# Chapter 3

# Sampling the Adult Resting Population

Direct bait catches and indirect ones in which the host is enclosed in a trap usually catch only unfed hungry females in search of a blood-meal. Traps employing attractants such as carbon dioxide, light or some other visual stimuli, also mainly attract mosquitoes primarily concerned with host seeking, and, moreover, are usually species selective. Non-attractant traps, such as suction traps, give less biased collections of mosquitoes and should sample all species more or less equally. However, they only sample the aerial population and unfed females being the most active category again predominate, but in this instance not all will necessarily be host seeking.

Although most trapping and sampling procedures are orientated to catching actively flying mosquitoes, adults probably pass more time resting in natural or man-made shelters than in flight. Collections of such resting populations usually provide more representative samples of the population as a whole than most other methods. In addition to unfed females, not all of which may be concerned with blood feeding, males and both blood-fed and gravid females are caught. The collection of blood-engorged adults is particularly useful as they can be used to study natural host preferences. There are other advantages in sampling the resting population, for example the age structure may be more representative than that based on samples of adults caught at bait or in traps.

Only very few mosquito species commonly rest in man-made shelters, most rest in natural shelters, such as amongst vegetation, in hollow trees, animal burrows, and crevices in the ground, etc. Unfortunately searches for outdoor resting mosquitoes have frequently proved time consuming and unrewarding (e.g. Bahang *et al.*, 1984; Bown & Bang, 1980; Downe, 1960; Magnarelli, 1977*b*; Muirhead-Thomson, 1956; Muirhead-Thomson & Mercier, 1952; Ree *et al.*, 1976; Rehn *et al.*, 1950). However, worthwhile numbers of mosquitoes have sometimes been caught from vegetation with motorised aspirators or by sweep-netting and from artificial resting shelters, especially from the 1980s onwards (Breeland, 1972*a,b*; Chandler *et al.*, 1976; Cordellier *et al.*, 1983; Day *et al.*, 1990; McHugh, 1989; Mani & Devaputra, 1988; Mutero *et al.*, 1984; Nasci & Edman, 1984; Nayar, 1982; Senior White, 1951; Wharton, 1950).

## THE INDOOR RESTING POPULATION

Although only comparatively few mosquito species regularly rest in human and animal habitations those that do are often important vectors of malaria, filariasis and more rarely arboviruses. For example, in Pakistan Reisen & Milby (1986) caught more than 14 species of mosquitoes resting inside houses and cattle sheds, the most common of which were *Anopheles stephensi, Culex quinquefasciatus, Anopheles culicifacies, Anopheles annularis* and *Anopheles subpictus*. In another study in Pakistan Reisen *et al.* (1982) caught 13 mosquito species from houses, and 17 species from cattle sheds, in both sites the most common two species were *Culex quinquefasciatus* and *Culex tritaeniorhynchus*. In El Salvador Lowe & Bailey (1979) found that 1-hr collections between 0800–1000 hr or 1845–2020 hr in stables provided good numbers of female *Anopheles albimanus*. In general more were collected in the mornings than in the evening collections, and morning catches provided a higher proportion of blood-fed individuals. Mosquitoes were transferred to gauze covered cages placed in a styrofoam cold box and covered with a damp cloth. Despite being kept in the field until late afternoon, there was a mortality of only about 8·2%. In Mexico in an evaluation of insecticidal residual house-spraying, aspirator collections of *Anopheles albimanus* for 15 min/house were made weekly between 0900–1100 hr in 32 selected village houses (Bown *et al.*, 1984). Similar collections were also made from animal corrals and from vegetation, but from 2000–2200 hr. In India Yasuno & Rajagopalan (1977) employed 15-min aspirator collections in store rooms of houses and in cattle sheds to monitor seasonal changes in *Culex quinquefasciatus* adult densities.

Mosquitoes are usually caught from houses and animal quarters with aspirators or by knock-down pyrethrum spray collections. Torches are used to both locate indoor resting mosquitoes and also to aid the collection of adults that have been knocked down by space spraying. Russell *et al.* (1963) recommended a torch that fits on to the head like a miner's lamp so that both hands are left free; such torches are often used by spaeleogists. The light should not be too powerful otherwise when it is shone on resting mosquitoes it may cause them to take flight. When collecting mosquitoes from African huts Haddow (1942) placed greaseproof paper over the torch to diffuse the light. Grimstad & DeFoliart (1974) collected mosquitoes visiting flowers at night with a torch strapped to the head that used a red filter over the bulb. The use of red filters or bulbs have also often been used in bait collections to reduce the likelihood of disturbing mosquitoes settling on the bait.

### Resting counts

This method has been used in studies on the effect of residual insecticides on house resting mosquitoes (Smith, 1964). The mosquitoes resting on the walls and roof of a hut are carefully counted but not caught, so that both the behaviour of the mosquitoes and the toxicity of the insecticide can be assessed. In East Africa Smith (1964) found that only about 47 and 61% of the total *Anopheles gambiae* population resting in grass and mud-roofed huts, respectively, were in fact counted. A large number are obviously missed by this method. The mosquitoes must not be disturbed to resettle elsewhere otherwise they may be recounted. Hibernating females are more easily counted than non-hibernating mosquitoes as they are not so readily disturbed. Weekly counts have been made of hibernating populations of *Culex pipiens* and/or *Culiseta annulata* in England in brick-built shelters (Service, 1969; Sulaiman & Service, 1983) and caves to show their seasonal build-up and decline in numbers, and also to study the rate of spread of fungal infections in *Culex pipiens* populations. In Japan populations of *Culex pipiens* form *pallens*, and a few other species, were studied by counting the resting adults in a cave every 7–10 days for 16 months (Shimogama & Takatsuki, 1967). The position of resting adults on a wall or roof can be marked with a pencil, so that the approximate proportion of the population that moves in-between sampling days can be determined (Service, 1969). In Japan Makiya & Taguchi (1982) studied the movement of overwintering *Culex pipiens* form *pallens* in underground air-raid shelters by marking off 37 squares (50 × 50 cm) and photographing adults resting in each square. Their numbers and position were checked weekly with their photographs. It was observed that about 70% of the adults changed their position from week to week. Also in Japan Natuhara *et al.* (1991) measured seasonal fluctuations of *Culex tritaeniorhynchus* by counting the numbers resting on grey sheets (100 × 145 cm) marked with grid lines and pinned to the walls of cow sheds. Temporal patterns of abundance agreed well with collections in light-traps.

## Tubes

The simplest, but not necessarily the most efficient, method of collecting resting adults is to carefully place a plastic or glass tube, such as a test tube, over them, and to plug the open end with a piece of cotton wool (Russell & Baisas, 1935; Russell *et al.*, 1963). If the cotton wool is pushed down to confine the mosquito to the end of the tube, several adults isolated by cotton wool plugs can be collected in each tube. A collecting tube can be made into a killing tube by placing cut-up pieces of rubber bands or bicycle inner tube in the bottom of a suitable sized test tube (e.g. 150 × 18 or 24 mm) and flooding them with chloroform. After allowing the fluid to be soaked up the excess is poured away and a layer of absorbent cotton wool inserted, which is then covered with 1–2 layers of white blotting or filter paper (Fig. 3.1a). This paper layer should remain dry; if it becomes wet too much chloroform remains in the tube which then should be left uncorked for it to evaporate. Such a killing tube remains effective for several weeks, if not months. To recharge it the cotton wool and filter paper discs are removed and the rubber bands resoaked with chloroform. Alternatively, crushed potassium or sodium cyanide is placed in the bottom 6 mm of the tube and covered with 1·3-cm layer of dry plaster of Paris or sawdust and 2–3 filter paper discs. Finally a wet mixture of plaster of Paris is added to the tube. A roll of paper, which can afterwards be withdrawn, can be inserted into the tube while adding the wet plaster of Paris to keep the sides clean. A small plastic conical funnel fixed into the mouth of the tube helps to reduce the evaporation of the killing agent and prevent mosquitoes from flying or falling out (King *et al.*,

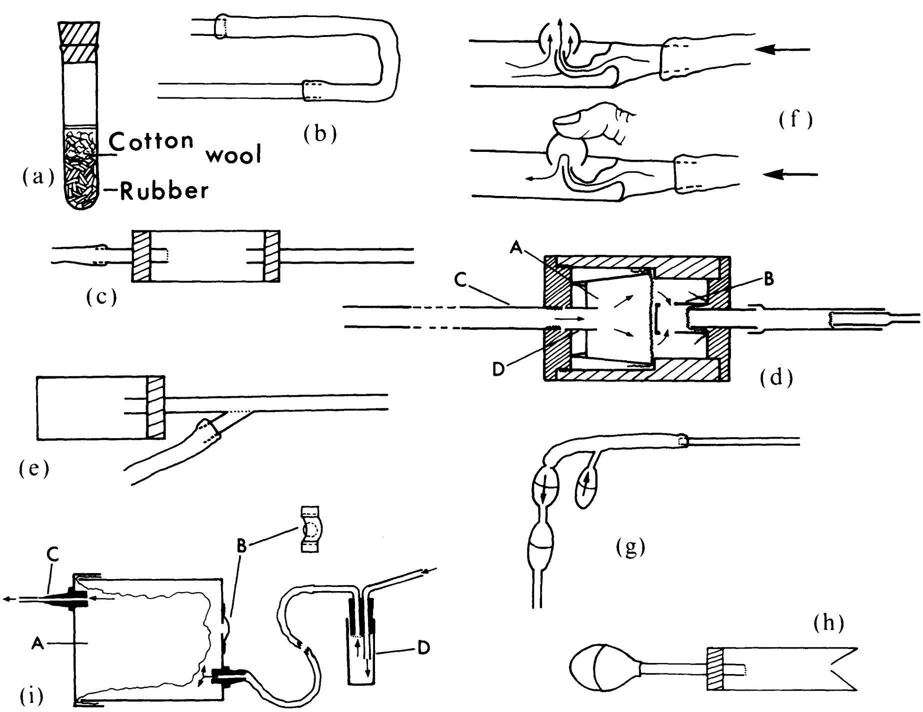


*FIG 3.1. Aspirators: (a) Collecting tube with rubber pieces soaked in chloroform; (b) sucking tube; (c) aspirator with reservoir; (d) aspirator with A — removable cardboard cup showing B — plastic tubing with holes near its end, C — plastic collecting tube, D — tubing glued to bottom of the cup (after Coluzzi & Petrarca, 1973); (e) aspirator after Barnes & Southwick (1967); (f) aspirator incorporating 'venturi' valve principle; (g) aspirator of Saliternik (1963a); (h) aspirator type of Russell et al. (1963); (i) aspirator after Wiens & Burgess (1972) showing A — plastic bag, B — cardboard carton having a one-way flap valve, C — mouthpiece, D — reservoir-type pooter.*

1939). A tube with cyanide lasts much longer than one with chloroform but is not rechargeable and is dangerous if broken. Furthermore, there may be difficulties in safely disposing it when it is no longer required. Burton (1954) made a killing tube from 'Lusteroid plastic' test tubes, 6 × 1¼ in. Killing agents such as carbon tetrachloride or chloroform were soaked up by either cut-up rubber bands placed at the bottom and held in place by a wedge of cellulose sponge or directly by the sponge. Chloroform, however, may gradually damage plastic test tubes and ether must be avoided as it dissolves both the sponge and test tube.

Mosquitoes can be collected directly into killing tubes or blown into them from aspirators. Practice may be needed in positioning a tube over a mosquito because toxic vapours escaping from it may cause the mosquito to fly away before it can be caught.

## Oral aspirators

Mosquitoes found resting on various surfaces are frequently collected with an aspirator, sometimes referred to as a 'pooter'. Such collections are commonly referred to as 'hand-catches'. The simplest and most widely used type of aspirator is made from a 30–45-cm length of 8–12-mm internal diameter plastic or glass tubing. A piece of mosquito netting is taped either directly over one end, or over a short piece of smaller diameter tubing which is inserted into the end of the larger tubing. A 50-cm length of rubber or pliable plastic tubing is slipped (often with the help of vaseline) over the end of the glass tubing (Fig. 3.1*b*), and a small section of plastic or glass tubing is inserted into the opposite end to form a mouthpiece. Several mosquitoes can be sucked up into the aspirator before they are gently blown into a suitable container. Blood-engorged adults must be sucked up gently otherwise they may smash on the barrier netting at the end of the tube. Unless the operator keeps sucking or closes the end of the tube with a finger or piece of cotton wool, captured mosquitoes are liable to fly out. To reduce the likelihood of their escaping and to enable larger numbers to be collected before they are discharged, aspirators with reservoirs are used (Fig. 3.1*c*). In using these the collecting tube has to be removed before the adults can be blown out. Ryan (1989) suggested a small modification to reservoir-type pooters to allow insects to be stunned or killed. He placed together citric acid crystals and sodium hydrogen bicarbonate at the bottom of the pooter and covered them with a thin plug of cellulose wadding. Insects were sucked up in the normal manner and then a few drops of water were carefully introduced down the glass entry tube of the pooter, this caused the production of carbon dioxide. When stunned, the insects were either tipped out and sorted, or the reservoir removed from the pooter and capped with a cork, and replaced with another tube containing citric acid and bicarbonate.

Examples of reservoir-type aspirators are illustrated in Figs 3.1*c–e,h* and Figs 3.2*b,c*.

Various sized plastic or paper cups covered with mosquito netting are widely used as inexpensive and expendible small holding containers for mosquitoes. Coluzzi & Petrarca (1973) developed an aspirator in which mosquitoes are collected directly into such cups (Fig. 3.1*d*). A plexiglas cylindrical chamber in which the cup (A) is housed forms the body of the aspirator. A short length of plastic tubing with a ring of five to six small holes near its end (B) is glued over a small central hole in the permanently fixed plastic base of the cylinder. A smaller diameter piece of tubing is inserted through this hole and is attached to a length of rubber tubing which forms the mouth-piece. A piece of fine barrier netting is inserted between the tubing with holes and the smaller diameter section. A plastic collecting tube (C) is threaded into the top of the cylinder which when screwed into position retains the paper cup within the cylinder. A short section of plastic tubing (D) glued to the inside of the top projects into a hole made in the bottom of the paper cup. By having the collecting tube threaded into the aspirator different shapes and lengths of tubing can be used, while the function of the five to six small holes in the tubing in contact with the netting of the cup is to ensure a more even aspiration, thus minimising damage to the

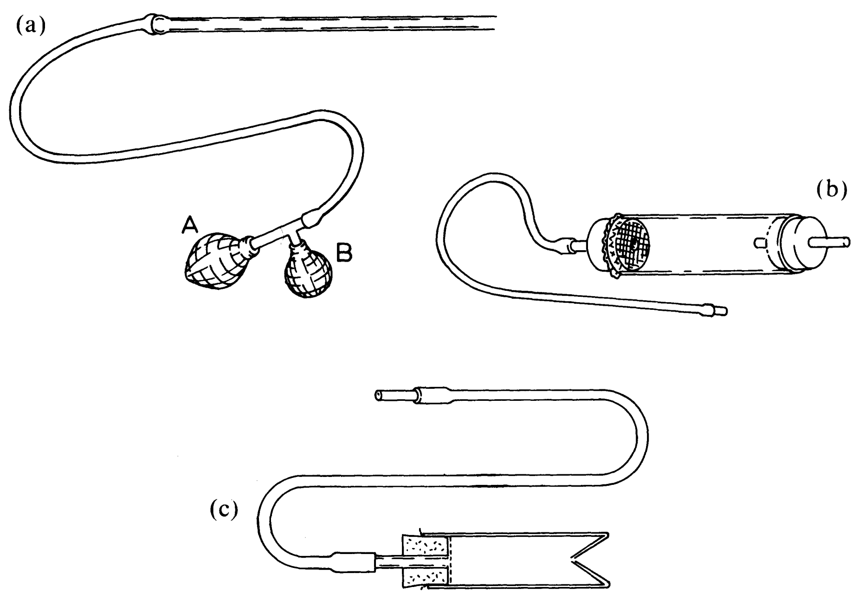


*FIG. 3.2 (a) Aspirator with aspirating bulb A — with valve, B — blowing bulb; (b) and (c) reservoir-type aspirators (after World Health Organization, 1975).*

mosquitoes. After mosquitoes have been sucked into the paper cup, the top of the plexiglas cylinder is unscrewed and the paper cup removed after the hole in its bottom has been plugged with cotton wool.

Cartridge-type aspirators were also described by Barnes & Southwick (1967). Their second model, which is the simplest, consists of a plastic collecting tube with a small side arm near the base, the entrance of which is covered with nylon netting. Insects sucked into the collecting tube pass into a removable plastic cartridge fitted at the end (Fig. 3.1*e*). Different sized and shaped cartridges can be used. Sholdt & Neri (1974) described how an aquarium dip tube of the type used to remove organic debris can be easily modified to form a mouth aspirator incorporating a holding cage.

The practice of collecting mosquitoes by sucking them up into aspirators is widespread, but over a period of time it can become tiring, and moreover the prolonged inhalation of mosquito scales, dust and fine debris may cause, or aggravate, bronchial allergies. A case is known of severe histoplasmosis in an entomologist who habitually sucked up *Phlebotomus* from caves with deep layers of bat excreta. A cotton wool plug can be inserted into mouth aspirators to minimise this hazard, but this can reduce suction power. Insertion of a wad of very fine plastic mesh as used in oxygen and anaesthetic lines in hospitals to deliver clean air to patients does not reduce suction to any appreciable extent. To collect phlebotomine sandflies Warburg (1989) modified a standard disposable bottle-top filler as used to sterilise laboratory cultures by removing the Millipore filter and inserting a double-layered paper tissue, so when the filter was placed into an oral aspirator it prevented dust being sucked up into the mouth. At the Liverpool School of Tropical Medicine we have made the much simpler modification of inserting a 'filter' (ThermoVent 1200, Portex Co.), made to fit into tubes delivering anaesthetic or air to patients in hospitals, directly into the rubber tubing of the simple aspirator shown in Fig. 3.1*b*. There is no appreciable reduction in suction, and the end of the filter forms a suitable mouth piece. Several more ingenious methods have been developed to overcome the problem of inhaling dust and dirt. For example, if a small glass valve is inserted towards the end of the glass tubing of an aspirator, inhalation of harmful particles is prevented because mosquitoes are caught by blowing, not sucking (Fig. 3.1*f*). The catch is discharged by placing a finger over the valve opening and again blowing. As long ago as 1939 Woodbury & Barnhart described an aspirator using the venturi principle to avoid the collection of insects by sucking. Spielman (1964) described and illustrated an aspirator operating from a source of compressed air which employed a venturi made from glass tubing.

In Israel collectors who habitually aspirated adults of *Anopheles sergentii* from limestone caves suffered from the inhalation of fine dust particles. Their aspirators were consequently modified to eliminate the need for oral sucking by connecting large rubber bulbs to the aspirator's long length of rubber tubing (Fig. 3.1*g*). When one of the upper bulbs was squeezed mosquitoes were sucked up, whereas pressure on the other bulb discharged them. Both bulbs were operated in one hand, the other held the glass part of the aspirator (Saliternik, 1963*a*). An alternative, but similar modification is shown in Fig. 3.2*a* where both the aspirator (A) and blowing (B) bulbs have valves. Another very simple aspirator that avoids sucking by mouth consists of a thick glass-walled, or plastic, cylinder 15 cm long and about 3 cm in diameter (Fig. 3.1*h*). An inverted plastic cone is inserted into one end and the opposite end is closed with a cork or rubber bung. A piece of narrow glass tubing which has its end covered with mosquito netting is connected to a large rubber suction bulb and is inserted through the cork or bung. Mosquitoes are sucked up into the cylinder by squeezing the bulb and are prevented from escaping by the cone at the opposite end. When sufficient mosquitoes have been caught the cylinder is removed, corked and replaced with another (Russell *et al.*, 1963).

Another type of aspirator (Fig. 3.1*i*) that avoids the inhalation of dust particles was developed for the collection of agricultural insects (Wiens & Burgess, 1972). A thin plastic bag (A) is positioned in a 3-pint capacity cardboard carton with its open end folded over the rim, the lid is then replaced and the joint between the lid and carton sealed with silicone rubber or adhesive tape. A one-way flap valve (B) is made in the bottom of the carton by cutting out a 1-cm hole and loosely taping a small piece of polythene plastic over it. A suitable mouthpiece (C) is inserted through the lid and a length of rubber tubing used to connect the bottom of the carton to a reservoir-type pooter (D). In operation air is first blown into the plastic bag to inflate it and this results in expelling air from the carton through the flap valve. Air is then sucked out of the bag which on deflation draws air, and insects, into the pooter.

Sometimes the end of the glass tubing of an aspirator is bent at an angle to facilitate collecting from awkward corners and surfaces.

A detailed description is given by Woke (1955) of an aspirator that can also serve as a holding cage for small delicate arthropods.

### Small battery operated aspirators

Small battery operated hand vacuum cleaners which are sold for removing dust from clothing, cars and house furniture etc. can be converted into aspirators (Dell'Uomo, 1967; Dyce *et al.*, 1972; Husbands, 1958; Husbands & Holten, 1967; Jackson & Grothaus, 1971; McCreadie *et al.*, 1984; Meek *et al.*, 1985; Nelson & Chamberlain, 1955; Saliternik, 1963*b*; Spencer, 1962; Sudia & Chamberlain, 1967; Trpis, 1968). Suction is usually produced by high speed rotation of a plastic or metal fan, usually having two or more blades, which is driven by a 3- or 6-V d.c. motor powered by dry-cell torch batteries, which are often of the rechargeable type. Those vacuum cleaners having a circle of bristles forming a brush around the air intake of the cleaner usually have these removed before having a glass or plastic tube or cone fixed over the intake. A useful version developed at the London School of Hygiene and Tropical Medicine is shown in Fig. 3.3*a*. After removal of the bristles a small plastic funnel is cut to the correct size and glued in position. Its short spout, which is covered with a piece of mosquito netting, projects into the glass or plastic collecting tube. The small dust bag originally present is replaced with a length of rubber tubing so that with the batteries switched off the catch of mosquitoes can be blown into a suitable container. An alternative method consists of glueing or screwing a 12-cm long, 4-cm diameter clear plastic cylinder (e.g. a plastic holding tube supplied with the WHO mosquito susceptibility kit) over the air intake to form a collecting reservoir. An aspirator of this design is shown in Fig. 3.3*d*. Because of its large size, mosquitoes may get damaged by being repeatedly blown around inside the reservoir. This can be reduced if three approximately equally spaced horizontal stiff cardboard baffles are inserted into the collecting reservoir to reduce air turbulence. Carver (1967) made an aspirator from a battery operated vacuum cleaner by clipping the bristles of the brush and fixing mosquito netting over the inlet with plastic bands. A plastic funnel about 7 cm in diameter was taped over the inlet with adhesive tape. Mosquitoes sucked into the funnel through its spout were removed by blowing through the outlet hole.

