7.6. a) tick mouthparts, b) feeding embedded in skin and c) female engorged *Ixodes scapularis*..

The act of blood feeding by a hard tick results in a "feeding wound". As it begins to feed, a tick secretes saliva containing compounds that increase blood flow, prevent clotting and suppress the host's immune response. Ticks imbibe the blood that pools in the wound. At the same time, they

regurgitate excess water that has been extracted from the blood meal into the wound. This process increases the possibility for the transmission of pathogens from a tick to its animal host.

Transmission of a pathogen typically does not occur until an infected tick has attached and fed for at least 24 hours, and transmission of some pathogens does not begin until an infected tick has fed for 48 hours or more (Figure 2.7.6.b).

Hard ticks commonly feed on their host for long periods of time, sometimes as long as several weeks, with feeding time depending on life stage, species of tick and the type of host. Larvae of hard ticks usually complete a blood meal within a day or two and engorge very little. Nymphs attach to a host and complete a blood meal within a few days. They engorge enough so that red blood can be seen through their body wall. Male adults feed much like nymphs, but do so repeatedly on one host animal. They are only intermittent feeders and do not engorge as the large scutum greatly limits the amount that can be consumed at one time.

Females will not undergo feeding and engorgement until they have mated with a male tick. They attach to a host feed to completion in a week or so. Engorgement is dramatic over the course of the last day or two of feeding, resulting in a huge increase in body size. Engorgement of female adults is facilitated by the lack of a large scutum and the possession of an expandable body wall. Fully engorged females of certain species may be over half an inch long and a quarter inch wide. The hard tick's cuticle will grow as the tick feeds, to allow for the increasing amount of blood consumed (Figure 2.7.6).

2.7.5 Soft ticks

Life Cycle

Figure 2.7.8. *Otobius megnini* nymph

In soft ticks, the life stages are not so easily distinguished. There is some variation in the number of nymphal moults (up to 7) where hard ticks consistently have only one nymphal stage.

In general, the life cycle of a soft tick is as follows: eggs are laid in several batches of hundreds. Once hatched the larvae require a blood meal from a host and moult to the first of their nymphal stages. For most soft tick species there are multiple nymphal stages. They gradually increase in size with each moult until they reach the adult stage.

Soft ticks will feed several times during each life stage and adult females will lay several small batches of eggs between each blood meal. They usually have a longer life cycle than hard ticks, lasting through several years. Some soft tick species are also able to survive for long periods of time without feeding.

Some species such as *Otobius megnini* (spinose ear tick) have adapted to a different life cycle. This species lives and feeds inside the ears of the host (naturally deer, mountain sheep and pronghorn antelope but now also cattle, domestic animals and humans) and employs a single host life cycle. The adult ticks do not feed and mating occurs on the ground.

Habitat

The majority of soft tick species are nest parasites, which emerge only briefly to feed at night and hide during the day in or near the nest of the host. They are often found in burrows, caves, nesting

material and nearby rock crevices. They have also been known to sometimes inhabit dilapidated dwellings and animal rearing shelters.

Many soft ticks thrive in hot and dry conditions.

Hosts

Soft ticks have high host and microhabitat specificity. They are most commonly associated with birds, particularly sea birds such as penguins. Some species are also parasitic on mammals, humans and occasionally bats.

Behaviour

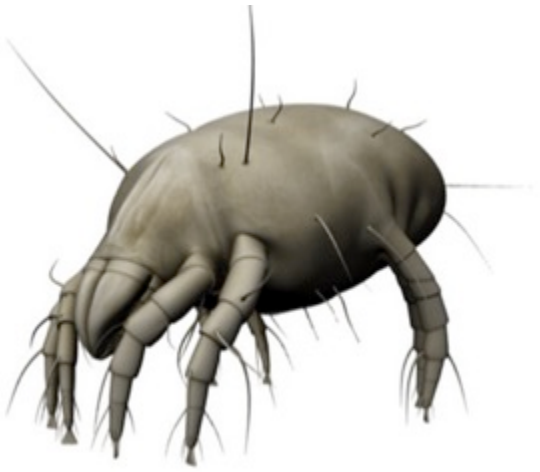
Figure 2.7.9. Soft tick.

Some soft ticks also quest for their host by climbing low lying vegetation. The majority of soft ticks however are nest parasites which inhabit sheltered environments such as nests, caves or burrows. They remain hidden amongst the nesting material of their host while they are away and will rapidly move to feed on the host once it returns to the nest. Therefore, they do not need to actively seek hosts.

Soft ticks usually feed rapidly and for only a short amount of time on their host, ranging from just several minutes to a few days. The cuticle of soft ticks will expand as they feed but it doesn't grow as in hard ticks, so they can't consume nearly as much blood at one time. Adults generally feed to repletion in minutes to hours, while larvae and nymphs feed for more extended periods. Soft ticks may expand to anywhere from 5-10 times their unfed body weight (Figure 2.7.9).

2.8. Mites

Mites are parasites which feed off plants and animals, including insects. There are over 45,000 described species of mites. It is believed that we have only found 5% of the total diversity of mites. Mites are believed to have existed for around 400 million years.



turbosquid.co

Figure 2.8.1. Body shape of a mite.

Mites belong to the Class Arachnida (eight legs), order Acarina (also known as Acari). Members of this order differ from other arachnids in that the body is not segmented and the cephalothorax and abdomen are combined into one body region.

Mites are among the most diverse and successful of all the invertebrate groups. They have exploited an incredible array of habitats, and because of their small size (most are microscopic, some can't be seen without a microscope) most go totally unnoticed (Figure 2.8.1).

A tropical species, *Archegozetes longisetosus,* is one of the strongest animals in the world, relative to its mass (100 µg): it lifts up to 1182 times its own weight, over 5 times more than would be expected of such a minute animal.

Figure 2.8.2. Spider mite (*Tetranychus urticae*) with two silk threads.

Some mite species are vectors of a number of human and animal diseases, while others are pests of plants (Figure 2.8.2). Some of the plant pests include the so called "spider mites" (family Tetranychidae), thread-footed mites (family Tarsonemidae), and the gall mites (family Eriophyidae), these will not be discussed further.

Some species that attack animals are obligate parasites, and have to eat the tissues of living animals to survive. Mites can be ectoparasites (feeding on the outer skin) or endoparasites (feeding on the underlying tissues). The ectoparasites live on the host's body surface, while the endoparasites dig tunnels under the host's skin in which they live and reproduce (Figure 2.8.3).

While some parasitic mites transmit disease organisms, many cause diseases themselves.

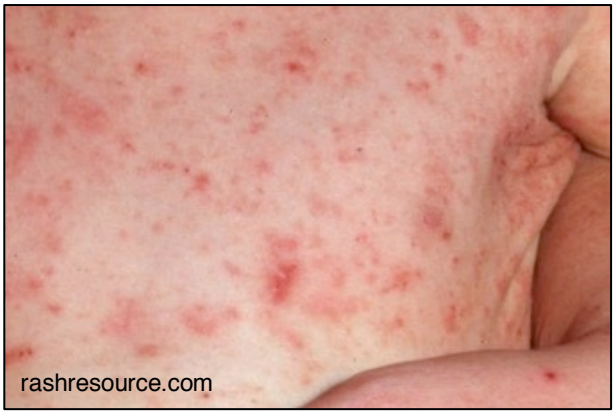


Figure 2.8.3. Rash produces by Sarcoptes scabie

These include scabies and mange-contagious skin diseases characterized by inflammation, irritation and intense itching. Members of the Sarcoptic Mange mites (family Sarcoptidae) burrow under the skin (Figure 2.8.3), and the *Demodex* mites (family Demodicidae) are parasites that live in or near the hair follicles of mammals including humans (Figure 2.8.4).

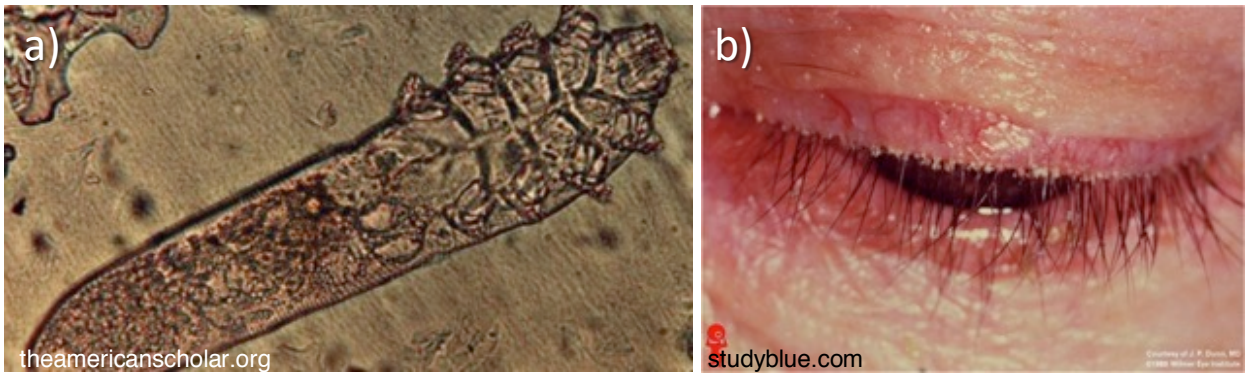


Figure 2.8.4. a) *Demodex* sp. mite, b) Demodex mites living in the hair folicies.

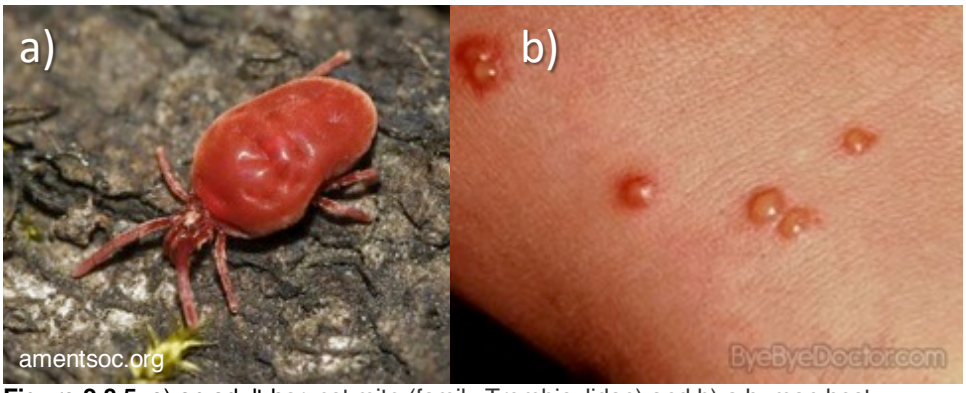


Figure 2.8.5. a) an adult harvest mite (family Trombiculidae) and b) a human host reaction after a chigger feed on thir skin cells

Many more mites are free living, that is, not parasitic. Some, such as the chigger, are parasitic as larvae but free-living in the nymph and adult stages (Figure 2.8.5).

Figure 2.8.6. a dust mite.

Perhaps the best-known is the house dust mite (family Pyroglyphidae). This mite is well known for causing allergic reactions in humans (Figure 2.8.6). The allergy is a hypersensitive reaction to proteins in the excretion of dust mites. The protein attacks the respiratory passages causing hay fever and asthma. It will also aggravate atopic dermatitis in people who have a tendency to this problem. Obviously these reactions are not as a result of a disease and will not be covered further.

Figure 2.8.7. Varroa destructor attached to a honeybee.

Insects also have parasitic mites, examples include *Varroa destructor* which attaches to the body of the honeybee (Figure 2.8.7), and *Acarapis woodi* (family Tarsonemidae) which lives in the tracheae of honey bees. There are hundreds of species of mites associated with other bee species and most are poorly described and understood. Some are thought to be parasites, while others beneficial symbionts. These types of mites will not be discussed further.

2.8.1 Life cycle

Reproduction in mites is highly variable. They have variable numbers of different developmental stages; one or more stages may be absent in some groups.

For the purposes of this manual, a 4 stage life cycle will be described. The first is the egg, which hatches into stage 2, the 6-legged larvae (sometimes called a chigger). In stage 3, they moult into the 8-legged nymph, and then again into the final stage, the adult (Figure 2.8.8). Each stage varies considerably throughout the mites, even between closely related groups. Due to their diverse appearance a general description is difficult to provide. Information of each stage is very sparse for some groups and therefore only a small amount of detail has been included for each stage.

Mites may take only a week to complete their life cycle, while some like the harvest mites only have one generation per year.

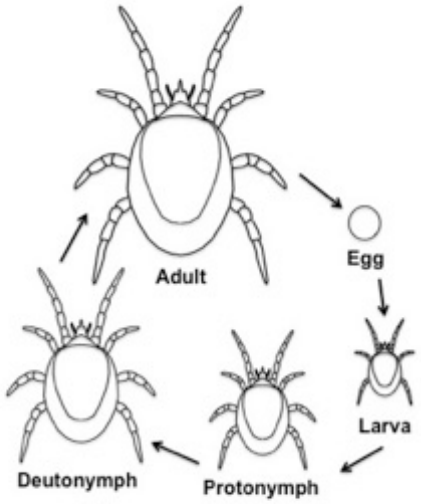
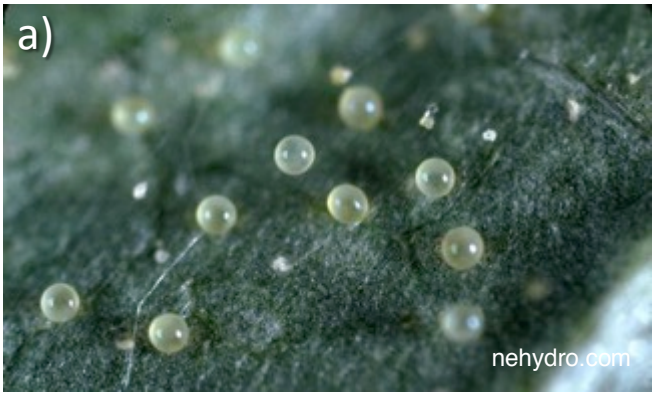


Figure 2.8.8. A four stage mite life cycle.

2.8.1.1 Eggs

Mite eggs are generally spherical-oval (Figure 2.8.9). Some are laid singly, while others in large batches. Most mites are oviparous, i.e. they lay eggs in which the embryos are at an early stage of development. Some mites develop their eggs within the female and live larvae are born (termed ovoviviparity).



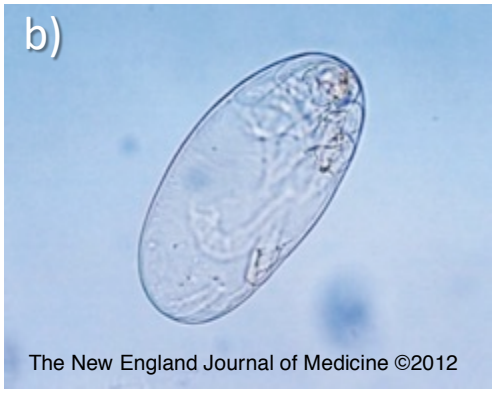


Figure 2.8.9. a) spider mite eggs on a plant, b) dust mite egg.

Mite eggs occur in a wide range of habitats, for example the adult females of harvest mites lay their eggs on the ground, while scabies and demodex mites burrow into the skin of humans to lay their eggs.

2.8.1.2 Larvae

Larval mites have 3 pairs of legs (Figure 2.8.10a) , whereas nymph and adult mites have 4 pairs. They are usually too small for a person to see, although you may be able to see groups of them.

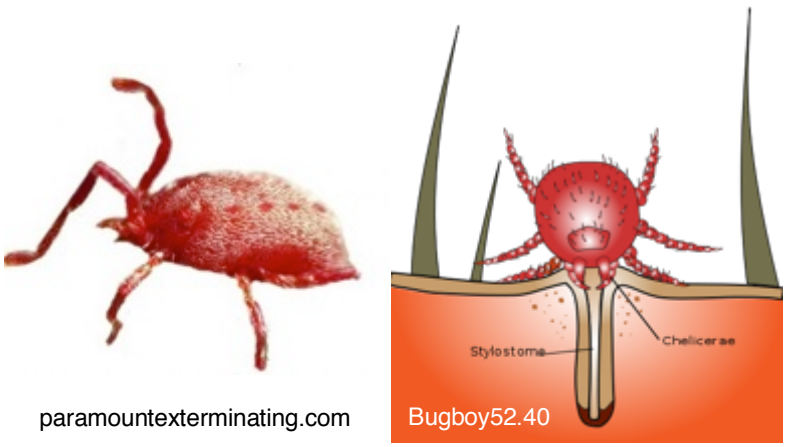


Figure 2.8.10. a) a Chigger, b) Chigger feeding.

A commonly used name for some mite larvae, e.g. harvest mites, is “chigger”. Chigger larvae do not burrow into the skin but inject a salivary fluid which produces a hardened, raised area around them (Figure 2.8.5b). Body fluids from the host are withdrawn through a feeding tube. Larvae feed for about 4 days and then drop off and moult to non-parasitic nymphs (Figure 2.8.10b).

Sarcoptes scabiei larvae spend their entire life on the host in a burrow beneath the skin. They consume dead skin cells, skin secretions, fungal spores and bacteria.

2.8.1.3 Nymphs

Mite nymphs can vary significantly in shape. Sometimes the nymphal stage is free-living in parasitic species, while in others like *Sarcoptes scabiei* nymphs moult into the adult while still in the burrow.

2.8.1.4 Adults

Some mites, including harvest mites, overwinter as adults. Females lay eggs on the ground in groups of several hundred, and the resulting clumps of larval mites that hatch from these eggs can result in severe infestations of their hosts. Sometimes, like the nymph, the adult stage is free-living even in some parasitic species.

2.8.2 Habitats

Mites exist in almost all habitat types including terrestrial, freshwater and marine environments, and ranging from deserts to rain forests, mountain tops to tundra, and saltwater ocean floors to freshwater lakes.

Terrestrial mites are commonly found in soil or leaf litter, under the bark of trees or feeding on the leaves and stems of plants. Many live freely in the soil or water, but there are also a large number of species that live as parasites on plants, animals and invertebrates, and even some that feed on mould.

2.8.3 Hosts

Most species of mites are predatory and will feed on a variety of small invertebrates, while others are herbivorous, often feeding on plant sap and sometimes causing damage to agricultural crops and garden plants.

Mites are generally host specific, meaning they will usually attack only a certain species of host, but will sometimes cross over from one species to another, particularly if their preferred choice of host is not available.

2.8.4 Behaviour

Parasitic mites normally live on the host or in their nests, burrows, building etc. When the animal dies or abandons the nest/burrow/building mites migrate to other areas of the structure in search of new hosts. For example, rodent mites often become a nuisance after an infestation of mice or rats has been eliminated and people become aware of the problem as they are being attacked by the mites searching for an alternate food source.

Many mites have needle-like piercing-sucking mouthparts, for example spider mites feed by penetrating the plant tissue with their mouthparts and are found primarily on the underside of the leaf. They spin fine strands of webbing on the host plant - hence their name.

Harvest mite larvae or chiggers have several instinctive behaviours that help them find food:

Light sensitivity: Chiggers move toward shady areas. This protects them from the sun, which can dry out their bodies. Also, when a potential host casts a shadow in a chigger-infested area, chiggers can flock toward it.
Temperature sensitivity: Once a chigger comes into contact with its host's skin it detects the host's body heat.
Touch sensitivity: Tiny, hair-like sensory organs cover a chigger's body. These organs help chiggers find hosts and find a good place to feed. They also help chiggers find each other and form clusters.
Upward mobility: Chiggers like to climb. They will climb into vegetation and wait for hosts to brush past.
Questing response: Like ticks, chiggers use a posture called questing to find food. In the right conditions chiggers will stand with their front legs outstretched so they can grasp potential hosts. Shadows and vibrations tend to provoke the questing response.

These behaviours all help chiggers find hosts, but finding a host is only half the problem. Chiggers also have to find places to feed where they won't be brushed or groomed away. On top of that, the skin in the feeding area has to be thin because a chigger's microscopic mouthparts can't get through tough, leathery skin. Chiggers often attach themselves to pores or hair follicles or any other natural depression in the skin.

2.9. Rats

Provisions for management of the risk posed by rats at borders are variously contained in the IHR 2005. References to rats are as follows:

INTERNATIONAL HEALTH REGULATIONS (2005)
PART I – DEFINITIONS, PURPOSE AND SCOPE,
PRINCIPLES AND RESPONSIBLE AUTHORITIES

Article 1 Definitions:

> "deratting" means the procedure whereby health measures are taken to control or kill rodent vectors of human disease present in baggage, cargo, containers, conveyances, facilities, goods and postal parcels at the point of entry;

Article 22 Role of competent authorities

> (c) be responsible for the supervision of any deratting, disinfection, disinsection or decontamination of baggage, cargo, containers, conveyances, goods, postal parcels and human remains or sanitary measures for persons, as appropriate under these Regulations;

1. Disinsection, deratting, disinfection, decontamination and other sanitary procedures shall be carried out so as to avoid injury and as far as possible discomfort to persons, or damage to the environment in a way which impacts on public health, or damage to baggage, cargo, containers, conveyances, goods and postal parcels.

Article 28 Ships and aircraft at points of entry

2. Subject to Article 43 or as provided in applicable international agreements, ships or aircraft shall not be refused free pratique by States Parties for public health reasons; in particular they shall not be prevented from embarking or disembarking, discharging or loading cargo or stores, or taking on fuel, water, food and supplies. States Parties may subject the granting of free pratique to inspection and, if a source of infection or contamination is found on board, the carrying out of necessary disinfection, decontamination, disinsection or deratting, or other measures necessary to prevent the spread of the infection or contamination.

ANNEX 5
SPECIFIC MEASURES FOR VECTOR-BORNE DISEASES

S3. States Parties should accept disinsecting, deratting and other control measures for conveyances applied by other States if methods and materials advised by the Organization have been applied.

2.9.1 Background to Rodents

Rodents are a large group of mammals (order: Rodentia) with about 2,277 species described worldwide. Latin *Rodare* (to gnaw) + *dentis* (tooth) = Rodent. > 40% of mammalian species are rodents. Including: rats, mice, guinea pig, hamster, squirrel, vole, lemming, capybara. Does not include rabbits and hares.

Commensal Rodents:
Commensal = "To eat from the same table" – rodents. Species that share a very close association with humans. The 3 most universal commensal rodent species are:

Rattus norvegicus (Norway Rat, Grey Rat, Brown Rat, Burrowing Rat, Sewer Rat)

Rattus rattus (Ship Rat, Black Rat, Climbing Rat, Roof Rat, Alexandrian Rat)
Mus musculus (common house mouse, domestic mouse)
In New Zealand (and Pacific Islands) also include the exotic *Rattus exulans* (Kiore, Pacific Rat, Native Rat, Figures 2.9.1.and 2.9.2, Table 2.9.1)

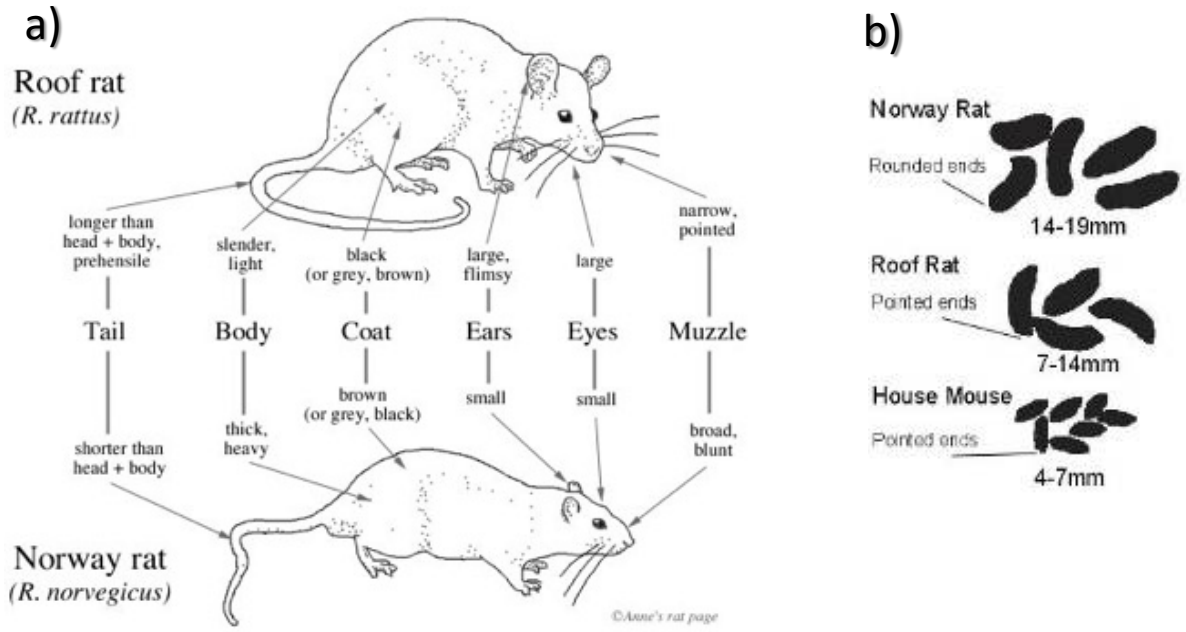


Figure 2.9.1.a) Roof rat and Norway rat description and comparison, b) Norway rat, Roof rat and House mouse droppings.

2.9.3 Behaviour

2.9.3.1 Rattus rattus

Food preferences: omnivorous – seeds, fruit, vegetables, eggs, grain, predatory on small bird chicks, scavenge on dead animals. In cold environments seeks out high fat diets.

- Excellent climbers, often in high places.
- Does not burrow.
- Excellent swimmers.
- Tend to nest in elevated positions.
- Nocturnal with most activity and feeding soon before sunset and sunrise.
- Strong social hierarchy.
- Neophobic.

Figure 2.9.2. Norway rat, roof rat, Kiore and house mouse.

2.9.3.2 Rattus norvegicus

Food preferences: omnivorous – meats, fish, flour, grains, fruits, vegetables, predatory on small animals, scavenge on dead animals. In cold environments seeks out high fat diets. Has been known to survive in cold-rooms eating frozen fat off butchered carcases.

- Excellent swimmers and climbers.
- In nature dig and nest in extensive burrows; in engineered environments – tend to nest low down.

- Nocturnal with most activity and feeding soon before sunset and sunrise.
- Strong social hierarchy.
- Neophobic.

Table 2.9.1. Comparison between Kiore, Roof Rat, Norway Rat and House Mouse.

	Kiore (*Rattus exulans*)	**Roof Rat (*Rattus rattus*)**	**Norway Rat (*Rattus norvegicus*)**	**House Mouse (*Mus musculus*)**
Other common Names	Maori rat, Polynesian rat, Native rat, Pacific rat	Black rat, Blue rat, Bush rat, House rat, Ship rat, Matapo	Brown rat, Water rat, Sewer rat, Pouhawaiki	Field mouse
Adult weight (g)	Typically 60-80; (up to 180)	Typically 120-160; (up to 225)	Typically 200-300; (up to 450)	Typically 15-20; (up to 30)
Max. head-and-body length ("HBL") (mm)	180	225	250	115
Tail	Slightly shorter or longer than HBL Thin and uniformly dark all over	Much longer than HBL Uniformly dark all over	Clearly shorter than HBL Thick, with pale underside	Slightly shorter or longer than HBL; Uniformly grey-brown
Ears	15.5-20.5 mm Cover eyes when pulled forward Fine hairs do not extend beyond edge of ear	19.0-26.0 mm; Cover eyes when pulled forward; Fine hairs do not extend beyond edge of ear.	14.0-22.0 mm; Do not cover eyes when pulled forward: Obvious hairs extend beyond edge of ear.	12.0-15.0 mm
Adult hind-foot length	24.5-31.0 mm	28.0-38.0 mm	30.0-41.5 mm	15.0-21.0 mm
Colour of upper-side of hind-foot **Fur on back**	Outer edge dark near ankle Rest of foot and toes pale Brown	Uniform colouring over whole foot, usually dark. Three colour variations: rattus: uniformly black; alexandrinus: brown with long black guard hairs frugivorous: brown with long black guard hairs.	Always completely pale. Uniformly grey. Brown	Brown Dull grey-brown
Fur on belly	White-tipped grey giving irregular colour	Three colour variations: rattus: uniformly grey; alexandrinus: uniformly grey; frugivorous: uniformly white or creamy-white;	White-tipped grey giving irregular colour.	Uniformly grey.
Length of droppings	6.4-9.0 mm	6.8-13.8 mm	13.4-19.1 mm	3.9-7.6 mm
Number of nipples	8	10-12 (usually 10)	12	10 to 12
Habits	Agile climber Digs small holes Nests mainly on the ground Feeds both on the ground and in trees Infrequent swimmer	Very agile and frequent climber Rarely burrows Nests mainly in trees and shrubs Infrequent swimmer	Burrows extensively; Climbs much less frequently than the other rats Strong swimmer Nests underground Very wary	Mainly ground dwelling, though capable climber Nests in small holes

3. Vector Borne Diseases

3.1. Mosquitoes

Although not all mosquitoes can or do act as vectors for all or any pathogens, mosquitoes are the arthropod group responsible for the most human deaths worldwide: more than 750,000 deaths every year!

Vector mosquitoes, and the parasites and pathogens that they transmit, are recognised to have played an important role in the development and dispersal of the human race, being responsible for some events that have shaped the course of history. Although vaccines, chemoprophylaxis, chemotherapy, genetics and vector control measures are becoming more sophisticated, even now, none of the major mosquito-borne diseases of the world can be said to be under complete control.

Mosquitoes are responsible for transmitting three types of human pathogenic organisms:
Arboviruses – viruses causing diseases such as dengue, Yellow fever and various encephalitides. (The term 'arbovirus' is derived from arthropod-borne-virus).
Plasmodia – protozoans which are the cause of malaria.
Filarial worms – nematodes that cause lymphatic filariasis.

Biological transmission refers to the situation where the pathogen or parasite undergoes a period of development and/or multiplication within the vector (which acts as a true intermediate host and is essential for the completion of the cycle) before being passed on to another host following this incubation period (sometimes called the 'intrinsic' incubation period to differentiate it from the incubation period in the vertebrate host which is the 'extrinsic' incubation period). There are three systems that apply (Table 3.1.1):

1. The pathogen develops and multiplies, e.g. malarial parasites – there is sexual union of the blood stages in the mosquito gut, encystation in the gut wall, multiple sporozoite formation in the cyst and movement of the sporozoites to the mosquito's salivary glands as infective stages for introduction into a new host during subsequent feeding.
2. The pathogen develops only, e.g. filarial parasites – the microfilarial blood stages taken into the mosquito gut escape from the gut and develop through three stages in the mosquito's tissues before entering the head as infective stages for introduction into a new host during subsequent feeding.
3. The pathogen multiplies only, e.g. arboviruses – virus particles taken in with blood into the mosquito gut invade the gut cells, disseminate and multiply in body tissues and penetrate the salivary glands to be introduced into a new host during subsequent feeding.

Table 3.1.1. Three types of biologically transmitted human pathogenic organisms**.**

Pathogen	**Biological changes suffered by the pathogen within the vector**	**Deseases**
Protozoan parasite - *Plasmodium* sp.	Pathogen develops and multiplies	Malaria
Filarial parasites - micro worms	Pathogen only develops	Elephantiasis
Arboviruses (Arthropod borne virus)	Pathogen only multiplies	Viral diseases, more than 1000 examples

Irrespective of the particular cycle, once a mosquito vector has picked up a pathogen, the vector needs to survive for at least the period of time required by the pathogen to complete its development cycle or multiply to the point that the mosquito becomes infective, before that mosquito can be involved in the transmission of the pathogen to the new host. This intrinsic

incubation period varies with pathogen and temperature, but in general is in the order of 1-2 weeks. Thus the mosquito must survive for at least 1-2 weeks for it to become infective and therefore mosquito longevity is a critical factor in the dynamics of transmission of disease pathogens.

In all cases, the crucial factor in transmission to man (the epidemiology of the disease) is the amount and type of contact between the mosquito vector and the human host. The incidence and prevalence of disease in an area will depend upon the presence of the disease, susceptible vectors and the amount of human-vector contact. The latter is a product of interaction between the habitat and behaviour of the mosquito vector and the habitat and behaviour of the human host. The more often that a potentially infective mosquito intrudes into the human environment, or that the humans intrude into the natural environment where mosquitoes harbour pathogenic organisms, the greater the risk of initiating an urban outbreak or epidemic.

Mosquito borne diseases are complicated communicable diseases as they involve the vector as an additional component of the disease system. Social, behavioural, environmental and immunological factors may affect the human component, yet with vector involvement further influences impinge on the system and still more may arise if the disease is a zoonosis, involving other vertebrates as well as humans. Such a complex system may seem formidable, however the more complex the system, the greater the number of opportunities exist for disrupting the disease cycle.

3.1.1 Parasites

3.1.1.1 Malaria

Malaria parasites are micro-organisms that belong to the genus *Plasmodium*. There are more than 100 species of *Plasmodium*, which can infect many animal species such as reptiles, birds and various mammals. Only four species of *Plasmodium* infect humans in nature (there are some other species which can, exceptionally or under experimental conditions, infect humans).

The four species infecting humans are:

- *Plasmodium falciparum*, which is found worldwide in tropical and subtropical areas. It is the only species that can cause severe, potentially fatal malaria. It is estimated that every year 700,000 to 2.7 million people are killed by *P. falciparum*, especially in Africa where this species predominates. *P. falciparum* can cause severe malaria because it multiples rapidly in the blood, and can thus cause severe blood loss (anaemia). In addition, the infected parasites can clog small blood vessels. When this occurs in the brain cerebral malaria results, a complication that can be fatal.
- *Plasmodium vivax*, which is found mostly in Asia, Latin America, and in some parts of Africa. Because of the population densities, especially in Asia, it is probably the most prevalent human malaria parasite. While *P. vivax* only exceptionally causes death (most often due to rupture of an enlarged spleen) it can cause symptoms that are incapacitating. Thus, *P. vivax* contributes substantially to the disease burden (morbidity) of malaria, with a resulting social and economic impact. *P. vivax* (as well as *Plasmodium ovale*) has dormant liver stages ("hypnozoites") that can activate and invade the blood ("relapse") several months or years after the infecting mosquito bite.
- *Plasmodium ovale* is found mostly in Africa (especially West Africa) and the islands of the western Pacific. It is biologically and morphologically very similar to *P. vivax*. However, differently from *P. vivax*, it can infect individuals who are negative for the Duffy blood group, which is the case for many residents of sub-Saharan Africa. This explains the greater prevalence of *P. ovale* (rather than *P. vivax*) in most of Africa.
- *Plasmodium malariae*, found worldwide, is the only human malaria parasite species that has a quartan cycle (three-day cycle). (The three other species have a tertian, two-day

cycle). *P. malariae* causes a long-lasting, chronic infection that in some cases can last a lifetime. In some patients, *P. malariae* can cause serious complications such as the nephrotic syndrome.

Malaria is transmitted among humans by female mosquitoes of the genus *Anopheles.* Female mosquitoes take blood meals to carry out egg production, and such blood meals are the link between the human and the mosquito hosts in the parasite life cycle. Of the approximately 430 known species of *Anopheles*, only 30-50 transmit malaria in nature.

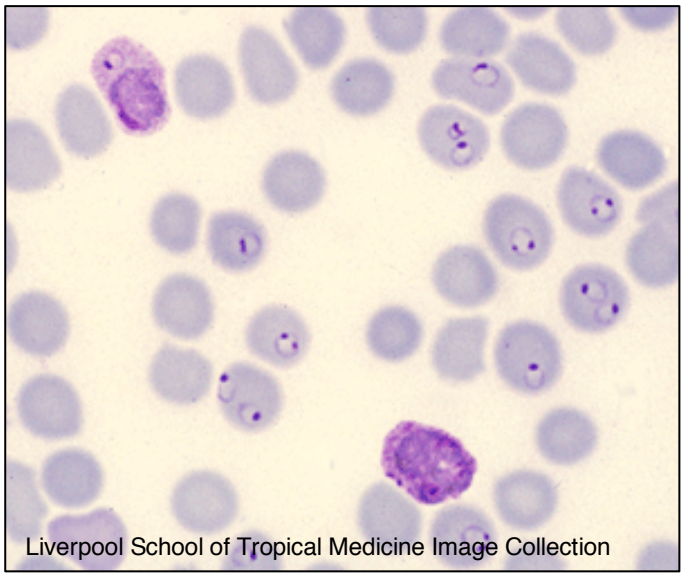


Figure 3.1.1. *Plasmodium falciparum* and *P. vivax* inside red cells.

Life Cycle of Malaria

In nature, malaria parasites spread by infecting successively two types of hosts: humans and female *Anopheles* mosquitoes. In the human hosts, the parasites grow and multiply, first in the liver cells and then in the red cells of the blood (Figure 3.1.1). In the blood, successive broods of parasites grow inside the red cells and destroy them, releasing daughter parasites ("merozoites") that continue the cycle by invading other red cells. The blood stage parasites are those that cause the symptoms of malaria.

When certain forms of blood stage parasites ("gametocytes") are picked up by a female *Anopheles* mosquito during a blood meal they start another, different, cycle of growth and multiplication in the mosquito. After 10-18 days the parasites are found (as "sporozoites") in the mosquito's salivary glands. When the *Anopheles* mosquito takes a blood meal from another human, the sporozoites are injected along with the mosquito's saliva and start another human infection once they parasitize the liver cells. Thus the mosquito carries the disease from one human to another (acting as a "vector"). In contrast to the human host, the mosquito vector does not suffer from the presence of the parasites (Figure 3.1.2).

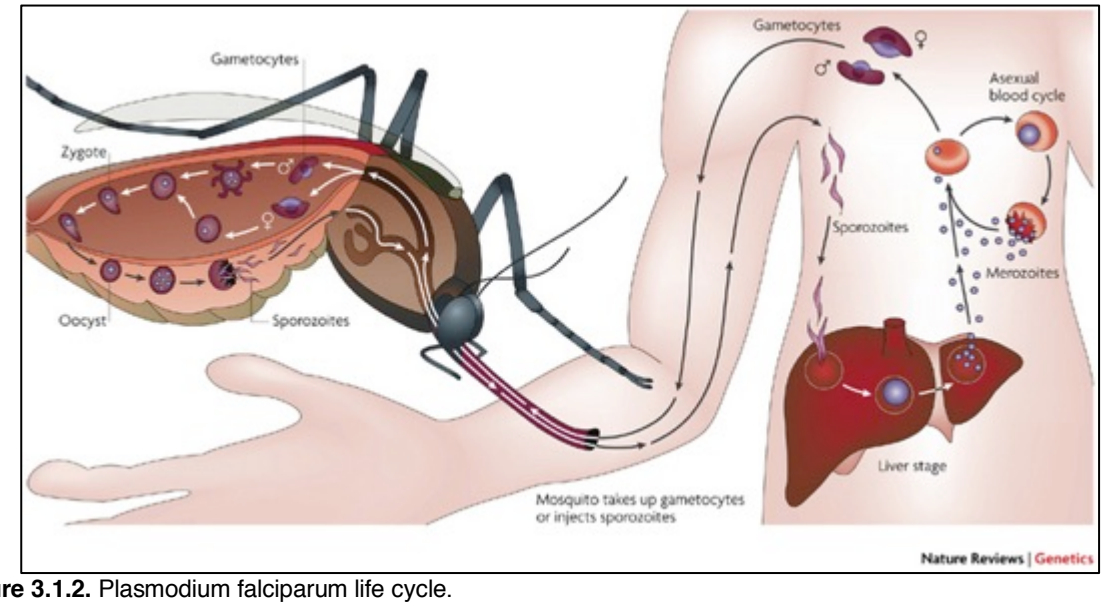


Figure 3.1.2. Plasmodium falciparum life cycle.

The malaria parasite life cycle involves two hosts. During a blood meal a malaria-infected female *Anopheles* mosquito inoculates sporozoites into the human host. Sporozoites infect liver cells and mature into schizonts, which rupture and release merozoites. (Of note, in *P. vivax* and *P. ovale* a dormant stage [hypnozoites] can persist in the liver and cause relapses by invading the bloodstream weeks, or even years, later.) After this initial replication in the liver (exo-erythrocytic schizogony) the parasites undergo asexual multiplication in the erythrocytes (erythrocytic schizogony). Merozoites infect red blood cells. The ring stage trophozoites mature into schizonts, which rupture releasing merozoites. Some parasites differentiate into sexual erythrocytic stages (gametocytes) (Figure 3.1.2). Blood stage parasites are responsible for the clinical manifestations of the disease.

The gametocytes, male (microgametocytes) and female (macrogametocytes), are ingested by an *Anopheles* mosquito during a blood meal. The parasites' multiplication in the mosquito is known as the sporogonic cycle. While in the mosquito's stomach, the microgametes penetrate the macrogametes generating zygotes. The zygotes in turn become motile and elongated (ookinetes) which invade the midgut wall of the mosquito where they develop into oocysts. The oocysts grow, rupture, and release sporozoites, which make their way to the mosquito's salivary glands. Inoculation of the sporozoites into a new human host perpetuates the malaria life cycle (Figure 3.1.2).

Humans infected with malaria parasites can develop a wide range of symptoms. These vary from asymptomatic infections (no apparent illness), to the classic symptoms of malaria (fever, chills, sweating, headaches, muscle pains), to severe complications (cerebral malaria, anaemia, kidney failure) that can result in death. The severity of the symptoms depends on several factors, such as the species (type) of infecting parasite and the human's acquired immunity and genetic background.

The successful development of the malaria parasite in the mosquito (from the "gametocyte" stage to the "sporozoite" stage) depends on several factors. The most important are ambient temperature (higher temperatures accelerate the parasite growth in the mosquito), humidity and whether the *Anopheles* survives long enough to allow the parasite to complete its cycle in the mosquito host.

Excerpts from http://www.cdc.gov/malaria

3.1.1.2 Elephantiasis or Lymphatic filariasis

Lymphatic filariasis, commonly known as elephantiasis, is a neglected tropical disease. Infection occurs when filarial parasites are transmitted to humans through mosquitoes and can result in an altered lymphatic system and the abnormal enlargement of body parts, causing pain, severe disability and social stigma. Infection is usually acquired in childhood causing hidden damage to the lymphatic system.The painful and profoundly disfiguring visible manifestations of the disease, lymphoedema, elephantiasis and scrotal swelling occur later in life and lead to permanent disability. These patients are not only physically disabled, but suffer mental, social and financial losses contributing to stigma and poverty.

Currently, 1.10 billion people in 55 countries are living in areas that require preventive chemotherapy to stop the spread of infection. Approximately 80% of these people are living in the following 10 countries: Angola, Cameroon, Côte d'Ivoire, Democratic Republic of the Congo, India, Indonesia, Mozambique, Myanmar, Nigeria and the United Republic of Tanzania.Globally, an estimated 25 million men suffer with genital disease and over 15 million people are afflicted with lymphoedema. Eliminating lymphatic filariasis can prevent unnecessary suffering and contribute to the reduction of poverty.

Elephantiasis is caused by parasitic worms, nematodes (roundworms) of the family Filariodidea. There are 3 species of these thread-like filarial worms:

- *Wuchereria bancrofti*, which is responsible for 90% of the cases
- *Brugia malayi*, which causes most of the remainder of the cases
- *Brugia timori*, which also causes the disease.

Adult worms lodge in the lymphatic system and disrupt the immune system. The worms can live for an average of 6–8 years and, during their life time, produce millions of microfilariae (immature larvae) that circulate in the blood.

Transmission

Mosquitoes are infected with microfilariae by ingesting blood when biting an infected host. Microfilariae mature into infective larvae within the mosquito. When infected mosquitoes bite people, mature parasite larvae are deposited on the skin from where they can enter the body. The larvae then migrate to the lymphatic vessels where they develop into adult worms, thus continuing a cycle of transmission.

Lymphatic filariasis is transmitted by different mosquitoes for example by *Culex* species, widespread across urban and semi-urban areas, *Anopheles*, mainly found in rural areas, and *Aedes*, mainly endemic on islands in the Pacific.

Symptoms

Lymphatic filariasis infection involves asymptomatic, acute, and chronic conditions. The majority of infections are asymptomatic, showing no external signs of infection. These asymptomatic infections still cause damage to the lymphatic system and the kidneys, and alter the body's immune system.

Acute episodes of local inflammation involving skin, lymph nodes and lymphatic vessels often accompany chronic lymphoedema or elephantiasis. Some of these episodes are caused by the body's immune response to the parasite. Most are the result of bacterial skin infection, however, where normal defences have been partially lost due to underlying lymphatic damage.

When lymphatic filariasis develops into chronic conditions it leads to lymphoedema (tissue swelling) or elephantiasis (skin/tissue thickening) of limbs and hydrocele (scrotal swelling) (Figure 3.1.3). Involvement of breasts and genital organs is common.

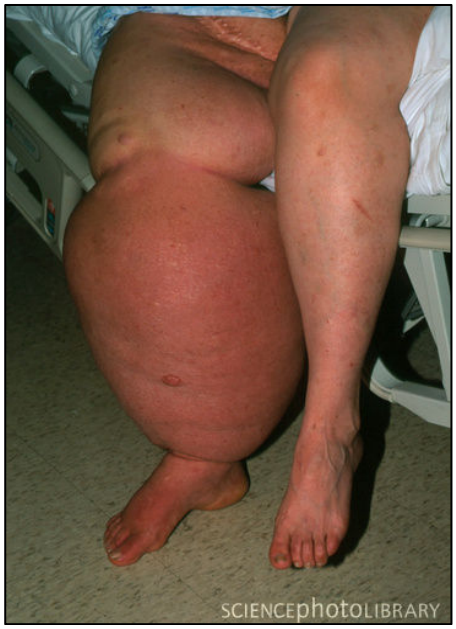


Figure 3.1.3. Patient with chronic lymphoedema or elephantiasis.

Treatment

Large-scale treatment involves a single dose of 2 medicines given annually to an entire at-risk population in the following way: albendazole (400 mg) together with either ivermectin (150-200 mcg/kg) or with diethylcarbamazine citrate (DEC) (6 mg/kg). These medicines have a limited effect on adult parasites but effectively reduce the density of microfilariae in the bloodstream and prevent the spread of parasites to mosquitoes. This recommended large-scale treatment strategy is called preventive chemotherapy when conducted annually for 4–6 years, and it can interrupt the transmission cycle.

Morbidity management and disability prevention are vital for improving public health and should be fully integrated into the health system to ensure sustainability. Surgery can alleviate most cases of hydrocele. Clinical severity and progression of the disease, including acute inflammatory episodes, can be reduced and prevented with simple measures of hygiene, skin care, exercise, and elevation of affected limbs. People with lymphoedema must have access to continuing care throughout their lives, both to manage the disease and to prevent progression to more advanced stages.

Excerpts from http://www.who.int/mediacentre/factsheets/fs102/en/ and http://www.newspakistan.pk/2012/04/23/diagnose-treat-elephantiasis/

3.1.2 Arboviruses

The table 3.1.2. shows the taxonomic classification of the arboviruses discussed in this section.

Table 3.1.2. Arboviruses taxonomic classification

Family	Flaviviridae	Family Togaviridae
Genus	*Flavivirus*	Genus *Alphavirus*
Species	*Dengue virus* (DENV) *Japanese encephalitis virus* (JEV) *Murray Valley encephalitis virus* (MVEV) *West Nile virus* (WNV) *Kunjin* *Zika virus* (ZIKV) *Yellow fever virus* (YFV)	*Eastern equine encephalitis virus* (EEE) *Ross River virus* (RRV) *Chikungunya virus* (CHIKV)

3.1.2.1 Dengue

Dengue (DF) and dengue haemorrhagic fever (DHF) are caused by one of four closely related, but antigenically distinct, virus serotypes (DEN-1, DEN-2, DEN-3, and DEN-4), of the genus *Flavivirus*. Infection with one of these serotypes provides immunity to only that serotype for life, so persons living in a dengue-endemic area can have more than one dengue infection during their lifetime. DF and DHF are primarily diseases of tropical and sub-tropical areas and the four different dengue serotypes are maintained in a cycle that involves humans and the *Aedes* mosquito. *Aedes aegypti*, a domestic day-biting mosquito that prefers to feed on humans, is the most common vector. Infections produce a spectrum of clinical illness ranging from a non-specific viral syndrome to severe and fatal haemorrhagic disease. Important risk factors for DHF include the strain of the infecting virus, as well as the age, and especially the prior dengue infection history of the patient.

Symptoms

Four to six days after infection and last for up to 10 days, may include;

- Sudden, high fever
- Severe headaches
- Pain behind the eyes
- Severe joint and muscle pain
- Nausea
- Vomiting
- Skin rash, which appears three to four days after the onset of fever
- Mild bleeding (such a nose bleed, bleeding gums, or easy bruising)

History of Dengue

The first reported epidemics of DF occurred in 1779-1780 in Asia, Africa and North America. The near simultaneous occurrence of outbreaks on three continents indicated that these viruses and their mosquito vector have had a worldwide distribution in the tropics for more than 200 years. During most of this time DF was considered a mild, non-fatal disease of visitors to the tropics. Generally, there were long intervals (10-40 years) between major epidemics, mainly because the introduction of a new serotype in a susceptible population occurred only if viruses and their mosquito vector could survive the slow transport between population centres via sailing vessels.

A pandemic of dengue began in Southeast Asia after World War II and has spread around the globe since then. Epidemics caused by multiple serotypes (hyperendemicity) are more frequent, the geographic distribution of dengue viruses and their mosquito vectors has expanded and DHF has emerged in the Pacific region and the Americas. In Southeast Asia, epidemic DHF first appeared in the 1950s but by 1975 it had become a frequent cause of hospitalization and death among children in many countries in that region.

Current Trends

In the 1980s, DHF began a second expansion into Asia when Sri Lanka, India, and the Maldive Islands had their first major DHF epidemics. Pakistan first reported an epidemic of dengue fever in 1994. The epidemics in Sri Lanka and India were associated with multiple dengue virus serotypes, but DEN-3 was predominant and was genetically distinct from DEN-3 viruses previously isolated from infected persons in those countries. After an absence of 35 years, epidemic dengue fever re-emerged in both Taiwan and the People's Republic of China in the 1980s. The People's Republic of China had a series of epidemics caused by all four serotypes and the first major epidemic of DHF, caused by DEN-2, was reported on Hainan Island in 1985. Singapore also had a resurgence of dengue/DHF from 1990 to 1994 after a successful control program had prevented significant transmission for over 20 years. In other countries of Asia where DHF is endemic, the epidemics have become progressively larger in the last years.

In the Pacific, dengue viruses were reintroduced in the early 1970s after an absence of more than 25 years. Epidemic activity caused by all four serotypes has intensified in recent years with major epidemics of DHF on several islands.

Despite poor surveillance for dengue in Africa, epidemic dengue fever caused by all four serotypes has increased dramatically since 1980. Most activity has occurred in East Africa and major epidemics were reported for the first time in the Seychelles (1977), Kenya (1982, DEN-2), Mozambique (1985, DEN-3), Djibouti (1991-92, DEN-2), Somalia (1982, 1993, DEN-2), and Saudi Arabia (1994, DEN-2). Epidemic DHF has not been reported in Africa or the Middle East, but sporadic cases clinically compatible with DHF have been reported from Mozambique, Djibouti, and Saudi Arabia.

The emergence of dengue/DHF as a major public health problem has been most dramatic in the American region. In an effort to prevent urban Yellow fever, which is also transmitted by *Ae. aegypti*, the Pan American Health Organization started a campaign that eradicated *Ae. aegypti* from most Central and South American countries in the 1950s and 1960s. As a result, epidemic dengue occurred only sporadically in some Caribbean islands during this period. The *Ae. aegypti* eradication program, which was officially discontinued in the United States in 1970, gradually weakened elsewhere and the mosquito began to re-infest countries from which it had been eradicated. As a result, the geographic distribution of *Ae. aegypti* in 2002 was much wider than it was before the eradication program.

In 2013, cases have occurred in Florida (United States of America) and Yunnan province of China. Dengue also continues to affect several South American countries, notably Costa Rica, Honduras and Mexico. In Asia, Singapore has reported an increase in cases after a lapse of several years and outbreaks have also been reported in Laos. In 2014, trends indicate increases in the number

of cases in the People's Republic of China, the Cook Islands, Fiji, Malaysia and Vanuatu, with Dengue Type 3 (DEN 3) affecting the Pacific Island countries after a lapse of over 10 years. Dengue was also reported in Japan after a lapse of over 70 years.

