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Host preferences of mosquitoes.

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HOST PREFERENCES OF MOSQUITOES

A Thesis Presented

by

Robert George Means

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University of Massachusetts in
partial fulfillment of the requirements for the degree of

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HOST PREFERENCES OF MOSQUITOES

A Thesis

by

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INTRODUCTION ·

In recent years, many workers in the field of medical entomology have realized the necessity of knowing preferred hosts of mosquitoes and other biting Diptera. Hess and Holden (1958) have stressed that the host preferences of mosquitoes and their abilities to deviate from these preferences are factors of great importance in the study of arthropod-borne encephalitis viruses, some of which are considered to be the most important mosquito-transmitted diseases of man in the United States today (Herms and James 1961; Kelser 1933; Lumsden 1958). Certainly mosquitoes which will feed only on birds cannot transmit viruses to man or other mammals. However, mosquitoes which feed on both birds and mammals can transmit viruses to any of the hosts, providing the mosquitoes are capable of infecting and the hosts are receptive to the viruses. Therefore, it is advisable to have as much knowledge as possible regarding the host preferences of mosquitoes as well as their abilities to transmit viruses to their hosts. Hayes and Parsons (1957) have pointed out that, although certain mosquitoes may be found to transmit viruses in laboratory experiments, field tests must be made to determine their true potential as vectors in nature.

Several outbreaks of Eastern (EE), Western (WE), St. Louis (SLE), and other encephalitides have occurred in the United States since 1933, and earlier epizootics were probably caused by the same viruses (Beadle 1952; Beadle 1959; Hansom 1957; U. S. Dept. of Health, Educ. & Wel. Feb. 1965). Eastern Encephalitis, the most virulent of these viruses, is also the most common along the Atlantic coast. About 60 percent of the

clinical human cases are fatal, and survivors usually suffer mental incapacities (Beadle 1959; Horsfall 1962). In Massachusetts alone, epidemics of EE were fatal to 25 of 34 victims, and 10 of 13 victims in 1938 and 1956 respectively (Alexander and Murray 1958; Getting 1941). An outbreak in New Jersey in 1959 resulted in 22 mortalities of the 33 cases reported (Kandle 1960). Several other outbreaks of EE have occurred in Louisiana, New York, Florida, and other states (Beadle 1959). In 1964 SLE, EE, and WE claimed the lives of at least 57 persons in 9 states (U.S. Dept. of Health, Educ., and Wel. Feb. 1965).

Ten Broeck first suggested in 1938 that birds could serve as reservoirs of EE and other encephalitides, and later the same year the virus was isolated from pheasants and a pigeon (Fothergill and Dingle 1938; Tyzzer, Sellards and Bennett 1938; Van Roekel and Clarke 1939). It is now generally accepted that birds are the main reservoirs for these viruses (Beadle 1959), although snakes and turtles may serve as overwintering reservoirs (Thomas and Eklund 1960; Thomas, Eklund and Rush 1958). Chamberlain (1958a, 1958b) has shown that EE infection is usually associated with swamp areas, having its reservoir in swamp-inhabiting birds such as blackbirds, starlings, grackles, catbirds, and others. Mosquitoes showing avian preferences maintain the infection in these birds. Outbreaks occur when an unusually high population of mosquitoes is present, along with the virus in reservoir birds and many other susceptible birds. According to Chamberlain, feeding specificity of the mosquitoes becomes less important at these times, resulting in transfer of the virus to horses and man.

In New York State, EE virus was first isolated from pheasants in 1952 and 1953 (Beaudette et al. 1954), and was again discovered in pheasants and White Pekin ducklings on Long Island in 1959 (Dougherty and Price 1960). Since both the duck industry and the tourist trade were economically important to Long Island, the findings were of concern to the inhabitants as well as to public health officials of the state.

At least sixteen species of mosquitoes are known to be potential vectors of the virus (Chamberlain et al. 1958; Collins 1960; Hayes 1961b; Hayes et al. 1960; Hayes et al. 1962; Howitt et al. 1949; Karstad et al. 1957; Wallis 1959; Wallis, Taylor and Henderson 1960). Thirteen of these, including Aedes atropalpus (Coquillett), Aedes sollicitans (Walker), Aedes triseriatus (Say), Aedes vexans (Meigen), Anopheles crucians Wiedemann, Culex restuans Theobald, Culex salinarius Coquillett, Culiseta melanura (Coquillett), Mansonia perturbans (Walker), Orthopodomyia signifera (Coquillett), Psorophora ciliata (Fabricius), Psorophora confinnis (Lynch Arribalzaga) and Psorophora ferox (Humboldt), are present in Suffolk County (Collins 1960; Jamnback 1961).

In 1962 and 1963 a study was made of the host preferences of mosquitoes in Suffolk County. The program was sponsored by the New York State Museum and Science Service, Albany, and was operated in conjunction with programs for determining the incidence of encephalitis virus in mosquitoes and in vertebrates of Long Island. The Cornell University Duck Disease Laboratory in Eastport and the Suffolk County Mosquito Control Commission in Yaphank assisted in the program by supplying laboratory space and materials for equipment. Test animals

were housed in cages at the Wildlife Refuge in Quogue through the cooperation of the New York State Conservation Department. This paper serves as a final report of the two year study.

REVIEW OF LITERATURE

The attractants for some mosquito species have been studied in an effort to better understand why one animal may be preferred over another by a particular mosquito species (Brown, Sakaria and Thompson 1951; Burgess and Brown 1957; Gilbert and Gouck 1957; Howlett 1910; Marshall and Staley 1932; Parker 1948; Peterson and Brown 1951; Rahm 1957; Rudolfs 1922; Tate and Vincent 1932; Thompson and Brown 1955). It has been found from these studies that the following factors contribute to the attraction of mosquitoes:

1. Blood-hunger. According to Rudolfs this is the main factor causing mosquitoes to attack and feed.
2. Color. With most species, dark colors are more attractive than light; however, the reverse is sometimes true. Hue, chroma, fluorescence, and sheen all play roles in the attractiveness of a certain color to a mosquito species.
3. Odor. The odors of sebum and carbon dioxide have both been shown to attract mosquitoes to their hosts.
4. Heat. Peterson and Brown have shown that convective warmth can be a chief attracting factor, particularly when the air temperature is below 60 degrees Fahrenheit.
5. Moisture. Many investigators have shown moisture to be a strong attractant. Peterson and Brown demonstrated that this is particularly true with air temperatures above 60 degrees Fahrenheit.

6. Light. Mosquito species vary in their preferred hours of feeding and the degree of their phototaxis.

Direct observation of mosquitoes feeding upon a host is sometimes used as a method of determining specific preferences. The method has many limitations however, and is usually used only to compare the relative attractiveness of different human hosts (Brown 1958; Clyde and Shute 1958; Laarman 1958).

One of the most commonly employed methods of determining specific host preferences of mosquitoes and other blood-sucking flies is the precipitin test (Adam 1956; Blanton, Keenan and Peyton 1955; Bull and King 1923; Bull and Reynolds 1924; Bull and Root 1923; Colless 1959; Downe 1960; Downe and Morrison 1957; Edman and Downe 1964; Jobbins, Burbutis and Crans 1961; Shemanchuk, Downe and Burgess 1963; Weitz 1956; Williams, Weitz and McClelland 1958). With this method, anti-sera prepared in advance from the blood of several possible host species are tested against the blood from an engorged mosquito. A process of elimination usually indicates the animal species or group of species upon which the mosquito has fed.

Engorged mosquitoes are usually obtained for this testing in one of the following ways:

1. Reared mosquitoes are offered a choice of hosts in a cage. After the mosquitoes have become engorged, their hosts are identified by the precipitin test.
2. Host animals are placed in the field, singly or in groups, in an enclosure. Mosquitoes which are attracted to the animals and become trapped in the enclosure are collected, identified and the hosts determined by the precipitin test.

3. Mosquitoes are collected in light traps or from the vegetation by sweep nets and are identified; the precipitin test is then used to determine the host species.

Two major problems are encountered with this method of host identification. First, if many species of mosquitoes are to be collected and there are several possible host animals, it is difficult to acquire the large stock of anti-sera necessary (Dow, Reeves and Bellamy 1957). Secondly, it is difficult to prepare anti-sera which will effectively distinguish between bird species (Dow, Reeves and Bellamy 1957; Hammon and Reeves 1947).

Various methods have been developed to expose animals in the field and to collect the mosquitoes (and other biting Diptera) which are attracted to them. One method devised to collect biting flies from sheep employed a tent which had one edge of the open bottom hinged to one edge of a rectangular pipe frame the same size as the bottom of the tent. The pipe frame was staked to the ground and a piece of canvas stretched over it. The remaining three sides of the tent and frame were connected by long springs. With the tent in a raised position and the bait (a sheep) staked on the canvas, biting flies were attracted. The tent was sprung from a distance, then the trapped flies were collected from the sheep and tent walls with an aspirator (Jones 1961).

To collect blood-sucking Diptera from birds, Bennett (1960) confined test animals in chicken wire or hardware cloth cages, set them on squares of plywood for a suitable exposure period, then covered them with collecting cages of fine mesh screening. The trapped flies were allowed to feed for 20 to 30 minutes, then were collected with an

aspirator through a cloth sleeve at the top of the cage. Anderson and DeFoliart (1961) used a method similar to Bennett's in 1957 and in 1959 to collect black flies from birds. In 1960 they modified the method by using the collecting cage as a "blackout box." A small transparent trap was connected to the box over a five-inch square hole in one of the upper corners. After engorging, most of the simuliids were attracted to the light emitted through the trap. They were confined in the trap by closing a slide at the bottom of it. The trap was then removed and placed in a freezer to anesthetize the flies which were then transferred to alcohol for identification and storage.

Both of the above methods work well with species of biting flies which are abundant in an area (as is usually the case with black flies and punkies) but not so well with insects which are less abundant. Since the investigator must be present to place the tent or collecting box over the test animal, the testing time and number of testing areas are necessarily limited. Also, attraction to the collector competes with attraction to the test host.

To eliminate the need to be present at the preference traps, some investigators have devised methods which will trap biting flies attracted to test animals. By so doing, longer exposure periods can be employed (overnight, for example) and more insects can be collected than would otherwise be possible. The simplest of these automatic methods was used by Fredeen (1961) to study black flies attracted to sheep. No animal was used; instead, a dark cloth or plywood frame the general size and shape of a sheep attracted the insects. The bottom of the frame was left open and the top was equipped with a glass collecting jar. Insects flying into the dark enclosure, presumably attracted by the shape, then

became attracted to the light source at the top and were trapped in the one-way collecting jar. This method had the disadvantages of (1) attracting only by shape and/or color, (2) providing no proof that the black flies would feed on the animal species being simulated and (3) being limited to larger animals such as sheep, cattle, or horses.

A more versatile method which could be used to test the scent attractiveness of small animals (or carbon dioxide) was described by Bellamy and Reeves in 1952. Holes were cut in the cover and base of a 50-pound lard can and fine mesh screen funnels were soldered to each end with the small holes directed into the can. Mosquitoes could readily fly into the trap but could not usually find their way out. After an exposure period, the small holes of the funnels were plugged with cotton and the entire trap placed into a larger can containing chloroform. The mosquitoes, thus anesthetized, could be collected for identification by removing the cover end of the trap.

