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NOTES ON THE OCCURRENCE AND HABITAT OF ANOPHELES PUNCTIPENNIS AND ANOPHELES MACULIPENNIS IN THE VALLEY OF THE ANDROSCOGGIN.

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The attempt to prevent malarial infection by combating the particular kind of mosquito that forms the intermediate host of the malarial parasite is not likely to meet with even moderate success unless fortified by a complete knowledge of the development and ecology of the various members of the incriminated genus. It is believed by many that one of the most hopeful measures for diminishing malaria lies in the discovery and reproduction of the natural conditions hostile to Anopheles. As pointed out by several writers, such conditions assuredly exist, or Anopheles would be far more generally distributed than is actually the case. What these conditions are can be determined only through systematic study of the local conditions in different regions by a number of different observers. The following notes of one summer's collections are recorded in the hope that they may be found useful in further comparative studies.

In many respects the history and distribution of malaria in New England offers a particularly inviting field for investigation. Malaria was not one of the diseases with which the early settlers had seriously to contend; according to Dr. Holmes,[1] the records of the first century of New England indicate that "indigenous intermittent fever can have pre-

[1] O. W. Holmes. Facts and Traditions respecting the Existence of Indigenous Intermittent Fever in New England. Boylston Prize Essay, 1836.

vailed but to a very limited extent, and the only place which we can clearly point to as giving origin to the disease is New Haven." Adams,[1] writing in 1880, states that "During the present century it has been, with very trifling exceptions, unknown as an indigenous disease. . . . But in 1850 this disease obtained a foothold in Connecticut, on the shore of Long Island Sound, and about 1864 began a northward march across the western half of Connecticut, and has steadily pushed forward, until, in 1876, it reached the northern border of the State, and thence, ignoring the boundary line, stepped over into Massachusetts, appearing in Sheffield in 1877. But, previous to this date, a few cases had appeared in three localities; viz., in Springfield since 1870, New Marlborough since 1874, and Holyoke since 1875. In 1878 the disease appeared at Agawam and New Lenox; and in 1879 and 1880 it made a rapid advance, invading a considerable number of towns."

Since 1880 malaria of the tertian type has prevailed quite extensively in many parts of New England and has invaded many districts previously exempt. Many of the surburban towns around Boston have suffered rather severely during the last decade. Prior to this time indigenous malaria, save a few scattered cases which attracted attention from their rarity,[2] had been practically unknown in Boston and its environs. The towns of Central and Western Massachusetts, especially those along the valley of the Connecticut and among the Berkshire Hills, appear to have harbored some

[1] J. F. Alleyne Adams. Intermittent Fever in Massachusetts. Second Annual Report of the Massachusetts State Board of Health, Lunacy, and Charity. Supplement, Public Health, 1880.

[2] An interesting Report on Intermittent Fever in Chelsea, by Drs. H. I. Bowditch, John Ware, and Ephraim Bush, in the Boston Medical and Surgical Journal, Vol. 47, p. 535, Jan. 26, 1853, records several cases of fever and ague originating near the edge of a "partially dried-up marsh." In this report the statement is made that data extracted from a diary kept by the Rev. Noadiah Russell in 1682–3, while a tutor at Harvard, indicate that at that date fever and ague were prevalent in the neighborhood; the diarist himself had an attack in 1682. Dr. Holmes (op. cit.) states that Urian Oakes, President of Harvard College from 1675 to 1681, suffered with a quartan ague perhaps contracted in this country. Five cases of intermittent fever originating in Boston between 1865 and 1868 are recorded by Treadwell (Boston Med. and Surg. Journal, 1868, Vol. 78, p. 227).

malaria in early times and to have suffered up to the last decade much more than the eastern portion of the State.

In Connecticut and Rhode Island it seems that malaria always prevailed more extensively than in Massachusetts. Dr. Holmes (op. cit.) refers to New Haven as the only locality in New England where the evidence of the existence of indigenous malaria in colonial times is incontrovertible. Adams (op. cit.) gives a list of thirty-five towns and villages in Connecticut in which malaria appeared between 1850 and 1878, and Chapin[1] gives a long list of Rhode Island towns in which malaria was known to occur in 1881; a "large number of cases" were reported as occurring in Providence in that year.