In 2015, Delhi, India, recorded its worst outbreak since 2006 with over 15 000 cases. The Island of Hawaii, United States of America, was affected by an outbreak with 181 cases reported in 2015 and ongoing transmission in 2016. The Pacific island countries of Fiji, Tonga and French Polynesia have continued to record cases.

The year 2016 was characterized by large dengue outbreaks worldwide. The region of the Americas reported more than 2.38 million cases in 2016, where Brazil alone contributed slightly less than 1.5 million cases, approximately 3 times higher than in 2014. 1032 dengue deaths were also reported in the region. The Western Pacific Region reported more than 375 000 suspected cases of dengue in 2016, of which the Philippines reported 176 411 and Malaysia 100 028 cases, representing a similar burden to the previous year for both countries. The Solomon Islands declared an outbreak with more than 7000 suspected cases. In the African Region, Burkina Faso reported a localized outbreak of dengue with 1061 probable cases.

In 2017 (as of Epidemiological Week 11), the Region of Americas have reported 50 172 cases of dengue fever, a reduction as compared with corresponding periods in previous years. The Western Pacific Region has reported dengue outbreaks in several Member States in the Pacific, as well as the circulation of DENV-1 and DENV-2 serotypes.

An estimated 500 000 people with severe dengue require hospitalization each year, and about 2.5% of those affected die.

Figure 3.1.4. Countries and areas affected by Dengue.

Figure 3.1.4 shows the countries or areas affected by Dengue in 2014. For an up to date view visit the health map website: http://www.healthmap.org/dengue/en/

The reasons for the dramatic global emergence of DF/DHF as a major public health problem are complex and not well understood. However, several important factors can be identified:

1. Major global demographic changes have occurred, the most important of which have been uncontrolled urbanisation and concurrent population growth. These demographic changes have resulted in substandard housing and inadequate water, sewer and waste management systems, all of which increase *Aedes aegypti* population densities and facilitate transmission of *Ae. aegypti*-borne disease.
2. In most countries the public health infrastructure has deteriorated. Limited financial and human resources and competing priorities have resulted in a "crisis mentality" with emphasis on implementing so-called emergency control methods in response to epidemics rather than on developing programs to prevent epidemic transmission. This approach has been particularly detrimental to dengue control because, in most countries, surveillance is (just as

in the US) passive; the system to detect increased transmission normally relies on reports by local physicians who often do not consider dengue in their differential diagnoses. As a result, an epidemic has often reached or passed its peak before it is recognized.
3. Increased travel by airplane provides the ideal mechanism for infected human transport of dengue viruses between population centres of the tropics, resulting in a frequent exchange of dengue viruses and other pathogens.
4. Effective mosquito control is virtually non-existent in most dengue-endemic countries. Considerable emphasis in the past has been placed on ultra-low-volume insecticide space sprays for adult mosquito control, a relatively ineffective approach for controlling *Ae. aegypti*.

Future Outlook

Status of vaccine development:

The first dengue vaccine, Dengvaxia (CYD-TDV) by Sanofi Pasteur, was first registered in Mexico in December, 2015. CYD-TDV is a live recombinant tetravalent dengue vaccine that has been evaluated as a 3-dose series on a 0/6/12 month schedule in Phase III clinical studies. It has been registered for use in individuals 9-45 years of age living in endemic areas.

Challenges to vaccine development:

Infection by one of the four dengue virus serotypes has been shown to confer lasting protection against homotypic re-infection, but only transient protection against a secondary heterotypic infection. Moreover, secondary heterotypic infection is associated with an increased risk of severe disease. This and other observations suggest an immunopathological component in dengue pathogenesis, which is referred to as immune enhancement of disease. Due to these dengue-specific complexities, vaccine development focuses on the generation of a tetravalent vaccine aimed at providing long-term protection against all virus serotypes. Additional challenges are posed by the lack of an adequate animal disease model and the resulting uncertainty around correlates of protection. In spite of these challenges, vaccine development has made remarkable progress in recent years, and the current dengue vaccine pipeline is advanced, diverse and overall promising.

Wolbachia

Wolbachia is a bacterium present naturally in up to 60% of all the different species of insects around us, including some mosquitoes. However, it is not usually found in the *Aedes aegypti* mosquito, the primary species responsible for transmitting human viruses such as dengue, chikungunya, and Zika.

For many years scientists have been studying *Wolbachia*, looking for ways to use it to potentially control the mosquitoes that spread human diseases. Researchers have shown that when introduced into the *Aedes aegypti* mosquito, *Wolbachia* can stop these viruses from growing inside the mosquito and being transmitted to people. This important discovery has the potential to transform the fight against life-threatening viral diseases.

Wolbachia is safe for humans, other vertebrates and the environment. It is a naturally occurring bacterium already found in the environment in many insect species. Two independent risk assessments have been conducted, both of which gave an overall risk rating of ‘negligible’ (the lowest possible rating) for the release of mosquitoes with *Wolbachia*.

Wolbachia can only be transmitted from parent to offspring inside the female’s egg. It spreads into insect populations by altering the reproductive success of the insect that carries it. When a male mosquito that carries *Wolbachia* mates with a female without the bacteria then that female’s eggs don’t hatch. *Wolbachia* infected female mosquitoes do not suffer from this effect and produce normal numbers of offspring – which carry *Wolbachia*. Initially, this reproductive effect will be very small as there will be few *Wolbachia* infected mosquitoes in the population, but over successive generations the numbers of males and female mosquitoes infected with *Wolbachia* will increase.

Sources:
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http://www.who.int/immunization/research/development/dengue_vaccines/en/
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3.1.2.2 Japanese encephalitis

Japanese encephalitis is a viral disease that infects humans and other vertebrates. It is transmitted by mosquitoes, and in humans causes inflammation of the membranes around the brain. Intensification and expansion of irrigated rice production systems in South and South-East Asia over the past 20 years have had an important impact on the disease burden caused by Japanese encephalitis. Where irrigation expands into semi-arid areas, the flooding of the fields at the start of each cropping cycle leads to an explosive build-up of the mosquito population. This may cause the circulation of the virus to spill over from their usual hosts (birds and pigs) into the human population (Figure 3.1.5).

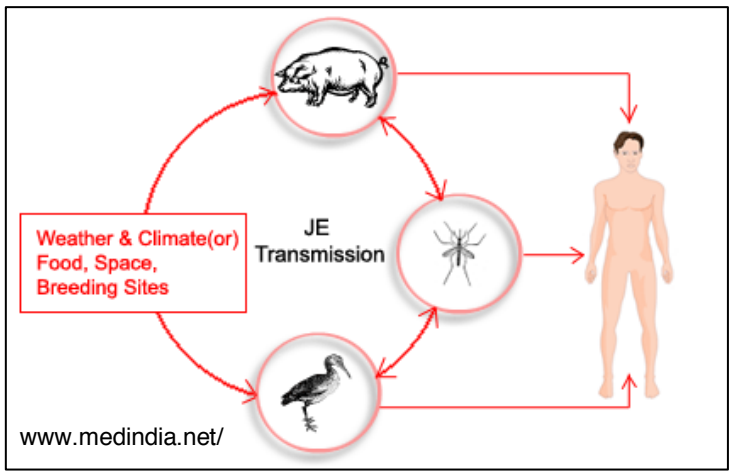

Figure 3.1.5. Transmission of the Japanese encephalitis virus.

Japanese encephalitis (JE) is caused by a flavivirus that affects the membranes around the brain. Most JE virus infections are mild (fever and headache) or without apparent symptoms, but approximately 1 in 200 infections results in severe disease characterised by rapid onset of high fever, headache, neck stiffness, disorientation, coma, seizures, spastic paralysis and death.

The case fatality rate can be as high as 60% among those with disease symptoms; 30% of those who survive suffer from lasting damage to the central nervous system. In areas where the JE virus is common, encephalitis occurs mainly in young children because older children and adults have already been infected and are immune.

The virus causing JE is transmitted by mosquitoes belonging to the *Culex tritaeniorhynchus* and *Culex vishnui* groups, which breed particularly in flooded rice fields. The virus circulates in ardeid birds (herons and egrets). Pigs are amplifying hosts, in that the virus reproduces in pigs and infects mosquitoes that take blood meals, but does not cause disease. The virus tends to spill over into human populations when infected mosquito populations build up explosively and the human biting rate increases (Figure 3.1.5).

JE is a leading cause of viral encephalitis in Asia with 30,000-50,000 clinical cases reported annually. It occurs from the islands of the Western Pacific in the east to the Pakistani border in the west, and from Korea in the north to Papua New Guinea in the south. Because of the critical role of pigs, its presence in Muslim countries is negligible. JE distribution is very significantly linked to irrigated rice production combined with pig rearing.

Japanese encephalitis is a patchy disease and important outbreaks have occurred in a number of places in the past 15 years, including South India (Arkot district in Tamil Nadu) and in Sri Lanka (Mahaweli System H).

Vaccination

An effective killed vaccine is available for Japanese encephalitis, but it is expensive and requires one primary vaccination followed by two boosters. This is an adequate intervention for travellers, but has limited public health value in areas where health services have limited resources. An inexpensive live-attenuated vaccine is used in China, but is not available elsewhere.

Excerpts from http://www.who.int/water_sanitation_health/diseases/encephalitis/en/

3.1.2.3 West Nile

West Nile virus (WNV) is in the genus *Flavivirus*, part of the Flavivirus Japanese Encephalitis Antigenic Complex. This complex includes the Alfuy, Cacipacore, Japanese encephalitis, Koutango, Kunjin, Murray Valley encephalitis, St. Louis encephalitis, Rocio, Stratford, Usutu and Yaounde viruses (Table 3.1.2).

Transmission Cycle

West Nile virus is amplified during periods of adult mosquito blood-feeding by continuous transmission between mosquito vectors (*Culex* sp. and *Aedes* sp.) and bird reservoir hosts (Figure 3.1.6). Infectious mosquitoes carry virus particles in their salivary glands and infect susceptible bird species during blood-meal feeding. Competent bird reservoirs will sustain an infectious viremia (virus circulating in the bloodstream) for 1 to 4 days after exposure, after which these hosts develop life-long immunity. A sufficient number of vectors must feed on an infectious host to ensure that some survive long enough to feed again on a susceptible reservoir host.

People, horses, and most other mammals are not known to develop infectious-level viremia very often and are probably "dead-end" or incidental-hosts.

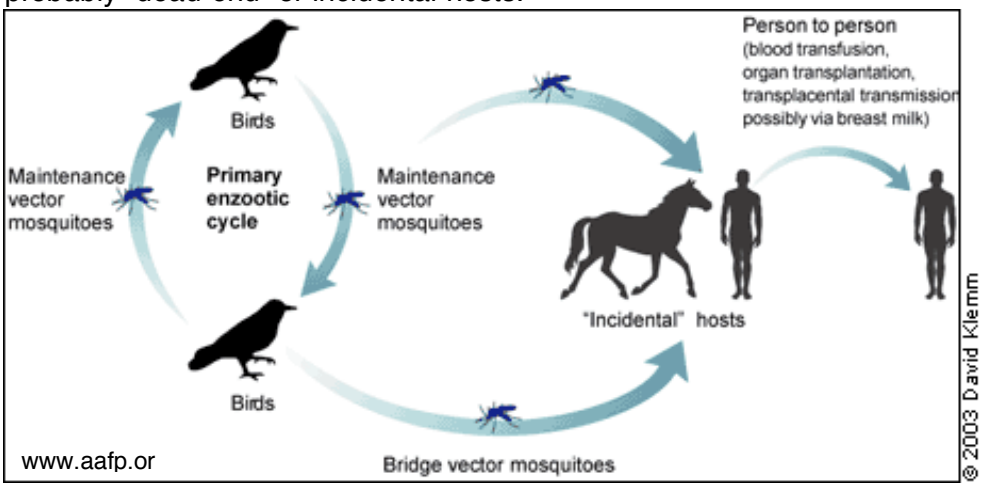

Figure 3.1.6. West Nile Virus Transmission cycle

WNV has emerged in recent years in temperate regions of Europe and North America, presenting a threat to public and animal health. The most serious manifestation of WNV infection is fatal encephalitis (inflammation of the brain) in humans and horses, as well as mortality in certain domestic and wild birds. WNV has also been a significant cause of human illness in the United States in 2002 and 2003.

Symptoms

Most people (70-80%) who become infected with West Nile virus do not develop any symptoms. About 1 in 5 people (roughly 20%) who are infected will develop a fever with other symptoms such as headache, body aches, joint pains, vomiting, diarrhoea, or rash on the skin covering the trunk of the body. Most people with this type of West Nile virus disease recover completely, but fatigue and weakness can last for weeks or months. Less than 1% (roughly 1 out of every 150 people infected, or 0.67%), of people who are infected will develop a serious neurologic illness such as encephalitis or meningitis (inflammation of the brain or surrounding tissues). The

symptoms of neurologic illness can include headache, high fever, neck stiffness, disorientation, coma, tremors, seizures, or paralysis.

People with certain medical conditions, such as cancer, diabetes, hypertension and kidney disease are also at greater risk for serious illness. Neurologic illness may also manifest as meningitis; an inflammation of the meninges that encases the brain. For reasons yet to be understood those over the age of 50 and immune compromised patients such as transplant recipients are at the highest risk of developing this disease after infection.

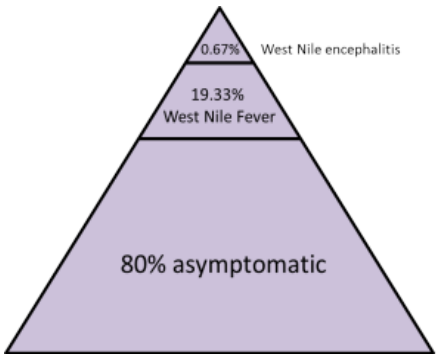

Figure 3.1.7. Clinical manifestations in patients with West Nile virus infection.

Recovery from severe disease may take several weeks or months. Some of the neurologic effects may be permanent. About 10 percent of people who develop neurologic infection due to West Nile virus will die.

It is important to remember that while this disease does have significant clinical manifestations the actual disease burden in infected individuals is lower, 80% of persons will not experience symptoms.

History

West Nile virus was first isolated from a febrile adult woman in the West Nile District of Uganda in 1937. The ecology was characterized in Egypt in the 1950s. The virus became recognised as a cause of severe human meningitis or encephalitis (inflammation of the spinal cord *and* brain) in elderly patients during an outbreak in Israel in 1957. Equine disease was first noted in Egypt and France in the early 1960s. WNV first appeared in North America in 1999, with encephalitis reported in humans and horses. The subsequent spread in the United States is an important milestone in the evolving history of this virus.

West Nile virus has been described in Africa, Europe, the Middle East, west and central Asia, Oceania (subtype Kunjin), and most recently, North America.

Outbreaks of WNV encephalitis in humans have occurred in Algeria in 1994, Romania in 1996-1997, the Czech Republic in 1997, the Democratic Republic of the Congo in 1998, Russia in 1999, the United States in 1999-2003, and Israel in 2000. Epizootics of disease in horses occurred in Morocco in 1996, Italy in 1998, the United States in 1999-2001 and France in 2000, and in birds in Israel in 1997-2001 and in the United States in 1999-2002. Since its introduction in 1999 into USA, the virus has spread and is now widely established from Canada to Venezuela.

Excerpts from http://www.cdc.gov/ncidod/dvbid/westnile/index.htm

3.1.2.4 Zika

Zika virus (ZIKV) is a member of the Flaviviridae virus family and the *Flavivirus* genus. In humans, it causes a disease known as Zika fever. It is related to dengue, yellow fever, West Nile and Japanese encephalitis, viruses that are also members of the virus family Flaviviridae (Table 3.1.2). Outbreaks of Zika virus have previously been reported in tropical Africa, in some areas in Southeast Asia and more recently in the Pacific Islands and currently in the Americas, especially Brazil.

Zika virus infection is symptomatic in only about one out of every five cases. When symptomatic, Zika infection usually presents as an influenza-like syndrome, often mistaken for other arboviral infections like dengue or chikungunya. Zika causes brain damage and microcephaly in babies born with the virus after their mothers have been infected during the pregnancy.

Transmission

Zika virus is transmitted by mosquitoes and has been isolated from a number of species in the genus *Aedes - Aedes aegypti*, *Aedes africanus, Aedes apicoargenteus, Aedes furcifer, Aedes luteocephalus* and *Aedes vitattus,* mosquitoes that are not normally found in New Zealand. The virus reservoirs are presumably monkeys. Zika virus is transmitted to humans mainly by certain species of *Aedes* mosquitoes. Some of these species bite during the day as well as in the late afternoon/evening.

In 2009, it was proved that Zika virus can be sexually transmitted between humans. Professor Brian Foy, a university biologist from the Colorado State University at the Arthropod Borne and Infectious Disease Laboratory, visited Senegal to study mosquitos and was bitten on a number of occasions during his research. A few days after returning to the USA he fell ill with Zika, but not before having vaginal intercourse with his wife. His wife subsequently showed symptoms of Zika infection, along with extreme sensitivity to light. Foy is the first person known to have passed on an insect-borne virus to another human by sexual contact.

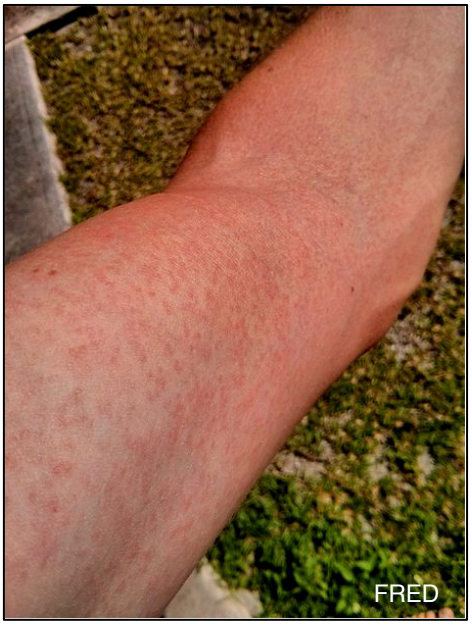

Figure 3.1.8. Rash on arm due to Zika virus.

Symptoms

The main clinical symptoms in patients are fever, conjunctivitis, transient arthritis/arthralgia (mainly in the smaller joints of the hands and feet) and maculopapular rash (that often starts on the face and then spreads throughout the body, Figure 3.1.8). In general, the disease symptoms are mild and short-lasting (2-7 days).

Studies show that the extrinsic incubation period in mosquitoes is about 10 days. The incubation period in human is typically 3–12 days. There is no specific therapy for Zika virus infection and acute symptoms typically resolve within 4-7 days

The first well documented case of Zika virus was in 1964, beginning with a mild headache and progressing to a maculopapular rash, fever, and back pain. Within 2 days, the rash was fading, and within 3 days, the fever was gone and only the rash remained. There is no vaccine or preventive drug for Zika virus, and only treatment of symptoms is possible. Usually non-steroid anti-inflammatories and/or non-salicylic analgesics are used.

- low-grade fever (between 37.8°C and 38.5°C)
- arthralgia, notably of small joints of hands and feet, with possible swollen joints
- myalgia
- headache, retro-ocular headaches
- conjunctivitis
- cutaneous maculopapular rash
- post-infection asthenia which seems to be frequent.

More rarely observed symptoms include digestive problems (abdominal pain, diarrhoea, constipation), mucous membrane ulcerations (aphthae), and pruritus.

Zika virus infection causes a mild disease and, other than notification, no particular action is

required. However, as Zika infection may cause a rash that could be confused with more serious diseases such as measles or dengue, these more serious diseases do need to be ruled out. Diagnosis of Zika will first and foremost be by exclusion, based on symptoms, travel history and exclusion of more serious diseases including measles, rubella and dengue. The pathogenesis of the virus is hypothesized to first infect dendritic cells near the site of inoculation, and then spread to lymph nodes and the bloodstream. In terms of replication, flaviviruses generally replicate in the cytoplasm, but Zika virus antigens have been found in infected cell nuclei.

Diagnosis

Zika virus can be identified by RT-PCR in acutely ill patients and from day 5 post onset of fever by serology (detection of specific IgM antibodies). Serological cross-reactions with closely related flaviviruses are possible.

Treatment

Symptomatic only (non-steroid anti-inflammatories, non-salicylic analgesics); no vaccine or preventive drug is available.

History

The first outbreak of the disease outside of Africa and Asia was in April 2007, on the island of Yap in the Federated States of Micronesia. This virus was characterized by rash, conjunctivitis, and arthralgia, and was initially thought to be dengue. The Chikungunya and Ross River viruses were also suspected. However, serum samples from patients in the acute phase of illness contained RNA of Zika virus. The Zika fever disease process was relatively mild: there were 49 confirmed cases, 59 unconfirmed cases, no deaths and no hospitalisations.

Zika virus could be considered an emerging pathogen, as it spread outside Africa and Asia for the first time in 2007. Thus far, it has been a relatively mild disease with limited scope, but its true potential as a virus and as an agent of disease is currently unknown.

Major Outbreaks

Serologic studies have shown that Zika infections are occurring from Africa to Southeast Asia; in 1978 a small outbreak of acute fever in Indonesia due to Zika virus infection was described.

An outbreak has been reported on Yap Island, Federated States of Micronesia (FSM) from April to July 2007. This was the first outbreak of Zika virus identified outside of Africa and Asia. A total of 108 cases were confirmed by PCR or serology and 72 additional cases were suspected. The most common symptoms were rash, fever, arthralgia and conjunctivitis, and no deaths were reported. The mosquito *Aedes hensilli*, which was the predominant species identified in Yap during the outbreak, was probably the main vector of transmission. While the way of introduction of the virus on Yap Island remains uncertain, it is likely to have happened through introduction of infected mosquitoes or a viraemic human.

Brazil: In May 2015, the public health authorities of Brazil confirmed autochthonous transmission of Zika virus in the north-eastern part of the country. As of 8 October, autochthonous cases of Zika virus had been detected in 14 states: Alagoas, Bahia, Ceará, Maranhão, Mato Grosso, Pará, Paraná, Paraíba, Pernambuco, Piauí, Rio de Janeiro, Rio Grande do Norte, Roraima, and São Paulo. Public health measures implemented by national and state authorities include the development and dissemination of sentinel protocol for Zika virus surveillance, development and validation of protocol for surveillance of neurological syndromes, and vector control activities.
Colombia: As of 16 October, 9 samples were laboratory-confirmed as Zika virus infections out of 98 samples from the Bolívar department (13 from Cartagena and 85 from Turbaco). These are the first cases of Zika virus infection detected in the country.

Outbreaks in the Pacific Islands

French Polynesia: Between early October 2013 and 21 March 2014, 8,700 suspected cases of

Zika. New Caledonia: Between 25 November 2013 and 25 March 2014 there have been 352 confirmed cases of Zika virus. Of these, 244 are locally transmitted cases while the other 32 cases were imported from French Polynesia.
Cook Islands: Between 13 February and 24 March 2014 there have been 648 dengue-like illness cases reported with 49 of these laboratory confirmed with Zika virus.
Easter Island: As of 07 March 2014 there have been 40 suspected cases and 1 confirmed case of Zika virus reported.
Cook Islands, Solomon Islands, Vanuatu, Fiji and Samoa in 2015.

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3.1.2.5 Yellow fever

Yellow fever is a viral disease that has caused large epidemics in Africa and the Americas. It can be recognized from historic texts stretching back 400 years. The "yellow" in the name is explained by the jaundice that affects some patients (Figure 3.1.9). The disease is caused by the Yellow fever virus, which belongs to the *Flavivirus* group. In Africa there are two distinct genetic types (called topotypes) associated with East and West Africa. South America has two different types, but since 1974 only one has been identified as the cause of disease outbreaks.

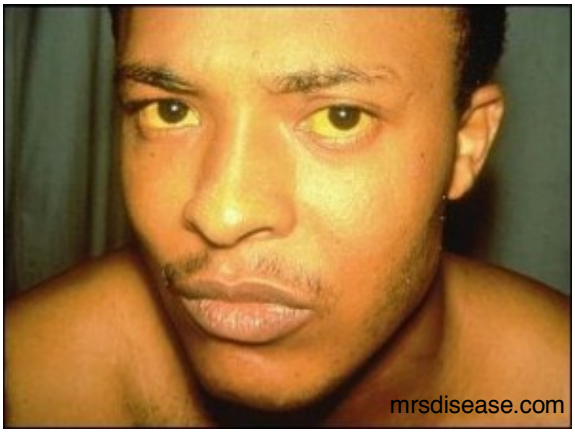


Figure 3.1.9. Jaundice caused by liver damage.

Symptoms
Infection causes a wide spectrum of symptoms, from mild to severe illness and death. The virus remains silent in the body during an incubation period of 3-6 days. There are then two disease phases. While some infections have no symptoms whatsoever, the first "acute" phase is normally characterised by fever, muscle pain (with prominent backache), headache, shivers, loss of appetite, nausea and/or vomiting. Often, the high fever is paradoxically associated with a slow pulse. After 3-4 days most patients improve and their symptoms disappear.

However, 15% enter a "toxic phase" within 24 hours. Fever reappears and several body systems are affected. The patient rapidly develops jaundice and complains of abdominal pain with vomiting. Bleeding can occur from the mouth, nose, eyes and/or stomach. Once this happens, blood appears in the vomit and faeces. Kidney function deteriorates; this can range from abnormal protein levels in the urine (albuminuria) to complete kidney failure with no urine production (anuria). Half of the patients in the "toxic phase" die within 10-14 days, the remainder recover without significant organ damage.

Yellow fever is difficult to recognise, especially during the early stages. It can easily be confused with malaria, typhoid, rickettsial diseases, haemorrhagic viral fevers (e.g. Lassa), arboviral infections (e.g. dengue), leptospirosis, viral hepatitis and poisoning (e.g. carbon tetrachloride). A laboratory analysis is required to confirm a suspect case. Blood tests (serology assays) can detect Yellow fever antibodies that are produced in response to the infection. Several other techniques

are used to identify the virus itself in blood specimens or liver tissue collected after death. These tests require highly trained laboratory staff using specialised equipment and materials.

Although an effective vaccine has been available for 60 years, the number of people infected over the last two decades has increased and Yellow fever is now a serious public health issue again.

Regions Affected

The virus is constantly present with low levels of infection (i.e. endemic) in some tropical areas of Africa and the Americas. This viral presence can amplify into regular epidemics. Until the start of this century, Yellow fever outbreaks also occurred in Europe, the Caribbean islands and Central and North America. Even though the virus is not thought to be present in these areas now, they must still be considered at risk for Yellow fever epidemics.

Thirty-three countries, with a combined population of 508 million, are at risk in Africa. These lie within a band from 15°N to 10°S of the equator. In the Americas, Yellow fever is endemic in nine South American countries and in several Caribbean islands. Bolivia, Brazil, Colombia, Ecuador and Peru are considered at greatest risk.

There are 200,000 estimated cases of Yellow fever (with 30,000 deaths) per year. However, due to underreporting, only a small percentage of these cases are identified. Small numbers of imported cases also occur in countries free of Yellow fever. Although Yellow fever has never been reported from Asia, this region is at risk because the appropriate primates and mosquitoes are present.

Transmission

Humans and monkeys are the principal animals to be infected. The virus is carried from one animal to another (horizontal transmission) by a biting mosquito (the vector). The mosquito can also pass the virus via infected eggs to its offspring (vertical transmission). The eggs produced are resistant to drying and lie dormant through dry conditions, hatching when the rainy season begins. Therefore, the mosquito is the true reservoir of the virus, ensuring transmission from one year to the next.

Several different species of the *Aedes* and *Haemogogus* (S. America only) mosquitoes transmit the Yellow fever virus. These mosquitoes are either domestic (i.e. they breed around houses), wild (they breed in the jungle) or semi-domestic types (they display a mixture of habits). Any region populated with these mosquitoes can potentially harbour the disease. Control programmes successfully eradicated mosquito habitats in the past, especially in South America. However, these programmes have lapsed over the last 30 years and mosquito populations have increased. This favours epidemics of Yellow fever.

Infection of Humans

There are three types of transmission cycle for Yellow fever: sylvatic, intermediate and urban. All three cycles exist in Africa, but in South America, only sylvatic and urban Yellow fever occur.

- *Sylvatic (or jungle) Yellow fever*: In tropical rainforests, Yellow fever occurs in monkeys that are infected by wild mosquitoes. The infected monkeys can then pass the virus onto other mosquitoes that feed on them. These infected wild mosquitoes bite humans entering the forest resulting in sporadic cases of Yellow fever. The majority of cases are young men working in the forest (logging, etc.). On occasion, the virus spreads beyond the affected individual.
- *Intermediate Yellow fever*: In humid or semi-humid savannahs of Africa, small-scale epidemics occur. These behave differently from urban epidemics; many separate villages in an area suffer cases simultaneously, but fewer people die from infection. Semi-domestic mosquitoes infect both monkey and human hosts. This area is often called the "zone of emergence", where increased contact between man and infected mosquito leads to disease. This is the most common type of outbreak seen in recent decades in Africa. It can shift to a

more severe urban-type epidemic if the infection is carried into a suitable environment (with the presence of domestic mosquitoes and unvaccinated humans).

- *Urban Yellow fever*: Large epidemics can occur when migrants introduce the virus into areas with high human population density. Domestic mosquitoes (of one species, *Aedes aegypti*) carry the virus from person to person; no monkeys are involved in transmission. These outbreaks tend to spread outwards from one source to cover a wide area.

Treatment

There is no specific treatment for Yellow fever. Dehydration and fever can be corrected with oral rehydration salts and paracetamol. Any superimposed bacterial infection should be treated with an appropriate antibiotic. Intensive supportive care may improve the outcome for seriously ill patients, but is rarely available in poorer, developing countries.

Vaccination

Vaccination is the single most important measure for preventing Yellow fever. In populations where vaccination coverage is low, vigilant surveillance is critical for prompt recognition and rapid control of outbreaks. Mosquito control measures can be used to prevent virus transmission until vaccination has taken effect.

Yellow fever vaccine is safe and highly effective. The protective effect (immunity) occurs within one week in 95% of people vaccinated. A single dose of vaccine provides protection for 10 years and probably for life. Over 300 million doses have been given and serious side effects are extremely rare. However, recently a few serious adverse outcomes, including deaths, have been reported in Brazil, Australia and the United States. Scientists are investigating the cause of these adverse events and monitoring to ensure detection of any similar incidents.

The risk to life from Yellow fever is far greater than the risk from the vaccine, so those who may be exposed to Yellow fever should be protected by immunisation. If there is no risk of exposure, for example, if a person will not be visiting an endemic area, there is no necessity to receive the vaccine. Since most of the other known side effects have occurred in children less than six months old, vaccine is not administered to this age group. The vaccine should only be given to pregnant women during vaccination campaigns in the midst of an epidemic.

Vaccination can be part of a routine preventive immunisation programme or can be done in mass "catch-up" campaigns to increase vaccination coverage in areas where it is low. The World Health Organization (WHO) strongly recommends routine childhood vaccination. The vaccine can be administered at age nine months, at the same time as the measles vaccine. Eighteen African nations have agreed to incorporate Yellow fever vaccine into their routine national vaccination programmes. This is more cost effective and prevents more cases (and deaths) than when emergency vaccination campaigns are performed to control an epidemic.

Past experience shows the success of this strategy. Between 1939 and 1952 Yellow fever cases almost vanished from French West Africa after intensive vaccination campaigns. Similarly, Gambia instituted mass routine vaccination after its 1979/1980 epidemic and later incorporated Yellow fever vaccine into its childhood immunisation programme. Gambia reported 85% vaccine coverage in 2000. No cases have been reported since 1980, yet the virus remains present in the environment.

To prevent an epidemic in a country, at least 80% of the population must have immunity to Yellow fever. This can only be achieved through the effective incorporation of Yellow fever into childhood immunisation programmes and the implementation of mass catch-up campaigns. The latter is the only way to ensure that coverage of all susceptible age groups is achieved and will prevent outbreaks from spreading. Very few countries in Africa have achieved this level to date.

Vaccination is highly recommended for travellers to high-risk areas. A vaccination certificate is required for entry to many countries, particularly for travellers arriving in Asia from Africa or South America. Fatal cases in unvaccinated tourists have been reported.

Surveillance

Because vaccination coverage in many areas is not optimal, prompt detection of Yellow fever cases and rapid response (emergency vaccination campaigns) are essential for controlling disease outbreaks. Improvement in Yellow fever surveillance is needed as evidenced by the gross underreporting of cases (estimates as to the true number of cases vary widely and have put the underreporting factor between three- and 250-fold). A surveillance system must be sensitive enough to detect and appropriately investigate suspect cases. This is facilitated by a standardised definition of possible Yellow fever cases, that is "acute fever followed by jaundice within two weeks of onset of symptoms, or with bleeding symptoms or with death within three weeks of onset". Suspect cases are reported to health authorities on a standardised case investigation form.

Ready access to laboratory testing is essential for confirming cases of Yellow fever, as many other diseases have similar symptoms. WHO has recently recommended that every at-risk country have at least one national laboratory where basic Yellow fever blood tests can be performed. Training programmes are being conducted and test materials are provided by WHO.

Given the likelihood that other cases have occurred (but have not been detected), one confirmed case of Yellow fever is considered to be an outbreak. An investigation team should subsequently explore and define the outbreak. This produces data for analysis, which guides the epidemic control committee in preparing the appropriate outbreak response (e.g. emergency vaccination programmes, mosquito control activities). This committee should also plan for the long term by implementing or strengthening routine childhood Yellow fever vaccination.

Future Outlook

Over the last 20 years the number of Yellow fever epidemics has risen and more countries are reporting cases. Mosquito numbers and habitats are increasing. In both Africa and the Americas there is a large susceptible, unvaccinated population. Changes in the world's environment, such as deforestation and urbanisation, have increased contact with the mosquito/virus. Widespread international travel could play a role in spreading the disease. The priorities are vaccination of exposed populations, improved surveillance and epidemic preparedness.

In March 1998, WHO held a technical consensus meeting in Geneva to identify obstacles to Yellow fever prevention and control. Priorities identified included: prevention through routine immunisation and preventive mass immunisation campaigns; detection, reporting and investigation of suspect cases; laboratory support; outbreak response; vaccine supply; and furthering research. Guidelines for investigation and control of Yellow fever outbreaks, and a background document reviewing topics of importance discussed at this meeting have been published.

Excerpts from http://www.who.int/mediacentre/factsheets/fs100/en/

3.1.2.6 Ross River & Barmah Forest Viruses

Traditionally known as Epidemic Polyarthritis, both Ross River (RR) and Barmah Forest (BF) disease are caused by viruses which are transmitted to humans through the bite of mosquitoes. A wide variety of symptoms may occur from rashes with fevers, to arthritis that can last from months to years with RR virus infection. There are no specific treatments; actions which reduce mosquito bites are the best form of prevention against these debilitating diseases.

RR disease is the most commonly transmitted mosquito-borne viral disease to humans in Australia. The number of cases has averaged >5,000 per annum during 1991-1997. The virus appears to be endemic in most rural areas, and there has been an increasing incidence near major cities. BF disease is less common, but the number of cases appears to be increasing annually, with several outbreaks occurring during the 1990's.

For most of Australia, peak incidence of the two diseases is through the summer and autumn months, particularly from January through to March, when the mosquito vectors are most abundant. However, in southwestern Australia and eastern Victoria, RR activity often begins in the spring months and peaks in early summer. Areas under intensive irrigation and localities close to saltmarshes are most productive for mosquito populations and hence tend to result in the highest number of human cases of disease. Outbreaks occur when local conditions of rainfall, tides and temperature promote vector abundance.

Serological studies and laboratory investigations have indicated that native mammals, most likely kangaroos and wallabies, are natural hosts for RR virus but little is known about the hosts of Barmah Forest virus.

RR virus transmission from human to mosquito to human (thus occurring without the involvement of an animal) has been proposed, and there is now little doubt that such a cycle involving only humans and mosquitoes occurs during periods of intense virus activity.

RR and BF viruses have been isolated from many mosquito species, indicating wide susceptibility among mosquitoes. In inland regions the major vector is *Culex annulirostris* which breeds in freshwater habitats, especially in irrigated areas. Along coastal regions saltmarsh mosquitoes represent the major threat, including *Aedes (Ochlerotatus) vigilax* and *Ae. camptorhynchus* in northern and southern coastal regions respectively. There is some evidence that 'floodwater' *Aedes* species such as *Ae. normanensis* play an important role in transmission in inland regions following heavy rains or floods, and *Coquillettidia linealis* is a secondary vector in areas with established wetlands. In the domestic urban situation there is evidence to suggest that *Ae. notoscriptus* may be a vector, while *Cx. quinquefasciatus* is not.

Symptoms

Human infection with RR virus or BF virus may result in the clinical condition known as polyarthritis. The effects range from: a symptom-less condition; through a transient rash (Figure 3.1.10) and mild illness with fever; to polyarthritis affecting chiefly the ankles, fingers, knees, and wrists, but other joints may be affected.

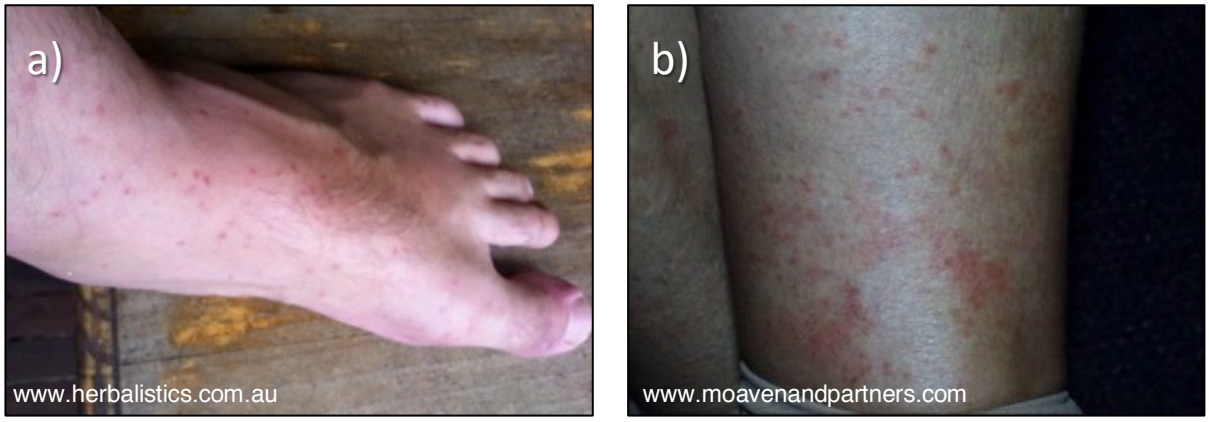


Figure 3.1.10. Rush due to a) RRV and b) BFV

The disease is not fatal. For RR virus symptoms become evident from 3-21 days (average 9 days) after infection, and mild cases may recover in less than one month but many persist for months to years. Recent studies have indicated that the rash may be more florid with BF virus infections

but that the arthritic symptoms are greater with RR virus infection (Figure 3.1.10). People of working age are most likely to be afflicted with the diseases, whilst symptoms are rare in children.

A variety of blood tests are used to demonstrate the presence of specific antibodies to RR and BF virus. Blood samples should be taken during the acute and convalescent phases of the illness and a four-fold rise in antibody levels will confirm the clinical diagnosis.

Specific therapies do not exist to treat the diseases, rather it is the symptoms that are alleviated. This includes various analgesics to reduce the pain and fevers and anti-inflammatory agents for the arthritic symptoms.

Excerpts from http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/viruses/rossriverbarmahforest.htm

3.1.2.7 Chikungunya

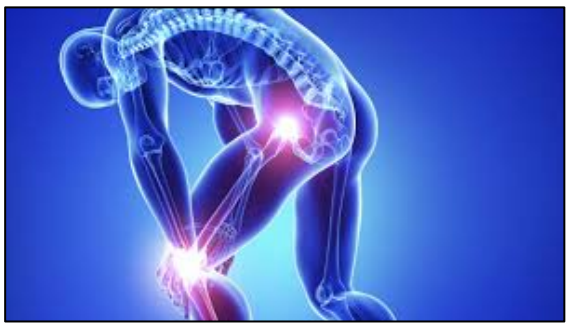


Figure 3.1.11. Severe joint and muscle pain produced by the Chikungunya virus.

Chikungunya fever is an RNA virus that belongs to the alphavirus genus of the family Togaviridae transmitted to humans by infected mosquitoes that is characterised by fever, headache, rash, and severe joint and muscle pain (Figure 3.1.11). The name chikungunya, which means “that which bends up,” is derived from the Kimakonde language of the Makonde people. This African tribe lives on the eastern border between Mozambique and Tanzania, where chikungunya virus was first detected in an epidemic that occurred in 1952–53. The disease was given its name because severe musculoskeletal pain caused affected persons to walk in a stooped posture.

Transmission

The virus is transmitted from human to human by the bites of infected female mosquitoes. Two important vectors are *Aedes aegypti* and *Aedes albopictus*, which also transmit dengue virus. These mosquitoes can be found biting throughout daylight hours, with peaks of activity in the early morning and late afternoon. Both species are found biting outdoors, but *Ae. aegypti* will also readily feed indoors. After the bite of an infected mosquito, onset of illness occurs usually between four and eight days but can range from two to 12 days. The proximity of mosquito breeding sites to human habitation is a significant risk factor for chikungunya.

Symptoms

Symptoms appear between 4 and 7 days after the patient has been bitten by the infected mosquito and these include:

- High fever (40°C/ 104°F)
- Joint pain and swelling (lower back, ankle, knees, wrists or phalanges)
- Rash
- Headache
- Muscle pain
- Nausea, Fatigue

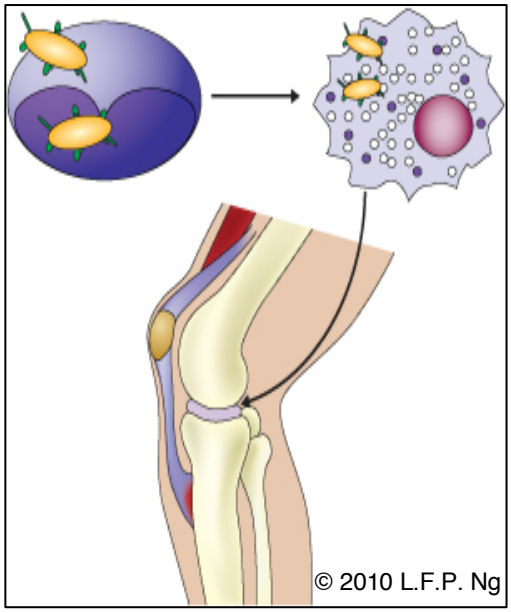


Figure 3.1.12. Chikungunya viruses (yellow) may hitch a ride to joints where they can cause ongoing joint pain long after the virus has been cleared from the blood.

Chikungunya is characterised by an abrupt onset of fever frequently accompanied by joint pain. The joint pain is often very debilitating, but usually lasts for a few days or may be prolonged to weeks. Most patients recover fully, but in some cases joint pain may persist for several months, or even years (Figure 3.1.12).

The disease shares some clinical signs with dengue and often symptoms in infected individuals are mild and the infection may go unrecognised, or be misdiagnosed in areas where dengue occurs.
It is rarely life-threatening however widespread occurrences can cause substantial morbidity and economic loss. Occasional cases of gastrointestinal complaints, eye, neurological and heart complications have been reported, as well as gastrointestinal complaints. Serious complications are not common, but in older people are at increased risk for severe symptoms and death, as well as new-borns, and people with medical conditions such as high blood pressure, diabetes, or heart disease. Recovery from an infection will confer life-long immunity.

Treatment

There is no cure for the disease – neither specific antiviral drugs nor commercial vaccine. Treatment is focused on relieving the symptoms, particularly the joint pain using anti-pyretics, optimal analgesics and fluids. Most patients feel better within a week as symptoms are generally self-limiting and last for 2–3 days but some people may develop longer-term joint pain high blood pressure, diabetes, or heart disease. The virus remains in the human system for 5-7 days and mosquitoes feeding on an infected person during this period can also become infected.

Diagnosis

Several methods can be used for diagnosis. Serological tests, such as enzyme-linked immunosorbent assays (ELISA), may confirm the presence of IgM and IgG anti-chikungunya antibodies. IgM antibody levels are highest three to five weeks after the onset of illness and persist for about two months. Samples collected during the first week after the onset of symptoms should be tested by both serological and virological methods (RT-PCR).

The virus may be isolated from the blood during the first few days of infection. Various reverse transcriptase–polymerase chain reaction (RT–PCR) methods are available but are of variable sensitivity. Some are suited to clinical diagnosis. RT–PCR products from clinical samples may also be used for genotyping of the virus, allowing comparisons with virus samples from various geographical sources.

Distribution

Chikungunya has been identified in nearly 40 countries in Asia, Africa, Europe and also in the Americas. The virus occurs in sub-Saharan Africa, south-east Asia and tropical areas of the Indian sub-continent, as well as islands in the south-western Indian Ocean. In recent decades mosquito vectors of chikungunya have spread to Europe and the Americas. In 2007, disease transmission was reported for the first time in a localised outbreak in north-eastern Italy.

Countries having documented, endemic, or epidemic chikungunya are:
Asia: Human chikungunya virus infection has been documented in Cambodia, East Timor, India, Indonesia, Laos, Malaysia, Maldives, Myanmar, Pakistan, Philippines, Réunion, Seychelles, Singapore, Taiwan, Thailand and Vietnam.

Africa: Chikungunya occurs in Benin, Burundi, Cameroon, Central African Republic, Comoros, Congo (DRC), Equatorial Guinea, Guinea, Kenya, Madagascar, Malawi, Mauritius, Mayotte, Nigeria, Senegal, South Africa, Sudan, Tanzania, Uganda and Zimbabwe.
Europe and the Americas: Aside from minor incidence rates caused by imported cases from travellers, Italy is the only European country which has had an outbreak. The Americas have not had any major outbreaks so far.

Major Outbreaks
This disease has been classified as re-emerging or spreading in recent years with a number of outbreaks occurring in often virgin territories. Human infections in Africa have been at relatively low levels for a number of years, but in 1999-2000 there was a large outbreak in the Democratic Republic of the Congo.

Chikungunya was first identified in Tanzania in the early 1952 and has caused periodic outbreaks in Asia and Africa since the 1960s.

Outbreaks are often separated by periods of more than 10 years. Since 2004, chikungunya fever has reached epidemic proportions, with considerable morbidity and suffering.

Starting in February 2005, a major outbreak of chikungunya occurred in islands of the Indian Ocean. A large number of imported cases in Europe were associated with this outbreak, mostly in 2006 when the Indian Ocean epidemic was at its peak. A large outbreak of chikungunya in India occurred in 2006 and 2007. Several other countries in South-East Asia were also affected. Since 2005, India, Indonesia, Maldives, Myanmar and Thailand have reported over 1.9 million cases. In 2007 transmission was reported for the first time in Europe, in a localized outbreak in north-eastern Italy. There were 197 cases recorded during this outbreak and it confirmed that mosquito-borne outbreaks by *Ae. albopictus* are plausible in Europe.

In December 2013, France reported 2 laboratory-confirmed autochthonous cases in the French part of the Caribbean island of St Martin. Since then, local transmission has been confirmed in over 43 countries and territories in the WHO Region of the Americas. This is the first documented outbreak of chikungunya with autochthonous transmission in the Americas. As of April 2015, over 1 379 788 suspected cases of Chikungunya have been recorded in the Caribbean islands, Latin American countries, and the United States of America. 191 deaths have also been attributed to this disease during the same period. Canada, Mexico and USA have also recorded imported cases.

On 21 October 2014, France confirmed 4 cases of locally-acquired chikungunya infection in Montpellier, France. In late 2014, outbreaks were reported in the Pacific islands. Currently chikungunya outbreak is ongoing in Cook Islands and Marshall Islands, while the number of cases in American Samoa, French Polynesia, Kiribati and Samoa has reduced. WHO responded to small outbreaks of chikungunya in late 2015 in the city of Dakar, Senegal, and the state of Punjab, India.

In the Americas in 2015, 693 489 suspected cases and 37480 confirmed cases of chikungunya were reported to the Pan American Health Organization (PAHO) regional office, of which Colombia bore the biggest burden with 356 079 suspected cases. This was less than in 2014 when more than 1 million suspected cases were reported in the same region.

In 2016 there was a total of 349 936 suspected and 146 914 laboratory confirmed cases reported to the PAHO regional office, half the burden compared to the previous year. Countries reporting most cases were Brazil (265 000 suspected cases), Bolivia and Colombia (19 000 suspected cases, respectively). 2016 is the first time that autochthonous transmission of chikungunya was reported in Argentina following an outbreak of more than 1 000 suspected cases. In the African

region, Kenya reported an outbreak of chikungunya resulting in more than 1 700 suspected cases. In 2017, Pakistan continues to respond to an outbreak which started in 2016 (Figure 3.1.1.3).

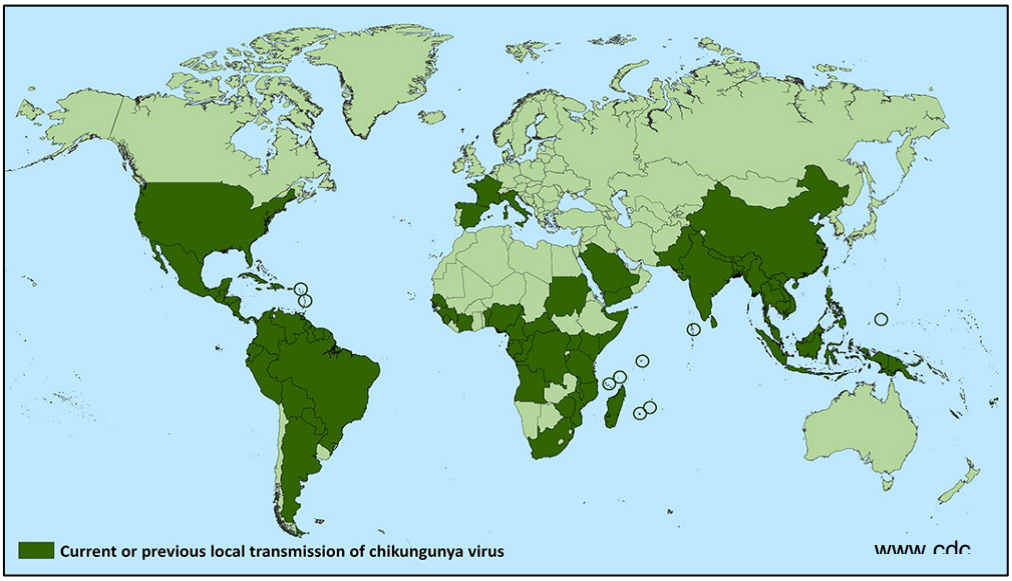


Figure 3.1.13. Countries and territories where chikungunya cases have been reported *(as of May 29, 2018).* Does not include countries or territories where only imported cases have been documented.

Resources
http://www.who.int/mediacentre/factsheets/fs327/en/
http://www.cdc.gov/chikungunya/
http://www.britannica.com/EBchecked/topic/1159461/chikungunya-fever
http://www.paho.org/hq/index.php?option=com_topics&view=article&id=343&Itemid=40931
http://www.ecdc.europa.eu/en/healthtopics/chikungunya_fever/pages/index.aspx
http://www.tropeninstitut.de/krankheiten/krankheit.php?kid=40
http://carpha.org/What-We-Do/Public-Healt

3.1.2.8 Other Diseases

Mosquito-transmitted viral diseases causing brain inflammation/encephalitis including virus that have been previously discussed but also:

- Murray Valley encephalitis virus (MVEV) and Kunjin
- Eastern equine encephalitis virus (EEE)
- Western Equine Encephalitis
- La Crosse Encephalitis
- St. Louis Encephalitis
- Powassan Encephalitis
- Venezuelan Equine Encephalitis

***Phlebovirus* in the family Bunyaviridae**

Rift Valley fever (RVF), which is of the *Phlebovirus* type, is an acute, fever-causing viral disease that affects domestic animals (such as cattle, buffalo, sheep, goats, and camels) and humans. It is spread by either touching infected animal blood, breathing in the air around an infected animal being butchered, drinking raw milk from an infected animal, or the bite of infected mosquitoes. There is a human vaccine; however, as of 2010 it is not widely available. There is no specific treatment and medical efforts are supportive. RVF is most commonly associated with mosquito-borne epidemics during years of unusually heavy rainfall.

3.2. Sandflies

Sand flies transmit viral and bacterial diseases, as well as protozoans. Sand fly-borne viral fevers include sand fly fever, changuinola fever, and vesicular stomatitis. However, phlebotomines are most widely known as vectors of Leishmania, causing cutaneous, mucocutaneous or visceral leishmaniasis.