A modification of this method was described in 1957 by Dow and co-authors. Four of the traps were suspended about five feet from the ground around a common axis, which was rotated by an electric motor. The rotation insured that the traps would each be in the same position an equal amount of time, thus minimizing the effects of varying population densities of mosquitoes. This method was employed by Henderson and Senior (1961) in California to test the attractiveness of reptiles, amphibians, and small mammals to Culex tarsalis. Hayes (1961a) used the method with slight modifications to test the preferences of Culiseta melanura in Massachusetts. Two disadvantages of this method are (1) the large size of the equipment, and (2) the need for electricity to run the motor.

A host-preference trap described by Edgar and Herndon (1957) also rotated about a common axis driven by an adjustable speed motor. This trap had six wedge-shaped aluminum compartments equipped with screen inlets having adjustable apertures, filters to regulate color and light intensity, and attachments for the introduction of carbon dioxide if desired. The trap was meant for use primarily in the laboratory rather than for field studies. In addition to the disadvantages given for the Dow method, this trap was also very expensive.

Worth and Jonkers (1962) described two useful portable host preference traps. One, a trap which had no moving parts, employed a baffle or funnel-type of entrance to trap the mosquitoes, similar to the trap described by Bellamy and Reeves (1952). The other trap employed many mechanical features which might fail to operate correctly although the writers claimed it worked well. The turning key of a wind-up alarm clock raised a wire, which in turn raised a horizontal wire holding two scoop-shaped flaps open. The flaps, hinged at the upper end, were released so that they closed together, enclosing a caged animal and any mosquitoes which had been feeding on the animal or resting on the cage. Both of these traps had the advantages of portability, operation without electricity, and low cost.

PROCEDURES

MATERIALS AND METHODS. In 1962 preliminary studies were conducted in Suffolk County to devise an efficient method for exposing test animals to natural populations of mosquitoes, collect any mosquitoes which might be attracted, and preserve them for future species determinations. A method of direct observation was first tried. Test animals were confined in tubes of 1/4 inch hardware cloth and placed in the field for one hour. At five minute intervals the engorging mosquitoes were collected from the animal with an aspirator. There were many disadvantages to this manual method of collection:

1. Only one (or very few) animals could be observed at a time and in only one area.
2. Observations had to be made in daylight hours.
3. Observations of longer than one or two hours were not feasible. In this length of time, unless the area was heavily populated with mosquitoes, few were collected. If the area was heavily populated, it soon became very uncomfortable for the investigator.
4. Many of the mosquitoes would escape before being collected.
5. The presence of the investigator may have influenced the results.
6. No host "preferences" were actually shown. Rather, it was shown that a mosquito was attracted to an animal, not that this host was preferred over another species.

Another method which was tried was the blackout-box trap employed by Anderson and DeFoliart in 1960 and described by them in 1961. This was soon found unsatisfactory for collecting large numbers of mosquitoes

from small hosts. Unlike black flies, few mosquitoes would feed at one time on the host. Some of the disadvantages of the manual method also applied here.

Because of the disadvantages of these methods it seemed advisable to devise an automatic mechanism for collecting the mosquitoes as they engorged on the test animals. It was desired that the trap should have the following qualities:

1. It should remove mosquitoes feeding on test animals and trap them in some sort of collecting jar.
2. This should be a continuous operation working for several hours at a time.
3. It should be suitable for use in woods and swamps where electricity is not available.
4. It should be small enough so that several traps could be transported in a station wagon and carried into the woods by one man.
5. It should be as inexpensive as possible.

The apparatus designed was similar to a large-sized New Jersey light trap, using a test animal instead of the light as an attractant. A circle of hardware cloth was forced about two-thirds of the way into an open metal cylinder sixteen inches in diameter and twenty inches deep. The cylinder was set into the wide end of a metal cone twenty-four inches deep which tapered from twenty inches down to two and three-fourths inches. The top ring of a mason jar was welded to the small end of the cone. When a test was to be made, a mason jar half filled with 80 percent alcohol was screwed to the bottom of the cone.

About midway in the cone, a truck fan was attached with the airflow directed downward. A twelve-volt storage battery supplied power for the fan. Experimentation showed, however, that the downrush of air produced by the fan would cause the alcohol to "bump." This hastened evaporation and also left some of the collected mosquitoes on the side of the cone, where they dried out and became unidentifiable. To provide an air outlet and eliminate the bumping, two 4-inch holes were cut near the bottom of the cone and were covered with fine mesh copper screening. The cone was then screwed into a wooden stand.

The test animal was bound in hardware cloth and placed in the cylinder on the hardware cloth shelf. It was observed that with the fan operating continuously, few insects were attracted to the animal and those which were had no chance to become engorged before being pulled into the alcohol. Therefore, the storage battery was wired to a 12-volt DC/110-volt AC converter, the converter connected to a 110-volt repeating cycle timer, and this in turn to a 50-amp. transformer to reduce the voltage to twelve volts. This timing device was then connected to the fan. The converter was necessary to feed the timer since a twelve-volt repeating timer was not available at the time. Under most circumstances, a cycle having the fan off for 10-15 minutes, then on for 1-3 minutes collected the most mosquitoes with the greatest percentage of engorgement.

Several sets, or triads, were constructed during the winter for use in 1964. A triad consisted of three traps, one six-volt storage battery and box, and one timer. Each trap was composed of the following parts (figs. 1 and 2):

1. A cone, 20 inches deep, the wide half made of galvanized steel and the narrow end of 20-gauge copper screening. The cone tapered from a diameter of about sixteen inches down to about two and three-fourths inches. The ring of a mason jar cover was welded to the small end.
2. About four inches from the wide end of the cone a six-volt fan was fastened to the inside with the airflow directed toward the small end. A six-volt system was used instead of the twelve-volt in hopes that fewer batteries would be stolen, since most automobiles no longer use six-volt batteries.
3. A galvanized steel cylinder, fourteen inches in diameter and twenty inches long, rested inside the top of the cone. About one-third of the way from the bottom of the cylinder a piece of half inch hardware cloth was forced in to form a shelf for the test animal.
4. Each trap was screwed into place in a wooden stand. Most of the fans were supplied with thirty feet of electrical cord and a plug.

For each test period three of the traps were arranged in an equilateral triangle having twenty-foot sides with one point directed south. A locked wooden box containing a six-volt storage battery and a timing mechanism was chained to a tree near the center of the triangle (figs. 3, 4, & 5). The thirty-foot cords of two traps were plugged into a duplex receptacle which was wired to the third trap and mounted on its stand. A thirty-foot wire from this receptacle was equipped with the plug of a two-pole universal utility connector, the receptacle of

which was mounted in the battery box and connected in series with the timer and battery. After all connections were made, the timer would engage the battery with the fans for a certain period before disengaging for a longer time.

The timers were constructed at the State Museum and consisted essentially of (fig. 4):

1. A watch mechanism operated by two size D flashlight cells.
2. A microswitch to engage and disengage the battery with the fans.
3. A plastic disc about two inches in diameter with slots cut out which caused the microswitch to make electrical connections for one minute out of every sixteen minutes.
4. A waterproof plastic case with mounting bracket.

With a cycle of one minute on - fifteen minutes off, a storage battery would last for about 48 hours without being recharged. When a battery required charging, it was removed from the box and carried to and from the testing area with a battery strap to minimize movement of the delicate timers.

Animals to be tested were confined in either one-inch chicken wire or one-fourth-inch hardware cloth, depending on their sizes (fig. 6), and placed on the hardware cloth shelves inside the cylinders (fig. 2). Pint mason jars half full of 80% ethyl alcohol were attached to the bottoms of the cones and the pieces of apparatus connected. The animals were left in an area at about 5:00 p.m. on the evening a test was to be run. The author always remained at the testing site through one complete cycle to make sure that all equipment was operating correctly. After a

nominal 24-hour period (usually 22-24 hours) the animals were picked up, returned to their cages at the wildlife refuge in Quogue, and not used again for at least four days. The mosquito collections were placed in vials, labelled, and stored for future identification.

STATIONS USED. Triads of the host preference traps were operated from May through August, 1963, at a total of twelve stations in Suffolk County. Brief descriptions of the test stations follow (see map)

STATION 1. The Wilcox duck farm was in Speonk, on Brushy Neck Lane, south of Route 27, approximately 1.7 miles west of the junction with Old Country Road in Westhampton. The Wilcox farm, typical of most of the duck farms in Suffolk County, was used as a testing site through the generosity and cooperation of its owner, Mr. Leroy Wilcox. The host preference traps were set up in high grass and weeds on the south side of a holding pen which usually contained 50-250 eight-week ducklings. The Speonk River, a shallow, slow-flowing stream about 30-feet wide crossed the west end of the holding pen. The stream, which was polluted from duck droppings and was filled with algae for about eight feet from each shore, served as an ideal breeding site for Culex pipiens. Nearby in the bordering woods, a large bucket of rain water bred Culex restuans all summer.

STATION 2. A large maple swamp was situated at the end of a dirt road running south from Route 24 in Flanders, about 4.5 miles west of Route 27 in Hampton Bays and 0.5 mile east of Pleasure Drive in Flanders. The dirt road ended at a 10-15 acre pond, which was owned by the Flanders Rod and Gun Club, about 1.5 miles south of Route 24. A large swampy area (400-500 acres) surrounding the pond contained

many windfallen maple trees, most of which had root holes varying in size from 5 to 20 feet in diameter by 1/2 to 6 feet deep. The root holes and snowpools throughout the swamp bred many species of mosquitoes until about the middle of July, at which time most of the water dried up. The host preference traps were operated at the northern end of the swamp about 300 yards east of the pond, in an area which was typical of the rest of the swamp.

STATION 3. The Quogue Wildlife Refuge (N. Y. State Conservation Department) was located on South Country Road, 0.7 mile north of Route 27 in Quogue. The refuge was rectangular, extending north from the road for about one mile and east/west for 0.4 mile. Mr. Donald Greely, manager of the refuge, kindly consented to house the test animals, which were located at the southwestern end of the refuge.

The southern one-fourth of the area was kept fairly free of standing water and had few mosquitoes. The northern end, however, was comparatively wild with many potholes amidst the scrub pine and scrub oak. In the approximate middle of the area a pair of excavations covered about 20 acres and were surrounded by about 70 acres of hardwood swamp. Another series of potholes about 1200 yards north of South Country Road covered 25-30 acres and was also surrounded by hardwood swamp. Host preference traps were operated at both of these swampy areas.

STATION 4. The testing site at Riverhead was located at the end of Center Drive, 0.5 mile west of the junction with Routes 24 and 113, Riverhead-Moriches Drive, and Peconic Avenue. Eventually Center Drive will continue westerly for about one mile further. Sixty feet west of the end of the present road, just within the edge of a heavily wooded

area, a drainage ditch ran north to the Peconic River and west along the proposed roadway for about one mile. The ditch varied in width from one to three feet, occasionally widening out to form large muddy pools. Its slowly moving water ranged in depth from a few inches to about two feet. Many species of aquatic and semi-aquatic plants grew in and beside the ditch. The host preference traps were operated along the western branch of the ditch, with one trap on the southern side and the other two on the northern side.