The three northern New England States appear to have been almost entirely exempt through their whole history. There is no good evidence that indigenous malaria has ever existed in Maine; one case occurring at Poland is mentioned by Dr. Holmes, but this is rejected by Adams on diagnostic grounds; one case occurring at Biddeford about 1760 was indigenous "according to tradition." Upon the map accompanying Dr. Holmes's paper, Burlington in Vermont, and Kensington in New Hampshire, are the sole localities in these States which are designated as places where intermittent fever is "supposed to have originated." Adams (op. cit.) says: "It is not known that intermittent fever has appeared farther north than Massachusetts." Chapin (op. cit.) reports a few cases occurring at Keene, New Hampshire, in 1883, and notes that some other cases have occurred in towns along the valley of the Connecticut. On the whole, however, it is clear that the disease has been far less prevalent in the northern than in the southern portion of New England.

Some interesting questions are raised by the historical record. It may be asked, for instance, whether the comparative exemption of the early settlers was due to the absence of Anopheles either wholly in some localities or in sufficient

[1] Chapin C. V. The Origin and Progress of the Malarial Fever now prevalent in New England. Fiske Fund Prize Dissertation, No. XXXII., 1884.

numbers in others, or whether it was due chiefly to the scarcity of individuals harboring the malarial parasite from whom the Anopheles might become infected. It may be asked also what was the cause of the malarial invasion of New England which began in the southern portion about the year 1850, and has progressively extended its range northward, until at the present time many localities, previously free throughout their history, such as the lower valley of the Charles River, are thoroughly impregnated with the germs of the disease? Why, again, have the more northern New England States escaped malaria almost wholly, although no more sparsely settled than other regions in which malaria has occurred? If, as seems probable, the full answers to the questions raised above are connected with the presence or absence of special topographical or seasonal conditions inimical to special kinds of mosquitoes, the importance of setting on foot coöperative investigations into the ecology of Anopheles cannot be mistaken.[1]

New England would seem to be an especially favorable field for such a study. Making all due allowance for the inadequacy of the historical record, it is nevertheless true that this portion of the United States contains a relatively large proportion of municipalities possessed of fairly precise information regarding their early medical history. A reëxamination of the available historical data might throw light on some of the obscurer points at issue. The recent extension of the disease, leading to the infection of communities which had hitherto enjoyed immunity, offers a definite and promising field for inquiry. The occurrence and especially the relative seasonal abundance of Anopheles in various localities is one of the more important points to be determined, and it would seem as if coöperative investigation along this line should be especially fruitful.

The locality in which my own observations have been made during the summer of 1901 is in the neighborhood of

[1] The employment of bands of Italian workmen for the construction of water-works and sewerage systems, whatever significance it may have for particular localities, can only partially explain the range and spread of the disease.

the town of Shelburne, N.H., in the valley of the Androscoggin River, and near the Maine border. The town is sparsely settled, and consists of scattered farm-houses strung along a strip of cultivable meadow-land barely thirteen hundred feet wide; the river bottom is here about seven to eight hundred feet above sea level. From the intervale, hills rise, sometimes quite abruptly, to a height of twenty-five to thirty-five hundred feet. The mean temperature in the summer is, as a rule, under 70° F. (cf. Hitchcock, "Geology of New Hampshire," p. 144. June, 62.9°; July 69.3°; August, 64.1°; September, 55.4°). The region is considered a healthy one, and so far as I can discover from the testimony of local physicians and others, no cases of malaria have ever originated in Shelburne or its vicinity.

My collections have extended over the period from June 21st to September 21st. Two related lines of work have been pursued: the collection of the winged insects, and the collection of the larvæ and study of the larval habitat.

RELATIVE ABUNDANCE OF ADULT ANOPHELES.

Very few winged insects belonging to the genus Anopheles were found during the entire season. I never observed adult Anopheles in the immediate neighborhood of their breeding-places, although I carefully scrutinized every mosquito that approached, and captured and examined hundreds of other mosquitoes in the course of the summer. The house occupied by the writer was the object of special search, since, as is well known, most species of Anopheles are semi-domestic, and tend to congregate in dwellings and their immediate neighborhood.

