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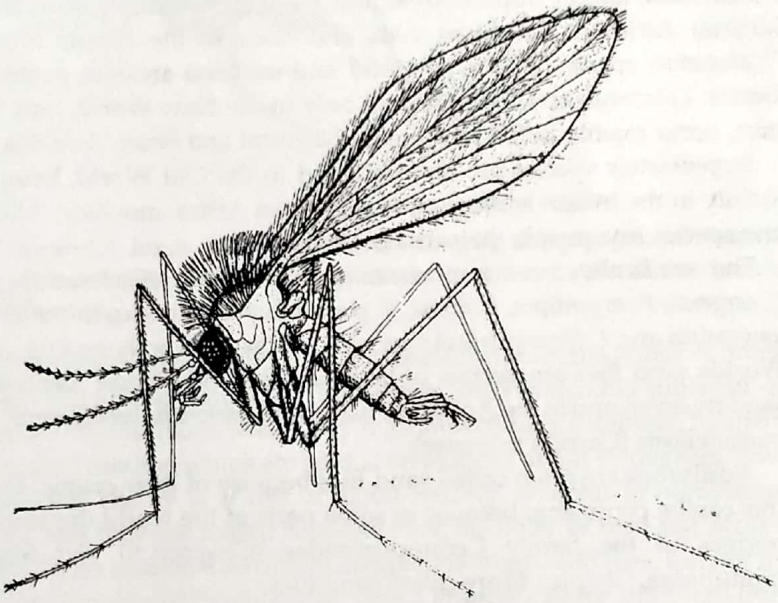
Phlebotomine sand flies (Phlebotominae)



Within the subfamily Phlebotominae of the family Psychodidae it is estimated that there are approaching 1000 species and subspecies of sand flies, in five or six genera (depending on whether *Psychodopygus* is considered a subgenus or genus). Three genera – *Phlebotomus*, *Lutzomyia* and *Sergentomyia* – suck blood from vertebrates, the former two being the more important because they contain disease vectors.

The genus *Phlebotomus* occurs only in the Old World, from southern parts of northern temperate areas, mainly the Mediterranean region, to central Asia, and in tropical areas, but there are not many species in sub-Saharan Africa or Southeast Asia and none in the Pacific area. Most *Phlebotomus* species inhabit semiarid and savanna areas in preference to forests. *Lutzomyia* species are found only in the New World, and, by contrast, occur mainly in forested areas of Central and South America.

Sergentomyia species are also confined to the Old World, being found mainly in the Indian subregion, sub-Saharan Africa and Asia. Although a few species bite people they are not vectors.

The medically most important species include *Phlebotomus papatasi*, *P. sergenti*, *P. argentipes*, *P. ariasi*, *P. perniciosus* and species in the *Lutzomyia longipalpis* and *L. flaviscutellata* species complexes. In both the Old and New Worlds sand flies are vectors of leishmaniasis and viruses responsible for sand fly fever, and in the Andes the bacterium *Bartonella bacilliformis*, causing bartonellosis (Carrión's disease).

Adult flies are often called sand flies because of their colour. However, this can be confusing, because in some parts of the world the small biting midges of the family Ceratopogonidae (Chapter 6) and black flies (Simuliidae, Chapter 4) are called sand flies.

5.1 External morphology

Adults of *Phlebotomus* and *Lutzomyia* are difficult to distinguish, but as the former genus is found only in the Old World and the latter in the New World this is not a problem.

Adult phlebotomine sand flies are readily recognized by their minute size (usually less than 5 mm long), hairy appearance, relatively large black eyes and long and ***stilt-like*** legs (Plate 6). The only other blood-sucking flies which are as small as this are some species of biting midges (Ceratopogonidae), but these have non-hairy wings and differ in many other details (Chapter 6). Phlebotomine sand flies have the head, thorax, wings and abdomen densely covered with long hairs. The 16-segmented ***antennae*** are long and composed of small bead-like segments having short hairs; antennae are similar in both sexes. The mouthparts are short and inconspicuous and adapted for blood-sucking, but ***only females*** bite. At their base is a pair of five-segmented maxillary palps which are relatively conspicuous and droop downwards.