Another modification was described by Husbands & Holten (1967). They removed the brush assembly of a battery vacuum cleaner and fixed a plastic plate over the air intake. A short stub of plastic tubing (28 mm internal diameter) with fine netting over the free end is cemented in the middle of the plate (Fig. 3.3*b*). A 20-cm extension tube of glass or rigid plastic is inserted over the short plastic stub. A number of small collecting tubes, that also serve as holding tubes, are made by cutting the end off a conical centrifuge tube and covering it with mosquito netting. One of these tubes is inserted into the end of the extension tube. After one or more mosquitoes have been collected the end of the centrifuge tube is closed with cotton wool and the tube removed and another inserted. For collecting in inaccessible places the extension tube is made of rubber or flexible plastic,

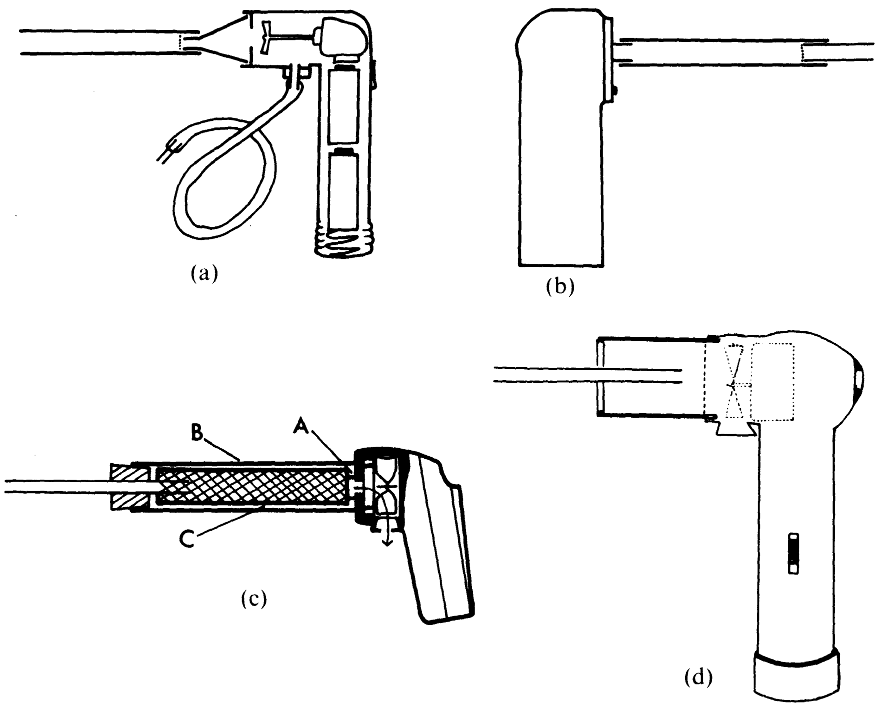


*FIG. 3.3. Battery operated aspirators using modified small hand vacuum cleaners: (a) Type developed by London School of Hygiene & Tropical Medicine; (b) after Husbands & Holten (1967); (c) after Trpis (1968) showing A — spacers, B — acrylic outer casing, C — holding tube; (d) battery-powered aspirator of the World Health Organization (1975).*

but two hands are then required to hold the aspirator. A larger type of portable car vacuum cleaner that operates from two 6-V dry-cell batteries was modified by Harden *et al.* (1970) for collecting mosquitoes at bait catches. A length of 1-in diameter plastic tubing with mosquito netting at one end is screwed into the suction hose of the cleaner; the end is closed periodically with a cork and the tube removed and replaced with another.

Trpis (1968) described a useful but more complicated aspirator (Fig. 3.3*c*). After removing the brush from a small portable vacuum cleaner a circular plastic base plate with five 10-mm diameter holes is permanently fixed over the air intake. Two 3-mm thick and 3-mm$^2$ spacers (A) are glued on to the face of the base plate. A rectangular acrylic plastic outer casing, 180 × 40 × 37 mm (B), is cemented on to the base plate. The opposite end is fitted with a rubber bung through which a length of 9-mm diameter glass tubing is inserted. A number of rectangular holding cages, 155 × 30 × 25 mm, are made with the two ends and upper and lower sections of 3-mm acrylic plastic but with the two sides of plastic mosquito screening (C). A 9-mm diameter hole is cut from one end of each holding tube. The tube is then inserted into the larger permanently fixed plastic

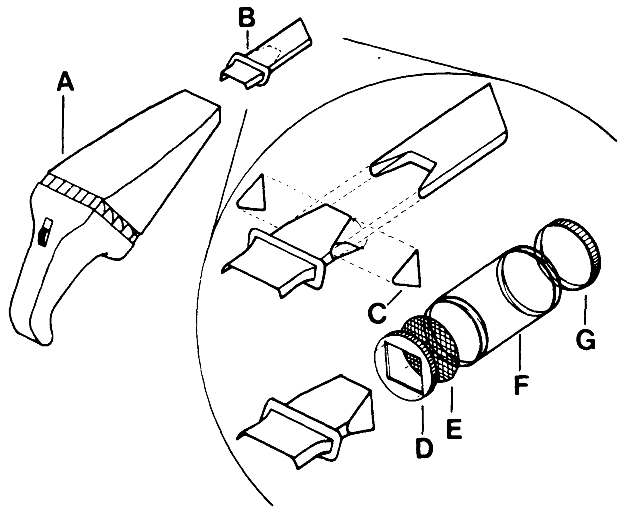


*FIG. 3.4. Black and Decker 'Dustbuster Plus' cordless vacuum cleaner modified for aspirating mosquitoes: A — vacuum cleaner, B — crevice tool, C — triangular piece of plexiglas, D — screw cap, E — 50-mesh plastic screen, F — transparent plastic vial, and G — screw cap (from Meek* et al.*, 1985).*

casing to rest on the two spacers on the base plate. When sufficient numbers of mosquitoes have been sucked up into the holding tube it is removed, corked and replaced with another.

McCreadie *et al.* (1984) modified a commercial Black and Decker 'Dustbuster' hand vacuum cleaner, by removing the filter from the distal end of its detachable nozzle and internal flap near the apex, to convert it into an aspirator. A rectangular cloth collecting bag (8 × 15 cm) is inserted into the apex of the nozzle, and two flaps sewn into the mouth of the bag are folded back on to the outside of the nozzle and held in place by elastic bands. After use the cloth bag is removed and its opening sealed with a spring clip.

Meek *et al.* (1985) described two very useful battery operated aspirators made from adapting Black and Decker power tools. Unfortunately the first model is no longer made by Black and Decker and so is not described here. The other model consists of a 'Dustbuster Plus' cordless vacuum cleaner (Fig. 3.4) powered by rechargeable batteries and has a large capacity 6-V motor thus providing greater suction than the regular Dustbuster model with its 3·6-V motor, as already described by McCreadie *et al.* (1984). This improved vacuum cleaner is supplied with a charger base equipped with a 120-V electrical charger, and several cleaning attachments including a so-called crevice tool. The crevice tool (B) is cut approximately 8·9 cm from its flanged end and two 0·32-cm thick triangular-shaped plexiglas pieces (C) are stuck on either side with hot glue as shown in Fig. 3.4. Each triangular section is about 2·79 cm at the base and 3·81 cm high. The screw cap (D) from a 20-dram transparent plastic prescription (F) vial has a 1·91 × 2·54-cm piece cut out to enable the end of the modified crevice tool to fit into it. Hot glue is applied to make a permanent bond. The ends of two such vials are removed and the non-threaded ends joined together with epoxy cement to form a 12·1-cm collecting tube (F). A 4·45-cm circular piece of plastic screening (E) is attached over one of the threaded ends with cement. This screened end of the tube can now be screwed into the vial cap attached to the adapted crevice attachment of the vacuum cleaner. When sufficient mosquitoes have been caught the open end of the tube is closed, and the tube replaced with another. An alternative type of collecting tube can be made from the transparent cylinders and screw caps comprising the exposure tubes of the WHO adult mosquito insecticide susceptibility kits.

Shroyer (1989) described a mechanical aspirator using a rotary vane pressure-suction pump for use in the laboratory to transfer virus-infected mosquitoes.

McGavin & Furlong (1981) designed a cheap electronic counter that could be attached to an aspirator to count insects. In the field it operates from rechargeable batteries providing about 10·8 V, in the laboratory mains output of 8–12 V d.c. at approximately 300 mA can be used. In tests it proved reliable for counting objects sucked up, ranging in size from wheat grains to *Calliphora* puparia. Another solid-state electronic insect counter was described by Pearson *et al.* (1975) but it is only useful in the laboratory, because it is not a portable aspirator. Bennett (1980) used this electronic counter to count *Aedes sierrensis* adults for mark–release experiments.

If vehicles can be parked near the collecting site battery powered aspirators that operate from 12 V can be plugged directly into the standard cigarette lighter socket located on the instrument panel of some cars, or into the sockets for inspection lamps (Harden *et al.*, 1970).

### Large mechanical aspirators

I have omitted references to larger mechanical aspirators described in the first edition of the book because most were cumbersome and operated from full-sized vacuum cleaners or from portable generators, and are now very rarely used to collect mosquitoes. They have been superceded by more efficient and smaller motorised insect aspirators which are described on pp. 249–60.

### Resting sites of mosquitoes in houses

The walls, ceilings, roof, clothing, furniture including the underside of beds, and other objects in huts should be examined with a torch to locate resting mosquitoes. In some areas where houses are raised on stilts mosquitoes may also rest underneath them (Spencer, 1965). In Tanzania Smith (1955) found that in both round and rectangular village huts about 62–66% of the females and 60–70% of the males of *Anopheles gambiae* rested below the thatched roof, mainly on the mud walls, but also on various objects and under the beds. Similarly, in later studies Smith (1962*a,b*) found that about 56–75% of *Anopheles gambiae* rested on hut walls and household objects. In The Gambia although Bryan (1979) caught some adults of the *Anopheles gambiae* complex from walls and ceilings of village houses, she caught most mosquitoes inside mosquito nets which are widely used in that country.

In Java most *Anopheles aconitus* found indoors were collected resting on walls just above the floor especially in crevices and in dark sheltered corners, very few rested higher up the walls or on the ceilings (Joshi *et al.*, 1977). Adults were also collected from hay and straw used as fodder and stored in animal sheds. In contrast in India Batra *et al.* (1979) had to use ladders in houses and other buildings to collect *Anopheles stephensi*, because they tended to rest high up on the walls and ceilings beyond the reach of the collectors. In Pakistan Reisen *et al.* (1979) using a mechanical aspirator (Davis & Gould, 1973) collected *Anopheles subpictus* resting on the floor and lower walls of animal sheds and houses. When these collections were finished two collectors caught any remaining adults with mouth aspirators for 15 min.

In Java Damar *et al.* (1981) in studying nocturnal resting height preferences collected, each hour, anopheline mosquitoes resting in cattle sheds in which the walls had been marked off at heights of 75, 250, 225 and 300 cm.

The local distribution of adults resting in huts may be affected by the type of hut construction, the building materials used, the availability of alternative resting places, such as clothing and furniture, and the presence of fires. There may also be differences between the resting pattern during the day and night, and also seasonal variations. Smith *et al.* (1966) found that in huts with grass roofs about 80% of *Anopheles gambiae* rested on the roofs during the day and night, but in huts with corrugated iron roofs, although 80% rested on them at night less than 10% rested on the roofs during the day, when temperatures were higher. In Japan Ogata *et al.* (1968) studied the hourly fluctuations throughout the night of the numbers of *Culex tritaeniorhynchus* resting on the walls of pigsties. In the D'Entrecastleaux Islands, Spencer (1965) found that about 60% of *Anopheles farauti* rested on the hut walls below the level of 3 ft.

To study the resting sites of *Anopheles darlingi* in houses in Brazil Roberts *et al.* (1987) caught blood-fed mosquitoes in the peridomicilliary environment during the early evening and marked them with USR fluorescent pigment number 1953. A hundred marked females were released inside a house at 2200 hr, and periodic searches with a 'Black-Ray, ULV 56' long-wave ultraviolet lamp showed their preferred resting places. Other times unfed females collected in entrance traps between 1800–2000 hr were marked and released inside houses at 2040 hr. Less than 50% of marked females were caught after 1 hr of release, and only 20% could be found in a house 7–8 hr post release.

In Kenya indoor resting densities of *Anopheles gambiae* complex and *Anopheles funestus* were based partly on weekly collections of adults resting on a 5 × 6 ft reed mat placed in houses to form a ceiling (Sexton *et al.*, 1990). In India Yasuno *et al.* (1973*b*, 1977) placed plywood boxes (50 × 30 × 28 cm) having one-half of the lid hinged and left overnight partially open—for entry of mosquitoes—in dark corners of bedrooms in 20 houses. The boxes were lined with black cloth and contained a sponge saturated with water to increase humidity. On one occasion as many as 325 *Culex quinquefasciatus* were collected overnight from a single trap, while the mean catch reached a peak of 40·7 in April. These traps were effective only when mosquito densities were large and humidities low. In Trinidad over about 16 months Nathan (1981) collected 1720 female *Culex quinquefasciatus* and substantial numbers of *Aedes aegypti* from resting shelters placed inside houses. These shelters comprised open-ended 30-cm square plywood boxes painted white outside and black inside and provided with a screened jar of water to improve their attractiveness. Basically they were similar to the boxes used by Yasuno *et al.* (1973*a*).

The distribution of mosquitoes resting in houses has been investigated by several other entomologists (Haddow, 1942; Joshi *et al.*, 1977; Pal *et al.*, 1960; Roberts *et al.*, 1987; Smith, 1964; Wattal & Kalra, 1960 etc.).

### Collecting efficiency

The numbers of *Anopheles gambiae* entering village houses was shown in The Gambia to be about 43% fewer in those with closed eaves, that is houses with no or very little space between the tops of the wall and the roof (Lindsay & Snow, 1988). Clearly these types of houses should be avoided if large collections of mosquitoes are required. The numbers of mosquitoes caught from huts are usually expressed as the mean number per hut, that is hut density. In India the number of *Anopheles culicifacies* caught resting in houses were expressed as the mean hour density (MHD)

$$\text{MHD} = \frac{n \times 60}{t \times p}$$

where $n$ = numbers caught, $t$ = collection time in minutes, and $p$ = number of collections (Subbarao *et al.*, 1988).

Mosquitoes caught from under over-hanging eaves of huts, from underneath huts or from their outside walls are sometimes included in the calculation of hut densities although they are not resting in the huts. Hut densities will nearly always represent an underestimate of the numbers of mosquitoes in a hut because of the inability to catch the entire resting population. Aspirator collections will recover a smaller proportion of the mosquitoes resting in huts than pyrethrum spray catches, but are essential if live individuals are required.

In a series of 30-min catches from huts in two unsprayed villages in India, Viswanathan *et al.* (1950) found that the mean female density of *Anopheles culicifacies, Anopheles fluviatilis* and *Anopheles stephensi* calculated from morning catches (0700–1100 hr) was about 30% greater than afternoon (1100–1500 hr) catches. The relationship between the two densities was shown to be:

$$\text{Mean afternoon density} = 0{\cdot}6455 \times \text{mean morning density} + 1{\cdot}3$$

In two villages that were sprayed with DDT the relationship was:

$$\text{Mean afternoon density} = 0{\cdot}488 \times \text{mean morning density} + 1{\cdot}27$$

Because of their greater activity it was more difficult to collect adults at night (2000–2100 hr), but despite this about twice as many were caught in both unsprayed and DDT sprayed huts than in morning collections (0700–1100 hr). In HCH sprayed huts, however, only about 40% more were caught during the night than in the morning.

The efficiency of hand-catching was assessed from 620 hut collections in six Indian villages (Viswanathan *et al.*, 1952). Aspirator collections were made in each hut with the doors and windows shut and all openings and cracks closed with paper and cloth. After completion of these collections white sheets were placed on the floor and the hut space sprayed with 0·2% pyrethrum in kerosene. Since all openings had been carefully closed it was considered that the pyrethrum collections recovered all the mosquitoes resting in the huts. While this might have been true, it is more probable that a few mosquitoes failed to fall clear of objects and drop on to the floor sheets, and that a few of those that did were not recovered. This is, however, unlikely to be a serious source of error. A total of 7785 anophelines and 4940 culicines were caught in these collections, and about 32% of the *Anopheles culicifacies* and 26% of *Anopheles subpictus* actually present in the huts were caught in the hand-catches. Of the 10 other *Anopheles* species and culicines caught from the huts 28% and 31%, respectively, of the total numbers resting in the huts were collected in the hand-catches. There was no attempt to collect all the adults resting in the huts and collections were terminated after 30 min irrespective of whether more mosquitoes could be caught. If collecting for a standard time (e.g. 30 min) catches a known proportion of the mosquitoes in huts, then so long as the collector's efficiency does not change over a sampling programme the actual numbers present can be estimated. The proportion of the total population caught by hand-catches from different types of huts may vary, and this should be accounted for in calculating mean hut densities. Viswanathan *et al.* (1952) concluded than when aspirator collections were made for 30 min and the results expressed as numbers caught per man-hour this gave a useful index for comparing mosquito densities between different catching stations.

Batra *et al.* (1979) found that aspirator catches collected 31·3% of the males and 28·9% of the females of *Anopheles stephensi* resting in houses, whereas with *Anopheles subpictus* only 8·8% of the males and 7·1% of the females were caught; the remainder being collected by pyrethrum spray catches.

In West Africa Ribbands (1946*a*) found that even when attempts were made to catch with aspirators all the *Anopheles* resting in a hut, pyrethrum catches still yielded about a further 28%. In India Senior White & Rao (1946) also found that despite attempts to catch with aspirators all anophelines resting in huts about 28% remained uncaught, and in Morocco pyrethrum collections in houses following aspirator collections showed that only about 47% of the *Anopheles labranchiae* were caught with aspirators (Bailly-Choumara, 1973). In Jamaica as little as about 10% of the indoor resting population of *Anopheles albimanus* were caught in hand collections (Muirhead-Thomson & Mercier, 1952). Ribbands (1946*a*) reported that the proportion of the total catch obtained in aspirator collections varied greatly (50–90%) on different days. Because male and unfed females are generally more difficult to locate, he concluded that they were more likely to be underestimated in aspirator collections than blood-fed and gravid individuals. Unfortunately he caught only 494 mosquitoes in his experiments, and consequently his results are not so meaningful as those of Viswanathan *et al.* (1952) who found no evidence that males were underestimated more than females.

Undoubtedly a greater proportion of the absolute numbers of mosquitoes are caught from huts if aspirator collections are continued until no further adults can be found (i.e. catching to completion), but in practice the method is too time-consuming for routine catches. It may take 2–3 hr for all the mosquitoes to be collected from a single hut (Rao, in Viswanathan *et al.*, 1952). Catching to completion is also probably subject to greater individual bias than collecting for a specific time, because of the wide variations in the abilities and conscientiousness of the collectors. The ease with which mosquitoes are found also varies between huts depending on the availability of different types of resting sites.

In Ethiopia Krafsur (1977) treated the successive monthly proportions of unfed, partly fed, blood-fed and all other gonotrophic categories of *Anopheles* caught resting in houses each in turn as a set of independent random variables having a binomial distribution. Then, the probabilities that monthly proportions in the different gonotrophic stages were homogeneous were estimated from the chi-squared statistic

$$\chi^2 = \Sigma \; \frac{(x_i - n_i\theta)^2}{n_i\theta\,(1 - \theta)}$$

where $n_i\theta$ = expected proportion of any gonotrophic condition, $n_i$ = sample size, $\theta$ = population mean, and $x_i$ = observed sample mean. He found considerable heterogeneity, which was caused in part by the proportional increase in unfed and partly fed females when the mosquito population was growing, and a decrease when the population was declining (See Krafsur (1977) for further details of his methods of calculation.)

Nagasawa (1976) using three years' data on the collection of *Culex quinquefasciatus* resting in houses (2000–2100 hr) and from human bait catches (2100–2400 hr) in the same seven catching stations in Myanmar firstly transformed the data into logs and calculated the mean numbers caught/man-hour. Then by harmonic analysis, following the method of Bliss (1970), two-term Fourier curves were plotted to show seasonal patterns of abundance obtained by the two methods, and estimates were derived of maximum and minimum numbers of mosquitoes caught by the two sampling methods.

It is not only often difficult to find outdoor resting populations of *Anopheles*, but generally even more problematic to estimate the proportion of the population that rests out of doors. In Nigeria Molineaux & Gramiccia (1980) estimated the proportion of *Anopheles gambiae* complex that fed on man indoors and then rested indoors as follows:

Let $IRD$ = the true indoor resting density
$I\hat{R}D$ = the indoor resting density estimated by pyrethrum spray-sheet collections
$I\hat{R}D = b_1 \times IRD$
$b_1 = I\hat{R}D/IRD$ = the bias of the estimated indoor resting density
$MBR$ = the true man-biting rate
$M\hat{B}R$ = the man-biting rate estimated by night-biting collections
$M\hat{B}R = b_2 \times MBR$

$b_2$ = $\hat{MBR}/MBR$ = the bias of the estimated man-biting rate
$HBI$ = the proportion of blood-meals in the pyrethrum spray-sheet collection positive for man (human blood index)
$N$ = the number of persons per hut (the population of the village, divided by the number of huts)
$x$ = the proportion of blood-meals followed by resting indoors (at least until the next morning when pyrethrum spray-sheet collections are made).
$T$ = period in days of resting indoors after feeding.

Then:

$$IRD \times HBI = MBR \times N \times x \times T$$

or

$$\frac{I\hat{R}D}{b_1} \times HBI = \frac{M\hat{B}R}{b_2} \times N \times x \times T \qquad (1)$$

$I\hat{R}D$, $M\hat{B}R$, $HBI$, $N$ are measured directly. $T$ is estimated by

$$T = 1 + \frac{G}{F},$$

where $G$ and $F$ are the proportions of gravid and blood-fed individuals, respectively, caught in the pyrethrum spray-sheet collection (if the maturation time is 2 days, as is suggested by the clear-cut bimodal distribution of the pyrethrum spray-sheet collection by abdominal appearance);

$x$ and $b_2/b_1$ (the relative bias of the two sampling methods) are unknown.

If we know the one, we can compute the other from eqn (1):

$$x = \frac{I\hat{R}D}{M\hat{B}R} \times \frac{b_2}{b_1} \times \frac{HBI}{N \times T} \qquad (2)$$

$$\frac{b_2}{b_1} = \frac{M\hat{B}R}{I\hat{R}D} \times x \times \frac{N \times T}{HBI} \qquad (3)$$

The values, $M\hat{B}R$, $I\hat{R}D$, $N$, $T$ and $HBI$ were readily obtained by Molineaux & Gramaccia (1980) and inserted in eqns (2) and (3). With *Anopheles funestus* past experience shows that nearly all blood-fed individuals rest indoors, thus $x$ can be given the value of 1, thus enabling the value of $b_2/b_1$ to be computed from formula 3. It is found to be 1·16, that is in comparison with the indoor-resting density, the man-biting rate is overestimated by 16%. Now, assuming that the relative bias of the two sampling methods is the same for both *Anopheles funestus* and *Anopheles gambiae* s.l., then the value of $x$ for *Anopheles gambiae* s.l. can be derived from formula 2, and is found to be 0·47. That means that only about half the blood-meals taken on man by *Anopheles gambiae* are followed by resting indoors.

Lines *et al.* (1986) studied the mixing of indoor and outdoor-resting adults of the *Anopheles gambiae* complex and *Anopheles funestus* in Tanzania by marking them with different coloured fluorescent powders. Out of 568 female *Anopheles gambiae* caught outside in pit shelters which were marked and released just 31 were recaptured, all from inside houses. An estimate of the proportion of outdoor-resting females which rest exclusively out of doors is 0/31. The 95% upper confidence limit ($f$), can be calculated from the binomial distribution by

$$(1 - f)^{31} = 0{\cdot}05 \quad \text{i.e.}\ f = 0{\cdot}092$$

Now, if the outdoor resting sample had contained 9·2% or more exclusively outdoor-resters, at least one should have appeared in the sample of 31 in 95% of the observations. But this estimate is based on unbiased sampling with respect to the endophilic and exophilic populations and fails to take into account various biological variables. A full analysis of the recapture data taking into account the probabilities that the outdoor resting collections consist of endophilic females that had 'strayed outside' together with a few exclusively exophilic females, and other mixes of the indoor and outdoor collections, is presented by Lines *et al.* (1986).

## Hand-net catches

Occasionally small hand-nets, about 15 cm in diameter, made of fine mosquito netting have been used to catch adult mosquitoes resting in human and animal habitations. In Taiwan for example Rosen *et al.* (1989) used a net once a week from July 1980 to December 1983 for 1 hr, or slightly longer, after sunset and collected 142 434 female *Culex tritaeniorhynchus* resting in a single shed used to rear pigs. Obviously very large numbers of mosquitoes must have been resting in this shed. In the Dominican Republic Tidwell *et al.* (1990) found that fluctuations in adult population levels of both *Aedes aegypti* and *Culex quinquefasciatus* could be monitored by two men using 12-in hand-nets to catch mosquitoes in houses for 5-min periods. Particular attention was paid to searching underneath beds, chairs and tables as well as in cupboards. They found the mean number of female *Aedes aegypti* per house ranged from 1·22–15·04, with several houses having more than 20 females, and one as many as 134. The average numbers of female and male *Culex quinquefasciatus* collected were 3·2 ± 4·8 and 3·0 ± 4·3, respectively; the highest catches were 68 females and 39 males.