From the beginning of the season daily search was instituted for the mosquitoes that had found their way into the house, which, although quite carefully screened, was not mosquito-proof. The following table gives the results of the findings:

	A. punctipennis.	C. triseriatus.	C. stimulans.
June 27	0	2	0
June 28	0	1	0
June 29	0	2	0
July 2	0	4	0
July 4	0	2	0
July 5	0	2	0
July 11	0	1	0
July 13	1	0	0
Aug. 8	0	1	0
Aug. 12	1	0	0
Aug. 15	0	0	1
Aug. 17	0	1	0
Aug. 25	0	0	1

A similar but less systematic hunt was made for mosquitoes on the porch of the house and on the outside of the screens in the evening, with the following results:

	A. punctipennis.	C. triseriatus.	C. stimulans.	C. pungens.
June 21	1[1]	15	0	0
June 22	1[2]	14	0	0
June 24	0	2	0	0
June 25	0	5	1	0
June 26	0	4	1	0
June 28	0	1	0	0
July 1	0	0	1	0
July 14	0	1	1	0
Aug. 5	1[3]	0	0	0
Aug. 21	0	0	4	0
Aug. 25	1[2]	0	0	1
Aug. 27	0	0	1	0
Sept. 4	0	0	2	0
Sept. 5	0	0	1	0
Sept. 7	0	0	0	0

[1] Killed in act of biting, on porch of house, 10 A.M.
[2] Caught on outside of screen with other mosquitoes in evening.
[3] Killed in act of biting, 7.30 P.M.

In the course of the whole season, therefore, only six specimens of A. punctipennis were captured in and about the house as against seventy-four specimens of Culex. The actual disproportion between the two genera is somewhat greater than these figures indicate, since many mosquitoes which were killed by a blow of the hand were injured too seriously for identification of the species and were not included in the table. It was, however, possible in these cases to distinguish readily between the genera Culex and Anopheles, and all the individuals so examined proved to be Culex with the two exceptions (June 21, August 5) above noted. Some fifty-three individuals belonging to unidentified species of Culex should be added to the total number of this genus. In this locality, therefore, during the season of 1901, the proportion of Anopheles found in the house and in its immediate neighborhood was about 4.5 per cent. of the mosquito fauna (6 Anopheles punctipennis, 58 C. triseriatus, 14 C. stimulans, 2 C. pungens, 53 C. sp?).

If the mosquitoes that were encountered along the roadside or in the woods were to enter into consideration, the proportion of mosquitoes belonging to the genus Anopheles would be lower still, since not a single Anopheles imago was captured away from the neighborhood of the house, whereas hundreds of mosquitoes belonging to the other genus were caught with the cyanide bottle or otherwise killed.

There are no definite statements regarding the relative abundance of the two genera in other parts of New England. The impression obtained from the Notes published by Theobald Smith[1] is that the proportion of Anopheles was somewhat greater in the neighborhood of Boston in 1900 than I have found it in Shelburne in 1901, and a few scattering observations made in the Boston suburbs (Newton, Mass.) by the writer in the early summer of 1900 tend to strengthen this impression. There is also very little to be found in the general literature of the subject

[1] Theobald Smith. Notes on the Occurrence of *Anopheles punctipennis* and *A. quadrimaculatus* in the Boston Suburbs. Journal of the Boston Society of Medical Sciences, 1901, V., p. 321.

regarding the numbers of Anopheles present in various localities, even in the more carefully studied malarious districts. (Cf. Nuttall, Cobbett, and Strangeways-Pigg, Journal of Hygiene, 1901, I., p. 9.) Kerschbaumer[1] has, however, recently published some interesting data for the locality of San Pelagio in Istria. From May 26 to August 6, 1900, there were captured of the species Culex pipiens 2,787 females and 91 males; of C. maculatus, 2 females; and of Anopheles maculipennis (claviger) 572 females and 1 male. From August 8th to October 14th there were captured in horse-stables, carriage-houses, and hay-lofts 108 females and 42 males belonging to the species C. pipiens, and 1,667 females and 248 males belonging to the species A. maculipennis. In pig-pens there were found 2 females of C. pipiens and 646 females and 18 males of A. maculipennis. On the island of Hong Kong, out of thirteen localities tabulated by Thomson (Brit. Med. Journ., Sept. 14, 1901, p. 684), the proportion of Anopheles ranged from 0.2 per cent. to 56 per cent., the percentage of Anopheles captured in five localities being smaller than the percentage captured by me in Shelburne. MacGregor (Brit. Med. Journ., Sept. 14, 1901, p. 682) states that 70 per cent. of the mosquitoes that haunt the Government House in Lagos, West Africa, belong to the genus Anopheles.