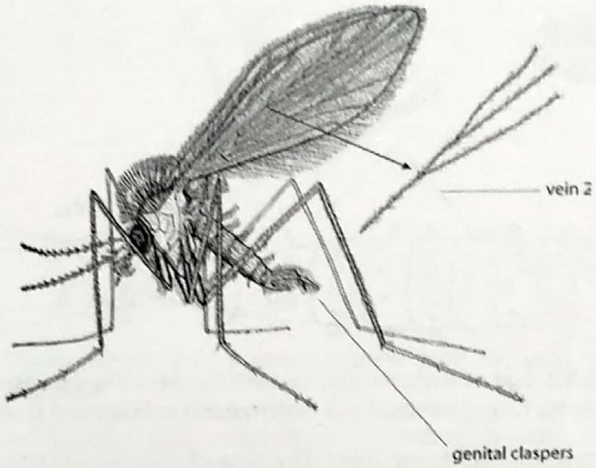


Figure 5.1 Adult male phlebotomine sand fly, showing genital claspers at end of abdomen, and a diagrammatic representation of the double branching of wing vein 2.

Wings are lanceolate in outline and quite distinct from the wings of other biting flies. The Phlebotominae can be distinguished from other subfamilies of the family Psychodidae, which they may superficially resemble, by their *wings*. In sand flies the wings are held at an angle of about 40 degrees over the body when the fly is at rest or blood-feeding, whereas in non-biting psychodid flies they are folded roof-like over the body or held flat across the body. Wing venation also differs. In phlebotomine sand flies, but not in the other subfamilies of Psychodidae, ***vein 2 branches twice***, although this may not be apparent unless most of the hairs are rubbed from the wing veins (Fig. 5.1).

The abdomen is moderately long and in the female more or less rounded at the tip. In males it terminates in a prominent pair of genital claspers (Fig. 5.1) which give the end of the abdomen an upturned appearance.

Identification of adult phlebotomine sand flies to species is difficult and usually necessitates the examination of internal structures, such as the arrangement of the teeth on the cibarial armature, the shape of the spermatheca in females, and in males the structure of the external genitalia (terminalia).

5.2 Life cycle

The minute eggs (0.3–0.4 mm) are more or less ovoid in shape and usually brown or black, and careful examination under a microscope reveals that they are patterned, as shown in Figure 5.2. Some 30–70 eggs are laid singly at each oviposition. They are thought to be deposited in small cracks and holes in the ground, at the base of termite mounds, in cracks in masonry, on stable floors, in poultry houses, amongst leaf litter and in the Americas

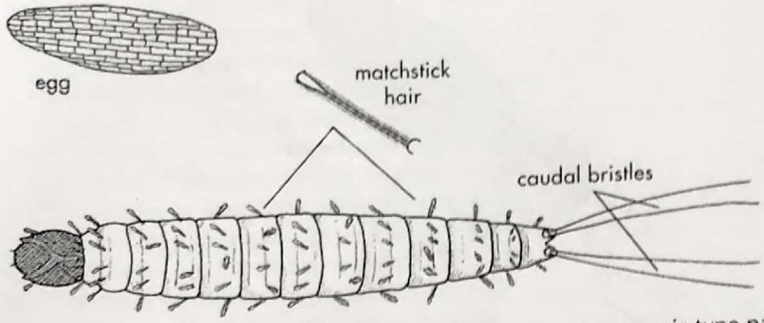


Figure 5.2 Egg of a phlebotomine sand fly, showing mosaic-type pattern, and a 12-segmented larva with matchstick hairs and caudal setae.

between buttress-roots of forest trees. The type of oviposition site presumably varies greatly according to species.

Although eggs are not laid in water they require a microhabitat with high humidity. They are unable to withstand desiccation and hatch after 4–20 days, although hatching may likely be delayed in cooler weather. Larvae are mainly scavengers, feeding on organic matter such as fungi, decaying forest leaves, semi-rotting vegetation, animal faeces and decomposing bodies of arthropods. Although some species, especially of the genus *Phlebotomus*, occur in semiarid areas, the actual larval habitats must have a high degree of humidity. Larvae may be able to survive by migrating to drier areas if their breeding places are temporarily flooded.

There are four larval instars. The mature larva is 3–6 mm long and has a well-defined black head which is provided with a pair of small mandibles; the body is white or greyish and has 12 segments (Fig. 5.2). Ventrally the abdominal segments have small pseudopods, but the most striking feature is the presence on the head and all body segments of conspicuous thick bristles with feathered stems, which in many species have slightly enlarged tips. They are called ***matchstick*** hairs, and they identify larvae as those of phlebotomine sand flies. In most species the last abdominal segment bears two pairs of conspicuous long hairs called the ***caudal setae***. First-instar larvae have two single bristles, not two pairs.

