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Studies on the biology and distribution of the Rockpool mosquito, Aedes atropalpus (Coquillett).

Seymour A. Maisey
University of Massachusetts Amherst

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University of Massachusetts

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INTRODUCTION

The rockpool mosquito, Aedes atropalous (Coquillett) is of interest to those engaged in public health work and to entomologists. It has been shown that this mosquito is capable of transmitting various disease-producing organisms to man and his domesticated animals. Its habitat is somewhat peculiar in that the species normally occurs in rock holes along streams or near other bodies of water. Comparatively few studies have been made of the life cycle or seasonal history of this mosquito. Preliminary investigations indicated that the only known method used prior to the undertaking of this study for determining the larval instars of this species was unsatisfactory. Many of the morphological features used in other species to distinguish fourth instar larvae from earlier instars cannot be utilized in A. atropalpus.

Earlier workers including Dyar (1903), Howard et al. (1912-1917), Carpenter et al. (1946) and Carpenter and LaCasse (1955) have described

the larval stages of this species as "slow in development," however Haufe (1952) indicated that development is probably rapid. All of the above-mentioned observations have been made in the field. Only a relatively small number of experiments have been conducted in the laboratory on the biology of A. atropalpus.

In order to attempt to solve some of the problems relating to this insect, Dr. Frank R. Shaw secured an exploratory grant from the United States Public Health Service in 1956. The writer was employed as a research assistant to conduct field and laboratory work. Certain aspects of the investigation were selected for his thesis. These related to an investigation of the criteria of value for distinguishing the larval instars, the determination of the effect of temperatures on the growth rate of the immature stages of this mosquito and observations on the distribution, both seasonal and geographical, of this insect.

REVIEW OF LITERATURE

Habits

The adult females of A. atropalpus are known to feed on both mammals and birds. They are persistent biters in their natural environment, and will attack during the daytime causing great personal discomfort. Davis (1940), Laird (1941) and Trembley (1947) found that the adults are reluctant to feed in captivity. During the daytime, adults are found resting under or on rocky ledges near their larval habitat as along rocky streams and ocean shores, and have been collected in homes and stables (1940 Massachusetts Mosquito Survey). Trembley (1947) reared A. atropalpus in the laboratory and observed that this species is autogenous, stenogamous, homodynamic and does not exhibit spanogamy. Earlier (1945), she had found that the adults required a high humidity and were quickly killed by drying. In the laboratory, mating was observed (Trembley, 1947) to take place at twilight. Despite almost

constant observation, she had never observed mating or oviposition in full daylight or under various intensities of light and colors. Trembley, therefore, concluded that mating and oviposition customarily takes place in total darkness.

The larvae are found throughout the summer in rockpools along streams or rivers, in rain-filled rockpools well removed from streams, near dams or natural waterfalls (See Fig. I and II) and along rocky shores of the ocean or lakes. On rare occasions, it has been found in tree-holes and in an artificial container (Hedeen, 1953). Kumm et al. (1940) observed larvae of this species in urns and flower vases, half full of rain water in a cemetery in Costa Rica. Shields (1938) recorded specimens from an abandoned septic tank; and Good (1945) reported specimens from a fire barrel on a temporary bridge. Kumm and Zuniga (1942) collected the larvae from a large cement water tank.

Dyar (1903) observed that the larvae have the ability to remain below the water surface for

Figure I. Typical habitat of Aedes atropalpus (Coq.).

Figure II. A rockpool in which Aedes atropalpus (Coq.) is found.

long periods feeding on algae and other minute organisms on the bottom and the sides of the rockpools. Hedeen (1953) observed that A. atropalpus has a definite preference for some of the common colonial fresh water algae. He found that if the common alga, Spirogyra, is introduced into a culture of atropalpus, the larvae will come from all corners of the container and begin to feed. He also noted that the blue-green alga, Oscillatoria, also attracts the larvae, but not to such a degree as Spirogyra. In order to determine other organisms that may normally be used for food by the larvae, Hedeen dissected intestines of many specimens collected in the field and found such organisms as diatoms, desmids, Pediastrum, Gonium, Euglena, Paramecium, Stentor, Blepharisma, Euplotes, and several species of rotifers. Various small worms were also found. Many small crustaceans were found with the Nauplius larval form of Cyclops, and Daphnia being the most prevalent. Hedeen concluded that the larvae are definitely omnivorous feeders although preferences

for the aforementioned fresh water algae were observed; and that the larval stage in nature does not limit itself to any particular diet but uses whatever food is available. Hedeen also observed in the laboratory that the larvae of this species is negatively phototropic. This he submits as a possible explanation of why this species spends long periods of time below the water's surface and is frequently observed under various objects found on the bottom.

Medical Importance

During 1939, workers at the Harvard Medical School demonstrated in the laboratory that *Aedes atropalpus* can transmit the Eastern Equine Encephalomyelitis (Carpenter *et al.*, 1946). This is a disease that infects both man and animals. The virus causes an inflammation of the brain resulting in nerve cell destruction, apathy, muscular weakness, and lethargy passing into more or less profound somnolence. Death is the final result among animal victims and a high percent mortality occurs

among young and old humans. Based upon collections made by the 1940 Massachusetts Mosquito Survey, of the six vectors of this disease known to exist in Massachusetts, it is about fourth in numerical importance. Laird (1941) found that A. atropalpus is susceptible to Plasmodium lophurae which infects ducks and other domestic fowl. Trembley (1946) found that A. atropalpus is highly susceptible to Plasmodium gallinaceum and can easily transmit this parasite to healthy fowl in the laboratory. At the present time, it is not known definitely whether or not it is a vector in nature. Because of the limited collections previously made of A. atropalpus and its restricted distribution, this species has not been considered to be an important vector of the disease.

General Taxonomy

Coquillett (1902) was the first to describe the species atropalpus although he placed it in the genus Culex. He described the adult male and female and gave a brief account of where the species

was initially discovered. Evidently, the larvae were not found because they were not described.

In 1906, Dyar and Knab transferred atropalpus from the genus Culex to Aedes. Theobald (1907) apparently was unaware of the Dyar and Knab paper when he stated that he was doubtful whether the species should be in the genus Culex. He said, "The structure of the male palp described by, and the long thin proboscis as figured by Smith seem to preclude it from Culex, and also the larval characters."

In 1908, Dyar and Knab described what they thought to be a new species, Aedes epactius from specimens reared from larvae collected in Mexico. They described the adult male and female and said that, "This species is closely related to Aedes atropalpus, but differs in the coloration of the thoracic markings." The larvae were not described. Howard, Dyar and Knab (1912-1917) still considered A. epactius to be separate species, ... "due to different coloration in the adults, although the

larvae are similar and the life history seems to be identical." Dyar in 1921 indicated that epactius might be a form of atropalpus when he said that atropalpus occurs "possibly in Mexico (epactius Dyar & Knab)." In 1922, Dyar reduced epactius in taxonomic rank by saying, "The form is doubtless to be considered a race of atropalpus."

In 1921 Dyar described what he thought to be a new species, Aedes perichares from specimens obtained from Costa Rica. Dyar (1928) recognized two races of A. atropalpus, A. atropalpus atropalpus from the Atlantic States and A. atropalpus epactius from Arizona and Mexico, although he still considered A. perichares a separate species. Edwards (1932) in his Genera Insectorum listed A. perichares as a synonym of the variety epactius of the species atropalpus. However, Lane (1953) still considers A. epactius to be a distinct species with perichares being a synonym of epactius. Other workers including Matheson (1944) and Carpenter and LaCasse (1955) do

not recognize Dyar's, Edwards' or Lane's interpretations as being valid but consider both epactius and perichares to be synonyms of A. atropalpus.

In summarizing, there are thre main opinions as to the relationships of A. atropalpus, A. epactius and A. perichares. They are:

1) Aedes atropalpus is not differentiated into races. The species has two synonyms, A. epactius and A. perichares. Matheson (1944) and Carpenter and LaCasse (1955).

2) The species A. atropalpus is represented by two distinct races, A. atropalpus atropalpus and A. atropalpus epactius. Dyar (1922). A. perichares is a synonym of the race epactius. Edwards (1932).

3) There are two species, A. atropalpus and A. epactius with A. perichares a synonym of epactius. Lane (1953).

Larval Instars

Dyar (1902) described the egg, fourth instar larva and pupa. In his description of the larva, he mentioned that on the anal segment the ventral brush is confined to the barred area, but that in the second and third instars the tufts are before the barred area. Dyar did not give any data to support this nor did he define the term barred area. Various culicidologists have used this term in their descriptions of other mosquito larvae, but this term has not been clearly defined.

In 1903, Dyar described the first, second, and third instar larvae, but did not give any criteria for distinguishing the larval instars except for repeating the statement concerning the barred area. Howard et al. (1912-1917) also mentioned that in the fourth instar the small tufts of the ventral brush do not precede the barred area.

Of the relatively few workers who have

published papers on A. atropalpus following Dyar, only Barr (1958) in his Mosquitoes of Minnesota gave a very brief description of the third instar. In this, he distinguished the fourth instar from the third by the head capsule width, range or number of comb scales, and number of branches in the siphonal tuft. Nothing was mentioned in his description about the second instar.