In determining the seasonal abundance of *Aedes aegypti* in Indonesian huts, adults were caught from some huts by 10-min aspirator collections and from others by sweep-netting for 5 min, but no details of the latter technique were given (van Peenen *et al.*, 1972). More *Aedes aegypti* were caught with aspirators than by sweep-netting, but no males were collected, whereas substantial numbers of males were caught with the sweep-nets.

In India Rajagopalan *et al.* (1977) were unable to use the 'hand–catch index' to sample indoor resting *Culex quinquefasciatus* in urban houses because their densities were too high, mosquitoes were therefore caught in 10-min collections from houses using an 8-in sweep-net (Yasuno *et al.*, 1973*b*). The mean numbers caught per house by this method varied from 56·4 (June) to 354·3 (October) during 1972–1973, and from 11·5 (January) to 117·9 (October) in 1973–1974. They also used a removal method to estimate the total population of *Culex quinquefasciatus* (see pp. 232–3).

Sometimes net-catches are preceded by gently spraying houses with non-toxic oils (e.g. Risella or citronella oil) paying particular attention to cracks and crevices. This causes adults that have escaped capture to take flight, and these are caught in the net.

### Pyrethrum spray sheet collections

Pyrethrum spraying of habitations was originally used as a control measure (Covell *et al.*, 1938; De Meillon, 1936; Eddey, 1944; Russell & Knipe, 1939, 1940, 1941), but the practice was discontinued in the 1940s, being replaced by spraying huts with DDT and other residual insecticides. Knock-down space spraying with pyrethrum is now used as a standard, quick and easy method of catching mosquitoes resting in huts and animal shelters. It is usually the most efficient of the available methods for collecting mosquitoes, but its efficiency depends on the type of hut construction.

The routine procedure is as follows. All occupants, animals, easily removable objects such as small tables and chairs, exposed food and drinking water are firstly removed from a hut that is to be sprayed. When water is stored in pots imbedded in the floor and cannot be removed, the pot opening should be covered with a lid. Two people carefully lay white sheets made of calico or some other strong fabric over the entire floor, over the bed, and over furniture and miscellaneous objects that have not been removed. Fires made in hut floors can present problems because a small area of floor space has to remain uncovered. All doors and windows are closed and the hut space-sprayed by one or two people, depending on its size, with 0·1–0·2% pyrethrum in kerosene (paraffin) usually synergised with piperonyl butoxide. In Egypt Kenawy *et al.* (1990) used 0·2% neopybuthrin to spray houses. In Tanzania Bushrod (1979) added 0·3% chloroform to her 0·3% pyrethrum spray as she considered this prevented mosquitoes that were knocked down from recovering. Certain proprietary brands of pyrethrum concentrate, such as Pycon 819E contain, in addition to piperonyl butoxide, emulsifiers enabling the concentrate to be mixed with water instead of kerosene. For this the diluent (water) should be reduced by 20% from that needed with kerosene. Strong spray-guns of medium capacity and preferably with a long plunger stroke are best. Each sprayer should have a suitable pin so that when, as invariably happens, a pump becomes blocked, it can be cleared. The small and cheap pumps that can often be bought locally for domestic purposes should generally be avoided, because they usually leak, are easily broken and often lack sufficient power to direct the spray to the top of high-roofed rooms or houses. However, suitable pumps are becoming increasingly difficult to obtain. Some operators in preference to buying cheap locally made 'flit-guns' have resorted to using larger sprayers, usually with a lance, that are sold for agricultural spraying. In India Batra *et al.* (1979) collected mosquitoes by space-spraying houses, huts, cattle sheds, firewood sheds and other shelters with a swingfog machine. Because of the open nature of the structures they had to use 1% pyrethrum, not 0·2% as generally recommended, to get a good knock-down.

Whatever type of spray gun is used the spray is first directed at all potential escape routes, such as closed doors, windows and eaves; it is then aimed towards the roof or ceiling. To reduce the numbers of mosquitoes escaping from the large gaps that exist in some huts between the tops of the walls and roof, one or two assistants walk round the outside of the hut spraying these gaps either before or while the inside is sprayed. In small huts, however, it may be possible to fill up these gaps with surplus spray sheets. If a hut has either no door or one that fits very badly two assistants stationed outside the hut hold a spray sheet over the doorway both during and after spraying. In Sri Lanka Büttiker (1958) pinned or pegged cotton sheets over the palm matting walls of improvised huts and shelters to prevent mosquitoes escaping. Mr R. B. Highton (pers. comm. 1974) has made use of the mosquitoes' attraction to light when irritated by insecticides. The door of a hut is left open and mosquito netting held over the doorway. On spraying adults fly towards the doorway and eventually fall on to the floor sheets placed at the base of the doorway. Whatever method is employed sprayers leave the hut after spraying. Usually about 90% of the mosquitoes fall on to the spray sheets within the first 7 min after spraying, and after about 10 min the sprayers re-enter the hut to retrieve the mosquitoes.

Mosquitoes can be collected from the spray sheets while they remain in position on the hut floor, but it is usually better for two people to carefully pick up each sheet at the corners and shake the contents on to a single sheet. Weather permitting this sheet is taken outside the hut and examined by two collectors. If it is windy or raining, torches are used and the mosquitoes are collected from the sheet inside the hut. Mosquitoes which have been carefully picked up from the sheets with forceps or fingers are placed in small convenient containers lined with damp filter paper or lint. Plastic petri-dishes, tobacco or cigarette tins make useful receptacles. It is usually convenient to count, sex and record the gonotrophic stages and species of mosquitoes after they have been collected from each hut, then those collected from several huts in a compound or village section can be pooled. In hot weather the containers holding the mosquitoes can be placed in wide-mouthed vacuum flasks or in commercial cold boxes to keep them cool. On return to the laboratory mosquitoes will still be in a sufficiently good condition to allow dissections for malarial and filarial parasites, ovarian age-grading, and the preparation of gut smears for immunological (e.g. ELISA) tests etc. If they cannot be dissected the same day mosquitoes can be stored overnight in a domestic refrigerator, or in a deep freeze for months for age-grading dissections (see pp. 814–15).

Furniture should be returned to a house after a spray catch has been completed, otherwise householders may refuse permission for their houses to be sprayed on future occasions.

It is convenient to have a good supply of both small (2 × 1 m) and large (2 × 2 m) spray sheets available; they should be counted after removal from each hut to ensure that none are 'lost'. They must be regularly washed, because mosquitoes are easily overlooked when collecting from dirty sheets. In Egypt sheets are frequently not placed over the floor in pyrethrum space catches, instead a 1-$m^2$ white sheet supported on two sticks is moved around a room and under furniture while someone sprays a kerosene solution of 0·2% pyrethrum (El Said *et al.*, 1986; Kenawy *et al.*, 1986, 1990). This so-called index-sheet method is also used in animal shelters. Clearly considerably fewer mosquitoes than obtained by conventional spray sheet methods will be collected, and there seems little to recommend this procedure, unless for some reason householders forbid sheets being spread over the floor and furniture.

Commercial aerosol containers containing pyrethrum or pyrethroids can be used to space spray huts, but the method is usually too expensive for routine use, although Krafsur (1971, 1977) used aerosols containing 0·6% pyrethrins synergised with 1·4% piperonyl butoxide to spray houses in Ethiopia. Many aerosol preparations contain residual insecticides and these should be avoided.

**Factors influencing spray sheet collections**

There may be large variations between the numbers of mosquitoes resting in different huts in the same compound. In general unfed mosquitoes are attracted to huts in numbers related to the number of human occupants (Haddow, 1942) but no general arithmetic relationship has been established between catch size and number of occupants. In addition to the number of people in a hut many other variables such as the presence of open doorways, fires and large gaps between the eaves also affect the numbers of mosquitoes resting in them. Spencer (1965) thought that the few *Anopheles farauti* found in huts with fires was due to a reduced humidity rather than the production of smoke. Village huts which are near larval habitats may contain more mosquitoes, especially males, than those further away (Service, 1964). Small village huts occupied by a number of people generally harbour large numbers of resting adults. These types of huts can be selected and sprayed when large numbers of mosquitoes are required, such as for determining infection rates, or seasonal changes in numbers. Selecting houses containing disproportionally large numbers of mosquitoes introduces a pronounced sampling bias. To obtain a reliable estimate of the mean hut density of mosquitoes in a village, or area, representative samples of the different types of huts must be sprayed, and not just huts known or suspected to contain large numbers of mosquitoes.

It is more difficult to efficiently spray very large huts, especially those that are divided into a number of rooms, and also those that have high roofs. A smaller proportion of the mosquito population is caught in huts with an incomplete ceiling below the roof, because some of the adults that are knocked down fall on to the ceiling partition and are not collected. In East Africa Gillies (1955) considered that pyrethrum collections underestimated the mosquitoes resting in huts by about 10–20%; but because of their more sluggish flight blood-fed individuals were less likely to escape capture than other categories. Freyvogel & Kihaule (1968) were advised by Gillies that they could expect to recover about 75% of the mosquitoes present in Tanzanian huts sprayed with 0·1% pyrethrum. The ability to escape capture may vary between different species as well as between different gonotrophic stages. If so, then pyrethrum catches may not give representative samples of either the relative hut densities of different species, or of the gonotrophic stages within a single species.

Swellengrebel & de Buck (1938) observed that heavy spraying with pyrethrum reduced the numbers of the *Anopheles maculipennis* complex resting in houses, and in India Senior White *et al.* (1945) noted that pyrethrum spraying exerted a repellent effect on *Anopheles minimus* resting in houses. In Assam Ribbands (1946*b*) observed a reduction of about 90% of *Anopheles minimus* resting in huts the night after they had been sprayed with 0·1% pyrethrum at the rate of 25 ml/1000 $ft^3$. Some degree of repellency was evident up to the fourth day. In West Africa Muirhead-Thomson (1948) found that when the village huts were sprayed about every other day it reduced the house resting *Anopheles* to a third of their original numbers. In unsprayed huts, for example, only about 20% of the *Anopheles* left as blood-fed individuals whereas in regularly sprayed huts there was an exodus of about 76%. It was concluded that the reduced catch of mosquitoes in sprayed huts was mainly due to the exodus of blood-fed females. In East Africa Haddow (1942) also reported that pyrethrum spraying had a repellent effect on the numbers of mosquitoes resting in huts. There is evidence that the kerosene in the spray mixture is itself repellent. Whatever the cause it is generally agreed that because the repellency is short lived huts can be sprayed weekly, possibly twice a week but not at shorter intervals, without there being a reduction in the number of mosquitoes caught.

It is often recommended that pyrethrum or aspirator collections are started before sunrise so as to include in the catch mosquitoes that will leave huts with the onset of daylight. If there is a dawn exodus then catching should be completed, not just started, before dawn, otherwise collections from the last few huts will be made after a proportion of adults have in fact left. However, unless huts are occupied by staff members it is usually difficult or impossible, to gain access to huts before dawn. If they cannot all be sampled before dawn it is better that collections are made from all huts after dawn, although in Nigeria no significant differences were observed between the numbers of *Anopheles gambiae* or *Anopheles funestus* caught in huts sprayed at different times from 0430–0730 hr (Service, 1964). Similarly, in Kenya there was no significant difference between the mean hut densities (70·9 and 68·4) of *Anopheles gambiae* calculated from pyrethrum catches made at 0730 hr or 1400 hr (Joshi *et al.*, 1973). In contrast far fewer adults were caught at 1400 hr in huts which had been sprayed with fenitrothion. The explanation is that around dawn, just before the morning collections were made, adults that had fed outside on cattle entered the huts. In the fenitrothion sprayed huts most of these were killed by the residual insecticide before the afternoon catches were made and consequently the mean catch was smaller.

## Interpretation of house catches

Mosquitoes caught from huts are usually expressed in terms of mean hut densities, but aspirator collections can also be expressed as the numbers caught/man-hr. Hut densities will nearly always be smaller than the real densities of resting adults, although under ideal conditions knock-down spray catches may recover almost all the mosquitoes resting in huts. A hut can be regarded as a sampling unit, and several huts (sampling units) must be sampled before the relative size of the indoor resting population in an area or village can be reliably indicated by the mean hut density. An obvious difficulty is that huts differ considerably in both size and attractiveness to mosquitoes, consequently the sampling unit is not standardised. This usually results in large variations between the numbers of mosquitoes caught from different huts, and leads to a very large variance of the mean hut density. As in other sampling programmes this means that a large number of samples (huts) must be taken to get a reliable mean value. This limitation is very frequently ignored, and in many surveys the comparison of mean hut densities between different areas is unreliable because it is based on too small a sample.

Hut densities are commonly used to measure changes in the seasonal and annual abundance of mosquitoes, to compare house resting densities in different villages or areas, and to assess the impact of control measures on the endophilic species. Very rarely have attempts been made to estimate the total population of an endophilic mosquito in a village from hut densities. In an East African village Gillies (1955) calculated the total indoor populations of *Anopheles gambiae* (15 577) and *Anopheles funestus* (15 002) not by taking a sample of huts but by spraying all the huts (119) on five separate days over a period of 11 days. Because the percentage of both species that rested out of doors was known (about 5%), Gillies was able to estimate their total populations in the village, an area comprising 380 acres. Regular human bait collections were made in a hut throughout a year to measure seasonal changes in relative population size, and by using these values Gillies (1955) was able to adjust his single population estimates of *Anopheles gambiae* and *Anopheles funestus* to give estimated figures for total populations throughout the year.

The ratios of the number of mosquitoes caught in indoor night human bait catches to the number of human blood-fed mosquitoes caught in pyrethrum spray sheet collections can be used as an index of exophily. The higher the index the greater the degree of exophily. In northern Nigeria Molineaux *et al.* (1976) found that this index increased in the rainy season. This could be due to: (i) progressive increases in anthropophagy (i.e. decrease in non-human feeds found in pyrethrum catches); (ii) decrease in time mosquitoes rested indoors; (iii) changes in the relative bias of the two sampling methods; or (iv) a decrease of the proportion of mosquitoes resting indoors after feeding (i.e. endophily). They concluded that the latter was the most likely explanation and was associated with an increase in vegetation during the rains which provided more out of door resting places.

Only rarely have mosquito sampling data been fitted to mathematical distribution models, but Nedelman (1983), in an interesting analysis of the entomological data from the World Health Organization study on the epidemiology and control of malaria in Garki, northern Nigeria (Molineaux & Gramiccia, 1980) tested the data against the negative binomial, Neyman Type A and Polya-Aeppli distributions. It was found that the numbers of adult *Anopheles gambiae* s.l. caught from houses in pyrethrum spray sheet collections were best described by a negative binomial distribution with temporally varying means but a constant $k$. Taking $k$ to indicate heterogeneity among the houses, then this is largest for pyrethrum collections, then for exit traps fitted to the huts and finally for indoor human bait collection. Molineaux & Gramiccia (1980) reported that the relative efficiencies of these three sampling methods, and also out of door collections, varied over time. Nedelman (1983), however, believed that except in one of eight villages these trapping methods maintained constant relative efficiencies throughout the rainy season, and in all but two of the villages the relative efficiencies of the four sampling methods were the same.

As discussed above Molineaux & Gramiccia (1980) considered that the pooled counts from all villages for indoor bait catches and out of door resting collections from pit shelters increased relative to the pyrethrum collections, through the rainy season. They believed that this increase was due to fewer *Anopheles gambiae* s.l. resting indoors as vegetation grew and so provided better out of door resting sites. However, Nedelman (1983) analysed data from all villages separately and concluded that apparent increases in exophily might depend on local ecology.

In Haiti Hobbs *et al.* (1986) compared the numbers of *Anopheles albimanus* recorded inside houses in human bait collections, with the numbers of blood-fed mosquitoes caught during 15-min search periods performed hourly throughout the night. Because considerably fewer blood-feds were caught resting in houses than were caught as unfeds in all night bait collections, it was concluded that engorged *Anopheles albimanus* rested for only short periods in houses after feeding. However, a problem with such comparisons is that often only a small percentage of mosquitoes actually resting in houses is caught. Nevertheless, in Jamaica Muirhead-Thomson & Mercier (1952) concluded that only about 10% of *Anopheles albimanus* breeding in houses remained in them during the day.

### Estimation of indoor population by the removal method

The application of the removal method to estimate population size is discussed in detail with reference to larval populations in Chapter 2. Yasuno *et al.* (1973*b*, 1977) applied the method to the collection of *Culex quinquefasciatus* resting in urban houses in Delhi. Basically the principle is that the numbers of individuals caught in a sample are related to the size of the population, and that if a number of successive samples are taken then the population decreases in size and consequently the numbers caught in the samples diminish. In their study Yasuno *et al.* (1973*b*) collected *Culex quinquefasciatus* from 10 houses using either four people per house collecting with aspirators or two people catching adults in hand-nets. The total numbers caught by the two methods were separated into seven 5-min units of time, but in one house collecting was extended for 18 time units (90 min) and 605 mosquitoes caught. Figure 3.5 shows that in this catch there was a reasonably good regression of the numbers caught against time. The estimate of total population present ($\hat{N}$) is given by:

$$\hat{N} = \sum_{i=1}^{k} (i-1)y_i \div (1-q)^k$$

where $y_i$ = numbers caught in $i$th time unit collection, $k$ = number of time unit collections, $p$ = probability of capture during a single time unit and hence $q = 1 - p$. A more complete explanation of the procedure is given in Chapter 2.

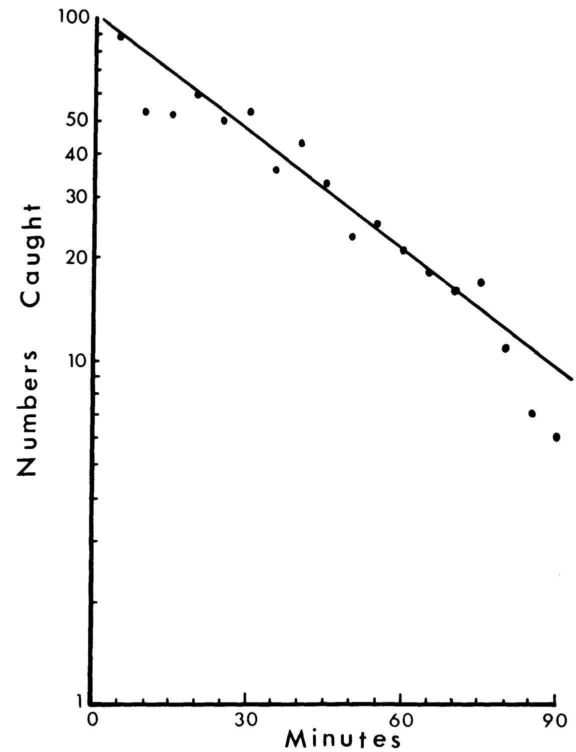


*Fig. 3.5. Number of* Culex quinquefasciatus *caught by successive 5-min collections with aspirators from a house in India (after Yasuno* et al., *1973*b*).*

The average probability of capture per unit time ($p$) based on collections from 10 houses was 0·124 for aspirator collections and 0·297 for hand-net collections. This difference might have been the cause of the greater standard error attached to the first type of collection (6·1–23·1% of $\hat{N}$) than to the latter (1·4–10·4% of $\hat{N}$). It was concluded that even four people collecting in a single house with aspirators was inefficient and did not give an accurate population estimate. However, as the standard error of the probability of capture ($p$) was quite small (0·17–0·035) it seemed that if more time unit collections were performed a more reliable population estimate could be obtained. For example, from Fig 3·5 seven unit collections give the estimate of the population of *Culex quinquefasciatus* in the house as 752 ± 145, whereas from 18 unit collections the population estimate has a much smaller error, 711·8 ± 21·6.

When two men caught mosquitoes with hand-nets the confidence limits were generally within ±10% of the estimates, and under the conditions of the experiment about seven time unit collections were sufficient. It was concluded that the sweep-net method was the better of the two in collecting a large proportion of the mosquitoes resting in houses.

In Papua New Guinea Charlwood & Bryan (1987) collected indoor resting *Anopheles punctulatus* from a house with aspirators and segregated the catch into twelve 5-min intervals; they then estimated the absolute population by the regression of numbers caught in the *i*th catch against previous total catch, and also by maximum and likelihood methods (Zippin, 1956, 1958). The regression method estimated the population as 192 and the Zippin method as 282 ± 53·1, whereas the actual total catch was 280. These very similar results indicated that aspirator collections were efficient in collecting most if not all *Anopheles punctulatus* resting in houses—or at least in this house.

The ability of removal trapping to provide an accurate estimate of the total population of mosquitoes resting in a collection of houses, or in a whole village or town, obviously depends on the number of houses sampled. For example, Yasuno *et al.* (1973*b*, 1977) had to collect a high proportion of the mosquitoes resting in houses before populations could be reliably estimated. This is a common disadvantage of this method (see Chapter 2). Although removal trapping can be used to obtain the total numbers of mosquitoes resting in houses, in most instances equally, if not more, reliable results could be obtained more conveniently by carefully performed pyrethrum space spraying.

## THE OUTDOOR RESTING POPULATION

It is usually more difficult to find those mosquitoes that rest out of doors (see p. 210 for references) than those that rest in buildings such as houses, shelters, barns and animal quarters. This is because outdoor populations are usually widely distributed over large areas and not concentrated in discrete units. Most mosquito species in fact rest exclusively out of doors in natural resting places, and only a relatively few species rest in man-made shelters. Even with a species such as *Anopheles gambiae* which is regarded as highly endophilic, a certain proportion of the population may be found resting out of doors (Gillies, 1954) although suitable indoor resting sites are available.

Gillies (1956) considered that three basic types of exophily could be exhibited by malaria vectors: (1) *obligatory*, where because of the absence of man-made shelters *Anopheles* were compelled to rest in natural outdoor shelters; (2) *facultative*, where human and animal habitations were available but adults rested in either natural outdoor shelters or in houses; and (3) *deliberate*, where mosquitoes specifically avoided resting in habitations although they were available. Mosquitoes in this category were further divided into: (a) those that in fact fed in houses (endophagic) and rested outside; and (b) those that both fed (exophagic) and rested out of doors. Gillies (1956) was principally concerned with the epidemiological importance of exophily exhibited by malaria vectors, and was little interested in outdoor resting populations of unfed females, or males. From an ecological point of view, however, it may be important to adequately sample all gonotrophic stages and age classes of mosquito populations.

It has usually proved very difficult to get reliable comparisons between either the relative or absolute sizes of the indoor and outdoor resting populations of mosquitoes. Molyneaux & Gramiccia (1980), however, tried to estimate the proportion of the *Anopheles gambiae* complex and *Anopheles funestus* resting out of doors in northern Nigeria by an indirect method which is summarised on pp. 224–5.

The size and importance of the exophilic population of species that commonly rest inside houses is probably often overlooked.

Apart from the daytime resting of males and gonoactive females, hibernating mosquitoes are also found in a variety of outdoor resting sites.