Larval development is usually completed after 20–30 days, the duration depending on the species, temperature and availability of food. In temperate areas and arid regions sand flies may overwinter as diapausing fully grown larvae. Prior to pupation the larva assumes an almost erect position in the habitat, the skin then splits open and the pupa wriggles out. The larval skin is not completely cast off but remains attached to the end of the pupa. The presence of this skin, with its characteristic two pairs of caudal bristles, aids in the recognition of the phlebotomine pupa. The pupal shape is as shown in Figure 5.3. Adults emerge from the pupae after about 6–13

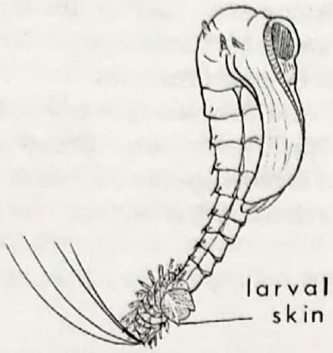


Figure 5.3 Pupa of a phlebotomine sand fly, with larval skin still attached.

days. The life cycle, from oviposition to adult emergence, is 30–60 days, but extends to several months in some species with diapausing larvae. In temperate areas adults die off in late summer or autumn and species overwinter as larvae, with the adults emerging the following spring. It is usually extremely difficult to find larvae or pupae of sand flies, and relatively little is known about their biology and ecology.

5.2.1 Adult behaviour

Both sexes feed on plant juices and sugary secretions, but ***females*** in addition suck blood from a variety of vertebrates, including livestock, dogs, urban and wild rodents, snakes, lizards and amphibians; a few species feed on birds. Females of many *Phlebotomus* species in the Old World and *Lutzomyia* species in the New World bite mammals, including humans. Biting is usually restricted to crepuscular and nocturnal periods, but people may be bitten during the day in darkened rooms, or in forests during overcast days. Most species feed out of doors (***exophagic***) but some species also feed indoors (***endophagic***). A few species are ***autogenous***, that is they can lay eggs without blood-feeding.

Adults are ***weak fliers*** and usually disperse 100 m or less from their larval habitats. Consequently biting is often very localized. However, adults of at least some species have been known to fly up to 2.2 km over a few days. When close to a host sand flies may have a characteristic hopping type of flight, so that there may be several short flights and landings before females settle on a host. Even a light wind inhibits flight activities and biting. Because of their very short mouthparts sand flies are unable to bite through clothing.

During the day adult sand flies rest in sheltered, dark and humid sites, but on dry surfaces, such as on tree trunks, on ground litter and foliage of forests, in animal burrows, termite mounds, tree-holes, rock fissures, caves, cracks in the ground and inside human and animal habitations. Species that commonly rest in houses (***endophilic***) before or after feeding on humans

are often referred to as domestic or peridomestic species. Examples are *Phlebotomus papatasi* in the Mediterranean area, *P. argentipes* in India and the *Lutzomyia longipalpis* species complex in South America.

In temperate areas of the Old World sand flies are seasonal and adults occur only in the summer months. In tropical areas some species are common more or less throughout the year, but in other species there may be well-marked changes in abundance of adults related to the dry and wet seasons.

5.3 Medical importance

5.3.1 Annoyance

About 70 species are vectors of disease to humans, but apart from their importance as vectors, sand flies may constitute a serious, but usually localized, biting nuisance. In the Americas up to 100 bites per night have been recorded. In previously sensitized people their bites may result in severe and almost intolerable irritations, a condition known in the Middle East as harara.

5.3.2 Leishmaniasis

Leishmaniasis is a term used to describe a number of closely related diseases caused by about 30 distinct species, subspecies and strains of *Leishmania* parasites. Worldwide there are about 1–2 million cases a year, with about 12 million people currently infected in 88 countries. The three main clinical forms are cutaneous, mucocutaneous and visceral leishmaniasis. A fourth, less common form is diffuse cutaneous leishmaniasis, while post-kala-azar dermal leishmaniasis is caused by *Leishmania donovani donovani* following cure of the initial visceral form. The epidemiology of leishmaniasis is complex, involving not only different parasite species but different strains of parasites and different reservoir hosts.