THE LARVÆ OF ANOPHELES.

Methods of Collection. — I have employed with complete success the simple outfit recommended by Nuttall, Cobbett, and Strangeways-Pigg (op. cit., p. 12), consisting of a white enamelled dipper (the one used by the writer was 14 cm. in diameter and of 600 cc. capacity), a wide-mouthed pipette, and collecting bottles. The use of the white dipper made it possible to detect the larvæ very quickly and readily and to distinguish between Culex, Dixa, and Anopheles with the greatest ease.

[1] Fritz Kerschbaumer. Malaria, ihr Wesen, ihre Entstehung und ihre Verhütung. Vienna, Braumüller, 1901, pp. 89–95.

List of Localities in which Anopheles Larvæ were found.— In the following table the abbreviations p. and m. in the column headed "species" signify respectively the species A. punctipennis and A. maculipennis. When the letter is in brackets the identification is based on the examination of larvæ only. The height above sea-level is taken from the sheets of the U.S. Geological Survey.

ANOPHELES LOCALITIES.

PLACE.	Ht. above sea-level.	Dates when Anopheles larvæ were collected.	Species.	Notes.
Shelburne, N.H. (1)	700 ft.	June 25 " 26 " 27 " 28 " 29 July 2 " 4 " 7 " 10 " 16	p. m.	Ditch in intervale near Androscoggin River, 1–2 ft. deep. Sluggish current. No shade. Many tadpoles. About 300 yds. from house occupied by the writer and 70–100 ft. lower. Adults of A. punctipennis and A. maculipennis raised.
		July 18 " 22 " 24 Aug. 3 " 12 " 24 Sept. 1 " 7 " 18 " 19 " 21		C. stimulans found July 18, and later (Aug. 3) found small Dixa larvæ (also Aug. 22).
Shelburne (2)	700 ft.	June 30 July 1 " 25 Aug. 9	[p]	Ditch in intervale. No current. June 30 found 1 larva to about 3 dips. Made a second visit July 1, and caught only 2 larvæ in about 25 dips. No shade. Little life of any kind. Several predatory larvæ. Visited again July 25, and found nearly dry. 1 A. larva and a few small C. larvæ (stimulans). Completely dry Aug. 28.

ANOPHELES LOCALITIES. — *Continued.*

PLACE.	Ht. above sea-level.	Dates when Anopheles larvæ were collected.	Species.	Notes.
Shelburne (3)	800 ft.	July 8 " 9 " 13 " 22 " 24 Aug. 8	[p]	Roadside pool. 1 dip gave 2 A. larvæ, 2 C. larvæ (pungens), and 1 Dixa larva. Shaded densely during most of day. Few tadpoles. Spirogyra abundant. A. larvæ never abundant.
Shelburne (4)	900 ft.	July 8	sp?	Small pool in hollow of rock filled with rain water. About 5 in. deep and 6 in. wide, 3 ft. long. About 1 A. larva and 1 C. stimulans larva per dip. C. stimulans very abundant in other pools close by. July 22 visited again and found it completely dry. Found small Anopheles larvæ with Culex in neighboring pool in the rocks. Only C. in this pool August 1.
Shelburne (5)	725 ft.	July 26 Aug. 8	sp?	Margin of small pond about 100 yds. in diameter. Many Culex (pungens) — only 2 A. larvæ in about 40 dips. Tadpoles. Nuphar advena.
Shelburne (6)	740 ft.	July 29	sp?	Roadside pool, grass-grown, shaded. Little life. In 10 dips 1 A. larva, 3–4 C. sp(?), 15 Dixa. Evidently much extended by recent heavy rain.
Shelburne (7)	720 ft.	Aug. 12	[p]	Margin of small pond about 300 yds. in diameter. South bank of Androscoggin. Water very deeply colored with peat. Unshaded. C. pungens and C. stimulans abundant. 3 A. larvæ per dip.