Exophilic mosquitoes shelter in many types of habitats, such as animal burrows, hollow trees, holes at the base of termite mounds, cracks and crevices in the ground, on tree trunks, under bridges, on fencing and brick walls, in abandoned mines, and amongst a variety of vegetation. These different habitats provide a wide range of microclimatic conditions. Generally, adults resting amongst vegetation will be afforded less protection from wind, sunlight and desiccation than those sheltering in rodent holes and tree-holes etc. Different species may exhibit marked preferences for resting in particular habitats. Harwood (1962) found *Anopheles freeborni* overwintered in animal burrows, whereas *Culex tarsalis* occurred mainly amongst vegetation and rock-holes and fissures. Breeland (1972*a*) gave a detailed list and description of the favoured natural diurnal resting sites in El Salvador of *Anopheles quadrimaculatus* and *Anopheles pseudopunctipennis*, both species favouring rock crevices and fissures. Also in El Salvador *Anopheles albimanus* can be found resting under bridges, in culverts and rock crevices and between the buttress roots of trees, but in Cuba extremely few adults are found in these situations (A. Navarro, pers. comm., 1983). In England *Coquillettidia richiardii* and several *Aedes* species rest primarily amongst vegetation, whereas various *Culiseta* spp., *Anopheles plumbeus* and *Anopheles claviger* mainly occur on tree trunks (Service, 1969, and unpublished results). In Guatemala Cupp *et al.* (1986) collected few *Mansonia titillans* until they discovered they rested in tall grasses growing in bunches around bases of trees in open fields. During surveys in Tennessee over a million *Anopheles quadrimaculatus* and numerous other *Anopheles* species were caught from barns, but although *Anopheles walkeri* comprised about 30% of the *Anopheles* caught at bait, at first none, and later only a few, were found in the barns (Snow & Smith, 1956). It was finally discovered that *Anopheles walkeri* rested during the day on the stems of various swamp plants.

Not all vegetation is equally attractive to mosquitoes. During collections of mosquitoes from various resting sites in England none was caught resting in bracken (*Pteris aquilinum*) although the plant would appear to offer attractive shelter (Service, 1971*a*). In Malaysia, however, bracken (*Glichenia* sp.) proved to be a favourite resting site of several *Anopheles* species (Wharton, 1950). In Tanzania the outdoor population of *Anopheles gambiae* was found sheltering amongst the dense growths of salt bushes and also in crevices in the ground, whereas *Anopheles pharoensis* occurred in less dense growths of salt bush (Smith, 1961). In some areas of Kenya *Anopheles arabiensis*, and to a lesser extent *Anopheles gambiae*, are commonly found in small granaries made of maize stalks (Clarke *et al.*, 1980; Githeko, 1992), while in other areas lacking such granaries adults of *Anopheles arabiensis* can be found in cracks and crevices of brick pits and in cracks in the ground (Service, 1985). In these highly favoured resting sites adults of *Anopheles pretoriensis, Anopheles pharoensis* and *Anopheles rufipes* can also be found, but surprisingly not *Culex quinquefasciatus* although this species is very common in nearby houses.

The distribution of mosquitoes resting amongst vegetation may change during the day so that they avoid direct sunlight (Senior White *et al.*, 1945; Service, 1971*a*). There may also be differences between day-time and night-time resting sites; for example in El Salvador *Anopheles albimanus* rests during the day in rock crevices, culverts and other natural sites, but at night adults accumulate in large numbers on walls and fences enclosing cattle corrals (Breeland, 1972*a*,*b*, 1974; Breeland *et al.*, 1974). Apart from movements within a habitat, changes in weather conditions may cause some species to seek shelter in more protected habitats. In an area in England where *Aedes cantans* normally rested amongst vegetation, numerous adults were found in rodent burrows during an exceptionally dry period. They were previously absent from these burrows and vacated them again after heavy rainfall (Service, 1973). In Tennessee *Anopheles walkeri* rests almost exclusively amongst vegetation, but during the hot summer months a few adults are found in barns (Snow & Smith, 1956). Similarly in Pakistan *Anopheles culicifacies* is found in exposed dryish resting sites during the cooler months of the year, whereas they tend to switch to resting in damp cattle sheds during hotter periods. In Turkey, *Anopheles sacharovi* rests principally in shelters during the winter months, but in more exposed places in the hotter months (Service, 1989).

Indoor spraying with residual insecticides can also affect the behaviour of mosquitoes, such as *Anopheles minimus* which was formerly an endophilic malaria vector over much of the Oriental region. But now in Myanmar after some 35 years of DDT house-spraying it is endophilic. Similarly, in India *Anopheles minimus* was formerly predominantly endophilic and endophagic, but it is now frequently found resting out of doors, and in Thailand this vector is now mainly exophilic and exophagic (Ismail *et al.*, 1974, 1975, 1978). In contrast, in Nepal, house-spraying has not selected out exophily, but resulted in the virtual elimination of the species (Parajuli *et al.*, 1981). Other cases of increased exophily have been reported for the *Anopheles gambiae* complex (Muirhead-Thomson, 1960), *Anopheles sundaicus* (Kalra, 1980; Kalra in Bang, 1985), *Anopheles philippinensis* (Bang, 1985), and *Anopheles farauti* (Taylor, 1975).

In studies on Japanese encephalitis in India Mani *et al.* (1991) collected adult female *Culex tritaeniorhynchus, Culex vishnui* and *Culex pseudovishnui* from in and around cattle sheds in the hour after sunset and multiplied the average number caught by the proportion parous to obtain a 'dusk index'. When this index was compared with human bait catches (1800–0600 hr) both exhibited a sharp increase after transplantation of rice seedlings, but the dusk index remained high after biting counts had decreased to a low level. This was due to the paucity of *Culex tritaeniorhynchus* at human bait, and its abundance in dusk collections around cattle sheds. The dusk index was therefore routinely used to monitor vector densities.

Since the first edition of this book there have been greater attempts, in both temperate and tropical countries, to collect out of door resting mosquitoes, and many such efforts have proved rewarding. The most widely used methods employ motorised aspirators (sweepers) or sweep-nets to collect mosquitoes resting in vegetation, while in the USA the use of new types of man-made resting shelters (e.g. walk-in red boxes) have proved rewarding.

## NATURAL RESTING SITES

### Crab holes

In Panama large numbers of several *Deinocerites* species were collected from crab holes by placing fine mesh cages over their entrances and then either dislodging the mosquitoes by blowing in smoke, or by forcing them up into the cages by flooding with water (Tempelis & Galindo, 1970). In Jamaica mosquitoes were also collected from crab holes by lightly spraying with pyrethrum and catching the escaping adults in cages placed over their entrances (Muirhead-Thomson & Mercier, 1952).

In Malaysia Rudnick (1986) reported the capture of 257 male and 666 female adult mosquitoes from nine aspirator collections from crab holes. All the males, except one, were *Uranotaenia lateralis*, and 55% of the females were also of this species.

### Caves

Mosquitoes resting in caves can usually be caught directly with aspirators, but in Israel Saliternik (1965) found it more convenient to catch *Anopheles sergentii*, which rested during the day in caves, crevices and fissures in limestone rocks, in exit cages. The cages, which consisted of a 20 × 20 × 18 cm wooden framework covered with mosquito netting, were placed over or close to the opening of crevices or caves during the day, and the catch retrieved the following morning. Large numbers of *Anopheles* were also collected from caves in Palestine by Shapiro *et al.* (1944).

### Tree trunks, fencing, culverts, banks etc.

Mosquitoes resting on relatively exposed surfaces can be collected with aspirators. Their distribution may be very patchy, but with practice favoured resting places, sometimes the basal 2–3 ft of tree trunks, may be identified. Gently tapping or prodding tree trunks, bromeliads, and earthen banks etc. with a stick usually disturbs resting mosquitoes which can then be caught in a small hand-net.

In Zika forest, Uganda from 20 collections performed throughout the 24-hr day from tree trunks McCrae *et al.* (1976) succeeded in catching 1328 male and 1777 female *Anopheles implexus*.

Mosquitoes resting on branches and foliage of trees have sometimes been collected by spraying the trees with insecticide. For example, in Australia Kay (1983) used a swingfog machine to spray 0·1% pyrethrins synergised with 0·6% piperonyl butoxide into trees. The operator had to sometimes stand on a ladder to ensure the insecticidal fog reached the tops of the trees. Mosquitoes and other insects were collected beneath the trees on plastic sheets. This method demonstrated that considerable numbers of *Culex squamosus* were resting in the trees, a species poorly represented in aspirator collections from vegetation and resting box catches. In the USA Simmons *et al.* (1989) used a backpack sprayer fitted with a ULV nozzle to spray the lower canopy of trees with 3·3% resmethrin insecticide. Resting simuliid flies were knocked down and collected from sheets spread on the ground under the trees. Shaking the trees helped dislodge insects which had fallen onto leaves. From 32 man-hr 84 simuliid blackflies ($\bar{x}$ = 2·63) were obtained, but truck trapping for 450 man-hr yielded 8730 blackflies ($\bar{x}$ = 19·4).

Papers written in Japanese during 1968 and 1971 have described how using commercial insecticidal smoke 'bombs' (canisters), which are normally used to fumigate houses and warehouses, a variety of arthropods can be collected from trees. An English description of this method was later given by the authors Yamashita & Ishii (1977). Basically the smoke canisters were ignited and either hand-held or placed in metal containers supported on a variety of extension tubes to reach high up in the tree canopy. Smoking lasted 1–3 min and insects knocked down were collected on white trays or sheets placed on the ground underneath the trees and bushes. Diptera formed 7·0–21·8% of the arthropods caught by this method.

Frank & Curtis (1977) give an interesting description of their frustrations in trying to catch large numbers of both sexes of *Wyeomyia vanduzeei* for mark–release experiments. The creation of artificial resting sites and 'honey pot' feeding stations proved unsuccessful. The only way of collecting adults was to aspirate them from their natural resting sites on the rough bark of buttonwood trees. With experience they were able to catch about 80 adults, with both sexes approximately equal, in 2 hr, and from catching for 2 hr daily 5 days/week for 42 weeks 5920 male and 3836 female *Wyeomyia vanduzeei* were caught.

## Tree-holes, rodent burrows, termite mounds, crevices etc.

### *Aspirators*

Adults resting in these types of recesses can be located with a torch and collected with aspirators. Büttiker (1958) collected 32 female and 22 male *Anopheles culicifacies* including blood-fed individuals from a tree-hole in Sri Lanka. In El Salvador Breeland (1972*b*) collected with aspirators an average of 42·4 adults of *Anopheles albimanus* and 12·9 *Anopheles pseudo-punctipennis* per hour from natural diurnal resting sites such as rock crevices, tree-holes and ground holes. In other surveys, 7670 *Anopheles albimanus* and 2344 *Anopheles pseudopunctipennis* were caught in 181 collections from natural shelters. These day-time collections of resting adults were much less time consuming than night captures, and were probably less influenced by climatic conditions. Furthermore, they were of particular value in catching adults of *Anopheles pseudopunctipennis* which are infrequently caught in light-traps, at human bait, in stable captures or in collections from houses. Breeland (1972*a*,*b*) concluded that the collection of adults from day-time resting sites provided reliable information on changes in population levels of *Anopheles albimanus* and *Anopheles pseudopunctipennis*. In southern California and Mexico hibernating adults of *Anopheles freeborni* and several other mosquitoes were collected from nests of wood rats (*Neotoma fuscipes*) (Ryckman & Arakawa, 1951, 1952).

In India 2910 anophelines belonging to nine species were collected in 588 man-hour collections using sucking tubes and torches to search in holes in damp mud banks, in crevices, amongst stones, on fences and in a culvert. The last proved to be the most productive site, yielding up to 400 *Anopheles culicifacies*/man-hr (Mani *et al.*, 1984). In Canada Hudson (1978) collected overwintering mosquitoes from man-made rock piles that were covered with snow. Firstly he removed the snow and then turned over the stones (19 to >50 cm) by hand and aspirated out adults. Twelve searches in seven rock piles (116·5 man-hr) yielded 108 *Anopheles earlei*, 102 *Culex territans*, 1 *Culiseta alaskaensis* and 1 *Culiseta minnesotae*.

### *Fumigation and spraying*

In many situations relatively few mosquitoes can be collected by direct searches, but more may be caught if they are disturbed from their resting sites and caught as they fly out. Tobacco smoke, smoke from a beehive fumigator or produced by burning corrugated paper soaked in potassium chlorate can be blown in, or Risella or citronella oil or a weak solution of pyrethrum (0·5%) can be sprayed into, the resting sites. Alternatively commercially available insecticidal aerosols can be used (Service, 1963). Büttiker (1958) collected *Anopheles culicifacies* from large cavities in tree-holes in Sri Lanka by placing spray sheets over the bottom and also over the opening and then spraying with pyrethrum. Zukel (1949*a*) collected adults from hollow trees by fumigating them with sulphur dioxide produced by burning cheese cloth impregnated with a paste of sulphur and fuel oil wrapped round a wire frame. This was lighted and placed in a hollow tree which had all openings covered with canvas, and the mosquitoes collected on a white sheet placed at the base of the hollow. Zukel (1949*a*) reported that whereas only a single female of *Anopheles quadrimaculatus* was collected from hollow trees prior to smoking, substantial numbers of *Anopheles quadrimaculatus, Uranotaenia sapphirina, Culex erraticus*, and a few *Anopheles punctipennis, Culex quinquefasciatus* and *Culex peccator* were caught as a result of fumigation. He also found that smoking was more efficient in driving them out of their hiding places than fumigating with hydrogen cyanide or pyrethrum. Love & Goodwin (1961) also found fumigation useful in collecting adults from tree-holes. Mosquitoes have also been caught from hollow trees, rodent burrows and other natural resting sites by spraying them with chloroform or acetone and catching the escaping adults in nets or small cages placed over their openings (Loomis & Green, 1959; Mortenson, 1953; Trapido & Aitken, 1953; Zukel, 1949*b*). In California on 30 occasions Reisen *et al.* (1989) lightly sprayed with atomized chloroform 605 rodent burrows, and on another three occasions they sprayed 15 burrows, but only four female and two male escaping *Culiseta inornata* were collected in sweep-nets.

In India Batra *et al.* (1979) tried several methods to collect exophilic mosquitoes, including lowering a canvas cloth fixed to an umbrella frame down wells and then space spraying above with 1% pyrethrum. Nine or more mosquito species, including 50 *Anopheles stephensi* and 924 *Culex quinquefasciatus*, were caught from 102 urban wells, and 214 *Culex quinquefasciatus* from rural wells. Most of these mosquitoes, however, were newly emerged individuals. Mosquitoes resting in culverts and in some other miscellaneous places were smoked out by lighting mosquito coils and catching escaping adults in hand-nets. In other shelters, such as tree-holes, bamboo, holes in walls and also other culverts, adults were caught in hand-nets after they had been flushed out by spraying with 1% citronella oil. From 273 culverts 45 *Culex quinquefasciatus* were caught when mosquito coils were lit, but none was caught from 60 culverts sprayed with citronella oil, and only seven mosquitoes were caught from all the other resting sites sprayed with citronella oil.

### *Traps*

Harwood & Halfhill (1960) developed a simple trap for collecting mosquitoes, mainly *Culex tarsalis* and *Anopheles freeborni*, from amongst vegetation, crevices in the ground and from animal burrows. Their trap consists of a 2-lb coffee can with a fine wire mesh inverted cone fixed into the base, and with a large circular hole cut from the top and covered with fine mesh. The can rests on a metal flange soldered to a circle of ½-in wire mesh (Fig. 3.6*a*). The trap is placed over an animal burrow, and earth is placed around the base of the trap so that light only enters through the top of the can. Mosquitoes fly out of the burrow through the inverted cone of the trap. A similar trap was used in Australia for collecting biting Diptera emerging from vertebrate burrows (Dyce *et al.*, 1972). Wire mesh over the entrance of the can prevents rodents entering the trap. The

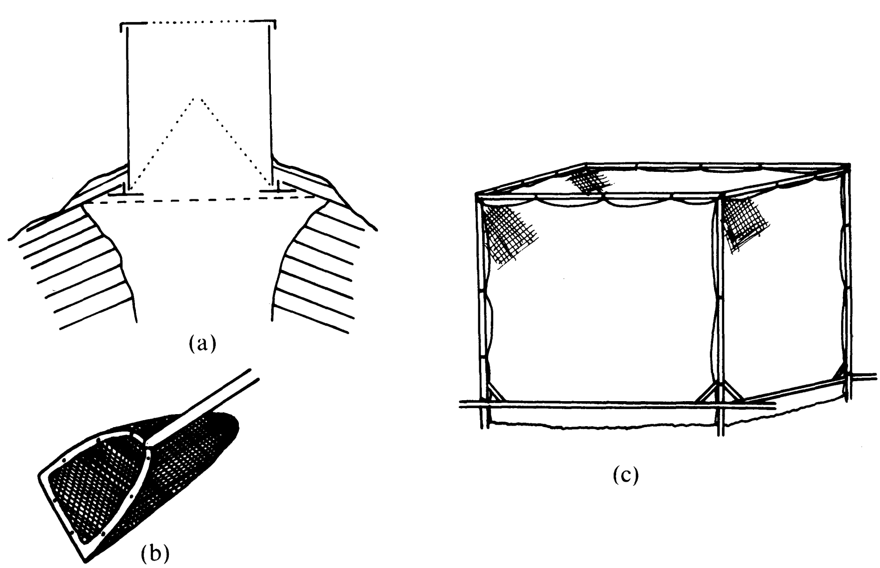


*FIG. 3.6. (a) Diagram of trap used by Harwood & Halfhill (1960) to collect mosquitoes resting in crevices in the ground; (b) sweep-net; (c) large drop-net cage.*

trap can be modified for collecting mosquitoes from vegetation and rocky outcrops and crevices in the ground (Harwood, 1962; Harwood & Halfhill, 1960). Canvas, black heavy duty polythene or tarpaulin is fixed to the trap's metal flange which is supported on a tripod having 1–1½-ft long legs. The bottom edges of the tarpaulin are held down with rocks so that a tent-like structure is erected over the area to be sampled. The tripod legs are coated with motor oil to prevent spiders ascending and spinning webs inside the trap. When 118 traps were placed in position over rodent holes during the mid-afternoon and recovered the following day an average of 2·0 female and 3·0 male *Culex tarsalis* and 12·7 female and 8·2 male *Anopheles freeborni* were collected per night. Some burrows gave consistently high catches. One that was probably inhabited, or used by wood rats, produced an average of 9·0 female and 13·1 male *Culex tarsalis* and 8·7 female and 11·0 male *Anopheles freeborni*, while a marmot burrow gave an average of 4·5 female and 7·0 male *Culex tarsalis* and 210 female and 46·5 male *Anopheles freeborni* per night. This type of trap was also used to study hibernating populations of *Culex tarsalis* and *Anopheles freeborni* in the USA (Harwood, 1962), and overwintering adults of *Culex tarsalis, Culiseta inornata* and *Anopheles earlei* in Canada (Shemanchuk, 1965).

Hudson (1978) also working in Canada used modified traps of Harwood & Halfhill (1960) to collect mosquitoes emerging from burrows of the badger (*Taxidea taxus*). Traps consisted of 15-cm long, 10-cm diameter cans, with the bottom removed and replaced with a black 1-mm mesh cone funnel, ending in a 1·5-cm diameter hole at the apex. A 2·5-cm hole was cut from the metal push-on lid and covered with 2-mm metal mesh screening. The base of the can was inserted through a hole cut from a 20-cm square piece of plywood, having a skirt of black plastic attached to its edges to help seal the trap over the badger's burrow. To prevent rodent damage traps were enclosed in protective 6-mm wire screening. Every 7–10 days the push-on lid was removed and the catch aspirated out, but 10–20% escaped; clearly a more efficient removal method is required. A total of 127 *Anopheles earlei* and 1 *Culex territans* were caught from 53 burrows, but the mean numbers of *Anopheles earlei* caught per burrow varied from 0·5 to 7·0, and in fact no mosquitoes were recovered from 49% of the burrows.

In Sri Lanka a few mosquitoes were collected by placing muslin gauze in and over holes in termite mounds and stimulating mosquitoes to fly out by pouring about 5 ml of mosquito repellent containing a benzoate derivative into some of the openings (Büttiker, 1958). Mosquitoes resting in nests of kingfishers, sand martins and bee-eaters, which consist of narrow burrows in earthen banks, were examined for mosquitoes by the insertion of a narrow strip of filter paper and spraying with 2% pyrethrum. The idea was that mosquitoes knocked down by the spray were caught on the paper strip which was then withdrawn. However, only four female culicines were recovered from 12 birds' nests by this method (Büttiker, 1958).

## Granaries

Clarke *et al.* (1980) were the first to show that in the Kisumu area of Kenya substantial numbers of the *Anopheles gambiae* complex (subsequently shown to be both *Anopheles arabiensis* and *Anopheles gambiae*) rested in grain stores, whose walls and roofs were made of maize or millet stalks plastered with mud. Mosquitoes resting in these granaries can be caught by pyrethrum spray sheet collections. For this a sheet is placed over the stored grain inside the granary while another is held over the entrance of the granary (Fig. 3.7). Alternatively a person with a torch enters the granary and collects mosquitoes with an aspirator.

*FIG. 3.7. Sheet being placed over entrance of a granary in Kenya prior to pyrethrum-spraying it (M. W. Service).*

In the study by Clarke *et al.* (1980) Muirhead–Thomson-type pit shelters were dug in the same compounds as those with grain stores. The mean monthly number of the *Anopheles gambiae* complex they collected per house was 17·0 times greater than those from pit shelters, but just 2·5 times more than those collected from granaries. Grain stores gave collections 6·8 times greater than from pit shelters. *Anopheles funestus* is a more endophilic species and few adults were collected from either grain stores or pits, the numbers in houses being 53·3 and 34·3 times greater than those found in pit shelters and granaries. The numbers in granaries were 1·6 times greater than those in pit shelters. These and later studies (Githeko, 1992) have shown that *Anopheles arabiensis* more commonly rests in granaries and other out of door structures than does *Anopheles gambiae* s.s.. Githeko (1992) constructed simple walk-in box-like shelters made of papyrus fronds in areas lacking granaries, and found they were very attractive out of door resting sites for *Anopheles arabiensis.*

## Vegetation

### *Sweep-netting*

Many mosquito species rest amongst grassy and shrubby vegetation and on the foliage of bushes and shrubs. Mosquitoes have sometimes been collected by slowly walking through vegetation and capturing them in small hand-nets as they are disturbed and fly out (McClelland, 1957; McClelland & Weitz, 1963; Teesdale, 1959). McClelland (1957), however, realised that this procedure was likely to be biased in favour of collecting unfed females of species that bite man, because having been disturbed they will tend to be attracted to the collector. In one series of catches in East Africa the catchers used an insect repellent (dimethylphthalate) to try and prevent unfed females of *Aedes aegypti* being attracted to them after they had been disturbed from their day-time resting sites (McClelland, 1957).

In the USA Copeland (1986) caught adults of *Aedes thibaulti* from a wood by disturbing vegetation with a stick and collecting them with a sweep-net. He also used two green resting boxes, but most adults were collected with aspirators (Nasci, 1981). In South Africa mosquitoes that were flushed out by walking through grassy vegetation were caught in test tubes as they resettled on nearby vegetation (De Meillon *et al.*, 1957). Some species may not be readily flushed out by walking through vegetation but can be caught if the vegetation is vigorously sweep-netted. In Kenya, van Someren *et al.* (1958) caught 65 species of mosquitoes by this method.

The most suitable sweep-net consists of a strong white calico bag fastened by pop studs over a D-shaped metal frame to which a 2–3-ft wooden handle is attached (Fig. 3.6*b*). A number of swift forward and backward strokes are made without interruption through the vegetation, then the bag, which is quickly folded over to prevent the catch from escaping, is lightly sprayed with chloroform and placed in large plastic bag (e.g. shopping bag). After 1–2 min the net is removed and the contents tipped into a white photographic dish. Leaves and debris are discarded and the mosquitoes collected. If mosquitoes caught in sweep-net collections are required alive the procedure advocated by Masner & Gibson

(1979) might be tried. This consists of quickly inverting the sweep-net over a strong 24-cm deep cloth bag fitted over a 24-cm metal ring mounted on a short tripod, and then closing the bag's opening with a transparent plastic lid. Mosquitoes, and other insects that are positively phototropic, will fly or crawl to the upper part of the bag from where they can be removed by inserting an aspirator into a small slit opening near the top of the bag. This is rather a cumbersome procedure and as an alternative the sweep-net bag can be placed for about 2–4 min in a cold box containing dry ice. The net is then removed and the contents tipped into a white dish, and the mosquitoes aspirated into paper or plastic cups.