STATION 5. The Laurel Lake testing area was located north of Route 25, about one mile east of Laurel Post Office. A 500-acre swamp, which bred many species of mosquitoes, including Culiseta melanura, lay between Route 25 and Laurel Lake, a distance of about 1/2 mile. The floor of the swamp was covered with sphagnum moss and decaying leaves. Until about the middle of July the many tree root holes held from 1/2 to 4 feet of water and the entire swamp floor was wet, with many large pools of standing water. After that time the area became dry and water was found only in deep holes near the tree roots. The host preference traps were set up about 1/4 mile west of Laurel Lake Road in an area typical of the swamp. Although this was a heavy breeding area, few mosquitoes were collected in 1963 because of larval treatments made by the Mosquito Commission.

STATION 6. The Wolf Swamp Wildlife Sanctuary in North Sea was located between Millstone Brook Road and the eastern shore of Big Fresh Pond. It consisted of about 125 acres of hardwood forest surrounded by an eight-foot wire fence. The only mosquito breeding site was a drainage ditch running from Millstone Brook Road to the pond. This

ditch occasionally had pools of stagnant water which bred Culex territans and Culiseta melanura in small quantities. Host preference traps were operated about 75 yards east of Millstone Brook Road and 30 yards north of the road leading to Big Fresh Pond. Permission to use this sanctuary and Dupont II Wildlife Sanctuary (Station 7) was obtained from the Nature Conservancy.

STATION 7. Dupont II Wildlife Sanctuary was located on Captain's Neck Lane, running south from Route 27A, about 1.3 miles west of Southampton. This 40-acre salt marsh was used only occasionally since the many drainage ditches and catch basins were treated heavily with DDT by the Mosquito Commission and few mosquitoes were present. When the area was used as a testing site the triad was operated in tall weeds and grass about 600 yards south of the entrance gate and about 15 yards west of a grass roadway leading to Taylor Creek Inlet.

STATION 8. Sears Pond Outlet crossed Route 24 in Flanders about 2.8 miles west of Route 27 in Hampton Bays. The outlet, which varied in width from 3 to 8 feet and in depth from 1/2 to 2 feet, ran north from Sears Pond, across Route 24 through a five-foot culvert, and emptied into Flanders Bay. On the northern side of Route 24 it widened out to form a marsh covering about 15 acres. This marsh, which contained an abundance of sphagnum moss and deep tree root holes, bred many species of mosquitoes, including Culiseta melanura, which rested in the culvert during daylight hours. The host preference traps were operated on the south side of the road about 30 yards west of the outlet and 10 yards south of the culvert.

STATION 9. The testing site at Sebonack Neck in North Sea was on the grounds of the National Golf Links of America, on Sebonack Inlet

Road, approximately 1 mile northwest of the junction with New North Highway Road. Bordered on the north by Sebonack Creek and on the south by Bullhead Bay, this area consisted entirely of salt marsh and sand. The area was used as a testing site only occasionally because of vandalism. When tests were run here the traps were set up in a sandy area near the end of the road, diagonally across from the golf clubhouse. Wooden pilings at the edge of the water gave some protection from the wind and also provided a stationary object to which the traps and battery could be chained.

STATION 10. Cow Neck in North Sea was also a salt marsh area, bordered on the north and west by Peconic Bay, on the south by Little Peconic Creek and West Neck Creek, and on the east by Scallop Pond. The traps were set up just off a dirt road, halfway between Scott Road and Scallop Pond. At this point there was a stand of hardwood trees which provided some shade for the test animals. Few tests were made in this area because of vandalism.

STATION 11. Spring Farm, a pheasant and Mallard Duck farm and hunting preserve, was located at the end of Claypits Road in Sag Harbor, approximately 3 miles north of Bridgehampton. The farm was owned and operated by Mr. George Scallinger and his son David, who donated pheasants for testing at various times through the summer. The area consisted mainly of open grass meadow, with wire cages for pheasants and ducks, surrounded by hardwood forest. At the eastern end, a large pool was used for the ducks, but since there was a continuous flow of water, few mosquitoes developed. Host preference traps were set up near the pheasant pens at both the eastern and western ends but few mosquitoes were ever collected.

STATION 12. The Cornell University Long Island Duck Disease Research Laboratories were located on the north side of Old Country Road, approximately 2.5 miles west of Route 27 in Westhampton. A small brook ran through grassy marshland just to the west of the laboratories. Several drainage ditches ran into the brook from the nearby duck pens. The area would have been an ideal breeding place for Culex pipiens except that the Mosquito Commission treated it often. Most traps were set up and operated occasionally about 25 yards north of the road and 30 yards west of the brook, but few mosquitoes were ever collected.

SELECTION OF TEST ANIMALS. For each test a different species of animal was placed in each of two traps; the third trap was left empty as a control. The animals were chosen whenever possible so that they were of different taxonomic classes but were of approximately the same size and color. Table I lists the animal species tested.

SPECIES DETERMINATIONS. Species determinations of the collected mosquitoes were made by the writer at the Entomological Laboratories at the University of Massachusetts during the fall and winter of 1963. It was soon found that the scale patterns necessary for identification were obscured on mosquitoes preserved in alcohol. Therefore, a method of staining was sought so that the light-colored scales would be more obvious. After experimentation with many dyes and stains the most effective method discovered was to stain the mosquitoes for 12-18 hours in a solution of safranin O stain, rinse them, and place them in a solution of malachite green stain. By this procedure the internal

structures and parts of the exoskeleton were colored a deep red but the scales were unaffected and contrasted well against the red background, particularly when placed in the malachite green solution (Means 1963).

ANALYSIS OF DATA

The "t" test was used to determine whether the mean averages of each mosquito species attracted to the various host animals were significantly different (Edwards 1950). In most cases the analysis was made by combining the results in all tests comparing one taxonomic host class with another, i.e. bird versus mammal. When three or more trials comparing species within a class were complete, these data were also analyzed. A trial was considered to be complete when (1) both animals lived through the test period, (2) all traps operated for the full test period, and (3) at least one specimen of the mosquito species being analyzed was attracted to either test animal. In many cases vandalism or accidental death of one or both of the animals rendered the test incomplete and the data were not included in the analysis.

The difference between the number of mosquitoes attracted to each test animal in each trial was squared. Then the sum of the squared deviations for the differences was found by the formula:

$$\Sigma(D-\bar{D})^2 = \Sigma D^2 - \frac{\Sigma(D)^2}{n}$$

when $D = X_1 - X_2$

n = number of differences, or trials

The standard error of the difference between the means was found by the formula:

$$s = \sqrt{\Sigma(D-\bar{D})^2/n(n-1)}$$

The value of "t" was then found by dividing the difference between the treatment means by the standard error of the difference between the means:

$$t = (\bar{X}_1 - \bar{X}_2) / s$$

when \bar{X}_1 = mean number of mosquitoes attracted to test animal #1

\bar{X}_2 = mean number of mosquitoes attracted to test animal #2

The level of significance of the difference between the two means was found by interpolating this value of "t", using n-1 degrees of freedom, in a "t" table (Arkin and Colton 1950).

RESULTS AND DISCUSSION

During the summers of 1962 and 1963, 125 tests comparing the degrees of attraction of various mosquito species to different species of small vertebrates were made in Suffolk County. In twenty-five percent of the trials vandals either destroyed equipment or killed, stole, or released test animals. Another ten percent of the trials were rendered incomplete because of the accidental death of one or both of the test animals. In the 80 completed trials, 4574 mosquitoes representing 10 species were collected (Table II). No mosquitoes were collected in the unbaited control traps which were operated during each trial. The following paragraphs present a discussion of each mosquito species; the previously recorded hosts and the animal species which were found to be hosts during the study are listed, and the host preferences are discussed.

Culex restuans Theobald. Culex restuans was attracted to all of the vertebrate species tested (Table II). In tests comparing birds and mammals, restuans was readily attracted to both classes and the rates of attraction were not significantly different. Warm-blooded animals were more attractive than cold-blooded animals in all cases, with reptiles particularly attracting very few restuans in tests comparing them with either birds or mammals (Table III). Garter Snakes were preferred over Box Turtles by C. restuans, indicating an intraclass specific host preference (Table IV). The percentages of attracted restuans which engorged on test animals varied little within the host classes, ranging from 80.0 to 87.6 percent (Table II).

Culex restuans has been recorded as feeding on the following vertebrates: chicken, pheasant, pigeon, birds in general, man, cattle, pig, Eastern Cottontail, domestic rabbit, Redback Vole, Northern Water Snake, Eastern Box Turtle (Barr 1958; Breeland, Snow and Pickard 1961; Carpenter and LaCasse 1955; Edman and Downe 1964; Felt 1904; Hayes 1961a; Ross 1947; Smith 1904). In the tests conducted in Suffolk County, in addition to biting man, restuans engorged upon the Ring-necked Pheasant, Bob-white Quail, Ruffed Grouse, Common Grackle, Purple Starling, Red-eyed Towhee, Blue Jay, Mallard Duck, White Pekin Duck, New England Cottontail, Meadow Vole, White-footed Mouse, Eastern Chipmunk, Eastern Box Turtle, Eastern Garter Snake, Blacksnake, and Leopard Frog (Tables I and II).

EE virus has been isolated in nature from Culex restuans (Hayes et al. 1960). In view of this and the data which show that the species will feed on a variety of hosts including man, restuans should be considered a possible endemic vector of EE virus.

Culex pipiens Linnaeus. Culex pipiens also fed on all species tested (Table II). Birds were preferred over mammals and also over amphibians, but there was no significant difference in the numbers of pipiens attracted to birds and to reptiles when these two classes were compared. In tests comparing bird species, Culex pipiens showed some preference for White Pekin Ducks and Mallard Ducks over Ring-necked Pheasants, but there was no significant difference in the relative attractiveness of Ring-neck Pheasant and Bob-white Quail (Tables III and IV). All of these tests were conducted at the Wilcox Duck Farm (Site 1), where, although immense numbers of Culex pipiens were present,

they seldom bit man. In contrast to this, pipiens specimens were occasionally collected biting man in wooded situations, but were collected in only 3 complete host preference tests. These were tests conducted at Maple Swamp (Site 2) comparing mammals with reptiles, in which the former were very significantly preferred over the latter (Tables III and IV). The rates of engorgement of attracted mosquitoes were similar for all animal classes, ranging from 84.9 percent for amphibians to 90.0 percent for reptiles (Table II).

The two populations of mosquitoes apparently differed in their host preferences. It is possible that the duck farm population was Culex pipiens pipiens, a man-ignoring, ornithophilic variety of the pipiens complex, and the sylvan population was Culex pipiens molestus, a man-biting variety (Horsfall 1955, Jobling 1938, Mattlingly et al. 1951). However, Dr. Hans Schober, who has been studying larval and adult specimens from the duck farms and swamps of Suffolk County, has concluded that neither of these populations of pipiens morphologically matches the previous descriptions of either Culex pipiens pipiens or Culex pipiens molestus (Personal communication). Until these mosquitoes have been studied further it would seem best to refer to them as the duck farm strain and the sylvan strain of Culex pipiens.