Temperature Studies on the Immature Stages

Since it is known that temperature greatly affects chemical and physical phenomena, it is not surprising to find it playing an important part in the life of organisms. Pearls (1914) proved Sanderson's (1910) theory that the rate of insect development is affected by temperature and, other factors being constant, this rate increases in direct proportion to the increase in temperature, within the normal limits of development. This is the present belief among entomologists and biologists.

Very little work has been done on the effect of temperature on the rate of growth of the immature stages of mosquitoes in general, and the only thorough investigation on the subject was made by Huffaker (1944) on Anopheles quadrimaculatus Say. He found a significant shift in the optimal temperature from about 91.9° F. to 86.5° F. as development progresses from the embryonic to the pre-imaginal phases. The optimal developmental temperature was found to be about 88° F. and the time required to complete development at this temperature was 7.3 days. The threshold of development was about 45° F. The lowest generally lethal high temperature for this mosquito was about 95° F., and the highest generally lethal low temperature was about 52° F. He also noted that high temperatures have an accumulative injurious effect.

Bar-Zeev (1958) studied the effect of temperature on the growth rate and survival of the immature stages of Aedes aegypti (L.). He

found that development from newly hatched larva to adult was most rapid at 90° F., and the time required to complete development at this optimum temperature was 6.4 days. The threshold of development was between 48° and 50° F. The lowest minimum lethal temperature was about 55° and the highest minimum lethal temperature was between 97° and 100° F. He also found that temperatures above 90° F. have a cumulative injurious effect on the immature stages of this mosquito.

Various workers have stated that the larva of A. atropalpus is slow in development, although Haufe (1952) said that"development is probably rapid since pool temperatures during the day rise quickly and remain high until the evening." These opinions were presumably based on comparisons with other species. Investigations prior to this study, on the relationship of temperature to the rate of development of the immature stages of A. atropalpus were those by Trembley and Hedeen.

Trembley (1945) reared larvae of A. atropalpus in the laboratory at temperatures fluctuating

from 74° F. to 84° F. and found that pupation occurred within nine days to two weeks.

Trembley (1947) in further studies on the biological characteristics of laboratory-reared A. atropalpus reared two separate colonies. The adults in one colony were offered blood at approximately two-week intervals; cellucotton soaked in a ten percent dextrose solution was always available. The adults in the other colony were not allowed access to blood, and were given only the dextrose solution on cotton. The larvae in both colonies were reared at temperatures fluctuating from 78.8° F. to 86° F., and maintained generally between 80.6° F. and 82.4° F. The larvae in the colony that was allowed access to blood reached the pupal stage in four to nine days, with a mean of six and three-tenths days and a mode of seven days. The larvae in the colony that was not allowed access to blood reached the pupal stage in five to twelve days, with a mean of seven and one-tenth days and a mode of eight days. The pupal stage in the colony

allowed access to blood ranged from twenty-four to fifty-three hours with a mean of forty-five and one-tenth, and a mode of forty-eight. The pupal stage in the colony not allowed access to blood ranged from twenty-four to fifty-three hours with a mean of forty-four and three-tenths, and a mode of forty-eight.

Hedeen (1953) reared A. atropalpus on a limited scale in the laboratory. His data showed that the length of life from newly hatched larvae to adults ranged from seven to ten days, with a mean of eight and one-quarter days. However, he did not indicate in his paper at what temperature the larvae and pupa were reared.

Trembley (1955) in her bulletin on mosquito culture techniques indicated that the larval stage of A. atropalpus at 71.6° F. ± 3.6° F. ranged from ten to twelve days, with a mode of ten days. The pupal stage ranged from one to three days.

Cartocraft Desk Outline Map, North America No. 18005

Distribution of Aedes atropalpus (Coq.); showing state distribution in the United States and Mexico, provinces in Canada, and countries in Central America.

(Based upon Carpenter and LaCasse, 1955)

Scale of Miles
0 200 400 600 800

Modified Conic Projection

Printed in U. S. A.

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GEOGRAPHICAL DISTRIBUTION

According to Carpenter and LaCasse (1955), A. atropalpus is known from the Eastern United States west to New Mexico, from southern Canada, Mexico and El Salvador. In addition Dyar (1921) had earlier recorded the species from Costa Rica and later (1928) extended the range to Nicaragua. Rees and Nielsen (1955) first reported this species from Utah. Harry D. Pratt, in October, 1927 collected larvae of Aedes atropalpus epactius from Guatemala City, Guatemala which represents a new distribution record.

Published Locality Records

Country	Locality
Canada(Haufe, 1952; Twinn, 1949; Carpenter, 1949; Gibson, 1937)	Goose Bay, Labrator-1949 Chaadiere River, Beauceville, Quebec-July 1936 Ottawa River, Fitzroy Harbour, Ontario-Sept. 1936
Costa Rica(Kumm et al., 1940; Dyar, 1921)	Liberia in the province of Guanacaste-1940 Ciruelas-Oct. 1920
El Salvador(Kumm and Zuniga, 1942)	Tunel de la Muralla, Chalatenango-1940 Sonsonate, Sonsonate-1940 Alegria, Usulutan-1940

Country	Locality
Mexico(Dyar and Knab, 1908; Aitken, 1942)	Almoloya, Oaxaca-1908 Cordoba, Vera Cruz-1908 Peninsula of Lower California-1942
Nicaragua (Dyar,1928)	
United States Arizona (Dyar, 1922)	Sabino Basin, Catalina Mts.-Aug. 1918
Arkansas(Carpenter, 1941)	Cedar Creek in Petit Jean State Park on Petit Jean Mt., Pope County-Sept. 1938
Connecticut(Matheson, 1945)	Double Beach, Bradford-July 1904
District of Columbia (Dyar, 1922; Good, 1945)	Chain Bridge-Aug. 1914 Roosevelt Island-Sept. 1942
Georgia(Carpenter, 1950; Stough et al. 1949)	Furman Shoals on the Oconee River, Baldwin County-March 1948 Tallulah River near Persimmon, Rabun County-July 1947 War Womans Creek near Pine Mt., Rabun County-July 1947
Kansas(Beadle, 1944; Anonymous, 1951)	Cowley County-1944 Allen County-1951 Cherokee County-1951 Montgomery County-1951

Country	Locality
Maine(Bean, 1946)	Bar Harbor-Aug. 1929 Georgetown-July 1944 Ft. Levett, Portland-Sept. 1944 Stillwater branch of the Penobscot River-Sept. 1906 Orono-1911
Maryland (Carpenter, 1950; Dyar, 1922)	Great Falls-Aug. 1903 Plummer's Island, Montgomery County-May 1902 Potomac River (one mile east of Harpers' Ferry) Washington County-Sept. 1947 Stubblefield Falls on the Potomac-Sept. 1904
Massachusetts(Dyar, 1922; Feemster and Getting, 1940)	Cummington-July 1903 Westfield-July 1903 Webster-1939 Barnstable County-1939 Essex County-1939 Franklin County-1939 Hampshire County-1939 Middlesex County-1939 Worcester County-1939
Minnesota(Owen, 1937)	Jay Cook State Park-July 1932 Knife River, Lake Superior-July 1934 Sucker River-July 1935
Missouri(Gurney, 1943)	Camp Crowder-July 1942
New Hampshire (Blickle, 1952; Johnson, 1925; Dyar, 1922)	Belknap-June 1952 Coos-June 1952 Grafton-June 1952

Country	Locality
New Hampshire (cont.)	Rockingham-June 1952 Stratford-June 1952 Center Harbor-Sept. 1902 Mt. Ossipee-1902 Mt. Monadnock, Cheshire and Hillsboro Counties-1917
New Jersey(Schmitt, 1942)	(No available locality records for this state)
New Mexico(Barber, 1939)	Last Chance Canyon-Aug. 1936
New York (Carpenter, 1950; Barnes et al., 1950)	Plattsburg, Clinton County-(No Date) Ausable Chasm, Essex County - (No Date) Haines Falls, Greene County - (No Date) Old Forge, Herkimer County - (No Date) Deferret, Jefferson County - (No Date) Forestport, Oneida County - (No Date) New York City, Queens County - (No Date) McLean, Tompkins County - (No Date) New Rochelle, Weschester County-(No Date) Crown Point, Essex County -May 1946 Tupper Lake, Franklin County -Aug. 1904
North Carolina(Carpenter, 1950; Schoof and Ashton, 1944)	Cullosja River between Franklin and Highlands, Macon County-July 1947

Country	Locality
North Carolina (cont.)	Cashiers, Jackson County -July 1947 Chimney Rock-Sept. 1939 Mt. Gilead, Eurie Dam-Aug. 1938
Oklahoma(Rozeboom, 1942)	Wichita National Forest -1942 Hinton-(No Date)
Pennsylvania(Dyar, 1922)	Shenks Ferry, Lancaster County-Oct. 1902
Rhode Island(Felton et al., 1950; Knutson, 1942)	Jamestown-(No Date) Point Judith-(No Date) Narragansett-1942
South Carolina(Carpenter, 1950)	Chattanooga River (approx. ¼ mile north of bridge on U.S. Highway 76 near Longcreek), Ocanee County -July 1947
Texas(Rueger and Druce, 1950; Hedeen, 1953)	Camp Bowie-April 1945 Camp Hood-May 1945 Camp Wolters-April 1945 Austin,Travis County-1953 Round Rock, Williamson County-1953 Fredericksburg, Gillespie County-1953 Junction, Kimble County-1953 Sheffield, Pecos County-1953 San Antonio, Bexar County -1953
Tennessee(Shields, 1938; Arnold, 1940)	Kenzel Springs-1934-1936 Knoxville-1934-1936 Neubert Springs-1940