Sweep-net bags made of calico or similar material are extremely strong and can be used to sweep holly bushes (*Ilex aquifolium*) and even bramble (*Rubus* spp.) without tearing. When, however, vegetation is wet the bags become soaked with water and the collection becomes a mass of sodden leaf litter and specimens, which, as a consequence, are often of little value. So, when sweeping wet vegetation a bag made of mosquito netting is preferable because this does not become so sodden, but of course it tears more easily. Sweep-net bags become dirty and sticky due to accumulation of sugary plant secretions and should be removed periodically from their frames and washed.

Some entomologists use a sweep-net having a detachable small bag fixed with velcro 'touch and close' fastener to the bottom (Fig. 3.8). After a predetermined number of sweeps the bag is transferred to a carton and a new one substituted. Mosquitoes caught in sweep-nets tend to become denuded of scales and consequently not too many sweeps should be made before the catch is removed from

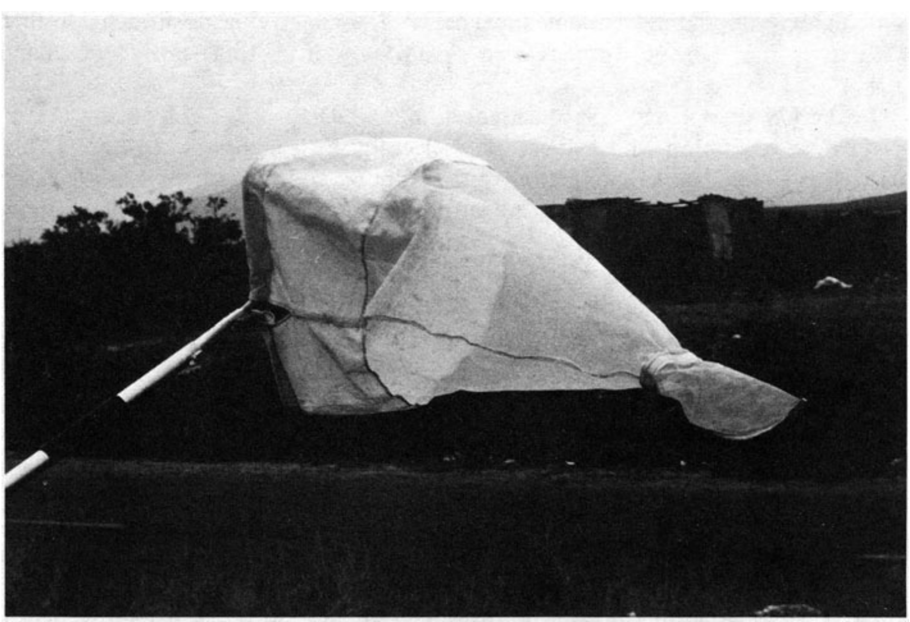

*FIG. 3.8. Sweep-net with detachable bag at bottom (M. W. Service).*

the bag and identified. In Texas standardised sampling, consisting of three series of 20 sweeps through rice fields with a 30·5-cm diameter sweep-net, were made every 30 min from 1900–2400 hr to study insemination and temporal abundance of newly emerged *Psorophora columbiae* (Robert & Olson, 1986). Magnarelli (1977*a*) collected *Aedes canadensis* by taking two 30-min sweep-net samples from the edge of its larval habitats each week during the adult season. Sweep-netting has been used in England to study the distribution of mosquitoes resting in various types of vegetation (Service, 1971*a*) and in many parts of the world to collect blood-engorged females for blood-meal identification (Aitken *et al.*, 1968; Ardö, 1958; Cordellier *et al.*, 1983; McClelland & Weitz, 1963; Pajot, 1977; Renshaw, 1991; Service, 1971*b*; Takahashi *et al.*, 1971; Wharton, 1950; Williams *et al.*, 1958).

Relative densities of different mosquito species resting in different types of vegetation, and changes in densities associated with different times of the day and year, can be obtained if the collecting technique is standardised and a known number of sweeps are taken; then the average number caught per sweep, or ten sweeps, can be calculated. In Nigeria, for example sweep-net collections were made in four different biotopes around compounds, mainly to collect *Aedes simpsoni* (Bown & Bang, 1980). But from 25 collections of 100 sweeps just 32 female and 22 male *Aedes simpsoni* were caught, mostly from bushes and from under trees, with fewer adults being caught in cocoyam and banana plantations where the species bred. In addition 47 females and 21 males belonging to other *Aedes* species, and 352 female and 881 male *Culex* species, were collected, most of which were caught from cocoyam and banana plantations. In contrast Pajot (1977) was able to collect blood-fed *Aedes simpsoni* by sweep-netting vegetation in banana plantations in the Central African Republic. In Polynesia Rivière *et al.* (1979) using small hand-nets caught a few (92 females and 7 males) *Aedes polynesiensis* resting at different heights amongst various types of vegetation.

In Louisiana Holck & Meek (1991) obtained absolute density measurements of *Psorophora columbiae* and *Culex quinquefasciatus* by placing 32-cm diameter 1-m long plastic area samplers with netting tops on the ground to enclose an area of 0·1 $m^2$. Adults were removed with aspirators from a side-hole fitted with a netting sleeve. At the same time D-vac and sweep-net samples were taken over 5-m transects. Using regression analysis it was concluded that both D-vac and sweep-net sampling could be used to estimate absolute population density (as obtained by the area sampler), but because the coefficient of variation was less for net samples and as less time was involved in taking these samples, sweep-netting was the method of choice for estimating population size.

Sometimes, however, relatively few mosquitoes are caught by sweep-netting. For example, Magnarelli (1977*b*) encountered difficulties in finding out of door resting blood-fed mosquitoes in the USA. Only 489 engorged females of *Psorophora ferox, Coquillettidia perturbans* and *Aedes* were caught in sweep-nets, while searches underneath fallen trees and collections from resting boxes of Goodwin (1942) yielded just 81 engorged *Culex* and *Culiseta* species. In the Ivory Coast although Cordellier *et al.* (1983) caught large numbers of *Aedes* and *Culex* mosquitoes (9555 females and about 10 670 males) by sweep-netting vegetation in forested areas very few females (82) of known or potential yellow fever or dengue vectors, species they particularly wanted, were collected.

In mosquito surveys in Bali Lee *et al.* (1983) collected 17 species during the evenings in sweep-nets and by aspirator collections of out of door resting adults, whereas 20 mosquito species (*Aedes, Anopheles, Armigeres, Culex* and *Mansonia*) were caught in CDC light-traps placed in or near animal shelters. However, 2.6 times as many mosquitoes were caught by sweep-nets and aspirator collections than in the light-traps.

Tonkyn (1980) quantified the process of sweep-netting as follows: the collector moves forward at a constant rate while swinging the net through a circular arc. The volume of air thus sampled is

$$\beta\left(\pi r + \frac{2\Delta y}{\pi}\right)$$

where $\beta$ = area of net opening, $\Delta y$ = distance the collector moves during the sweep, and $r$ = radius of net. So, with a sweep of 180° of radius 1·25 m with a net area of 0·073 m$^2$ (12-in diameter), made by a collector who takes two steps totalling 1·7 m with each sweep, the volume of air sampled ($V$) will be

$$V = 0{\cdot}073\left(1{\cdot}25\,\pi + \frac{2\,(1{\cdot}7)}{\pi}\right)$$
$$= 0{\cdot}366 \text{ m}^3$$

Consequently, say 25 sweeps will sample 9·15 m$^2$ of air. For sweep angles different from 180° the reader should consult this paper for the appropriate formula. Southwood (1978) gives other references on the efficiency of sweeping for non-medical insects.

However, there are many variables that can make sweep-netting an imprecise sampling method. For instance the efficiency of sweeping is likely to vary considerably between different workers; some species may be dislodged and collected more easily than others; there may be differences between vertical distribution of different species in vegetation; differences between the availability of species due to diel cyclical activities; and variations in collecting efficiency due to weather conditions. Nevertheless, despite these limitations I believe that sampling mosquitoes by sweep-netting has been underused, and that in several, if not many, situations sweeping vegetation would prove rewarding.

### *Drop-net cages*

De Zulueta (1950) in Colombia and Rehn *et al.* (1950) in Puerto Rico were amongst the first to collect mosquitoes resting in vegetation by drop-nets. Rehn *et al.* (1950) caught very few mosquitoes but de Zulueta (1950) had more rewarding results. His procedure consists of driving four vertical poles into the ground, taking care not to disturb the vegetation between them, and suspending a 2-m high and 2-m square muslin net (or tent) from the poles, the sides are then quickly pulled down. Two men enter the tent and spray the vegetation with citronella oil to disturb the mosquitoes, which are collected with aspirators from the sides of the net. De Zulueta (1950) found it took only about 10 min to catch all the mosquitoes flushed up from the 4 $m^2$ of ground enclosed by the net. From 104 drop-net catches performed from 0700–1700 hr 1106 culicines (mainly *Culex chrysonotum, Culex (Melanoconion)* spp. and *Psorophora confinnis)* and 128 anophelines (*Anopheles braziliensis, Anopheles peryassui* and *Anopheles parvus*) were collected. This gave an average of 11·9 mosquitoes/4 $m^2$, which is equivalent to about 3 million mosquitoes/$km^2$. About 23% of the anophelines and 52% of the culicines caught in the cages were males; blood-fed females formed about 11% of the catch.

To shorten the time taken to make a catch de Zulueta (1952) constructed a portable cage made from a wooden framework covered with mosquito screening. The cage, which was 1·73 m square and 2 m high, enclosed an area of about 3 $m^2$, and was fitted with a door and handles to facilitate carrying. Using this already erected cage, collections could be made about every 10 min. These collections were made in savannah grassland in an inhabited area, as distinct from the previous catches which were made in an uninhabited area (de Zulueta, 1950). Of those collected 40·0–68·8% of the female *Anopheles* and 24·8–31·9% of the culicines were blood-fed. There was a higher proportion of engorged specimens in catches made near houses. From 732 collections 2396 culicines and 220 *Anopheles* were caught, of which about 59 and 48% respectively were males. The smaller density of mosquitoes, 1·2/$m^2$, obtained in these catches was thought to reflect a real diminution in their population size.

In Jamaica Muirhead-Thomson & Mercier (1952) placed a mosquito net over vegetation but waited until sunset to collect adults that emerged of their own accord from the vegetation. They also used a large canvas tent having a single opening facing the sun and covered with a mosquito netting exit trap to collect mosquitoes resting in vegetation. In Trinidad Senior White (1952) found that drop-nets were useful for collecting mosquitoes resting amongst grassy vegetation, but not for *Anopheles aquasalis*, as adults did not respond by flying out of the vegetation in the presence of sunlight to be caught on the walls of the net.

Modified cages consisting of light wooden frames, 6-ft square and 5-ft high, constructed in four sections to facilitate carrying, were used by McClelland in East Africa (McClelland, 1957; McClelland & Weitz, 1963). A net was suspended within the framework in the manner of a Barraud cage (Fig. 3.6*c*). The top of the net and lower part of each side panel was made of calico while the upper sections were of white mosquito netting. Mosquitoes were flushed from the vegetation with either a bee smoker in which a mixture of charcoal and coarse sawdust was burnt, or with a fine spray of citronella and sesame oil. A single catch took 5–20 min depending on the numbers of mosquitoes present. Smoke was superior to spraying in driving mosquitoes out from the vegetation. Larger cages enclosing about 10 $m^2$ of ground were made by suspending nets within a hexagonal frame, 2 m high and 2 m wide, constructed from 18 sections which were assembled in the field by five people. Three assistants entered the cage by a sleeve on one side to collect the mosquitoes. Smoke was produced outside the cage and blown into it by a fan through a 15-ft length of reinforced rubber hose. Unfortunately the fan rapidly became heavily coated with resinous deposits and required frequent washing. Over 24 mosquito species belonging to eight genera were collected from vegetation by these cages (McClelland, 1957). About 23% of the females were blood-fed whereas only about 8% of the adults collected by sweeping vegetation were engorged. McClelland (1957) suggested that the lower proportion of engorged adults obtained by sweep-netting was due to the dilution effect of unfed mosquitoes attracted to the catchers and included in the collections.

A useful lightweight drop-net cage can be made from a framework of light alloy ½-in tubing, with a nylon net suspended within it. It is convenient to have a zip fastener sewn down one of the vertical edges of the cage so that collectors can enter it without having to crawl in underneath. The erected cage can be easily carried short distances and resited elsewhere by two to four people, or dismantled in about 15 min and carried to other areas.

At the Kenya coast drop-net cages 2 m square and 2 m high and having cloth loops at the corners to enable them to be easily manoeuvred by two people were placed over coconut husks, fallen coconut fronds and other debris which formed attractive resting places for *Anopheles merus.* The enclosed debris was disturbed with a stick or by kicking and adults caught for 15-min periods with powered aspirators (Mutero *et al.*, 1984). Up to 100 *Anopheles gambiae* complex, including *Anopheles merus*, per man-hr were caught. Of 724 females trapped 64% were unfed, 27·4% blood-fed, 4·8% half-gravid and 3·7% were gravid. In contrast no mosquitoes were caught in box shelters of Gillies (1954).

Drop-net cages may not always be suitable for collecting mosquitoes. For example in Guyana, Symes & Hadaway (1947) found that when adults of *Anopheles darlingi* were introduced into a 6 × 12 × 7-ft cage positioned over low scrub vegetation and grass, very few could be collected on the sides of the cage after the vegetation had been beaten. They discovered that adults were resting not on the vegetation but on the ground and on fallen leaves. In Sardinia, although a number of culicines were caught from vegetation enclosed within a mosquito net, *Anopheles* were not collected (Trapido & Aitken, 1953), and in Indonesia Bahang *et al.* (1984) tried collecting out of door resting mosquitoes by dropping a net over bushes, but very few were caught.

Drop-nets cannot be easily used in wooded areas with emergent scrubby vegetation, although there may be concentrations of day-time resting mosquitoes in such habitats. Sweep-netting is more economical in manpower than using drop-nets and can be carried out in a greater variety of habitats, but is less easy to quantify.

### *Plastic tent*

To collect adults of *Culex modestus* which in the Carmague overwinter in reed (*Phragmites*) piles and amongst the dense vegetation in reed swamps, plastic tents have been used (Mouchet *et al.*, 1969). These are made from tough white transparent plastic sheeting pulled over a 1·5-m high metal or wooden frame which is erected over swamp vegetation. An area of 3 × 3 m is enclosed by the plastic tent. The interior becomes very humid and hot, and this combined with the action of disturbing the enclosed vegetation results in mosquitoes flying out from their resting places and settling on the tent walls or roof. In addition to

*Culex modestus*, adults of *Uranotaenia unguiculata, Culex pipiens, Culex impudicus, Culiseta annulata, Culiseta subochrea* and *Anopheles hyrcanus* and a variety of other Diptera including *Culicoides* spp. have been collected in France in these plastic tents.

In attempts to collect overwintering *Culex tritaeniorhynchus* in Korea plastic tents were placed over rock piles of stones that were dismantled. A temperature of about 10–12°C was needed to induce mosquitoes to fly and settle on the roof and walls of the tent, and when the sun failed to raise the temperature to this level a gas stove was used to heat the interior. Plastic tents were also placed over piles of straw, reeds and rock walls. Ree *et al.* (1976) considered the technique was not very effective in disturbing mosquitoes resting near the ground because the temperature there remained low. The only mosquitoes collected by this method were *Anopheles sinensis* (15), *Culex pipiens* (31), *Culex orientalis* (4) and *Anopheles pullus* (1).

Plastic tents have not been widely used, but this method might prove useful in collecting non-hibernating mosquitoes resting in vegetation, especially species such as *Anopheles darlingi* which normally rest on or near the ground and are not retrieved with drop-net cages. The increase in temperature within a plastic tent might force such mosquitoes from their hiding places up into the tent, although as noted above (Ree *et al.*, 1976) found temperatures near the ground may remain low.

## Mechanical suction sweepers

### *Dietrick's machine ('D-vac')*

Several types of suction machines have been developed to collect arthropods from vegetation, but many of the earlier models required either mains supply of electricity or large cumbersome generators. Many of these suction machines have narrow diameter (1–3 in) suction hoses, convenient for extracting insects from short grassland but totally unsuitable for collecting insects, such as mosquitoes, resting in tall grass and herbage. The models developed by Dietrick *et al.* (1959) and Dietrick (1961) have much larger diameter suction hoses. The latter, which is available commercially under the name of the 'D-vac', is the more useful model as it is portable. It operates from a single-cylinder, air-cooled, two-cycle petrol engine. By altering the engine throttle air displacement can vary from about 500–1500 $ft^2$/min. Total weight is about 27 lb. A 5-ft length of 8-in diameter flexible wire ribbed canvas air ducting is connected to the fan intake. The other end is joined by an 18-in length of tapering plastic-coated nylon ducting to a fibre glass collecting head 11 in in diameter and about 11 in long (Fig. 3.9*a*). A fine 18-in long tapered organdie bag, into which the insects are sucked, is positioned within the fibre glass collecting head.

I have sometimes successfully used this machine in England to collect mosquitoes resting in grassy and shrubby vegetation and it was employed by Mitchell & Chen (1973) in Taiwan and by Tempelis *et al.* (1970) in Hawaii to collect blood-fed mosquitoes for precipitin testing. Kuntz *et al.* (1982) successfully used D-vac aspirators to collect reasonable numbers of blood-fed *Anopheles crucians* and *Psorophora columbiae*, as well as a few engorged adults of six

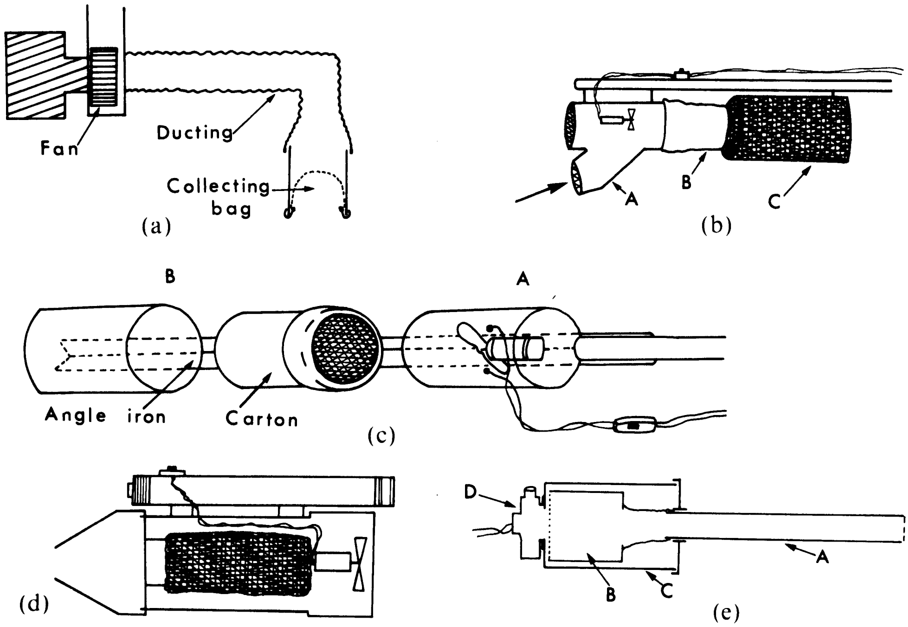


*FIG. 3.9. Mechanical suction sweepers: (a) Diagram of Dietrick type; (b) model of de Freitas, A — collecting tube, B — netting sleeve, C — collecting chamber; (c) CDC-type, A and B — cylindrical tubing, C — holding carton in middle; (d) model of R. Garcia; (e) model of Davis & Gould (1973), A — mailing tube, B — collecting chamber, C — housing for collecting chamber, D — fan unit.*

other species. However, because of its cost and unavailability in some regions Sheldahl (1974) converted a petrol-driven, backpack insecticide mistblower into an efficient and powerful aspirator. The petrol tank and sprayer hose are removed and the air intake connected to a flexible 4-in diameter, 8-ft length of hose ending in an aluminium collection chamber which houses the collection carton. The conversion of a standard mistblower to power aspirator apparently takes only some 15–20 min. The machine should be operated on about quarter throttle. Unwanted insects are excluded by placing a mesh screen over the collecting tube, and also by varying the force of the air intake. Thornhill (1978) described how a machine much cheaper than the D-vac, but very similar, could be made by using a lawn mower 4 h.p. air-cooled two-cycle 98 cc engine.

Summers *et al.* (1984) found the D-vac heavy, cumbersome and expensive and developed an improved sampler called the UC-VAC, and described it in detail. The basic unit consists of an Echo-PB-400 Power Blower with a special venturi attachment—Echo-PBAV-400—which uses energy of the exhaust to create suction in a separate section of piping. The metal collection canister has an internal diameter of 30·48 cm and is 21·10 cm long, with 'lips' 6·03 cm in diameter to which suction pipes are attached. These measurements are important in ensuring that there is maximum suction. As in the D-vac there is a removable cloth bag in the collection canister to retain the insect catch.

The total weight of the D-vac is 19·5 kg, the UC-VAC weighs 15·0 kg, at full throttle the rev/min are 3000 and 8000 respectively, in addition the UC-VAC is less noisy, vibrates less, and is less than half the price of the D-vac.

Moreby (1991) described a very simple modification that can be made to motorised vacuum samplers of the Dietrick (1961) and Thornhill (1978) type to facilitate removing the catch from the collecting bag (Fig. 3.10*a*). This consists of taping the net collecting bag on to the apical removable nozzle ring A, thus dispensing with the hooks or clips that normally secure the bag to nozzle B. Ring A now fits tightly into ring B, without the need of hooks or clips. The insect collection can be easily emptied into a plastic bag by turning the bag inside out. Only one collecting bag is therefore needed.

In England attempts to use a smaller type of motorised machine developed by Arnold *et al.* (1973) and known as the 'Univac' to collect mosquitoes from woodland scrub vegetation have not proved successful. When placed near the ground the strong air-intake through the 6·3–15-cm diameter nozzle leading to a 6·3-cm suction hose caused much ground debris and soil to be collected, which ruined the collected mosquitoes. Even when it was not placed near the ground, the few mosquitoes that were caught were cut into pieces (Renshaw, 1991). Southwood (1978) described how two bags can be used in the machine, one of nylon netting to retain insects and an outer larger cloth bag to prevent dust passing through the fan.

### *Suction sweeper of de Freitas*

This machine (de Freitas *et al.*, 1966) is composed of three main parts: (1) a 19·5-cm length of 9-cm diameter plastic tubing carrying on its lower side a 12-cm long side port made of the same material (A); (2) a 9-cm long nylon netting sleeve (B); and (3) a 26-cm long, 15-cm diameter collecting chamber (C) made from 32 × 32 mesh wire screening (Fig. 3.9*b*). A small 3–4½-V d.c. motor, such as used in CDC light-traps, having an eight-bladed propeller mounted at the rear is positioned within the upper section of the plastic tubing. The tube's opening is covered with 14 × 14 mesh wire to prevent mosquitoes entering. These are collected through the lower side port which is covered with 2 × 2 screening to prevent leaves and other debris being sucked in. The complete machine is suspended underneath a 1-m long wooden handle. The motor is wired through an on/off switch to a 3-V power supply consisting of torch batteries or a rechargeable wet cell battery attached to the belt of the operator.