Recorded hosts of Culex pipiens include the following: birds, man, cattle, pig, rodents, frog (Barr 1958; Breeland, Snow and Pickard 1961; Carpenter and LaCasse 1955; Edman and Downe 1964; Felt 1904; Flemings 1958; Headlee 1945; Jobling 1938; Ross 1947; Roubaud 1933; Smith 1904; Stage, Gjullin and Yates 1952; Steward and McWade 1961). In the tests conducted in Suffolk County pipiens fed on the Ring-necked Pheasant, Bob-white Quail, Mallard Duck, White Pekin Duck, Common

Grackle, Purple Starling, Red-eyed Towhee, Blue Jay, New England Cottontail, White-footed Mouse, Eastern Chipmunk, Eastern Box Turtle, Eastern Garter Snake, Blacksnake, and Leopard Frog (Table II).

Since at least one strain of arbovirus has been isolated from Culex pipiens (Chamberlain et al. 1958; Hayes et al. 1962), the species should be regarded as a possible enzootic and endemic vector of encephalitis. It is possible that pipiens was involved in the transmission of EE to ducks on Long Island in 1959 (Dougherty and Price 1960).

Aedes canadensis (Theobald). Aedes canadensis was also a general feeder (Table II), but showed a definite preference for mammals over either birds or reptiles. There were no significant preferences shown for either birds or cold-blooded hosts when canadensis was collected in tests comparing these classes. Neither were there any appreciable differences in attraction to species within any of the animal classes (Tables III and IV). The rates of engorgement varied little among the host classes, ranging from 83.0 percent for reptiles to 88.4 percent for mammals (Table II).

In many areas this species is considered a common pest mosquito which readily attacks man (Armstrong 1960; Carpenter and LaCasse 1955; Headlee 1945; Horsfall 1955; Matheson 1944; Rempel 1953; Ross 1947; Smith 1904; Stage, Gjullin and Yates 1952; Steward and McWade 1961; Wallis 1960). It has also been recorded feeding on domestic duck, grouse, raven, sparrow, Red-winged Blackbird, Catbird, chicken, grackle, pheasant, pigeon, English Sparrow, starling, "fledgling birds" in general, Little Brown Bat, Chipmunk, White-footed Mouse, Norway Rat,

Redback Vole, Grey Squirrel, Cottontail, domestic rabbit, Eastern Garter Snake, Northern Water Snake, box turtles, Eastern Painted Turtle, Eastern Spotted Turtle, Bullfrog, Green Frog, Redback Salamander, American Toad (Bennett 1960; Hayes 1961a; Hayes 1965; Nolan, Moussa and Hayes 1965; Wallis 1960). In general, it has been reported as feeding on both warm and cold-blooded animals (Barr 1958). In the field tests conducted in Suffolk County, in addition to biting man, the species was attracted to and engorged upon the Ring-necked Pheasant, Bob-white Quail, Ruffed Grouse, Common Grackle, Purple Starling, Red-eyed Towhee, New England Cottontail, Meadow Vole, White-footed Mouse, Eastern Chipmunk, Eastern Box Turtle, Eastern Garter Snake, and Leopard Frog (Tables I and II).

Since WE virus has been isolated from Aedes canadensis in nature (Hayes 1961b), and the species appears to have a wide range of hosts, canadensis should be considered a possible endemic vector of encephalitis.

Culiseta melanura (Coquillett). Culiseta melanura was definitely ornithophilic, preferring birds over mammals, reptiles or amphibians (Tables III and IV). In tests comparing Ring-necked Pheasants with Mallard Ducks a strong preference was indicated for the pheasants, but when other birds were compared, i.e. Ring-necked Pheasant versus Bob-white Quail, Red-eyed Towhee versus Common Grackle, or male pheasant versus female, no differences were shown. In four tests comparing mammals with reptiles the latter were significantly more attractive to Culiseta melanura, further indicating that mammals were non-preferred hosts.

Of the few Culiseta melanura which were attracted to mammals (a total of 10 specimens) only 40 percent had taken blood. This contrasted markedly with the percentages of melanura which engorged upon birds, reptiles and amphibians (92.1 percent, 81.0 percent and 74.1 percent respectively) (Table II).

Culiseta melanura has been recorded as feeding on several bird species, both in nature and in the laboratory. Among them are the following: chicken, pheasant, sparrow, starling, pigeon, grackle, Catbird, Cowbird, Cardinal, duck, quail, Robin and Red-winged Blackbird (Chamberlain, Sudia and Nelson 1955; Hayes 1961a; Jobbins, Burbutis and Crans 1961; Wallis 1959). The species has occasionally been recorded engorging on man (Hayes and Doane 1958; Jobbins, Burbutis and Crans 1961; Schober 1964; Wallis 1959) and other mammals including the Norway Rat, rat, Redback Vole, white mouse, mouse, Eastern Chipmunk, Eastern Grey Squirrel, Eastern Cottontail, domestic rabbit, deer, dog, Raccoon, Opossum (Chamberlain, Sudia and Nelson 1959; Hayes 1961a; Jobbins, Burbutis and Crans 1961; Wallis 1959). Although Hayes (1961a) collected many Culiseta melanura which had been attracted to cold-blooded vertebrates, few of these mosquitoes had taken blood. The cold-blooded hosts of Culiseta melanura which have been recorded include the Eastern Milk Snake, Northern Water Snake, Eastern Spotted Turtle and frog (Hayes 1961a; Jobbins, Burbutis and Crans 1961). In the tests conducted in Suffolk County, melanura was attracted to and fed upon the Ring-necked Pheasant, Bob-white Quail, Ruffed grouse, Common Grackle, Purple Starling, Red-eyed Towhee, Blue Jay, Mallard Duck, New England Cottontail, Meadow Vole, Eastern Box Turtle, Eastern Garter Snake, Blacksnake, and Leopard Frog (Tables I and II).

Culiseta melanura has been considered a primary enzootic vector of EE among birds for several years (Chamberlain 1958a). Since it will also feed on a variety of mammals, including man, it must be considered a possible endemic vector. Also, since it feeds on both reptiles and amphibians, it could be involved in the over-wintering of the virus, if cold-blooded vertebrates are actually over-wintering reservoirs as suggested for WE (Thomas and Eklund 1960; Thomas, Eklund and Rush 1958). Other strains of arbovirus have also been isolated from this mosquito (Hayes et al. 1962).

Mansonia perturbans (Walker). Mansonia perturbans was not greatly attracted to reptiles, and showed a strong preference for any of the other host classes with which reptiles were compared, i.e. birds, mammals, and amphibians. In tests comparing two reptiles, the Eastern Garter Snake and the Eastern Box Turtle, some perturbans were attracted to each species and there was no significant preference for one over the other.

Large numbers of Mansonia perturbans, with no significant differences, were collected in tests comparing birds with mammals, birds with amphibians, Ring-necked Pheasant with Bob-white Quail, and male pheasant with female (Tables III and IV). There was one complete test each comparing a Ring-necked Pheasant with a Mallard Duck, a Red-eyed Towhee with a Common Grackle, and a White-footed Mouse with an Eastern Chipmunk. In the first two tests the pheasant and the grackle each attracted a few specimens of Mansonia perturbans while their respective test partners attracted none. Approximately equal numbers of specimens of Mansonia perturbans were attracted to each of the mammalian hosts in

the third test (Tables II and III). Rates of engorgement were considerably lower for the cold-blooded hosts than for the warm-blooded, ranging from 75.5 percent for amphibians to 90.7 percent for birds (Table II).

Records of Mansonia perturbans feeding on man are numerous and in many areas it is considered an important pest species (Armstrong 1941; Breeland, Snow and Pickard 1961; Barr 1958; Carpenter and La Casse 1955; Horsfall 1955; Matheson 1944; Remple 1953; Ross 1947; Smith 1904; Stage, Gjullin and Yates 1952; Steward and McWade 1961; West and Hudson 1960). Mansonia perturbans has also been recorded feeding on the chicken, fowl, domestic duck, grackle, raven, grouse, pheasant, pigeon, sparrow, heron, Eastern Cottontail, domestic rabbit, White-footed Mouse, Redback Vole, Norway Rat, horse, Northern Water Snake, Bullfrog, and Green Frog (Armstrong 1941; Bennett 1960; Hayes 1961a; Hudson et al. 1958; Snow, Pickard and Sparkman 1960). In the tests conducted on Long Island the species was attracted to and engorged upon the Ring-necked Pheasant, Bob-white Quail, Common Grackle, Purple Starling, New England Cottontail, White-footed Mouse, Eastern Chipmunk, and Leopard Frog (Tables I and II).

Mansonia perturbans has been found infected with EE virus in nature (Howitt et al. 1949). Considering this and its wide host range, the species could be an important endemic vector of encephalitis virus.

Other species. Relatively few individuals of Aedes aurifer, abserratus, excrucians, cinereus and Culex territans were collected, but some of these species showed very significant host preferences (Tables III and IV).

Aedes aurifer (Coquillett). Aedes aurifer definitely preferred birds over either mammals or reptiles and also was significantly more attracted to Bob-white Quail than to Ring-necked Pheasant. In one test comparing two mammals, a New England Cottontail attracted four aurifer while its test partner, four White-footed Mice, attracted none. The difference in this latter test may have been due merely to the difference in the size of the test animals. Engorgement rates were high for both birds and mammals (89.1 percent and 100.0 percent respectively) (Table II).

Aedes aurifer has been recorded feeding on man and grouse (Armstrong 1960; Bennett 1960; Carpenter and LaCasse 1955; Headlee 1945; Horsfall 1955; Matheson 1944; Smith 1904; Steward and McWade 1961; West and Hudson 1960). In the tests conducted in Suffolk County, in addition to man and grouse, aurifer also fed upon the Ring-necked Pheasant, Bob-white Quail, Common Grackle, and New England Cottontail, but not on the White-footed Mouse, Eastern Box Turtle, or Eastern Garter Snake (Table II).

Aedes abserratus (Felt and Young). Aedes abserratus was collected only in tests comparing birds with mammals and birds with reptiles and in one test comparing a New England Cottontail with four White-footed Mice. Mammals were strongly preferred in the first test and, although birds attracted a few abserratus and reptiles none when these classes were compared, the difference was not significant. In the single test comparing the two mammals, the rabbit attracted four abserratus and the mice none, but this might have been due to the difference in the relative sizes of the hosts (Tables III and IV). Only one out of the three

abserratus attracted to reptiles was engorged, while 80.0 and 83.3 percent of those attracted to birds and mammals, respectively, were engorged.

Aedes abserratus is also a man-biter (Carpenter and LaCasse 1955; Wallis 1960; West and Hudson 1960). Bennett (1960) listed it as feeding on the domestic duck and Blue Jay, and Wallis (1960) reported that it attacks many fledgling birds. In addition to feeding on man in Suffolk County, abserratus was also collected from the Ring-necked Pheasant, New England Cottontail, Eastern Chipmunk and Eastern Garter Snake. It was not attracted to the Bob-white Quail, Ruffed Grouse, Common Grackle, Blue Jay, White-footed Mouse, or Eastern Box Turtle (Table II).