Country	Locality
Vermont(Felton et al., 1950; Johnson, 1925)	Pittsford-(No Date) Hanover Area-1908
Utah(Rees and Nielsen, 1955)	Along the Colorado River-1955
Virginia(Carpenter, 1950; Dyar, 1922)	George Washington National Forest near Rawley Springs -Oct. 1947 Richmond-1901 Difficult Run-Aug. 1906
West Virginia (Carpenter, 1950)	Shenandoah River (approx. 5 miles east of Charles Town), Jefferson County -Sept. 1947
Wisconsin(Dickinson, 1944)	Jim Falls-1944 Chippewa River-1944

New Locality Records

Canada	Cooper, Ontario-Aug. 1955 (J. R. Vockeroth) Tadoussac, Quebec-Aug. 1939 (M. E. Smith) Norway Bay, Quebec-Aug. 1938 (G. E. Shewell) St. Gedeon, Lake St. John, Quebec-July 1939 (M. E. Smith)
Guatemala	Guatemala City (water around tree bases)- Oct. 1957 (H. D. Pratt)
United States Georgia	Milledgeville-(No Date) H. D. Pratt) Tallulah Gorge, Tallulah- Mar. 1950 (Cole & Wall)

Country	Locality
Massachusetts	Along the coast in the towns of Hull and Scituate-1955 (L. F. Wells, Jr.)
	Chesterfield Gorge, Chesterfield-May 1957 (S. A. Maisey & F. R. Shaw)
	Charlemont-Sept. 1934 (Mass. Mosq. Survey)
	Cohasset-Aug. 1958 (L. F. Wells, Jr.)
	Green River, Colrain-May 1956 (F. R. Shaw)
	Loudville (7 miles west of Northampton on Rt. 66 below dam)-April 1957 (S. A. Maisey & F. R. Shaw)
	Rockport-Aug. 1957 (M. E. Smith)
	Turners Falls (below Turners Falls dam)-Aug. 1957 (S. A. Maisey)
	Waconah Falls, Dalton-Sept. 1956 (F. R. Shaw)
	Windsor Jams, Windsor-Sept. 1956 (F. R. Shaw)
Maine	Along shore near Anemone Cave, Mt. Desert I.-July 1955 (F. R. Shaw)
	Bernard-July 1956 (F. R. Shaw)
	Otter Cliff, Mt. Desert I.-June 1956 (F. R. Shaw)
	Pophan Beach-July 1956 (F.R. Shaw)
	Salmon Falls, Saco River, Bar Mills-June 1939 (J. F. Hanson)
	Sand Beach, Mt. Desert I.-June 1956 (F. R. Shaw)
	Thunder Hole, Mt. Desert I.-July 1955 (F. R.Shaw)

Country	Locality
Maine (cont.)	Big Brother Mt., Baxter State Park-July 1948 (M. E. Smith)
New Hampshire	North Monroe-April 1957 (F. R. Shaw) Woodsville-Aug. 1956 (F.R. Shaw)
Maryland	Gunpowder River, Baltimore County -May 1956 (G. B. Craig, Jr.)
New York	South Inlet, Webb-May 1951 (H. A. Jamnback)
Vermont	East Barnett-April 1957 (F. R. Shaw) Halifax Gorge, Halifax-Sept. 1956 (F. R. Shaw) Passumsic-Sept. 1956 (F. R. Shaw) Rt. 103, near Ludlow-Aug. 1956 (F. R. Shaw & R. J. Lavigne)
Texas	Onion Creek, Travis County-Sept. 1943 (D.E.E.)

METHODS AND PROCEDURES

Collection and Preparation of Larvae

Larvae of A. atropalpus were collected in various stages of growth during the summer of 1957 from rockpools located at Loudville, Chesterfield, Turners Falls and Westfield, Massachusetts, at East Barnett, Vermont and North Monroe, New Hampshire. The specimens were collected from the pools by using either a baster or dipper, depending upon the depth and width of the rockpool (see Fig. 3). The larvae were carried back in glass jars to the laboratory where a portion of them were killed by placing them in hot water and the remainder were used for biological observations.

The larvae that were killed were placed into a series of Syracuse watch-glasses, each one containing a particular concentration of alcohol (70%, 85%, 95% and 100%). The specimens remained in each concentration for ten minutes. They were next immersed in oil of wintergreen for twenty to thirty minutes and then mounted in Canada Balsam on microscope

Figure III. Collecting equipment.

slides. After approximately twenty-four hours on a slide warmer, the specimens were ready for microscopic examination.

Additional mounted specimens were contributed by Dr. Frank R. Shaw of the Department of Entomology, University of Massachusetts from his collection of atropalpus larvae obtained from Mt. Desert, Maine. Dr. Shaw also contributed to the study alcohol-preserved larvae sent to him by Dr. Harold B. Craig, Jr., formerly with the Army Chemical Center in Maryland. Dr. Craig collected the specimens at Gunpowder River in Baltimore County, Maryland. These were also mounted and examined.

Rearing Larvae to Known Instar

Eggs of A. atropalpus were collected from the water surface of rockpools that contained larvae of only this species of Aedes at Loudville, Massachusetts and brought back to the laboratory for rearing. Collection of the eggs was made either by placing a jar just below the water surface next to the scattered eggs and allowing the water

and eggs to flow into the jar or by placing a piece of paper toweling directly below the eggs and lifting the toweling slowly, allowing the water to drain off with the eggs remaining on the toweling. Rockpool water, which was green with algae, was also taken back to the laboratory. In the laboratory, the eggs were placed in a large glass jar containing water from the rockpools and allowed to hatch at room temperature. Upon hatching, the larvae were removed with an elongated eye-dropper and placed in individual numbered vials containing rockpool water (see Fig. 4). The vials were placed in a constant temperature cabinet at 80° F. The larvae were observed thereafter every twenty-four hours for molting. At this time the water in the vials was replenished with rockpool water and a very small amount of a mixture of powdered yeast and dogfood added. During the first few days the vials containing larvae were observed under a dissecting microscope for exuviae. If exuviae were found, they were removed with an

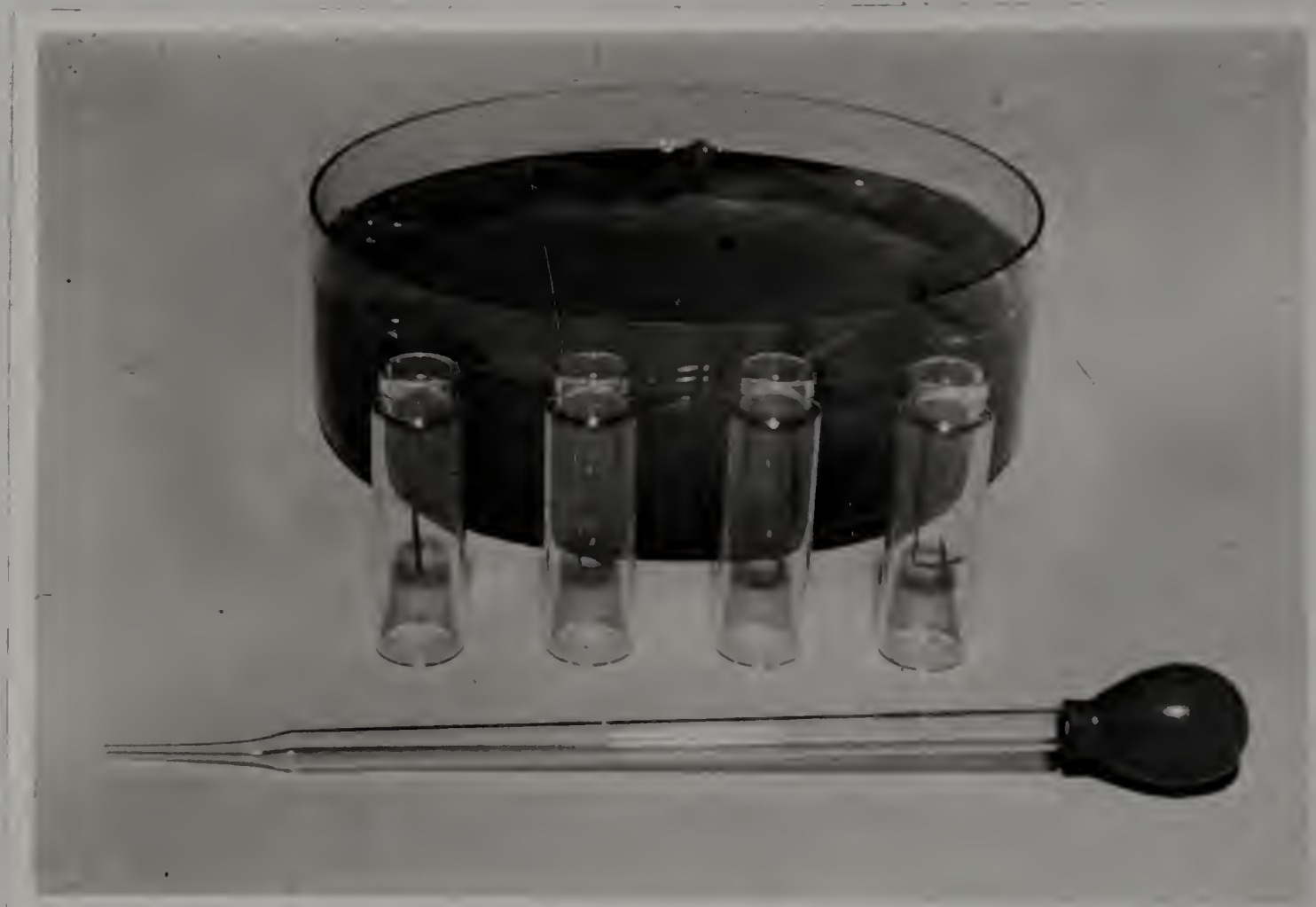


Figure IV. Rearing equipment.

eye-dropper, preserved in small vials which were marked with the corresponding number of the vial containing the larvae. The larva at this time was either killed in hot water or allowed to progress to the next instar. In this way, between twenty to twenty-five larvae of the second, third and fourth instar, with exuviae, were preserved in 70 percent alcohol for later examination. Due to the limited number of eggs available, first stage larvae were not killed and mounted. However, in order to secure data on this stage a number of first instar larvae were collected in the field, mounted and studied. Measurements were made of characters that had previously been shown to be of value for determining the later larval instars.