This suction sweeper has been successfully used in Brazilian forests, to collect mosquitoes resting amongst vegetation, from rodent burrows and from the bases of trees. Mosquitoes are first gently disturbed by prodding vegetation or rodent burrows with a stick and then caught as they take flight. When sufficient mosquitoes have been caught, the nylon sleeve is closed with a clip or piece of string and the collection chamber and sleeve removed. Mosquitoes collected by this machine are apparently in a good taxonomic condition (de Freitas *et al.*, 1966). A greater number of species (at least 63) were caught by this apparatus than obtained by light-traps (27) and a variety of other collecting methods (42–59). The machine was used to collect mosquitoes for virus isolation studies,

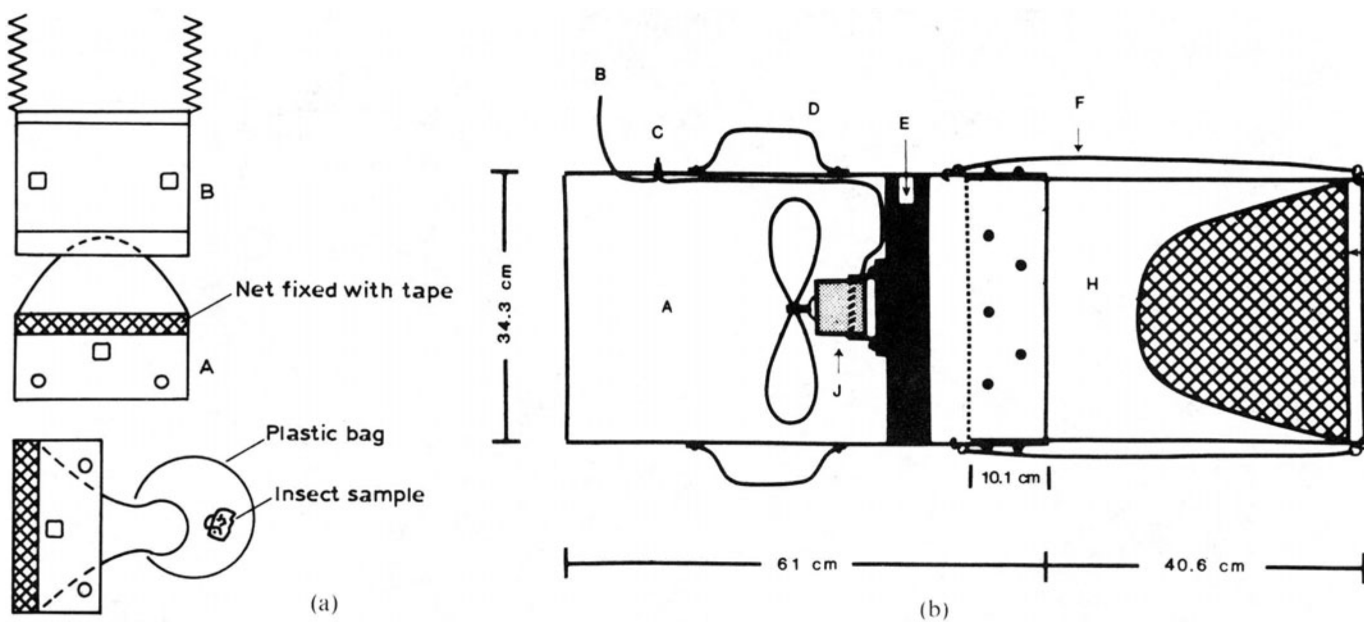


*Fig. 3.10.* *Mechanical aspirators: (a) Modification to Dietrick aspirator proposed by Moreby (1991); (b) Nasci-type aspirator, A — aluminium tube, B — wire to battery, C — push-button switch, D — heavy duty gate handle, E — wooden, motor support, F — heavy rubber band with 'S' clips at either end to secure collecting bag. G — is a D-vac bag folded over rim of plastic tube, H — plastic tube partially inserted and bolted inside aluminium tube, J — motor mount assembly (Nasci, 1981).*

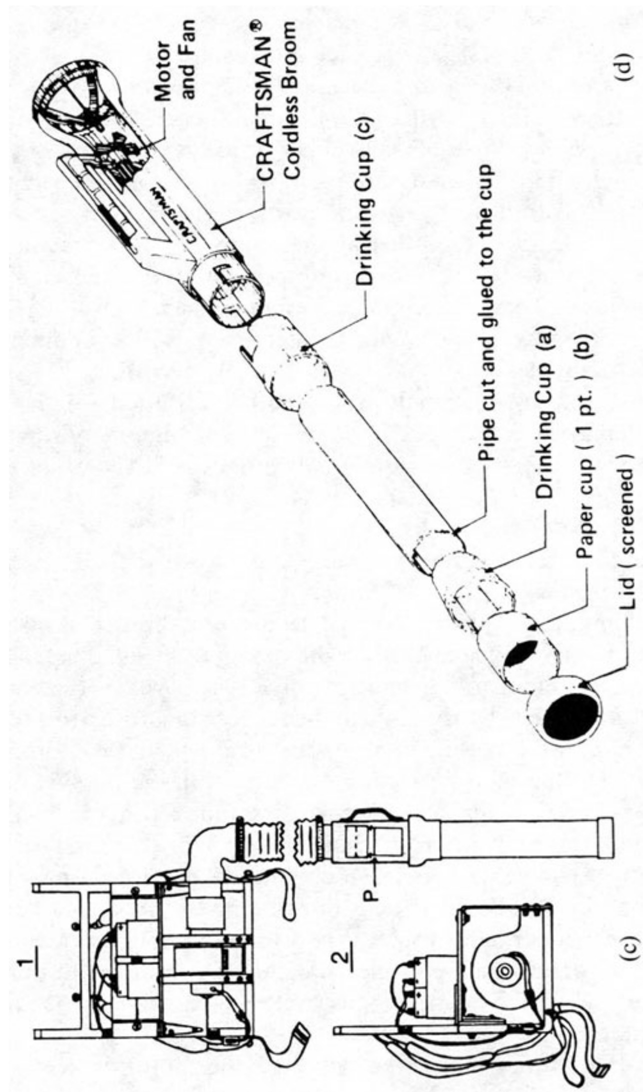


*Fig. 3.10*—contd. *(c) AFS-sweeper, 1 — rear view of sweeper showing collection carton (P) inserted into handle part of tubing, 2 — side (left) view (Meyer* et al.*, 1983); (d) cordless broom aspirator (after Perdew & Meek, 1990).*

to obtain engorged females for blood-meal determination, and to study the daytime distribution of mosquitoes resting in different types of vegetation. The inventors stressed that much of the success of collecting adults depended on the ingenuity of the operator in locating natural resting places.

Burkot & DeFoliart (1982) in Wisconsin, USA used the de Freitas *et al.* (1966) aspirator and also the D-Vac machine to collect mosquitoes. In South Africa Jupp & McIntosh (1987) constructed an aspirator, basically the same as that of de Freitas *et al.* (1966), to collect *Aedes circumluteolus* from vegetation. It consists of an 18-cm length of 15-cm diameter perspex tubing housing a 12-V, 8-W d.c. motor with a three-bladed rubber propeller. The end of the perspex tube is covered with 13 × 13-mm wire mesh to prevent debris being sucked up. Their procedure was to disturb vegetation with a stick held in one hand and then with the aspirator held in the other about 10 cm from the ground to aspirate insects in flight. About 10% of the mosquitoes that were sucked up through the fan blades into a 20-$cm^3$ collecting cage were damaged. Over a 16-month period the numbers of *Aedes circumluteolus* caught per trap-hour in different months varied from 0–416 males and <1–561 females. By recording the numbers of unfed, blood-fed and gravid females the monthly variations in the percentage of engorged females were plotted. Other relatively common mosquitoes collected were *Culex neavei, Culex zombaensis, Culex insignis* and *Culex antennatus.*

### *The CDC sweeper*

Like the previous machine, suction is provided by a 3–4½-V d.c. motor, but this operates from a 6- not a 3-V battery (Hayes *et al.*, 1967). A 3-in diameter two-blade aluminium propeller is fixed to a motor housed in a 3½-in outside diameter 4-in length of clear cast acrylic tubing (A) (Fig. 3.9*c*). The basal ⅜ in of a 1-in wide, 3½-in inner diameter ring of acrylic plastic is cemented over the opposite end of the fan housing. A pint-sized cardboard carton (C), 3⅜ in in outer diameter, with the bottom end removed and covered with no. 24 nylon mesh, is seated within the ⅝-in projecting rim of this plastic ring. This comprises the removable collection chamber. A collecting tube (B) made from acrylic tubing of the same shape and size as the fan housing is fitted to the top of the cardboard carton by a similar 1-in wide 3½-in inside diameter ring of plastic. A 15-in length of 1-in aluminium angle is fitted along the outside of the complete collecting unit, being permanently fastened to the fan housing by a circular metal ring and to the collecting tube by elastic bands. The 3-in length of angle that extends from the back of the fan housing is riveted to a 24-in long handle of 1-in diameter aluminium tubing.

After mosquitoes have been collected the motor is kept running while the front tube is removed and a lid is placed on the cardboard carton, which is then removed and replaced with another. A quart-size carton or two 1-pint ones taped together can be used for larger catches, but increasing the capacity of the collection chamber decreases the suction power at the intake. The original aim was to collect mosquitoes resting amongst vegetation by slowly moving the apparatus backwards and forwards in a sweeping motion, hence its name. The motor, however, provides insufficient suction for this and the machine is normally used as a powered aspirator for collecting mosquitoes resting in shelters and traps. In Hawaii Tempelis *et al.* (1970) found this machine very useful in collecting mosquitoes, including blood-fed individuals.

Yamashita & Ishii (1977) described, together with a diagram and photographs, an aspirator for collecting insects which is based on the CDC sweeper of Hayes *et al.* (1967) but also somewhat resembles the aspirator made by Hall *et al.* (1968) to collect ants.

### *Suction sweeper of Garcia*

A long torch provides both a convenient handle and housing for five 1·5-V dry-cell batteries which are needed to power a small fan placed at the rear of a metal cylinder mounted on brackets beneath the torch. A small conical section forms the intake and is a sliding fit over the cylindrical chamber (Fig. 3.9*d*). A detachable fine muslin collecting bag is tied on to an inner metal tube projecting from the conical section. A metal cap covered with fine mesh is placed over the end of the intake to prevent mosquitoes escaping when the collecting bag is withdrawn from the cylinder. Mosquitoes are sucked into the collecting bag which is tied across with string and removed. I am indebted to Dr R. Garcia for showing me this suction sweeper.

### *Suction sweeper of Davies*

The following description of this powered sweeper which has not been published is based on information kindly supplied by Dr J. B. Davies. It consists of a modification of the suction trap described by Davies (1973). A wooden handle is attached to a 6-in diameter fan housing unit, which can conveniently be made from a 5-lb dried milk tin (Fig. 3.13*a*). A small electric motor with a three-bladed plastic propeller, such as sold for model aircraft, is mounted towards the rear of the metal housing unit. Mosquitoes are sucked up from vegetation into a 9-in diameter, 13-in long mosquito netting cage attached to the rear of the fan housing and tied to the wooden handle. A plastic foam flap which partially opens when the fan is operating falls back to prevent mosquitoes escaping from the netting cage when the motor is switched off. Leaves and large debris are excluded from the aspirator by a coarse wire mesh screen clipped over the entrance of the fan unit. The aspirator can be powered by a 6-V gel cell battery attached to the operator's belt. This suction sweeper has proved very useful in collecting mosquitoes from vegetation in Trinidad.

### *Davis & Gould's machine*

This small portable, battery operated aspirator was developed in Thailand to collect mosquitoes resting amongst vegetation (Davis & Gould, 1973). It consists of four basic parts, a collecting tube (A), a collecting chamber (B), a housing for the collecting chamber (C) and a fan unit (D) (Fig. 3.9*e*). The collecting tube consists of a 36-in length of 3½-in diameter plastic water pipe. Half-inch wire mesh can be taped over the entrance to prevent leaves and large insects being sucked up. This tube fits into a 3½-in diameter metal sleeve fitted to the hinged lid of the aluminium collecting chamber housing, which is 8 in in diameter and length. A 1-gal paper ice cream carton with bottom replaced with nylon netting and with an 18-in length of 6-in wide surgical stockinette taped around the other end forms the collecting cage and is inserted into the aluminium housing. The free end of the stockinette is slipped over the section of the collecting tube that extends about 4 in through the metal sleeve in the lid of the metal housing. Suction is provided by a 'squirrel cage' fan blower from a Volkswagen car heater unit. This is bolted through a gasket, made from a car inner tube to the bottom of the metal housing, which has a 3½-in diameter hole cut from the middle. The blower fan operates from a 6-V motor cycle battery which can be attached to a belt worn by the operator. This aspirator was successfully used to collect mosquitoes from a variety of habitats during a study of Japanese encephalitis in Thailand (Gould *et al.*, 1974).

This aspirator or a modified version was used in several studies in Pakistan by Reisen and his colleagues. For example, using it Reisen *et al.* (1982) caught 12 mosquito species resting in fields and 14 species resting in the forest, including *Culex tritaeniorhynchus, Culex quinquefasciatus, Aedes lineatopennis, Culex pseudovishnui, Culex fuscocephala, Aedes culicinus, Aedes indicus* and *Aedes yusafi*. Adults of all species were more common in forest collections, and in fact *Aedes yusafi* was collected only in the forest. In other studies the Davis & Gould (1973) aspirator caught *Culex tritaeniorhynchus* and a few adults of 11 other species from fields in Pakistan (Reisen *et al.*, 1978), while Reisen & Milby (1986) used the aspirator to collect more than 14 species of exophilic mosquitoes, the most common again being *Culex quinquefasciatus*, while *Culex pseudovishnui* was also common.

### *Suction sweeper of Kay*

Kay (1983) made an aspirator from a 1-m length of 10-cm diameter PVC drain pipe (Fig. 3.11). A 12-V motor drawing 37 W and a fan are powered by a 9-Amphr motor-cycle battery carried in a bag on the hip. The velocity of air through the intake is 5 m/s and mosquitoes are collected in terylene bags inserted into the intake of the PVC tubing. These bags can be secured with rubber bands and are removed every 15 min. This aspirator is similar to that of Davis & Gould (1973) but is simpler in that the enlarged holding cage is omitted. In Australia mosquitoes were collected from inside houses and from natural out of door resting places. In one site 21 591 mosquitoes, mainly *Culex quinquefasciatus*, were caught, the mean catch being 35·2 mosquitoes. In another site the total catch was 55 961 and the mean catch was 48·6 mosquitoes. Using this aspirator Kay (1983) collected some 7552 blood-engorged mosquitoes of 13 species, but mainly *Culex annulirostris* from 2119 collections from vegetation in sylvan and urban areas of Queensland.

### *Aspirator of Nasci*

Disadvantages of the D-vac machine are its weight, its noise, need for refuelling with petrol, and its cost. For these reasons Nasci (1981) constructed an aspirator from locally available materials that was some seven times cheaper, and weighed less than 5 kg. It consists of sheet metal (aluminium) rolled round to form a 61-cm

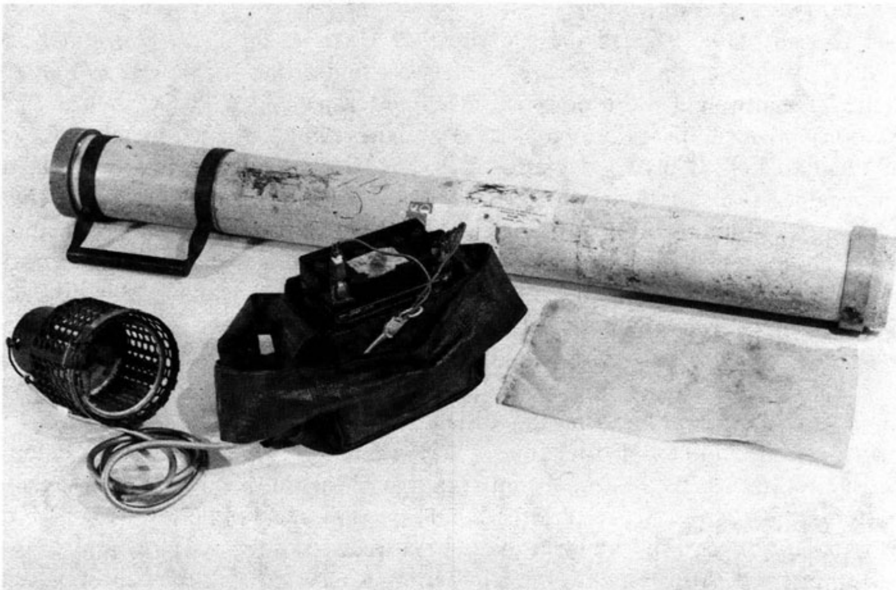

*FIG. 3.11. Mechanical aspirator of Kay (1983) (photograph courtesy of B. H. Kay).*

long and 34·3-cm diameter tube (Fig. 3.10*b*). A 12-V d.c. motor with a 25-cm fan blade is mounted on a 14-cm square piece of plywood screwed onto a 34·3-cm length of 4 × 10-cm strip of wood rounded at the ends to fit into the base of the metal tube. Transparent sheet plastic is rolled and bolted to form a 61-cm and 34·3-cm diameter tube which can be inserted 10·1 cm into the metal tube. A D-vac collecting bag is folded over the other end of the plastic tube and held in position by S-shaped clips attached to large rubber bands. Two metal gate handles are fixed on the metal cylinder. Wire from the motor passes to a push-button switch on the metal tube and then to two 6-V gel batteries connected in series, which will power the aspirator for at least 6 hr.

Using this type of aspirator Beier *et al.* (1982) collected 2220 female and 1127 male mosquitoes belonging to more than nine species from 15 daily collections of 5-min durations from 15 different areas in a wood. *Aedes vexans* formed 60·4%, and *Culex* spp. formed 23·9% of the catch. In Florida Nayar (1982) succeeded in using this type of aspirator to collect large numbers of male and female *Wyeomyia mitchellii* resting on trunks of oak trees. Also in the USA weekly sampling over 3 months from woodlots collected 841 *Aedes triseriatus* group mosquitoes (Nasci, 1982).

This machine has proved very successful in collecting a variety of mosquitoes from amongst vegetation, and appears to be one of the more widely used aspirators in the USA, although in some places it tends to have been replaced by the aspirator described next.

### *Suction sweeper of Meyer*

Meyer *et al.* (1983) found the aspirators of Davis & Gould (1973) and Nasci (1981) unsuitable, mainly because of their size and/or lack of power, so they designed a lightweight more powerful machine termed the AFS (Arbovirus Field Station) sweeper. Basically it consists of a lightweight L-shaped metal backpack adapted to hold a plywood platform (8 × 13 in) supporting two 6-V d.c. rechargeable gel cell batteries (Fig. 3.10*c*). These are connected in series to deliver 12-V at a minimum of 16 A, which is needed to operate the blower for about 1 hr. The blower is a 12-V Dayton model (2C646) and is mounted underneath the plywood platform on the horizontal section of the backpack. Air displacement of the blower is about 2·12 $m^3$/min, which represents improved suction over most previous battery operated aspirators.

The suction tube is composed of various sections of 4-in diameter black PVC piping (as used by plumbers and called ABS tubing), and about 4-ft of 4-in diameter automobile defroster hose, making altogether a tube about 8 ft long. Mosquitoes are sucked into a 1-pint sized paper carton having a nylon screened bottom which is inserted where the handle section and nozzle join (Fig. 3.10*c*). This collection carton can be rapidly removed, its lid placed on and a new carton inserted. A screen of ¼ or ½-in netting is inserted over the collecting tube to prevent leaves and twigs being sucked into the collection carton.

This aspirator was used in California to catch mosquitoes resting in various out of door shelters and amongst vegetation (Fig. 3.12) (Reisen *et al*, 1988).

*Fig. 3.12. AFS-sweeper in action (photograph courtesy of R. P. Meyer).*

Generally substantially more *Culex quinquefasciatus, Culex tarsalis, Culex stigmatosoma* and *Culiseta incidens* were collected from shrubbery and flower beds than from house eaves, porches, miscellaneous buildings and debris littering yards. Resting sites were characterised by high humidity and shade provided by trees or buildings. The numbers of *Culex quinquefasciatus, Culex tarsalis, Culex stigmatosoma* and *Culiseta incidens* caught by aspirators, and also in gravid mosquito traps, in different house compounds was very aggregated and mimicked a negative binomial distribution. In later studies in California vegetation was sampled with the AFS sweeper and with the more powerful D-vac machine, but these collections generally proved unrewarding, except when vegetation near breeding sites was sampled when large numbers of males and fewer newly emerged females of *Culiseta inornata* were caught (Reisen *et al.*, 1989). From 31 collections with the sweeper, mainly near breeding sites, 217 females ($\bar{x}$ = 7·0) and 12 150 males ($\bar{x}$ = 391·9) were collected, while from five collections with the D-vac 189 female ($\bar{x}$ = 37·8) and 1527 male ($\bar{x}$ = 305·4) *Culiseta inornata* were caught.

### *Perdew & Meek aspirator*

Perdew & Meek (1990) argued that their aspirator, made by modifying a commercial cordless 'broom', provides greater suction and requires less modification than many previous battery powered aspirators. The basic equipment is a blower-type broom that consists of a plastic body incorporating a fan and a motor, and in its handle rechargeable cadmium batteries, and an extension tube tapered and curved distally. To convert the broom into an aspirator the wires to the motor are reversed and the tapered end of the extension tube cut off. Two rigid plastic tapered beakers (474 ml) having a minimum 9-cm inner diameter at the top are required. To make an elongated version of the aspirator a selected length of the bottom of the plastic cup (a) is cut off to leave a 6·9-cm opening that can be glued to the inside of the extension tube (Fig. 3.10*d*). A 473-ml paper cup (b) has a 6·4-cm diameter hole cut from both its lid and its bottom, and both holes covered with screening of suitably sized mesh. This forms the collection carton, and without its lid, is pushed tightly into the plastic cup stuck in the end of the extension tube. This extended version of the aspirator is 118-cm long. A shorter (65-cm) version can be made by cutting the bottom from a similar plastic cup (c) to leave a 7·4-cm diameter opening. After two opposing rectangles are cut out (Fig. 3.10*d*) the cup is fitted over the end of the main body of the cordless broom and secured with four screws. The paper cup (collection carton) is fitted into the end of the plastic cup, the extension tube is not used. After using either the extended or short aspirator the screened lid is pushed on the paper cup which is then removed and replaced with another.

The aspirator can operate efficiently for about 45 min, with a minimum air flow of 21 $m^3$/min.

### *Miscellaneous aspirators*

In Kenya Chandler *et al.* (1975) collected at least 21 mosquito species, including 3237 blood-feds, from vegetation in the Kisumu area using battery powered aspirators (no description given). Using the same type of aspirators in other studies in a heronry near Kisumu, they caught considerable numbers of mosquitoes (8592 females and 2700 males of 25 species) from amongst grass and bushy vegetation beneath trees in which birds were resting (Chandler *et al.*, 1976). The most common species were females of *Culex antennatus* (37·2%), *Culex univittatus* group (18·6%), *Mansonia uniformis* (17·3%), *Aedes circumluteolus* (12·5%) and *Mansonia africana* (6·9%). Only three *Anopheles* were caught, the most common was *Anopheles pharoensis* (1·4%), while *Anopheles gambiae* s.l. and *Anopheles ziemanni* formed just 0·3 and 0·2% of the female catch. Unfed mosquitoes formed 54·6% of the catch of females, while blood-fed and gravid individuals formed 36·1 and 9·3%. Similarly in another series of catches in the heronry a variety of mosquitoes resting on vegetation beneath trees were collected by their battery powered aspirators (Chandler *et al.*, 1977). From 116 catches they collected 31 445 female and 21 732 male mosquitoes belonging to 30 species in eight genera, the principal species being *Culex univittatus* group, *Culex antennatus*, and *Aedes circumluteolus*. Blood-fed individuals formed 11·7% of the total female catch.