Aedes excrucians (Walker). Aedes excrucians showed no significant preferences when it was collected in tests comparing birds with mammals, mammals with reptiles, and Ring-necked Pheasant with Bob-white Quail (Tables III and IV). Two tests comparing pheasant with quail indicated that the former species may be more attractive to excrucians; however, when each of these birds was used in tests with various mammals, excrucians was attracted to both. Therefore, more data are necessary before any definite conclusions can be drawn concerning the relative attractiveness of these two species. It was also indicated that mammals might be preferred over reptiles, since the latter did not attract any excrucians, but for the data available the difference was not significant. In one test comparing a New England Cottontail with four White-footed Mice, excrucians was attracted to and fed on both hosts (Tables III and IV). The rates of engorgement were similar for birds and mammals (Table II).

In many areas Aedes excrucians is considered a serious pest species, attacking man in large numbers (Carpenter and LaCasse 1955; Horsfall 1955; Matheson 1944; Steward and McWade 1961; West and Hudson 1960), and in laboratory tests it has fed on the rabbit (Hudson et al. 1958). In Suffolk County, excrucians fed on man and also engorged upon the New England Cottontail, White-footed Mouse, Ring-necked Pheasant, and Bob-white Quail, but not on the Eastern Box Turtle (Table II).

Aedes cinereus Meigen. Aedes cinereus was collected in only two tests. One compared a bird (Bob-white Quail) with a mammal (New England Cottontail), in which the bird attracted four cinereus and the mammal none. In the other test, both hosts, a Ring-necked Pheasant and a Bob-white Quail, attracted a few cinereus (Tables II and III). Because of the scarcity of data no conclusions can be drawn from these tests except that cinereus was attracted to and fed on both birds tested.

Aedes cinereus has been recorded feeding on man, cattle, pig, sheep, dog and fowl (Barr 1958; Carpenter and LaCasse 1955; Horsfall 1955; Matheson 1944; Ross 1947; Shemanchuk, Downe and Burgess 1963; Stage, Gjullin and Yates 1952; Steward and McWade 1961; West and Hudson 1960).

Culex territans Walker. Culex territans was not collected in any tests comparing two warm-blooded animals (Table III). When given a choice of a warm or cold-blooded vertebrate, the latter was very significantly preferred. Comparisons of reptiles with amphibians and Blacksnakes with Eastern Box Turtles indicated no differences in the relative attractiveness of the different cold-blooded animals (Tables III and IV). These data support previous evidence that reptiles and amphibians are greatly preferred over birds and mammals by Culex territans.

Culex territans (apicalis auct.) is generally considered to prefer cold-blooded hosts and has been reported as feeding on frogs and snakes (Barr 1958; Breeland, Snow and Pickard 1961; Burgess and Hammond 1961; Carpenter and LaCasse 1955; Dyar 1928; Horsfall 1955; Matheson 1944; Shannon 1915; Stage, Gjullin and Yates 1952; Steward and McWade 1961; Wallis 1960). There have been some records of territans feeding on warm-blooded vertebrates including man, water rats, cattle, and birds (Edman and Downe 1964; Horsfall 1955; Means 1965; Wallis 1960; West and Hudson 1960). In the tests in Suffolk County, territans engorged upon the Leopard Frog, Eastern Box Turtle, and Blacksnake, but not on the Ring-necked Pheasant, Bob-white Quail, White-footed Mouse, or New England Cottontail (Table II).

SUMMARY AND CONCLUSIONS

In 1962 and 1963 a study of the host preferences of mosquitoes was conducted in Suffolk County, New York. A new and different type of trap was designed to collect mosquitoes attracted to small animals, and several of these traps were operated during the summer of 1963. A total of 4554 mosquitoes, representing 10 species, was collected in 80 trials comparing vertebrates. Using the "t" test to evaluate data, there were some mosquito species which had significant host preferences, while other species fed readily on a wide range of hosts.

In confirmation of previous evidence, Culiseta melanura and the duck farm strain of Culex pipiens were both ornithophilic but also engorged readily on reptiles. Both of these species also fed occasionally on some mammals, including rabbits and mice, and some workers have collected C. melanura biting man. In agreement with previously published data, the sylvan strain of C. pipiens preferred mammals over reptiles. Although there were no complete tests comparing birds with mammals when individuals of the sylvan strain of C. pipiens were collected, it is assumed that these mosquitoes were mammalophilic, since they readily bit man while the duck farm strain did not, and since they preferred mammals over reptiles while the duck farm strain preferred reptiles over mammals.

Since encephalitis viruses have been isolated from both of these species, the evidence is great that they might have been involved in the 1959 EE outbreak in ducks on Long Island. Since many swamp-nesting birds feed diurnally in the duck pens, they could have received the

virus from *C. melanura* in the swamps and transferred it via *C. pipiens* to the ducks.

Since both of these mosquito species feed on reptiles to a great extent, the reptiles could serve as overwintering reservoirs for EE virus, as has been suggested for WE. Also, since the species do bite mammals, the sylvan strain of *C. pipiens* to a great extent and *C. melanura* occasionally, they could serve as endemic vectors of the virus.

Both *Culex restuans* and *Mansonia perturbans* preferred either birds or mammals over reptiles. *Mansonia perturbans* also preferred amphibians over reptiles, but *C. restuans* was not collected in any tests comparing these two classes of cold-blooded vertebrates. Birds were somewhat preferred over amphibians by *C. restuans* but there were no differences in the attraction of *M. perturbans* to either of these host classes.

In tests comparing Box Turtles with Garter Snakes the former were preferred by *C. restuans* but there were no differences indicated for *M. perturbans*. These results are in agreement with the results of other workers with the notable exception that *C. restuans* engorged to a great extent on Leopard Frogs when they were compared with birds in Suffolk County. Previous records have not listed amphibians as being hosts for this species.

Since EE virus has been isolated from both of these species, they should be considered potential endemic vectors of the virus. This is particularly true since birds are known to be reservoirs of the virus and both *C. restuans* and *M. perturbans* feed readily on both birds and mammals, including man.

Aedes canadensis preferred mammals over either birds or reptiles but fed readily on all taxonomic classes of vertebrates. These results are in agreement with other reports that the species is a man-biting mosquito which feeds on mammals, birds, reptiles and amphibians. Since A. canadensis has a wide range of hosts, and since WE virus has been isolated from the species, it should be considered a potential enzootic and endemic vector of encephalitis viruses.

In confirmation of the published records of the hosts of Culex territans, this species was found to feed almost exclusively on reptiles and amphibians. In fact, when two warm-blooded animals were compared, no C. territans were collected, although they were known to be in the area. Since C. territans very rarely bites man or other warm-blooded vertebrates, the species is probably not involved in the transmission of encephalitis viruses to man.

Few specimens of Aedes aurifer, abserratus, excrucians or cinereus were collected in the tests; therefore, little can be said concerning their host preferences. The data collected indicated that A. aurifer may be ornithophilic, although some individuals were collected from rabbits also. The species has been recorded as feeding on birds and also as a pest of man. Aedes abserratus seemed to prefer mammals over birds, but did feed on the latter and also on reptiles to a certain extent. These data are in agreement with previous published records which state that A. abserratus is a general feeder and a common pest of man.

So few specimens of Aedes excrucians and cinereus were collected that no conclusions can be drawn concerning their host preferences.

Table I. Vertebrate Species Used in Host Preference Tests in
Suffolk County, New York, 1962-1963.

Vertebrate species*	Common name	No. of nights exposed	No. of successful trials
BIRDS			
<u>Phasianus colchicus</u>	Ring-necked Pheasant	41	31
<u>Colinus virginianus</u>	Bob-white Quail	24	17
<u>Anas platyrhynchos</u>	Mallard Duck	16	6
<u>Anas boschas</u>	White Pekin Duck	15	10
<u>Quiscalus quiscula</u>	Common Grackle	19	13
<u>Sturnus vulgaris</u>	Purple Starling	9	3
<u>Pipilo erythrophthalmus</u>	Red-eyed Towhee	8	4
<u>Bonasa umbellus</u>	Ruffed Grouse	7	2
<u>Cyanocitta cristata</u>	Blue Jay	3	2
Total birds		142	88
MAMMALS			
<u>Sylvilagus transitionalis</u>	New England Cottontail	24	18
<u>Peromyscus leucopus</u>	White-footed Mouse	12	8
<u>Microtus pennsylvanicus</u>	Meadow Vole	6	4
<u>Tamias striatus</u>	Eastern Chipmunk	6	4
<u>Procyon lotor</u>	Raccoon	2	0
Total mammals		50	34

* Scientific and common names from American Ornithologist's Union (1957), Burt (1957, Conant (1958).

Table I. (pg. 2)

Vertebrate species	Common name	No. of nights exposed	No. of successful trials
REPTILES			
<u>Terrapene carolina</u>	Eastern Box Turtle	22	18
<u>Coluber constrictor</u>	Blacksnake	9	6
<u>Thamnophis sirtalis</u>	Eastern Garter Snake	10	6
Total reptiles		41	30
AMPHIBIANS			
<u>Rana pipiens</u>	Leopard Frog	10	8
<u>Rana palustris</u>	Green Frog	5	0
Total amphibians		15	8
Total, all vertebrates		248	160

Table II. Numbers of Mosquitoes Collected from Animals in Host Preference Tests
 Conducted in Suffolk County, New York, 1962-1963.

Host	Mosquitoes Collected				
	<u>Culex</u> <u>restuans</u>	<u>Culex</u> <u>pipiens</u>	<u>Aedes</u> <u>canadensis</u>	<u>Culiseta</u> <u>melanura</u>	<u>Mansonia</u> <u>perturbans</u>
BIRDS					
Pheasant	286/314 (22)*	173/183 (20)	116/132 (21)	400/441 (28)	174/186 (17)
Quail	73/87 (10)	52/62 (9)	37/44 (11)	190/203 (14)	117/129 (12)
Mallard Duck	37/45 (5)	55/60 (5)	0/0 (1)	38/40 (4)	0/0 (1)
White Pekin Duck	10/13 (2)	228/342 (9)	---	---	---
Grackle	5/7 (2)	84/86 (4)	58/68 (11)	135/147 (9)	58/67 (3)
Starling	10/12 (1)	16/16 (3)	4/4 (1)	24/24 (2)	10/14 (1)
Towhee	9/13 (2)	18/21 (2)	40/45 (4)	36/38 (3)	0/0 (1)
Grouse	3/4 (1)	---	8/12 (2)	23/24 (2)	---
Blue Jay	4/4 (1)	22/33 (2)	9/13 (1)	10/12 (1)	---
Totals, birds	437/499 (46)	711/803 (54)	272/318 (52)	856/929 (63)	359/396 (35)
Percent feeding	87.6	88.5	85.5	92.1	90.7
MAMMALS					
Cottontail	108/115 (10)	58/67 (7)	138/155 (16)	4/8 (10)	84/95 (5)
Meadow Vole	24/35 (4)	---	15/21 (4)	0/2 (2)	11/13 (2)
White-footed Mouse	27/33 (5)	6/8 (1)	67/74 (8)	0/0 (2)	22/25 (2)
Chipmunk	30/39 (4)	11/13 (2)	24/26 (3)	0/0 (4)	24/30 (2)
Totals, mammals	189/221 (23)	75/88 (10)	244/276 (31)	4/10 (18)	141/163 (11)
Percent feeding	85.1	85.2	88.4	40.0	86.5
REPTILES					
Box Turtle	3/4 (7)	65/71 (8)	51/60 (15)	21/27 (10)	11/14 (7)
Garter Snake	9/11 (2)	6/7 (1)	12/16 (4)	6/7 (2)	10/13 (5)
Blacksnake	4/5 (4)	10/12 (4)	10/12 (2)	7/8 (2)	0/0 (5)
Totals, reptiles	16/20 (13)	81/90 (13)	73/88 (21)	34/42 (14)	21/27 (17)
Percent feeding	80.0	90.0	83.0	81.0	77.8
AMPHIBIANS					
Leopard Frog	28/35 (4)	90/106 (7)	50/59 (4)	20/27 (5)	40/53 (5)
Totals, amphibians	28/35 (4)	90/106 (7)	50/59 (4)	20/27 (5)	40/53 (5)
Percent feeding	80.0	84.9	84.7	74.1	75.5
Totals, all animals	670/776 (86)	957/1087 (84)	639/741 (108)	914/1008 (100)	551/639 (68)
Percent feeding	86.3	88.1	86.2	90.7	86.2

* Number engorged/Number attracted (Number of trials when at least one specimen was collected in any trap of the triad). Dashes indicate the mosquito species was not collected in any trials involving this animal species.