ANOPHELES LOCALITIES. — *Concluded.*

PLACE.	Ht. above sea-level.	Dates when Anopheles larvæ were collected.	Species.	Notes.
Shelburne (8)	720 ft.	Aug. 12	sp?	Roadside pool near (7). Grass-grown. Unshaded. C. pungens and Dixa found also.
Gorham, N.H. (1)	800 ft.	July 3	p.	Stagnant pool near Androscoggin River. About 6–8 in. deep. Larvæ very abundant, especially among grass. 5–14 per dip. Pool partly shaded by large tree in afternoon. Few surface algæ (filamentous). Adult raised. Visited again July 25, and found completely dry. A little water on August 16, but no A. larvæ.
Gorham (2)	800 ft.	Aug. 16	sp?	Ditch in meadow near (1). Grass-grown. 2–3 larvæ per dip. C. pungens also.
Gilead, Me. (1)	720 ft.	July 29 Aug. 9	p?	Small pond in meadow. Grass-grown. Tadpoles. Nuphar. 1 A. larva per dip. About 20–30 (C. pungens) larvæ per dip. Some Lemna.
Gilead, Me. (2)	700 ft.	Aug. 9	p?	Meadow pool. Grass-grown. 1–4 A. larvæ per dip. No Culex.
Randolph, N. H. (1)	1,270 ft.	Aug. 14 Aug. 30	p.	Small ditch in meadow near Israel River. A. larvæ very abundant, 5 to 21 larvæ per dip. 10 pupæ captured. Spirogyra. Tadpoles. Much floating alga. 1 C. stimulans, a few Dixa. Adult raised.

The Relative Abundance of Larvæ. — The number of larvæ varied greatly in the different localities and in the same locality at different times. The smallest number of Anopheles larvæ found in any place where they were found at all was in Shelburne (6), where Anopheles larvæ were so scarce that on the average only one was found in about twenty dips; in this locality at the same time the larvæ of C. pungens were very abundant, averaging forty to one hundred to a dip. The other extreme was Randolph (1), where as many as twenty-one Anopheles larvæ were found per dip. The extreme of variation in one and the same collecting ground was observed in Shelburne (1), where on September 7th as many as twelve per dip were found, and on July 14th only one was brought up in about five dips. In this locality the vicissitudes of collection are indicated by the following field notes:

June 25. "Very abundant."
June 26. "About 3 per dip."
June 27. "About 6 to 7 per dip."
June 28. "Abundant."
June 29. "1 to 4 per dip."
July 2. "1 per dip."
July 4. "3 to 9 per dip."
July 7. "1 per dip."
July 10. "1 to 2 dips."
July 14. "Very scarce," 3 larvæ in 15 dips.
July 18. "Very scarce," 7 larvæ in 25 dips.
July 22. "Very scarce," 1 larva in 3 dips.
July 24. "Very scarce."
Aug. 3. "Very scarce." Only 27 larvæ in half-hour.
Aug. 12. "More abundant than on Aug. 3."
Aug. 24. "Much more abundant than for some weeks, 35 larvæ in 15 dips."
Sept. 1. "24 larvæ in 10 dips."
Sept. 7. "38 larvæ in 10 dips."
Sept. 18. "36 larvæ in 10 dips."
Sept. 21. "32 larvæ in 10 dips."

Numerical estimates of this sort are somewhat fallacious, owing to inevitable irregularities of distribution, caused by winds, growth of water-plants, etc., but probably have some value. It is important, however, that each observer at least should always follow substantially the same method of collection in his own work, since a slight divergence in the manner of "dipping" will lead to a difference in the number captured "per dip." The writer attempted to use in his work a uniform method, consisting in just immersing the lip of the dipper below the surface of the water and then swinging it rather rapidly through an arc of two to three feet, so that the surface quarter-inch of water would rush into the vessel.