In Florida Day & Curtis (1989) monitored the abundance of *Culex nigripalpus* by undertaking two 10-min collections with battery powered aspirators from vegetation three times a week for 3 years. In some months more than 5000 females were collected. A time series analysis of the numbers caught and rainfall revealed a significant cross correlation between the numbers of blood-fed females and rainfall. In 1985 there was best correlation between engorgement rates 2–13 days after rain, but in 1986 trends were not so clear, and in 1987 the only positive association was on the day of rainfall. In Guatemala Cupp *et al.* (1986) collected *Mansonia titillans* from amongst grass growing at the base of trees with a suction sweeper. In Brazil Natal & Marucci (1984) constructed a simple 3-in diameter aspirator from PVC drainpipe operating from a 6-V battery attached to a belt from around the waist to collect mosquitoes resting either indoors or amongst vegetation.

Gary & Marston (1976) describe a mechanical aspirator for collecting insects from trees or dense undergrowth unsuited to sweep-netting. This machine is made by modifying a commercial 12-V, 120-W vacuum cleaner (14 000 rev/min) which is operated from a 12-V car battery pushed around in a folding golf cart.

### *Vehicle-mounted aspirators*

The following describes two vehicle-mounted aspirators developed in the 1960s for collecting mosquitoes, and Southwood (1978) refers to a few others that have been used to sample non-medical arthropods. However, such machines appear to have rarely been used in recent years.

Stern *et al.* (1965) described a power aspirator which was mounted on a three-wheeled vehicle originally manufactured for high clearance pesticide spraying. The machine was used to collect agricultural insects from alfalfa fields. Bidlingmayer & Edman (1967) were, however, the first to design vehicle-mounted aspirators specifically for collecting mosquitoes. The original paper should be consulted for detailed description and photographs of their two machines. In both models a rigid rectangular transparent intake unit some 8 in from the ground is mounted

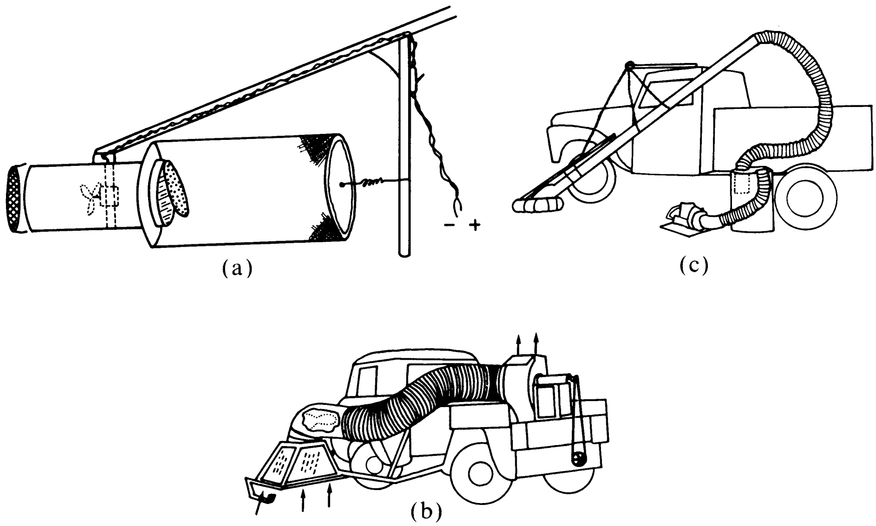


*FIG. 3.13. (a) Suction sweeper of J. B. Davies; (b) jeep-mounted aspirator (from Bidlingmayer, 1974); (c) truck-mounted aspirator of Rupp & Jobbins (1969).*

in front of a vehicle which is driven at 2–3 mph. An air displacement blower powered by the vehicle sucks up insects through the intake into a collecting bag fixed at the end of a screen cone. The larger of their two models is mounted on a jeep (Fig. 3.13*b*) and the smaller one on a 10 h.p. single-cylinder garden tractor. Although the air velocity through the 16-in diameter hose attached to the jeep-mounted aspirator is some 300–400 linear ft/min, suction at the air intake is nevertheless too weak to collect resting mosquitoes. Increasing the suction power to overcome this would also result in picking up considerable debris and leaf litter. Both models work on the principle that mosquitoes are first disturbed from rest by the approach of the vehicle, and are then sucked up into the aspirator. Short lengths of chain can be welded to the frame of the intake unit to help disturb mosquitoes resting among ground litter.

In Florida these machines have been used in pastures, pine woods and in hardwood hammocks; the tractor version is more suited to areas with soft soil, closely spaced trees and areas near buildings. Because freshly blood-fed mosquitoes are not readily disturbed Bidlingmayer & Edman (1967) considered that they would tend to be missed by the machines. However, considerable numbers of older blood-fed females which have regained some of their mobility are caught by these vehicle mounted aspirators. Edman (1971) found that powered aspirators were invaluable in collecting large numbers of blood-fed mosquitoes of many genera for determination of their host preferences.

Rupp & Jobbins (1969) developed a prototype of a vehicle mounted aspirator which they used to collect mosquitoes from undulating pastures traversed by deep ruts, where neither of the two machines of Bidlingmayer & Edman (1967) would have been able to operate. Their aspirator is powered by a 'Homelite Blower' (model 22B1) which displaces about 600 ft$^3$ air/min. It is connected by a 4-ft section of 5-in diameter flexible hose to a plastic-lined 30-gal fibre drum, in which there are two 5-in diameter openings (Fig. 3.13*c*). The one through which the hosing from the pump is inserted is covered with 20-mesh screening to prevent the catch being sucked through into the pump. A collection chamber is fixed within the drum beneath the other opening. This is connected to an 8-ft length of flexible hose attached to a 10-ft section of 5-in diameter aluminium tubing which terminates in the collecting head of the aspirator. This consists of a 22-in diameter piece of ½-in thick plywood with a 6 × 7-in intake hole covered with ½-in mesh to exclude unwanted debris. An inflated 15-in diameter car inner tube is attached by cords underneath the plywood disc to serve as both a shock absorber and an enlarged extension of the suction line. The pump and fibre drum are placed in the back of a ¾-ton four-wheeled drive pick-up truck, and the metal tubular intake is suspended alongside the driver's cab by chains fixed to an 8-ft length of angle iron mounted on top of the cab. An operator stationed in the back of the truck collects mosquitoes by moving the aspirator from side to side over the ground in a sweeping motion. This is done by pulling on a cord tied to one of the chains suspending the aspirator. Another operator presses down on the end of the aluminium tubing to lift the aspirator head clear of the obstacles.

Rupp & Jobbins (1969) list some of the improvements they consider are needed to make the aspirator more efficient. One of the requirements is that the collection chamber is removed from the drum and placed nearer the collecting head, as in the models developed by Bidlingmayer & Edman (1967).

No details are given of the mosquitoes caught by this machine in trials in New Jersey, except that both unfed and blood-fed individuals were collected from pastures from which they apparently would not have been collected by sweep-netting.

## ARTIFICIAL RESTING PLACES

Because of the difficulties usually encountered in locating outdoor mosquito populations which are often distributed over wide areas of vegetation, specially constructed artificial shelters have been used to try to attract mosquitoes to specific sites from which they can conveniently be collected. This idea is not new. Nuttall & Shipley (1902) appear to have been the first to have suggested that traps could be used to attract and catch resting adults. As a result various types of box-like shelters were used in both in and outdoors by later workers in efforts to control mosquitoes (see Service, 1976 for references). It was not until Russell & Santiago (1934) constructed their earth-lined box trap that it was fully appreciated that artificial resting places could be used to study and sample exophilic mosquitoes.

Since then several different types of artificial resting shelters have been made and evaluated, the variety of which depends much on the ingenuity of the entomologist. Some, such as the box shelters of Edman *et al.* (1968), have been used mainly to collect specific species or genera, such as *Culiseta melanura*, whereas others, such as pit shelters, are used to collect a broader range of species and genera. Loomis & Aarons (1954) coined the term 'artificial resting unit' to differentiate between an artificial shelter specifically made to collect mosquitoes and other man-made constructions such as bridges, and culverts which are used as resting sites by mosquitoes. This term has not, however, generally been adopted.

## Keg shelters

Field observations in the Tennessee Valley area of the USA showed that in general more *Anopheles quadrimaculatus* entered unbaited traps with the door left open than baited traps with a closed door (Smith, 1942). This together with the discovery that adults readily entered empty small box-like structures led to the development of the keg shelter as an artificial diurnal resting site for *Anopheles quadrimaculatus*. The shelter consists of an ordinary, untreated wooden barrel-shaped nail keg with the lid removed, placed on its side on the ground in a deeply shaded area, especially near the edge of larval habitats. Its entrance should be protected from wind and the sun by siting the keg at the base of a tree, alongside or under a log, under bushes or amongst scrub vegetation. In nine collections from a keg placed underneath a fallen tree Smith (1942) collected as many as 4006 *Anopheles quadrimaculatus*. On another occasion 1129 adults were caught from a single keg. Kegs also attracted large numbers of *Anopheles punctipennis*. Both males and females in all stages of gonotrophic development were collected from the kegs whereas collections from barns comprised mostly freshly blood-fed females. Smith (1942) considered that the kegs accurately reflected changes in the population size of *Anopheles quadrimaculatus*. He realised that in areas with patchy vegetation mosquitoes congregated into relatively small areas, and only one or two kegs were needed to sample the population. Whereas in areas of more uniformly dense vegetation adults were more evenly distributed, and more kegs were needed to adequately sample the resting population. Kegs, like other artificial resting shelters, have to compete for attractiveness with natural resting sites. Unsuccessful attempts were made by Smith (1942) to try and make the kegs more attractive by adding either raisins, small quantities of dry ice, ammonia or sucrose and by painting their insides black or lining them with earth or black cloths.

It is now very difficult to obtain wooden nail kegs. Because of this and the effectiveness of other simple and cheaper shelters (see below) kegs are now little used as artificial resting sites.

## Small red box shelters

Goodwin (1942) reported that nail kegs were not successful in Georgia, USA, for collecting *Anopheles quadrimaculatus* so he experimented with various wooden boxes as alternative resting sites. Boxes open at one end and measuring 1 $ft^3$, or 1 $ft^2$ in cross section, and 2 ft deep caught more *Anopheles quadrimaculatus* than smaller boxes, and more adults were caught in a 1-$ft^3$ box placed below 6 ft than in those placed higher. Boxes facing east, that is towards the rising sun, always caught fewer mosquitoes than those whose opening faced other directions. Goodwin (1942) found that the mean catch of *Anopheles quadrimaculatus* was greater in boxes painted red inside (24·13) than those painted white (0·04), yellow (0·28), blue (0·43), black (12·79) or green (0·47). Moreover, mosquitoes were more easily seen and collected from red boxes than from black ones where torches were sometimes needed. It was concluded that the best shelter was a 1-$ft^3$ wooden box painted dull black on the outside, red inside and positioned on the ground in a sheltered position, preferably not facing east. In comparative trials these red boxes were on average about 2·8 times more attractive than nail kegs (Goodwin, 1942).

Red boxes were used by Zukel (1949*a*) to collect *Anopheles quadrimaculatus* in Georgia and by Breeland (1972*a*) to collect *Anopheles albimanus* and *Anopheles pseudopunctipennis* in El Salvador. Both Burbutis & Jobbins (1958) and Moussa *et al.* (1966) found they were useful in collecting *Culiseta melanura*, and in fact could replace light-traps for monitoring seasonal changes in population size of this mosquito (Burbutis & Jobbins, 1958). Boxes on the ground facing approximately due west caught more mosquitoes than those facing other directions, or placed at a height of about 4 ft. Few mosquitoes were found in boxes inspected between 0600–0700 hr. Burbutis & Jobbins (1958) considered that most entered between 0800–0900 hr. In addition to *Culiseta melanura*, adults of *Anopheles crucians, Anopheles quadrimaculatus, Anopheles punctipennis, Culex salinarius, Culex restuans, Culex pipiens, Aedes canadensis, Aedes sollicitans, Coquillettidia perturbans*, and *Uranotaenia sapphirina* were caught in the boxes. Loomis & Sherman (1959) found that the boxes were very useful for collecting and measuring population changes of *Anopheles freeborni* and *Culex tarsalis* in California. In fact both they and Bradley (1943) caught more *Anopheles freeborni* in red boxes than in light-traps. Hayes *et al.* (1958) also caught large numbers of *Culex tarsalis* in box shelters.

Although Carpenter *et al.* (1946) reported better catches of mosquitoes in light-traps than in the resting boxes of Goodwin, Gusciora (1961) in 33 comparable tests in New Jersey, caught 13 240 mosquitoes in box shelters but only 6260 in light-traps. A ¼-in mesh screen was placed over the openings of the box shelters to reduce the large numbers of tipulids that sought shelter in them. Collections were made by placing a cloth net bag with elastic around the opening over the entrance of the box and spraying with chloroform. Gusciora (1961) suggested that increased catches might be obtained if ½-in thick plastic foam pressed into 3-in deep folds was wetted and tacked to the floor of the box, so as to increase humidity and the resting area available for mosquitoes. In India Yasuno *et al.* (1973*a*) did in fact place a sponge soaked in water in boxes placed inside houses to attract endophilic *Culex quinquefasciatus*.

Pletsch (1970) reported that the red boxes of Goodwin were useful in El Salvador for collecting *Anopheles*, but as they were very bulky and cumbersome he developed a collapsible box shelter made of ¼-in plywood. The four sides (12-in square) are joined together with strips of canvas, leather or plastic, stapled or nailed to the sides. The back of the box (12 × 13½ in) is joined to the bottom by a canvas hinge, and a transverse strip of wood (12 × 1½ × ¾ in) fastened to

*FIG. 3.14. Walk-in red box (M. W. Service).*

the inner face ½ in from the top gives rigidity to the box when the back is folded into position. This back panel is held in position with string or cord. Sixteen or more of these traps can be transported or stored in the space required to stack two conventional red boxes of Goodwin.

## Walk-in red boxes

In California Nelson & Spadoni (1972) developed much larger (6 ft tall, 4 ft wide, 6 ft deep) boxes painted red inside and out and called walk-in red boxes. These were later modified by Meyer (1985) to have a curtain that can be pulled across the entrance when a person enters to aspirate the catch (Fig. 3.14). These generally trap more mosquitoes than the smaller red boxes having 1 $ft^2$ entrances.

In California Reisen *et al.* (1983) compared the numbers of male *Culex tarsalis* caught in standard (0·3 × 0·3 × 0·03 m) red boxes (Goodwin, 1942) and larger (2 × 1 × 2 m) walk-in red boxes with those caught in CDC light-traps, truck traps and those caught from swarms. The red boxes, especially the larger ones, caught the most males, and were considered the best method for monitoring their seasonal abundance. These boxes have been used to sample other mosquitoes such as *Culex quinquefasciatus* and *Culex stigmatosoma* (Reisen *et al.*, 1990), and *Anopheles freeborni* (McHugh, 1989). In California 30 827 male and 22 813 female *Anopheles freeborni* were collected from 33 daily collections from 15 walk-in red boxes (1·8 × 1·2 × 1·3 m). Blood-fed females formed 19·6% of the catch, but most females (75·3%) were unfed (McHugh, 1989). Nelson *et al.* (1978) found that Goodwin (1942) type red boxes and larger walk-in ones sited near breeding sites caught almost exclusively newly emerged *Culex tarsalis*, whereas older adults were collected by carbon dioxide-baited light-traps. Possibly older mosquitoes used other resting sites, or those further away from the emergence sites.

Reisen *et al.* (1989) made concerted efforts to collect out of door resting *Culiseta inornata* in California. From 314 collections from 1-$ft^3$ red boxes 190 female ($\bar{x}$ = 0·61) and 30 male ($\bar{x}$ = 0·10) *Culiseta inornata* were obtained, compared to 867 females ($\bar{x}$ = 0·49) and 846 males ($\bar{x}$ = 0·47) from 1784 collections from walk-in red boxes. In this instance the larger boxes were no better in collecting *Culiseta inornata* than the smaller ones.

McNelly & Crans (1982) rightly emphasised that catches from different walk-in red boxes should be recorded separately for statistical tests, and not all box catches in an area combined. This advice, however, to record catches separately, applies to virtually all sampling methods.

## Red cloth shelter

Breeland & Glasgow (1967) devised a cheaper, lighter and more portable version of the red box shelter of Goodwin (1942). A 52-in length of red broadcloth which is 36 in wide is folded down the middle and stitched across at each end, so that when the material is cut between the middle seams two sacks are formed. Each sack is stapled to a 1-$ft^2$ unpainted wooden frame which is nailed to a tree (Fig. 3.15*a*). The sack is held out horizontally by tying the ends with string to a

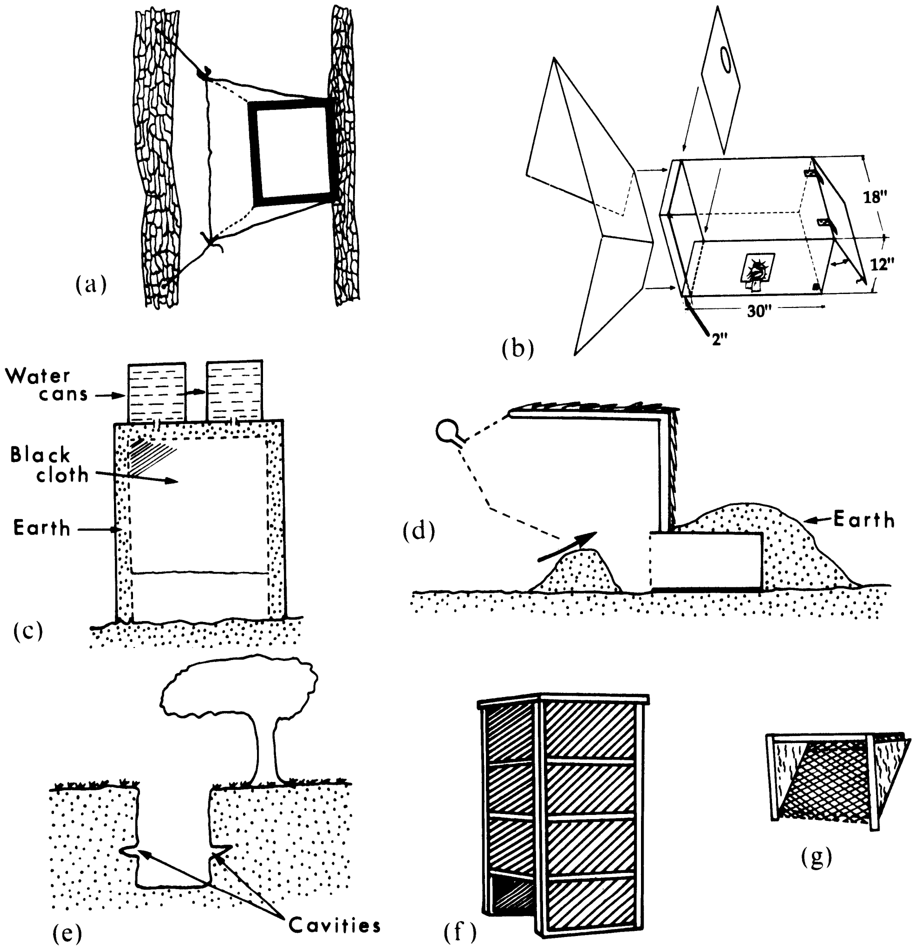


*Fig. 3.15. Artificial outdoor resting shelters: (a) Red cloth shelter of Breeland & Glasgow (1967); (b) resting box of Edman* et al. *(1968); (c) earth-lined box shelter; (d) metal drum-type shelter (after Vale, 1971); (e) Muirhead-Thomson pit shelter; (f) privy-type shelter; (g) ingress baffle fitted to privy-type shelter of Snow (1949).*

convenient tree or stake. In two separate series of trials in Alabama these red cloth shelters caught significantly more *Anopheles quadrimaculatus* than the conventional red boxes.

## Resting box of Edman

This resting box was developed by Edman *et al.* (1968) specifically for collecting blood-fed adults of *Culiseta melanura* in Florida. The final version of their trap consisted of a plywood box 30 in long, 18 in wide and 12 in high, painted matt grey on the outside and matt black inside. A red cotton collecting bag slightly smaller in width and height than the box but 8 in longer was put in the box and held in position by placing elastic loops sewn at the bottom four corners over hooks screwed into the bottom of the box. The open end of the cloth bag which projected from the box was folded back over its opening, and secured by elastic tape threaded through its end seam. A plywood concave frame, painted matt grey outside and matt black inside, with a 28 × 52-in opening tapering to a 10 × 16 base was placed just inside the opening of the box (Fig. 3.15*b*). Mosquitoes were collected from the box shelters by first carefully withdrawing the concave frame and then pulling the turned over end section of the collecting bag from the box and closing it.

Boxes without a cloth bag caught similar numbers of mosquitoes as those with the bag, but it was more time consuming to remove them with an aspirator than by removing the entire catch within the bag. Larger numbers of *Culiseta melanura*, especially males, were caught in boxes facing west than east. Boxes with a concave frame caught about 44 mosquitoes per day, whereas those without a frame caught only about nine per day but the proportion of blood-fed females increased from about 11 to 30%. However, because of the larger catch the absolute numbers of engorged females was higher in the boxes having a concave frame. It appeared that when the frame was positioned in the entrance of a box mosquitoes were attracted from a greater distance. Most *Culiseta melanura* entered the boxes before 0830 hr, but there was some flight activity during most of the day. Optimum collection time was influenced by weather conditions; in general, the drier and windier the weather the earlier adults entered the boxes. In addition to *Culiseta melanura*, more than 14 other mosquito species were collected from the boxes.

Nasci & Edman (1981*b*) used these resting boxes to collect *Culiseta melanura*; from one survey comprising just four nights 177 adults were caught, about 15% of which were blood-fed. In more prolonged collections the boxes were placed with their open-bottoms on damp soil at the bases of trees, with their openings facing west to avoid the morning sun. Several thousand blood-engorged *Culiseta melanura* were caught over 2 years from 10 boxes (Nasci & Edman, 1981*a*). Anderson *et al.* (1990) also readily caught blood-fed adults of this species in similar boxes. Later in Massachusetts, Nasci & Edman (1984) caught 12 094 female *Culiseta melanura* from 10 resting boxes. The mean number of females per trap night varied from less than 10 to about 75; blood-engorged adults ranged from less than 10% to about 30%.

### Resting box of Morris

A great variety of very simple (Goodwin, 1942) and more complicated (Edman *et al.*, 1968) resting shelters, including walk-in red boxes (Meyer, 1985; Nelson & Spadoni, 1972) have been used. It is difficult, however, to compare their efficiencies because they have been operated in different areas to collect different mosquito species, and have varied in colour and in location. Morris (1981) therefore undertook experiments in New York to study the effect of size and shape, location, and colour on mosquitoes caught.

In his experiments the standard shelter was a 30-cm wooden box painted matt black. Experimental designs included 10-, 20- and 40-cm cubes, the 30-cm box with the front opening partially closed, a box partitioned with one or two vertical or horizontal partitions, large boxes (20 cm high, 40 cm wide, 20 cm deep) on 10-cm high legs, and a less deep box (10 × 40 × 20 cm) on 20-cm high legs. Boxes were made of plywood or masonite, painted black, red, blue, brown or green, and had their openings facing in different directions. Morris (1981) concluded that a shelter 30 cm high, 40 cm wide and 20 cm deep caught about the same numbers of mosquitoes (*Culiseta melanura, Culiseta morsitans, Anopheles punctipennis, Anopheles quadrimaculatus* and *Culex territans*) as the standard 30-cm cube box, but smaller shelters caught fewer mosquitoes. Making boxes from masonite reduces cost and weight, and prevents damage by plywood-eating porcupines. There were no differences between the numbers caught in shelters painted black, red, brown, blue or green, but shelters painted white, grey or partially white and black attracted fewer mosquitoes. Presence of vertical or horizontal partitions had no effect on the catch. West-facing shelters consistently had more mosquitoes than east-facing ones when collections were made in the morning, but in the evenings there was no such difference. Obviously catches are increased by employing more shelters, but (Morris, 1981) considered they should be placed at least 1·5 m apart.