Table II. (pg. 2)

Host	Mosquitoes Collected				
	<u>Aedes aurifer</u>	<u>Aedes abserratus</u>	<u>Aedes excrucians</u>	<u>Aedes cinereus</u>	<u>Culex territans</u>
BIRDS					
Pheasant	23/24(8)	4/5(5)	7/9(4)	5/7(1)	0/2(4)
Quail	11/13(4)	0/0(1)	8/8(3)	8/8(2)	0/0(2)
Grouse	6/6(1)	0/0(1)	---	---	---
Grackle	17/21(4)	0/0(2)	---	---	---
Blue Jay	---	0/0(1)	---	---	---
Totals, birds	57/64(17)	4/5(10)	15/17(7)	13/15(3)	0/2(8)
Percent feeding	89.1	80.0	88.2	86.7	0.0
MAMMALS					
Cottontail	5/5(5)	29/35(7)	18/22(3)	0/0(1)	0/0(2)
White-footed Mouse	0/0(3)	0/0(1)	17/21(4)	---	0/0(2)
Chipmunk	---	6/7(1)	---	---	---
Totals, mammals	5/5(8)	35/42(9)	35/43(7)	0/0(1)	0/0(4)
Percent feeding	100.0	83.3	81.4	---	---
REPTILES					
Box Turtle	0/0(4)	0/0(2)	0/0(2)	---	25/30(5)
Garter Snake	0/0(1)	1/3(1)	---	---	29/35(6)
Blacksnake	---	---	---	---	19/19(4)
Totals, reptiles	0/0(5)	1/3(3)	0/0(2)	---	73/84(15)
Percent feeding	---	33.3	---	---	86.1
AMPHIBIANS					
Leopard Frog	---	---	---	---	40/43(7)
Totals, amphibians	---	---	---	---	40/43(7)
Percent feeding	---	---	---	---	93.0
Totals, all animals	62/69(30)	40/50(22)	50/60(16)	13/15(4)	113/129(34)
Percent feeding	89.9	80.0	83.3	86.7	87.6

Table III. Numbers of Mosquitoes Collected in Host Preference Tests Conducted in Suffolk County, New York, Comparing Animal Classes, Species and Sexes, 1962-1963.

Hosts Compared	Mosquitoes Collected				
	<u>Culex restuans</u>	<u>Culex pipiens</u>	<u>Aedes canadensis</u>	<u>Culiseta melanura</u>	<u>Mansonia perturbans</u>
Birds	59/69	95/104	72/84	147/156	65/73
Mammals	83/98 (12)*	4/6 (7)	165/185 (19)	3/6 (14)	58/69 (5)
Birds	32/37	136/145	42/55	105/118	55/60
Reptiles	0/0 (6)	75/82 (9)	57/64 (11)	13/17 (9)	2/3 (4)
Birds	60/67	201/229	40/49	78/81	32/34
Amphibians	28/35 (4)	87/102 (6)	50/59 (4)	18/23 (4)	25/34 (2)
Mammals	75/84	71/82	29/31	1/4	70/78
Reptiles	7/9 (3)	2/3 (3)	10/15 (4)	18/21 (4)	1/2 (4)
Reptiles	---	4/5	---	3/4 (1)	2/3
Amphibians	---	3/4 (1)	---	2/4 (1)	15/19 (3)
Pheasant	86/96	52/62	27/28	104/118	71/77
Quail	46/57 (5)	44/51 (8)	15/18 (4)	101/114 (9)	53/58 (8)
Pheasant	16/19 (2)	32/42 (4)	13/14 (1)	94/96 (4)	6/8 (1)
Mallard Duck	15/17	45/49	0/0	38/40	0/0
Pheasant	6/8 (1)	45/51 (4)	---	---	---
White Pekin Duck	6/7	61/70	---	---	---
Pheasant (♂)	48/50 (2)	---	---	52/55 (2)	28/30 (2)
Pheasant (♀)	49/52	---	---	40/44	32/34 (2)
Towhee	9/13 (2)	---	40/43 (4)	36/38 (3)	0/0
Grackle	5/7	---	23/27	61/69	17/22 (1)
White-footed Mouse	8/11 (2)	---	14/18 (2)	---	6/7 (1)
Chipmunk	7/9	---	16/20	---	7/9
White-footed Mouse	10/11 (2)	---	12/13 (2)	---	---
Meadow Vole	6/9	---	8/9	---	---
Garter Snake	9/11 (2)	---	2/3 (3)	---	7/8 (3)
Box Turtle	0/0	---	4/6	---	9/11
Totals, all trials	670/776 (43)	957/1087 (42)	639/741 (54)	914/1008 (50)	551/639 (34)

* Number engorged/Number attracted (Number of trials when at least one specimen was collected in any trap of the triad). Dashes indicate the mosquito species was not collected in any trials comparing these animal classes or species.

Table III. (pg. 2)

Hosts Compared	Mosquitoes Collected				
	<u>Aedes aurifer</u>	<u>Aedes aberratus</u>	<u>Aedes excrucians</u>	<u>Aedes cinereus</u>	<u>Culex territans</u>
Birds	28/30 (6)	0/0	8/8	4/4 (1)	---
Mammals	1/1	31/38 (7)	17/22 (3)	0/0	---
Birds	23/26 (5)	4/5 (3)	---	---	0/0 (4)
Reptiles	0/0	1/3	---	---	19/24 (4)
Birds	---	---	---	---	0/2 (4)
Amphibians	---	---	---	---	25/28 (4)
Mammals	---	---	7/8 (2)	---	0/0 (4)
Reptiles	---	---	0/0	---	19/22 (4)
Reptiles	---	---	---	---	10/11 (3)
Amphibians	---	---	---	---	15/15 (3)
Pheasant	0/0 (3)	---	7/9 (2)	5/7 (1)	---
Quail	6/8	---	0/0	4/4 (1)	---
New England Cottontail	4/4 (1)	4/4 (1)	8/10 (1)	---	---
White-footed Mouse	0/0	0/0	3/3	---	---
Blacksnake	---	---	---	---	15/16 (2)
Box Turtle	---	---	---	---	10/11 (2)
Totals, all trials	62/69(15)	40/50(11)	50/60(8)	13/15(2)	113/129(17)

Table IV. A Comparison of the Attraction Rates of Nine Mosquito Species to Various Vertebrates in Tests Conducted in Suffolk County, New York, 1962-1963.

	No. of trials (n)	Mosquitoes Attracted			Standard error (s)	t ($\bar{X}_1 - \bar{X}_2 / s$)
		min.	max.	mean per trial (\bar{X})		
<u>Culex restuans</u>						
Birds	12	0	22	5.75	3.57	0.6779
Mammals		0	23	8.17		
Birds	6	1	13	16.67	1.75	3.5257**
Reptiles		0	0	0.00		
Birds	4	3	29	16.75	1.78	4.4944*
Amphibians		0	18	8.75		
Mammals	3	23	35	28.00	3.05	8.1967**
Reptiles		2	4	3.00		
Pheasant	5	6	53	19.20	7.78	1.0026
Quail		0	22	11.40		
Pheasant	2	9	10	9.50	2.00	0.5000
Mallard Duck		6	11	8.50		
Pheasant (♂)	2	24	26	25.00	1.00	1.0000
Pheasant (♀)		26	26	26.00		
Towhee	2	6	7	6.50	1.00	3.0000
Grackle		2	5	3.50		
White-footed Mouse	2	5	7	6.00	0.50	6.0000
Chipmunk		4	5	4.50		
White-footed Mouse	2	4	7	5.50	1.00	1.0000
Meadow Vole		4	5	4.50		
Garter Snake	2	5	6	5.50	0.50	11.0000*
Box Turtle		0	0	0.00		
<u>Culex pipiens</u>						
Birds	7	0	40	14.86	2.76	5.0725***
Mammals		0	3	0.86		
Birds	9	0	50	16.11	5.42	1.2915
Reptiles		0	42	9.11		
Birds	6	15	63	38.17	5.16	4.0930***
Amphibians		10	30	17.00		
Mammals	3	25	31	27.33	11.67	18.8071***
Reptiles		0	2	1.00		
Pheasant	8	1	16	7.75	1.41	0.9787
Quail		0	12	6.37		
Pheasant	4	6	16	10.50	0.44	3.9773*
Mallard Duck		7	18	12.25		
Pheasant	4	8	17	12.75	1.30	3.6538*
White Pekin Duck		12	24	17.50		

* Difference between means significant at 5% level.
 ** Difference between means significant at 2% level.
 *** Difference between means significant at 1% level.

Table IV. (pg. 2)

	No. of trials (n)	Mosquitoes Attracted			Standard error (s)	t ($\bar{X}_1 - \bar{X}_2 / s$)
		min.	max.	mean per trial (\bar{X})		
<u>Aedes canadensis</u>						
Birds	19	0	24	4.42	1.40	3.8000***
Mammals		1	23	9.74		
Birds	11	0	21	5.00	2.74	0.2993
Reptiles		0	17	5.82		
Birds	4	10	16	12.25	1.85	1.3514
Amphibians		10	21	14.75		
Mammals	4	2	12	7.75	0.82	4.8780**
Reptiles		0	8	3.75		
Pheasant	4	0	27	7.00	3.59	0.6964
Quail		0	14	4.50		
Towhee	4	4	16	10.75	2.35	1.7021
Grackle		0	19	6.75		
White-footed Mouse	2	8	10	9.00	1.00	1.0000
Chipmunk		8	12	10.00		
White-footed Mouse	2	4	9	6.50	1.00	2.0000
Meadow Vole		3	6	4.50		
Garter Snake	3	0	2	1.00	0.81	1.2346
Box Turtle		1	3	2.00		
<u>Culiseta melanura</u>						
Birds	14	2	29	11.14	2.23	4.8700***
Mammals		0	2	0.43		
Birds	9	0	24	13.11	2.64	4.2500***
Reptiles		0	7	1.89		
Birds	4	15	26	20.25	2.40	6.0416***
Amphibians		3	9	5.75		
Mammals	4	0	3	1.00	1.18	3.6017
Reptiles		4	7	5.25		
Pheasant	9	0	29	13.11	1.63	0.2699
Quail		1	24	12.76		
Pheasant	4	21	28	24.00	3.08	4.5454**
Mallard Duck		5	16	10.00		
Pheasant (♂)	2	26	29	27.50	2.50	2.2000
Pheasant (♀)		21	23	22.00		
Towhee	3	4	26	12.67	6.85	1.5080
Grackle		8	32	23.00		
<u>Aedes aurifer</u>						
Birds	6	2	7	5.00	0.70	6.9000***
Mammals		0	1	0.17		
Birds	5	3	7	5.20	0.73	7.1233***
Reptiles		0	0	0.00		
Pheasant	3	0	0	0.00	0.47	5.6809*
Quail		2	3	2.67		