The Length of Larval and Pupal Life.[1] — Although freshly reared female Anopheles were confined in breeding jars under what appeared to be favorable conditions and were fed with human blood, — and also with bananas and blueberries, — I did not succeed in getting them to lay eggs in captivity, and so did not secure data bearing directly upon the length of larval life. Very young larvæ — 1.5 mm. to 3 mm. — captured in the open did not for some undiscovered reason thrive very well under the conditions in which I kept them, and almost invariably died before reaching the pupal stage, in spite of frequent changing of the water and the addition of abundant supplies of water-weed. The older larvæ fared somewhat better, and in the course of the season twenty larvæ were transformed into pupæ and hatched. In no case, however, did the Anopheles larvæ prove as hardy as Culex larvæ under observation at the same time and under the same conditions. Anopheles larvæ were not as a rule kept alive for more than two to three weeks, whereas larvæ of C. stimulans and C. triseriatus could be kept alive almost indefinitely with little care, and in one case full-sized Culex larvæ (C. triseriatus) were kept in water containing little or no food for twenty-five days and pupated normally at the end of that time, normal adults developing from these pupæ.

In the open, more small larvæ than large ones are captured. At different times during the season, three hundred

[1] All statements made in this paper refer to A. punctipennis unless A. maculipennis is expressly mentioned.

and fifty-four larvæ were captured in eight localities in Shelburne. Measurements showed that two hundred and thirty-four of them were between 1 and 4 mm. and one hundred and twenty were between 4 and 8 mm. long. This falls in line with the observations of Nuttall and Shipley[1] in Cambridgeshire, England, so far as the excess of small larvæ is concerned, although in Shelburne the large larvæ were relatively more numerous than in England (34 per cent. of large larvæ in Shelburne, 22 per cent. in Cambridgeshire). Wide variations are noticed in the size of larvæ captured at different times in one and the same locality. This is illustrated in the notes of collection at locality Shelburne (1). (The actual numbers at the different dates have no significance, since the time spent collecting on the different days was not uniform.)

SIZE OF ANOPHELES LARVÆ.

Shelburne (1).

	1 to 4 mm.	4 to 8 mm.	
July 2	5	4	
" 4	14	10	
" 7	2	3	2 pupæ
" 10	2	7	
" 14	0	3	
" 18	5	4	
" 22	8	8	
" 24	0	3	
Aug. 3	23	4	
" 12	10	12	1 pupa
" 24	27	8	
Sept. 1	18	6	
" 7	29	9	1 pupa
" 18	29	7	1 pupa
" 21	21	11	
	193	99	

[1] Journal of Hygiene, 1901, I., p. 70.

The proportion of pupæ to larvæ was very low. In Shelburne (1), as the table shows, only five Anopheles pupæ were found in the entire season, as against two hundred and ninety-two larvæ collected in the same locality. The only other locality where pupæ were found was Randolph (1), where ten pupæ were collected on August 14. Larvæ, however, as well as pupæ, were more abundant in this latter locality than in any other place where collections were made.

The Anopheles pupa can be readily distinguished from that of Culex by the difference in position when resting at the surface of the water, as pointed out by Howard (Notes on the Mosquitoes of the United States. U.S. Dept. of Agriculture, Divis. of Entomology, Bulletin No. 25, p. 40). They are sometimes quite green[1] when young, but always darken on approaching maturity. The aquaria in which the larvæ were placed were examined at frequent intervals between 6 A.M. and 9 P.M., and in several instances the hour of pupation was determined with considerable precision. The pupæ were removed to tumblers of clear water immediately after their transformation from the larval state. In the majority of cases, although by no means invariably, both pupation and emergence from the pupa occurred during the night. In experiments made with pupæ kept in the ice-box at a low temperature (8–13° C.), the pupæ of A. maculipennis persisted two days in the pupal state after those of A. punctipennis had hatched. The exact duration of the pupal stage of A. punctipennis was determined in seventeen instances, the longest pupal life being nine days at 8 to 13° C. and the shortest being thirty-six to forty hours at a temperature of 23–32° C. Eight hatched in thirty-eight to forty-eight hours, five in forty-eight to seventy-two hours, one in about seventy-two hours, and three, kept at an artificially lowered

[1] Kerschbaumer (op. cit., p. 79) states that the spring brood of Anopheles pupæ observed by him at San Pelagio were almost all green, but that he never encountered a green Culex pupa. All the young pupæ of C. pungens that were found in Shelburne, and those of C. stimulans from some pools, were quite as green as any Anopheles pupæ.

temperature, in six to nine days; one imago died in emerging from the pupa after forty-eight hours. Out of fourteen pupæ captured in the open, four hatched within twenty-four hours, five within forty-eight hours, two within sixty hours, one in four days, and two died during transportation.