Detailed data is only available for Leishmania, not for viral or bacterial infections.

3.2.1 Leishmaniasis

This disease is also known as Leichmaniosis, Leishmaniose and formerly, Orient Boils, Baghdad Boil, kala azar, black fever, sand fly disease, Dum-Dum fever or espundia.

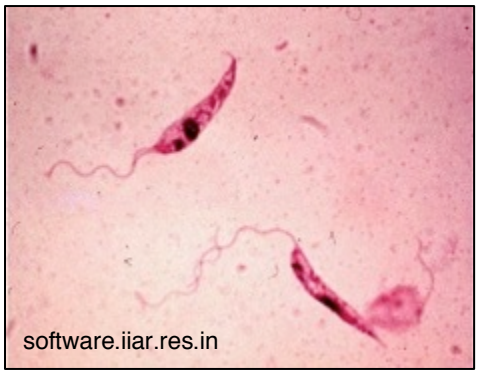


Figure 3.2.1. *Leishmania* sp.

Leishmaniasis is a vector-borne disease caused by obligate intracellular protozoa of the genus *Leishmania* and is transmitted by sand flies (Figure 3.2.1).

Human infection is caused by about 21 of the 30 species that infect mammals. These include the *L. donovani* complex with 3 species (*L. donovani, L. infantum,* and *L. chagasi*); the *L. mexica*na complex with 3 main species (*L. mexicana, L. amazonensis,* and *L. venezuelensis); L. tropica; L. major; L. aethiopica*; and the subgenus *Viannia* with 4 main species (*L. (V.) braziliensis, L. (V.) guyanensis, L. (V.) panamensis*, and *L. (V.) peruviana).*

The different species are morphologically indistinguishable, but they can be differentiated by isoenzyme analysis, molecular methods, or monoclonal antibodies. Leishmaniasis is transmitted by the bite of infected female phlebotomine sand flies. The sand flies inject the infective stage (i.e. promastigotes) from their proboscis during blood meals (Figure 3.2.2).

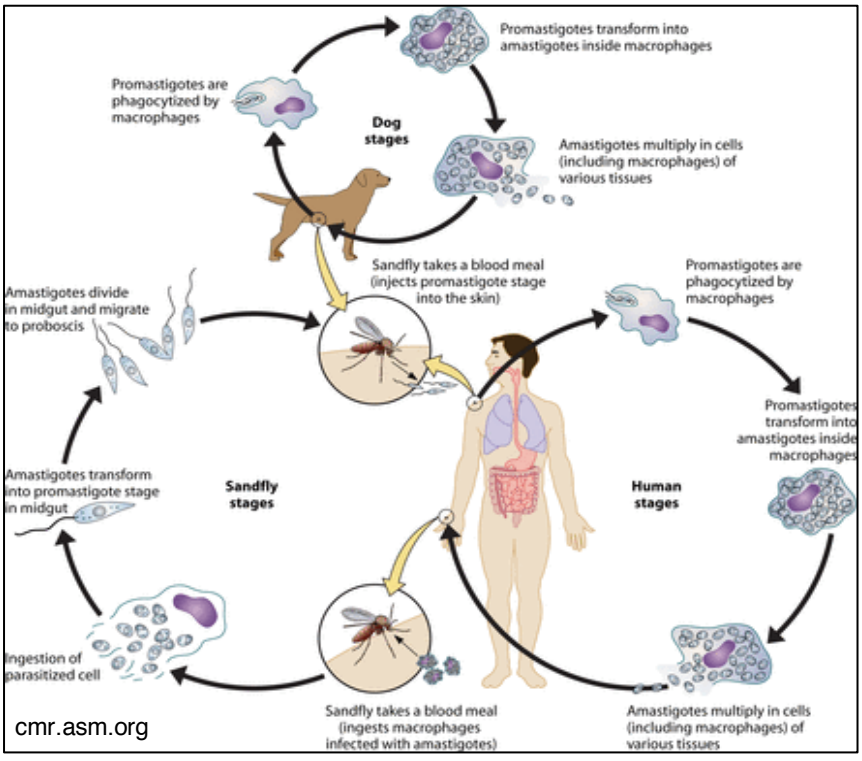


Figure 3.2.2. The life cycle of *Leishmania* species.

Promastigotes are phagocytosed by resident phagocytes, transform into tissue-stage amastigotes, and multiply within these cells through simple division. The parasite continues to infect phagocytic cells either at the site of cutaneous infection or in secondary lymphoid organs, with eventual parasitemia.

Parasite, host, and other factors affect whether the infection becomes symptomatic and whether cutaneous or visceral leishmaniasis results.

Sandflies become infected through feeding on a host either with an active skin lesion in cutaneous leishmaniasis or with parasitemia in visceral leishmaniasis. Parasites convert to promastigotes within the sandfly midgut. Promastigotes migrate from the midgut and transform into highly infectious metacyclic promastigotes.

In sandflies amastigotes transform into promastigotes, develop in the gut (in the hindgut for leishmanial organisms in the *Viannia* subgenus; in the midgut for organisms in the Leishmania subgenus) and migrate to the proboscis.

Distribution

Leishmaniasis is found in parts of about 88 countries. Approximately 350 million people live in these areas. Most of the affected countries are in the tropics and sub-tropics, and habitats range from rain forests in Central and South America to deserts in West Asia. This disease is found in Mexico, Central America, and South America - from northern Argentina to Texas (not in Uruguay, Chile, or Canada), southern Europe (leishmaniasis is not common in travellers to southern Europe), Asia (not Southeast Asia), the Middle East, and Africa (particularly East and North Africa, with some cases elsewhere). More than 90% of the world's cases of visceral leishmaniasis are in India, Bangladesh, Nepal, Sudan, and Brazil.

Symtoms

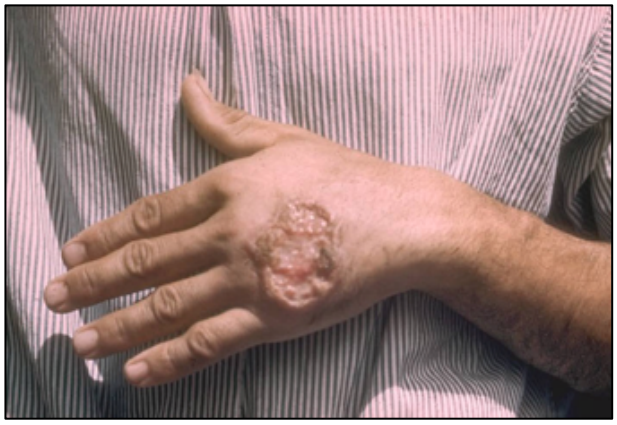


Figure 3.2.3. Cutaneous leishmaniasis

Human leishmanial infections can result in two main forms of disease: cutaneous leishmaniasis (Figure 3.2.3) and visceral leishmaniasis (kala-azar, Figure 3.2.4). The factors determining the form of disease include leishmanial species, geographic location and immune response of the host.

Cutaneous leishmaniasis is characterised by one or more cutaneous lesions on areas where sand flies have fed (Figure 3.2.3).

People who have cutaneous leishmaniasis have one or more sores on their skin. The sores can change in size and appearance over time. They often end up looking somewhat like a volcano, with a raised edge and central crater. A scab covers some sores. The sores can be painless or painful. Some people have swollen glands near the sores (for example, in the armpit if the sores are on the arm or hand).

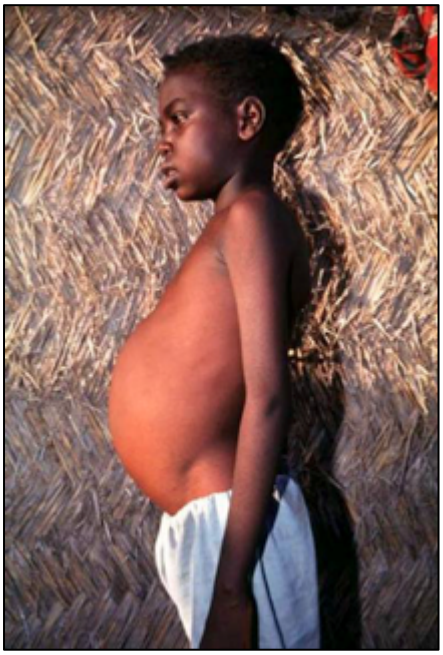


Figure 3.2.4. Visceral leishmaniasis.

Visceral leishmaniasis is a severe form in which the parasites have migrated to the vital organs (Figure 3.2.4). People who have visceral leishmaniasis usually have fever, weight loss and an enlarged spleen and liver (usually the spleen is bigger than the liver). Some patients have swollen glands. Certain blood tests are abnormal, for example, patients usually have low blood counts, including a low red blood cell count (anaemia), low white blood cell count and low platelet count. Some patients develop post kala-azar dermal leishmaniasis. Visceral leishmaniasis is becoming an important opportunistic infection in areas where it coexists with HIV.

Excerpts from
http://www.dpd.cdc.gov/dpdx/HTML/Leishmaniasis.htm
http://pathmicro.med.sc.edu/parasitology/blood-proto.htm.

3.2.2 Sandfly (*Phlebotomus*) Fever

Sandfly fever is a disease of considerable military importance because, although it is never fatal, it may nevertheless suddenly incapacitate large numbers of troops who may be urgently required for important operations.

It has long been endemic in the Mediterranean area and persists chiefly in the lowlands of those tropical and sub-tropical countries which have long periods of hot dry weather. The native population seems to be immune, probably because of infection in childhood. Newcomers, however, usually succumb to the disease during the first epidemic.

Sandfly fever is due to a small virus which is present in the blood of a patient from 24 hours before till 24 hours after the onset of the disease. The virus can be readily transferred to volunteers by intravenous or intracutaneous injection. It has not so far been transmitted to other animals. In nature the disease is transmitted by the bite of the female sand fly and there is no evidence that any other insect is implicated in transmission.

Following an incubation period of 3-6 days the disease has a rapid onset with fever for between 2-4 days. Occasional cases may be febrile for longer periods. Other symptoms include severe headaches, malaise and general aching of the limbs and back. Many patients complain of stiffness in the neck and occipital pain. After the fever has abated weakness, depression and loss of appetite may persist with resulting weight loss

3.2.3 Other Diseases

Additional disease organisms transmitted by sand flies include:

Chandipura virus - causes fever, symptoms similar to those of flu and acute encephalitis (inflammation of the brain). Chandipura virus was first isolated in 1965 in a village in Maharashtra State, India. Since then the virus has been reported in adjoining states in central India. The likely vector of the virus is the female phlebotomine sand fly. The virus has been detected in sand flies in Senegal and Nigeria, as well as in India. In 2003 Chandipura virus was responsible for an outbreak in southern India in which 329 children developed acute encephalitis and 183 died. The disease progressed rapidly from an influenza-like illness to coma and death. Chandipura virus is a member of the Vesiculovirus genus of the family Rhabdoviridae. This virus should be considered as an important emerging pathogen.

Carrion's disease (Oroya fever or Peruvian Wart) - is a rare infectious disease found only in Peru, Ecuador, and Colombia. It is endemic in some areas of Peru and is caused by infection with the bacterium *Bartonella bacilliformis* and transmitted by sand flies of genus *Lutzomyia*.

Toscana virus - occurs in the northern and western Mediterranean, while Chagres and Punta Toro viruses are found in the New World.

3.3. Fleas

Fleas are capable of transmitting pathogens that cause disease in humans and other animals.

3.3.1 Plague

Figure 3.3.1. *Yersinia pestis*.

Plague is an infectious disease of humans and other vertebrates caused by the bacterium *Yersinia pestis* (Figue 3.3.1). People usually get plague after being bitten by a flea that is carrying the plague bacterium or more rarely by handling an infected animal. Since fleas bite both people and domestic animals, especially cats and rodents, an infected flea can pass plague to animals or people.

Millions of people in Europe died from plague in the Middle Ages, when human homes and places of work were inhabited by flea-infested rats. Today, modern antibiotics are effective against plague but if an infected person is not treated promptly the disease is likely to cause illness or death.

There are several forms of plague: Pneumonic, Bubonic and Septicemic. Depending on circumstances these forms may occur separately or in combination. People usually show symptoms 2-6 days after being infected. Symptoms include fever, chills, weakness, and swollen and painful lymph nodes. A few people get pneumonia as a first symptom of plague. The infection then spreads to other parts of the body. If this disease is not treated early, it is often fatal.

Pneumonic plague occurs when *Y. pestis* infects the lungs. This type of plague can spread from person to person through the air. Transmission can take place if someone breathes in aerosolised bacteria, which could happen in a bioterrorist attack.

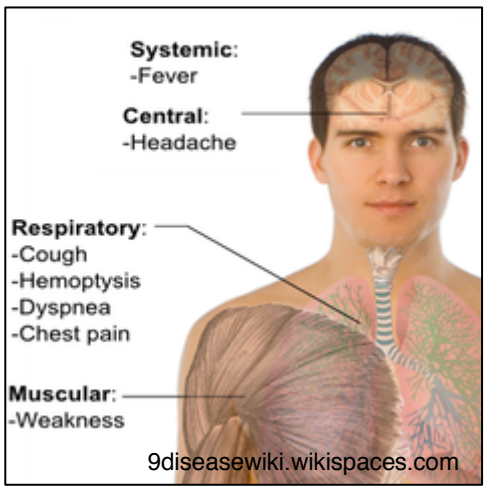


Figure 3.3.2. Main symptoms of Pneumonic plague.

Pneumonic plague is also spread by breathing in *Y. pestis* suspended in respiratory droplets from a person (or animal) with pneumonic plague. Becoming infected in this way usually requires direct and close contact with the ill person or animal. Pneumonic plague may also occur if a person with bubonic or septicemic plague is untreated and the bacteria spread to the lungs.

Bubonic plague is the most common form of plague. This occurs when an infected flea bites a person or when materials contaminated with *Y. pestis* enter through a break in a person's skin. Patients develop swollen, tender lymph glands (called buboes, Figure 3.3.3) and fever, headache, chills, and weakness. Bubonic plague does not spread from person to person.

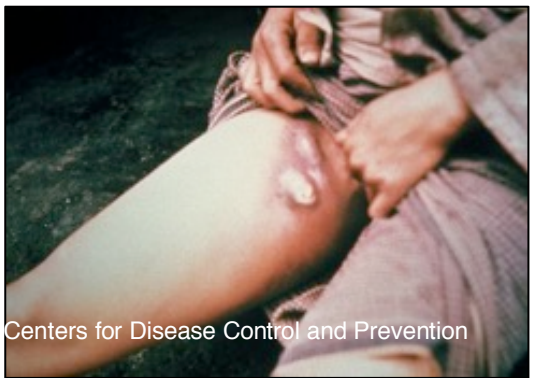


Figure 3.3.3. Bubonic plague.

Septicemic plague occurs when plague bacteria multiply in the blood. (Figure 3.3.4.) It can be a complication of pneumonic or bubonic plague or it can occur by itself. When it occurs alone, it is caused in the same ways as bubonic plague; however, buboes do not develop. Patients have fever, chills, prostration, abdominal pain, shock, and bleeding into skin and other organs. Septicemic plague does not spread from person to person.

Figure 3.3.4. Septic plague.

Figure 3.3.5. Rats killed during a clean-up in Dunedin during the 1900's Plague outbreak.

Wild rodents in certain areas around the world are currently infected with plague. Outbreaks in people still occur in rural communities or in cities. They are usually associated with infected rats and rat fleas that live in the home.

In the United States the last urban plague epidemic occurred in Los Angeles in 1924-25. Since then human plague in the United States has occurred as mostly scattered cases in rural areas (an average of 10 to 15 persons each year).

Globally the World Health Organization reports 1,000 to 3,000 cases of plague every year. In North America plague is found in certain animals and their fleas from the Pacific Coast to the Great Plains, and from southwestern Canada to Mexico. Most human cases in the United States occur in two regions: 1) northern New Mexico, northern Arizona, and southern Colorado; and 2) California, southern Oregon, and far western Nevada. Plague also exists in Africa, Asia, and South America (Figure 3.3.6).

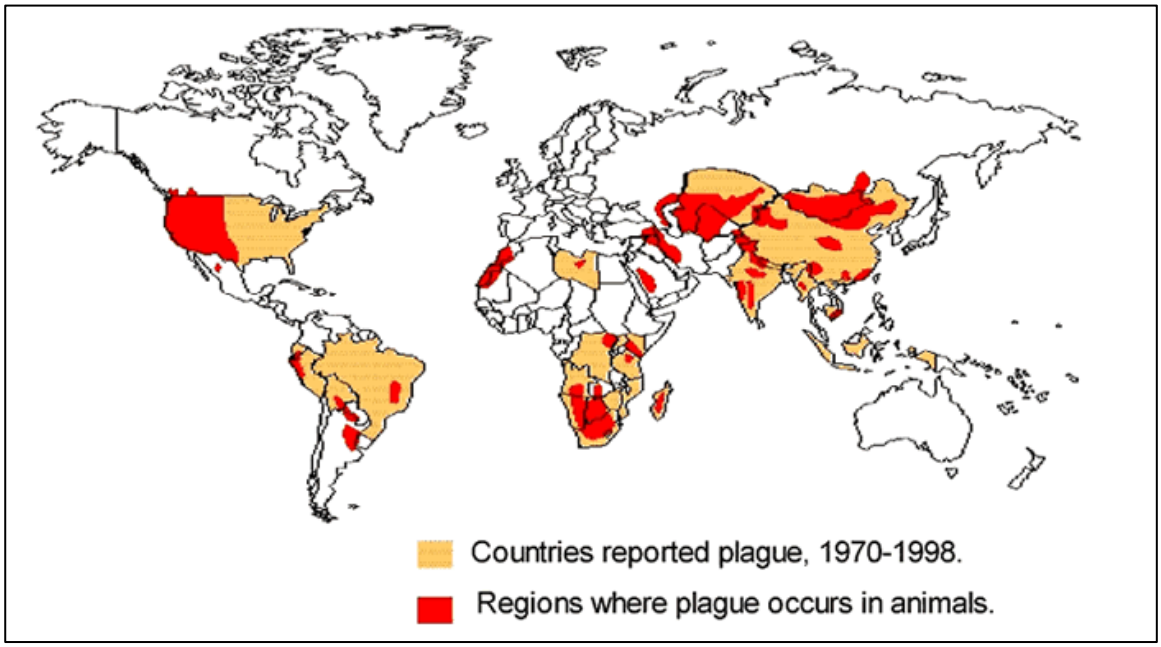


Figure 3.3.6. World distributin of Plague, 1998

3.3.2 Dipylidiasis

As mentioned earlier (section 2.3), *Ctenocephalides* flea species are an intermediate host for the tapeworm *Dipylidium caninum* that infects dogs, cats and occasionally humans, and causes Dipylidiasis. *D. caninum* is the most common tapeworms in dogs. Like all tapeworms, it requires an intermediate host and in this case it is the flea.

Figure 3.3.1. Adult tapeworm.

The adult tapeworm is flat, segmented and can be over several metres long (Figure 3.3.1). It is attached to the small intestine by suckers and has its own reproductive organs. The eggs are produced in the lower segments of the worm which break off and pass out with the faeces, containing the eggs. The intermediate stage once ingested by the dog can become an egg producing adult in 2-3 weeks.

Because human infection is the result of ingestion of infected dog or cat fleas, it occurs more often in children who kiss or are licked by their infected pets. Human infections exist worldwide, having been reported in Europe, the Philippines, China, Japan, Argentina and the United States.

Most infections with *Dipylidium caninum* are asymptomatic. Pets may exhibit behaviour to relieve anal pruritis (such as scraping anal region across grass or carpeting). Mild gastrointestinal disturbances may occur. The most striking feature in animals and children consists of the passage of proglottids (tape worm segments). The proglottids are motile when freshly passed and may be mistaken for maggots or fly larvae. Only high numbers appear to produce problems such as loss of appetite, weight loss and diarrhoea. Diagnosis is by the presence of rice like segments or eggs in the faeces.

Gravid proglottids are passed intact in the faeces or emerge from the perianal region of the host. Subsequently they release typical egg packets. On rare occasions, proglottids rupture and egg packets are seen in stool samples. Following ingestion of an egg by the intermediate host (larval stages of the dog or cat flea *Ctenocephalides* spp.), an oncosphere is released into the flea's

intestine. The oncosphere penetrates the intestinal wall, invades the insect's hemocoel (body cavity), and develops into a cysticercoid larva.

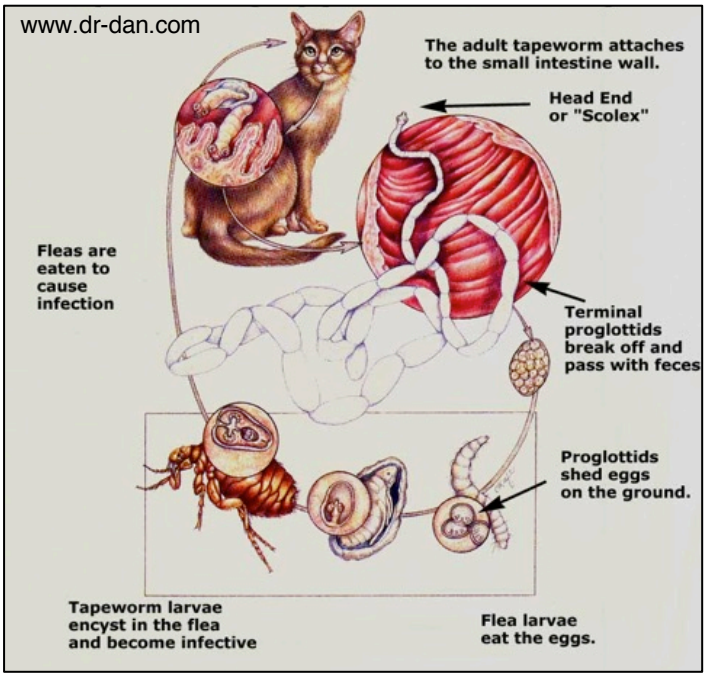


Figure 3.3.8. Tapeworm (*Dipylidium caninum*) life cycle.

The larva develops into an adult, and the adult flea harbours the infective cysticercoid. The vertebrate host becomes infected by ingesting the adult flea containing the cysticercoid. The dog is the principal definitive host for *Dipylidium caninum*. Other potential hosts include cats, foxes and humans (mostly children).

Humans acquire infection by ingesting the cysticercoid contaminated flea. This can be promulgated by close contact between children and their infected pets. In the small intestine of the vertebrate host the cysticercoid develops into the adult tapeworm which reaches maturity about 1 month after infection.

The adult tapeworms (measuring up to 60 centimetres in length and 3 millimetres in width) reside in the small intestine of the host, where they each attach by their scolex. They produce proglottids (or segments) which have two genital pores (hence the name "double-pored" tapeworm). The proglottids mature, become gravid, detach from the tapeworm, and migrate to the anus or are passed in the stool.

3.3.3 Other Diseases

The rabbit flea spreads the myxomatosis virus within rabbit populations, and the Oriental rat flea is the primary vector of *Yersinia pestis*, the bacterial pathogen for bubonic plague.

Despite the large number of flea species there are no known flea-borne arboviruses.

3.4. Lice

Lice can be of both direct and indirect importance with regard to humans. For example, human body lice carry and spread bacteria that cause the diseases: louse-borne typhus, trench fever, and louse-borne relapsing fever – indirect importance. They also cause pediculosis (see section 3.4.4) - direct importance.

Although chewing lice have little involvement in human disease one species, the common dog louse (*Trichodectes canis,* Figure 3.4.1), serves as an intermediate host of the dog tapeworm *Dipylidium caninum* which can be transmitted to humans. Some chewing lice species are known to transmit pathogens to birds and others are suspected to transmit pathogens to wild mammals.

Figure 3.4.1. Common dog louse.

Four families of sucking lice contain species that are of direct or indirect importance to humans. Examples of lice of direct importance include: the body louse, the head louse and the crab or pubic louse. Of indirect importance to humans are certain rodent-infesting sucking lice which are vectors of zoonotic pathogens.
Serious louse infestations commonly occur among the homeless, or persons in refugee camps and other crowded conditions that result from war and natural disasters.

Lice also infest domesticated mammals and poultry. Infested animals and louse control cost farmers and breeders hundreds of millions of dollars every year in lost production and the purchase of expensive chemical controls. For example, infested chickens will lay fewer eggs, resulting in less money earned by poultry farmers.

3.4.1 Louse-borne relapsing Fever

Also known as epidemic relapsing fever. This disease is caused by infection with the spirochaete bacterium *Borrelia recurrentis*, vectored by body lice. It is most common in Asia, Africa, and Central and South America. The lice become infected after feeding on an infectious person. No other animals are affected by this disease.

After ingestion by the louse, some spirochaetes pass through the gut wall and colonise the haemocoel where they multiply into huge populations. They are effectively trapped in the louse and the only way they can be transmitted to another person is by crushing the louse on the skin and causing a small abrasion through which the spirochaetes can enter the body.

Symptoms include head and muscle aches, nausea, anorexia, dizziness, coughing, vomiting, decrease in blood platelets and abrupt onset of fever. The most characteristic symptom is the presence of afebrile periods followed by periods of fever. These relapses usually occur 2-5 times before the disease dissipates. In severe infections, the liver and spleen become swollen, breathing becomes painful and the patient typically lies prostrate, shaking and taking shallow breaths. Mortality in untreated cases 5-40%. Antibiotics combat this disease.

Louse-borne relapsing fever occurs in epidemics amid poor living conditions, famine and war in the developing world. Historically this disease was responsible for 5 million deaths in eastern Europe and Russia during an epidemic lasting from 1919-1923.

3.4.2 Louse-borne Typhus

Also known as jail fever, epidemic typhus and exanthematic typhus. This disease is caused by the rickettsial bacterium *Rickettsia prowazekii* and is vectored by body lice which become infected after feeding on an infectious person. Rickettsiae ingested by the louse colonise the cells that line the gut, replicate and burst free into the gut. Some are then voided through the louse's faeces, which are typically deposited on the host while the louse is feeding and are able to penetrate the skin when the bite site is scratched by the host and infection begins. Infectious rickettsiae can remain viable in louse faeces for up to 30 days and it has been suggested that aerial transmission may also be possible.

Symptoms usually appear 10-14 days after the initial exposure: malaise, muscle aches, headaches, coughing, rapid onset of fever and a blotchy rash on the chest or abdomen. In severe cases, the rash will cover much of the host body. Later-stage symptoms in untreated cases include: delirium, prostration (total exhaustion or weakness), low blood pressure and coma which may result in death. Fatalities are usually 10-20% but can be up to 50% in untreated outbreaks. Prompt antibiotic administration is usually curative.

Several forms of epidemic typhus have been recognised in addition to the classic case outlined above:

- Recrudescent typhus or Brill-Zinsser disease is a recurrence of the disease after individuals were infected months or years previously. Infectious rickettsiae can remain dormant in the human tissues after the host has recovered from the initial bout of the disease, and later cause a second bout of disease in the presence or absence of lice. Intervals as great as 30 years have been recorded. If a patient experiencing a bout of recrudescent typhus also became infested with body lice, the lice could become infected during blood feeding and transfer it to other individuals initiating an outbreak.
- North American flying squirrels also harbour a zoonotic strain of this bacterium which is infectious to humans. The exact mode of transmission is unknown however, as the principle vector the flying squirrel louse, *Neohaematopinus sciuropteri,* doesn't bite humans. Flying squirrels often colonise attics or eaves of houses and it has been suggested that humans might become infected when frequenting these areas by inhaling aerosolised rickettsiae from infectious louse faeces.

Louse-borne typhus is relatively rare today, but persists in some parts of the world including United States, Russia, Asia, Africa, Central and South America. A recently outbreak in refugee camps in Burundi in 1996-7 was the largest epidemic of this disease since World War II and may have involved as many as 500,000 people. This demonstrates that although this disease appears to be less prevalent, it has the potential to emerge rapidly under certain conditions.

3.4.3 Trench Fever

Figure 3.4.2. Trench fever – homeless person's leg.

This disease is also known as 5-day fever or wolhynia and is caused by the bacterium *Bartonella quintana*. Body lice become infected with this bacterium when feeding on the blood of an infectious person who may or may not show clinical symptoms. The bacterium invades the midgut of the louse, replicates and is eventually voided in the faeces. It is transmitted to a new host when the faeces are scratched into the skin. Infection ranges from asymptomatic through mild to severe, but death is a rare outcome

This disease was unknown prior to World War I when in 1916 more than 200,000 (British cases – other countries over and above this number)

European troops engaged in trench warfare presented with symptoms including headache, muscle ache, fever and nausea, with disease episodes alternating with afebrile periods. The disease agent was identified, body lice implicated and then the disease virtually disappeared until World War II when it resurfaced in troops under similar conditions.

Trench fever is generally considered rare today, although it has been recorded recently as an opportunistic infection of homeless, chronic alcoholic or immuno-compromised individuals. In these cases, it manifests as mainly vascular tissue lesions, chronically swollen lymph nodes and endocarditis (inflammation of the heart valves) and has been called urban trench fever.

3.4.4 Pediculosis

Infestation by lice is termed pediculosis:

- body lice - *Pediculus corporis*
- head lice - *Pediculus capitis*
- crab (pubic) lice – *Pediculus inguinalis*

Pediculus corporis

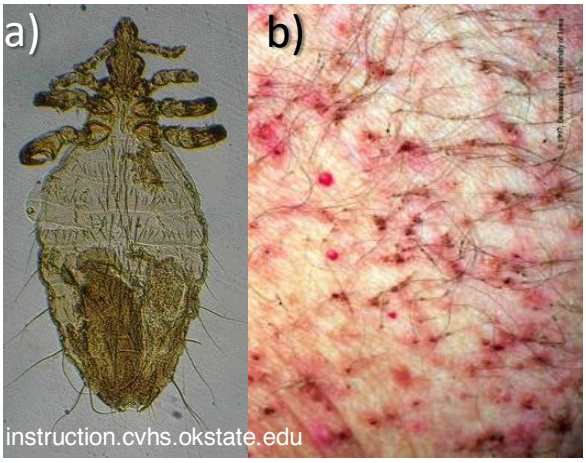


Figure 3.4.3. a) body Louse, b) body lice bites and bite sites infected with *Streptococcus pyogenes*

Body lice (Figure 3.4.3) were once common throughout the world. They are now rare in the developed world, but persist in parts of Africa, Asia and Central and South America. Their bites often cause intense irritation for a few days, with each bite site developing into a small red elevation of the skin. Persons subjected to bites over a prolonged period may develop desensitisation and little or no reaction occurs at the bite site. Persons with chronic body louse infestations often develop a generalised skin discolouration and thickening known as vagabond disease or hobo disease.

They may also develop swollen lymph nodes, edema, elevated body temperature, headaches, joint and muscle pain and a rash. Occasionally people become allergic to the bites and develop generalised dermatitis or a form of asthmatic bronchitis.

Pediculus capitis

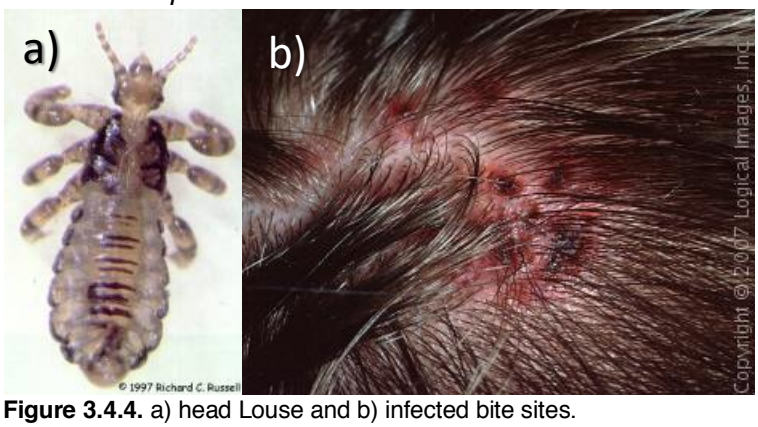


Figure 3.4.4. a) head Louse and b) infected bite sites.

Head lice (Figure 3.4.4) are almost morphologically indistinguishable from body lice (Figure 3.4.3), but have a clear predilection for head hair. They are still common throughout the world with an estimated 6-12 million people, primarily children, infested each year in the United States alone. They are passed from one host to the next through infested clothing and from heads in contact.

Head lice are not directly involved in pathogen transmission, but heavy infestations cause significant irritation and the resultant scratching can lead to secondary infections such as blood poisoning. Swollen cervical lymph nodes may accompany severe head louse infestations, as well, a scabby crust may form on the scalp with large numbers of head lice typically living beneath it.

Pediculus inguinalis

Crab lice are squat lice with robust claws for gripping thick body hairs (Figure 3.4.5). Other than pubic regions, these lice can be found in armpits, eyebrows, eyelashes of both sexes, and also in male beards, moustaches and chest hairs.

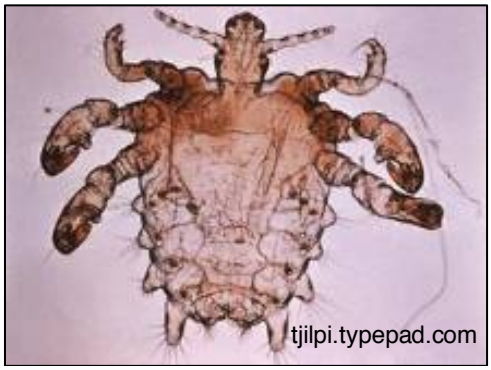


Figure 3.4.5. Pubic louse.

They are common worldwide and are often identified at STD clinics. Purple lesions frequently develop at the intensely itchy bite sites. These lice are not vectors for pathogens but secondary infections may occur at bite sites.

3.5. Bedbugs

Although bed bugs can harbour various pathogens, transmission to humans has not been proven and is considered unlikely. At least 27 agents of human disease have been found in bed bugs, including viruses, bacteria, protozoa, and parasitic worms. None of these agents reproduce or multiply within bed bugs, and very few survive for any length of time inside a bed bug. There is no evidence that bed bugs are involved in the transmission (via bite or infected faeces) of any disease agent, including hepatitis B virus and HIV.

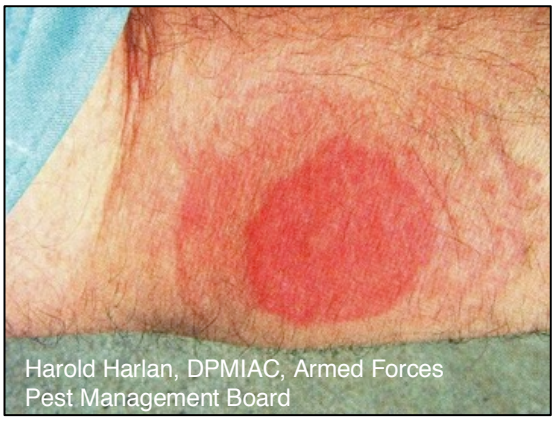


Figure 3.5.1. Delayed reaction from bed bug feeding on arm of researcher.

Though not known to carry diseases, bed bugs can substantially reduce quality of life by causing discomfort, sleeplessness, anxiety, and embarrassment. Heavy rates of feeding in children can result in significant blood loss and eventually lead to anaemia, especially in malnourished children.

Their medical significance is most commonly attributed to itching and inflammation from their bites. Many people have mild to severe allergic reaction to the bites, in rare cases anaphylaxis (severe, whole-body reaction). These bites can also lead to secondary infections of the skin such as impetigo, ecthyma and lymphanigitis.

3.6. Cockroaches

Although cockroaches are not usually associated with widespread disease outbreaks, their presence is a sign of poor sanitation procedures and they are known to carry a number of bacteria which could give rise to serious illness in humans. They may also induce allergies and asthma symptoms in susceptible people.

They are known to be able to carry a vast array of bacteria which may lead to wound infections, food poisoning and gastric upset. Amongst the organisms known to be carried by cockroaches are Salmonella spp. including Salmonella typhi causing typhoid, Entamoeba histolytica causing amoebiasis, Shigella dysenteriae causing dysentery, and potentially also the poliomyelitis virus. Also carried are: Proteus spp., Staphylococcus aureus. Staphylococcus epidermalis, Streptococcus faecali, and *Escherichia coli.*

3.6.1 Typhoid

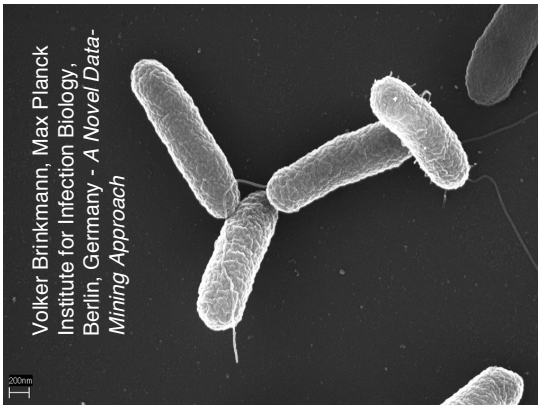


Figure 3.6.1. *Salmonella typhi.*

Typhoid fever is a life-threatening illness caused by the bacterium *Salmonella typhi* (Figure 3.6.1). Typhoid fever is still common in the developing world where it affects about 21.5 million persons each year. Most cases in developed countries can be attributed to infections picked up overseas. Typhoid fever can be prevented and can usually be treated with antibiotics.

Typhoid fever is common in most parts of the world except in industrialised regions such as the United States, Canada, Western Europe, Australia and Japan. Therefore, if you are traveling to the developing world you should consider taking precautions. Over the past 10 years travellers from the United States to Asia, Africa and Latin America have been especially at risk.

The easiest ways to avoid typhoid are by being vaccinated and by avoiding risk food and drinks. Water should be bought bottled or should be thoroughly boiled before consumption. Raw fruit and vegetables that cannot be peeled should be avoided, and anything which can be peeled should be peeled by the intended consumer with clean hands. Hot food should be freshly cooked and still steaming hot.

If infected with the disease the three commonly prescribed antibiotics are ampicillin, trimethoprim-sulfamethoxazole, and ciprofloxacin. Persons given antibiotics usually begin to feel better within 2-3 days, and deaths rarely occur. However, persons who do not get treatment may continue to have fever for weeks or months, and as many as 20% may die from complications of the infection.

Signs and symptoms of typhoid fever

Persons with typhoid fever usually have a sustained fever as high as 103° to 104° F (39° to 40° C). They may also feel weak, or have stomach pains, headache, or loss of appetite. In some cases, patients have a rash of flat, rose-coloured spots. The only way to know for sure if an illness is typhoid fever is to have samples of stool or blood tested for the presence of *Salmonella typhi*.

Transmission

Salmonella typhi lives only in humans. Persons with typhoid fever carry the bacteria in their bloodstream and intestinal tract. In addition, a small number of persons, called carriers, recover

from typhoid fever but continue to carry the bacteria. Both ill persons and carriers shed *Salmonella typhi* in their faeces (stool).

You can get typhoid fever if you eat food or drink beverages that have been handled by a person who is shedding *Salmonella typhi* or if sewage contaminated with *Salmonella typhi* bacteria gets into the water you use for drinking or washing food. Therefore, typhoid fever is more common in areas of the world where handwashing is less frequent and water is likely to be contaminated with sewage.

Even if your symptoms seem to go away, you may still be carrying *Salmonella typhi*. If so, the illness could return or you could pass the disease to other people. In fact, if you work at a job where you handle food or care for small children, you may be barred legally from going back to work until a doctor has determined that you no longer carry any typhoid bacteria.

Excerpts from http://www.cdc.gov/nczved/divisions/dfbmd/diseases/typhoid_fever/

3.6.2. Amoebiasis

Amebiasis (also known as *Entamoeba histolytica* infection) is a disease caused by the parasite *Entamoeba histolytica*. It can affect anyone, although it is more common in people who live in tropical areas with poor sanitary conditions. Diagnosis can be difficult because other parasites can look very similar to *E. histolytica* when seen under a microscope. Infected people do not always become sick. If your doctor determines that you are infected and need treatment, medication is available.

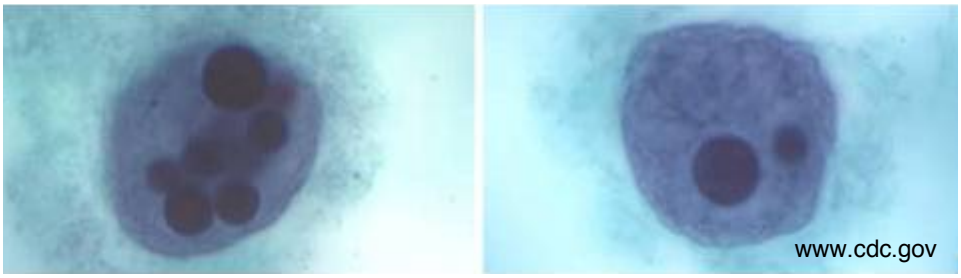


Figure 3.6.2. Trophozoites of *E. histolytica* with ingested erythrocytes (red blood cells) stained with trichrome.

Biology of disease

Several protozoan species in the genus *Entamoeba* colonise humans, but not all of them are associated with disease. *Entamoeba histolytica* is well recognised as a pathogenic amoeba, associated with intestinal and extraintestinal infections. The other species are important because they may be confused with *E. histolytica* in diagnostic investigations.

Life cycle

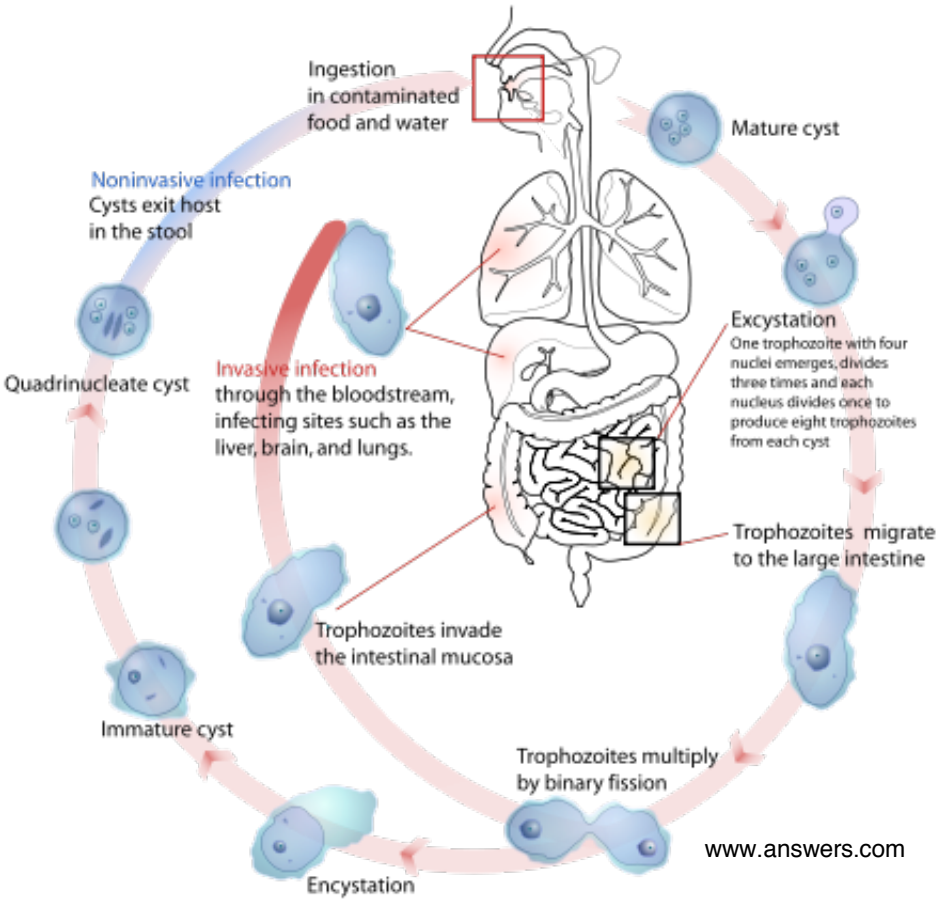


Figure 3.6.3. *Entamoeba histolytica* life cycle.

Cysts and trophozoites are passed in faeces. Cysts are typically found in formed stool, whereas trophozoites are typically found in diarrheal stool. Infection by *Entamoeba histolytica* occurs by ingestion of mature cysts in fecally contaminated food, water or hands. Excystation occurs in the small intestine and trophozoites are released, which migrate to the large intestine. The trophozoites multiply by binary fission and produce cysts, and both stages are passed in the faeces. Because of the protection conferred by their walls, the cysts can survive days to weeks in the external environment and are responsible for transmission. Trophozoites passed in the stool are rapidly destroyed once outside the body, and if ingested would not survive exposure to the gastric environment. In many cases, the trophozoites remain confined to the intestinal lumen (non-invasive infection) of individuals who are asymptomatic carriers, passing cysts in their stool. In some patients the trophozoites invade the intestinal mucosa (intestinal disease), or through the bloodstream, extraintestinal sites such as the liver, brain, and lungs (extraintestinal disease), with resultant pathologic manifestations. It has been established that the invasive and non-invasive forms represent two separate species, respectively *E. histolytica* and *E. dispar*. These two species are morphologically indistinguishable unless *E. histolytica* is observed with ingested red blood cells (erythrophagocystosis). Transmission can also occur through exposure to fecal matter during sexual contact (in which case not only cysts, but also trophozoites could prove infective).

E. histolytica infection can occur when a person ingests or puts anything into their mouth which has touched faeces or been otherwise contaminated with *E. histolytica,* or swallows cysts of *E. histolytica* picked up from contaminated surfaces or fingers.

Only about 10% to 20% of people who are infected with *E. histolytica* become sick from the infection. Symptoms usually develop within 2-4 weeks. The symptoms are often quite mild and can include loose faeces (poop), stomach pain and stomach cramping. Amoebic dysentery is a severe form of amebiasis associated with stomach pain, bloody stools and fever. Rarely *E. histolytica* invades the liver and forms an abscess. In a small number of instances, it has been shown to spread to other parts of the body, such as the lungs or brain, but this is very uncommon.

Diagnosis

Diagnosis of amebiasis can be very difficult. One problem is that other parasites and cells can look very similar to *E. histolytica* when seen under a microscope. Therefore, sometimes people are told that they are infected with *E. histolytica* even though they are not. *Entamoeba histolytica* and another amoeba, *Entamoeba dispar*, which is about 10 times more common, look the same when seen under a microscope. Unlike infection with *E. histolytica*, which sometimes makes people sick, infection with *E. dispar* does not make people sick and therefore does not need to be treated.

If you have been told that you are infected with *E. histolytica* but you are feeling fine, you might be infected with *E. dispar* instead. Unfortunately, most laboratories do not yet have the tests that can tell whether a person is infected with *E. histolytica* or with *E. dispar.* Until these tests become more widely available it usually is best to assume that the parasite is *E. histolytica*.

A blood test is also available but is only recommended when your health care provider thinks that your infection may have spread beyond the intestine (gut) to some other organ of your body, such as the liver. However, this blood test may not be helpful in diagnosing your current illness because the test can be positive if you had amoebiasis in the past, even if you are no longer infected now.

Treatment

Several antibiotics are available to treat amoebiasis. Treatment must be prescribed by a physician. You will be treated with only one antibiotic if your *E. histolytica* infection has not made you sick. You probably will be treated with two antibiotics (first one and then the other) if your infection has made you sick.

Excerpts from http://www.cdc.gov/parasites/amebiasis/

3.6.3 Shigellosis

Shigellosis is an infectious disease caused by a group of bacteria called *Shigella*. Most who get infected with *Shigella* develop diarrhoea, fever and stomach cramps starting a day or two after they are exposed to the bacteria. The diarrhoea is often bloody. Shigellosis usually resolves in 5-7 days. Persons with shigellosis in the United States rarely require hospitalisation. A severe infection with high fever may be associated with seizures in children less than 2 years old. Some persons who are infected may have no symptoms at all, but may still pass the *Shigella* bacteria to others.

Shigella were discovered over 100 years ago by a Japanese scientist named Shiga, for whom they are named. There are several different kinds of *Shigella* bacteria: *Shigella sonnei*, also known as "Group D" *Shigella*, accounts for over two-thirds of shigellosis in the United States. *Shigella flexneri*, or "Group B" *Shigella*, accounts for almost all the rest. Other types of *Shigella* are rare in this country, though they continue to be important causes of disease in the developing world. One type found in the developing world, *Shigella dysenteriae* type 1, can cause deadly epidemics.

Many different kinds of germs can cause diarrhoea, so establishing the cause will help guide treatment. Determining that *Shigella* is the cause of the illness depends on laboratory tests that identify *Shigella* in the stools of an infected person. The laboratory can also do special tests to determine which antibiotics, if any, would be best to treat the infection.

People with mild infections usually recover quickly without antibiotic treatment. However, appropriate antibiotic treatment kills *Shigella* bacteria and may shorten the illness by a few days. The antibiotics commonly used for treatment are ampicillin, trimethoprim/sulfamethoxazole (also known as Bactrim* or Septra*), ceftriaxone (Rocephin*) or, among adults, ciprofloxacin. Some *Shigella* bacteria have become resistant to antibiotics. This means some antibiotics might not be effective for treatment. Using antibiotics to treat shigellosis can sometimes make the germs more resistant. Therefore, when many people in a community are affected by shigellosis, antibiotics are sometimes used to treat only the most severe cases. Antidiarrheal agents such as loperamide (Imodium*) or diphenoxylate with atropine (Lomotil*) can make the illness worse and should be avoided.

People with diarrhoea usually recover completely, although it may be several months before their bowel habits are entirely normal. About 2% of people who are infected with one type of *Shigella*, *Shigella flexneri*, later develop pain in their joints, irritation of the eyes and painful urination. This is called post-infectious arthritis. It can last for months or years, and can lead to chronic arthritis. Post-infectious arthritis is caused by a reaction to the *Shigella* infection that happens only in people who are genetically predisposed to it. Once someone has had shigellosis, they are not likely to get infected with that specific type again for at least several years. However, they can still get infected with other types of *Shigella*.

Every year about 14,000 cases of shigellosis are reported in the United States. Because many milder cases are not diagnosed or reported the actual number of infections may be twenty times greater. Shigellosis is particularly common and causes recurrent problems in settings where hygiene is poor, and can sometimes sweep through entire communities. It is more common in summer than winter. Children, especially toddlers aged 2-4, are the most likely to get shigellosis. Many cases are related to the spread of illness in child-care settings, and many are the result of the spread of the illness in families with small children. In the developing world shigellosis is far more common and is present in most communities most of the time.

<u>Transmission</u>

The *Shigella* bacteria pass from one infected person to the next. *Shigella* are present in the diarrheal stools of infected people while they are sick and for up to a week or two afterwards.

Most *Shigella* infections are the result of the bacterium passing from stools or soiled fingers of one person to the mouth of another person. This happens when basic hygiene and handwashing habits are inadequate and can happen during certain types of sexual activity. It is particularly likely to occur among toddlers who are not fully toilet-trained. Family members and playmates of such children are at high risk of becoming infected.

Shigella infections may be acquired from eating contaminated food. Contaminated food usually looks and smells normal. Food may become contaminated by infected food handlers who forget to wash their hands with soap after using the bathroom. Vegetables can become contaminated if they are harvested from a field with sewage in it. Flies can breed in infected faeces and then contaminate food. Water may become contaminated with *Shigella* bacteria if sewage runs into it, or if someone with shigellosis swims in or plays with it (especially in splash tables, untreated wading pools or shallow play fountains used by day-care centres). Shigella infections can then be acquired by drinking, swimming in, or playing with the contaminated water. Outbreaks of shigellosis have also occurred among men who have sex with men.

Prevention

Currently, there is no vaccine to prevent shigellosis. However, the spread of *Shigella* from an infected person to other people can be stopped by frequent and careful handwashing with soap. Frequent and careful handwashing is important among all age groups. Handwashing among children should be frequent and supervised by an adult in day-care centers and homes with children who have not been fully toilet trained.

Basic food safety precautions and disinfection of drinking water prevents shigellosis from contaminating food and water. However, people with shigellosis should not prepare food or drinks for others until they have been shown to no longer be carrying the *Shigella* bacterium, or if they have had no diarrhoea for at least 2 days. At swimming beaches, having enough bathrooms and handwashing stations with soap near the swimming area helps keep the water from becoming contaminated. Day-care centres should not provide water play areas.