Table IV. (pg. 3)

	No. of trials (n)	Mosquitoes Attracted		mean per trial (\bar{X})	Standard error (s)	t ($\bar{X}_1 - \bar{X}_2 / s$)
		min.	max.			
<u>Mansonia perturbans</u>						
Birds	5	0	28	14.60	2.29	0.3493
Mammals		0	25	13.80		
Birds	4	11	18	15.00	1.49	9.5693***
Reptiles		0	2	0.75		
Birds	2	15	19	17.00	0.00	0.0000
Amphibians		16	18	17.00		
Mammals	4	14	24	19.50	2.38	7.9832***
Reptiles		0	2	0.50		
Reptiles	3	0	2	1.00	0.67	7.9552***
Amphibians		6	7	6.33		
Pheasant	8	0	21	9.63	2.38	1.0000
Quail		1	21	7.25		
Pheasant (♂)	2	13	17	15.00	1.00	2.0000
Pheasant (♀)		14	20	17.00		
Garter Snake	3	0	5	2.66	1.00	1.0000
Box Turtle		3	6	3.66		
<u>Culex territans</u>						
Birds	4	0	0	0.00	0.41	14.6341***
Reptiles		5	7	6.00		
Birds	4	0	1	0.50	0.96	6.7708***
Amphibians		4	9	7.00		
Mammals	4	0	0	0.00	1.19	4.6218**
Reptiles		3	8	5.50		
Reptiles	3	2	6	3.67	0.34	2.2059
Amphibians		3	7	5.00		
Blacksnake	2	7	9	8.00	0.50	5.0000
Box Turtle		5	6	5.50		
<u>Aedes abserratus</u>						
Birds	7	0	0	0.00	1.38	3.8986***
Mammals		1	12	5.43		
Birds	3	1	4	2.67	0.88	3.0341
Reptiles		0	0	0.00		
<u>Aedes excrucians</u>						
Birds	3	0	8	2.67	2.05	2.2780
Mammals		5	10	7.33		
Mammals	2	3	5	4.00	1.00	4.0000
Reptiles		0	0	0.00		
Pheasant	2	1	8	4.50	3.50	1.2857
Quail		0	0	0.00		

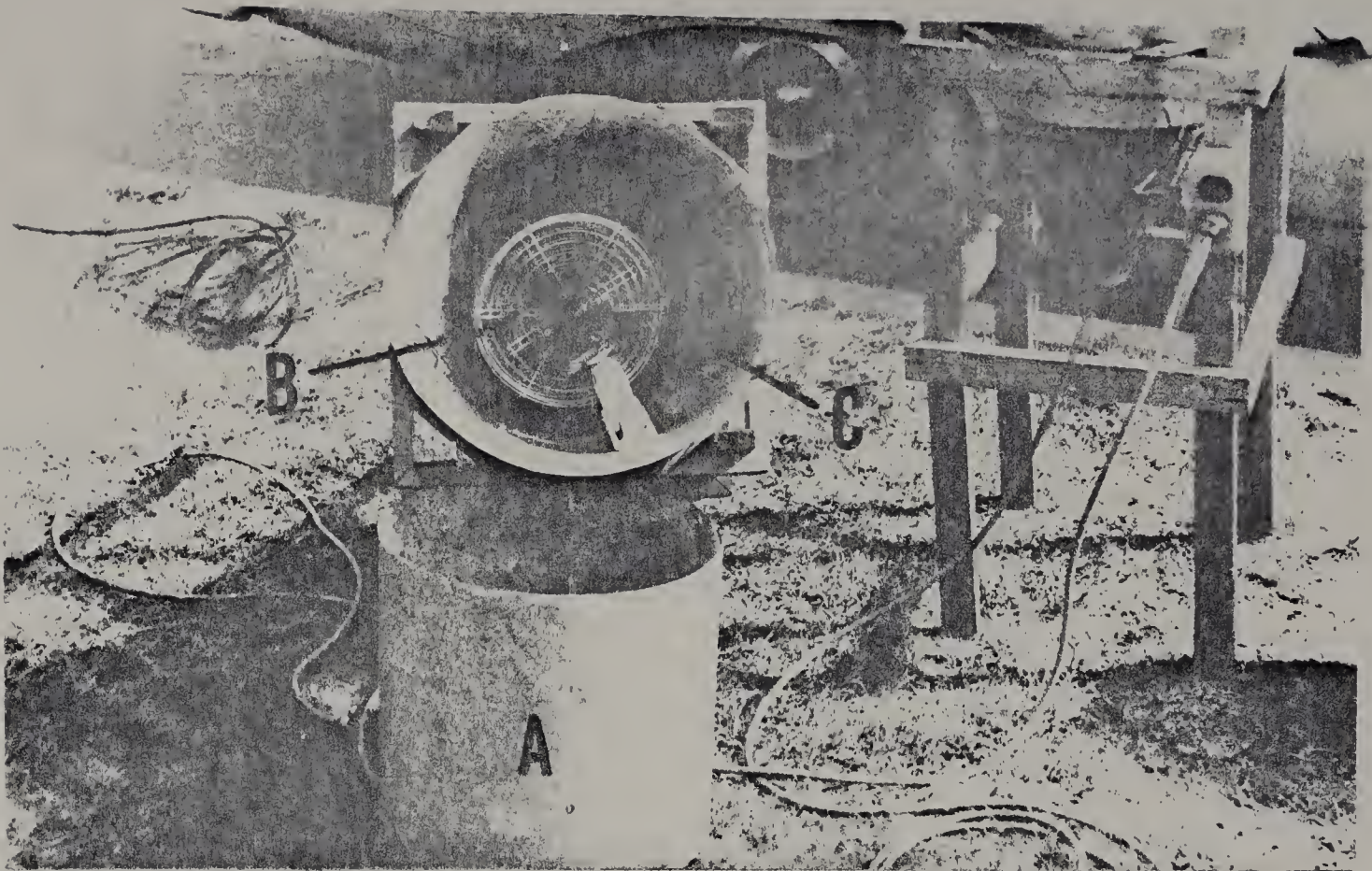


Fig. 1. Sections of host preference trap

A = cylinder, B = fan, C = cone.



Fig. 2. Test animal (Bob-white Quail) being placed
in wire holder.

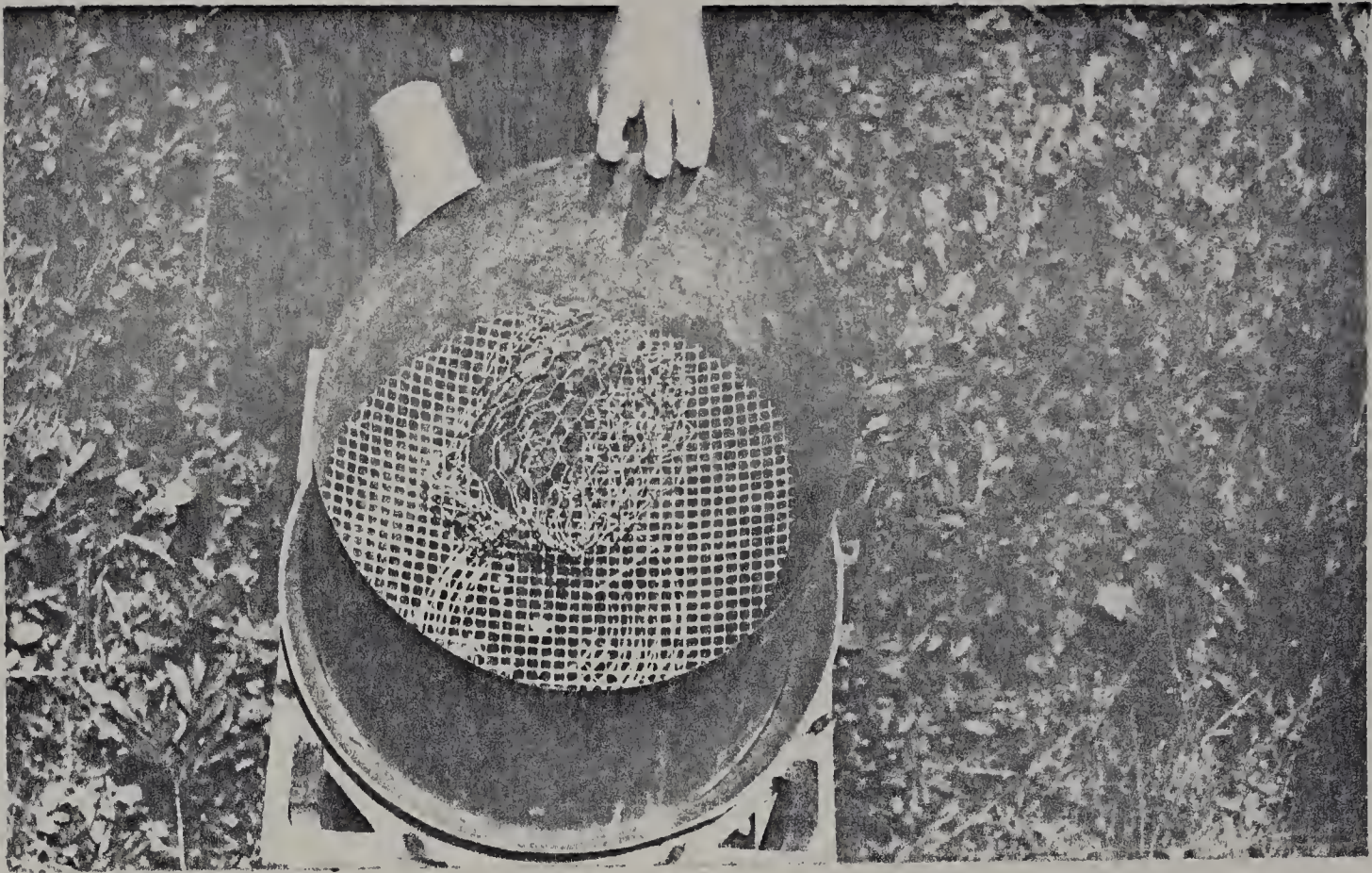


Fig. 3. Host preference trap with animal in place.