Basically ***amastigote*** parasites ingested by female sand flies with a blood-meal multiply in the gut and develop into ***promastigotes***, which are elongate, have a flagellum and attach to the mid-gut or hind-gut wall and multiply rapidly. Many, however, are voided when the fly defecates. After further development the survivors migrate to the anterior part of the mid-gut and then to the fore-gut. Here some parasites become ***metacyclic*** forms. Four to 25 days after the sand fly has taken an infective blood-meal the metacyclic forms are found in the mouthparts, and are introduced into a new host during feeding. Infective flies frequently probe more often than uninfected flies, thus maximizing transmission of parasites during blood-feeding. Previous feeding by females on sugary substances, mostly obtained from plants, is essential not only for the survival of the sand fly but also for the development of the parasites to the infective form.

Most types of leishmaniasis are ***zoonoses***. The degree of involvement of humans varies greatly from area to area. The epidemiology is largely

determined by the species of sand flies, their ecology and behaviour, the availability of a wide range of non-human hosts, and also by the species and strains of *Leishmania* parasites. In some areas, for example, sand flies will transmit infections almost entirely among wild or domesticated animals, with little or no human involvement, whereas elsewhere animals may be important ***reservoir hosts*** of infection for humans. In India infections may be transmitted between people by sand flies, with animals taking no identifiable part in transmission. The epidemiology of the leishmaniases is complex, and only simplified accounts are given below.

Cutaneous leishmaniasis (CL)

In the Old World, CL is known also as ***oriental sore***. It occurs mainly in arid areas of the Middle East to northwestern India and central Asia, in North Africa and various areas in East, West and southern Africa. The principal parasites are *Leishmania major*, transmitted mainly by *Phlebotomus papatasi*, and *Le. tropica*, transmitted by *P. sergenti*. *Leishmania major* is usually zoonotic and in most of its range gerbils (e.g. *Rhombomys opimus*) are the reservoir hosts; *Le. tropica* occurs in densely populated areas and humans appear to be the main reservoir hosts. In the New World, CL is found mainly in forests from Mexico to northern Argentina, and is caused by *Leishmania braziliensis*, *Le. amazonensis* and *Le. mexicana*. Rodents and dogs appear to be reservoir hosts. Vectors include *Lutzomyia wellcomei* and *L. flaviscutellata*.

Mucocutaneous leishmaniasis (ML) (espundia)

A severely disfiguring disease found from Mexico to Argentina. It is mainly caused by *Leishmania braziliensis*. Dogs may be reservoir hosts. *Lutzomyia wellcomei* is an important vector.

Diffuse cutaneous leishmaniasis (DCL)

A form that causes widespread cutaneous nodules or macules over the body. It is confined to Venezuela and the Dominican Republic and the highlands of Ethiopia and Kenya. In South America the parasite is *Le. amazonensis*, transmitted by *Lutzomyia flaviscutellata*, and spiny rats (*Proechimys* species) are reservoir hosts. In Ethiopia and Kenya the parasite is *Le. aethiopica*, transmitted by *Phlebotomus pedifer* and *P. longipes*, with rock hyraxes (*Procavia capensis*) as reservoir hosts.

Visceral leishmaniasis (VL)

Often referred to as ***kala-azar***. It is caused by *Leishmania donovani donovani* in most areas of its distribution, such as India, Bangladesh, Sudan, East Africa and Ethiopia. Among the vectors are *Phlebotomus argentipes* and *P. orientalis*. Rodents, wild cats and genets (*Genetta genetta*) may be reservoir hosts. In

the Mediterranean basin, Iran and central Asia, including northern and central China, *Leishmania donovani infantum* is the parasite, and the vectors include *P. ariasi* and *P. perniciosus*. Dogs and foxes (*Vulpes vulpes*) are reservoir hosts. Visceral leishmaniasis also occurs sporadically in Central and South America, where the parasite is *Le. donovani infantum* (*Le. chagasi* of some authors), transmitted by species in the *Lutzomyia longipalpis* complex.

5.3.3 Bartonellosis

Bartonellosis, sometimes called Oroya fever or Carrión's disease, is encountered in arid mountainous areas of the Andes, mainly in Peru, but also in Ecuador and Colombia. It is caused by the bacterium *Bartonella bacilliformis* and is transmitted in Peru by *Lutzomyia verrucarum* and *L. peruensis*, and by *L. colombiana* in Colombia, but there are probably other vectors. Transmission is possibly only by contamination of the mouthparts. Apart from humans there are no other vertebrate reservoir hosts.