All examinations and measurements were made with a compound microscope having an ocular of 10x and an objective of 10x. A micrometer in the ocular was calibrated to 0.14 mm. per small division. The measurements were rounded off to three places and recorded in thousandths of a millimeter. Measurements of reared larvae were made without the use of

coverslips on the specimens. After the measurements of a particular structure were completed, the data were arranged in tabular form. By inspection, a class interval was then selected. The range in millimeters of each group shows the extremes of the measurements found for that particular group or instar, and does not necessarily coincide with the extremes of that group indicated by the class interval.

Rearing Larvae at Constant Temperatures

In order to conduct laboratory experiments on the effect of temperatures on the growth rate of the immature stages of A. atropalpus, constant temperature cabinets were needed. This posed a problem since this basic equipment for the desired range of temperatures was not available in the Department of Entomology. The problem was finally resolved through the graciousness and kindness of Dr. Warren Litsky, of the Department of Bacteriology, who permitted the use of their temperature cabinets for this study.

Most of the eggs used to rear larvae were obtained from Dr. Craig, who maintained a laboratory colony of A. atropalpus started from larvae collected at Big Gunpowder Falls, Maryland. Some eggs were collected at Loudville, Massachusetts, as mentioned previously. Because of the small number of eggs collected in the field, the larvae hatching from these eggs were all reared at 80° F.

The eggs were placed in a glass jar containing tap-water, or water deoxygenated by boiling, and allowed to hatch at room temperature. When deoxygenated water was used there was a larger percentage of hatch than when tap-water was used. This technique had been suggested by Dr. Craig and proved to be of value.

The newly hatched larvae were put into individual vials containing tap-water. The vials were then placed in a constant temperature cabinet (see Fig. 5) and the larvae reared to the adult stage. Thirty larvae were reared at each temperature

Figure V. Temperature cabinet.

except at 90° F., at which twenty-four larvae were reared. The number of larvae used in this test was less because of the small number emerging from eggs immersed in tap-water. The vials containing larvae in the constant temperature cabinet were observed thereafter at twenty-four hour intervals for exuviae. At the same time, the larvae were fed a small amount of powdered yeast and dogfood. Any food remaining from the previous feeding was removed with an eye-dropper to prevent the water from becoming turbid. Larvae were reared at constant temperatures of approximately 93°, 90°, 80°, 70°, 60°, 50°, and 45° F. ± 1° F.

BIOLOGICAL OBSERVATIONS IN THE FIELD

Since the general biology of *Aedes atropalpus* is known from observations made and recorded by Dyar (1903, 1904), Howard *et al.* (1912-1917), Trembley (1947), and Hedeen (1953) there seemed little point in repeating this work. However, in the course of these investigations, some seasonal observations of the life cycle were made and these are presented herewith.

During the summer of 1957, this investigator made numerous periodic collections of *Aedes atropalpus* at Loudville, Massachusetts. The larvae were first observed on April 17th and seemed to be in second and third instar. Thereafter, larvae were found throughout the summer in the rock-pools. The last observed occurrence of the immature forms was recorded on November 8th.

Dr. Shaw has collected larvae of this species in East Barnett, Vermont, as early as April 4th and as late as September 15th. In North Monroe, New Hampshire, the same collector has found specimens

from April 18th through September 15th. On Mt. Desert Island, Maine, this species was collected from May 12th through October 22nd inclusive.

During a collecting trip to Loudvills, Massachusetts on May 17th, 1958 this investigator was unable to get close to the rockpools because of water flowing over the dam and cascading over the rocks. At this time the rockpools were being thoroughly flushed out so that no larvae could have been present. On the next visit, which was on May 23rd, the water had been diverted by an opening or "gate" in the dam so it was possible to make observations of the rockpools. At this time the larvae in them were mainly in second instar. In a conversation with the owner of this area, the writer was informed that the owner personally had diverted the flow of water from over the dam on May 18th. On May 29th, the rockpools were observed again, and found to contain fourth instar larvae. Although the exact time between the diversion of the water and hatching of the eggs was not known, it had taken

a maximum of eleven days from egg to fourth instar larva under natural conditions in late May.

Observations by this investigator as to the site of oviposition confirm Dyar's (1904) observations. In the late fall the eggs are laid in patches firmly adhering to the sides of the rockpool while during the summer they are deposited on the surface of the water loosely and separately.

During the course of this study, other species of mosquito larvae were found in association with Aedes atropalpus. The most prevalent of such species was Culex restuans Theobald. Other members of the genus Culex that are found to occupy the same specific habitat are Culex pipiens Linn. and Culex territans Walker. Anopheles punctipennis (Say) is another common species found to be in association with Aedes atropalpus. The only other Aedes species found (on one occasion) by this investigator was Aedes vexans (Meigen). On one occasion Dr. Shaw found Aedes canadensis (Theobald) in rockpools near Sand Beach, Mt. Desert, Maine.

DEFINITION OF TERMS

The following is a list of terms and their definitions as they were applied in the measurements and observations of various morphological characteristics:

1. Width of head capsule - the widest portion of the head capsule, between the larval eyes (Fig. 7. HI).

2. Length of siphon- from its point of juncture with the eighth abdominal segement to its tip on the dorsal surface excluding the valves and acus (Fig. 6. AC).

3. With of siphon- the basal diameter of the diameter at its point of juncture with the eighth abdominal segment (Fig. 6. AB).

4. Antennal tuft- the tuft of hairs located between the distal and proximal areas of the antennae (Fig. 7. F)

5. Preantennal tuft- the tuft of hairs located on the head near the base of the antennae or head hair #7 (Fig. 7. C).

6. Siphonal tuft- one of a pair of ventro-laterally opposed hair-tufts located between one-half to two-thirds the length of the siphon Fig. 6. E).

7. Pecten- one of a pair of subventral, longitudinal rows of pointed teeth or spines extending towards ths apex from the base of the siphon (Fig. 6. D).

8. Barred area*- the entire area consisting of the bases of those hairs of the ventral brush each of which has a sclerotized transverse bar with an enlarged portion where the alveolus of the hair is inserted. In addition there are lateral sclerotizations (except in the first and second instar larvae) which may enclose all or most of these bars.