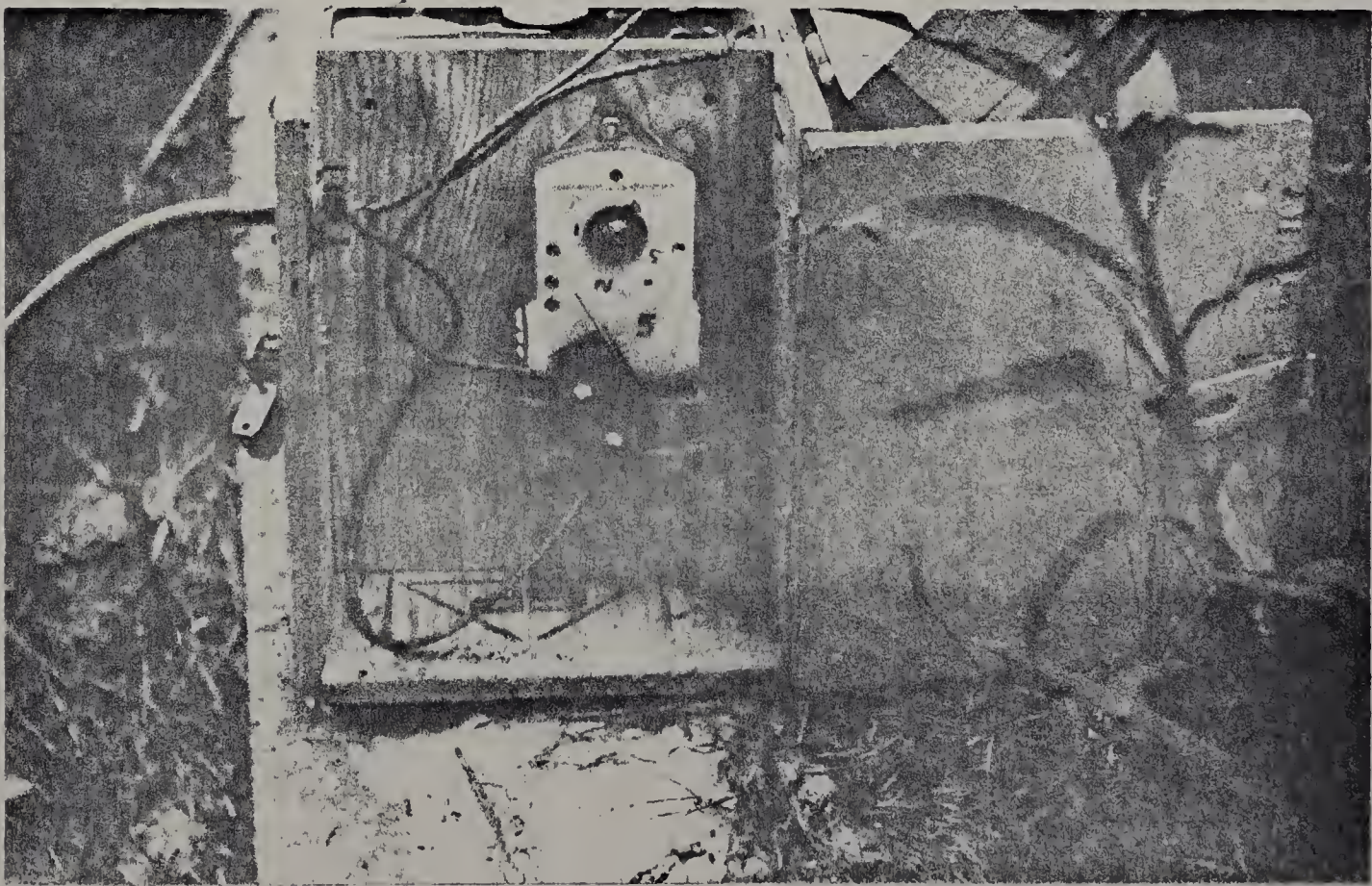


Fig. 4. Open battery box showing timing mechanism.

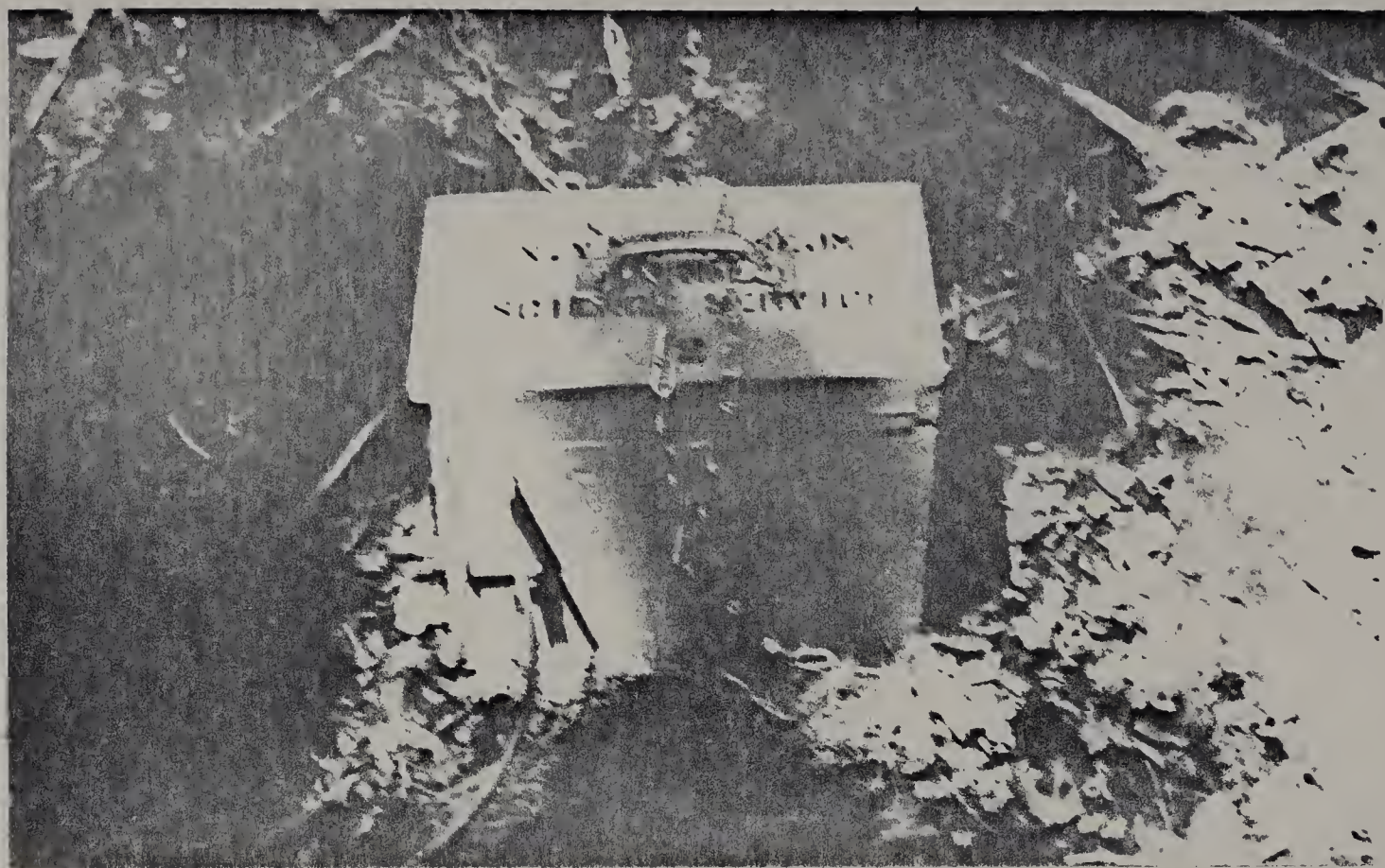


Fig. 5. Locked and chained battery box.

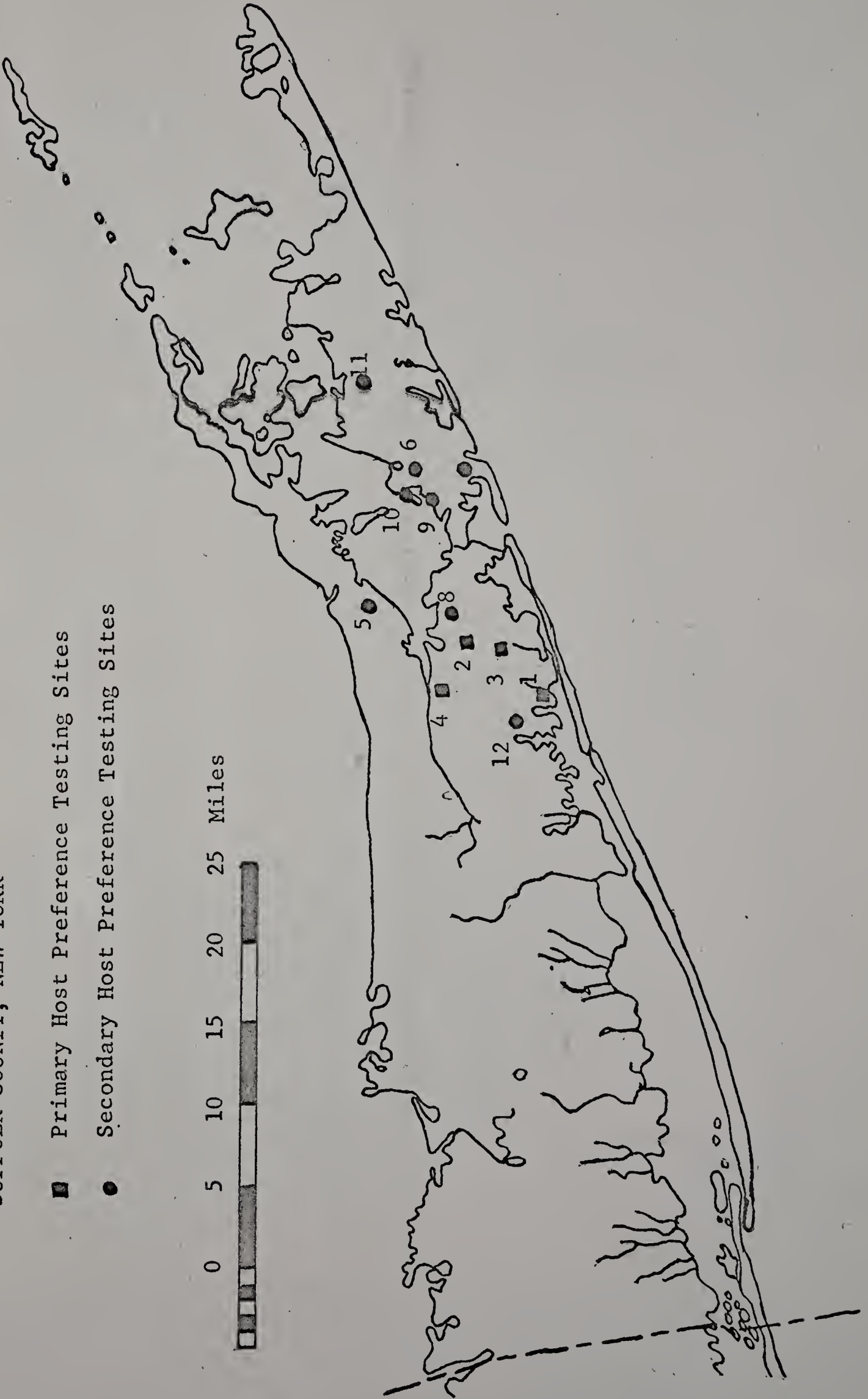


Fig. 6. A triad of host preference traps
operating in Maple Swamp, Flanders.

SUFFOLK COUNTY, NEW YORK

- Primary Host Preference Testing Sites
- Secondary Host Preference Testing Sites

0 5 10 15 20 25 Miles



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