Simple precautions taken while traveling to the developing world can prevent shigellosis. Drink only treated or boiled water, eat only cooked hot foods or fruits you peel yourself. The same precautions prevent other types of traveller's diarrhoea. Some prevention measures in place in most communities help to prevent shigellosis. Making municipal water supplies safe and treating sewage are highly effective prevention measures that have been in place for many years.

Excerpts from http://www.cdc.gov/nczved/divisions/dfbmd/diseases/shigellosis/

3.6.4 Polio

Polio is an infectious disease caused by a virus that lives in the throat and intestinal tract. It is most often spread through person-to-person contact with the stool of an infected person and may also be spread through oral/nasal secretions. Polio used to be very common in the United States and caused severe illness in thousands of people each year before the polio vaccine was introduced in 1955.

Approximately 95% of people infected with polio will have no symptoms. About 4-8% of infected people have minor symptoms, such as fever, fatigue, nausea, headache, flu-like symptoms, stiffness in the neck and back, and pain in the limbs, which often resolve completely. Fewer than 1% of polio cases result in permanent paralysis of the limbs (usually the legs). Of those paralyzed, 5-10% die when the paralysis strikes the respiratory muscles. The death rate increases with increasing age.

Poliovirus is a member of the enterovirus subgroup, family Picornaviridae. Enteroviruses are transient inhabitants of the gastrointestinal tract, and are stable at acid pH. Picornaviruses are small, ether-insensitive viruses with an RNA genome. There are 3 poliovirus serotypes (P1, P2 and P3). There is minimal heterotypic immunity between the 3 serotypes. That is, immunity to one serotype does not produce significant immunity to the other serotypes. The poliovirus is rapidly inactivated by heat, formaldehyde, chlorine and ultraviolet light.

Pathogenesis

The virus enters through the mouth, and primary multiplication of the virus occurs at the site of implantation in the pharynx and gastrointestinal tract. The virus is usually present in the throat and in the stool before the onset of illness. One week after onset there is less virus in the throat, but the virus continues to be excreted in the stool for several weeks. The virus invades local lymphoid tissue, enters the bloodstream and then may infect cells of the central nervous system. Replication of poliovirus in motor neurons of the anterior horn and brain stem results in cell destruction and causes the typical manifestations of poliomyelitis.

Clinical features

The incubation period for poliomyelitis is commonly 6-20 days with a range of 3-35 days. The response to poliovirus infection is highly variable and has been categorised on the basis of the severity of clinical presentation. Up to 95% of all polio infections are unapparent or asymptomatic. Estimates of the ratio of unapparent to paralytic illness vary from 50:1 to 1,000:1 (usually 200:1). Infected persons without symptoms shed the virus in the stool and are able to transmit the virus to others. Approximately 4%–8% of polio infections consist of a minor, nonspecific illness without clinical or laboratory evidence of central nervous system invasion. This clinical presentation is known as abortive poliomyelitis, and is characterised by complete recovery in less than a week. Three syndromes observed with this form of poliovirus infection are upper respiratory tract infection (sore throat and fever), gastrointestinal disturbances (nausea, vomiting, abdominal pain, constipation or, rarely, diarrhoea) and influenza-like illness. These syndromes are indistinguishable from other viral illnesses.

Nonparalytic aseptic meningitis (symptoms of stiffness of the neck, back and/or legs) usually following several days after a prodrome similar to that of minor illness, occurs in 1%–2% of polio infections. Increased or abnormal sensations can also occur. Typically, these symptoms will last from 2-10 days, followed by complete recovery.

Fewer than 1% of all polio infections result in flaccid paralysis. Paralytic symptoms generally begin 1-10 days after prodromal symptoms and progress for 2-3 days. Generally, no further paralysis occurs after the temperature returns to normal. The prodrome may be biphasic, especially in children, with initial minor symptoms separated by a 1-7 day period from more major symptoms. Additional prodromal signs and symptoms can include a loss of superficial reflexes, initially increased deep tendon reflexes and severe muscle aches and spasms in the limbs or back. The illness progresses to flaccid paralysis with diminished deep tendon reflexes, reaches a plateau without change for days to weeks, and is usually asymmetrical. Strength then begins to return. Patients do not experience sensory losses or changes in cognition.

Many persons with paralytic poliomyelitis recover completely and, in most, muscle function returns to some degree. Weakness or paralysis still present 12 months after onset is usually permanent. Paralytic polio is classified into 3 types, depending on the level of involvement. Spinal polio is most common and during 1969–1979 accounted for 79% of paralytic cases. It is characterised by asymmetric paralysis that most often involves the legs. Bulbar polio leads to weakness of muscles innervated by cranial nerves and accounted for 2% of cases during this period. Bulbospinal polio, a combination of bulbar and spinal paralysis, accounted for 19% of cases. The death-to-case ratio for paralytic polio is generally 2%–5% among children and up to 15%–30% for adults (depending on age). It increases to 25%–75% with bulbar involvement.

Excerpts from http://www.cdc.gov/polio

3.7. Ticks

Ticks are excellent vectors for disease transmission. They are second only to mosquitoes as vectors of human disease, both infectious and toxic. They can transmit a wide variety of pathogens, including bacteria, protozoans and viruses to humans and other vertebrates.

As our native ticks do not attach to humans, ticks would not be of any concern to human health in New Zealand if it was not for the introduced species of the cattle tick (*Haemaphysalis longicornis*) that is known to transmit theileriosis, babesiosis, tick-borne encephalitis and more.

Every year there are interceptions at the border of other tick species (Heath and Hardwick, 2011) which can carry diseases of greater concern to human health including Lyme disease, ehrlichiosis, babesiosis, Rocky Mountain spotted fever, tularemia, tick-borne encephalitis and tick-borne relapsing fever, as well as major animal diseases include babesiosis and anaplasmosis. Therefore, personal protection and tick control is very important, as well as biosecurity and border control.

3.7.1 Lyme disease

Lyme disease is caused by *Borrelia burgdorferi*, a bacterium carried by the blacklegged tick or deer tick (*Ixodes scapularis*), the western black legged tick *Ixodes pacificus* and the sheep tick (*Ixodes ricinus*) in Europe and by *Ixodes persulcatus* in Asia. These ticks are usually found feeding on cattle, sheep, horses, dogs and cats.

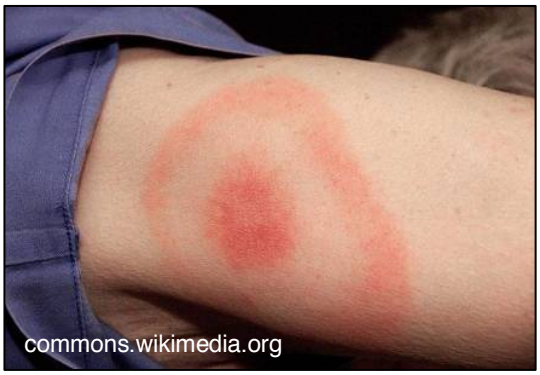


Figure 3.7.1. *Erythema migrans* rash.

Lyme disease is endemic to North America and Eurasia. Symptoms include fever, headaches, fatigue and a distinctive “bullseye” rash on the skin (*Erythema migrans*, Figure 3.7.1). Infection can spread into the heart, joints and nervous system if it remains untreated.

Lyme disease can usually be confirmed through a blood test to detect the presence of antibodies designed to fight the disease. However, it takes 6-8 weeks for the antibodies to show up, and so a blood test done soon after contracting the disease may be negative (falsely indicating absence of Lyme disease). Even after the disease has progressed and antibodies are present, the tests may sometimes still be negative when the disease is present (a result called a "false negative"). If there are signs of early symptoms, especially the tell-tale rash, immediate treatment is usually advised. The blood test will continue to be positive for life.

There is no certain cure for Lyme disease; however, it can be effectively treated with antibiotics. The earlier the disease is treated the better the prognosis for complete recovery. However, successful treatment of the disease will not prevent getting Lyme disease again. A Lyme disease vaccine is now available.

3.7.2 Rocky Mountain spotted fever (RMSF)

Rocky Mountain spotted fever (RMSF), like all rickettsial infections, is classified as a zoonosis. Zoonoses are diseases of animals that can be transmitted to humans. RMSF is caused by *Rickettsia rickettsii*, a bacteria that is transmitted to humans by the American dog tick (*Dermacentor variabilis*) and the Rocky Mountain wood tick (*Dermacentor andersoni*) in the United States, and *Amblyomma cajennense* in South America. Despite its name, RMSF is found in many areas outside of the Rockies, occurring throughout North, Central and South America.

These ticks are vectors and primary reservoirs for this bacterial pathogen. Mice, deer, grounding-feeding birds, wild rodents and dogs are also reservoirs. Risk factors for contracting the disease are those who are frequently exposed to dogs and who live near wooded areas or regions with tall grass.

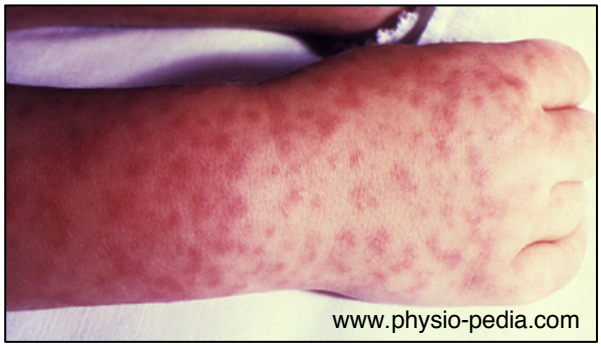


Figure 3.7.2. Typical rash seen with RMSF.

Symptoms of RMSF include sudden onset of fever, headache, and muscle pain, followed by the development of a rash (Figure 3.7.2). It can be difficult to diagnose in the early stages of the disease and can prove fatal if it is not treated. As the name implies, the illness presents with a very distinctive rash that, indeed, looks like spots.

The rickettsia is introduced into humans after an infected tick feeds for more than 6 hours. The tick bite is painless and frequently goes unnoticed.

3.7.3 Tick-borne relapsing fever (TBRF)

TBRF is a disease characterised by relapsing or recurring episodes of fever, often accompanied by headache, muscle and joint aches and nausea.

Tick-borne relapsing fever (TBRF) is caused by several species of spiral-shaped bacteria (*Borrelia* spp.) that are transmitted to humans through the bite of infected soft ticks of the genus *Ornithodoros*. Most cases occur in the summer months in mountainous areas of the Western United States.

3.7.4 Tularemia

Tularemia (also known as "rabbit fever" and "deer fly fever") is a disease that was first recognised as a plague-like disease of rodents in 1911 in Tulare, California. It is caused by a highly infectious bacterium (*Francisella tularensis*) that is widespread "in nature", occurring in a variety of wild animals, in water, and even in soil. The bacterium is not dependent on arthropod transmission, but can be transmitted by the lone star tick (*Amblyomma americanum*) and also from deer flies.

This disease has a worldwide distribution, but exists primarily in the northern hemisphere, including Asia and North America. Most cases occur in the south central United States.

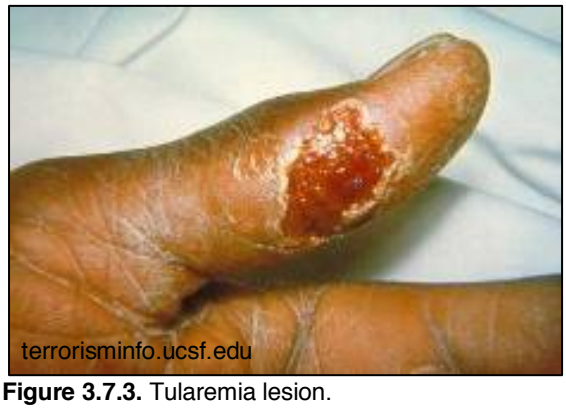


Figure 3.7.3. Tularemia lesion.

General symptoms include fever, headache, chills, nausea, and dry cough. An ulcerated lesion at the site of the bacteria inoculation (e.g. a tick bite) occurs in about 80% of patients (Figure 3.7.3).

There is no vaccine for the general public, but one is available for people in high-risk occupations.

3.7.5 Ehrlichiosis

Ehrlichiosis is a tick borne disease caused by several species of bacteria in the genus *Ehrlichia*, which are pathogens that cause disease in humans, dogs, cattle, sheep, goats, and horses. Currently, 3 species of *Ehrlichia* in the United States and 1 in Japan are known to cause disease in humans.

There are 3 distinct ehrlichioses in the United States. The first, caused by the bacteria *Ehrlichia chaffeensis*, is transmitted by the lone star tick (*Amblyomma americanum*) which occurs in south eastern and southern central parts.

Human granulocytic ehrlichiosis (HGE) represents the second recognised ehrlichial infection of humans in the United States. The name for the species that causes HGE has not been formally proposed, but is carried by the black legged tick (*Ixodes scapularis*) and the western blacklegged tick (*Ixodes pacificus*) in the United States (Figure 3.7.4).

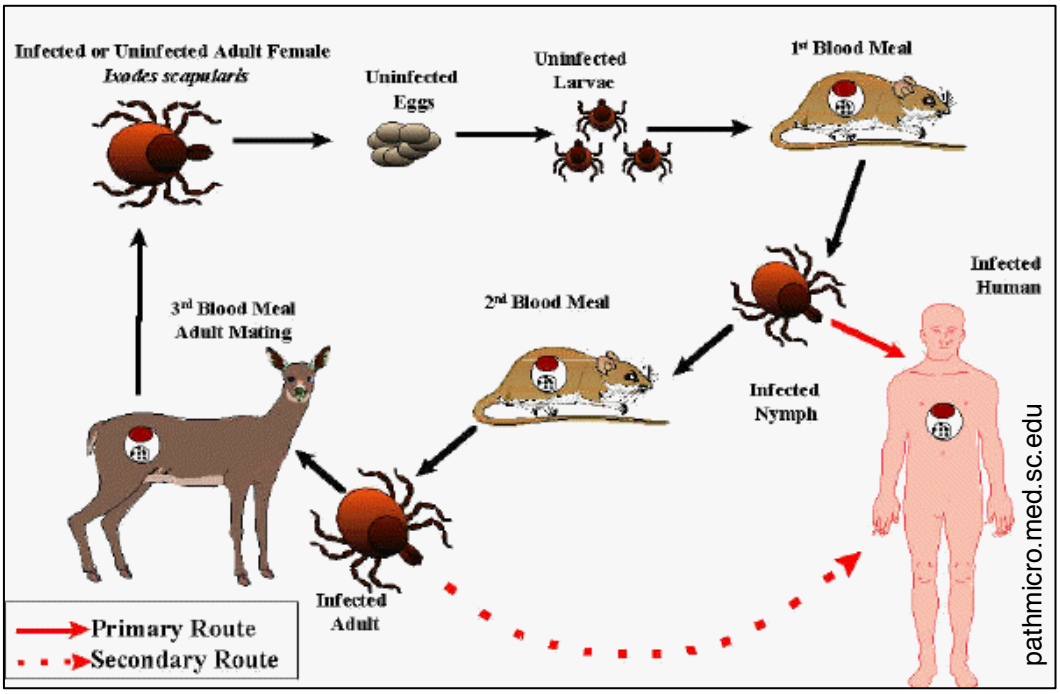


Figure 3.7.4. Proposed life cycle for the agent of Human Granulocytic Ehrlichiosis.

The third and most recently discovered ehrlichiosis is caused by *Ehrlichia ewingii* and has so far been limited to a few patients in Missouri, Oklahoma, and Tennessee. The full extent of the geographic range of this species, its vectors and its role in human disease is currently under investigation.

Symptoms of Ehrlichiosis include headaches, myalgia, rigors and vomiting. Sennetsu fever, caused by *Ehrlichia sennetsu*, in Japan is characterised by fever and swollen lymph nodes. This disease is very rare outside the Far East and Southeast Asia, and most cases have been reported from western Japan.

Canine ehrlichiosis is caused by *Ehrlichia canis* which is transmitted by the brown dog tick (*Rhipicephalus sanguineus*). Symptoms of canine ehrlichiosis include lameness and fever.

3.7.6 Babesiosis

Babesiosis is an uncommon malaria-like parasitic disease caused by piroplasms, protozoan parasites of the genus *Babesia*. *Babesia microti* uses the same tick vector (*Ixodes scapularis*) as Lyme disease and HGE, and frequently occurs in conjunction with them. Babesiosis in humans

is a rare, potentially fatal disease, but is a common infection in other vertebrates. People can be infected with both babesiosis and Lyme disease at the same time.

Babesiosis occurs in the north east of the United States, especially the offshore islands of New York and Massachusetts. Cases have also been reported in Wisconsin, California, Georgia, and in some European countries.

Babesiosis causes a disease very similar to malaria. Infection with *Babesia* parasites can be asymptomatic or cause a mild non-specific illness, and therefore many cases go unnoticed. In mild cases, people may experience mild fevers and anaemia. In more severe cases, fevers go up to 105°F/40°C with shaking chills, and anaemia (haemolytic anaemia) can become severe. Organ failure may follow, including adult respiratory distress syndrome.

In animals, there are a number of *Babesia* species which cause disease. *Babesia canis rossi* causes canine babesiosis and is vectored by the brown dog tick (*Rhipicephalus sanguineus*). This tick will feed on a wide variety of mammals, but dogs are the preferred host in the United States and appear to be required to develop large infestations. Canine babesiosis symptoms include fever, anorexia and anaemia. Out of at least 6 *Babesia* species that have a considerable impact on livestock health and productivity, 2 species (*Babesia bovis* and *Babesia bigemina*) have the greatest affect. Both of these piroplasms cause bovine babesiosis (tick-fever or cattle-fever) in cattle, economically the most important tick-borne disease of cattle worldwide.

Babesia bigemina is transmitted by *Boophilus* ticks, while *Babesia bovis* is vectored by ticks of the genera *Boophilus*, *Rhipicephalus*, and *Ixodes*. Particularly severe forms of this disease can include a severe haemolytic anaemia. *B. bigemina* is distributed wherever *Boophilus* ticks are encountered, which includes North and South America, Southern Europe, Africa, Asia and Australia.

No vaccine against babesiosis is available.

3.7.7 Tick-borne encephalitis (TBE)

Tick-borne encephalitis (TBE) or tick-borne meningoencephalitis (FSME) is a tick-borne viral infection of the central nervous system affecting humans, as well as most other mammals. Caused by a member of the genus *Flavivirus*, the tick-borne encephalitis virus (TBEV) which has 2 subtypes: the European subtype, vectored by the sheep tick (*Ixodes ricinus*); and the Far Eastern subtype (Russian spring-summer encephalitis virus (RSSEV), vectored by the taiga tick (*Ixodes persulcatus*).

The ticks act as both the vector and reservoir for the TBEV. The main hosts are small rodents, with humans being accidental hosts. Large animals are feeding hosts for the ticks, but do not play a role in maintenance of the virus. The virus can chronically infect ticks and is transmitted both transtadially (from larva to nymph to adult ticks) and transovarially (from adult female tick through eggs).

The virus can infect the brain (encephalitis), the membrane that surrounds the brain and spinal cord (meningitis) or both (meningoencephalitis). The disease is incurable once manifest but infection can be prevented by vaccination, and the virus can be inactivated, halting disease progression. In humans the disease can be fatal. Person-to-person transmission has not been reported. Vertical transmission from an infected mother to foetus has occurred.

TBE is an important infectious disease in many parts of Europe, the former Soviet Union, and Asia, corresponding to the distribution of the ixodid tick reservoir (Figure 3.7.5).

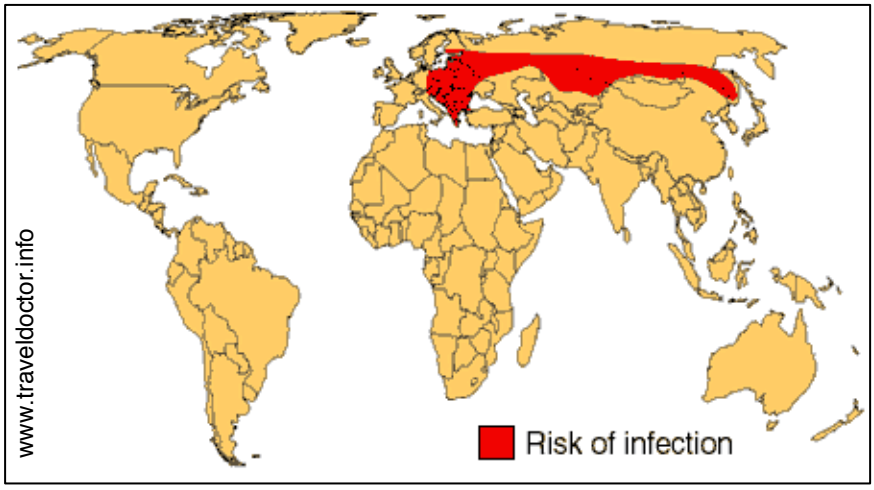


Figure 3.7.5. TBE – Tick Borne Encephalitis distribusion.

3.7.8 Some other tick-borne diseases

Table 3.7.1. Epidemiologic features and symptoms of rickettsial diseases

Antigenic group	Disease	Agent	Vector or acquisition mechanism	Animal reservoir	Geographic distribution outside the USA
Spotted fevers	African tick bite fever	*Rickettsia africae*	Tick	Rodents	Sub-Saharan Africa
	Aneruptive fever	*R. helvetica*	Tick	Rodents	Old World
	Australian spotted fever	*R. marmionii*	Tick	Rodents, reptiles	Australia
	Far Eastern spotted fever	*R. heilongjiangensis*	Tick	Rodents	Far East of Russia, Northern China
	Flinders Island spotted fever, Thai tick typhus	*R. honei*	Tick	Not defined	Australia, Thailand
	Lymphangitis associated rickettsiosis	*R. sibirica* subsp. *mongolotimonae*	Tick	Rodents	Southern France, Portugal, Asia, Africa
	Maculatum infection	*R. parkeri*	Tick	Rodents	Brazil, Uruguay
	Mediterranean spotted fevers	*R. conorii*	Tick	Dogs, rodents	Africa, India, Europe, Middle East, Mediterranean
	North Asian tick typhus	*R. sibirica*	Tick	Rodents	Russia, China, Mongolia
	Oriental spotted fever	*R. japonica*	Tick	Rodents	Japan
	Queensland tick typhus	*R. australis*	Tick	Not defined	Australia, Tasmania
	Tick-borne lymphadenopathy (TIBOLA), Dermacentor-borne necrosis and lymphadenopathy (DEBONEL)	*R. slovaca*	Tick	Lagomorphs, rodents	Europe, Asia
	Unnamed rickettsiosis	*R. aeschlimannii*	Tick	Domestic and wild animals	Africa
Coxiella	Q fever	*Coxiella burnetii*	Most human infections are acquired by inhalation of infectious aerosols; tick	Goats, sheep, cattle, domestic cats, other	Worldwide
Anaplasma	Anaplasmosis	*Anaplasma phagocytophilum*	Tick	Small mammals, and rodents	Europe, Asia, Africa

*This represents only a partial list of symptoms. Patients may have different symptoms or only a few of those listed.

Table adapted from http://wwwn.cdc.gov/travel/yellowBookCh4-Rickettsial.aspx.

3.8. Mites

Mites are associated with disease either as vectors or, as in the case of scabies, as the cause through burrowing into human flesh. Many mites also cause several forms of allergic diseases, including hay fever, asthma and eczema and also aggravate atopic dermatitis. It is thought that inhalation of mites during sleep exposes the human body to some antigens which eventually induce hypersensitivity reaction. Examples of mites causing allergic reactions such as itching and dermatitis include: chicken mite (*Dermanyssus gallinae*), Tropical rat mite (*Ornithonyssus bacoti*) and the Northern fowl mite (*Ornithonyssus sylviarum*).

[Excerpts from Thomson, M.C. 1995, Disease Prevention Through Vector Control: Guidelines for Relief, Oxfam. 96pp.]

3.8.1 Scrub typhus

Scrub typhus is an infectious disease that is transmitted to humans from field mice and rats through the bite of mites that live on the animals. It is a rickettsial disease caused by *Rickettsia tsutsugamushi* (*Orientia tsutsugamushi*), transmitted by the larvae of trombiculid mites that parasitise rodents. The larva is the only stage that can transmit the disease to humans and other vertebrates.

The tiny chiggers (mite larvae) attach themselves to the skin. During the process of obtaining a meal they may either acquire the infection from the host or transmit the rickettsiae to other mammals or humans. In regions where scrub typhus is a constant threat a natural cycle of *R. tsutsugamushi* transmission occurs between mite larvae and small mammals (e.g. field mice and rats). Humans enter a cycle of rickettsial infection only accidentally.

Figure 3.8.1. Trombiculid mite under SEM (*Leptotrombidium pallidum*).

The two main vectors are *Leptotrombidium akamushi* and *L. delicense* (Figure 3.8.1).

In Malaysia, Sumatra, New Guinea and tropical Queensland mite islands are associated with the coarse, fire-resistant kunai grass (*Imperata cylindrical*). Limited studies have shown that rat control may exacerbate scrub typhus transmission because the mites with fewer hosts upon which to feed are more likely to feed on humans.

Scrub typhus is also known as tsutsugamushi disease (Figure 3.8.2). The name tsutsugamushi is derived from two Japanese words: tsutsuga (meaning something small and dangerous) and mushi (meaning creature). The infection is called scrub typhus because it generally occurs after exposure to areas with secondary (scrub) vegetation.

It has recently been found, however, that the disease can also be prevalent in such areas as sandy beaches, mountain deserts, and equatorial rain forests. Therefore, it has been suggested that the names mite-borne typhus, or chigger-borne typhus, are more appropriate. Since the disease is limited to eastern and south-eastern Asia, India, northern Australia and the adjacent islands, it is also commonly referred to as tropical typhus.

The seasonal occurrence of scrub typhus varies with the climate in different countries. It occurs more frequently during the rainy season. Certain areas such as forest clearings, riverbanks, and grassy regions provide optimal conditions for the infected mites to thrive. These small geographic regions are high-risk areas for humans and have been called scrub-typhus islands.

Some of the islands are essentially 'man-made' and are those environments where field rodents have built up large populations, for example, in neglected areas of cultivation.

The incubation period of scrub typhus is about 10-12 days after the initial bite. The main symptoms are fever, a wound at the site of the bite, a spotted rash on the trunk, and swelling of the lymph glands. Infections in humans can result in a high rate of mortality. This disease is found in south, central, eastern and southeast Asia and Australia.

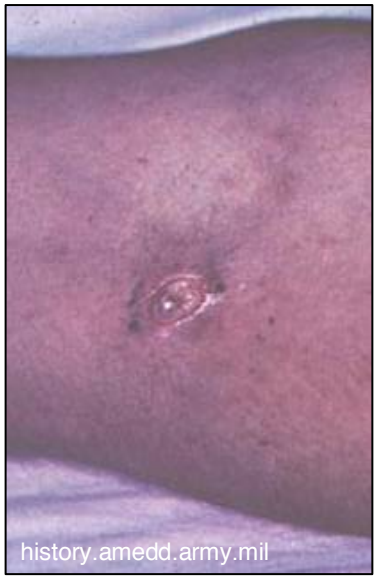


Figure 3.8.2. Scrub typhus.

3.8.2 Rickettsial Pox

The house mouse mite (*Liponissoides sanguineus*) is primarily a parasite of mice. It leaves its rodent host to wander throughout buildings and bite people. Its major importance is that it has been identified as the vector of rickettsial pox (*Rickettsia akari*), a mild and nonfatal human disease.

Symptoms include fever, adenopathy and chicken-pox like rash. The disease begins at the site of the mite bite as a painless, firm, red nodule that develops into a fluid-filled blister that bursts and crusts over. This lesion may be large, almost up to an inch wide. Several days later the patient develops a fever and chills with sweating (diaphoresis) and muscle pain (myalgia). Over the next 2-3 days a rash that looks like chickenpox develops. This rash clears up within a week.

This disease is present in the United States, Russia, South Africa, Korea, Turkey and Balkan countries.

3.8.3 Scabies

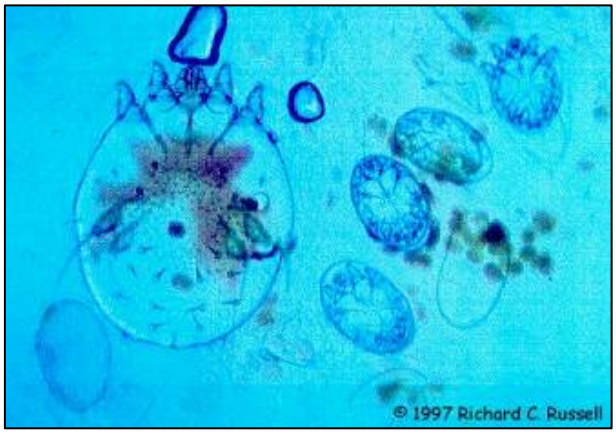


Figure 3.8.3. Scabies mite adult and larvae.

Scabies is not caused by a microorganism but the minute parasitic 'itch' mite (*Sarcoptes scabiei,* Figure 3.8.3) burrowing into the surface layer of a person's skin. This mite also causes 'mange' in a wide range of domestic and wild animals.

The inflammation and itching typical of a scabies infestation is caused by the body's response to the activities and faecal debris of mites. Scabies infections can be extremely unpleasant because of the intense itchiness they cause. Secondary infections can occur as a result of scratching.

Transmission is mostly through prolonged close contact with other people that are infected with the mite; quick contact (as in a hug or a handshake) is unlikely to spread the mite. The scabies

mite is only active above 20°C and is transmitted during host contact under warm conditions, for example in bed. The mites can survive up to 24 hours outside the skin.

In scabies mites the adult fertilised female mite is usually the infective life stage. She adheres to the skin using suckers on her legs and burrows into the skin, chewing her way through the skin surface, feeding on skin cells and excavating a tunnel beneath where she lays her oval eggs. In 3-5 days these eggs hatch into larvae and move freely over the skin. Soon they transform into nymphs and reach maturity 10-14 days after hatching. The adults can then either stay in that host or be scratched off and transmitted to a new host.

Eventually they mate; then the females quickly borrow back into the skin to tunnel and lay eggs. Females live about 2 months and never return to the skin surface. Adult females are usually short-lived after they have laid their eggs.

Domesticated animals can serve as reservoir hosts, but usually different strains have distinct host preferences so infections that are contracted from animals may cause irritation and itching, but are usually short-lived.

Signs include a vesicular rash, visible burrows in the skin, intense itching of infected areas, caused by allergic reaction to activities and secretions of the mites (Figure 3.8.4a). Intense itching may result in disturbed sleep; bleeding and scab formation from scratching can allow for secondary bacterial infection. Itching may be especially bad at night.

Figure 3.8.4. a) Scabies, b) Norwegian Scabies.

Immuno-compromised individuals may experience Norwegian scabies, which involves extensive scaling and crusting (Figure 3.8.4b).

Scabies is a major public health problem worldwide and can reach epidemic proportions in refugee camps, where crowding and poor environmental conditions enable the mite which causes the infection to spread rapidly. It is related to shortage of water for washing. There are an estimated 300 million cases a year, with immuno-compromised people more likely to develop Norwegian (crusted) scabies.

3.9. Rats

Table 3.9.1. Rat transmitted diseases

Disease	Transmission /association	Human disease
Plague	*Yersinia pestis* vectored by Oriental rat flea *Xenopsylla cheopis.*	Bubonic plague – bacteria arrested in lymph nodes. Septicaemic plague – blood infection "Black death".
Sylvatic plague	*Yersinia pestis* – mainly in Nth American rodents.	Less virulent form.
Pneumonic plague	Direct contact and droplet transmission.	Bacterial infection of lungs.
Murine typhus	*Rickettsia typhi* vectored by *Xenopsylla cheopis.* Also directly via infected rat urine and faeces.	*R. typhi* enters blood via flea bite. Less virulent hand louse borne typhus.
Weil's disease	Leptospira icterohaemorrhagiae in rat blood and urine ingested in contaminated food.	Chronic debilitation with fever, elevated heart rate and liver involvement (jaundice).
Rat-bite fever/ Relapsing fever	Mechanical transmission of *Spirillum minus* and/or *Streptobacillus moniliformis* infection of rat salivary glands causing contamination on their teeth/gums.	Swelling of lymph glands, muscular pain. Relapse after apparent recovery.
Trichinosis	*Trichinella spiralis* via eating contaminated pork from pigs eating infected rats.	Worms become encysted in muscles of humans. High mortality in USA.
Lymphocytic choriomeningitis	Virus latent in mice – transmission by contamination of food by infected mouse faeces.	Produces mild meningitis.
Rickettsial pox	Transmission of Rickettsia vectored by bite of *Allodermanyssus sanguiensis* mites infesting mice.	Mild, non-fatal, resembling chicken pox.
Mouse typhoid	Salmonella typhimurium from mice to man via faeces.	Severe food-poisoning, particularly in young children
Poliomyelitis	Two polio viruses excreted by infected mice.	Poliomyelitis.
Favus	Fungus transmitted from infected mice to man, or via cats.	Ringworm.

Transferred by:

- Contamination of food or utensils with rodent urine or faeces.
- Contamination by direct contact with urine or faeces.
- Indirect contamination via bloodsucking ectoparasites e.g. fleas.
- Indirect contamination via pets to humans.
- Contamination by rat bites.

4. Medical Vector Surveillance and Integrated Pest Management

Since most medical vectors are not abundant in New Zealand yet, the importance of surveillance programmes at Point of entries (POE), such as sea- and airports, are absolutely necessary.

Part IV of the Biosecurity Act 1993 provides for the continuous monitoring of New Zealand's status in regard to pests and unwanted organisms.

Article 22 of the International Health Regulations (WHO 2005) requires that, as far as practical, facilities used by travellers at point of entry are maintained in a sanitary condition and kept free of sources of infection or contamination, including vectors and reservoirs.

Article 34 requires, as far as practicable, containers and container loading areas to be kept free from sources of infection or contamination, including vectors and reservoirs.

This is realised by
- checking for the arrival/establishment of unwanted organisms of public health significance.
- identifying potential breeding sites and arranging for these sites to be eliminated or to be the subject of active, ongoing control measures.
- recording the distribution and habitat preference of pest species in New Zealand.

4.1. Pathways of medical vectors

Increased imports and international travel has meant there are several pathways of entry for medical vectors into New Zealand.

Shipping
New Zealand imports a large number of goods, from cars to food stuffs. The large number of vehicles and containers that arrive at our borders contain endless sites for medical vectors esp. mosquitoes to stowaway in. There have been several discoveries of exotic mosquito adults in containers related to used vehicles/machinery.

Aircraft
This mode of transportation is the second major pathway which, as well as transporting a wide variety of import goods, is also responsible for bringing countless travellers and their luggage into New Zealand. Interception specimens have been found in air freighted goods and in cabins of aircraft entering New Zealand for engineering work.

Postage
The posting of goods is another pathway, associated with both shipping and aircraft. There is a continuous supply of materials entering the country.

4.2. Ship Sanitation

Although there are general techniques for sampling for vectors which we will cover through this section, it should always be remembered that there are ship specific considerations to be factored into surveillance while conducting ship inspections for vectors following WHO 'Training Toolkit for Ship Inspection under IHR 2005 and issuance of Ship Sanitation Certificates'(Table 4.2.2.)

4.2.1 Planning

Previous visits in ports known to have infected vectors or endemic to vector borne diseases should be taken into consideration by inspectors and a more thorough inspection for such vectors would be essential in this case.

Different vectors can be found on board ships such as: cockroaches, flies, mosquitoes, bed bugs, fleas, bees, mites, ants, beetles, pests of stored products, fruit flies and rodents.

Each of these vectors has different biology and behaviour. Inspectors should take consideration of the biology and behaviour of vectors in order to identify harbourage places when inspecting ships and be able to recognise evidence of their presence.

It is possible to find vectors in all areas of the ship and therefore, all areas should be inspected:

- Food areas (preparation, service, dishwashing, refrigerators and other equipment, utensil storage, food stores)
- Garbage room (between and under ragbags and food packaging)
- Cabins (mattresses, under bed and other furniture, carpets, coatings, lockers)
- Engine room (drainage systems, joints of decks and bulkheads, among pipe lines)
- Open decks (lifeboats and any standing water)
- Ventilation system
- Laundry
- Cargo holds

However, some areas are most probable to be infested by specific species (Table 4.2.1).

Table 4.2.1. Examples of places where vectors can be found.

Location	Vector
Cabin mattress, curtains	Bedbugs
Lifeboats, open decks	Mosquito eggs and larvae
Garbage, underneath refrigerators	Cockroaches
Food stores	Fruit flies, rodents, pests of stored products
Dining room	House fly
Garage	Rodents, mosquitoes
Cargo holds	Rodents, mosquitoes

Moreover, specific loads can be associated with specific vectors, for example mosquito eggs can be found in used tyres.

Inspectors should check in all areas (Area 13 Other systems and areas, Table 2.4.2) of the ship for:

- Presence of vectors at all stages of their life cycle (e.g. eggs, larvae, pupa and adult mosquitoes; eggs, nymphs, adult bed bugs);
- Other evidence of their presence such as droppings, faeces, dead vectors, cast skins;
- Conditions supporting vector harbourage such as inadequate deck drainage, pooling of water, accumulated garbage, poor hygiene, cracks and cervices in food preparation areas;
- Entry points.

Table 4.2.2. WHO Handbook checklist for vector management system and evience for standing water.

Code of areas	Inspection results: evidence found, sample results, documents reviewed	Control measures and corrective actions	Required	Recommende
13.1 Overall vector management system				
13.1.1 ☐	No rat-proof guard.	Place rodent-proof guard to prevent rodents from boarding ships via the mooring lines.	☐	
13.1.2 ☐	No integrated vector management plan.	Develop an integrated vector management plan.	☐	
13.1.3 ☐	No vector control inspection records and logs available (including pesticide application).	Conduct routine surveillance on vectors and reservoirs; for example, deploy and check rodent traps and other devices.	☐	
		Develop vector control inspection records and logs, including pesticide application logs.	☐	
13.2 Standing water				
13.2.1 ☐	Evidence of standing water in different areas of the ship's open spaces (e.g. lifeboat covers, bilges, scuppers, awnings, gutters, air-treatment plants) that can hold insect larvae. Evidence of depressions or culverts that can collect standing water.	Implement operational procedures to control public health risks to crew and passengers, and communities that could be affected by ships and cargoes arriving in ports.		☐
13.2.2 ☐	Evidence of live vectors or their larvae in standing water inside lifeboats.	Eliminate standing water and apply vector control measures.	☐	

4.3. Exclusion of exotic mosquitoes of public health significance

New Zealand has a small number of mosquito species, meaning there are a number of under-utilised mosquito habitats. There is the potential for exotic species to exploit these habitats, with very little local competition from our own mosquitoes. New Zealand has no endemic diseases of public health significance and the population is largely unaware of mosquitoes and their roles as vectors of disease. Therefore, the population is highly susceptible to exotic mosquito-borne diseases, both in terms of the lack of inherent resistance and ignorance regarding controlling mosquito numbers to prevent epidemics.

- First there would be a potential risk of transmission of disease - not only to humans but to other animals and birds.
- There would of course be nuisance biting from any human-biting species that arrived, to a degree most New Zealanders would not be familiar with.
- There is also the cost of control and mitigation of all types from monitoring, to sprays, to repellents.

There are approx. 3000 species of mosquito described in the world at present, as well as over 1000 arboviruses which are known to cause disease in humans, other animals or both. Even Australia, our neighbour, has over 300 species of mosquito as well as several mosquito-borne diseases including Dengue fever, Ross River virus disease, Barmah Forest virus disease, Murray Valley Encephalitis and Kunjin virus disease.

There are two separate components of a programme to exclude exotic mosquitoes, both should be recognised as being critical and both need to be well maintained:

1. Border control activities:
Mosquito exclusion activities should be integrated with surveillance inspection and disinsection of vessels, aircraft and risk goods to deter the occurance of an exotic species.

2. Surveillance of mosquitoes:
Regular monitoring of controlled breeding habitats in and near New Zealand port environs may be of use in detecting and preventing incoming mosquitoes from escaping the port areas It also detects any presence of local species to identify potential habitat that may be utilised by exotic species if they escape detection on arrival.

4.3.1 (Pre)- Border Inspection and Clearance of Risk Goods

The first lines of defence against the importation of exotic mosquitoes include the pre border clearance of risk goods conducted by MPI Quarantine Service (MQS) staff at off-shore sites. The inspection of sea vessels (including yachts) arriving at seaports and the disinsection of untreated aircraft arriving at airports, and the inspection of their high-risk cargo is the next line of defence.

4.3.2.1 Aircraft and airports

Every aircraft arriving from a foreign place is to be sprayed in every compartment so as to kill all mosquitoes (if any) using an approved insecticide and methodology.

4.3.2.2 Seaports and ships

Inspection and Treatment of First Port of International Call Vessels (including yachts) will, as far as is practicable, be inspected to identify the presence of exotic mosquitoes (all life stages) or of actual or potential breeding sites for exotic mosquitoes.

Risk items associated with ships, yachts, other pleasure craft
The following items are considered high risk with respect to the detection of any exotic mosquito of public health significance:

- soil, sand, clay and water from any country
- tyres from any country
- forestry and agricultural equipment from any country
- used buses, cars, motor cycles, trucks, utility vehicles and vans from any country
- sea containers from any country
- air containers from any country
- water tanks and other drinking water storage containers
- discarded items (such as tins, cans, bottles, cups, drums)
- plastic sheeting and tarpaulins with pooled water
- bilge water
- fenders, especially used tyres
- life boats with pooled water
- decks with pooled water (remaining for 5-10 days); swimming pools
- accommodation area including pot plants, bathrooms, blocked basins, closets, under bunks, dark parts of rooms

MPI will advise the relevant public health unit of all suspected interceptions of exotic mosquitoes.

4.3.2 Mosquito Surveillance

The monitoring of mosquito populations at seaports and airports is the third line of defence against importations of exotic mosquito species. Public health units or port companies may do this but in the latter case the results of surveillance should be audited by public health units. This surveillance also provides critical data regarding the potential for ports/airports to export indigenous species to other countries.

Where and when?
Locations should be defined by risk assessment relating to entry and establishment potential of exotic mosquitoes (such as evaluation of entry pathways, volume and type of risk goods, life stage of incoming mosquitoes, availability in New Zealand of blood hosts and larval habitats, suitability of the climate, species biology, vector importance, human population size).

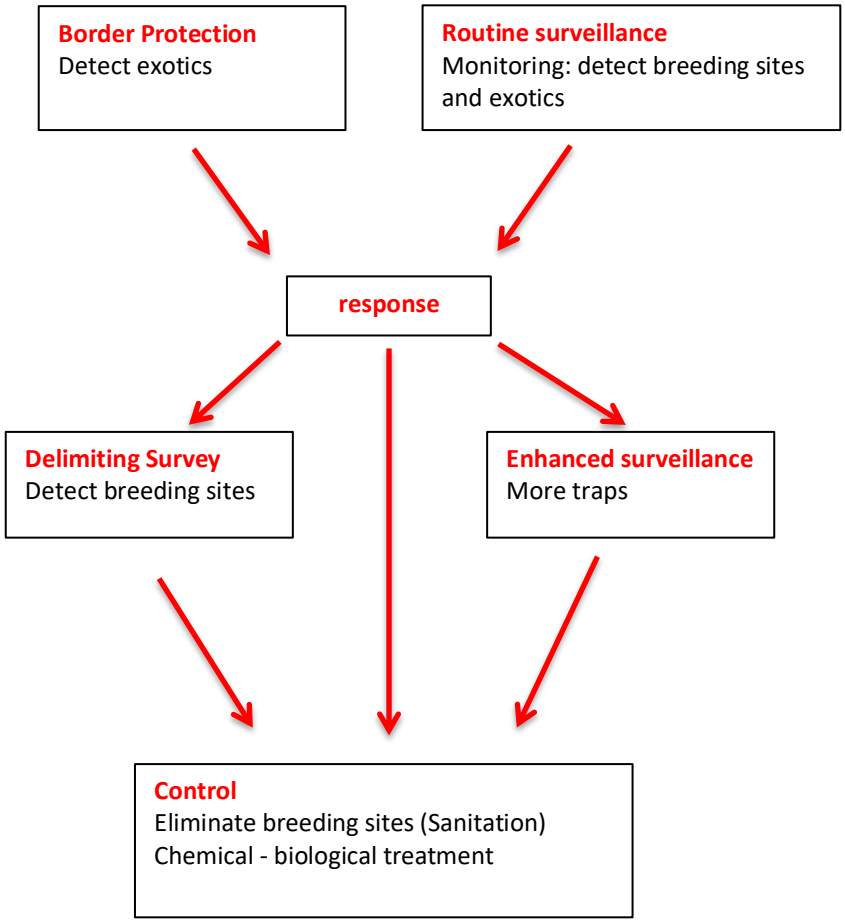
- high risk locations: larval sampling and adult trapping annually (weekly in summer / fortnightly in winter)
- low risk locations: larval sampling on a seasonal basis (from October to May)
- ongoing surveillance of habitats (or potential habitats) for fresh water container-breeding mosquitoes within a 400 m zone
- ongoing surveillance around international yacht berths (first port of call), and premises of importers of high risk goods such as used tyres
- surveys at least once a year in a surrounding five km buffer zone of ports and airports as well as other high risk sites and favoured habitats within the metropolitan region

Mosquito Surveillance

If a live exotic mosquito of public health significance (unwanted mosquito) is detected at the border we speak of an **interception**.

If an interception had been undetected and an exotic mosquito population has established we speak of an **incursion.**

In both cases a strategy for an adequate **response** is necessary as well as the awareness of dealing with media and educating the public.



What are we looking for?
The answer is "unwanted mosquitoes". Those are exotic mosquito species which are highly vector competent and are therefore of public health significance (Table 4.3.1).

Table 4.3.1. New Zealand unwanted mosquitoes, scientific name, common name and the diseases they are able to transmit.

Organism	**Common name**	**Public health significance**
Aedes aegypti	Yellow fever mosquito	Barmah Forest virus disease, Dengue fever, yellow fever, Chikungunya, Zika
Aedes albopictus	Asian tiger mosquito	Chikungunya, Dengue fever, Eastern equine encephalitis, Japanese encephalitis, Ross River virus disease, Venezuelan equine encephalitis, Western equine encephalitis, yellow fever, Zika
Aedes atropalpus	Rock pool mosquito	Eastern equine encephalitis, LaCrosse encephalitis, possibly West Nile virus disease
Aedes camptorhynchus	Southern saltmarsh mosquito	Ross River virus disease, possibly Barmah Forest virus disease
Aedes japonicus	Japanese rockpool mosquito	Japanese encephalitis
Aedes polynesiensis	Polynesian mosquito	Bancroftian filariasis, Chikungunya, Dengue fever, Murray Valley encephalitis, Ross River virus disease
Aedes scutellaris	[No common name]	Dengue fever
Aedes vigilax	Saltmarsh mosquito	Bancroftian filariasis, Kunjin virus disease, Murray Valley encephalitis, Ross River virus disease
Aedes sierrensis	Western tree hole mosquito	Western equine encephalitis
All mosquitoes of the genus *Anopheles*	Malarial mosquitoes	Malaria, may transmit filariasis and several viral diseases
Culex annulirostris	Common banded mosquito	Ross River Virus disease, Barmah Forest virus disease, Murray Valley and Japanese encephalitis and Kunjin virus disease
Culex gelidus	Frosty mosquito	Japanese encephalitis
Culex pipiens pallens	Common house mosquito	Bancroftian filariasis, Chikungunya, Japanese encephalitis, West Nile virus disease
Culex sitiens	[No common name]	Japanese encephalitis, possibly Ross River virus disease

Which technique?
There is no single method ideally suited to sampling all mosquito populations. Each technique employed will be more suitable for specific species and unsuitable for others. In any mosquito surveillance the programme should be developed to monitor the target species at various stages of the insect's life cycle.

For example, if you are in an area where the primary concern are *Aedes aegypti*, you would not focus your efforts on light trapping at significant distances from the area of concern as *Ae. aegypti* has a short flight range and is not greatly attracted to light traps. Whereas if you were concerned with *Aedes vigilax* you would not waste resources sampling small containers in urban areas as it is a saltmarsh breeder. Most programmes will be concerned with a range of mosquito species and a suite of sampling methodologies should be employed.

What to do?
All specimens collected during surveillance should be submitted to the Ministry's mosquito identification experts by overnight courier.

All larval habitats where feasible for mosquitoes within the 400 m or 1000 m port/airport zone need to be removed or treated within three days of detection.

4.3.2.1 Routine surveillance

Regular surveillance for the presence of exotic mosquitoes at sea and air ports (including devanning and transitional facilities) is necessary throughout New Zealand at selected locations.

The generic monitoring methodologies that are currently employed variously across the range of seaports and airports are:

1. larval surveys (ground pools and receptacles)
2. larval traps (sentinel tyre traps)
3. adult traps (CO_2 light traps, BG traps, GAT and Dominator).

4.3.2.2 Larval sampling

Regular sampling for larvae should be carried out in all natural (e.g. tree-holes, leaf axils) and artificial containers (e.g. tyres, drums), ground pools, ponds, sumps, drains, and marshes (fresh and saline), building gutters and any other accumulation of water within the 400m exclusion zone for sea- and airports. Sampling frequency should be at least fortnightly during warmer months, but preferably weekly. Thorough searches should be made for above and below ground, habitats, as these are often hidden but ideal sites for breeding to take place.

Sampling Equipment

Seaports and airports are often dangerous places to work in, and so it is essential that personnel involved in surveillance are familiar with the site Health and Safety requirements. Hi-Viz vests and other personal safety equipment should be used where required.

- Mosquito Sample Collection Sheets (or equivalent)
- Map
- GPS
- Tubes, labels, pencil
- Dippers: The standard ladle-type dippers of either 100-150ml or 400-500ml are used widely and are acceptable and effective for sampling various habitats, particularly surface water accumulations, depending on the relative size.
- Pipettes: Large (turkey baster) and small plastic Pasteur pipettes (1ml and 3ml) are useful for removing larvae from smaller habitats/receptacles, transferring larvae from ladles to collection tubes.
- White plastic tray

4.3.2.2.1 Groundwater sampling

Ground water sampling incorporates most of the habitat categories excluding container type habitats and will include field drains and runnels, swamps, marshes, mangrove, ponds, lake edges etc.

For field sampling of ground water habitats, it is important to walk around the entire margin of the site to determine the entry or exit points and possible source of the water. Permanent sample points may be chosen after initial sampling if longer term surveillance is likely to occur. In a larger site, this may include a point in each vegetation or water type. These permanent sample points should be at sites where there is year-round access. All sample sites should be marked on a map. These permanent sample points will provide an assessment of how the breeding habitat, species type and numbers of mosquito larvae change over time, as well as potentially important factors in that habitat that may lead to fluctuations in mosquito numbers.

Additional larval samples should also be taken at different points throughout the habitats during each visit to make sure the permanent sample points are efficient indicators of larval breeding sites.

The following procedures relate to sampling for mosquitoes in natural water bodies such as ponds, marshlands, drains, oxidation ponds and so on, but may also apply to containers being checked for the presence of container-breeding mosquito larvae.

Though a few mosquito species can live in water with a gentle current or flow, larvae of most species will be found in still water.

Mosquito larvae are usually found where surface vegetation or debris is present. In larger bodies of water, larvae are confined to marginal areas or floating surface materials (e.g. weed mats). Examine maps, and satellite imagines when available, for vegetation patterns and likely areas of mosquito breeding. Plan the access route and plan your specific search sites.

When searching for mosquito larvae be prepared to walk and push through thick vegetation into the selected sites chosen on the aerial photographs or satellite images. There is no substitute for leg work and perseverance. Please, however, beware of your personal safety at all times.

When approaching a margin of a water body it is important to note the vegetation patterns. The different types of grass, reeds or other vegetation may be clues to deciding exactly where the mosquitoes are likely to be and which habitats must be sampled. When you have selected particular habitats, look at the water before disturbing it with the feet, ladle or shadow. Note the presence of fish and other predators, and look for larval activity. Remember that wind may cause larvae to gather at the down-wind end of a pool.

- Do not cast your shadow over the water as this will disturb the larvae. Look through the water carefully for wriggling or hanging larvae before you disturb the water with your feet or ladle. Quickly skim the larvae into your ladle. NB: *Aedes* larvae generally dive to the bottom of the water (and usually they swim down at an angle) when disturbed. Some species spend a lot of time near the bottom of the water column, this should be factored in when sampling any habitat.
- Begin sampling at the water edge. Dip into any grass or vegetation and let the water run into the dipper (ladle). Scoop the dipper up just before it completely fills with water. If there is a chance that larvae have been disturbed prior to dipping, then wait for approximately 30 seconds for larvae to resurface then continue with sampling. If the habitat allows, move forward three paces, stop and take another 2 samples and so on. You can cover a greater area by zigzagging as you move through the habitat.
- Take time to search for first instar larvae. You may have to look for a shadow of movement in the bottom of the dipper as they are very small and are very difficult to see.