9. Siphonal index- the quotient derived from dividing the length by the basal diameter of the siphon.

10. Pentad hairs- the hairs located on the eighth abdominal segement sublaterally and posteriorly to the comb scales.

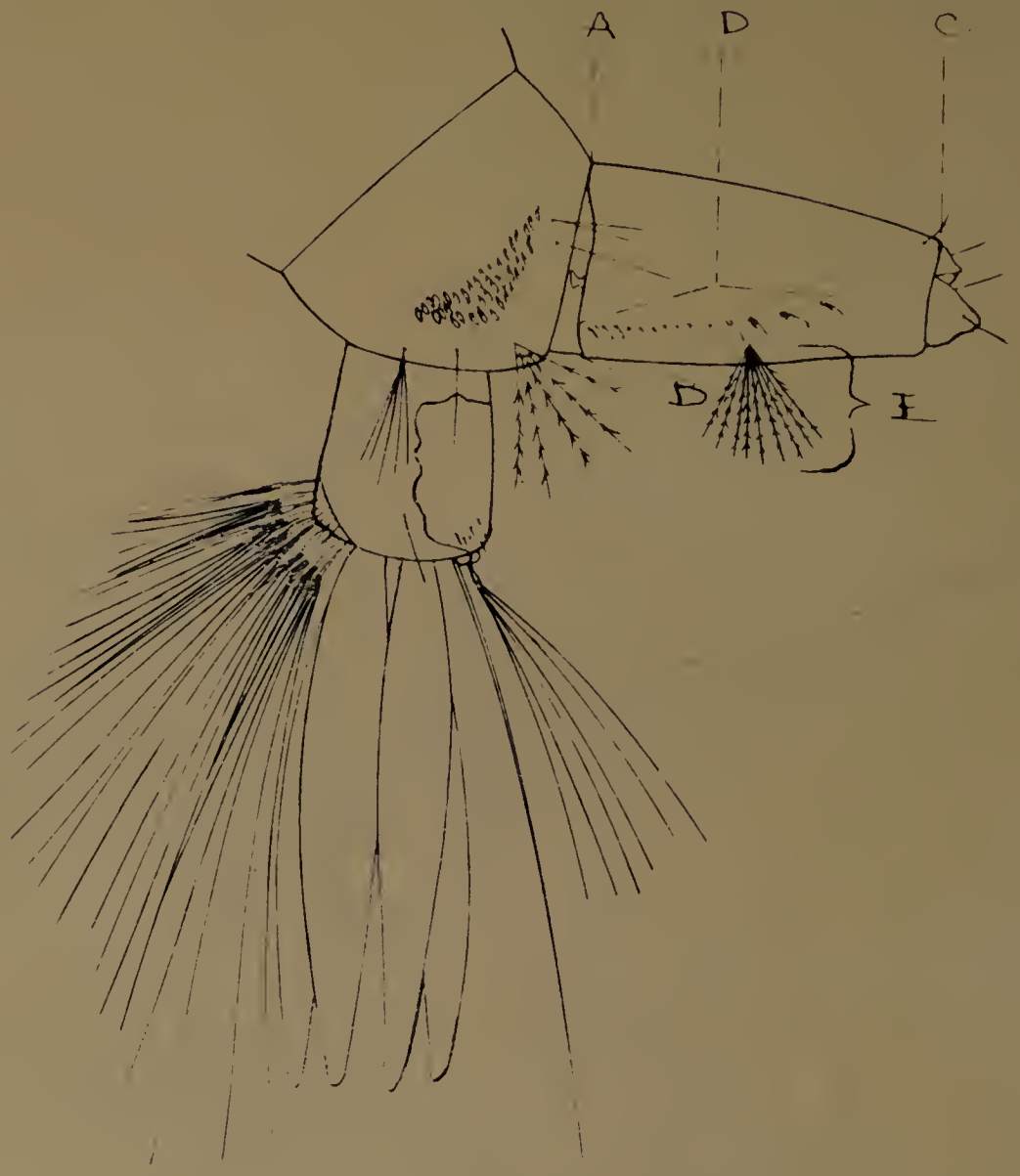
11.	Distance (%) of tuft along siphon-	the length of the siphon divided into the distance of the siphonal tuft from the base of siphon; that quotient multiplied by 100.
12.	Cratal brush-	tufts of the ventral brush arising from the barred area.

*The problem of the more proximal hairs in the ventral brush is a puzzling one and various workers (personal correspondence, 1958) have not been consistent in defining this term. Stone, Breland, and Bohart in their definitions of this term have omitted reference to the proximal hairs of the ventral brush which may or may not have sclerotized bars but are detached from the lateral sclerotizations. Belkin, however, in his definition considers the detached hairs to be outside the barred area and states that they have a very small basal sclerotization which is not extended as a transverse bar. In the current investigation,

observations have disclosed that usually the detached hairs of the third and fourth instar larvae do have transverse bars (see Table #8 in RESULTS). In addition, in most second instar larvae the hairs of the ventral brush have transverse bars but lack the lateral sclerotizations. Therefore, according to the definition of earlier workers, second instar larvae would not possess a barred area. In the present definition of the barred area, this investigator has included the phrase 'all or most' of the bars, so that if the alveolus of the proximal hairs consists of a sclerotized bar or portions of the bar, even though not enclosed within the lateral sclerotizations, those hairs are considered to lie within the barred area.

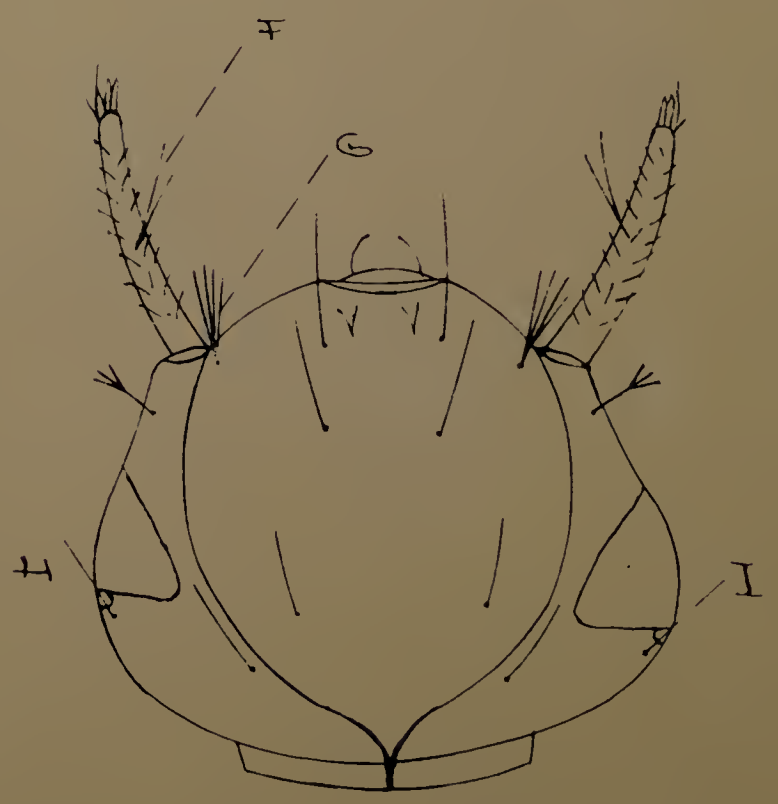
AEDES ATROPALPUS (COQUILLETT)

FIGURE #6



VENTRAL VIEW OF TERMINAL SEGMENTS

FIGURE #7



DORSAL VIEW OF HEAD CAPSULE

RESULTS AND THEIR INTERPRETATIONS

Head Capsule Width

Table #1 shows the results of measurements made of the head capsule width of larvae of A. atropalpus reared under controlled conditions to a known instar. Upon examination of the data it becomes apparent that these measurements are relatively constant within each of the three later instars. This observation corresponds with that of Dyar (1890) who, in his study of lepidopterous larvae, found that the sclerotized parts did not change in area during a stadium but rather that the change occurred with ecdysis. In the three instars indicated there is a marked difference between the extremes.

Measurements made of 249 specimens of field-collected larvae (Table #2) show that they also separate into four distinct groupings, when the same class interval is used. Measurements of first instar larvae were included for the purpose of completion. As one may observe, there is no overlap of the

Table #1- Head Capsule Widths of Reared Larvae

Width in mm.	Number of Individuals 2nd Instar	3rd Instar	4th Instar
.378-.419	2		
.420-.461	15		
.462-.503	8		
.504-.545			
.546-.587		2	
.588-.629		3	
.630-.671		5	
.672-.713		8	
.714-.755		6	
.756-.797			
.798-.839			1
.840-.881			3
.882-.923			5
.924-.965			7
.966-1.007			4
1.008-1.049			2
Total No. of Individuals	25	24	22
Range in mm. of Each Group	.384-.484	.567-.742	.832-1.064
Mean in mm.	.448	.672	.953

Class Interval of .042 mm.

Table #2- Head Capsule Widths of Field-Collected Larvae

With in mm.	Number of Individuals 1st Instar			
.252-.293	9			
.294-.335	4			
.336-.377				
.378-.419		6		
.420-.461		14		
.462-.503		9		
.504-.545				
.546-.587				
.588-.629			1	
.630-.671			5	
.672-.713			12	
.714-.755			15	
.756-.797			2	
.798-.839				1
.840-.881				17
.882-.923				21
.924-.965				21
.966-1.007				30
1.008-1.049				40
1.050-1.091				25
1.092-1.133				3
1.134-1.175				1
Total No. of Individuals	14	29	35	159
Range in mm. of Each Group	.252-.322	.389-.489	.616-.784	.819-1.170
Mean in mm.	.285	.442	.707	.980

Class Interval of .042 mm.

extremes although the difference between field-collected third and fourth instars is not as marked as in those reared under laboratory conditions.

Width of Siphon

Measurements of the width of the siphon (Table #3) also show a distinct grouping of each instar, with no overlapping of the extremes, although the difference between second and third instar is not as conspicuous as that between third and fourth instar. When these are compared with the measurements made from field-collected specimens (Table #4), a similar grouping of larvae occurs, indicating a separation of the instars, although there is less difference between the extremes than in the reared larvae.

Length of Siphon

The measurements of the length of siphon of reared larvae (Table #5) show that there is an overlap between second and third instar although the

Table #3- Width of Siphon of Reared Larvae

Width in mm.	Number of Individuals 2nd Instar	3rd Instar	4th Instar
.098-.125	6		
.126-.153	19		
.154-.181		2	
.182-.209		6	
.210-.237		14	
.238-.265		2	
.266-.293			
.294-.321			5
.322-.349			1
.350-.377			7
.378-.405			4
.406-.433			4
.434-.461			1
Total No. of Individuals	25	24	22
Range in mm. of Each Group	.109-.150	.162-.265	.294-.448
Mean in mm.	.129	.213	.368

Class Interval of .028 mm.