5.3.4 Sand fly fevers

Sand flies transmit the seven viral serotypes responsible for sandfly fevers, also called papataci fever (sometimes spelt papatasi or pappataci fever), three-day fever or *Phlebotomus* fevers. The classical form of the disease is found in the Mediterranean region, but it also extends up the Nile into Egypt, and from the Middle East to northern India, Pakistan, Afghanistan and China. The most important vectors in the Old World are *P. papatasi* and *P. perniciosis*. Other forms of the virus in Central and South America are transmitted by *Lutzomyia* species such as *L. trapidoi* and *L. ylephiletor*.

Female sand flies become infective 7–10 days after taking a blood-meal. Infected females lay eggs containing the virus, and these eventually give rise to infected adults. This is an example of *transovarial* transmission, a phenomenon that is more common in the transmission of various tick-borne diseases (Chapters 16 and 17). There are possibly mammalian reservoir hosts, and in fact infected gerbils (*Rhombomys opimus*) have been found in Iran, but in many areas it is likely that humans are the main reservoir of infection.

Sand fly fever virus is recognized as a potential bioterrorism agent.

5.4 Control

Although phlebotomine sand flies are very susceptible to insecticides, until recently there have been few organized attempts to control them. However, in most areas where house-spraying has been used to control *Anopheles* vectors there have been large reductions in sand fly populations followed by interruption of leishmaniasis transmission. When houses in Kabul, Afghanistan, and in the Peruvian Andes were sprayed with the pyrethroid lambda-cyhalothrin, cutaneous leishmaniasis was reduced by 60% and 54% respectively. In the Americas, in countries such as Brazil and

Venezuela, spraying houses with lambda-cyhalothrin has substantially reduced the vectors of cutaneous leishmaniasis.

In areas where sand flies rest indoors, cattle sheds and chicken houses should also be sprayed. Obviously where sand flies bite and rest out of doors house-spraying will have little effect. However, if the outdoor resting sites are known (e.g. animal shelters, stone walls, tree trunks, termite hills) they can be sprayed with residual insecticides. Insecticidal fogging of outdoor resting sites may also give some, but temporary, control of vectors.

Personal protection can be achieved by applying efficient insect repellents such as DEET, piperidene-based ones and neem oil. Insecticide-impregnated bed-nets such as the long-lasting Olyset nets (see Chapter 2) will give protection from sand fly biting. For example, in Afghanistan and Syria insecticide-treated polyester bed-nets gave good protection against *Phlebotomus sergenti*, an important vector of cutaneous leishmaniasis (*Le. tropica*).

Control of sand fly larvae remains impossible, because the breeding sites of most species are unknown.

Because most leishmaniasis transmission involves ***reservoir hosts***, such as rodents and dogs, attempts have been made to destroy them. In China leishmaniasis was effectively eliminated in the 1950s by killing dogs, but although similar culls have been made in parts of Brazil and the Mediterranean region results have been disappointing. Dogs have sometimes been dipped or sprayed with pyrethroids such as deltamethrin, but repeated treatments, typically every 2–3 months, are needed. Deltamethrin-treated collars on dogs, which can remain effective for eight months, have also given good, albeit local, control of *Le. donovani infantum* in Italy and Iran. In the Mediterranean there are millions of dogs, of which 1–40% are infected with visceral leishmaniasis. Recently a new vaccine has given complete and lasting protection against leishmaniasis in dogs, thus reducing the reservoir of infection. In Russia and Jordan zoonotic cutaneous leishmaniasis has been controlled by destroying rodent colonies, but elsewhere results have not been encouraging.

Resistance to DDT was found in *P. papatasi* and *P. argentipes* in India and parts of Nepal, and greater tolerance or resistance to several insecticides, such as pyrethroids, has been reported in other sand flies.

Over 60% of visceral leishmaniasis occurs in India, Bangladesh and Nepal, and an estimated 150 million people are at risk. In 2005 these countries signed an agreement to eliminate VL by 2015, mainly based on integrated vector control.

Further reading

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Ashford, R, W. (2001) Leishmaniasis. In M. W. Service (ed), *The Encyclopedia of Arthropod-Transmitted Infections of Man and Domesticated Animals*. Wallingford: CABI, pp. 269–79.