4.3.2.2.2 Artificial and Natural Container Habitats

Container sampling incorporates artificial and natural container habitats, and arguably also includes artificial subterranean habitat. Natural containers such as tree holes, leaf axils and coconut shells etc., and artificial containers such as discarded rubbish, tyres, tin cans, plastic sheeting, oil drums, buckets and guttering etc.

- Examine the area, for any containers and note the type of container and the presence of water or larvae in each container.
- Each container should be sampled for larvae.
- The first step in sampling larvae is to look carefully at the surface of the water for larvae or pupae.
- With a ladle, quietly lower the ladle as deep into the container as possible so that larvae can be seen against the white background of the ladle. The ladle can then be slowly extracted with the larvae and water.
- Depending on size, the container can be emptied carefully into a white tray for further examination.
- Tyres can be prised open and the water can be scooped with a ladle, ensuring that the ladle rim makes contact with the bottom of the tyre.

General

- If you find larvae transfer them to a specimen tube using a plastic pipette.
- Number the tube and complete the sampling form.
- Always keep sample tubes and sheets together in the field.
- Pipette all larvae collected from one site into a single sampling tube where possible.
- Record the number of dips made, this information will give a guide as to how big any future adult populations may be.
- If no larvae are found it is still important that the sample is recorded.
- Upon return to the office follow the procedure for sample handling.

4.3.2.3 Adult sampling

4.3.2.3.1 Biting/Landing Collections

The "bait" subject rolls up his shirtsleeves or trouser legs and sits quietly for 10-15 minutes, collecting the mosquitoes that settle on the exposed skin. Various techniques are used such as test tubes (Figure 4.3.2) and small vacuum devices (Figure 4.3.1). NB: For medical/ethical reasons, this method should only be used under the supervision of senior environmental health staff. There is no legal barrier to collecting samples from personnel who are fully clothed.

4.3.2.3.2 Aspirating Adults

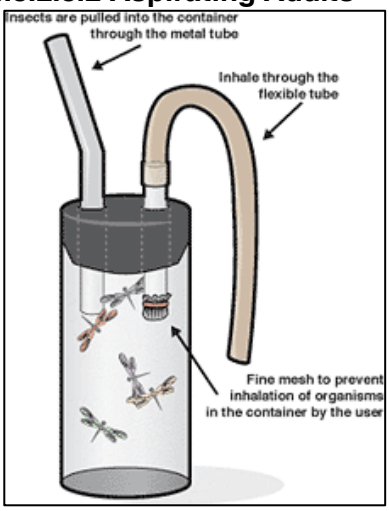


Figure 4.3.1. Mouth-operated aspirator

Adults on the wing or resting on a surface may be collected using an aspirator (Figure 4.3.1), often called a pooter. It is a device for collecting small insects or spiders using light suction. Motorised versions are available but normally the suction produced is by the lungs.

When using the type of aspirator depicted above, air is sucked through the thick end of the tube while the thinner end is held near the mosquito. The mosquito is sucked into the thin tube as far as the join with the larger tube where movement is stopped by a mesh barrier. The mosquito may then be blown into a collection tube and processed.

4.3.2.3.3 Collecting Mosquitoes using a tube

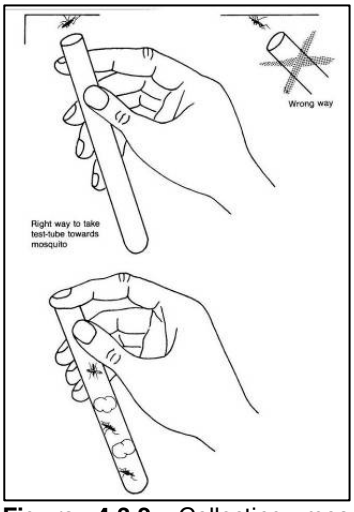


Figure 4.3.2. Collecting mosquitoes with a tube.

Hold the mouth of the tube directly over the mosquito. When the mosquito is disturbed it will fly into the tube.

Close the mouth of the tube with your index finger or thumb. Remove your finger and push a plug of cotton wool into the tube.

Push the plug down until the mosquito is trapped in the bottom 2 cm of the tube (Figure 4.3.2).

4.3.2.4 Larval trapping

For the detection of *Aedes aegypti* at airports and seaports, the standard practice endorsed by the World Health Organisation (WHO) has been the deployment of ovitraps. Although targeting *Ae. aegypti,* they may also attract *Aedes albopictus* and other container-breeding exotic mosquito species, which arrive on foreign vessels, or local species such as *Aedes notoscriptus* or *Culex quinquefasciatus* which may be present within the port environs.

4.3.2.4.1 Tyre Traps

Some mosquito species have evolved from ovipositing in leaf axils and tree holes and are known to find used tyres particularly attractive oviposition sites. An example is *Aedes albopictus* which has been spread globally as its desiccation resistant eggs were transported in used tyres.

Some Aedine mosquitoes e.g. *Aedes aegypti* are known to prefer darkened breeding areas and so a tyre trap works by providing an attractive oviposition surface above nutrient-enhanced water for a mosquito to oviposit on within a darkened environment. The attractiveness of these traps for *Aedes aegypti* can be enhanced by utilising aged water, which is produced by supplementing the water to be used, with organic nutrients such as grass or rabbit food pellets (especially lucerne-based), several days prior to use.

Aedine mosquitoes will lay eggs above the water line inside a tyre and the eggs will hatch after being flooded. A disadvantage of this method is that eggs laid in week one may not be flood hatched until week two (through the replacement of water in the trap) and therefore cannot be sampled until week three. This may allow an exotic mosquito up to two weeks to establish more widely within an area, before its discovery.

Tyre traps pose a high risk of breeding large numbers of mosquitoes, and so a few pellets of S-methoprene, a mosquito juvenile hormone, should be added weekly to prevent the emergence and escape of adults.

Due to the S-methoprene the mosquitoes will only develop through to pupal stage. Identification of species at the pupal life stage is difficult, so it is important that the mosquitoes are sampled while they are present as one of the four larval instars. The addition of S-methoprene to the traps has been shown to have no deterrent effect on ovipositing female mosquitoes (Ritchie & Long, 2003). S-methoprene pellets are able to be purchased from the MoH Contractor New Zealand BioSecure

The following represents best practice for the construction and maintenance of ovitraps and tyre traps for the surveillance of exotic *Aedes* spp., and sampling of eggs, larvae (and adult mosquitoes if caught). (Adapted from guidelines prepared by P. Whelan, G. Hayes and J. Carter, Territory Health Services).

Equipment

- Tyre, used car or small aviation type
- Map with exact locations of ovitraps to be set up/serviced
- Mosquito Trap Collection Sheets (Appendix 6)
- Processing larval tyre traps Checklist (Appendix 5)
- Aged water - tap water containing rabbit pellets (preferably lucerne-based) left to stand for a week or so
- Brush
- S-methoprene
- White plastic tray
- 1 or 3 millilitre pipette
- Sample tubes with label
- Pencil

Trap Position
When deployed as larval traps, whether in parallel with or as replacements for conventional ovitraps, tyre traps should be placed close to the activity wishing to be monitored, and protected from the wind when outdoors.

Traps should be deployed indoors or outdoors in relatively secluded, sheltered, shady, low to the ground (0-1 metres above ground) sites, near vegetation (where possible), protected from rain and animal disturbance, but near areas where there is regular human activity. They should not be placed near spider webs or inside very thick vegetation.

Trap Construction
Used automobile or light aircraft tyres (<500 millimetre diameter) can be placed outdoors in sheltered shaded areas, near vegetation if possible, in an upright position. The vertical alignment of the tyre should be marked so it can be replaced in the same aspect and filled with at least 1 litre of water to the same marked level (or to the drainage hole) on each occasion. This will ensure that eggs laid previously will be covered for hatching and not left away from or above the water line. Chaining, or otherwise fixing, the tyre in position for security purposes can facilitate the correct positioning.

Tyre traps are typically old tyres with a drainage hole cut into the tread or side wall. Cutting into the side wall is easier in the field and can be done with a box cutter or other sturdy knife. The tyres may be left black or marked with red paint.

Tyre traps remain in the field and need to be serviced regularly. They should be marked to aid placement, e.g. it is suggested that a weather-proof label be attached to the tyre that is:
a. readily identifiable;
b. describes the purpose of the tyre e.g. "*Mosquito Surveillance Trap*" and requests that individuals desist from interfering with the trap; and
c. includes appropriate name and contact information.

When not in use, tyre traps should be stored in an area where they can't get wet and subsequently provide a breeding site for nuisance mosquitoes.

Tyres should be inspected weekly over the warmer months of the year and fortnightly at other times. All the water is drained from the tyre and into a white shallow tray for larval inspection. NB: It is important to carry out this step from week 1, and not just from week 2, as Culicine mosquitoes lay their eggs on the water surface and they may hatch within hours of oviposition.

Trap Placement
Ensure that each time the trap is serviced it is repositioned at the same orientation and same angle of lean as previously. Any relocation of trap positions should be recorded on sample sheets and the field map and the new positions allocated new site numbers.

Fill the trap with water to the same level as previously filled and add a S-methoprene pellet or ~30 S-methoprene pellets. S-methoprene does not act as a repellent in traps.

Trap Collection
Empty the trap completely into a white tray (this may take more than one fill of the tray). Remove all larvae (instars 1-4), pupae and exuvia using plastic pipettes and transfer to a labelled sample tube. The label should be written in pencil and contain the trap sample number with the collector's initials and the collection date. NB: Larval skins (exuvia) can also be used for species identification. If any adults are present in the water remove carefully and place into a separate labelled specimen tube. Complete the Trap Collection Sheet (positive or negative). Cross contamination between trays should be avoided by washing the pipettes (inside and out) between each.

Trap Maintenance

As mentioned earlier, tyre traps should be regularly brushed and flushed in the field. If the trap becomes very dirty and there are no facilities for cleaning it on site, transport it back to base and clean properly. For details of water quality and sample size refer to Table 4.3.2.

Table 4.3.2. Tyre trap water quality maintenance and sample size.

Water quality	
Aged water (Chlorinated water, storage, additives)	PHUs are requested to store water that is chlorinated for a week before usage in tyre traps. 5-10 lucerne pellets or a handful organic matter/grass is to be added to the water in tyres.
Changing water in tyres	The water of the tyre must to be flushed and emptied entirely through the drill holes into a white dish and is to be discarded after all mosquito larvae have been sampled. The water is not to be reused. New aged water must be used to refill the tyre.
Scrubbing tyre (no, yes, frequency)	Do not scrub if the trap is negative **and** the water is clear despite the organic additives. Or: HPOs should occassionally scrub the tyres. If the change of water is not sufficient to obtain a natural water quality, and the water shows signs of bloom or water fouling (great increase of phytoplankton or in a water body as a response to increased levels of nutrients or brown smudge) – and as this can lead to negative environmental effects include hypoxia, the depletion of oxygen in the water, it is repellent for most mosquito species – **please do scrub**.
Sample size	
Sample size – Routine larval traps	If there are too many larvae to pipette them into one sample container, the use of a fine-meshed sieve is recommended. If a sieve is used the sample should be proceed in the HPU laboratory to clean up all the debris. If a trap is always negative for a year, a change of the location should be considered and discussed with NZ BioSecure.
Sample size –enhanced larval traps (after an interception/incursion/public complaint)	Sampling larval traps after a response to an exotic mosquito, all specimens must be collected.
You may use the checklist for processing larval traps Appendix 5.	

4.3.2.4.2 Ovitrap

Ovitraps are normally not used by PHUs in the routine surveillance any longer (Review of New Zealand Mosquito Surveillance: Russell and Ritchie, 2013) but they are important for enhanced surveillance (see section 4.3.3 Response to Suspected or Confirmed Exotic Mosquitoes of Public Health Significance).

They provide an attractive artificial habitat for adult female mosquitoes to lay their eggs in. The combination of a smooth container and a rough surface has been found to focus mosquitoes for laying eggs on removable oviposition sites. The imprinted surface on masonite or wooden paddles (tongue depressors roughened by coarse sandpaper) provides a rough and a light coloured surface against which eggs can be more readily seen.

- 1 litre blackened, screw-top, glass/plastic jars (painted or inside black casing)
- 3 sets of numbered, masonite or wooden paddles; 16 centimetres x 3 centimetres
- Aged water - tap water containing rabbit pellets (preferably lucerne-based) left to stand for a week or so

- Mosquito Trap Collection Sheets
- Map with exact locations of ovitraps to be set up/serviced
- Compartmentalised, plastic, paddle transport box
- Trap Transport Container

Trap Construction

Ovitraps are typically made out of black, glass/plastic containers of approximately 500-750 millilitres and which contain a paddle (Figure 4.3.3a). They should be prepared in the laboratory or workroom prior to going out in the field.Each ovitrap should have a different number and each component (jar, paddle, black casing (if required)) should be numbered identically.

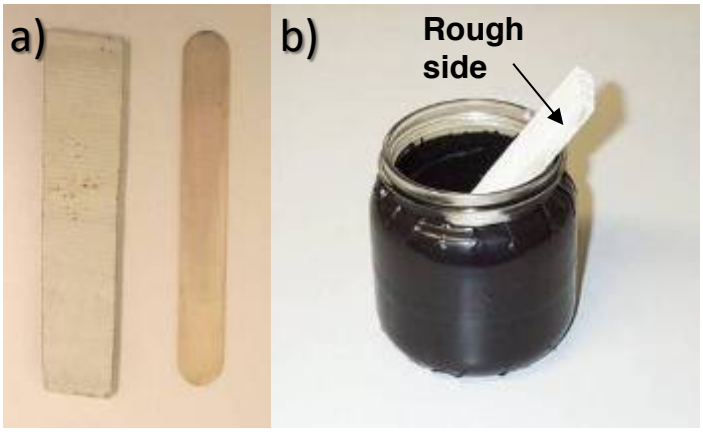


Figure 4.3.3. a) paddle types, b) paddle position in the ovitrap.

Fill the clean ovitrap jars with aged water to depth of 10 centimetres or near to the top (dependant on the size of jar used). Place the numbered, clean, masonite (or wooden) paddle (rough side up, Figure 4.3.3b) in the corresponding numbered jar.

Trap Placement

Place the jar into the black plastic casing (where required) and load the completed ovitraps into containers ready for transportation to the field sites.

Ovitraps should be stabilised by situating between bricks or stones, or behind/under a suitable object. Replacement ovitraps are positioned at the same time the already exposed ovitraps are collected. Any relocation of trap positions should be recorded on sample sheets and the field map and the new positions allocated new site numbers.

Ensure that the replacement ovitrap jar, masonite paddle and black plastic casing have corresponding site numbers, that the water in the jar is at the correct level and top-up if required. Also, that the paddle is placed rough side up and out (Figure 4.3.3b). Place the ovitrap in the designated position and add a S-methoprene pellet.

Trap Collection

Exposed ovitraps are collected at the same time the replacement ovitraps are being positioned. Record any disturbance to the ovitrap in the additional information section of the Trap Collection Sheet, such as invasion of trap by ants or frogs, trap stolen or vandalised, no water left in ovitrap, trap tipped/blown over, paddle lost etc.). Remove the paddle and place rough side up in the compartmentalised plastic paddle transport box (Figure 4.3.4). Remove the trap, record the presence of larvae, pupae or dead adults (Additional Information section of the Trap Collection Sheet) and place in the Trap Transport Container.

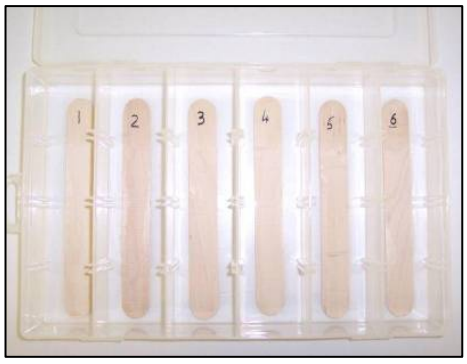


Figure 4.3.4. Paddle transport box.

NB: the presence of any pupa skins indicates that inspections have been too far apart and the ovitrap servicing period needs to be shortened. This aspect should be brought to the attention of the supervisor immediately on return from the field.

Trap Cleaning
All trap parts (jars, paddles, casings) should be cleaned with boiling water, whether mosquitoes were observed or not. Do not use detergent as this can make the traps too clean and less attractive to ovipositing females. Scrubbing of the jars and paddles will be required to remove any old hatched egg cases. Use a dedicated scrubbing brush, which has not been used for washing with detergent.

Wooden paddles may be reused, providing there was no evidence of mosquito eggs before cleaning. It should be noted however, that these will not last long and a supply of new paddles should be available in advance.

4.3.2.4.3 Trap treating

Since traps provide breeding habitat we are responsible to control the breeding. We want to detect and monitor mosquito activity but not increase an existing population. Therefore, we need to treat our traps with S-methoprene (for legislation please refer to the MoH Manual Section 5 Biosecurity).

S-methoprene is available in a number of formulations, including sustained release pellets, boluses and briquettes. Various formulations have improved persistence of S-methoprene, especially in water. Unformulated methoprene has a short half-life in water and soil (<10 days), but with the use of sustained release formulations, activity against mosquitoes has been detected for over 100 days in water. Persistence is affected by water quality, salinity and temperature. UV light rapidly degrades methoprene.

It is recommended to use a few pellets per tyre trap until they are dissolved (you can reuse the pellets after checking or cleaning the traps). The pellets are provided by NZ BioSecure.
See also section 4.3.10 Mosquito Control.

4.3.2.5 Adult Trapping

There are many different types of adult traps that can be used to attract and trap mosquitoes. The type should aim for the “fit for purpose”, depending on a target species, the purpose of monitoring breeding activity, or reducing population numbers, searching for egg laying females or host seeking females.

Many traps have been invented by researchers and are custom made for field work. But there are also multiple commercial traps available - not all of them are suitable for use.

Any given area might have several different mosquito species flying around and each species has a different life history -therefore responds to different attractants-. For example, some *Aedes* mosquitoes feed on people during the day and rely more on sight when seeking a meal. *Culex* mosquitoes also attack birds, are normally nocturnal and track prey by smell, such as nonanal for example.

While different brands and models of mosquito traps utilise different methods of attracting and killing mosquitoes, there are several common features found on mosquito traps. How well those features work, or whether they are available on all models, vary.

4.3.2.5.1 Traps for host seeking mosquitoes

<u>Light Traps (LT)</u>

Figure 4.3.5. Light trap setted up in the field.

Light is an attractant and can be used by itself, but is more effective when used in combination with CO_2, which is a powerful attractant because it represents the respiratory product of an animal host.

There are several types of "light traps" available; generally using dry ice or a CO_2 gas cylinder:

- CDC (Centres of Disease Control) light trap
- EVS (Encephalitis virus surveillance) traps
- PB Light Trap

The advantages of these traps are that they can be operated all night and can collect large numbers of insects. They can be large and permanent, or simple and portable, and there is a variety of power sources available.

There is a variation in species and numbers collected using light alone or light plus carbon dioxides as attractants. Traps baited only with light usually collect a variety of insects.

To target host seeking insects such as adult female mosquitoes, CO_2 baited light traps can be employed. Experiments have shown that CO_2 used in conjunction with other items may increase the number of mosquitoes and the range of species collected. Other chemicals such as "Octenol" have been shown to be a useful attractant for certain species.

The use of CO_2 –baited light traps is recommended for adult trapping at all ports. Where the ports are close to saltmarsh areas, and saltmarsh adults are being targeted for surveillance, the additional attractant octenol must be used. It should be noted that octenol may be repellent to some exotic mosquitoes so it is important the purpose of trapping is established before traps are set out.

These traps attract adults from a considerable area through a screened funnel into a killing jar or mesh bag suspended below the trap.

The light trap is usually made up of a small incandescent bulb, a fan and a catching container. Traps may be powered by batteries or AC power via an adaptor or inverter. CO_2 gas may be provided via dry ice or from bottled gas via a suitable calibrated regulator system. Expert advice on attractants and appropriate trapping methods is obtainable from the MoH contractor, New Zealand BioSecure.

Trap positions

It is suspended about 1.2-1.5 m above the ground in a sheltered position but with a wide range of 'view'. It should be placed in a position where it does not compete with other light sources and care may have to be taken to prevent ants from invading the trap. To be effective the traps should be placed near vegetation, out of windy exposed conditions and well away from congregations of animals such as cattle yards or horse paddocks. They should have a relatively clear line of sight to all the surrounding areas (that is they should not be placed in the middle of very thick tall grass or at the side of a building).

The trap site should be selected with care keeping in mind the purpose for which the trap is to be set and the possibility of vandalism or theft. There are few reasons for adult traps to be always sited routinely at the same location, rather than moving them around the port area or rotating them around different sites in order to more comprehensively sample the local or introduced fauna. Fixed site monitoring is usually undertaken to monitor relative abundance in an area, rather than to detect the presence/absence of particular species, and where the latter is the principal objective, the rotation of traps between sites within a locality can give a more effective coverage of risky areas, if compatible with the workforce available.

Well-vegetated harbourage areas should be sought out for trap placement, but traps should not be placed directly within dense shrubbery.

Traps are hung from tree branches or other supports and sited in sheltered areas away from competing light sources and animal hosts, and operated from at least one hour before sunset to at least one hour after sunrise.

Traps should not be in direct contact with vegetation, and the string or chain can be coated with Vaseline, to prevent ants from getting into the trap and destroying the collection. Traps should be sited upwind of any major mosquito habitats in the area, and multiple traps should be separated by at least 50m. Traps may be located within large buildings such as warehouses used for devanning as *Aedes aegypti* and *Aedes albopictus* are often found within or close to buildings and people.

A major disadvantage of adult traps is their security at the ports. Security is not usually a problem at the major airports however. Respect private property!

Erecting a Light Trap

- Position as closely as possible to a high producing breeding site.
- If at all possible place trap away from where it may be tampered with.
- Hang trap in a tree where possible, but if there are none suitable nearby, erect a tripod.
- Ensure the tripod is placed on level ground and is secured upright.
- Hang the light trap from the tree or tripod, using the chain.
- Secure the collection cup onto the light trap with a rubber band.
- Secure the gas bottle to a metal stake with another chain and padlock.
- Attach the regulator to the gas bottle using the spanner.
- Tape the tube from the regulator to the light trap and position correctly.
- Secure the red and black leads on to the battery – red to positive, black to negative.
- If required place the Octenol tube in the clip provided.
- Turn the gas on and check it is flowing.
- Check the trap fan is operating and the light bulb is working.

The ideal positioning of light traps will vary depending on the species being targeted but in general:

- Within sight of potential habitat.
- Trees provide shelter from wind and rain and sometimes provide food for adult mosquitoes.
- Avoid too much competition from other light sources.
- Avoid direct competition from other sources of blood feeds - a trap placed in the middle of a room full of people will catch a much lower proportion of mosquitoes.

Refer to Appendix 1 and 2 for the standard operating procedure.

Equipment

- Light trap with light bulb
- CO_2 gas cylinder
- Gas regulator and flow regulator
- Spanner
- GPS unit
- Charged Battery
- Chain and Padlock
- Labels for collection cup
- Toothbrush for cleaning
- Tubing (at least 3m per kit)
- Collection cup with stocking
- Spare bulbs, stockings, catch cups, rubber bands
- National Mosquito Trap Collection Sheets

- **If applicable:**

- 3 metal stakes (or wooden) for a tripod
- Hammer (Sledge) – for set up
- Insulation tape, wide and thin
- Leatherman – multi tool for cutting
- Octenol bottle and tube with pipe cleaner

Before you leave, organise charged batteries and equipment needed for current servicing run and place in vehicle. NB: Ensure that as many clean collection cups as are required, as well as a couple of spares, are taken on each trip and the stockings do not have any holes.

Processing

- Close the catch cup by twisting the stocking in a loop.
- Always remove collection cup before disconnecting the battery [disconnecting the battery will stop the fan that prevents any captured mosquitoes from escaping].
- Use the rubber band (or pegs) to secure the cup, do **NOT** tie a knot (this might cause holes in the socks).
- Ensure the cup is labelled correctly (date of collection, site ref, sample number, sampler name).
- All collection cups removed from the field should be stored in a chilly bin or similar, and placed in the freezer on arrival back at base.
- Check the fan housing is clear of debris such as insects and clean with a toothbrush if necessary.
- Attach the collection cup at base of trap. **Ensure the stocking does not have any holes in it** and that the rubber band is sufficient to hold the cup in place.
- Change the battery - Attach trap connections to battery – Red to positive terminal and Black to negative terminal.
- Change gas bottle and reconnect the regulator.
- Record any changes in habitat or location at which light trap is placed on the National Mosquito Trap Collection Sheet. Significant changes to the location could reduce the likelihood of the trap collecting mosquitoes.
- Before leaving the trap, **ensure that the gas is on, the fan is blowing, the light is going, and that the light source is not blocked by any object**.
- Ensure the National Mosquito Trap Collection Sheet is completed before leaving each site.
- In addition, staff working near to light trap sites should be encouraged to report mosquito biting activity as this may be more sensitive than the trap itself.
- *On return p*lace collection cups in freezer to kill mosquitoes – these need to stay there at least for 2 hours.
- Recharge batteries as necessary (Refer to battery instructions for direction on recharging). Keep uncharged batteries in designated spot, away from those already charged.
- You may use the checklist for processing adult traps Appendix 2.

Trap results can vary markedly from one site to another due to the proximity of the vegetation, exposure to wind, the effect of lights and other less obvious factors. Trap results should be recorded on a collection form.

Sample identification relies on the microscopic examination of specimens. Most identification relies upon the position of morphological features such as scales and hairs. These can easily be damaged or removed completely so careful handling and packaging of all specimens is required. Follow the instructions in the standardised guidelines to avoid unnecessary handling of and damage to specimens (see 4.3.6 Sample Handling).

Therefore, it is preferable that they are not examined before being sent through. There have been a number of occasions when specimens have been unable to be identified fully due to damage sustained during previous examination.

Trap Maintenance

- You may send the traps and regulators to NZ BioSecure for servicing once a year or if not working properly. Otherwise:
- Ensure the fan is unclogged and the fan housing is clean.
- Unscrew the motor and fan.
- Apply CRC to the joints and screws.
- Check the battery clamps are clean.
- Ensure that the regulator and all parts and leads are working and the regulator is calibrated to release the appropriate amount of gas over the duration of the trap placement (e.g. one week). The gas bottle needs to be near empty so you can determine the trap has been working when a trap is serviced (change in weight).

BG Sentinel Trap

Figure 4.3.6. BG Sentinal trap. Arrows show the convection currents.

The BG sentinel trap was designed to attract *Aedes aegypti* but has since been found to be effective at attracting a range of other mosquitoes. It utilises a fan system to draw mosquitoes into a collection pot in a similar way to a light trap; however, it is the way the attractants are emitted that is different. The trap mimics convection currents created by a human body, releasing attractants through a large surface area. The trap generally employs a BG-Lure, which releases a combination of other attractants: ammonia, lactic acid, and fatty acids as an attractant, however you may use CO_2 in conjunction with the lure.

One of the main drawbacks is that the trap has been designed for indoor use and its current design is not suitable for use outdoors in inclement conditions and should be placed away from strong winds, rain and direct sunlight, preferably 1m away from walls.

Target species without CO_2: *Aedes aegypti, Aedes albopictus, Aedes polynesiensis, Culex quinquefasciatus, Culex pipiens.*
With CO_2: *Ochlerotatus* spp., *Anopheles* spp. and *Coquillettidea* spp. and also Simuliidae, Ceratopogonidae.

For instructions in how to assemble and process a BG trap see appendix 3.

4.3.2.5.2 Gravid traps

Figure 4.3.7. Sticky ovitrap.

Sticky ovitraps capture the gravid females as they attempt to lay eggs but are stuck to the glue surface (Figure 4.3.7). They are actually not used anymore as the sticky film destroys the ID features. In Cairns, Australia this method has been employed for trapping blood fed adult mosquitoes which may be sent for PCR analysis to identify presence of arboviruses.

Figure 4.3.8. Standard gravid trap.

Standard gravid traps employ a combination of ovitrap attractants and light trap mechanics. The trap consists of a body of water similar to a large ovitrap which attract females ready to oviposit. A fan and collection pot system is mounted over the water and is designed to suck the females up into the collection pot as they fly over the water body (Figure 4.3.8).

As with lethal ovitraps this system removes the mosquitoes from the environment so is effectively also a form of control.

GAT (Gravid Aedes Trap)

Figure 4.3.9. GAT trap.

A recently developed novel trap (passive trap) has proven successful in capturing large numbers of Aedes, *Anopheles*, *Culex* and *Verrallina* mosquitoes without the use of power. These rely upon luring mosquitoes into a translucent “passive” trap from which those mosquitoes, attracted to the light entering the translucent body, cannot find their way out.

The passive trap approach was adopted to capture gravid females of container breeding mosquitoes. The new trap called GAT (Gravid Aedes Trap) uses a translucent top above a black bucket of infusion. Knock-down surface spray, or the optional blue LINNs, can be used to reduce the potential for escape of adult mosquitoes once trapped.

The GAT is a useful tool for capturing adult *Aedes aegypti* and may be suitable for other container-inhabiting species such as *Aedes albopictus* and *Culex quinquefasciatus*.

The GAT, used for surveillance programs is inexpensive, do not require electrical power, are not labour intensive, nor do they require difficult usage of adhesives, which damage the specimen

(sticky ovitraps). The GAT is useful wherever container-breeders are suspected. We suggest using GATs mainly at seaports but also at airports. Since they are very light weight they should be secured or positioned somewhere sheltered from the wind.

For instructions in how to assemble and process a GAT see appendix 4.

4.3.2.5.3. Traps for virus testing

Passive box trap

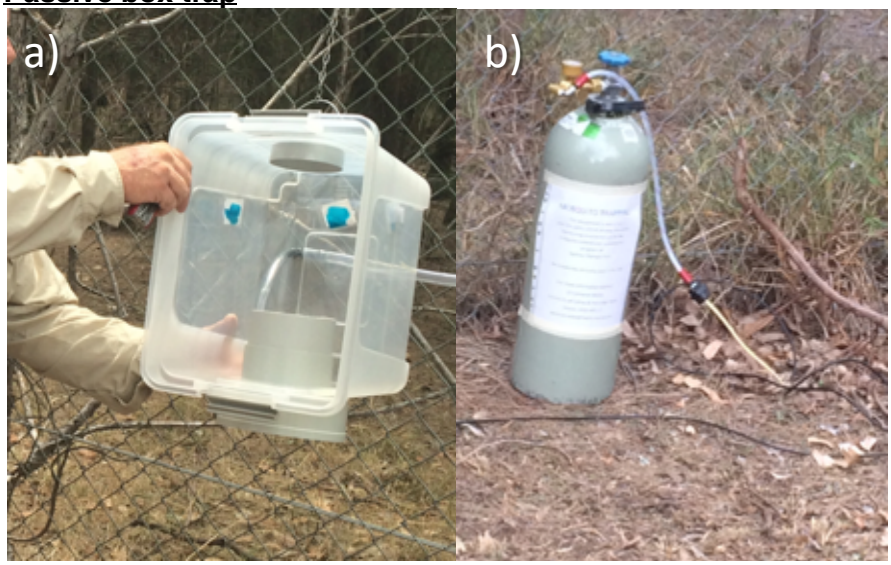


Figure 4.3.10. Passive box trap (a) connected to a CO_2 cylinder (b).

This trap has no light attractant but carbon dioxide. Inside the box there are several pieces of sticky paper with a sugary solution for the Mosquitoes to lick at. The saliva can be directly tested for any viruses (Figure 4.3.10).

4.3.2.5.4 Commercial traps

These traps attract a vast variety of insects and are, in general terms, useful to reduce insect populations. The use of these traps in the surveillance context is under discussion. If the trap is already available, we recommend to set the trap indoors and strongly recommend do not set it up outdoors since the trap is also attractive to a number of native insects that play important roles in nature such as pollination and biological control of pests.

Dominator

Figure 4.3.11. Dominator trap.

This trap uses a combination of mosquito lures: black light, heat and colour to lure and eliminate mosquitoes. Optional Octenol sachets are available to control mosquitoes in your back yard.

Actuated Gate System employs a plastic board downwind of the motor which is opened by air from the fan and closed by spring. When trap is powered "off", captured insects cannot fly away upwards due to the plastic board.

The location of the trap is a critical variable in maximising capture rates. Most mosquito species avoid direct sunlight and wind so we recommend shaded, sheltered areas (Figure 4.3.11).

Mega-Catch™

A line of traps that includes four models: the Alpha, Premier, Premier XC and Ultra. These traps are multi-attractant, run off 12 Volt power and rely on an integrated lighting display, infrared heat

and fragrance strips (Octenol or the combination Octenol/Lactic acid lure) to attract mosquitoes. The Ultra and Premier XC models can also use Carbon Dioxide (CO_2).

Pro 900 ALPHA Mosquito Trap (MCA-900)

Figure 4.3.12. Pro 900 ALPHA mosquito trap.

Ideal for small yards, patios and gardens. Lightweight, compact and portable. Constructed from resilient and weatherproof ABS. Heat pulsing from a central core heat system with digital pulse width modulation channel (simulates subtle changes in human body temperature).
Maximum range 27 metres, maximum coverage - 0.2 hectare. Rated for both indoor/outdoor use. Central light array incorporating multi-frequency UV LEDs (light emitting diodes).Designed to work with or without attractants, Octenol fragrance strips have been specifically formulated to boost capture rates of nuisance mosquitoes as well as sand flies, black flies and biting midges (no-see-ums).

All weather fan design with stainless steel ball bearings.

PRO 900 ULTRA Mosquito Trap

Figure 4.3.13. PRO 900 ULTRA Mosquito trap.

CO_2 comes ready with Mega-Catch's™ patented 'Variable Quantity Slow CO_2 Gas Release System'. The PRO 900 PREMIER XC can be made CO_2 capable with the addition of a CO_2 Gas Upgrade Kit (sold separately).

Operation of the optional CO_2 Gas Attractant System enables the traps to replicate a key feature of vertebrates respiration by releasing small quantities of CO_2 into the air plume emanating from the base of the trap.

Dragonfly

Figure 4.3.14. Dragonfly trap

This brand produces only one model, the Dragonfly II Biting Insect Trap. The trap is electric and also uses CO_2 from small canisters, along with infrared heat, a night light and attractant lures. The Dragonfly II is programmable with 8 operating modes and the CO_2, which is optional, is released in pulses every 5 seconds. Made by BioSensory Inc, in 2009 the Consumer Product Safety Commission (CPSC) announced a voluntary recall of the Dragonfly II mosquito trap. The company and the CPSC said that the "carbon dioxide (CO_2) pressure sensors inside these products can crack and leak or burst, causing the release of CO_2."

CO_2 is released every 5 seconds simulating the "breath" of a small animal. CO_2 is the most powerful attractant for mosquitoes and biting flies. The Dragonfly II monitors the amount of CO_2 remaining in the bottle and lets you know when the bottle is getting low by flashing a low CO_2 warning.

The thermal lure heats up, producing the infrared image of blood near the surface of the skin and the body temperature of a small animal. Octenol, and other natural host-odour attractants, are activated by the heat of the thermal lure, and evaporate into the CO_2 plume. In scientific terminology host odour attractants are called kairomones. Both CO_2 and octenol are kairomones. CO_2 is the most powerful attractant, and octenol is the second most powerful

attractant for mosquitoes and biting flies. When additional host-odour attractants such as lactic acid are added to the mix, the effectiveness increases still further. BioSensory biting insect lures use several combinations of host-odour kairomones, depending on the targeted insect.

Koolatron™

(Formerly Lentek™ International Inc.) currently markets 3 traps including the Guardian MK14 Bite Shield (Cordless), Guardian MK12 and Koolatron™ MK05 Champion Mosquito Trap.
The Guardian and Champion models are both electric and use octenol, heat and a lighting array. The Guardian models also burn propane to produce CO_2. In 2003, Koolatron™ purchased the business assets of Lentek™ International Inc., and took over the company.

Figure 4.3.15. Koolatron trap

Mosquito Magnet®

This brand now consists of the Executive, Independence, and Patriot models. Earlier models which included the Defender, Pro and Liberty, attracted a number of poor reviews from customers indicating problems with start up, and propane blockage/clogging issues. The traps use CO_2 together with secondary attractants in the form of Octenol or Lurex cartridges. Although some models are battery powered or electric, they all burn propane to produce CO_2. Created by American Biophysics Corp., when the company went into receivership in 2006, Woodstream Corporation purchased its assets.

Mosquito Magnet® Executive

4 fuel saving modes controlled via an easy-to-read LCD panel and a durable rigid net.T hese features can extend the life of accessories and propane up to 40%. Cordless. The coverage includes one full acre of land.
Automatically adjusts to external conditions ensuring optimal mosquito control when it's needed while virtually eliminating propane and attractant waste.

Figure 4.3.16 Executive Mosquito Magnet trap.

SkeeterVac SV5100

The SkeeterVac SV5100 is a cordless unit that creates its own power to energize the suction fan by converting propane to electricity using a catalytic converter. The by-product of the propane is CO_2 once ran through the catalytic converter.

When the fan powers up the exhaust from the fan blows out the unit, CO_2 and an attractant called Octenol used in the BaitBlock along with heat and moisture is expelled from the SV5100.

Figure 4.3.17. SkeeterVac SV5100

Mosquitoes are drawn to the CO_2, Octenol, moisture and heat thinking it is a mammal. Some mosquitoes will get stuck on the sticky TacTrap which is the black and white portion of the trap surrounding the SV5100 engine.

Mosquito research has shown that mosquitoes are attracted to dark colours. The design of the TacTrap with the white and black contrast tends to make mosquitoes fly toward the wider portions of the TacTrap which is where the suction from the fan pulls the mosquitoes into the permanent net where they will dehydrate and die. Eliminates mosquitoes and other backyard insects.

Easily lights with the AA battery powered electronic ignition. CO_2 rating is 35,000 cc/hour. Assembles easily in minutes; no tools required.

Measures 38 x 20 x 26 inches; weighs 34 pounds.

Flowtron MT275

Figure 4.3.18. Flowtron MT275 trap.

Attracts and kills mosquitoes and biting flies.
Uses bright LEDs and powerful vacuum to draw bugs in.
Uses no open flames, nets, or glue panels.
Dispose of insects with no sticky mess.
Measures 17 x 14 x 35 inches; 1-year limited warranty; 30' cord included.

Mosquito Slayer Pro

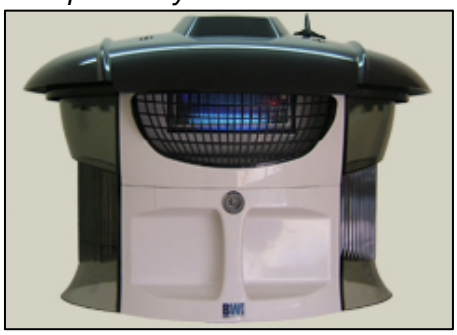


Figure 4.3.19. Mosquito Slayer Pro trap

Large capacity commercial and residential mosquito and biting midge trap.
It is successful in large or smaller areas - uses a suite of scientifically proven attractants – includes a Gas Regulator Hose - to maximise the catching of female mosquitoes and midges (biting). CO_2 is regarded as one of the primary attractants in mosquito traps.
Slayer Pro will cover up to 1 acre and comes with a 10m 12v ext.

Slayer Series 4 Gas

Figure 4.3.20. Slayer Series 4 Gas trap

The Mosquito Slayer Series 4 (Gas) Unit dispenses a controlled flow of carbon dioxide from a gas regulator hose which connects to a bottle of carbon dioxide.
Unit comes with an all-weather transformer with 5M electrical cord and a CO_2 Modulator 38.

The Slayer 4 gas will cover 1/2 acre.

Black Hole Mosquito Trap

Figure 4.3.21. Black Hole Mosquito Trap

By producing carbon dioxide, the Black Hole lures and captures mosquitoes and sandflies (but also other flying insects such as moths and flies that are attracted by black light). The Black Hole is easily installed around homes, gardens, swimming pools and outdoor eating areas, farm buildings, restaurants and bakeries, schools and military compounds etc. Especially suited to areas with high mosquito populations and areas with mosquito borne diseases.
Effective area 66-165m^2. Units should be placed outside and not exposed to the weather. For optimum performance, lamps should be changed every 3000 hours.

Tests
BG sentinel traps have proven to be the most efficient ones (Williams et al., 2006; Irish et al., 2008) but they need a power source and are not to be used in hard weather conditions.CDC and EVS light traps are the standard traps used in Australia and most effective (Ritchie and Kline, 1995). The passive box trap is used in Australia and has a very positive catch rate (e.g. for *Aedes aegypti* and other *Aedes* species).

In independent testing, the Mega-Catch brand usually seems to come out on top, and the results are impressive – thousands of insects in a single night, in many cases. At those rates, it only takes about two months to collapse a local mosquito population.

4.3.2.5.5 Lures
Fragrance
Most traps offer some variety of fragrance strip or lure. Mega-Catch, Dragonfly, Mosquito Magnet and Koolatron for instance, all offer octenol strips as an accessory, which is effective on some, but not all mosquito species. In 2009 Mega-Catch released a combination lure, which incorporates synthetic octenol, lactic acid and other ingredients specifically formulated to attract the Asian Tiger (*Aedes albopictus*) and other nuisance mosquitoes, as well as sand flies, black flies and biting midges (no-see-ums). Octenol attracts *Aedes* species in particular but is not qualified for *Culex* species.

A pheromone attractant (perspired decay enzyme) derived from mushrooms (fungi bi-product/composting slime) in tablet form is available. A close analogy is human body odour (or the smell of sweaty socks).

Like Lactic acid, the reviewed literature indicates that octenol seems to work best in conjunction with CO_2 and other physical attractants. Evidence shows that it may be selectively attractive for some mosquito species and varieties of insects (eg. *Aedes* but not *Culex*). However, some of the literature indicates that Octenol is a “must have” and seems to react synergistically with CO_2. Lactic acid, synthesise body odours (major component of sweat) are most effective when coupled with warm moist surfaces/airflows. Without CO_2 it is reported to be ineffective. Lactic acid excites neurones in a wide range of female mosquitoes and biting insects.

Investigations are continuing for information on other components of skin-pore emissions (NaCl) as the formulation of the “Lactic-lure” used in the traps appears to be about 10-15% glucose-based syrup, 1-2% surfactant (non-odorous industrial detergent), <1% citric acid and 83% water.

Heat
A heating element coupled with the light unit produces a radiating warm surface and probably increases volatilisation of attractant molecules. There is some conjecture in the literature on the

actual role of moisture and humidity. Perspiration at body temperatures seems ideally attractive. (NB: Accumulated sweat/BO is attractive to mosquitoes).

Heat alone will not attract mosquitoes but moist heated airflow in the range of 30-40 deg C may, and it appears to complement molecular dispersion and the catching capabilities of other attractants.

Mega-Catch, Dragonfly, Mosquito Magnet and Koolatron use some form of heat emission to help attract mosquitoes once they are close. The heat source usually is located near the trap intake system.

Light
The Mega-Catch has the most sophisticated of the light systems, with an array that flashes both visible and invisible spectrums at oscillating frequencies tested and proven to appeal to mosquitoes. The Dragonfly II has a night-light and Koolatron traps use a blue light system. Mosquito Magnet traps do not seem to offer this feature.

Mosquitoes have a complex ocular system that is sensitive in low light conditions covering the approximate spectral regions 320-620nm (UV- green/yellow not into the red-IR) with sensitivity peaks in the UV (330-345nm) and in the green (523nm). Some work suggests that lights covered with yellow and red filters (630+nm) make the light invisible to most mosquitoes. A 12-volt UV (fluorescent) tube in the 350-370 nm region is well-accepted technology and entomologically supported. The light used has a peak wavelength of 365 nm. An additional multi-spectral source is provided by 4 Light-Emitting Diodes (LEDs) connected into a collimated 60 deg prismatic disperser/refractor. These white, blue (468), green (520) and red (626) are sequentially pulsed producing the perception of movement that is also thought to assist mosquito orientation. It is not clear what this cycling does in terms of spectral attraction but the disperser may provide a wider spectrum of attractive wavelengths that may be selective to some other insects. Black-surfaced traps catch most mosquitoes.

Carbon Dioxide
Carbon dioxide cylinders/dry ice, available at the local air product supplier and BOC Gases, should always be considered. It increases the numbers of captured specimens, as well as the number of species, to include the unwanted exotic *Aedes albopictus* and *Aedes aegypti*.

EVS, PB, BG traps use CO_2. The CDC light trap can also run with Carbon dioxide. The usage of carbon dioxide is highly recommended to increase the number of the species as well as specimens. Mega-Catch offers a CO_2 system as an additional component of its Ultra and Premier XC traps. The Mega-Catch system has 5 settings that allow the owner to program a slow, timed release of CO_2 from cylinders like those used in soda fountains. Mosquito Magnet and Lentek/Koolatron burn tanks of propane for CO_2. Dragonfly II uses pure CO_2 (in canister form). Ambient background CO_2 levels are well mixed and relatively uniform in the atmosphere at <0.5%. Human breath usually contains <5% CO_2, (approximately a ten-fold increase). Mosquitoes can detect changes of the order of 0.01%.

Food-grade bottles of CO_2 (20-30 kgs = approx up to 10,000 minutes of operation, or 100 minutes per day for 100 days). Estimates in advertising of a bottle lasting 2-3 months (US$10/mth bottle rental and US$30-40/refill) seem reasonable. Competitors (Mosquito Magnet Pro) use BBQ-grade Propane (9.5kg) to produce CO_2 after burning. The combustion also produces moisture and heat and a bottle lasts 10-20 days, cost $25/refill.

Bottled CO_2 needs to be supplied to users at regular intervals and appears to have some advantages over manually controlled and ignited Propane based systems.

Trolley mounted unit and gas bottle is possible with trailing power cord.

4.4. Response to Suspected or Confirmed Exotic Mosquitoes of Public Health Significance

A mosquito response includes any actions taken to prevent the establishment and spread in New Zealand of an exotic mosquito of public health significance e.g. in the event of a (suspected) interception or incursion.

MPI has the lead role in responses to interceptions and incursions of all exotic organisms other than mosquitoes or rats (aboard a craft or in direct association with a craft only), which are the responsibility of the MoH.

A contingency plan should be developed and maintained by each public health unit for responding to biosecurity risks of public health significance (refer to the MoH manual Section 5 Biosecurity).

4.4.1 Exotic Mosquito Interception Response

Detection and Triggers

- If a live exotic mosquito of public health significance (unwanted species) is detected at the border we speak of an **interception** (Figure 4.4.1);
- This may be through an adult exotic mosquito being found during an inspection of imported item(s) at the border (seaport or transitional facilities) or flying in the terminal of an international airport, associated or not, wth a piece of luggage;
- Once a detected specimen has been recognised as being a mosquito, the response becomes the responsibility of the MoH;
- Most, but not all, interception events are initiated by MPI Border Staff who will on detecting evidence of a possibly exotic mosquito:
  - implement immediate containment action and
  - notify the PHS on call staff.

Phase 1 Notification

- PHU is notified of a suspected exotic mosquito being detected at a POE or TF;
- On Call HPO takes facts of finding including notifier's details, location, risk goods, flight number or Vessel name, life stage detected;
- The HPO should report the identification and location of exotic mosquitoes of public health significance to the CTO (Health), or the Deputy CTO (Health) within one hour and notify the NZB on call entomologist as soon as possible on-call phone 021 522 476.
  In preparation to visit index site, following equipment should be packed:
  - PPE
  - Traps, adult & larval (+CO_2 cylinders and gas fitting tools)
  - S-methoprene pellets
  - Knock down insecticide spray
  - Sample collection kit
  - Smart phone with macro lens

Phase 2 Deployment – Delimiting Survey

- Before entering the site MPI or AP should be contacted, and the Owner/Operator should be notified;

- The specimen needs to be picked up (after check of mosquito-features and a completed MPI Handover Certificate) ;
- If not already done, information about the circumstances are to be requested:
  - Dead or alive when found?
  - What development stage, adults possibly escaped?
  - Where and when found?
  - Associated with goods from which country?
- A picture of the specimen should be taken and sent (with any information about the case) to taxonomy@nzbiosecure.net.nz (notification of the delivered picture by text to on-call phone);
- After receiving the pre-screening results, the CTO/DCTO needs to be notified what the specimen is;
  - Lab confirms it is not a mosquito. No handover is required.
  - Mosquito possibly of local origin (endemic or introduced species) – no further action is required for the time being, the specimen needs to be shipped to New Zealand BioSecure for proper ID, overnight courier can be discussed and needs approval of DCTO.
  - Possibly exotic - The specimen needs to be shipped via urgent courier to New Zealand BioSecure for identification as soon as possible with advise of shipment details. Exotic identification results should be received within 24 hours; independent verification will be obtained and reported when available (this may take some time).
- Meanwhile the site/risk goods need to be examined;
- At least one trap should be assembled and an **initial survey** of breeding habitats should be conducted including checking all traps in place and treatment (S-methoprene) of potential mosquito breeding habitat. The record of GPS coordinates data is necessary;
- Course of Action (COA) needs to be developed for the case of a confirmed interception by seeking advice, organising additional resources if they are needed, (staff, material or equipment);
- After confirmation of an intercepted unwanted species (see flowchart below highlighted in red) fumigation of risk goods or risk conveyance needs to be arranged;
- A **full delimit survey** of the airport, port and any vessel is to be executed, as well as eliminating potential habitat, installing additional traps for adults and larvae and treating of the area;
- Directives for cleansing and disinsection needs to be issued.

Phase 3 Administration

- CTO/DCTO update on activities;
- Manager update on actions taken;
- Occupier, Port company and Port stakeholders briefing and advise on measures taken and follow up action;
- Equipment and material check and service;
- Contact the shipping agent and obtain voyage details including transit ports and onward destinations (domestic and foreign) and advise any public health units where the vessel is berthing and/or unloading risk goods;
- If an exotic mosquito was intercepted a SITREP needs to be sent to MoH and New Zealand BioSecure on the first working day unless requested to expedite this report and send it earlier (Appendix in the Manual section 5 Biosecurity provides a sitrep template for

exotic invertebrates). In case the mosquito is native or introduced an e-mail report should be sent;

- Planning to sustain the Response Surveillance program (if an exotic was detected).

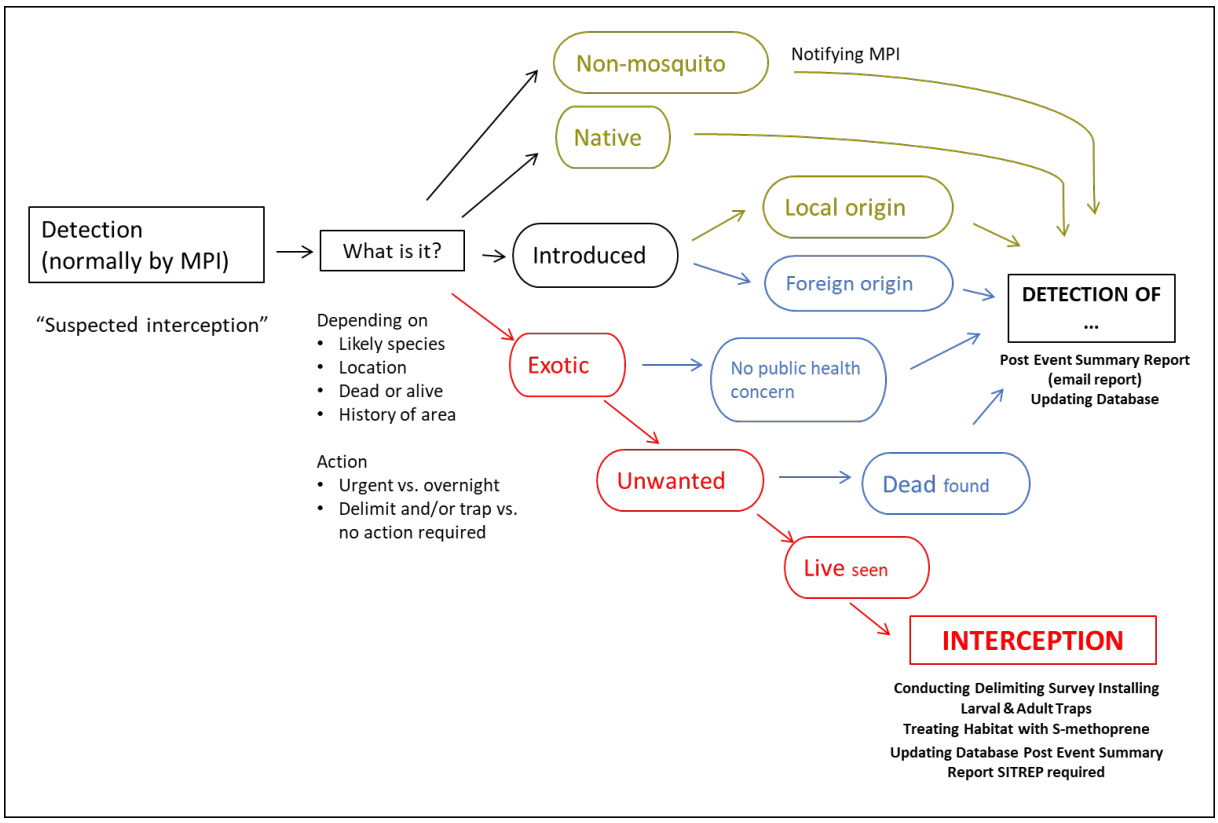


Figure 4.4.1 Interception response flow chart.