Table #4- Width of Siphon of Field-Collected Larvae

Width in mm.	Number of Individuals			
	1st Instar			
.042-.069	6			
.070-.097	15			
.098-.125	3	11		
.126-.153		20		
.154-.181			5	
.182-.209			12	
.210-.237			17	
.238-.265			18	
.266-.293				24
.294-.321				33
.322-.349				25
.350-.377				22
.378-.405				22
.406-.433				11
.434-.461				5
.462-.489				1
.490-.517				3
.518-.545				1
Total No. of Individuals	24	31	52	147
Range in mm. of Each Group	.056-.102	.111-.153	.154-.263	.266-.529
Mean in mm.	.075	.127	.219	.346

Class Interval of .028 mm.

Table #5- Length of Siphon of Reared Larvae

Length in mm.	Number of Individuals 2nd Instar	3rd Instar	4th Instar
.210-.237	1		
.238-.265	5		
.266-.293	18	1	
.294-.321	1		
.322-.349		3	
.350-.377		2	
.378-.405		4	
.406-.433		8	
.434-.461		6	
.462-.489			
.490-.517			1
.518-.545			1
.546-.573			1
.574-.601			2
.602-.629			4
.630-.657			3
.658-.685			5
.686-.713			2
.714-.741			2
.742-.769			1
Total No. of Individuals	25	24	22
Range in mm. of Each Group	.210-.294	.286-.448	.504-.742
Mean in mm.	.270	.398	.638

Class Interval of .028 mm.

separation of third and fourth is apparent. In field-collected larvae (Table #6) there is no overlap between presumably second and third instar larvae with the separation of third and fourth instar being slight.

One may observe in comparing the measurements made of reared and field-collected larvae that the intervals between the ranges in the second, third and fourth instar of field-collected larvae are smaller than in the reared larvae. The reason for this may be due to the variety of ecological factors that affect the normal population causing individuals to be further from the mean as compared with specimens reared under controlled conditions. In general, the field-collected larvae showed greater variation, particularly in the fourth instar, which may have been due to more optimum conditions in the natural environment.

Other Morphological Characters Studied

Table #7 is a summary of other morphological characters observed and recorded by this investigator.

Table #6- Length of Siphon of Field Collected Larvae

Length in mm.	Number of Individual 1st Instar			
.042-.069	1			
.070-.097	17			
.098-.125	6			
.126-.153				
.154-.181		1		
.182-.209		9		
.210-.237		4		
.238-.265		15		
.266-.293		4		
.294-.321		4		
.322-.349				
.350-.377			2	
.378-.405			9	
.406-.433			10	
.434-.461			7	
.462-.489				2
.490-.517				1
.518-.545				4
.546-.573				10
.574-.601				22
.602-.629				15
.630-.657				13
.658-.685				14
.686-.713				34
.714-.741				17
.742-.769				13
.770-.797				11
.798-.825				2
.826-.853				2
.854-.881				2
Total No. of Individuals	24	37	28	162
Range in mm. of Each Group	.046-.112	.168-.311	.378-.448	.462-.868
Mean in mm.	.085	.239	.412	.666

Class Interval of .028 mm.

Table #7- Observations on some of the Morphological Characteristics of Reared Larvae of Aedes atropalpus (Coq.).

	Branches of Siphonal Tuft	Number of Pecten Teeth	Branches of Antennal Tuft	Branches of Preantennal Tuft	Branches of 3rd Pentad	Branches of 5th Pentad	Siphonal Index	Distance of Tuft Along Siphon-Percentage of Length	No. of Tufts in Cratal Brush
2nd Instar									
Range	2-5	9-17	1-3	2-3	2-4	2-3	1.68-2.49	30.58-62.78	7-12
Mean	2.96	11.6	1.84	2.04	2.88	2.2	2.09	48.42	9.57
No. of Ind.	25	25	25	24	25	25	25	25	23
3rd Instar									
Range	4-7	12-24	1-3	2-5	3-8	2-5	1.29-2.5	41.28-61.11	8-13
Mean	5.33	17.26	2.05	3.25	5.04	2.7	1.88	49.15	10.59
No. of Ind.	24	23	21	20	23	23	24	23	22
4th Instar									
Range	7-9	9-26	1-3	3-5	6-12	2-6	1.43-2.10	44.47-62.26	8-13
Mean	8.18	18.45	2.10	4.24	8.61	3.7	1.75	52.04	11.05
No. of Ind.	22	22	20	21	23	23	22	21	21

The number of branches of the siphonal tuft was used by Barr (1958) in distinguishing the fourth instar from the third. Studies by this investigator of second, third, and fourth instar larvae indicated that this feature exhibits considerable overlapping in the succeeding instars and therefore cannot be used as a reliable criterion. Another character used by Barr was the number of comb scales. Although no data were recorded, preliminary investigations showed also results similar to those recorded for the branches of the siphonal tuft. The other characters observed and recorded show clearly that they also cannot be used as reliable criteria for distinguishing the larval instars of A. atropalpus due to extreme variation and overlapping.

The Barred Area

Dyar (1902) claimed that the fourth instar can be distinguished from the third and second by the ventral brush being confined to the barred area. Table #8 shows the results of observations

Table #8- Distribution of Ventral Brush on the Barred Area

REARED LARVAE

Larval Instar	Confined	Not Confined	No. of Specimens Observed
2nd	1	24	25
3rd	9	13	22
4th	20	1	21

FIELD-COLLECTED LARVAE

Larval Instar*	Confined	Not Confined	No. of Specimens Observed
2nd	0	35	35
3rd	37	13	50
4th	52	3	55

*The grouping of field-collected larvae was based on observations made on reared larvae of known instars of A. atropalpus and other species.

made on the barred area of second, third and fourth instar larvae. On fourth instar larvae in both reared and field-collected specimens the ventral brush is confined to the barred area with the exception of four individuals out of seventy-six examined. In third instar larvae of reared specimens, 41 percent of the individuals had the ventral brush confined to the barred area. In field-collected specimens of presumably third instar, 74 percent of the individuals were confined to the barred area. These results repudiate Dyar's claim, showing that the confinement of the ventral brush to the barred area cannot be used as a reliable criterion.

The first instar larva of this species can be readily distinguished from later instars by the presence of the egg breaker on the dorsal surface of the head. In addition the siphonal tuft is composed of a single hair and there is no ventral brush. Therefore first instar larvae were not extensively examined in this investigation.

Temperature Studies

The results of the temperature experiments (Table #9 and Fig. 8) conducted by this investigator on the immature stages of A. atropalpus corroborate the fact that there is an increase in the rate of development of the immature stages as the temperature increases, within the normal limits of development. The observed data for five temperatures for the time required from eclosion to emergence varied from 744 hours at 60° F. to 184.5 hours at 93° F.

There are definite indications that the optimum temperature from eclosion to emergence for this species is near 90° but less than 93° F. At approximately 93° F., the maximum number of hours required by the larval stage begins to increase (177 hours at 93° F. as opposed to 168 hours at 90° F.). The average number of hours in the larval stage exhibits a smaller proportional increase in the time required for development when compared to the increases at other temperatures. At this same temperature, the fourth instar larvae shows an increase

Table #9- Duration of Immature Stages of *A. atropalpus* at Different Constant Temperatures.