Morris *et al.* (1980) placed his shelters 3 m apart with one of the longer sides on the ground and the entrance facing west. Trapped mosquitoes were killed by placing a chloroform-soaked pad in a shelter and closing it with a lid. From 640 shelter-days in New York 1403 female *Culiseta melanura* were collected. Depending on the collecting area, means of 32, 36 and 42% were parous, while blood-fed and gravid females formed 38·5, 52·0 and 84·9% of the catches. Later Howard *et al.* (1989) found these resting shelters useful in collecting unfed, blood-fed, gravid and males of *Culiseta melanura* and *Culiseta morsitans* in their mark–recapture studies on flight range and dispersal.

### Resting box of Weathersbee

Although box-type resting stations of Edman *et al.* (1968) have proved useful in collecting *Anopheles quadrimaculatus* (Weathersbee *et al.*, 1986), they were considered cumbersome and rather expensive to make, consequently a more portable and cheaper resting unit was designed (Weathersbee & Meisch, 1988). It consists of a 113·5-litre plastic refuse container (72·3 × 41·9 × 80·6 cm) placed on its side on the ground, to the front entrance of which is attached a tent-like 3-ft extension made from a sheet of black plastic (3 × 5 ft) (Fig. 3.16). The narrower end was fixed with waterproof insulating tape to the refuse box, while cord threaded through holes along the ridge of the plastic tent and its two sloping ends (all reinforced with insulating tape) were attached to a centre 3-ft dowel. Two large nails passed through loops in the cord from the sloping ends and secured the canopy to the ground.

The mean numbers of *Anopheles quadrimaculatus* per day per box ranged from 118·3 ± 22·8 to 347·3 ± 93·4 males, and from 107·8 ± 27·1 to 227·5 ± 46·1 females. Numbers compared favourably with mean catches per day of 316–423 of the same species recorded by Snow (1949) from privy-type shelters. These boxes of Weathersbee & Meisch (1988) were used in later studies, again to collect *Anopheles quadrimaculatus* (Weathersbee & Meisch, 1990).

*FIG. 3.16. Resting box of Weathersbee & Meisch (1988) (photograph courtesy of A. A. Weathersbee).*

## Resting box of Kay

Kay (1983) used brown cardboard cartons (33 × 27 × 25 cm) which had the top flaps folded and tapered to form an ingress aperture (Fig. 3.17) as out of door resting boxes in Australia. The boxes were stacked in groups of 10–25 on top of each other. In the dry season wet cheesecloth was sometimes introduced into the boxes, while during the wet season they were sheltered against rain. In one site the mean catch was 26·3 mosquitoes and in another just 4·1. The most common mosquito species collected was *Culex quinquefasciatus*.

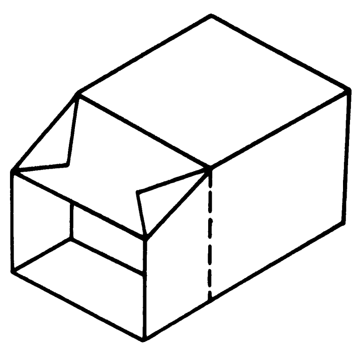

*FIG. 3.17. Cardboard resting box of Kay (1983) (figure courtesy of B. H. Kay).*

## Resting box of Charlwood

In Papua New Guinea Charlwood *et al.* (1985) reported that at dusk they caught *Anopheles farauti*, in all gonotrophic conditions, in a portable resting trap. This consisted of a 2-m long, 1-m wide, 1·5-m high wooden frame covered with black plastic with an exit trap fitted on one side and another on top. No mosquitoes were caught in the trap during the day.

### Rolled up mattresses

Khin (1964) reported that in Myanmar during residual house-spraying programmes it had proved difficult to collect out of door resting *Anopheles culicifacies* until artificial resting shelters were made by rolling bamboo mattresses (6 × 4 ft) into hollow cylinders. These were stood vertically on the ground and the open tops covered with thatch. From 21 man-hours 535 *Anopheles culicifacies* were collected from these artificial shelters.

### Plastic dustbin bags

In very dry regions of Dubai with scant vegetation I made resting shelters by placing black dustbin (trashbin) bags over 1-ft cube wire frames, leaving the bag partially open to form an entrance (Service, 1986). Cotton wool soaked in water was placed in the bag to increase humidity. These shelters were placed in position at 1700 hr and inspected the following day at 0800 hr. From 22 trap-nights 84 male and 264 female *Aedes caspius* were collected, 12·9% of the females were blood-fed, and from 44 trap-nights 614 males and 1745 female *Culex quinquefasciatus* were obtained, of which 12·6% were blood-fed. Although in wooded areas in England no mosquitoes were caught in these very simple and cheap artificial resting shelters they nevertheless deserve further evaluation.

### Earth-lined box shelters

Earth-lined boxes were first used by Russell & Santiago (1934) in the Philippines to collect *Anopheles flavirostris* and other *Anopheles* species. Their trap consists of a 3-ft long and 2-ft square, or larger, wooden framework with a 1-in thick layer of soil held in place against the inside walls and roof by 16-mesh/in screening. There is no bottom to the trap, which is placed directly on the ground. A black cloth hangs down over the entrance to within about ½ ft of the ground. Drip cans full of water keep the earth lining the box moist (Fig. 3.15*c*). When similar boxes without an earth lining were used the average catch of mosquitoes per night per box was 4·4–12·4, whereas in earth-lined boxes an average of 18·8–69·5 mosquitoes were caught. Further, none or very few *Anopheles flavirostris* were caught from the unlined boxes. Placing a light in the earth-lined box produced a maximum mean catch of 137·3 mosquitoes per night but no adults of *Anopheles flavirostris* entered the boxes.

In India Rao (1984) collected 'reasonable numbers' of *Anopheles culicifacies* from earth-lined wooden boxes placed out of doors.

In studies on the outdoor resting populations of *Anopheles gambiae* and *Anopheles funestus* in a humid sub-coastal belt of Tanzania Gillies (1954) used a modified version of the earth-lined shelter of Russell & Santiago (1934). His box consists of a 3-ft long, 3-ft high and 2-ft wide wooden frame covered with plastic mosquito gauze, with a black cloth hanging down from the front entrance to leave a 6–8 in entry gap underneath. These boxes are buried in a suitably shaded earth bank, and mosquitoes collected from them early in the morning. None of these boxes caught large numbers of *Anopheles*, the mean catch varied from 0·3–3 *Anopheles* per shelter per day, although occasionally up to 20 females were collected from a shelter. A comparison of the mosquitoes caught in these artificial shelters with those from natural outdoor resting sites showed a similar composition of unfed (32·7%), blood-fed (7·5%) and gravid (59·8%) individuals of *Anopheles gambiae*. In contrast a higher proportion of unfed females of *Anopheles funestus* were caught from natural (80·0%) than artificial shelters (40·3%). Exophily in *Anopheles gambiae* was also studied in an arid area of Tanzania which was interspersed with large swamps and irrigation areas. Cattle were more abundant in this area than the coastal region and larval habitats were larger but more concentrated. During peak populations 'well over 3000 *Anopheles gambiae*' were caught from 23 box shelters over a 10-day period (Gillies, 1956). Occasionally over 100 *Anopheles gambiae* were collected from a single shelter. A considerably high proportion of the females caught in these shelters were blood-fed than in those in the coastal areas. This apparent higher degree in exophily was probably due to the combination of a larger mosquito population, a scarcity of suitable natural resting sites and the greater availability of cattle in the area (Gillies, 1956).

Gillies (1954) thought that the box shelters sited near houses might compete with them in offering suitable alternative resting places, and in fact contain some adults which might otherwise have entered houses. A comparison of the ratio of half-gravid:gravid females caught in boxes placed near houses and in boxes some distance from houses with the ratio of half-gravid:gravid adults caught in houses did in fact indicate that boxes near houses in part reflected the endophilic population. Moreover, the construction of a hut near a box shelter caused a very marked reduction in the numbers of mosquitoes resting in it. It was concluded that whereas catches of mosquitoes resting in shelters away from houses gave a valid sample of the exophilic population, those collected from boxes placed near houses might not do so.

As with other artificial resting shelters the numbers of mosquitoes caught in these box shelters is greatly influenced by their location, with the result that identical boxes often catch greatly different numbers of mosquitoes. But, whereas portable resting shelters can be easily moved around so that locations giving the highest catches can be selected, the box shelters of Gillies are not easily resited. To overcome this difficulty portable mud-lined boxes similar to those of Russell & Santiago (1934) were used in Nigeria (Service, unpublished data). They were 3 ft long and 2 $ft^2$ in cross-section and covered on the inside with plastic mosquito gauze but unlike the original earth-lined boxes they had a bottom. Earth was packed on all four sides and bottom in between the 2-in thick wooden framework and held in place against the mosquito gauze by plywood panels nailed to the box. A black cloth hung down over the entrance to leave a gap of about 6 in. Water was sprinkled through the gauze sides of the box to dampen the earth and the boxes sited in shaded places in village compounds. During the wet season in Nigeria a mean of 4·7 female and 1·9 male *Anopheles gambiae* and 2·2 female and 2·6 *Anopheles funestus* were collected per night from eight such boxes. In the dry season the mean catch was reduced to 1·4 female and 0·4 male *Anopheles gambiae*, and 1·1 female and 0·8 male *Anopheles funestus*. The maximum overnight catch was 37 female *Anopheles gambiae* from eight boxes, recorded in the wet season.

Some of the difficulties that may be experienced in using these artificial resting sites includes theft of the black cloth hanging over the openings of the boxes, and the destruction of the boxes by termites, goats and vandalism, and the building of wasp nests in them.

## Pipe traps

Nelson (1980) developed an artificial rodent burrow termed the pipe trap for collecting *Culex tarsalis*. The device consists of three parts, a pipe, a plunger and a collecting carton (Fig. 3.18). The pipe consists of a 91-cm length of 18·5-cm outside diameter asbestos-cement sewer pipe, both ends of which taper to 16·2 cm in diameter. The pipe absorbs moisture. The plunger is a 91-cm length of 4·76-mm steel rod having a 14-cm wooden disc fixed at the end, to which is glued a piece of 16·5-cm diameter foam rubber. The collecting component consists of a 16·5-cm long, 3·8-litre cardboard carton. The bottom is removed and a 30·5-cm length of tubular stockinette is taped over the outside of the carton. The other end (top) is removed or modified so that the sleeve section of a CDC collecting bag is fitted over the end. Alternatively a sleeve of stockinette or mosquito netting is fixed over the top end of the carton.

A hole is dug in the ground (by a post-hole digger if available) and the pipe inserted at an angle of 20–25° from the horizontal so that about 7·5 cm projects from the hole. The piston-like plunger is pushed down to the bottom of the pipe. The following day any mosquitoes that have rested in the pipe are removed, by firstly fitting the stockinette sleeve on the bottom of the carton over the end of the pipe. The collector then reaches through the top sleeve and slowly pulls up the plunger, while squeezing the top sleeve to prevent mosquitoes escaping. When the foam rubber disc is flush with the end of the pipe the stockinette tube is eased off the pipe and pinched off across the face of the disc. The carton is then slipped off from the rod (Fig. 3.18) and both ends tied. The trap is then reset by pushing the plunger down to the bottom of the pipe. Rodents sometimes enter pipe traps, and possibly scorpions and snakes may enter them, so caution may be needed in withdrawing the plunger.

In California Nelson (1980) collected a mean of 13·8 male and 11·2 female (unfeds, blood-feds and gravids) *Culex tarsalis* per pipe compared to means of 28·8 and 24·6 from 1-ft red box shelters (Goodwin, 1942). Other species resting in the traps were *Culex quinquefasciatus, Culiseta inornata* and *Anopheles franciscanus*. However, Reisen *et al.* (1989) found these traps ineffective in their studies, as only one male and one female were collected from 160 collections, but then only a single female was collected from 100 collections from cone traps fitted to rodent burrows, whereas previously such traps had caught considerably more *Culex tarsalis* (Reisen *et al.*, 1985).

It seems that pipe traps will prove useful in other areas of the world, especially in hot dry regions, for a variety of mosquito species that normally rest in rodent burrows and other holes, or in cracks and crevices in the ground.

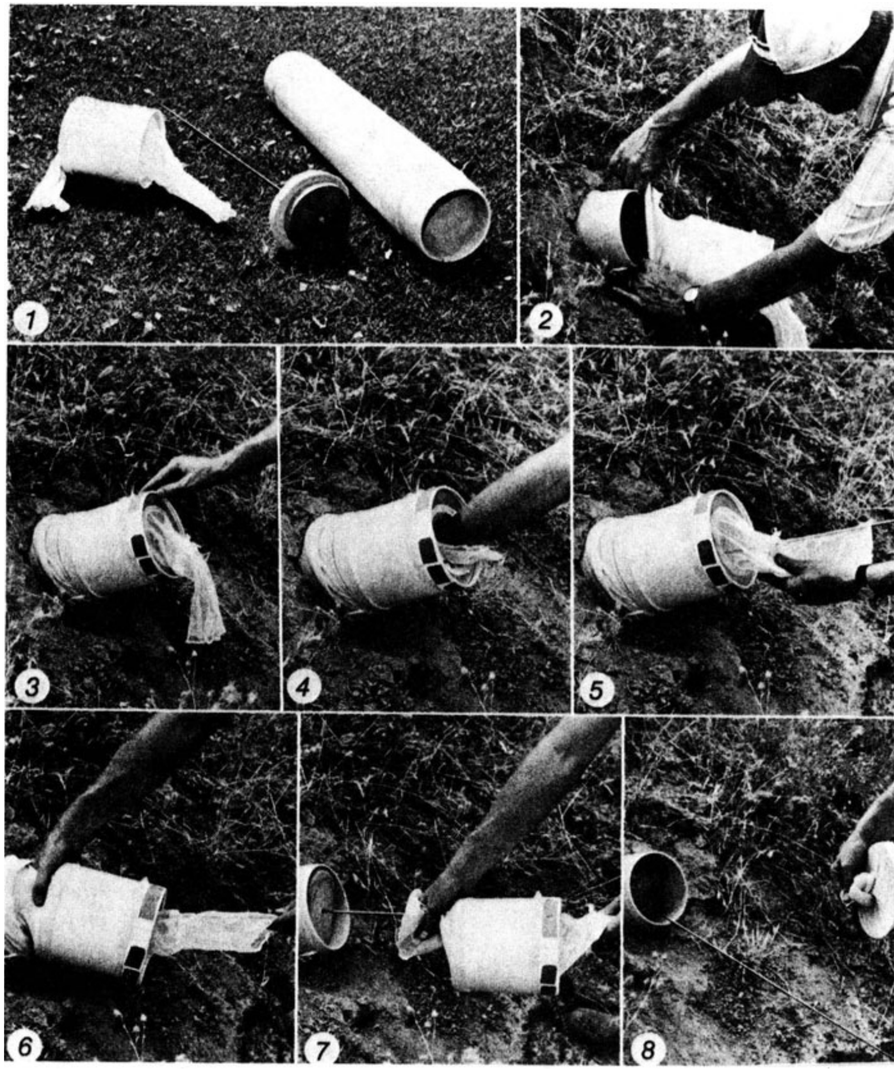


*FIG. 3.18. Pipe trap: 1 — showing components, 2–8 — step by step processes of removing catch (Nelson, 1980).*

**Metal drums, village pots, etc.**

Empty petrol drums (Laarman, 1959), village clay pots and other containers may also serve as useful resting places if they are buried in an earth bank or partially covered with a pile of earth. Vale (1971) described a useful resting place for the tsetse, *Glossina morsitans*, that he used in Rhodesia, and which might prove attractive to mosquitoes. It consists of a metal drum covered in earth, and a box with a thatch roof positioned over the entrance (Fig. 3.15*d*). Dichlorvos (DDVP) strips were placed in a small flask to kill the catch.

### Pit shelters

At the beginning of this century Blin (1908) described how *Anopheles* could be caught, and he considered controlled, by digging small holes in the ground, but it was many years later that Muirhead-Thomson used a similar method to sample outdoor resting mosquitoes. In Tanzania Muirhead-Thomson (1951) failed to collect any *Anopheles gambiae* or *Anopheles funestus* resting on natural earth banks, but discovered that if horizontal pits or channels were dug into them, unfed, blood-fed and gravid females of both species sought refuge in these dark niches. He also created attractive resting places for both species by undercutting the wall near the bottom of an abandoned excavation pit. These observations led to the development of artificial pit shelters (Muirhead-Thomson, 1958).

These pits are 5–6 ft deep and if possible are dug under trees or large bushes so that their openings (4–5 × 3–4 ft) are shaded from above; failing this a suitable cover should be placed partially over the pit entrance. About 1½–2 ft from the bottom of the pit a small cavity, about 1 ft deep, is dug out horizontally from each of the four sides (Fig. 3.15*e*). Mosquitoes are collected from both these small cavities or from the wall of the pit itself. It is advisable to encircle the pits with a thorn or fence enclosure to prevent cattle or young children falling into them, or them being used as toilets. In Tanzania four pit shelters sited 30–150 m from a village yielded, after about a month, 674 male and 626 female *Anopheles gambiae* complex and 130 male and 150 female *Anopheles funestus*. This represented just 20·8 and 4·6%, and 20·0 and 7·6%, respectively, of the total catches of females and males of these two species collected from inside houses and in pit shelters combined. No anophelines were caught by searching vegetation and earth banks, nor in box-type traps made from tea chests (Lines *et al.*, 1986). Muirhead-Thomson (1958) found pit shelters very useful in Zimbabwe for collecting *Anopheles gambiae*, the *Anopheles funestus* group and also several other *Anopheles*. More recently in Zimbabwe Mpofu (1985) found that pit traps caught both *Anopheles quadriannulatus* and *Anopheles funestus*, 87·9% of the latter found in these traps were blood-fed. Nevertheless, only 589 *Anopheles funestus* and 404 *Anopheles quadriannulatus* were collected from 12–13 pits inspected on 14 days/month for a year. Although D. A. Muir found them useful in Java, they failed to attract mosquitoes in Sarawak (Graham, 1969). Muirhead-Thomson was little troubled with problems of the pits becoming flooded, but I have found that in both Kenya and Nigeria most pits become inoperative each rainy season because of flooding. This can be difficult to prevent. The erection of suitable roofing over the pits may help to keep out rainwater, but it does not alleviate flooding by seepage water. Pits are not easily dug or maintained in areas where the soil is either rocky or sandy.

In India Shalaby (1971, 1972) dug pit shelters which were 160 cm deep, 130 × 120 cm wide and had 30-cm deep pockets dug in all four walls about 50 cm from the bottom of the pits. Whenever possible these were located under a tree, but when not, shade was provided by erecting thatch roofs over the pit entrances. These shelters proved very successful, a total of 12 *Anopheles* species were collected from them. Also in India Mani *et al.* (1984) dug pit traps (1 × 1 × 1 m) in riverine villages and over 2 years caught 12 *Anopheles* species, the most common being *Anopheles varuna, Anopheles subpictus* and *Anopheles culicifacies*, the maximum number of any species caught was 30·61 *Anopheles varuna*/10 trap-nights during July, 1981. The pits were made more attractive to mosquitoes by watering them the evening before collections were made. Later 2836 female *Culex vishnui* subgroup mosquitoes were collected from larger pit shelters (4 × 4 × 4 ft) having 6-in deep brick-lined recesses dug into the sides at 2 ft from the bottom of the pits (Anon, 1988; Mani & Devaputra, 1988). The pits were shaded by bushes or by a thatch roof supported on poles. Despite problems of pits being used as latrines and flooding they were considered very useful, because from 177 trap-nights they attracted 2836 blood-engorged mosquitoes which were used in host identification studies. The mean catch/trap-night over all months was 16·02; in October a mean of 86·32 female *Culex vishnui* subgroup was recorded. As the bricks lining the recesses held moisture for a long time they were considered useful in keeping these refuges damp.

In Gujarata state, India Bhatt *et al.* (1989) caught 4998 anophelines belonging to 10 different species and 1855 culicines from 20 pit shelters (1·2 × 1 m, and 1·5 m deep) which had 15-cm wide and 30-cm deep hollows cut in each side some 30–40 cm from the pit bottom. These shelters were examined fortnightly for a year, so in total 480 pit shelter collections were made. The most common anophelines collected from these pits were *Anopheles tessellatus* (32·32%), *Anopheles subpictus* (22·37%) and *Anopheles culicifacies* (11·44%).

In Iran shelters (120 × 90 and 150 cm deep) have been used with some success to catch *Anopheles stephensi, Anopheles fluviatilis* and *Anopheles dthali*, as well as unidentified culicines (Zaim *et al.*, 1986).

In Central Java Barodji & Supratman (1983) found pit shelters very useful in collecting substantial numbers of *Anopheles aconitus*, the lowest mean catch from four to eight pits was 2·9 in November, and the highest mean catch was 34·6 *Anopheles aconitus* in June. More adults/man-hour were collected from the traps in the dry season than in collections made from a variety of natural shelters, but the reverse was true in the rainy season. They concluded therefore that pit shelters did not accurately reflect population changes of exophilic adults.

## Privy-type shelters

In California, Schoof (1944) constructed 6-ft high wooden structures with 3-ft$^2$ bases and overhanging wooden roofs, which he placed in cool dark places near larval habitats and termed ‘anopheline houses’. He considered that they were better than red boxes or nail kegs in collecting *Anopheles*, such as *Anopheles crucians, Anopheles punctipennis* and *Anopheles quadrimaculatus*.

A small privy-type shelter made of wood and about 7 ft high, 4 ft square with a sloping roof and partially open front was mounted on wooden legs and used by Carpenter *et al.* (1946) to catch *Anopheles quadrimaculatus*. A modified privy shelter (Fig. 3.15*f*) was designed and used by Snow (1949) in the Tennessee Valley. It consists of a wooden framework 6·5 ft high with a 4-ft$^2$ base and with the roof and four sides covered with weatherproof cellulose board (‘Celotex’). The inner surfaces are lined with a moisture proof backing of black asphalt-like material. Each side is composed of four panels, any of which can be removed to either leave an open space or provide an opening for a Bates (1944) type ingress baffle (Fig. 3.15*g*). The trap is easily dismantled for transportation to the field. The biggest catch of *Anopheles quadrimaculatus* was obtained when the lowest panel on the west face of the trap was removed and the third panel on the opposite side replaced by an ingress baffle. The mean catch per day then varied from 316–423 *Anopheles quadrimaculatus*, some 15–24 times greater than that from nail keg barrels. When the lowest panel was not removed only about 4% of the *Anopheles quadrimaculatus* attracted to the trap actually entered it (i.e. through the baffle), 96% rested on the outside, but with the lower panel removed 71% of the catch was collected from inside the trap. The behaviour of *Anopheles crucians* differed from that of *Anopheles quadrimaculatus*. More adults were collected from the trap if the two panels were removed from all sides. In addition to *Anopheles quadrimaculatus, Anopheles crucians* and *Anopheles pictipennis* about 15 culicine species were caught in the shelters.

In Texas Hayes *et al.* (1973) used privy-type shelters and other shelters to collect outdoor resting mosquitoes, including blood-fed specimens.

### General considerations

Different mosquito species and different gonotrophic stages of the same species may require different types of resting sites, consequently artificial shelters are unlikely to give unbiased samples of the exophilic population of all species in an area. The efficiency of an artificial shelter in catching a particular species may vary in different areas, or in the same area at different times of the year. An artificial resting shelter must compete with natural outdoor resting sites. In areas, for example where these are minimal, artificial shelters may attract larger numbers of mosquitoes than similar shelters located in areas with abundant natural resting sites. It follows that these larger catches do not necessarily reflect a larger exophilic population. In fact during periods of dry weather mosquito populations may be greatly reduced, but because of the reduction of suitable outdoor resting sites greater numbers of mosquitoes may seek refuge in artificial shelters.

Artificial shelters provide resting places not only for mosquitoes but also for numerous other animals such as lizards, spiders and scorpions, and as these may be predators of mosquitoes they should be removed every time collections are made from the shelters.

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