Phase 4 Post deployment – Enhanced Surveillance

- Post Incident Surveillance maintaining enhanced monitoring for three "life cycles" of the species (21 – 28 Days) plus more delimiting surveys;
- Frequency of trap processing needs to be discussed with CTO/DCTO and New Zealand BioSecure and depends on the season;
- Collection of any samples of the traps are to be dispatched to New Zealand BioSecure for ID;
- Maintaining ongoing reports to CTO/DCTO of activity at Interception site including nil returns;
- If no additional exotic mosquitoes are found following three weeks of enhanced monitoring, this would indicate that a population is either absent or below the levels of detection. At this time, the response would be terminated and routine surveillance resumed;
- Internal review of Response.

➢ Exotic mosquitoes found during routine surveillance at the point of entry

- Alternatively, exotic mosquitoes can be found during routine surveillance (probably detected during the identification process at New Zealand BioSecure lab);
- In this situation an exotic mosquito may have breached the quarantine system and breeding may have already occurred;
- A delimiting survey and enhanced monitoring must be activated (see above);

- Adulticide treatments of appropriate harbourage sites must be considered supplemented by the prophylactic treatment of larval habitat with slow release or residual products;
- These decisions need to be technically based and tailored to suit the specific profile of the exotic mosquito or mosquitoes being dealt with;
- The response will refer to an exotic mosquito incursion response. Public health staff are expected to cooperate with, and work alongside, MPI staff until a transition is seamlessly completed;
- If, after appropriate surveillance, no additional exotic mosquito biomass was found, this would indicate that a population is either absent or below the levels of detection. At this time, MPI will terminate the incursion response routine public health unit surveillance is resumed. The finding of additional exotic biomass will mean the incursion response continues.

4.4.2 Exotic Mosquito Incursion Response

In this situation, exotic mosquitoes are found breeding in New Zealand. Immature or adult stages of exotic mosquitoes may be confined to one small area (e.g. a property or suburb) or may be found at multiple locations or over a widespread area.

MPI will lead any incursion response but may require support from public health staff. This support may include alerting stakeholders (particularly medical practitioners), to field assistance. Public health staff may also be required to initiate secondary disease control measures such as promotion of mosquito avoidance behaviour, provision of advice on arboviral disease risks and management and enhanced arboviral surveillance.

4.4.3 Investigation of public information or complaints

Follow-up investigation and taxonomic identification of suspected exotic mosquitoes submitted by the general public should be undertaken within three working days of the complaint being received. (Unusual biting activity led to the discovery of *Aedes camptorhynchus* in Napier in 1998 and in Blenheim in 2004).

4.4.4 Mosquito Personal Protection

Mosquito repellents applied to exposed skin surfaces, bed nets, or window curtains are the most widely used and successful of these measures. Nearly all repellents applied to skin contain DEET; bed nets and curtains are usually treated with a synthetic pyrethroid, such as permethrin or deltamethrin. Loose fitting, long sleeved and long legged clothing, as well as bed nets, can be effective even without the use of repellents, if kept in good repair and used properly.

Vaccines and prophylactic drugs also fall under this category. Unfortunately, there are few effective vaccines available for human use, exceptions being the highly effective 1 7-D Yellow fever and Japanese encephalitis vaccines. Prophylactic drugs are available that both prevent and *cure* malaria infections, but widespread resistance by parasites to many of the drugs in various parts of the world continues to hamper their use.

4.5. Sample Handling

DO NOT ATTEMPT TO REAR OUTSIDE OF AN APPROVED FACILITY. ALL REARING OF MOSQUITO LIFESTAGES MUST BE PERFORMED WITHIN A CTO (HEALTH) APPROVED INVERTEBRATE CONTAINMENT FACILITY.

The following details the procedure representing best practice handling of mosquito specimens on returning from the field to the office:

4.5.1 Mosquito Larvae

Figure 4.5.1. Sample tube containing larvae.

- Working with only ONE sample tube at a time, carefully pipette out as much of the habitat water as possible (without removing any larvae or pupae).
- Immediately after add ethanol into the tube. The final concentration should be 70%.
- DO NOT put cotton wool, tissue or any other padding in the sample tube with the larvae as they can get trapped in the padding and become damaged or desiccated. The tube should contain larvae and ethanol ONLY (Figure 4.5.1).
- DO NOT allow the larvae or pupae to dry out at any stage.

4.5.2 Mosquito Adults

Figure 4.5.2. Sample tube containing adult specimen

- Place all light trap collection cups in the freezer overnight or at least 2 hrs to kill the adults.
- Work with only ONE collection cup at a time, leaving the rest in the freezer if possible, to prevent the condensation wetting the specimens.
- All adults should be handled carefully to ensure no wings or legs break off and as few scales are removed as possible as these are important features for accurate species identification.
- If necessary to handle an adult specimen, use fine forceps to carefully hold the femur of the middle leg on either side of the body.
- Wet adults must be carefully removed and placed to air dry on blotting paper. Place the specimens out of direct sunlight and in a draft-free area and leave for half an hour before packaging for dispatch. [Do not attempt to wipe or blot any excess moisture as this will remove scales and damage the specimens.]
- Do not add any alcohol.
- Place the specimens in a sample tube of the appropriate size for the number of specimens present (small, medium or large).
- Place the specimens OUT of direct sunlight and in a draft-free area. (If possible, place in an incubator or hot-water cupboard) and leave for half an hour before carrying on with the rest of this procedure.
- Using fine forceps, taking care to remove any larger non-mosquito specimens such as moths. When in doubt as to whether a specimen is in fact a mosquito adult, always place it into the tube so the New Zealand BioSecure laboratory staff can make an accurate identification.

- Carefully position tissue paper (NO COTTONWOOL as the legs can get tangled in the fibres) in the top of the tube to help prevent the adults being shaken around the tubes too much during transit.
- DO NOT squash the tissue paper down onto the adult specimens.
- Complete for all collection cups.

4.5.3 Ovitrap Processing

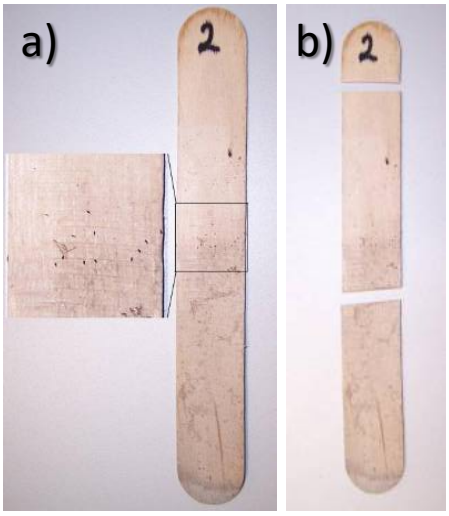


Figure 4.5.3. a) wooden paddle with eggs at watermark and close up of eggs, b) paddle cut off.

- Remove the paddle from the plastic transport box and examine for eggs. They should be near the water mark on the paddle (Figure 4.5.3a). Use a dissecting microscope or magnifying glass if available to examine the paddles more closely.
- If eggs are visible, wooden paddles should be packaged into individual slide mailers for transport (Figure 4.5.4b and c).
- Cut off the lower part of the paddle just below the water line using scissors and trim off the top. Be sure not to cut off any eggs. (Figure 4.5.3b).
- Write on the sample number and date and lay the remaining part onto a piece of tissue (Figure 4.5.4a).
- Wrap the tissue around the paddle and place into a slide mailer (Figure 4. 5.4b).
- Close the slide mailer and make sure it is labelled correctly (Figure 4.5.4c).

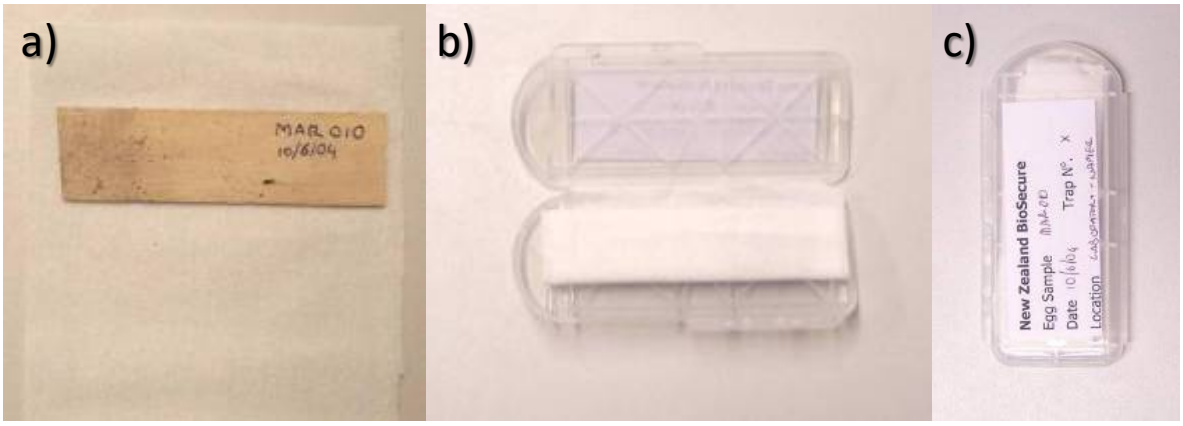


Figure 4.5.4. a) wooden paddle with sample number and date, b) Tissue wrapped around the paddle and c) paddle inside a slide mailer.

- Tip all the water from the jar into a white plastic tray.
- The trap jars should be examined by eye for mosquito eggs that may have been laid at the water surface. If present, attempt to remove the eggs without damaging them, so they can be sent through to the laboratory. If this is not possible, sterilise the jar with boiling water.
- Examine the water in the tray for the presence of larvae, pupae, exuvia (skins) or adults.
- Remove all larvae (instars 1-4), pupae, and exuvia using plastic pipettes and transfer to a labelled sample tube. The label should be written in pencil, contain the trap sample number with the collector's initials and the collection date. NB: Larval skins can also be used for species identification.
- If any adults are present in the water, remove carefully and place into a separate labelled specimen tube.
- Complete the corresponding Trap Collection Sheet (positive or negative).

- At least two sets of jars and paddles are required for each trap site and should be used on a rotational basis. One set is in use, while another set is being sterilised and dried.
- Additional paddles will be required if any are sent through to the laboratory for hatching.

4.5.4 Packaging for Transport

All collections, larval or adult, should be labelled with sample number, date, site of collection and collector name, before transported for identification. Refer to Appendix 6. Mosquito Sample Collection Sheet for the required information that should be registered in the field.

Accurate identification of exotic mosquitoes intercepted at importation on vessels or after introduction to ports, should be seen to be a critical component of the programme. The effectiveness of remedial actions will be dependent on the efficiency of the reporting system that, in turn, is dependent on the veracity of the identification process.

The importance of accurate identification cannot be stressed enough, and there must be a confidence within the system that the reported identifications from all sites are accurate. To achieve this, the Ministry contracts in expert mosquito identification services. This service is required to have their identifications of exotic mosquitoes independently verified by an external, overseas expert. The practice of local identification by public health units should not be undertaken as it leads to process delays, may damage potentially exotic specimens prior to arrival at the laboratory, is not backed by quality standards and duplicates the use of expensive equipment and resources.

When packaging paddles containing eggs for transport to the laboratory it is important to ensure the paddles do not touch each other and damage the eggs. The paddles must be lying flat with the eggs on the upper surface and be wedged so that if the package is inverted they cannot move and rub on the lid. Paddles should be placed in a dedicated transport box like those used for transferring the paddles from the field and fastened in place with two corks per paddle, which are stabilised with Blutack. If only one paddle is being sent, it may be desirable to use a smaller box.

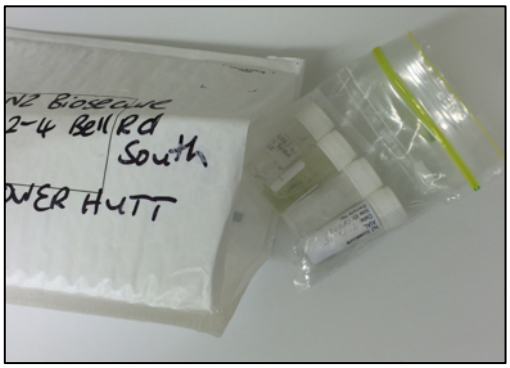


Figure 4.5.5. Samples ready to be sent to NZB.

All samples must be securely packaged and carefully labelled for transport.

1. All tube and container lids need to be secured tightly to ensure there is no leakage or loss of liquid or specimens in transit.
2. Tubes and containers should be clearly labelled with the sample details written **in pencil**. [If a tube leaks, the alcohol will not affect labels written in pencil].
3. Place all tubes of each type into plastic zip lock bag(s), i.e. dry adult specimens separate from larval samples in alcohol.
4. Ensure the sample tubes are also separate from any sample or information sheets to prevent them getting damaged if any tubes leak during transport.
5. Place the zip lock bags containing sample tubes in either plastic, padded envelopes (courier or mail), HANDIboxes, or in bubble-wrap in an unpadded courier bag.
6. Mark the package as FRAGILE and send to the following address:

Mail	New Zealand BioSecure PO Box 38-328 Wellington Mail Centre Lower Hutt	**Courier**	New Zealand BioSecure 2-4 Bell Road South Gracefield LOWER HUTT

7. Email the New Zealand BioSecure entomologists to let them know a package will be arriving, include the sampling date. taxonomy@nzbiosecure.net.nz
8. If you have not received a reply within three working days, contact New Zealand BioSecure to check the sample has been received.

Try to organise your routine surveillance sampling early to mid-week so samples will arrive at the laboratory before Friday. As there is no post on Sundays results could be delayed if samples are sent late in the week.

If specimens are to be sent to New Zealand BioSecure for identification and further work such as genetic analysis or storage, they need to be packaged appropriately so they arrive in good condition.

4.5.5 Non-routine samples (interception or incursion specimens)

All mosquito specimens that have been found at a POE outside the routine surveillance need to be packaged and sent by courier as soon as possible after alerting the laboratory (urgent courier - same day courier or as discussed on phone with New Zealand BioSecure laboratory staff). See also section 4.3.3.

4.6. Record Keeping and Reporting

Mosquito surveillance data collection in the field has historically been carried out using paper sample forms or notebooks (from which the data is then transferred over to sample forms), and then the data inputted into computer databases. It cannot be stressed enough how important accurate record keeping is for mosquito surveillance. It is needed to allocate any sample to a date and a location to understand the distribution and sometimes even the behaviour of the species. This applies especially to historic data. If the records are accurate people that joined the program later can still understand the data from many years ago.

4.6.1 National mosquito surveillance database

The Online National Mosquito Surveillance Database is a single, centrally housed database for all District Health Board mosquito surveillance which is accessible via the internet. The database is securely stored by an independent IT company, supplying a degree of physical security that should protect it against all but the most extreme disasters. It is frequently backed up, and has a power supply with several backup contingencies, and is also protected by an industrial firewall which has continually updated virus protection.

There are several advantages associated with the database system:

- Single, consistent national dataset with regional access to regional data subsets - All the data is stored as a single dataset, however access to data can be controlled regionally
- Reduced data handling - Reduced risks of data integrity loss. Also auditing of data occurs as results are added, ensuring the data is accurate and correct once the record is complete
- Ability to produce ad hoc reports - The database has the facility to run ad hoc reports based on combinations of any parameters included in the dataset
- Increased data control – The database rules can preclude submission of sample information until all critical information is supplied
- Data is current - All data is complete and up-to-date as it is added as the samples are collected as well as when the specimens are identified

There will be several user levels which will be allocated as appropriate.

New Zealand BioSecure laboratory staff will have access to the full dataset for all regions. They are able to add results, update records after auditing if necessary, run ad hoc queries on the full dataset, and allocate permissions to other users.

Each public health unit will have their own Health Board Administrator as well as Sample Creators who have access to the data for their region only. Health Board Administrators: will have the ability to:

- add new records (excluding results)
- run ad hoc queries
- edit previously entered records
- allocate other staff with ‘Sample Creator’ permissions

Once the data has been entered and submitted, it can only be viewed and queried, but cannot be changed or deleted, except by those users with the appropriate permission.

Sample creators chosen by the administrator have the ability to add new records (excluding results), and run ad hoc queries.

After the samples have arrived to the laboratory and are identified, the results are entered into the database by the laboratory staff and emails are sent to all relevant public health units to advise of the results.

New Users

If you are not a registered user, and require access to the database, you should contact your local Health Board Admin or the NZBEL Entomologists via the taxonomy email (taxonomy@nzbiosecure.net.nz). They will provide you with an access user name and password.

Once registered, open your internet browser and navigate to database login page, at http://www.nzbiosecure.neocom.geek.nz/Sampling (National Online Mosquito Database, Figure 4.6.1). It is recommended that you bookmark this page or create a shortcut from your desktop, to save typing the internet address each time you access the database. The site can also be accessed through the SMSL Website
https://www.smsl.co.nz/NZBEL/Entomology+Laboratory.html.

Logging In

Enter your username and your password and click the "Login" button. The menu page will appear.

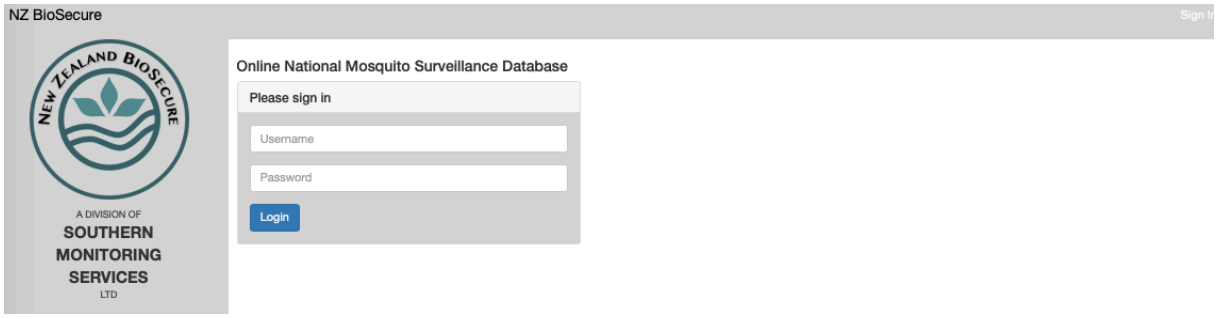


Figure 4.6.1. Login page for the National Online Mosquito Database.

Entering Sample Information

Samples can be entered singularly or in multiple by uploading a csv file.

Entering New Samples

On the welcome page (Figure 4.6.2), select “New Sample” from the left hand menu, a new “Create Sample” form will appear (Figure 4.6.3).

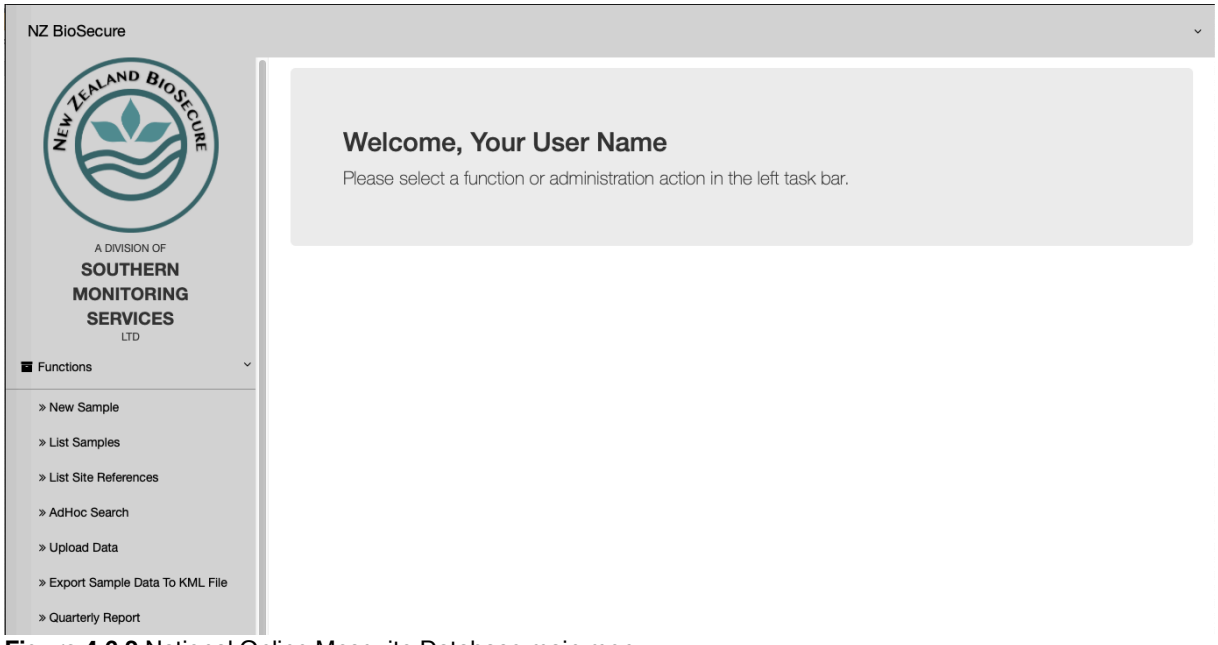


Figure 4.6.2 National Online Mosquito Database main menu.

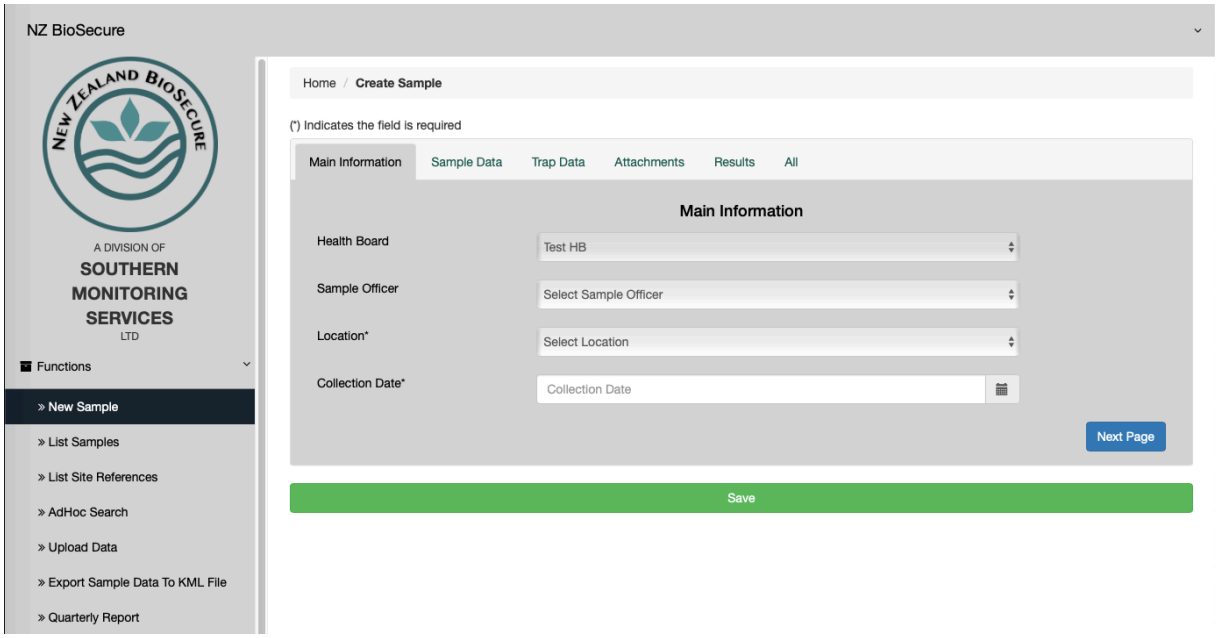


Figure 4.6.3. "Create a Sample" Main Information Tab.

"Main Information"

Select your name under "Sample Officer", "Location" and complete the "Collection Date" by clicking on the collection date or calendar and choose the date. Then select "Next Page" to view Sample Data Tab. Then select "Next Page" to view Sample Data Tab (Figure 4.6.4)

"Sample Data"

Several fields are now compulsory. They are denoted by the (*) beside the field name (Figure 4.6.4). Fields may contain drop-down menus where you can select from a list. Some of these are accompanied by an option to add new information to the drop down lists - Green Tabs positioned to the right.

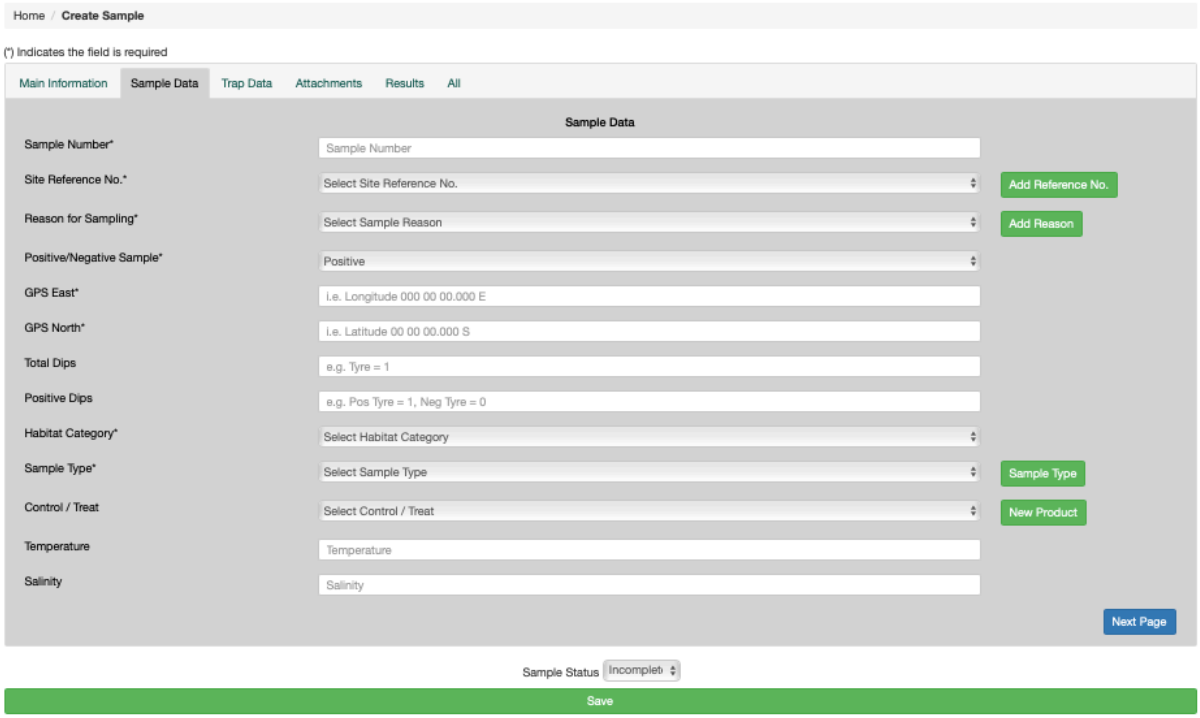


Figure 4.6.4. "Sample Data" Tab.

NB: The coordinates must be recorded and entered using Latitude and Longitude with degrees, minutes and seconds (Lat Lon (DMS)) under WGS84 datum following this example.

“GPS East”: 000 00 00.0000 E
“GPS North”: 00 00 00.000 S

It will read like the following on your GPS unit:
Longitude: 174º 54’ 57.836” E; GPS North: 41º 13’ 59.826” S

The coordinates should be entered using the following format in the online database e.g.
GPS East: 174 54 57.836 E; GPS North: 41 13 59.826 S.

Note that when entering the coordinates the following characters are not required (º “ ‘ -).

For larval samples, both the “Total Dips” and “Positive Dips” are required to be entered manually, and for all adult and larval trap samples, the number of “Trap Nights” must be entered.

NB: Enter habitat category: For traps select “Trap option”

NB. The “Positive/Negative” field will automatically display “Positive”, and needs to be changed to “Negative” manually, where required.

“Trap Data”
When complete select “Next Page” the “Trap Data” tab will be displayed (Figure 4.6.5)

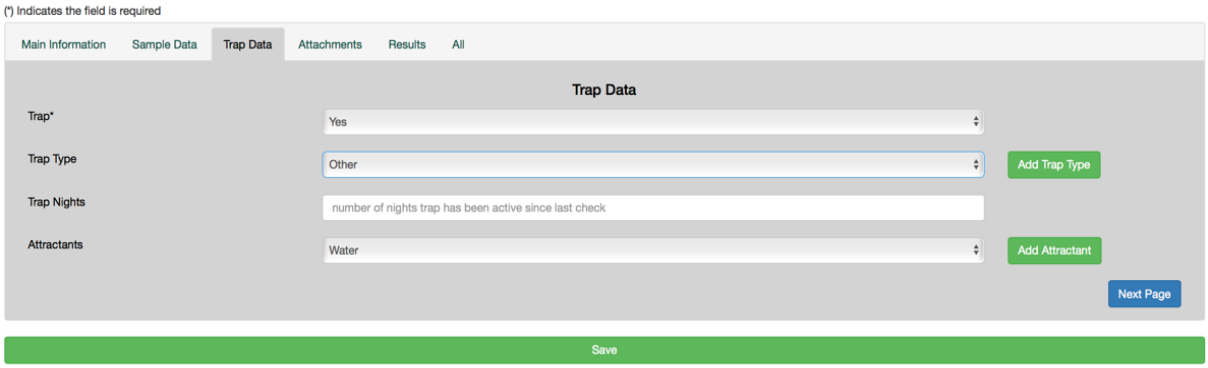


Figure 4.6.5. “Trap Data” tab.

NB. The “Trap” field will automatically display “Yes”, and needs to be changed to “No” manually, where required.

Select the correct option from the drop-down list for “Trap”, “Trap Type”, “Attractants” and enter the number of trap nights.

“Attachments”
Select “Next Page” to upload “Attachments” (Figure 4.6.6).
You may attach a file, a photograph or report to samples, by selecting “Choose File”. Your browser will direct to your file source for selection. When you save the sample form, the file will copy onto the database and be saved with the sample data. This is ideal for interception sitreps and specimen images. You can add comments in the comments section.

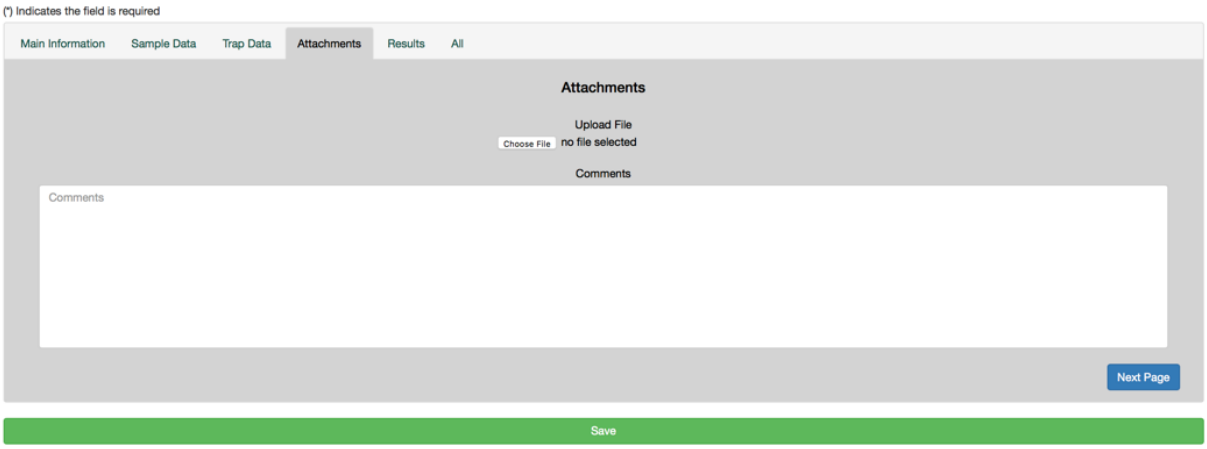


Figure 4.6.6. To attach a file click on “Choose File”.

Saving the Sample Information

When all the information for the sample has been entered, click the Green "Save" Bar at the bottom of the page.

If there are compulsory fields not entered a pop-up will occur (Figure 4.6.7) indicating missing data. Navigate to the correct tab and enter the missing data. Saving can occur at any page.

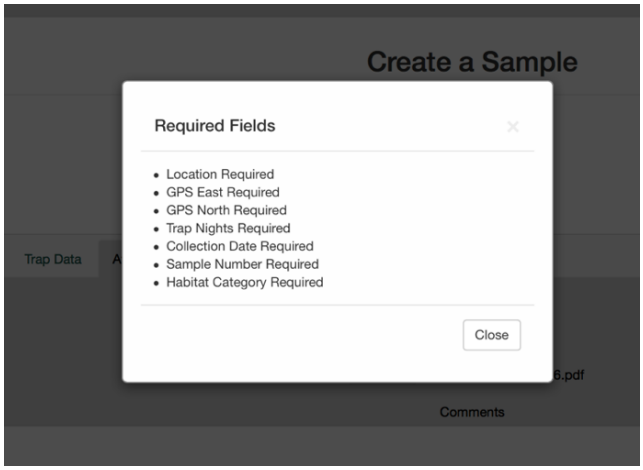


Figure 4.6.7. Missing data fields.

“All”

Alternatively, all the sample information can be entered in the “All” tab (Figure 4.6.8).

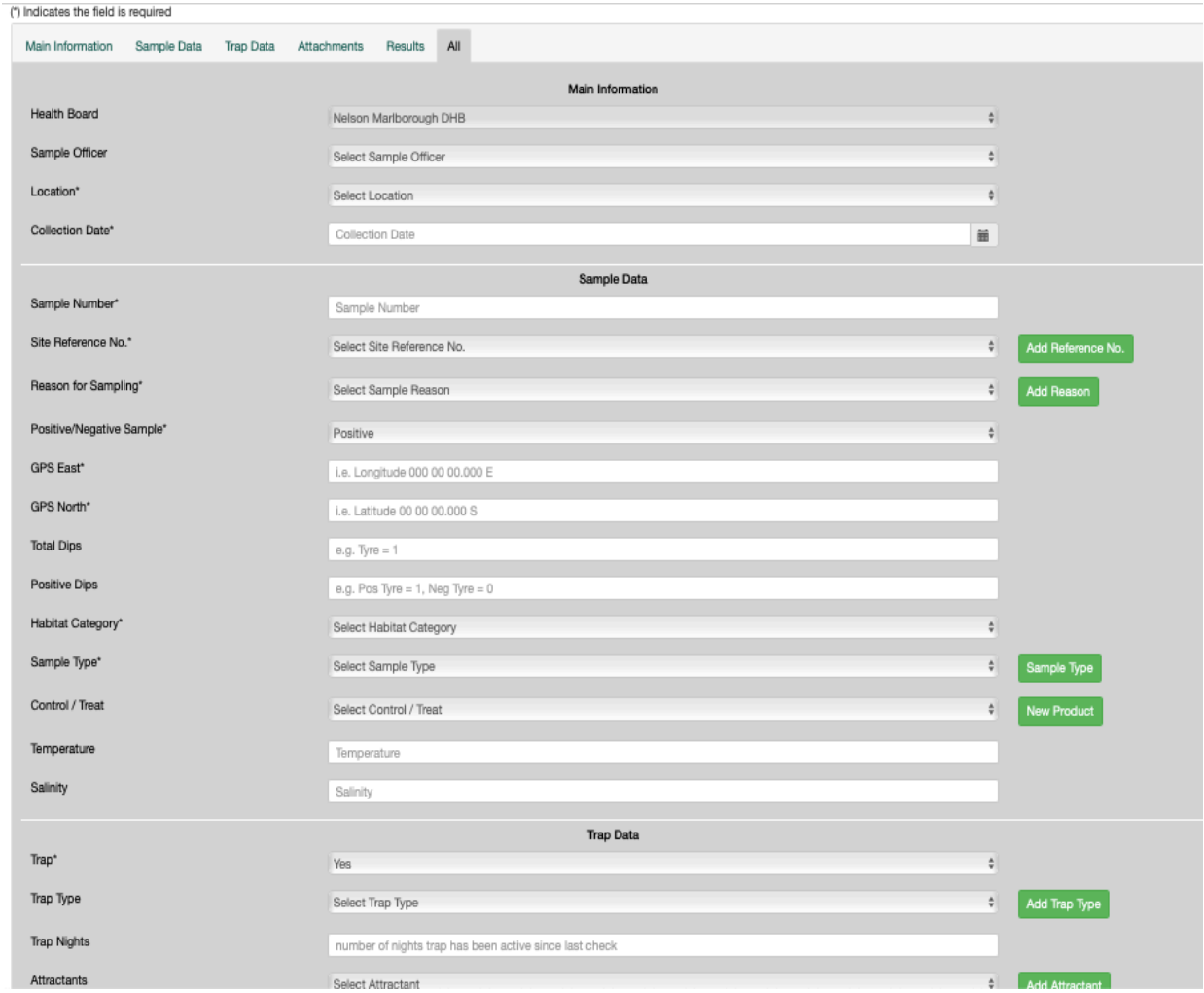


Figure 4.6.8. "All" Tab.

When all the information for the sample has been entered, click the "Save" button at the bottom of the page to save the record. The sample is then saved onto the database (Figure 4.6.9).

Figure 4.6.9. This message will appear if the samples have been saved.

If your sample does not save, i.e. you do not have a "Sample Saved" displayed at the top of the page (Figure 4.6.9). Click on the "back" icon on your internet browser and try clicking "Save" again. Repeat until the "Sample Saved" message appears on the screen. Failing this, contact your Health Board Admin or the NZBEL entomologists for assistance.

Entering Multiple Samples – Upload a CSV

The database has an upload function to add multiple samples in a CSV file. The CSV file format and example headers can be downloaded from the SMSL website (Entomology Laboratory page): http://www.smsl.co.nz/NZBEL/Entomology+Laboratory.html

The CSV file must follow a specific format (Figure 4.6.10), this cannot be altered.

NB: Samples from new site references need to be entered manually the first time. Alternatively, a new site reference may be created in the database prior to upload a csv file.

NB: The date entered must be in the following format yyyyMMddHHmmss.
e.g. Original Date: 2017/07/27 09:05:02;
Import Date Format: 20170727090502

SAMPLE_OFFICER	COLLECTION_DATE	LOCATION	SURVEILLANCE_TYPE	SITE_REFERENCE_NO	POSITIVE_NEGATIVE_SAMPLE	SAMPLE_NUMBER	TOTAL_DIPS	POSITIVE_DIPS	CONTROL_TREAT	TRAP	TRAP_TYPE	TRAP_NIGHTS	REASON_FOR_SAMPLING	ATTRACTANTS	HABITAT_CATEGORY	SAMPLE_TYPE	COMMENT
The name of the sample officer which existing in	The sample collection date, the date is UTC.	The location name which existing in SMSL's	Port/Airport s Saltmarsh sur General Routine Surve Public complaint Coastal MAFQS inspection Response Public enquiry Interception Sentinel site Post-interception Audit Survey Delimiting survey TLA Sentinel Site Post-audit check MPI sighting Seaport Survey Airport Survey Review Enhanced surveillanc	The site name which existing in SMSL's	positive negitive	Any number o	Any number.	Any number	Bti S-Methoprene Bti + S-Methoprene Aquatain Agnique Mortien Barrier Spray/Bleac	yes no	New Trap Typ Adult Adult Light Trap Adult Trap BG trap Bucket Dominator Trap Electric Trap Front Door Light GATrap MPI Sticky Trap MPI Sticky Tras Not a Trap Other Ovitrap - Jar Ovitrap - Tyre Ovitrap Tyre/Jar Square Tins Tyre Trap	Any number.	5 km annual s Akld Council S alternate site Audit Survey Cesspit sampl Complaint Delimiting sur EnhancedSurv Event Followup to s Fumigation sa Ground truthi Interception MAF sighting Mega Survey MPI sighting NSP Place of First Arriv ponding water Post-audit check Post-interception	BG Lure CO2 CO2 & Li Human Light Light + Glue methoprene Octenol Octenol &am Octenol &am Octenol &am person Water Other New Attractant	Flowing Str Ponded Str Lake Edge Swamp Ma Permanent Temporary Intermittar Natural Co Artificial Cr Subterrane Subterrane	Adult beetle Bycatch capture by container casings Delimiting eggs Larvae mite Negative Pupae no New Sample Type	Any string comment s.

Figure 4.6.10. Headers and related values the database can read.

To upload multiple samples, download the CSV file from the website (Figure 4.6.10). Open as excel and enter sample data to each of the rows as specified in the header using the correct format. Save and name the file as a CSV to a known location.

NB: Fields and values in CSV should be separated by comma.

Login and select “Upload Data” from the left hand menu (Figure 4.6.2 and 4.6.11)
Select “Browse” to locate your file, and choose. Select “Upload”.

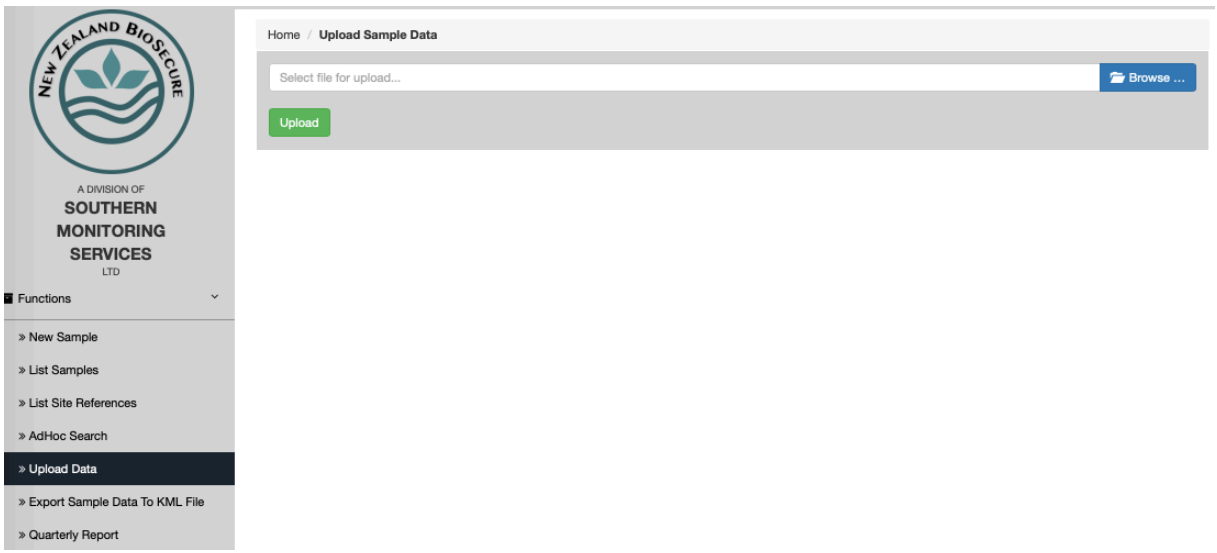


Figure 4.6.11. Upload Data.

A popup will display confirming upload success (Figure 4.6.12).

Figure 4.6.12. Popup display confirming upload and import data successfully.

Listing Samples

The "List Samples" link on the left hand menu allows you to view some of your samples for the current year or viewable archived datasets year. You can list your samples based on their "Status" (All, Complete or Incomplete), "+VE Sample" (All, Positive, Negative) or "Entry Method" (All, Uploaded or Manual). (Figure 4.6.13).

In “List Samples” be as selective as possible to find the samples, make your selections and click "Search". Your samples will appear in a summary table, from which you can check the results or edit your samples.

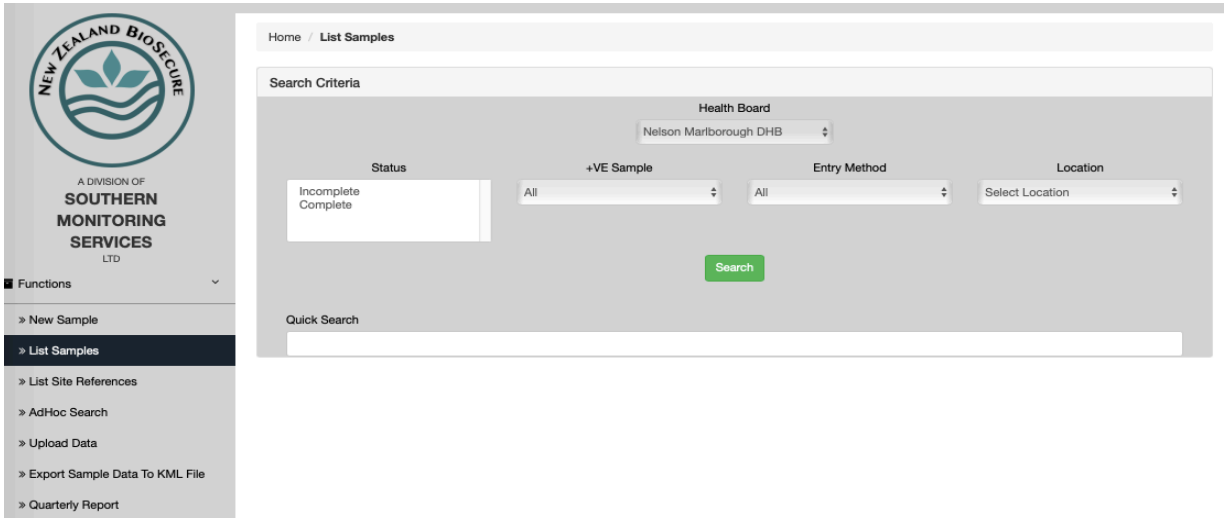


Figure 4.6.13. “List Samples” menu.

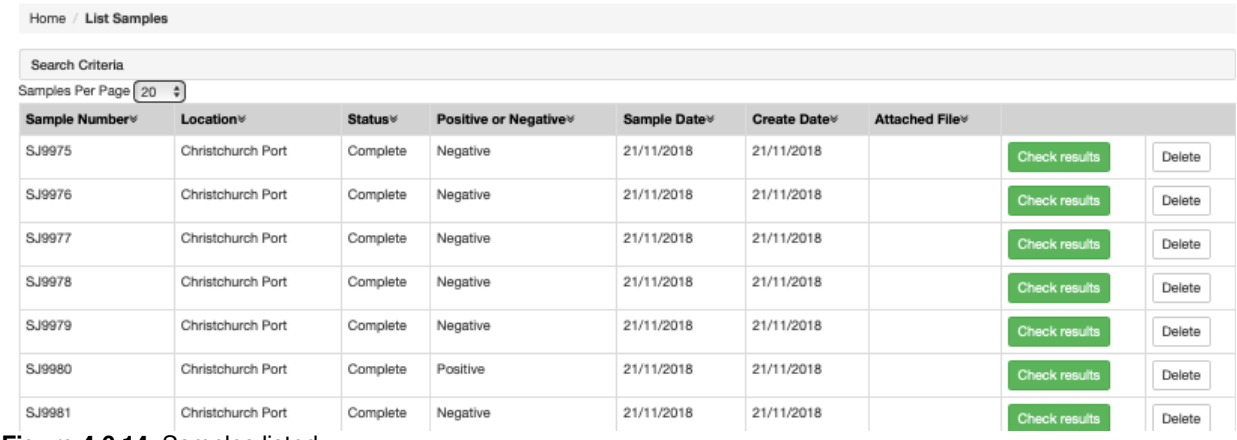


Figure 4.6.14. Samples listed.

You can also export a dataset to your computer by clicking from the “AdHoc Search” “Export CSV” (Figure 4.6.15). See below for instructions.

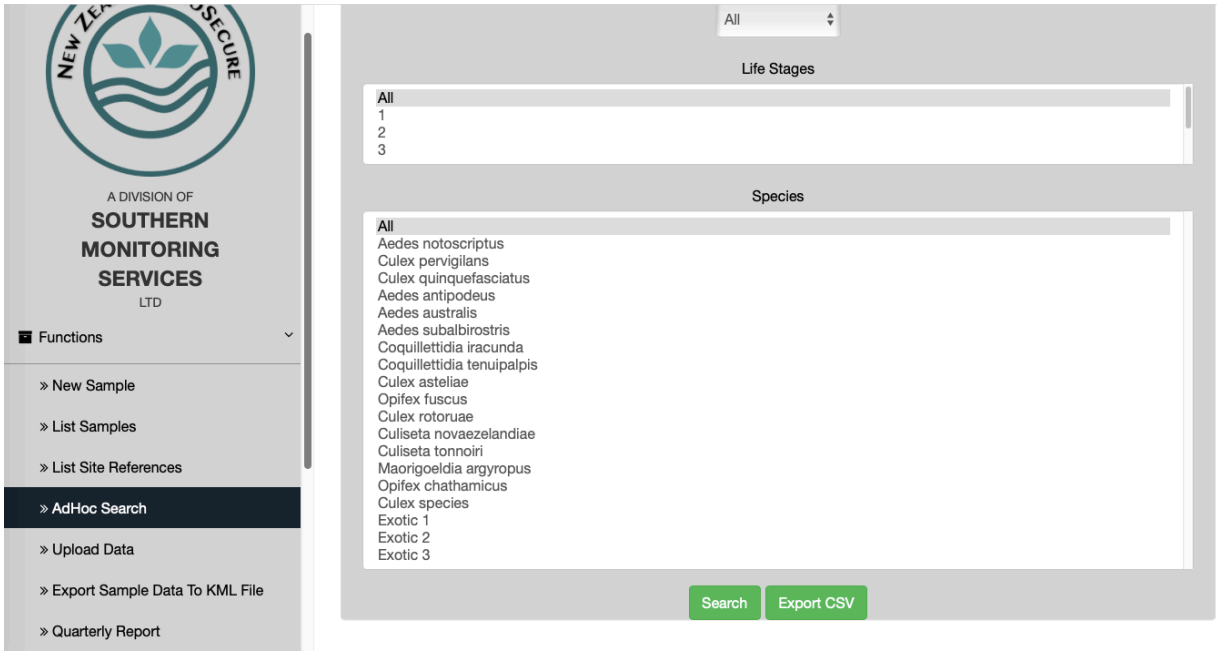


Figure 4.6.15. Export CSV function.

Site References

The database utilises an automated system for reducing the amount of data entry required for samples from fixed trap sites. The site details are manually entered into the database once, and then become automatically linked to the “Site Reference No.” field. From then on, the site detail fields are automatically filled in, once the appropriate “Site Reference No.” has been selected from the drop-down list.

Listing Site References

Click on the 'List Site References' link on the left hand manu, and all the site references for your Health Board will be displayed. From this page, you may add to the information relating to a particular site reference by clicking on 'Edit' adjacent to it (Figure 4.6.16).