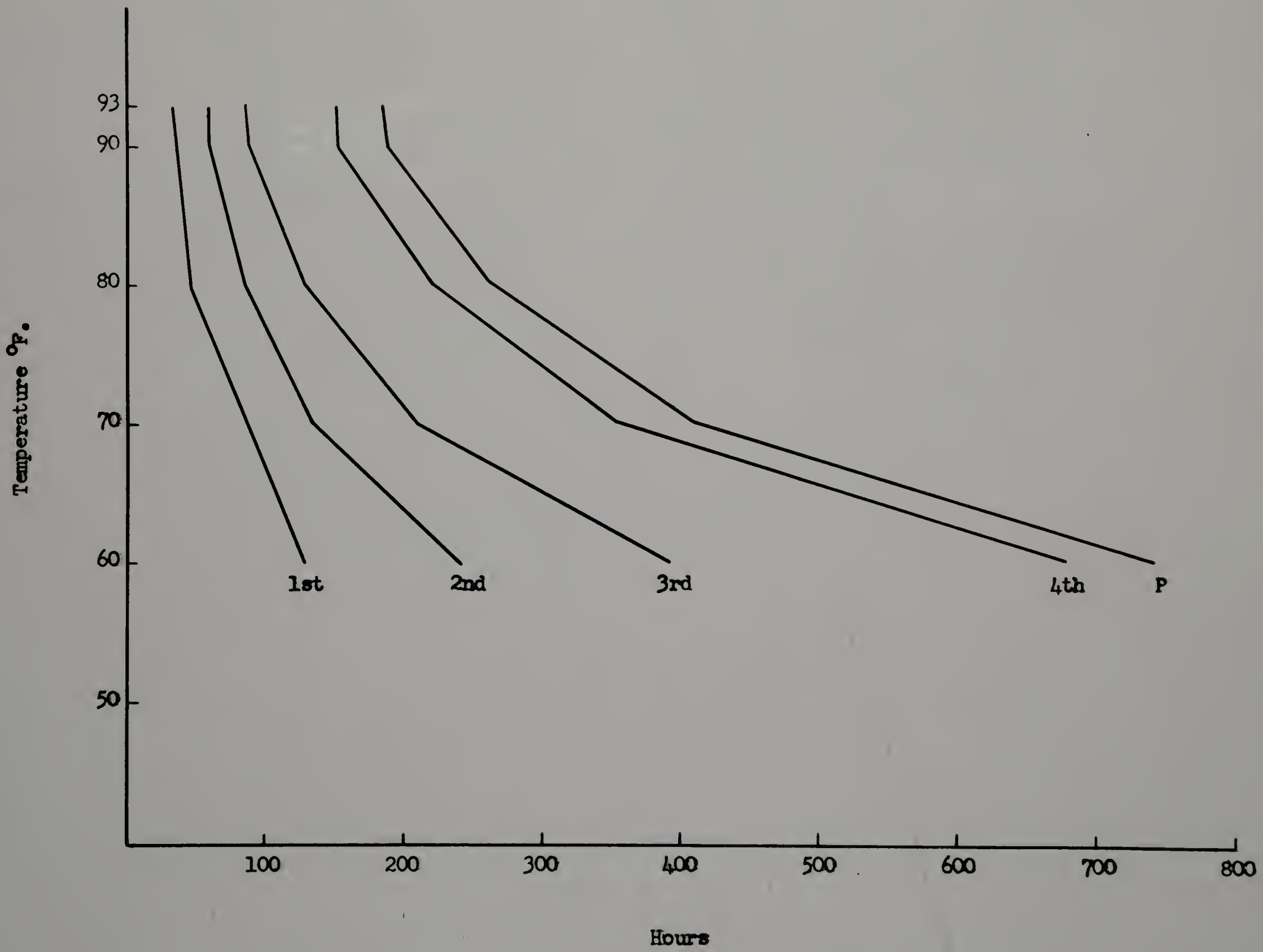
Temp. (°F.)	No. of Newly Hatched Larvae	LARVAL STAGE I H	I %	II H	II %	III H	III %	IV H	IV %	Range of Larval Stage in Hrs.	Ave. No. of Hrs. in Larval Stage	PUPA Ave. No. of Hrs.	PUPA %	PUPA Range of Pupal Stage in Hrs.	Percent of Initial Population that completed development	T
45	*1 30														0	
50	*2 30														0	
60	30	128	17.2	112	15.1	152	20.4	288	38.7	648-720	680	64	8.6	48-72	10	744
70	30	86.4	20.9	48	11.6	76.8	18.6	146.4	35.5	312-456	357.6	55.2	13.4	24-72	33.3	412.8
80	30	46.4	17.4	37.2	14.0	46.8	17.6	91.2	34.3	168-312	221.6	44.4	16.7	36-48	33.3	266
90	24	35.5	18.2	24	12.3	32.7	16.8	62.6	32.2	144-168	154.9	39.8	20.5	24-48	45.8	194.6
93	30	33	17.9	24	13	27	14.6	66	35.8	129-177	150	34.5	18.7	24-48	26.6	184.5

H = Average no. of hours for the stage indicated.
% = Percent of total time it took for the stage indicated - $\frac{100H}{T}$

T = Average no. of hours from hatching to emergence of adult stage.

*1 - None of the newly hatched larvae was able to reach the next stage.
*2 - Only one specimen reached 2nd instar after 408 hrs.

Figure 8. Average number of hours taken for newly hatched larvae of Aedes atropalpus to complete successive stages of development (1st-4th larval stages; P = Pupa).



of 3.4 hours in developmental time, which indicates that at this temperature there is a cumulative injurious effect. Although the average number of hours from hatching to emergence of the adult continues to decrease, the decrease is much less proportinally than at the lower temperatures. The percent of the initial population that completed development at 93° F. showed a significant decrease (26.6 percent) from those completing development at 90° F. (45.8 percent) which also indicates that at 93° F. there is a detrimental effect. The time required to complete development near the optimum temperature is 194.6 hours.

At 50° F. only one of the newly hatched larvae was able to reach the second instar after 408 hours, and it died after 504 hours. At 45° F. none of the newly hatched larvae reached the second instar. Thus the threshold of development for this mosquito is between 45° and 50° F.

The general action of temperature as it is related to the speed of development is presented in Figure 8. The horizontal distance between

successive curves represents the duration of successive stages at the given temperature. The fourth stage was the longest. The time involved is nearly double that of the other instars; it averaged 35.3 percent of the total time of development. This is to be expected, for it is general knowledge that the fourth instar is the one in which a great portion of the body size is attained and which requires much more time in its completion.

The first instar was the second longest in duration of development, averaging 18.3 percent of the total time of development, followed by the third with 17.6 percent, then the pupal stage 15.6 percent, and finally the second instar which averaged the shortest with 13.2 percent. Although the data do not show the optimum temperature for the first, second, third larval and pupal stages, there are indications which show that it is in the vicinity of 93° F.

For many years it has been known that variable and constant temperatures produce different rates of insect metabolism. The majority of workers

(Huffaker, 1944) seem to favor the idea that insect development under variable temperatures is faster than that under constant temperatures. Therefore, under natural conditions, in which the temperature of the rockpools is variable, it is possible that the rate of development of A. atropalpus may be faster than that of the optimum constant temperature in the laboratory.

The above results on the temperature studies indicate that the speed of development of the immature stages of this species is influenced to a great extent by the temperature of the water, all other things being equal; and that the rate of development is not slow as was the belief of earlier workers. Similar experiments conducted by Bar-Zeev (1958) with Aedes aegypti, which is considered fairly rapid in development, and Huffaker (1944) with Anopheles quadrimaculatus when compared with A. atropalpus, indicate that atropalpus is slower in development at its optimum temperature (1.7 days and 0.8 days slower respectively) but not so much slower as to be considered "slow in development."

SUMMARY

The larvae of the rockpool mosquito, Aedes atropalpus (Coquillett), were reared and collected for the purpose of investigating whether there are criteria of value for distinguishing the larval instars. Preliminary studies indicated that the only known method used prior to this investigation was unsatisfactory. Larvae were reared from eggs to a known instar, killed and examined. Specimens were also collected in the field and brought back to the laboratory for examination. Of the numerous morphological characters examined, three were found to be of value in distinguishing the instars. They are; the width of the head capsule, width of siphon and length of siphon.

In reviewing the literature, this investigator found that the majority of previous workers indicated that this species is slow in development. This seemed contrary to what the writer has observed in the field. To clarify the matter, this investigator reared larvae at various constant temperatures

and observed the rate of development. It was found that at 90° F., which is the approximate optimum temperature for this species, A. atropalpus took 8.1 days from eclosion to emergence. When compared with other species, such as Aedes aegypti and Anopheles quadrimaculatus, which are fairly rapid in development at their optimum temperature, atropalpus is 1.7 and 0.8 days slower respectively, but not so much slower as to be considered slow in development.

Observations in the field as to the seasonal distribution in Massachusetts indicate that the larval form of this species can be found from mid-April to the beginning of November. During these observations, certain other species of mosquito larvae were found in association with A. atropalpus. They were Culex restuans, Culex pipiens, Culex territans, Anopheles punctipennis and on single occasions, Aedes vexans and Aedes canadensis were found.

Since A. atropalpus has been shown to be capable of transmitting various disease-producing

organisms, its distribution is of importance. Observations on the locality records of this species indicate that it is more common than previously thought; and that it is usually prevalent in rocky areas near bodies of water, although on rare occasions it has been found in artificial containers.

CONCLUSIONS

The results obtained by this investigator from the examination of the barred area of reared and field-collected larvae of Aedes atropalpus repudiate Dyar's (1902) claim that the fourth instar can be distinguished from the third and second by the ventral brush being confined to the barred area.

The data support Barr's (1958) findings that the fourth instar can be distinguished from the third by the width of the head capsule but repudiate his claim that the number of branches in the siphonal tuft can also be used. Although no data were recorded on the number of comb scales, (another character used by Barr to separate fourth instar from third) preliminary investigations by this investigator indicated that this character is variable, and therefore cannot be used as a criterion.

Based upon the examinations of reared and field-collected larvae, the evidence seems to indicate that there are three characters of value in

determining the larval instars. They are the width of the head capsule, the width of the siphon and the length of the siphon. Although any one of these characters may be used to distinguish the larval instars, it is the opinion of this investigator that all three characters should be used.

The experiments conducted on the effect of temperatures on the growth rate of the immature stages of Aedes atropalpus indicate that the speed of development of the immature stages is not slow as was the belief of earlier workers. The data indicate that the optimum temperature for development is in the vicinity of 90° F. At this temperature, 8.1 days were required for development from egg to adult.

Finally, the locality records on the distribution of this species indicate that it is more common than previously thought. Its presence seems to be commonly associated with rock outcroppings near and in streams or other bodies of water.

In Massachusetts, this investigator has observed that the larvae of A. atropalpus can be usually found from mid-April to the beginning of November.

LITERATURE CITED

Anonymous.
1951. Mosquito records from the Missouri River basin states. Survey Section, Communicable Disease Center, Publ. Hlth. Serv., Fed. Sec. Agcy., Kansas City, Mo. Mimeo. unnumbered pub. 1-93 p.

Aitken, T. H. G.
1942. Contributions toward a knowledge of the insect fauna of Lower California. 6. Diptera: Culicidae. Proc. Calif. Acad. Sci., 4th series 24:161-170.

Arnold, F. T., Jr.
1940. The mosquitoes of East Tennessee. Unpublished Masters Thesis. Univ. Tenn., Knoxville, 80 p.

Barber, M. A.
1939. Further observations (1938) on the Anopheles of New Mexico. Amer. Jour. Trop. Med. 19:345-356, 1 tbl.