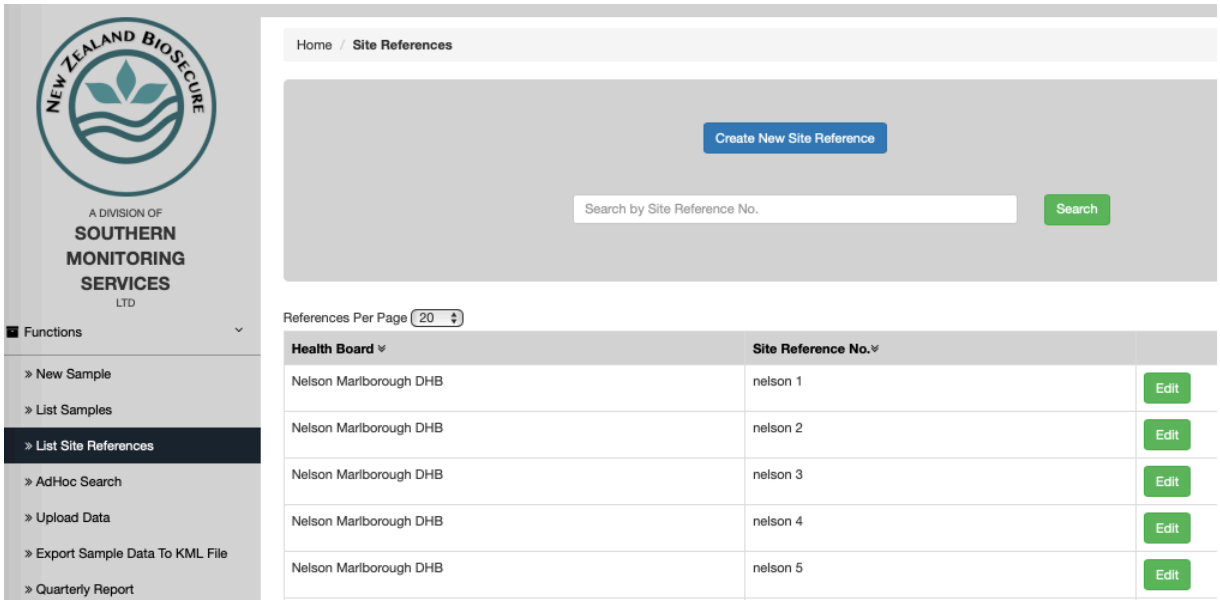


Figure 4.6.16. List Site References.

IMPORTANT NOTE: If you change ANY information in the site references, the changes will be reflected through ALL of the current sample records in the database which use that site reference. **If the change involves a moved trap, it is necessary to create a new site reference.**

Old site references can be removed by NZBEL, at the end of a database year once the archiving has been completed. Email taxonomy with a list of site references to be removed, and you will be advised by email when this has been completed.

To add a new reference, click on the blue button “Create New Site Reference” from the “List Site References” (Figure 4.6.16). Fill in the appropriate fields and click save (Figure 4.6.17). The new site reference will automatically appear in the dropdown menu for the next new sample you enter.

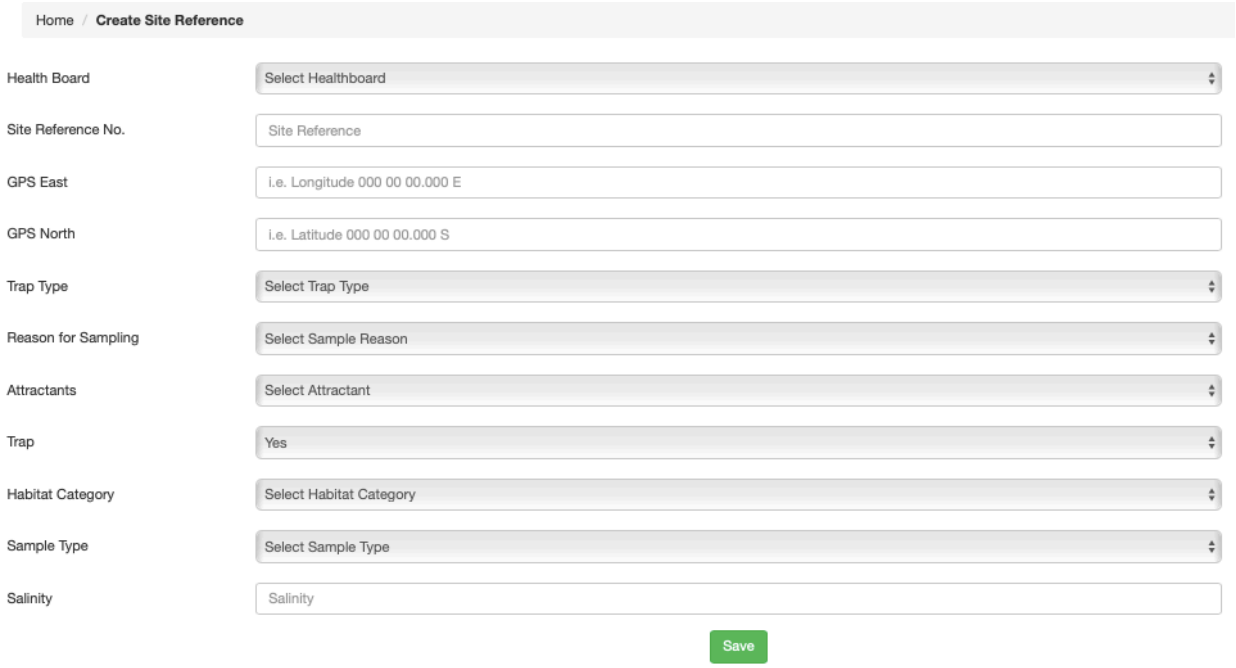


Figure 4.6.17. “Create Site Reference”.

Ad hoc searches

On the left hand Menu, click on "Ad hoc Search" to produce the Search Samples page (Figure 4.6.18).

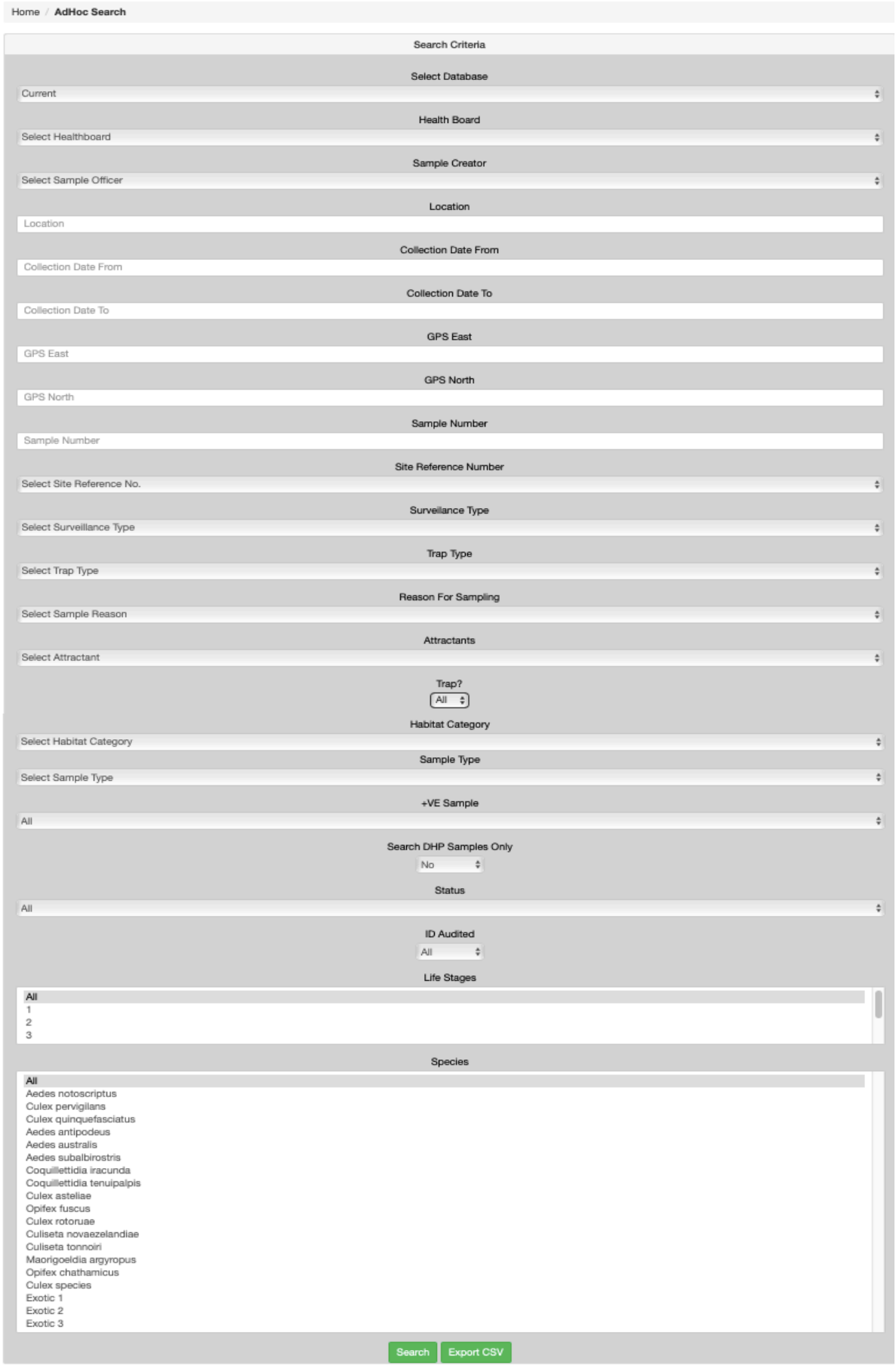


Figure 4.6.18. In the Ad Hoc Search menu refine your search.

Most fields from the sample records are included in this page, as they can be used to search and select the sample records. A single field or a combination of several fields can be used for searching. This is often the easiest way to search for samples.

Examples of searches; all samples collected on a particular date, all positive samples collected by a particular sampling officer during the month of April.

NB: A period of time must to be entered into the date fields to obtain the search results.

Once you have entered your criteria, click "Search" and you will be given the listing options as above in the Listing samples section. You can also export this dataset by clicking the "Export to CSV" button.

Finishing a Session
When you have finished with the database, click on your name at the top right of the current screen, click “Logout” to terminate your connection (Figure 4.6.19) and you will be returned to the login page (Figure 4.6.1) .

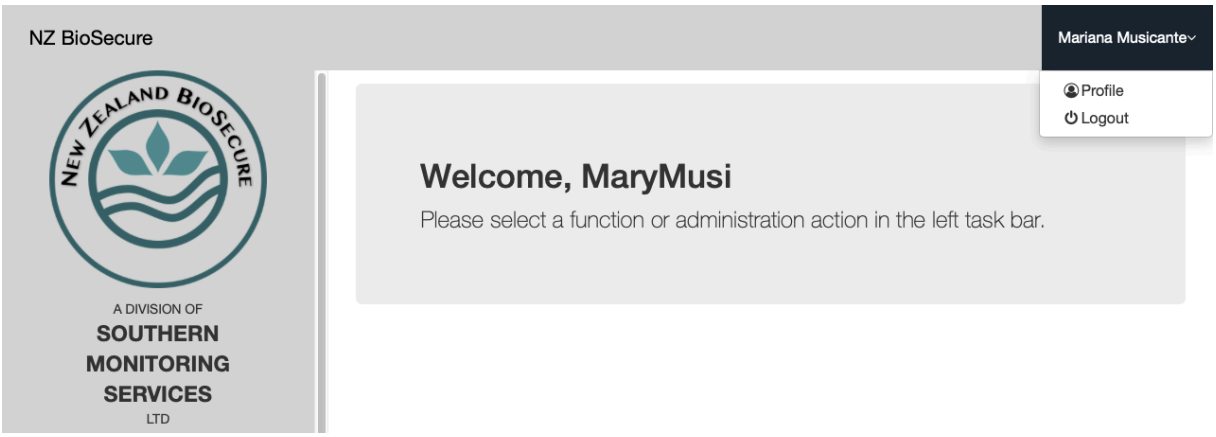


Figure 4.6.19. Functions shoing the Export data to KML file option.

Exporting Datasets

Data can be exported readily. The system is windows optimised and is the preferred option.

Ad Hoc Search Export Option
After you have searched or listed the sample records and obtained the data you wish to export, click on “Export to CSV” (Figure 4.6.20). The CSV should automatically download to your PC. Once downloaded it can be imported into excel for viewing.

Figure 4.6.20. “Ad hoc Search” results. Export to CSV.

Opening Exported Datasets in Microsoft Excel
Datasets exported as CSV files, can be imported into Microsoft Excel, for use in data analysis etc. This may not be automated, as the commonly used separator characters are often used in the data fields, the data may need to be imported manually depending on software versions.

Once you have exported the CSV file to a known location on your computer, open a new file in Microsoft Excel. Click on the “Data” menu, or “File” select the “Import” or similar option (e.g. Get data From Text) or “CSV file” (Figure 4.6.21).

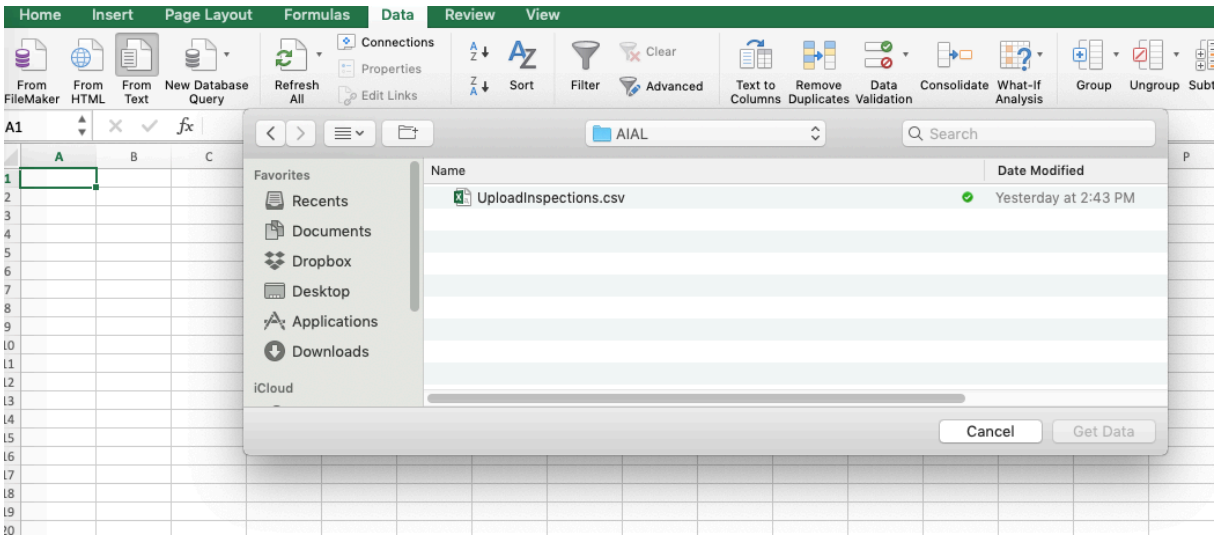


Figure 4.6.21. "Ad hoc Search" results. Export to CSV

A window will appear and prompt you to browse to the CSV file you wish to open in Excel. Once selected it will open a wizard that you can follow which assists with opening the file (Figure 4.6.22).

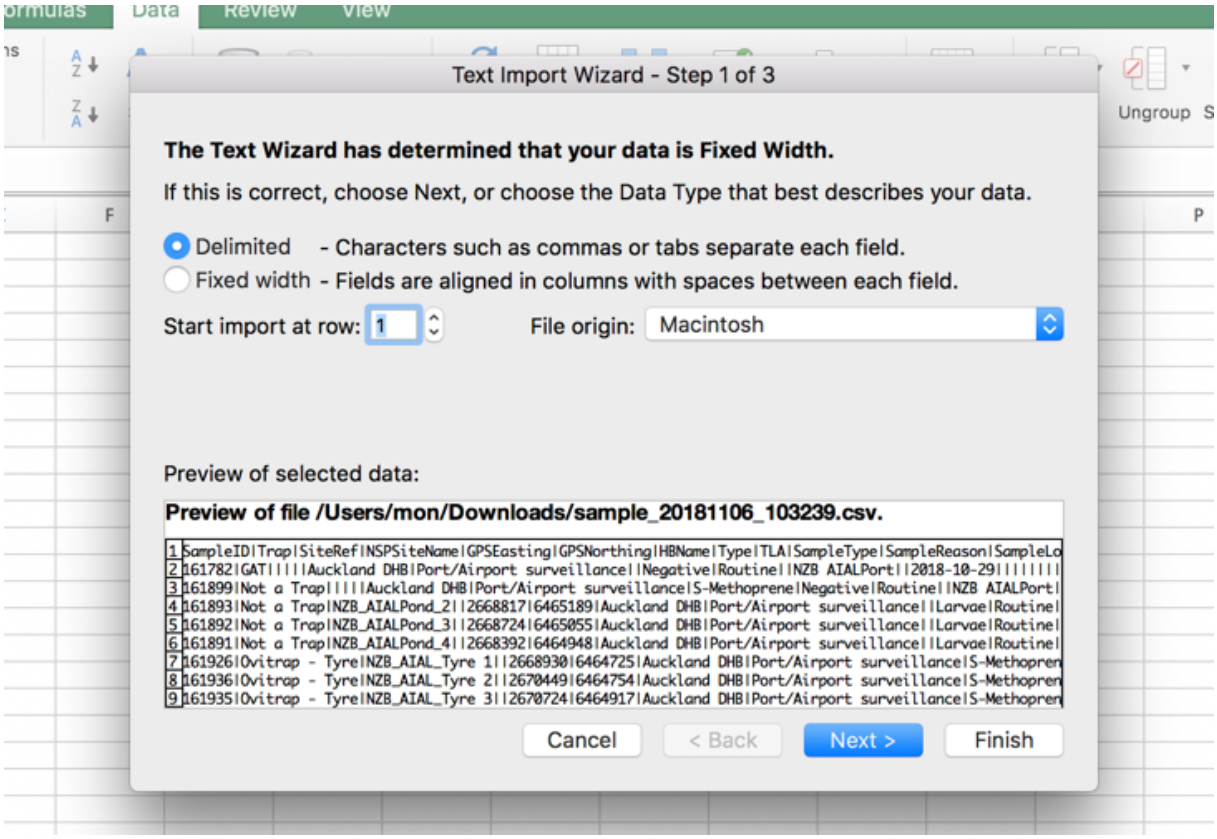


Figure 4.6.22. Select "Delimited" and then click "Next.

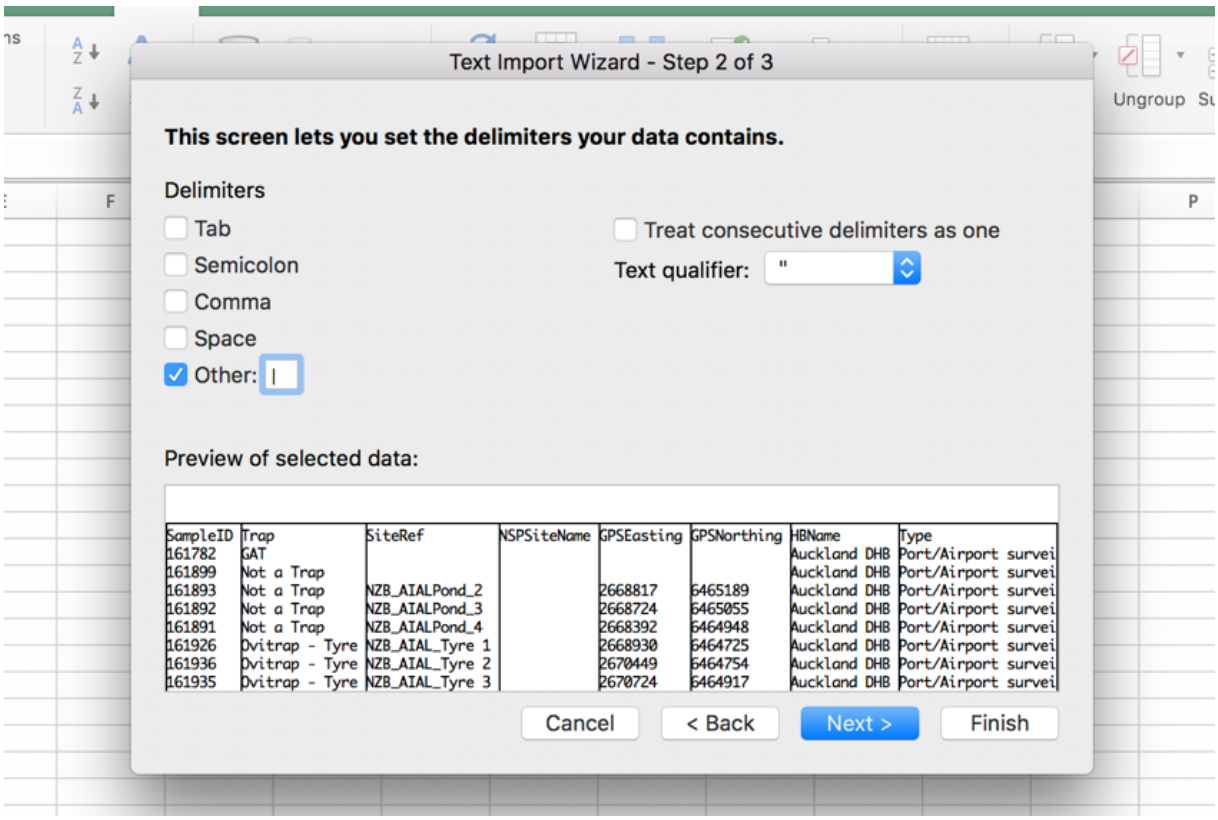


Figure 4.6.23. Uncheck the “Tab” box, check the “Other” box and add type in “|” (this is usually the shift \ button, above 'enter' on the keyboard). Click “Next”..

Click “Finish” and add the data to your worksheet where you can review and analyse (Figure 4.6.23). Save it as a Microsoft Excel spreadsheet file.

Exporting to KLM

This tool was designed for PHOs, Managers and the MOH to assist in the spatial visualization of the information collected in the field and the results provided by the NZB Entomology Laboratory.

KLM export tool allows the user to display the location and status for a sample or groups of samples on a map through Google Earth Pro.

NB In order to display accurately GPS data entered must be accurate and the coordinate information needs to follow the format described.

Select the export option from the left hand menu (Figure 4.6.24) and complete the search parameters, as minimum collection dates must be selected. Then select the “Export KML” tab at the bottom of the page, a file will automatically be download.

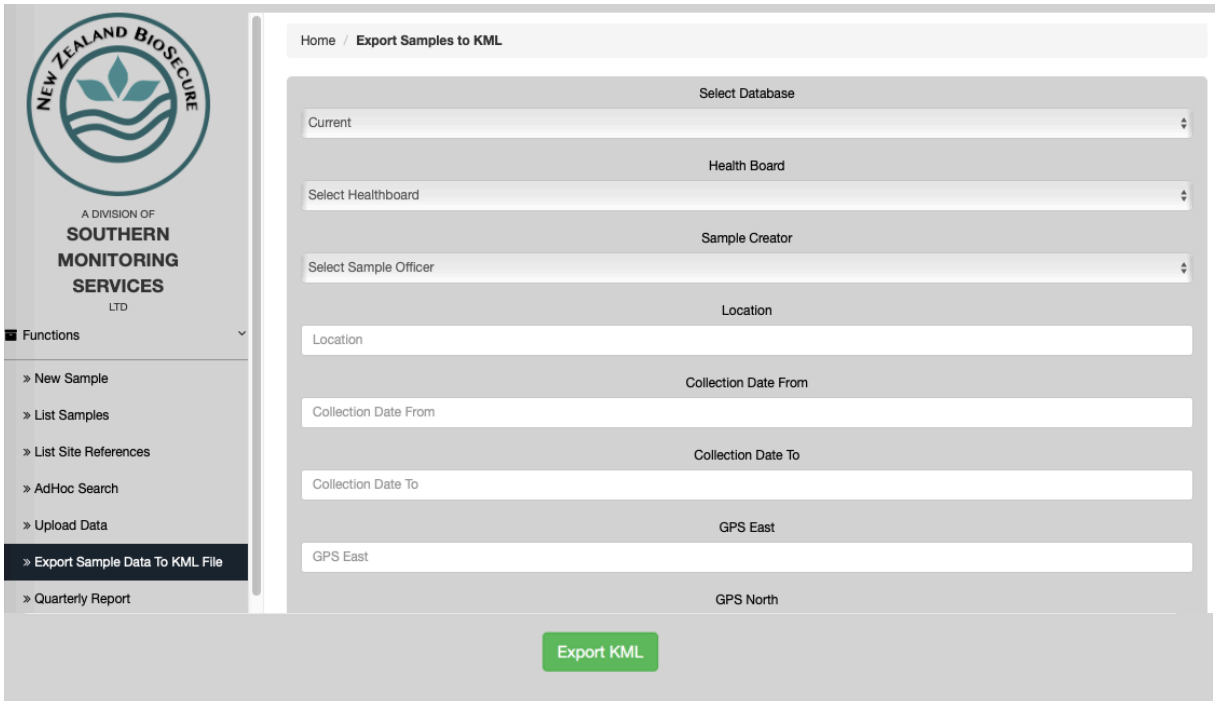


Figure 4.6.24. Export Sample Data to KLM.

Open the downloaded file with google earth and the samples will automatically load. Red pins – Exotic Species, Yellow Pins – non-exotic positive results and Green Pins – Negative. (Figure 4.6.25).

Figure 4.6.25. Display of positive samples with local species (yellow), positive sample with exotic species (red) and negative samples (green) at Ports of Auckland.

Each marker can be accessed to display the results (Figure 4.6.26).

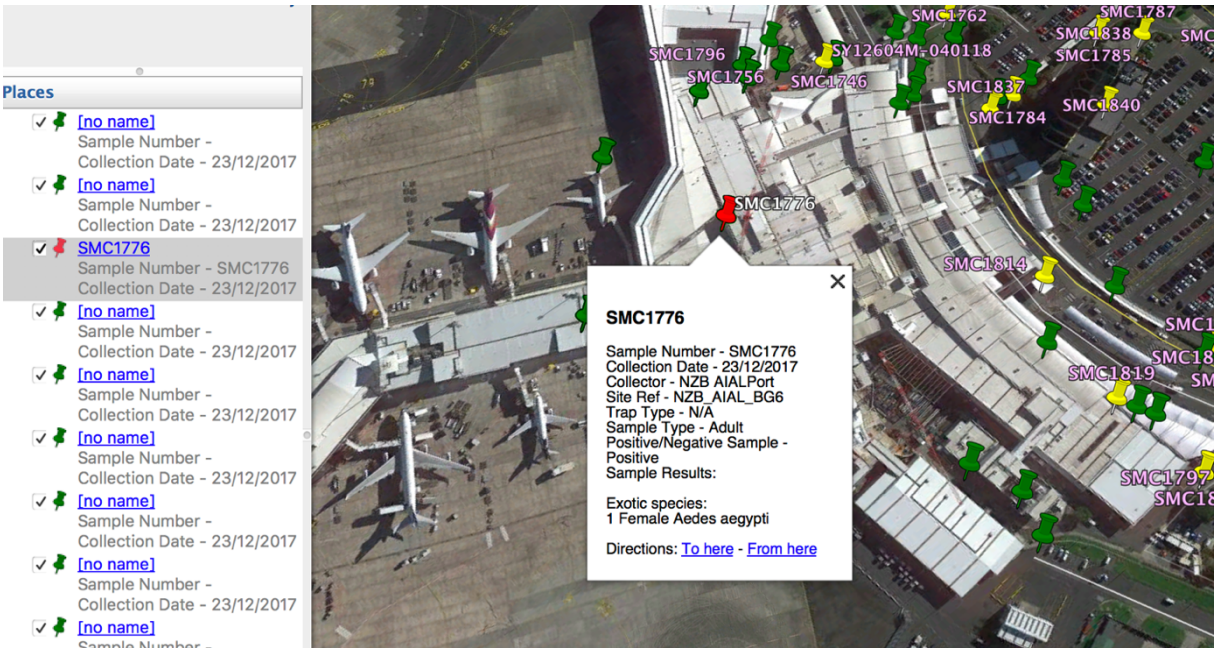


Figure 4.6.26. Results displayed in each sample, showing the sample number, positive or negative status, sample type (adults or larvae), and the sample results (number of mosquitos in each species).

4.7. Mosquito Control

4.7.1 Chemical control

The most widely used approach for mosquito abatement and prevention of mosquito-borne diseases worldwide continues to be the application of chemical pesticides to aquatic larval sources (larvicides) and into the air for adult mosquito control (adulticides). Such applications are done with aircraft-mounted, truck-mounted or manual equipment. Two popular methods for pesticide application are spreading of granular formulations and fogging with small amounts of concentrated insecticides broken into very small particles, Thermal Fogging and Ultra-Low Volume, or ULV.

In the years following World War II highly effective and persistent insecticides, such as DDT (an organochlorine), were used to control both mosquito larvae and adults. The worldwide malaria eradication effort of the 1950s and 1960s was based on treatment of interior walls of houses to kill indoor resting female Anopheline mosquitoes. This program was responsible for the complete disappearance of malaria in some countries and the reduction of human cases in others. A combination of factors, including resistance to DDT, increasing costs, political instability, and environmental safety eventually doomed the program, and malaria has returned to nearly all the formerly malaria-free areas at incidences as high as, or higher than, before.

Organochlorines were phased out in most areas of the world and replaced by newer classes of conventional insecticides, such as organophosphates (e.g. malathion), carbamates (e.g. carbaryl) and synthetic pyrethroids (e.g. resmethrin). Some of the same problems that arose with DDT (resistance, environmental safety) have occurred with nearly all the classes of synthetic organic insecticides and few chemical companies are developing new products for mosquito control. This has led to the use of pesticides known collectively as third-generation insecticides. These include synthetic materials that affect mosquito development (insect growth regulators), microbial insecticides such as *Bacillus thuringiensis israelensis* (Bti) and chitin inhibitors such as diflubenzuron. The use of oils to kill mosquito larvae predates the use of synthetic organic chemicals, and such use continues. Third-generation pesticides are more expensive than conventional pesticides but generally are less toxic to humans and other vertebrates. Because many are highly specific for mosquitoes, they are less disruptive to the environment.

4.7.2 Biological control

Biological control is the depression of population levels of mosquitoes using biological organisms or their products. In practice this involves the introduction of biological agents into mosquito habitats or the management of habitats to optimize the effect of such agents that occur there naturally. Most biological control agents are parasites or predators of mosquitoes; many different organisms have been tested for their effectiveness. However, only a few organisms can be considered to be effective as biological control agents. Two successful examples are mosquitofish *(Gambusia affinis)* and microbial toxins (Bti and Bs). Some investigators do not consider toxins to be biological control agents, but rather a type of insecticide.

S-methoprene

Methoprene is a larvicide - an insect growth regulator. By acting like an insect hormone, it interferes with insect growth and development. It can prevent normal moulting, egg-laying, egg-hatching, and development from the immature (larval) phase to the adult phase. This prevents the insects from reproducing.

It kills by disrupting metamorphosis and most mortality occurs during the larval and pupal moults. As well as indirect lethal effects, methoprene can cause a number of effects in insects at sub lethal doses, such as reduced fecundity, abnormal morphologies and altered pheromone production.

The lethal dose required to kill common mosquitoes is generally around 1 part per billion. Field application rates used against mosquitoes would be unlikely to be lethal to many other insects. When adult emergence is measured, methoprene generally performs as well or better than organophosphates and Bti.

Bti
Bacillus thuringiensis israelensis is a bacteria used for the biological control of mosquito larvae (1st instar - to early 4th instar). Bti produces a toxin in the gut of the mosquito larvae, which destroys the larvae gut lining causing death, usually within 12 hours. 4th Instar larvae and pupae are non-feeding and thus won't be affected by Bti. Bti is non-toxic to humans and non-target organisms.

4.7.3 GMO

Considerable research emphasis has been placed recently on the use of molecular approaches. Oxitec (orig. Oxford Insect Technologies), a British biotechnology company has developed methods for control of insect populations, in which genetically modified insects are used as a "living insecticide".

Oxitec has produced a genetically engineered line of the mosquito *Aedes aegypti* (OX513A) with the intent of suppressing the population of that mosquito at the release site(s). Open field trials of the OX513A genetically engineered mosquito have been conducted in Brazil, the Cayman Islands, Panama, and Malaysia and is now planned in Florida.

OX513A males, which do not bite or transmit disease, are released to mate with wild females. The offspring of such matings die before becoming adults. With repeated releases of sufficient numbers of these self-limiting males, there is a reduction in the wild population to below the level needed to transmit disease according to models of disease transmission.

Before release, male and female pupae are separated mechanically, exploiting the fact that naturally they are significantly different in size. The strain also contains a fluorescent marker, termed DsRed, which is a useful tool for quality control in production and effective monitoring in the field.

For the near future the most promising approach for area-wide control seems to be the selective use of combinations of modern, environmentally safe pesticides, source reduction, and biological control. For personal protection locally implemented programs for repellent-treated bed nets and window curtains will continue to be important.

4.7.4 Resistance

The future of insecticides for mosquito abatement is uncertain. Physiological resistance to pesticides in general has been a problem since the introduction of DDT, and resistance is now beginning to show up even among third-generation products including *Bacillus sphaericus* (Bs) - a microbial insecticide, and Altosid® - an insect growth regulator. The greatest threat to the continued use of pesticides for mosquito control is economics. The costs involved in conducting vertebrate and environmental safety tests on new pesticides are rarely justified on the basis of a relatively small market for public health pesticides. Consequently, few products based on new active ingredients have become available over the past 10-15 years.

4.7.5 Removal or treatment of larval habitats

Source reduction, e.g. the management of standing water to avoid mosquito development, is an important tool in mosquito control. In the early days of mosquito control source reduction usually

meant draining of swamps and marshes, and vast areas of wetlands were permanently lost. As appreciation of the value of wetlands increased in the latter part of the 20th century a more balanced approach to source reduction was adopted by mosquito abatement agencies.

Research has shown that mosquito breeding can be minimized by the timing of flooding in artificial freshwater wetlands and by restructuring of water channels in salt marshes in a way that restores natural tidal action. Such approaches are desirable because they actually improve aquatic habitats while minimizing mosquito problems.

Within the 400 m zone of international ports/airports, larval habitats must be removed or treated. Habitats that could be removed from ports include discarded receptacles such as tins and bottles, tyres and drums, and plastic sheeting that can hold water. The mitigation of mosquito habitat within the “international gateways” should be an ongoing campaign that needs to be pursued vigorously. All agencies and stakeholders need to be involved in ensuring this action.

Habitats that might be otherwise eliminated include blocked sumps, drains and roof gutters, waterlogged and poorly draining land, and surface depressions holding rainwater. Habitats that might need to be treated with anti-mosquito agents include marshes, ponds, channels, drains and sumps - treatment options (depending on circumstances) include

• Bti products and insect growth regulators such as S-methoprene products (as approved by regional councils)
• synthetic pyrethroids including Etofenprox, Bifenthrin, Lambda-cyhalothrin, Delta-methrin, Tetra-methrin, permethrin. Check the product is labelled for use against mosquitoes. Do not use in areas where the product may enter natural waterways.
• kerosene with castor oil to assist spreading (needs to be approved by regional councils)
• chlorine solution (5-10% A.I chlorine). Do not use in areas where the product may enter natural waterways.

Essential containers that cannot be removed, filled with sand, or otherwise precluded from offering a larval habitat for exotic mosquitoes at ports can be periodically treated with s-methoprene or residual pyrethroids.

At some airports and at most seaports, urban development and/or naturally vegetated areas exist within the 400m zone prescribed in the International Health Regulations. The cooperation of local authorities should be sought with respect to an extension of surveillance and intervention activity if so required. However, the realities of this situation should serve to further emphasise the importance of the border control procedures, particularly vessel inspection and port sanitation with respect to receptacles.

4.8. Sand Fly Surveillance

4.8.1. Sampling Sandflies

Sand flies are easiest to collect as adults in traps. Recent studies conducted by the United States Military in Tallil Air Base in Iraq have assessed the effectiveness of several traps in collecting sand flies (Appendix 19.2). Two of the more effective traps for collecting sand flies they examined, are discussed below.

CDC light traps
The CDC light traps used are very similar to the adult traps discussed in 8.1 for collecting mosquitoes. Though they have been run without CO_2 when collecting, CO_2 use may increase the effectiveness of the traps. These traps have been effective but the study showed that their effectiveness was improved if the bulb was replaced with a UV bulb. This type of trap collects the largest number of sand flies but also a large amount of other by-catch. Less by-catch will be collected using a standard bulb but also less sand flies - the reasoning for trapping should be considered when choosing trap types.
Traps should be placed about 0.5 metres above the ground and 30 metres or more apart.

Sticky traps
The other trap used was a 20 centimetre by 7 centimetre sticky cockroach trap and was found to be more effective when used in conjunction with a chemical light stick. All colours resulted in an increased catch, but red and blue sticks seemed to be the most effective.

The sticky trap / light sticks should be placed on the ground with the sticky surface and light stick facing up.

4.8.2. Processing sandflies

Sand fly adults should be processed as per mosquito adults (see section 4.3.8 and appendix 2).

4.8.3. Sand Fly Personal Protection

• If possible stay in well-screened or air-conditioned areas as much as possible. Avoid outdoor activities, especially from dusk to dawn, when sand flies are the most active.

• When outside, wear long-sleeved shirts, long pants, and socks. Tuck your shirt into your pants.
• Apply insect repellent (DEET based) on uncovered skin and under the ends of sleeves and pant legs. Follow the instructions on the label of the repellent to ensure effective application. Spray clothing with permethrin-containing insecticides. The insecticide should be reapplied after every 5 washes.
• Spray living and sleeping areas with an insecticide to kill insects.
• If you are not sleeping in an area that is well screened or air-conditioned, use a bed net and tuck it under your mattress. If possible, use a bed net that has been residually treated. Fine-mesh netting (<1.5mm) is needed for an effective barrier against sand flies. This is particularly important if the bed net has not been treated with permethrin.

4.8.4. Sand Fly Control

Unfortunately, due to the lack of understanding of the habitat utilised by immature stages of the sand fly and difficulties associated with surveillance of these stages, chemical control of sand flies before their adult stage is impractical. For controlling the adult stages barrier treatment of vegetation in areas frequented using a residual insecticide such as bifenthrin or permethrin may provide the most effective control. Fogging/ULV type treatments will also provide some control but, as with mosquito control, timing of the treatment is vital to its effectiveness.

4.9. Flea Surveillance

4.9.1. Sampling fleas

The direct collection of fleas generally uses the mimicking of a blood source's movements to attract the flea to jump to the source. Inserting a roll of flannel into a rodent's burrow and moving it around for several seconds before retrieving will cause any fleas to become excited and jump onto the material. The flannel may then be removed and sealed into a bag or container and frozen. When the fleas are incapacitated they may be removed from the material into appropriate containers

Alternatively, a yellow-coloured plate coated with a tacky substance is mounted above and below the teeth of a rake or between rollers and passed over an area infested by fleas. The vibrations and motion caused by the movement of the teeth or the rollers against the supporting surface excite and attract the insects which jump toward the brightly coloured plate where they become entrapped in the tacky substance. The apparatus may be used to either determine the nature and extent of the infestation prior to treatment.

Collection of fleas through rodent hosts is another method. Unfortunately, flea behaviour is such that they will quickly abandon their host if it dies, so the use of sticky rat boards or some form of humane trap is required for this method.

The sticky trap placement along known rodent corridors is the most ideal option. They need to be well anchored and regularly collected/inspected. One benefit of the sticky board is that if fleas leave the animal they may still become trapped in the glue. Other humane traps tend to be baited and should be placed appropriately. In both cases the host will be alive when collected and will need to be dispatched appropriately, bearing in mind that the fleas will abandon the body shortly afterwards.

4.9.2. Processing fleas

Fleas should be stored in 70% ethanol for their preservation and in preparation for identification.

4.9.3. Flea Personal Protection

Repellents such as DEET or permethrin-impregnated clothing may afford some personal protection against fleas.

4.9.4. Flea Control

Flea control is typically undertaken for one of two reasons: (1) to reduce the risks of disease transmission, or (2) to address a pest problem or economic losses associated with parasitisation of domestic animals by fleas. The strategies used for each situation are often different, and the best results are achieved when the biology and behaviour of the host are taken into account.

Flea control is an effective means of reducing the risks of flea-borne plague and murine (flea-borne) typhus. Because both diseases can be transmitted by rat fleas, the same control techniques can be used to control both plague and murine typhus. Typically, this involves using insecticidal dusts to treat rat runways and burrows. In emergencies, liquid spray formulations of insecticides can be applied to runways and burrow entrances, but these should be used only when dust formulations are unavailable. In some situations, insecticide can be applied to hosts through the use of bait stations that contain food or some other attractant, along with an appropriate insecticide that is placed on the floor of the station or applied to the host's body by forcing it to brush against an applicator as it enters the station. Limited attempts have been made to use

insecticide-treated cotton or other material that rodents take back to their nests. The advantage of this last method is that fleas are controlled not only on the animal but also in the nest.

Controlling fleas on pets and domestic animals is of great concern, as suggested by the fact that more than $1 billion US is spent annually in attempts to control cat flea infestations and the problems they cause. Flea control on pets and other domestic animals can take many forms, including the use of insecticidal dust formulations, granules, sprays, flea collars, topically applied liquids, shampoos, and oral systemics. Insect growth regulators (IGR5) that act as chitin synthesis inhibitors or mimic insect juvenile hormones are also popular, especially for controlling fleas on pets. Control measures do not always have to involve the use of insecticides. Vacuuming, when properly done with a suitable vacuum, has been shown to remove about 90% of cat flea eggs and 50% of larvae from carpets. Cleaning or removal of bedding or nests and other environmental modifications also can have favourable effects.

4.10. Louse Surveillance

4.10.1. Sampling lice

Because most lice exist predominantly on the host, the best way to sample them is to target any hosts in the area. Samples should be plucked or scraped from the skin or brushed from the hair of any symptomatic hosts, using a fine comb, and analysed under a microscope. Lice can occur all over a host's body, but some species prefer particular areas, e.g. hair, eyebrows, beard, armpits, groin etc.

4.10.2. Processing Lice

Lice should be stored in 70% ethanol for their preservation and in preparation for identification.

4.10.3. Personal Louse Protection

The following precautions can also be taken to reduce the likelihood of a head louse infestation:

- Cut your hair short or bind it into a pony tail.
- Do not use hats that are used by many people (e.g. selling items, costumes hire)
- Do not share hats with anyone that may have a louse infestation.

In the case of a body louse infestation:

- Destroy any clothing or bedding that may be infested by lice.

4.10.4. Louse Control

The reduced incidence of people infested with body lice has been achieved mainly through insecticidal intervention and increased hygiene standards, predictably accompanied by a global reduction in the prevalence of louse-borne diseases.

Louse control will not work if a cleaned person puts on infested clothing, both the infested person and their clothing needs to be treated.

Head lice are generally treated using pediculicidal shampoos. These are not completely affective against the nits, so the treatment should be repeated 1 week later to get any newly hatched nymphs. The use of louse combs is strongly recommended also.

Chemical pesticides are commonly used to kill lice on poultry and livestock; however, there are concerns over the safety of using these chemicals on large numbers of animals on a regular basis. There is also evidence that some lice are becoming resistant to pesticides. Louse resistance to pesticides was noted by the fact that fewer and fewer lice are killed with each application of the same amount of chemical.

4.11. Bed Bug Surveillance

Bed bugs are very elusive so a thorough search must be undertaken to determine the extent of the infestation and target control measures.

4.11.1. Advice to be given before inspection

Bed bug inspections need to include thorough searches behind several objects in a property (behind pictures, headboards, under floorboards, paint chips etc.). It is important, therefore, that the inspection processes is explained to the residents and provide:

- Instructions that it will be necessary to inspect the room, including looking through cupboards and drawers.
- Instructions that it will be necessary to remove bed heads, lift carpets and dismantle other items to access all bed bug harbourages.
- Instructions on any activities the client will be required to undertake prior to the inspection.
- Advice to the client that follow up inspections after the initial inspection and treatment will be necessary.

4.11.2. Surveillance Tools and Equipment

Useful tools for a bed bug inspection include:

- A powerful torch
- A 10x magnifying lens (to inspect for live bed bugs and eggs)
- Collection tubes (for gathering bed bugs for later confirmation of identity, sticky tape can also be used for gathering bugs)
- Fine tipped forceps (for picking up bed bugs)
- Gloves
- Screwdrivers and spanners for dismantling items
- An inspection mirror
- Plastic bags (large and small) to hold bottles, tape, infested items, etc.
- Notepad, for recording details of the infestation
- A digital camera (for recording infested sites, the digital images or printouts can also be given to the client in a report or provided as part of an educational package)

4.11.3. Indications of a Bed Bug Infestation

Indicators of infestation include:

- Live or dead bed bugs, and cast skins. Live bed bugs will confirm that the infestation is currently active (Figures 4.11.2, 4.11.3a).
- Faecal spotting. This is digested blood defecated by the bed bugs. It may be initially observed on the sheets, but will be commonly noticed along the mattress seams and other places where bed bugs hide. On light coloured surfaces individual faecal marks appear as small dark round spots, however the spotting may be in colour from cream, through grey to almost black. Generally, the spotting, will occur in groups and appear as splotches of dark marks. Note that the faeces of nymphal cockroaches appear similar, however bed bug blood spotting tends to occur in groups as the insect by nature aggregates. Red blood coloured spots or smears on the sheets may occur which can be the result of bed bugs passing sera, or engorged bugs being squashed by movements of the sleeping host (Figures 4.11.1 and 4.11.3a).
- Eggs (cream in colour with a slight bend, approx. 1 millimetre, which tend to be laid in crevices in dark areas (Figure 4.11.3b).
- A bed bug smell sometimes described as ‘sickly sweet’ but is akin to that of stink bugs. This is usually only noticed in heavy infestations, if close to the bugs or during the treatment process. There are specially trained dogs for detecting infestations.

Figure 4.11.1. Dark spots on mattress and box spring are a tell-tale sign of bed bugs.

Figure 4.11.2. a) Bed bugs between slat and frame of an infested bed. b) Bed bugs often reside along baseboards.

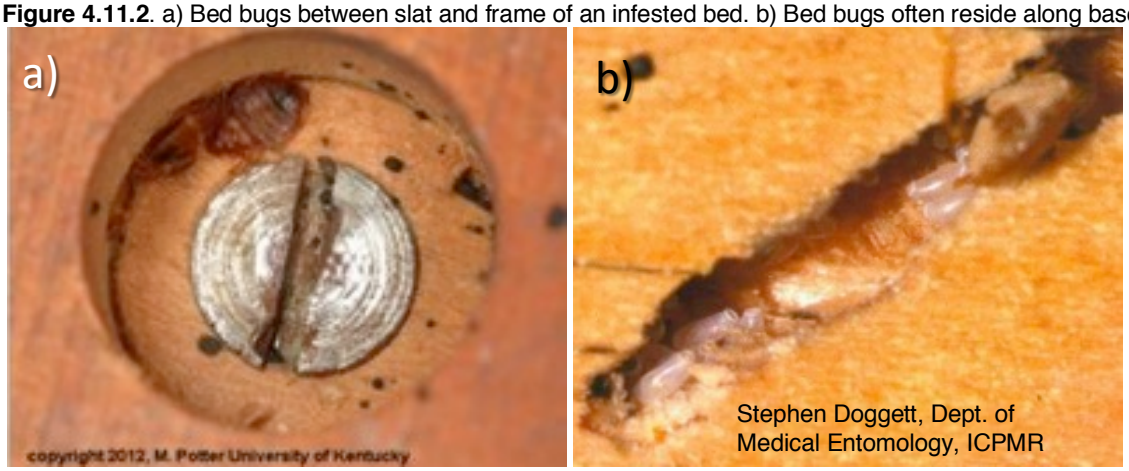


Figure 4.11.3. Bed bugs also congregate along seams of sofas and recliners. a) Bugs hiding near a recessed screw under a night stand (note the presence of faecal spots). b) Bed bug eggs glued in hole made by Furniture staple.

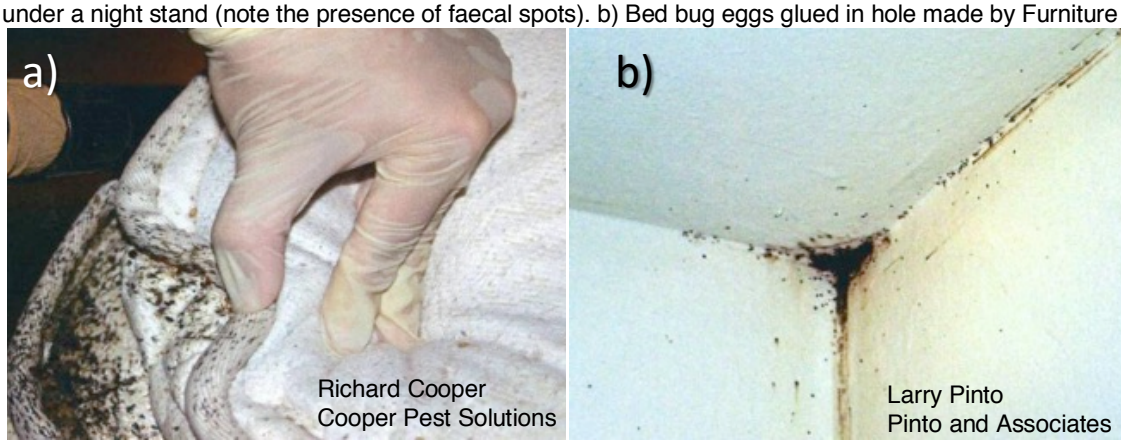


Figure 4.11.4. a) Bed bugs and faeces, shed skins, and eggs in fold of pillow top mattress in heavy infestation. b) Bed bugs clustering on ceiling- wall intersection in a very heavy infestation; note smear marks from residents crushing some with a broom

4.11.4. Other methods used for detection

Specialised sniffer dogs can be used to detect the sweet smell which infestations produce.

Traps are available but many are impractical (large, require electricity or a carbon dioxide bottle, not viable for hotel rooms where guests may be deterred from staying). As yet no traps have been devised which are particularly effective (sticky traps placed in heavily infested areas often catch no bed bugs) so are not advised as a sampling tool at this stage.

4.11.5. Important Information

Bed bugs spread readily and any live stage that is transported has the potential to start a new infestation elsewhere, Therefore:

- No infested items should be removed from the property before they have been thoroughly treated or encased. Otherwise bed bugs can easily be spread throughout the building or to other buildings.
- Any item removed from the property must be properly disposed of or treated to kill all stages which may be present.
- Samples should be properly bagged/placed in secure tubes for identification.

4.11.6. Bed Bug Personal Protection

Conventional insect repellents, like those used to deter ticks and mosquitoes, do not appear to be as effective against bed bugs. Therefore, attempting to avoid being bitten by applying insect repellent at bedtime is not recommended. Sleeping with the lights on is also not likely to deter hungry bed bugs, as they will adjust their feeding cycle to the host's sleeping patterns.

The best option for preventing bites is by reducing the likelihood of exposure to bed bugs. This can only be achieved by reduction of potential habitat, and regular inspections. Decluttering is one of the best ways of reducing potential harbourage sites and also makes detection easier. General maintenance (i.e. keeping paint, walls, flooring etc. in good condition to reduce number of potential harbourage sites, Figure 4.11.5) can also help.

To protect yourself when travelling and to reduce the likelihood of collecting "hitch-hikers" it is wise to put your luggage on a stand (or other hard surface) while you inspect the room for signs of bed bugs.

Figure 4.11.5. One of the biggest obstacles to success in bed bug control is excessive clutter, which provides unlimited areas for bed bugs to hide and to lay eggs (and protect them from pesticides!).

4.11.7. Bed Bug Control

One of the major factors for the degree of the bed bug resurgence has been poor pest control and the failure of industry associations around the world to provide guidance to their members on 'best practice' in the management of modern insecticide resistant strains of bed bugs (Doggett et al. 2011). Incorrect preparation for insecticidal treatment, as well as poor application of insecticide, has led to the development of resistance in some areas. In other cases, poor application has simply lead to dispersal of bugs and compounding of problems.

Because bed bugs can shelter under a number of surfaces from pesticides, it is important a thorough inspection is conducted to determine the spread of the infestation, and to prevent dispersal following pesticide application due to the repellent effect of some pesticides. Control is best undertaken by a pest control firm experienced in bed bug control. Even with experienced operators 100% eradication can never be honestly declared due to the biology and evasive nature of bed bugs.

Usually several visits for inspection and control are needed for effective control. A range of Integrated Pest Management techniques may be used to control the infestation, including:

Reduction of habitat and disposal of items
Heat treatment
Steam
Extreme cold
Encasement
Physical removal
Vacating a room
Traps/Barriers
Insecticides

<u>Reduction of habitat and disposal of items</u>
Reducing the overall biomass of a bed bug infestation can be achieved through discarding infested furnishing, although complete control will not be achieved. This option can be very expensive to the property owner and not always necessary. The exceptions are mattresses that are torn; these are difficult to treat by insecticides and steam, and should be discarded. However, they can be covered with an appropriate mattress encasement, heat treated or fumigated. Any item to be removed must be sealed in plastic before removal, ensuring that all openings are securely taped shut. Such furnishings should be treated before discarding. To avoid others acquiring bed bugs from discarded infested items the furniture should be destroyed or rendered unusable, for example mattresses and bases should be slashed. They should also be clearly labelled with obvious signs indicating that the items are infested with bed bugs and must be destroyed. Disposal of items should be coordinated with waste disposal collection. Around the world heat is being employed to effectively treat infested mattresses and furniture, and such processes are now becoming available in New Zealand.

Decluttering (being careful not to spread the infestation) is a necessary first step to achieving control. Items which can't be encased for treatment off-site should be opened in such a way to allow for surveillance and subsequent penetration of insecticide or heat treatments.

<u>Heat Treatment</u>
Bed bugs are very sensitive to heat and are rapidly killed when exposed to temperatures over 45^{o}C. If heat is used for bed bug control it is important that the high temperatures are applied suddenly; a gradual rise in temperature may cause the bed bugs to disperse, thereby potentially spreading an infestation.

Laundering:

Studies from the United Kingdom (Naylor & Boase, 2010) have shown if the water is at 60°C, then every bed bug stage will be killed in the wash. However, a temperature of 40°C will not be lethal to all the eggs. Many tap outlets will be below 60°C for safety reasons, so clearly if hot water is to be relied on for bed bug disinsection the temperature must be confirmed at or above 60°C. Bed linen, curtains and clothing can be bagged (and sealed) before removal from the room and washed in the hottest water possible (>55°C) and/or dried in a hot air clothes drier for at least 30 minutes. Alginate bags are preferable for infested linen, as the bags with the linen enclosed can be placed directly into the washing machine and the bags will dissolve. If alginate bags are not available, then plastic bags should be used.

For tumble drying, the Naylor and Boase investigations found that the dryer had to be operated on the 'hot' setting for 30 minutes for dry clothes to achieve a complete kill of all stages. If clothes are wet, then they should be left in the machine until completely dry.
All wardrobes, drawers and cupboards should be emptied and the contents treated as above. After clothing and materials have received the heat treatment, these should not be returned to wardrobes but kept sealed in plastic bags away from the infestation until eliminated.

Heating:
Large electric or gas driven heating units are increasingly being employed for bed bug control around the world. The most efficient are 'bubble treatments', where infested items are treated in a small contained area. Heat treating whole rooms is rarely successful without the use of insecticides as there are many harbourages that can protect the bed bugs, and control is especially difficult in heavily cluttered rooms. Thermal control for bed bugs in large spaces requires a high level of skill; there have been a series of fires resulting in the complete destruction of dwellings caused by the inappropriate use of heating units. This method should only be undertaken by trained individuals.

Solar Heating:
It is often claimed that bed bugs can be killed via heat by placing infested materials into black plastic bags and then into the sun. However, a scientific investigation has shown that this can be ineffective with large items such as mattresses, which have a high thermal inertia (Doggett et al., 2006). Since this method cannot be relied upon to disinfest items it is not recommended.

Steam
One practical method of exploiting heat is through the use of steam. The great advantage is that it will kill all bed bug stages including the eggs (most insecticides are non-ovicidal). However, control cannot reliably be achieved with steam alone.

It is important to note that there are many different brands and types of steam machines on the market, and not all are appropriate; the unit must be able to produce steam of a low vapour flow and high temperature.

As with all equipment, the steam machine must be properly maintained and the operating temperatures should be regularly checked with the aid of an infrared thermometer. Immediately after steam treatment the surface should be recording at least a temperature of 70-80°C.

An experienced operator should be used to ensure bed bugs are not blown about by the steam and that all areas are treated thoroughly with the correct flow of steam - inspections must be diligent and the treatment process must be meticulous.

As with any technology, steam has its limitations. Being water based, electrocution is a potential issue; thus power points and other electrical fittings should not be steam treated. Steam may damage heat and water sensitive materials, thus the Pest Manager should always test the item to be treated in a non-conspicuous area. Steam will raise the humidity in a room, which can lead to mould growth leading to other potential health issues. Steam treatments are very time consuming.

The greatest disadvantage is that steam is non-residual. Bugs that are not directly killed (and it is prudent to assume that a certain percentage will not be contacted) will not be exposed to any further control influence unless an insecticide is present. It is always necessary to complete the control process by following up any steam treatment with a residual insecticide.

Extreme Cold

The alternative to extreme heat is extreme cold, i.e. freezing the bugs. This has the advantage that heat sensitive materials will not be damaged. While this method can often not be directly used by the Pest Manager, it can be recommended to the home owner and hotelier for small items. Any item for freezing should be placed loosely into a bag, and as always, this must be done in the infested room prior to removal. The amount of time in the freezer would be dependent on the size of the item; the larger the item the longer in the freezer. If the freezer is operating at or around -20^{o}C, then 2 hours at this temperature will kill all stages. However, for the average household freezer, studies have indicated that 10 hours will be required (Naylor & Boase, 2010).

Dense items may take several days for the centre to cool sufficiently to kill the bugs and the longer an item is kept frozen the more likely the bugs will be destroyed. Naylor and Boase suggest around 8 hours of freezing is required per 2.5kg of dry weight of laundry. Many modern freezers are of the 'frost-free' type and go through cycles of varying temperatures. As a result, bed bugs will require a much longer time in the freezer to be killed, even up to several days.

Encasement

Seamless mattress covers provide fewer potential harbourage areas than mattresses, thus making them less susceptible to an infestation. The covers can also be readily removed for laundering thereby making control easier, and being white makes bed bugs and their spotting easier to notice. The benefits provided by mattress covers have been further extended with the recent development of specialised anti bed bug mattress encasements, which are now available in New Zealand. These encasements have incorporated an in-built membrane that is impervious to bed bugs; not only can bed bugs not penetrate these encasements, they are even unable to bite through the material.

Encasements may be used in two modes: to completely contain and hence inactivate an existing infestation in a mattress and ensemble base, or to prevent the mattress and base becoming infested in the first place.

As bed bugs can live for up to 6 months without feeding at 22^{o}C, or even longer in cooler climates, if used in containment mode the encasements must be left in place for much longer than this as removal represents a re-infestation risk. Thus users need to be made aware that encasements should not be removed if being used for bed bug containment. In these circumstances, bed sanitation can be improved by covering the mattress encasement with a seamless mattress cover which can be regularly removed and hot washed and hot dried.

It is important to note that mattress encasements cannot by themselves stop bed bugs and should be used as part of an overall bed bug management program. They will not stop a bed bug from climbing up onto a mattress but will make them easier to spot and treat. The desirable features of mattress encasements include: small zipper teeth that stop juvenile bed bugs passing through, few seams and tightly stitched joins, an in-built bite-proof membrane, end zipper stops that prevent bed bug escape or entry, and anti-removal devices.

Physical Removal

Bed bugs can be physically removed using vacuuming (or by sticky tape if numbers are small on a mattress). However, it is important that the vacuum cleaner does not become the source for further infestations so it must be properly 'disinsected' following use and only be used for pest control. Vacuum units that have the base and all hoses composed of solid plastic can be readily sterilised in hot water. This should be done as soon as possible after use.

A vacuum machine that has a disposable dust bag should be used. A crevice nozzle can be used along carpet edges, bed frames, mattress seams and in ensemble bases, furniture, and other potential harbourages. Vacuuming cracks and crevices prior to insecticide treatment will not only remove the bugs but dirt as well, which will allow the chemicals to penetrate better and improve their residual effect. After vacuuming is complete, the contents must be sealed within a plastic bag. This should then be destroyed by incineration if possible, rather than just being placed into the general rubbish. If incineration is not possible, then apply insecticide dust to the contents and seal in a plastic bag prior to disposal. Under no circumstances should an insecticide aerosol or spray be applied to an operating vacuum machine as this may cause an explosion and/or fire. The allergens from bed bugs are known to trigger asthmatic reactions and dispersal of the allergens can occur through vacuuming. Repeated exposure to the allergens can lead to a sensitisation thereby increase the risk of adverse respiratory effects, thus it is important that a vacuum machine fitted with a HEPA filter is used to protect the health of the client and the Pest Manager. When not in use the vacuum unit itself should be stored in a sealed bag.

Vacuuming is useful but has limitations so must be combined with a number of other measures to achieve control.

Stiff brushes are sometimes suggested for removing bed bug eggs; however, they are not recommended as they can disperse the eggs and make control more difficult.

Vacating a Room

Leaving an infested room vacant for extended periods is not an option to control the bugs as they can live for many months without a blood meal. Infested rooms must be inspected and treated.

Traps/Barriers

There have been a number of devices coming onto the marketplace claiming to capture or detect bed bugs (traps and monitors) or that aim to prevent the insects crawling onto beds (barriers). Neither will eliminate an infestation by themselves and must be used as part of an IPM program.

Most traps are active devices that attempt to catch host seeking bed bugs via the use of various attractants such as heat, humidity, carbon dioxide, and/or various other attractants.

There are many practicality and health and safety issues with traps currently available and as of June 2014, none of the traps have been tested and found efficacious in an independent scientific study. Thus presently, they are not recommended. The use of sticky tapes for the monitoring of bed bugs has been found ineffective (Doggett et al. 2011). Bed bugs tend to react negatively to gels and other sticky surfaces, and avoid capture.

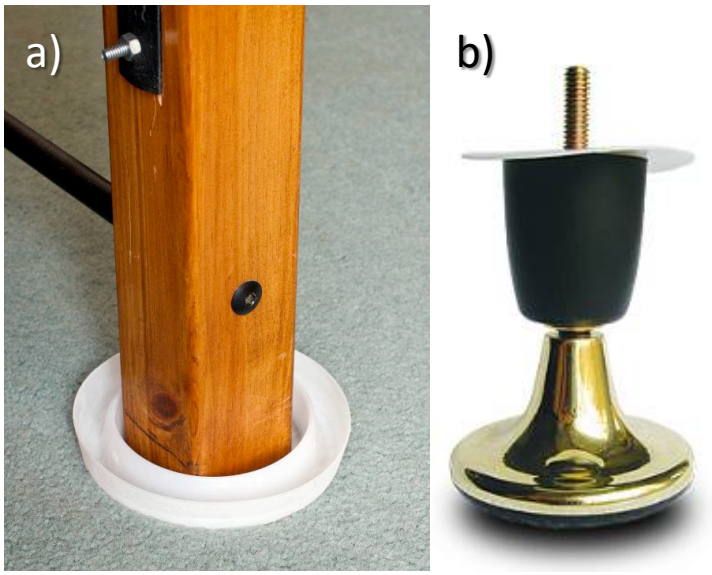


Figure 4.11.7. a) The 'Climbup Interceptor' underneath a bed leg, b) the BB Secure Ring on a bed leg.

There are simple passive units that aim to protect the sleeper by preventing bed bugs climbing beyond the bed legs, as the Climbup interceptor. Bedbugs, trying to climb up the leg are trapped between the inner and the outer ring (Figure 4.11.7a). While this approach is not a complete bed bug treatment solution, they can at least give you an indication of 1) is there an infestation and 2) how large is the infestation. After treating a room, an empty bowl means no more bed bugs.

Leave the Climbup interceptors in place for 2 weeks, to catch any late hatching bed bugs.

Barriers are placed either underneath the bed legs/casters or on top of the casters of ensemble bases (Figure 4.11.7b). Added to the barrier may be additional security devices to reduce the risk of the bed bugs gaining access to the sleeper including various dusts (insecticidal or talc) and/or sticky substances that entrap the insects. For barriers to be effective the bed must be kept away from the wall and valances and sheets must not touch the ground, otherwise bed bugs can then access the bed.

The 'BB Secure Ring' is a very simple barrier that fits between the bed leg and the bed, and is constructed from an ultra-smooth plastic which bed bugs cannot climb over. In laboratory trials, the device was able to prevent access by bed bugs of all strains and stages (Figure 4.11.7b, Doggett et al. 2011).

Insecticides

Chemical control is necessary as part of an IPM strategy, and effective control is unlikely to be achieved without the use of pesticides. However, these should only be undertaken by operators experienced in bed bug control as there are various repellency effects and resistance to certain pesticides. Various surfaces need to be treated with different products and application rates and it is important all harbourage sites are reached by the chemical. Thus the experienced operator needs to survey and prepare the room correctly before any chemicals are applied.

Bed bug control can only be maintained through a treatment strategy that includes a variety of techniques plus careful attention to monitoring. Proper use of pesticides may be part of the strategy, but will not by itself eliminate bed bugs. In addition, bed bug populations in different areas (both between and within countries) have developed resistance to the ways many pesticides work to kill pests. When dealing with a resistant population some products and application methods may only make the problem worse. It is necessary to consult a qualified pest management professional experienced in bed bugs to achieve control.

4.11.8. Processing and Sending

Bed bugs should be stored in 70% ethanol for their preservation and in preparation for identification.

4.12. Cockroach Surveillance

4.12.1. Sampling Cockroaches

Trapping can determine resting areas and infestation severity, monitor effectiveness of chemical controls, and detect population increases which may then require insecticide treatment. Several types of traps can be purchased. Most are about the size of a large matchbox, have openings at both ends, and have the inside surface covered with a very sticky adhesive and slow-release food attractant (Figure 4.12.1). Cockroaches detect the food odour, enter the trap, and become immobilised by the adhesive. Traps can also be made from deep glass jars with a layer of petroleum jelly on the inside to prevent escape, and either commercially available bait or a piece of fruit as an attractant.

Figure 4.12.1. Sticky trap for cockroaches, with nymphs and adults attached.

Traps should be positioned with (both) ends open and accessible to intercept cockroaches as they travel to and from harbourage and feeding areas. For maximum efficiency they should be placed in dark areas such as along bulkheads and in tight spaces. Traps should be left out for a minimum of 24 hours including an overnight period. A suitably placed trap can catch numerous cockroach adults and/or nymphs daily (Figure 4.12.1). Traps are relatively inexpensive, convenient to use, disposable, and do not need to contain toxic chemicals. If 2 or more cockroaches are caught within a 24 hour period, this may signal that a pesticide control operation is required.

Any live cockroaches still in traps can be killed with a 3% solution of dishwashing liquid in water.

Cockroaches may also be detected by physically searching resting sites. Commonly inhabited areas on ships include around false bulkheads, holes for electrical wiring and plumbing, in lagging and torn insulation, behind bulletin boards, around supports in serving lines and around other kitchen equipment and fittings, behind fridges, in deck drains, ventilation grating, fuse boxes, food stores and oncoming food supplies or other products that may be brought on to the ship and already contain infested materials such as wood and paper products.

Looking for signs of cockroach faeces is also a good way to spot past or present cockroach activity. A screwdriver for opening grates and hatchings will be required, as is a keen eye, and a flashlight for illuminating dim or unlit areas. Cockroaches will oftentimes be disturbed by the light and run away, making them even easier to spot. A flushing agent, usually a pyrethroid may also be used to check for cockroach activity. This is typically sprayed into a harbourage area and monitored for 3-5 minutes for any signs of cockroaches. Because of the repellent properties in these agents they should not be used in areas where traps or insecticide controls are going to be used.

4.12.2. Processing Cockroaches

Cockroaches should be stored in 70% ethanol for their preservation and in preparation for identification.

4.12.3. Cockroach Personal Protection

To protect humans from infective diseases associated with cockroaches, all areas likely to attract or harbour the insects should have some form of cockroach control applied (Figure 4.12.2). This

Figure 4.12.2. Chemical control.

could mean filling cracks in external walls and foundations, fixing or replacing leaking plumbing, keeping areas dry and clean, removing wastes in an efficient manner, ensuring food and human waste is not accessible by insects, setting baited traps or boards to trap or kill any cockroaches that may be present, or applying an insecticide. An effective cockroach control program is essential to prevent infestations.

In areas or situations where dysentery and other gastric diseases are present, good personal hygiene can help mitigate risks of further spread to the individual.

Drinking bottled or thoroughly boiled water, washing all raw fruits and vegetables, and thorough cooking will also help to prevent ingestion of food contaminated with faeces or bacteria tracked around by cockroaches. Where cockroach eradication of a site is not possible, and diseases such as typhoid and polio are endemic, individuals should seek immunisation against these.

4.12.4. Cockroach Control

It helps to know the species of cockroach involved in any infestation as this allows for a targeted approach, exploiting their susceptibilities to more effectively eradicate them from the site. Where multiple species of cockroach are involved, a range of different control options may have to be used to effectively tackle the problem.

German cockroaches, in particular, are a noted pest as they have a high reproductive rate, 3-4 batches of eggs per female per year and are well adapted to a life associated with humans and their structures. They are also very hardy and are known to hitchhike their way on to ships via the new stores coming aboard.

Prevention is the best way to ensure successful cockroach control. Elimination or reduction of food, water and shelter helps prevent breeding and spread of these insects and smaller outbreaks or invasions can be more easily contained. Inspections of oncoming goods should also be undertaken.

The presence of several stages of nymph, eggs and adults in an area suggest the population has become well established. A large infestation will need to be chemically treated before other control methods or environmental management procedures can be used to control the population and eliminate the problem. Smaller infestations can be treated with the use of traps and bait stations. Environmental management can help to eliminate small numbers of cockroaches and prevent new infestations from occurring. All food, including food waste, should be kept in secure containers and bins or disposed of promptly. All dishes and utensils should be cleaned after use and not left out overnight. Areas used in food preparation or consumption should be thoroughly cleaned often, and gaps behind appliances also cleaned. Cupboards, drawers, ovens, sinks and refrigerators should be regularly cleaned. Cockroaches will also eat paper and board if no other food is readily available so accumulations of organic waste, such as waste paper, should also be contained or removed.

Care and attention should be paid to all corners, spaces beneath cupboards and appliances, and small gaps and cracks around flooring and fittings where food waste may accumulate (Figure 4.12.3). Gaps around boards, flooring, piping and other fittings should also be sealed to eliminate harbourage sites. Goods being brought in from elsewhere should be inspected for signs of

cockroaches and be treated or kept separately so as not to introduce insects into areas vulnerable to infestation. Leaking pipes, windows and other sources of water and damp should also be remedied to reduce the amount of damp habitat.

Figure 4.12.3. Large cockroach infestation.

To control a large infestation residual or non-residual insecticides can be used. Non-residual insecticides will need to come into direct contact with the cockroach during application to kill it. No matter what form of insecticide is used multiple applications will be necessary as a single treatment is not likely to destroy all of the insects. How often a treatment is required will depend a little on what other control and sanitisation techniques are being employed, as well as how vulnerable the area or structure is to re-infestation.

Insecticides work best when applied to areas where cockroaches hide during the day or areas they regularly pass through at night to maximise the chance of exposure to the chemical. Cockroaches may become difficult to control as some species may be repelled by certain compounds and so can avoid the insecticides, while others are resistant to the active ingredient. For this reason, a combination of chemicals can be more effective rather than one single insecticide. The German cockroach, in particular, has developed resistance to a range of organophosphates, organochlorides and pyrethroids. Chemical control should always be followed up with environmental management to provide a well-rounded control program against cockroaches.

To lower cockroach numbers quickly a non-residual spray can provide immediate action. However, this in itself will not effectively control the population. Used in conjunction with a residual spray this is a very effective control regime. Insecticidal dusts can also be useful as they can be placed deeper into crevices and voids, and are also safe around electrical outlets where liquid sprays would not be safe to use. Dusts usually provide longer lasting residual control than sprays but are not effective in wet or damp locations. Dusts can be applied in squeeze bottles, or in bulb or bellow type dusters.

Where a liquid spray is to be used, it should be taken into consideration whether or not to use oil versus water based spray. Oil based sprays adhere better to smooth surfaces such glass and metal, but may damage painted surfaces, plaster or lino.

Figure 4.12.4. Cockroach trap located under the sink.

Traps are another option to reduce cockroach populations, especially when used in conjunction with poisonous baits, spray or liquid insecticides and other preventative measures. Traps should not be placed in exposed locations such as the deck of a ship (Figure 4.12.4). Exposure to too much water could destroy the trap and degrade the bait inside. They are a good option where aerosols and sprays are unable to be used such as around electrical equipment.

Care should be taken to dispose of dead cockroaches as egg capsules may be unaffected by the poisons and hatch out at a later date

when any residual treatments have become inactive. All control activities should be undertaken by a certified control person.

4.13. Sampling ticks

Ticks are usually large enough to be seen with the naked eye, but it is advisable to use a hand lens in the field and a dissecting microscope in the laboratory to search for small species and immature stages. Straight-shafted forceps are useful for collecting nymphal and adult ticks, but tick larvae are more easily picked up on the end of a moistened tip of a fine paintbrush. A fine probe or minute spatula or similar is a useful tool for manoeuvring specimens.

4.13.1. Removing ticks from hosts

The safest and most effective way to remove an attached tick is to grasp it behind the mouthparts with fine forceps and pull gently and steadily away from the skin until the tick releases its hold. Do not twist, jerk or crush the tick's body as this may release body fluids harbouring pathogens, directly into the wound.

Barbed mouthparts of the tick help to anchor it in the flesh of its host and ticks secrete compounds in their saliva that help to cement them in the feeding wound. Pulling too strongly or twisting while attempting to remove a tick may result in tearing of the tick, leaving the hypostome embedded in the skin. This can lead to bacterial infection in the feeding wound.

Do not attempt to remove an attached tick with caustic chemicals or by applying heat. This can kill the tick before it disengages its mouthparts. It can also cause the tick to regurgitate into the feeding wound and therefore increase the chance of transmitting a pathogen.

The removed tick should always be saved for identification, place into a sealed container and place in the freezer or add ethanol to the container. The attachment site of the tick should be washed thoroughly with warm soapy water and rubbing alcohol to remove any possible pathogens. Wash your hands as well as the tweezers or any other object the tick (or fluids from the tick) may have contacted. Objects used to remove or dispose of ticks as well as the site of the tick bite should be disinfected.

4.13.2. Free-living ticks

Ticks on vegetation or in a pasture can be collected by “dragging”, i.e. pulling a flannel sheet approximately 1.5 metres x 1 metre slowly across the ground. The tick attaches to the sheet, mistaking it for a passing host. These can then be removed using forceps.

Material can be removed from host environments such as nests and burrows to search for ticks in a white tray. Ticks can be recovered from the loose material by gentling shaking the material in a tray or sieve. This action can also stimulate the ticks to right themselves and move to a more protected location making them more visible amongst the debris.

Rocks adjacent to nests and burrows should also be overturned as some species prefer to live in this type of habitat.

4.13.3. Trapping ticks

Figure 4.9.1. Dry ice trap.

Soft ticks can be readily collected via dry ice traps (Figure 4.13.1). Blocks of dry ice emit large amounts of carbon dioxide, a host seeking stimulant. Traps are set in and around nesting areas of animal hosts. Soft ticks can be observed running along the surface of the ground towards the trap and are collected by hand, or inside a collection chamber in the trap.

Where dry ice is not readily available or there are inappropriate storage facilities it is possible to make dry ice snow using a CO_2 bottle and a Sno Pack. The liquid faction is run out of the bottle through the Sno Pack and a 500g compressed CO_2 block is produced.

4.13.4. Tick Personal Protection

- Wear light-coloured clothing with long pants tucked into socks to make ticks easier to detect and keep them on the outside of the clothes. Unfortunately, surveys show the majority of individuals never tuck their pants into their socks when entering tick-infested areas. It is unclear just how effective this prevention measure is without the addition of a repellent. Larval and nymphal ticks may penetrate a coarse weave sock. Do not wear open-toed shoes or sandals.
- Use of a DEET or permethrin-based mosquito and tick repellent can substantially increase the level of protection. This approach may be particularly useful when working in areas with a high risk of tick exposure.
- When walking keep to the centre of trails to minimise contact with adjacent vegetation.
- Unattached ticks brought in on clothing can potentially result in a later tick bite. Blacklegged ticks can survive for many days in the home depending upon the humidity. In the laboratory, nymphal *Ixodes scapularis* can survive for over 6 months at 93-100% relative humidity (RH) but over half will die in less than 4 days at 65% RH (RH in modern homes is generally <65%). On returning home remove, wash and dry the clothing. Many blacklegged ticks and lone star ticks can survive a warm or hot water wash, but they cannot withstand one hour in a hot dryer.
- Carefully inspect the entire body and remove any attached ticks (see section 4.13.1). Ticks may feed anywhere on the body. Tick bites are usually painless and consequently most people will be unaware that they have an attached tick without a careful check. Also carefully inspect children and pets. A hypersensitivity reaction to a tick bite may aid detection in a few individuals but most people will be unaware a tick is attached and feeding.

4.13.5. Tick Control

The removal or regular maintenance of ground cover that provides shelter for ticks, cutting grass and removing covering shrubbery will assist in the control of tick populations.

Residual treatment of the ground cover in areas walked over regularly should remove some of the risk of individuals picking up ticks. However, ticks are able to detect and avoid pesticides. A barrier treatment of the populated area with a residual insecticide such as bifenthrin or permethrin, from the centre out, would provide for good control.

These organisms should be preserved in 70% ethanol in small containers or vials with leak-proof lids.

Excerpts from Baker, 1999. Mites and ticks of domestic animals: an identification guide and information source. The Stationery Office, London. 240pp.

4.14. Mite Surveillance

4.14.1 Attached mites

Because most parasitic mites exist predominantly on the host, the best way to sample them is to target any hosts in the area. Samples should be scraped from the skin of any symptomatic hosts and analysed under a microscope. Mites can occur all over a host's body, but some species prefer particular areas. For example, on mammals: the ears, muzzle, chin and ventral midline are commonly affected areas; while on birds: the scaly areas of the legs and face, inside of feather quills etc.

Dislodged specimens are easier to see when combing or brushing is carried out over a white sheet or tray. If the host is dead, the results may be improved by first anaesthetising any mites still living by leaving the body in a box with a wad of ether-soaked cotton wool for about 30 minutes.

An alternative method is flotation. Specimens can be separated from dead hosts by shaking the body in a bowl or plastic bag containing a weak detergent solution and then filtering the liquid. Acari have a water repellent cuticle and the detergent helps to free the specimens by reducing the surface tension.

Additional methods may be required to separate specimens that are either too firmly attached or are actually buried in the skin. A simple method which can work for skin scrapings:

- mount tissues in a drop of 10% KOH or NaOH on a flat glass microscope slide;
- add a coverslip and clear by warming for 5-10 minutes;
- apply gentle pressure to the coverslip to separate the specimens from the softened tissues.

4.14.2 Unattached mites

Unattached mites can be sucked up into an aspirator (often called a pooter). This is particularly effective for catching rapidly moving mites without harming them. Specimens can be collected directly into preservative such as 70% ethanol, however this is not recommended for mouth operated aspirators as the user will inhale ethanol fumes. The entire aspirator can be placed in the freezer for a few minutes to slow down the specimens and then transfer them directly to a tube containing preservative or remove the top of the aspirator and fasten a lid to the vial and replace in the freezer.

4.14.3. Mite Personal Protection

- To avoid picking up mites, use insect and mite repellents which contain e.g. DEET on exposed skin.
- Permethrin treatment of clothing provides protection against mites.
- Wear protective boots, long sleeved clothes and trousers.

4.14.4. Mite Control

Indoor treatments with residual sprays or dusts may provide some degree of control against mites.

Proper treatment and control of a scabies problem requires:

- **Positive diagnosis of the problem by a physician**. Scabies mites are extremely small; females measure about 1/60th inch. In the case of both scabies and straw itch mites, the rash or bites associated with these mites is the primary diagnostic characteristic.
- **Application of an insecticide-containing prescription lotion to the body**.
- **Because there is time lag between the initial mite infestation and the appearance of symptoms, family members or people coming in close contact with infested persons may require treatment**.
- **Sanitation is extremely critical to successful control**. An infested person's undergarments and bed linen should be washed regularly in hot, soapy water. NOTE: Human scabies mites cannot survive off a host for more than about 24 hours. Therefore, insecticide foggers ("bug bombs") and sprays do not help eliminate the problem and are unnecessary.

these organisms should be preserved in 70% ethanol in small containers or vials with leak-proof lids.

A white tray is a useful piece of equipment for collecting mites as they show up well on a white background. Also when dropped on the ground, trays can act as a lure to some larval trombiculid mites.

Medium or heavy gauge plastic bags should be used to transport small dead animals or habitat samples for further investigation.

Excerpts from Baker, 1999. Mites and ticks of domestic animals: an identification guide and information source. The Stationery Office, London. 240pp.

4.15. Rat Surveillance

4.15.1. Indications of Rodent Infestation

Look, smell and listen: Top 10 indications of rat infestation

Very distinct smell (a social asset?)
Rat infestations are often first suspected (especially in enclosed places) by the presence of a highly distinctive odour. The smell is a combination of accumulated dried urine, faeces, body secretions (oils) and pheromones. Rats advertise sexual availability by copious urination in many locations across their home range. It tells other rats its gender, age, social status, reproductive status.

Sounds of activity
Large populations of rats can be quite loud. Calling, running, gnawing, and fighting noises especially at night when background noise may be reduced and rat activity is greatest.

Live and/or dead rats
For every rat you see – there may be several you don't.

Grease marks
Rats produce greasy secretions that penetrate their fur to aid scent marking, insulation and fur condition. Rats habitually rub against surfaces to mark their presence (Figure 4.15.1).

Figure 4.15.1. Grease marks produced by rats.

Runways - rat droppings
The greasy secretions will tend to rub off along the pathway used by rats (safety from the unknown). Over time the pathway becomes distinctly marked (Figure 4.15.2a).

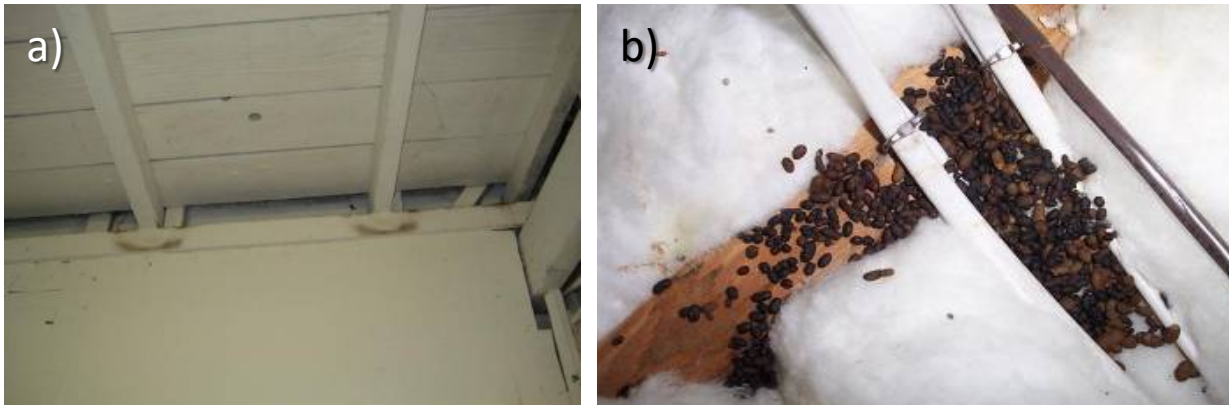


Figure 4.15.2. a) Classic rat runs under flooring joists, b) rat droppings.

Nesting signs and gnawing damage

Figure 4.15.3. Nest and gnawing damage.

Nesting material will be carried to a secluded location. Nests are usually located in warm dry areas with access to food and water. Mechanical areas can be made suitable by bringing in nesting materials (Figure 4.15.3).

4.15.2. Trapping Rats

There are available a number of rat traps (Figure 4.15.4)

Glueboards

Repeating traps

Snap traps

Cage traps

Humane traps
Figure 4.15.4. Different types of rat traps.

General Rules for Trap and Bait Station Placement

- Pre-bait traps (unset) for 1-2 days. Use food not in competition with what is present in the area.
- Set traps close to wall, in dark corners, behind and under objects.
- Set traps perpendicular to walls so that the catch bar snaps towards wall.
- Set glue.

4.15.3. Processing Rats

Rats may be preserved by freezing them; however, a simple ID should be possible on site allowing for destruction rather than preservation unless the specimen needs to be kept for any reason. Please never forward rats to the New Zealand BioSecure Laboratory.

4.15.4. Rat Control

Main sites of infestation in ships:
Tank top ceiling: if, as often happens, cracks appear between the ceiling boards, food material may be forced down into the underlying space and serve as a focus of infestation for an indefinite period. Insects bred in this space can readily move out to attack food cargoes and establish their progeny in them.

Between-deck centre lines, wooden feeders and bins are often left in place for several voyages and because of their construction is a frequent source of infestation. After unloading a grain cargo, burlap and battens covering the narrow spaces between the planks should be removed and discarded before the holds are cleaned or washed down. These coverings should be replaced by new material in preparation for the next cargo.

Transverse beams and longitudinal deck girders which support the decks and hatch openings may have an L-shaped angle-bar construction. Such girders provide ledges where grain may lodge when bulk cargoes are unloaded. The ledges are often in inaccessible places overlooked during cleaning operations.

Insulated bulkheads near engine rooms: when the hold side of an engine room bulkhead is insulated with a wooden sheathing, the air space and the cracks between the boards often become filled with grain and other material.

Sometimes the air space is filled with insulating material which may become heavily infested and serves as a place for breeding. Cargo battens: the crevices at the sparring cleats are ideal places for material to lodge and hide.

Electrical conduit casings: sometimes the sheet-metal covering is damaged by general cargo and when bulk grain is loaded later, the casings may become completely filled. This residual grain has often been found to be heavily infested. Casings that are damaged should be repaired immediately or, where possible, they should be replaced with steel strapping, which can be cleaned more easily.

Other places where material accumulates and where rats breed and hide include:

- The area underneath burlap, which is used to cover limber boards and sometimes to cover tank top ceilings.

- Boxing around pipes, especially if it is broken.
- Corners, where old cereal material is often found.
- Crevices at plate landings, frames and chocks.
- Wooden coverings of manholes or wells leading to double-bottom tanks or other places.
- Cracks in the wooden ceiling protecting the propeller shaft tunnel.
- Beneath rusty scale and old paint on the inside of hull plates.
- Shifting boards.
- Dunnage material empty bags and used separation cloths.
- Inside lockers.

4.15.5. Integrated Pest Management

Integrated pest management recognises that pest (in this case - rat) born risk generally has multiple causes and attempts to provide effective control by applying multiple management strategies in response. Management strategies are selected to best fit the specific context of these risks and can be generally recognised as:

- Organisational Controls
- Cultural Controls
- Physical Controls

Organisational Controls

Organisational controls are most often developed at the macro level. They can include:

- Design specifications and construction, and operational standards to eliminate rodent harbourage or resources in the first instance. Minimising concealed and/or inaccessible structural voids where rats may harbour; engineering specifications including rat proofing at the design stage; protocols for effective food storage and protection, waste management and cleaning; response protocols in the event of rat activity.
- Development and enforcement of regulations including IHA, food hygiene provisions, waste management provisions, immigration/border inspection provisions.
- Cross organisational matters including relationship building between Ministry of Health, port authorities and companies, importers/exporters, shipping companies.
- Reporting and evaluation of import health trends and case studies to provide a feedback loop to improving organisational controls.

Cultural Controls

Cultural controls focus on the human factors that enable or prevent effective application of organisational controls. Having high quality operational protocols is of little point if they are not followed by those responsible for their implementation. There are several key elements to getting the best out of cultural controls. These include:

- Knowledge of the technical issues around pest action and control.
- Motivation to act on that knowledge.
- Capacity to act. Without resources – nothing happens.
- Collective responsibility for the pest management outcomes.

Physical Controls

Maintenance and sanitation:

Ship cargo spaces, tank top ceilings and other parts of the ship should be kept in a good state of repair to avoid infestation. Cleanliness, or good housekeeping, is an important means of controlling pests on a ship. Since pests in general on ships become established and multiply in debris, much can be done to prevent their increase by simple, thorough cleaning. Box beams and stiffeners, for example, become filled with debris during discharge of cargo and unless kept clean can become a source of heavy infestation. It is important to remove thoroughly all cargo residue from deckhead frames and longitudinal deck girders at the time of discharge, preferably when the

cargo level is suitable for convenient cleaning. Where available, industrial vacuum cleaners are of value for the cleaning of cargo spaces and fittings.

Physical controls aim to prevent and eliminate rat infestations at the operational level subject to actual interaction with the physical environment. A number of sub-controls can be considered with operational actions aimed at managing rat risks. Some of these may include:

Exclusion of Rats

- Preventing rats entering ships at port.
- Rat proofing.
- Food protection.
- Cargo protection.

Detection of Rats

- Interview Captain or senior crew regarding any reports or signs of rat activity.
- Perform inspection of the vessel for signs of rat activity.
- Think 3 dimensions – high, low and hidden spaces.
- Lighten dark areas by torch or other artificial means.
- Check food and waste storages in particular.
- Tracking powder can reveal low levels of rat activity.

Elimination of Rats

- Correct structural defects.
- Correct procedural defects.
- Control current rat population.
- Rodenticides (use needs to be considered in context with cargo, risk of cross-contamination and competing food-sources).
- Traps. Can also be used for detection.

<u>Rodenticide application</u>

When the situation permits, rodenticides usually provide the most cost-effective approach to rodent control. Select a rodenticide with an active ingredient and formulation that works well for the particular environment. Correct bait placement is key to an effective integrated pest management program. Proper placement ensures rapid rodent control and protects children, pets and non-target animals from bait contact.

Two primary types of rodenticide bait are available – non-anticoagulants (acute) and anticoagulants.

Non-anticoagulants: Bromethalin and zinc phosphide based products are examples of acute baits which have no antidote. Palatability is generally low with products containing these active ingredients. Non-anticoagulants are considered single-feed baits because rodents typically stop feeding after one meal. If a lethal dose is ingested, rodents usually die within 24 hours. If a sub-lethal dose is eaten, rodents tend to develop bait shyness.

<u>Rodenticide Application Tips</u>

- Neophobia – the fear of new objects – makes Roof rats and Norway rats extremely nervous about changes in their territory. It takes several days for rats to accept a new object in their environment, including bait stations.
- Place rodenticides in areas inaccessible to children and non-target animals, preferably in properly installed, tamper-resistant bait stations. Bait stations not only provide added security for children and non-target animals, but also protect bait from the elements and provide a comfortable place for rodents to feed and groom.

- Use the proper rodenticide for the target rodent and the best formulation for the environment. Wax block types are best in wet areas.
- Using information obtained during the inspection process, place baits in rodent runways as close to their nest as possible.
- Use a sufficient amount of product to assure an uninterrupted supply of bait between service visits.
- In areas of identified mice activity, rodenticide bait placements should be no further than 2-4 meters apart due to their limited home range. Place control material as close to the nest as possible, and between the nest and food source.
- In areas of identified rat activity, rodenticides should be placed every 5-10 meters.
- Pre-baiting is the process of placing non-toxic bait prior to toxic bait in order to increase product acceptance. This practice generally is used for acute baits (e.g. zinc phosphide) with low palatability.

Trapping Tips

- In sensitive areas where rodenticide use is not permitted, traps are especially useful. Traps also prevent rodent deaths in inaccessible areas. After rodents and their patterns have been identified, follow the appropriate trapping methods.

  Trapping methods:
  - Store snap traps away from insecticides and chemicals that may impart a flavour. Remember, rodents have a keen sense of taste.
  - Bait snap traps with food that is more attractive than other readily available food sources, such as gumdrops, peanut butter, bacon, nutmeats or dried fruit (raisins). Secure bait to the snap trap trigger – a length of thread works well. For rats, fish (tuna) and meat (cat/dog food) may be used to bait traps. Glue boards can be baited, if necessary, with non-oily foods. The use of peanut butter, bacon and other oily, greasy foods will cause the glue to lose its stickiness.
  - Bait some mouse snap traps with nesting materials, such as cotton or dental floss, with a drop of vanilla. Mice constantly look for nesting material.
  - Place mechanical or snap traps and glue boards in areas unsuitable for rodenticide applications.
  - Position snap traps and glue boards to intercept rodents in runways. Place snap traps with the trigger toward the runway – generally along a wall, in corners, behind and under objects and near abundant tracks and droppings. Snap traps also may be attached to pipes and beams used as runways.
  - More traps are better than fewer traps.
  - Pre-bait traps until rodents, especially rats, overcome their fear and take bait readily. This may take several days for mature rats.
  - Glue boards shouldn't be used in areas with excessive dust or wetness – both elements make glue boards ineffective.
  - Check glue boards frequently to prevent rodents from escaping.
  - For mice, repeating or automatic mechanical traps may be used. Watch for tracks in the dust on the top of low-profile traps, which indicate mice are running over the top of them.

Rat guards

- Rat guards are required to be placed when visiting ports where plague is endemic. Moreover, the ship operator policy of the local regulation may require the placement of rat guards. The proper placement of rat guards is shown below:
  - Rat guards should have a 36-inch (91.4 centimetre) minimum outside diameter, a cone angle of 30 degrees, and be made of 18-gauge steel or aluminium.

- Rat guards must be mounted with the point of the cone toward the ship on all tending lines, at least 6 feet (1.8 metres) from the pier and greater than 2 feet (0.6 metres) from the ship.
- Use rags to plug gaps, securing the rags tightly to prevent loosening or being pulled apart by the rat.
- Ensure stray lines are kept out of the water.
- If 2 lines are in close proximity to each other, either group the lines to pass through a single rat guard, or install the rat guards side-by-side or touching to prevent rats from jumping from one line to another, skirting the rat guards and making them ineffective.

Source: US Navy shipboard pest control manual (2003). USA Department of the Navy and Navy Disease Vector Ecology and Control Center (Bancor, Washington) reviewed by J.A. Corneil, Washington.

A good vector management plan should contain the following:

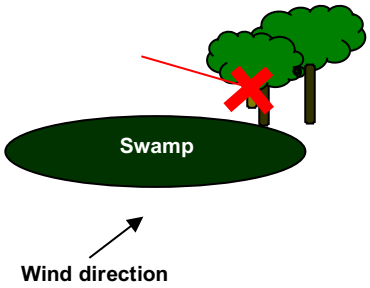
- Training of management team
- Crew positions and responsibilities
- List of vectors that can be found onboard
- Active surveillance (e.g. visual inspections using a flash light at night in high risk areas)
- Passive surveillance (placement of traps)
- Schedule of visual inspections
- Schedule of trap placement (locations) representative of the ship
- Records of findings
- Surveillance and control measures applied
- List of pesticides carried onboard
- Revision as appropriate

5. Appendix

Appendix 1 Setting up a new LT

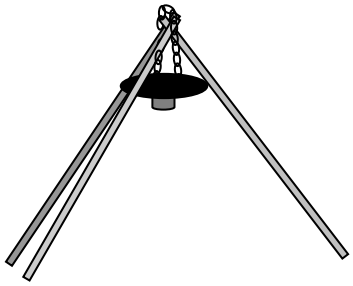
Find a suitable site for your trap

- Trap should be placed as near to a larval site as possible
- When positioning the trap check for vegetation nearby where mosquitoes will seek shelter, and if possible use trees to hang your trap from.
- If there are no trees near to the sites, assemble a tripod to hang your trap from.



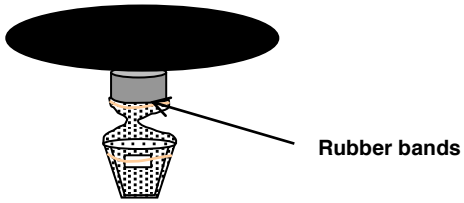
Assemble the Tripod and Hanging the Trap

- Assemble tripod using the three stakes and duct tape or wire.
- Hang the chain over the tripod (or tree branch) and attach the light trap using the miniature carabina clips.



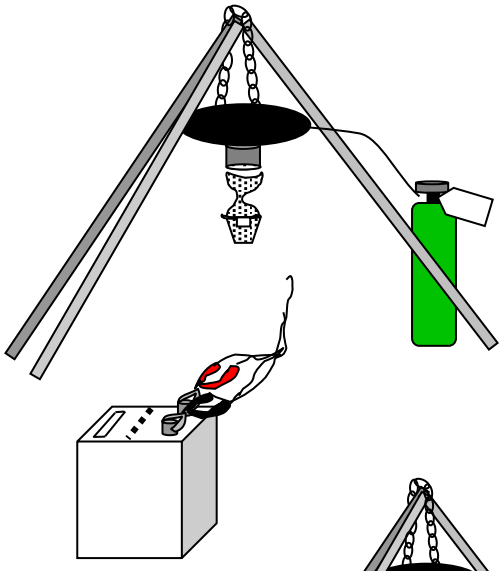
Attach collection cup to base of trap

- Using a rubber band. Ensure there are no holes in the stocking and that the rubber band will hold the cup in place.



Attach CO_2 cylinder

- screw in regulator & tighten using spanner
- attach gas hosing to light trap fan housing with tape
- turn gas on
- padlock gas bottle to tripod or tree using a chain

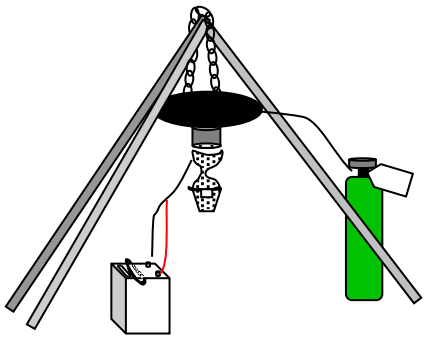


Attach battery

- Red to positive terminal
- Black to negative terminal

Final Checks

- Check that the light is on and the fan is working.
- Check the gas is on and flowing through the tube.



Appendix 2 Processing a Light Trap (LT) - Checklist

- ❑ Notify site security of your intention to visit surveillance sites

Processing LT

- ❑ Twist sock
- ❑ Remove capture cup
- ❑ Close sock with rubber band or a peg
- ❑ Mark or label cup
- ❑ Remove battery clamp
- ❑ Swap old battery for new one
- ❑ Attach battery clamp
- ❑ Turnoff CO_2 cylinder
- ❑ Remove regulator from cylinder
- ❑ Replace CO_2 cylinder with full cylinder
- ❑ Attach regulator to cylinder
- ❑ Turn on CO_2 cylinder

  - If you realise that either the battery or the cylinder last shorter or longer than usual, take in for service. The regulators can be sent in any time to NZBEL but should be at least once a year for calibration (0.4 l/hr)

Processing LT samples

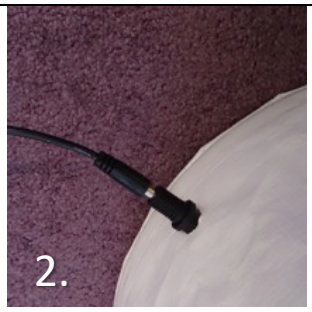
- ❑ Empty sample in to dish.
- ❑ Remove non mosquitoes.
- ❑ Put sample back into tube.
- ❑ Add label with unique identifier, sampler, date and site ID written in pencil to tube.
- ❑ Put tubes in package to send to lab.
- ❑ Add sample records to database (incl trap location!).
- ❑ Send samples to the lab.

Appendix 3 Assembling and Processing a BG Trap

<table>
<tr>
<td rowspan="2">1. To set up trap first place the uprights in the slots around the inside edge of the trap.

2. Place cable from power source through opening in bottom of trap.

3. Place attractant under mesh inside main body of trap – If being used.

4. Cover top of trap with white gauze mesh.</td>
<td> 
1.</td>
<td> 
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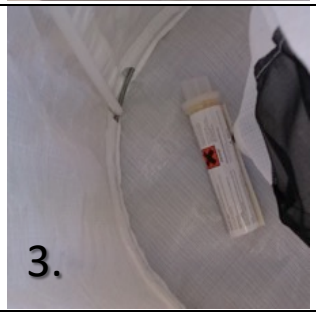
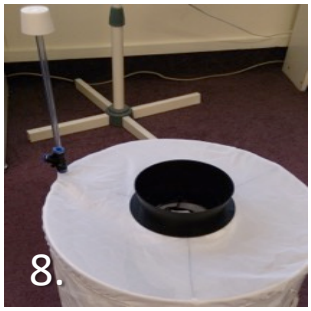
<td> 
3.</td>
<td> 
4.</td>
</tr>
<tr>
<td>5. Place the netting and catch bag over the bottom of the funnel.

6. With the netting inside the catch bag. Ensure seams of catch bag are on the outside to avoid specimens getting caught in seam.</td>
<td> 
5.</td>
<td> 
6.</td>
</tr>
<tr>
<td>7. Insert funnel inside trap.
8. If CO_2 being used, add CO_2 diffuser and attach CO_2 Cylinder.
9. If needed trap can be secured using tent pegs through hoops on outside.
10. Connect main power plug or battery clamps -Red to positive terminal- Black to negative terminal</td>
<td> 
7.</td>
<td> 
8.</td>
</tr>
</table>

Processing the BG trap

- Partially lift the funnel out of the trap so suction still affecting catch bag.
- Remove catch bag and tighten cord of catch bag.
- After removal of catch bag trap can be turned off or re-set.
- Place catch bag in freezer for at least an hour.
- Remove specimens from catch bag with tweezers.

Appendix 4 Assembling and Processing the GAT

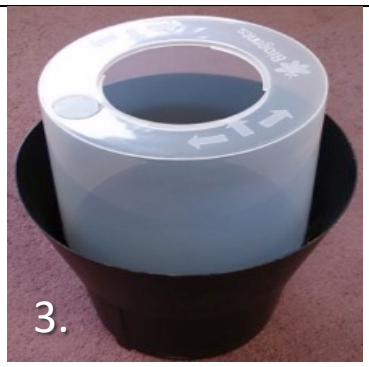
1. The base of the trap will need to be filled with water and a rabbit pellet added to the water.

2. Attach the mesh to the clear plastic container with the mounting ring.

   The inside of the container should be sprayed with a contact insecticide.

3. Place clear plastic container inside black container above water.

4. The funnel will be placed last with mesh, if used, also sprayed with a contact insecticide. No power or CO_2 needs to be connected.



Processing the GAT

- Due to the insecticide any adults inside the trap should be dead.
- Use forceps to collect specimens found on mesh inside trap.
- Old water should be removed and then new water and rabbit pellets added.
- Inside of trap can then be re-sprayed with insecticide.
- Once re-sprayed trap can be replaced.

Appendix 5 Processing larval tyre traps - Checklist

- ❑ Notify site security of your intention to visit surveillance sites

Processing tyre traps

- ❑ Pour contents of trap into a white tray.
- Do not scrub if tyre was negative and water from tyre is clear.
- ❑ Remove larvae from white tray using pipette.
- ❑ Take all larvae (or the majority) into one standard sample tube (Siphon off extra water to allow more larvae to be added. Disturbing the larvae will move them to the bottom of the tube allowing more water to be removed).
- ❑ Add label with unique identifier, sampler, date and site ID written in pencil to tube.
- ❑ Add details to collector's notebook.
- For tyres number of dips is always 1
- Trap nights will be 7 if surveillance occurs on same day each week.
- ❑ Refill tyre with unused but aged water.
- ❑ Add a Lucerne (rabbit) pellet to tyre.
- ❑ Add 2 pellets of S-methoprene.
- ❑ Replace tyre and check signage.

Processing larval samples

- ❑ Remove water from tube.
- ❑ Add 70% ethanol in to tube.
- ❑ Add label with unique identifier, sampler, date and site ID written in pencil to tube.
- ❑ Put tubes in package to send to lab.
- ❑ Add sample records to database (complete all the information required).
- ❑ Send samples to the lab.

Appendix 6 Mosquito Sample Collection Sheet

Organisation: _____
Collector: _____
Date: _____
Time: _____
Location: _____
Site reference number/trap number: _____
GPS E: _____
GPS N: _____
Sample number: _____

Surveillance type: ◯ Port ◯ Airport Other _____
Reason: ◯ Response ◯ Routine Other _____
Sample type: ◯ Larvae ◯ Adult Other
Habitat category: _____ Salinity (ppt): Temp (°C):
Trap type _____

Total # dips: _____ # +ve dips: _____
Trap nights: _____
Attractant: _____
Treatment: _____

Background Information (habitat description, weather, water event triggering hatch etc.)

HABITAT CATEGORIES

1. **Flowing stream**
2. **Ponded Stream**
3. **Lake edge**
4. **Swamp/Marsh**
5. **Permanent Pond**
6. **Temporary Pond**
7. **Intermittent ephemeral puddle**
8. **Natural container**
9. **Artificial Container**
10. **Subterranean habitats- natural**
11. **Subterranean habitats- artificial**

Results

	Larvae-Instar				Adults	
Species	1	2	3	4	Female	Male
Other:						
Exotic:						
Identified by (print):				Date identified:		
				Date results sent:		

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