Barnes, R. C., H. L. Felton and C. A. Wilson
1950. An annotated list of the mosquitoes of New York. Mosq. News 10(2):69-84.

Barr, R. A.
1958. The mosquitoes of Minnesota. Univ. Minn. Agric. Exper. Sta., Tech. Bull. 228, 154p, illust.

Bar-Zeev, M.
1958. The effect of temperature on the growth rate and survival of the immature stages of Aedes aegypti (L.). Bull. Ent. Res. 49:157-163, 2 figs., 3tbls.

Beadle, L. D.
1944. Report of malaria control in war areas.
Investigations in Kansas, 1944.
Unpub. Rpt. U.S. Pub. Hlth. Serv.

Bean, J. L.
1946. A preliminary list of the mosquitoes of Maine.
Canad. Ent. 78:25-28.

Belkin, J. N.
1958. Personal correspondence.

Blickle, R. L.
1952. Notes on the mosquitoes (Culicinae) of New Hampshire.
J. J. Mosq. Exterm. Assoc. Proc., 39: 198-202.

Bohart, R. M.
1958. Personal correspondence.

Breland, O. P.
1958. Personal correspondence.

Carpenter, S. J.
1941. The mosquitoes of Arkansas.
Ark. State Board of Health, Little Rock, Ark. 1-87 rev. ed., illust.

Carpenter, S. J., Middlekauff, W. W. and R. W. Chamberlain
1946. The mosquitoes of the Southern United States East of Oklahoma and Texas.
Amer. Mid. Nat. Monag. 3, Univ. Notre Dame Press:1-292, illust.

Carpenter, S. J.
1949. Notes on mosquito collections in Pennsylvania and Canada during 1948.
Mosq. News 9(4):172-173.

Carpenter, S. J.
1950. Notes on mosquitoes in North America: New distribution records for Eastern United States during 1946 and 1947. Mosq. News 10(2):64-65.

Carpenter, S. J. and W. J. LaCasse
1955. Mosquitoes of North America. Univ. of Calif. Press. 360 p., illust.

Coquillett, D. W.
1902. Three new species of Culex. Canad. Ent. 34(11):292-293.

Dickinson, W. E.
1944. The mosquitoes of Wisconsin. Milwaukee Pub. Mus. Pubb., 8(3):269-365, illust.

Dyar, H. G.
1890. The number of molts of Lepidopterous larvae. Psyche 5:420-422.

_____.
1902a. Illustrations of the larvae of North American Culicidae - II. Jour. N. Y. Ent. Soc. 10:194-201.

_____.
1902b. Notes on mosquitoes in New Hampshire. Proc. Ent. Soc. Wash. 5(1):140-148, 1 pl.

_____.
1903. Culex atropalpus Coquillett. Ent. News 14:180-182.

_____.
1904. Brief notes on mosquito larvae. Jour. N. Y. Ent. Soc. 12:172-174.

Dyar, H. G. and F. Knab
1906. The larvae of Culicidae classified as independent organisms.
Jour. N. Y. Ent. Soc. 14:169-230.

_____.
1908. Descriptions of some new mosquitoes from tropical America.
Proc. U.S. Nat. Mus. 35:(1632):53-70.

Dyar, H. G.
1921a. The mosquitoes of Canada.
Trans. Royal Canad. Institute 13, pt. 1, 71-120.

_____.
1921b. Three new mosquitoes from Costa Rica.
Insector Inscitiae Menstruus 9(1-3): 34-36.

_____.
1922. The mosquitoes of the United States.
Proceed. U.S. Nat. Mus. 62, Art. 1, (2447):1-119.

_____.
1938. The mosquitoes of the Americas.
Carnegie Inst. Wash. Pub. 387:1-616, illust.

Edwards, F. W.
1932. Genera Insectorum.
Diptera Fam. Culicidae fasc. 194, 258p.

Felton, H. L., R. C. Barnes and C. A. Wilson
1950. New distribution records for the mosquitoes of New England.
Mosq. News 10(2):84-91.

Getting, V. A. and H. F. Foemster
1940. Survey of mosquitoes of Massachusetts.
Mass. Dept. of Public Health and Work Projects Administration 1-138, illust.

Gibson, A.
1937. Mosquito suppression work in Canada in 1936.
Proc. N. J. Mosq. Extern. Assoc. 24: 96-108, 2 tbls.

Good, N. E.
1945. A list of the mosquitoes of the District of Columbia.
Proc. Ent. Soc. Wash. 47(6):168-179, 1 tbl.

Gurney, A. B.
1943. A mosquito survey of Camp Crowder, Missouri during 1942.
Jour. Econ. Ent. 36:927-935, 2 tbls.

Haufe, W. O.
1952. Observations on the biology of mosquitoes Diptera: Culicidae) at Goose Bay, Labrador.
Canad. Ent. 84:254-263, 3 figs., 1 tbl.

Hedeen, R. A.
1953. The biology of the mosquito Aedes atropalpus (Coq.).
Jour. Kansas Ent. Soc. 26:1-10, 2 tbls.

Howard, L. O., H. G. Dyar and F. Knab
1912-17. The mosquitoes of North and Central America and the West Indies.
Carnegie Inst. Wash. Pub. 159, Vol. 1-4, 1064 p., illus.

Huffaker, C. B.
1944. The temperature relations of the immature stages of the malarial mosquito, Anopheles quadrimaculatus Say, with a comparison of the developmental power of constant and variable temperature in insect metabolism.
Ann. Ent. Soc. Amer. 37:1-27, 2 figs., 4 tbls.

Johnson, C. W.
1925. Fauna of New England-15, list of the Diptera or two-winged flies. Boston Soc. of Nat. Hist. 7:1-326.

Knutson, H.
1943. The status of the mosquito of the Great Swamp in Rhode Island during 1942. Jour. Econ. Ent. 36:311-319, 2 figs.

Kumm, H. W., W. H. W. Komp and H. Ruiz
1940. Mosquitoes of Costa Rica. Amer. Jour. Trop. Med. 20:385-422.

Kumm, H. W. and H. Zuniga
1942. The mosquitoes of El Salvador. Amer. Jour. Trop. Med. 22:399-415, 2 figs., 4 tbls.

Laird, R. L.
1941. Observations on mosquito transmission of Plasmodium lophurae. Amer. Jour. Hyg. Sec. C., 34:163-167.

Lane, J.
1953. Neotropical Culicidae. Univ. Sao Paulo, Brasil Vol. 1-2:1-1098, illus.

Matheson, R.
1944. Handbook of the mosquitoes of North America. Comstock Publish. Co., Inc., 2nd ed., 314 p., Ithaca, N.Y.

_____.
1945. Guide to the insects of Connecticut. Part 6, The Diptera or true flies of Connecticut., Fasc. 2, Family Culicidae. Conn. State Geol. & Nat. Hist. Surv. Bull. 68:1-46, illus.

Owen, W. B.
1937. The mosquitoes of Minnesota with special reference to their biologies. Univ. Minn. Agric. Exper. Sta. Tech. Bull. 126, 75 p., illus.

Pearis, L. M.
1914. The relation of temperature to insect development. Jour. Econ. Ent. 7:174-179, 6 figs.

Rees, D. M. and L. T. Nielsen
1955. Additional mosquito records from Utah. (Diptera: Culicidae). Pan-Pacific Ent. 31:31-33.

Roseboom, L. E.
1942. The mosquitoes of Oklahoma. Okla. Agr. Exper. Sta. Tech. Bull. 16, 56 p.

Rueger, M. E. and S. Druce
1950. New mosquito distribution records for Texas. Mosq. News 10(2):60-63.

Sanderson, E. D.
1910. The relation of temperature to the growth of insects. Jour. Econ. Ent. 3:113-139, 21 figs.

Schmitt, J. B.
1942. Five species of mosquitoes new to New Jersey, found in the last five years. Mosq. News 2(4):26-29.

Schoof, H. F. and D. F. Ashton
1944. Notes and new distribution records of the mosquitoes of North Carolina. Jour. Elisha Mitchell Sci. Soc. 60(1): 1-10.

Shields, S. E.
1938. Tennessee Valley mosquito collections.
Jour. Econ. Ent. 31:426-430.

Stone, A.
1958. Personal correspondence.

Stough, B.D., M. A. King and D. E. Eyles
1949. Extension of known range of Aedes atropalpus (Coquillett) southeastward in Georgia.
Mosq. News 9(4):173.

Theobald, F. V.
1907. A monograph of the Culicidae of the World.
The British Museum, London, Vol. 1-5, 2459 p., illus.

Trembley, H. L.
1945. Laboratory rearing of Aedes atropalpus.
Jour. Econ. Ent. 38:408-409.

_____.
1946. Aedes atropalpus (Coq.) a new mosquito vector of Plasmodium gallinaceum Brumpt.
Jour. of Parasit. 32:499-501.

_____.
1947. Biological characteristics of laboratory reared Aedes atropalpus.

_____.
1955. Mosquito culture techniques and experimental procedures.
Amer. Mosq. Contr. Assoc. Bul. 3:1-73, 17 figs., 1 tbl.

Twinn, C. R.
1949. Mosquitoes and mosquito control in Canada.
Mosq. News 9(2):35-41, 2 tbls.

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APPROVED:

Frank R. Shaw

Jay R Fraver

Marion E. Smith

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