Only one pupa of A. maculipennis was captured; the imago emerged sixty hours later; in addition, two larvæ of this species were captured, and the pupal stage lasted seventy-two hours (temp. 20–25° C.) and eleven days (temp. 8–13° C.) respectively. The observations of Grassi cited by Nuttall and Shipley (op. cit.), of Howard (op. cit., p. 40), and of Nuttall and Shipley (op. cit., p. 275) indicate that the pupal stage of this species averages at least three to four days, and, according to Howard (op. cit., p. 41), usually lasts longer than this (a minimum of five days in June!). In my own experiments with pupæ kept in the ice-box at a low temperature (8–13° C.), the pupæ of A. maculipennis remained two days longer in the pupal state than those of A. punctipennis. It would seem to be true, therefore, that the pupal stage of A. punctipennis is perceptibly shorter than that of A. maculipennis under similar conditions,[1] although further observations where material is abundant are needed to establish this point.

Sex of Mosquitoes Hatched in Laboratory. — Thirty larvæ and pupæ of A. punctipennis collected at intervals through the season gave birth to eleven male and nineteen female adults. There was on the whole a preponderance of female adults among the mosquitoes hatched out in the laboratory, although the numbers dealt with are not large.

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[1] G. W. Herrick (Science, N.S., XIV., No. 348, Aug. 30, 1901, p. 330) states that the duration of the pupal stage of A. punctipennis, as observed by him in Mississippi, was two days (temperature of the water not given).

	Males.	Females.
A. Punctipennis	11	19
" *Maculipennis*	2	1
C. Stimulans	10	14
" *Triseriatus*	3	16
" *Impiger*	1	0
" *Pungens*	2	2
Aedes sp.	0	6
	29	58

Seasonal influence, if it exists, is not marked. It does not appear that any more males than females are hatched during the early part of the summer. The effect of temperature and other causes upon the proportion of the sexes should be made the subject of experimental work in localities where the material is sufficiently abundant, as the matter has obvious practical bearings.

Habitat, etc. — Anopheles larvæ were found in thirteen different localities — eight within the town limits of Shelburne, N.H., two in the adjoining town of Gorham, N.H., one in Randolph, N.H., and two in Gilead, Me. In most cases the collecting-grounds were visited several times during the course of the season. Nine out of the thirteen bodies of water remained permanent through the season, while four pools in which Anopheles was found breeding in the early summer dried up completely before the close of August.[1]

The season of 1901 was above the average in amount of rainfall in this region, and I think it probable that at least three more of the breeding-places would disappear in a dry year.

In eleven out of the thirteen localities Culex larvæ were found together with Anopheles larvæ; only a single collection was made in each of the other two. There were several

[1] One pool in which drying-up occurred became again filled with water later in the season, but no mosquito larvæ of any kind could be found in it.

localities, notably Shelburne (1) and (2), in which only Anopheles larvæ were found in the early part of the summer, no Culex larvæ appearing until later. In four places the Culex larvæ associated with Anopheles were those of C. stimulans, in five those of C. pungens, in one place both species were present, and in one case the species of Culex inhabiting the same pool with Anopheles was undetermined. It is to be noted as a fact not devoid of significance that wherever the larvæ of one genus were found very abundantly, those of the other genus were notably rare. Thus in the ditch where Anopheles larvæ were found in greatest numbers (Randolph (1)), only a single Culex larva was brought to light in fifty dips, whereas in Shelburne (5) only two Anopheles larvæ were captured in about fifty dips, the Culex larvæ (pungens) averaging at the same time thirty to forty per dip. In other cases, however, where neither genus was very plentiful, the two seemed to be on substantially the same numerical footing (cf. Shelburne (3) and (4)). It is doubtless true that the preponderance of one or the other genus in a particular breeding-pool is due not so much to instinctive selection of a spot for egg-laying by the female mosquito as to the presence or absence of conditions favorable for larval development. At all events, there can be no question that straggling Anopheles females sometimes lay eggs in bodies of water where the conditions are more suitable for the development of Culex larvæ than for that of their own young. Dixa larvæ were found in five of the Anopheles localities, in one instance (Shelburne (6)) in much larger numbers than either Anopheles or Culex. Anopheles larvæ thrive especially in unshaded waters, although three out of the thirteen localities in which I have collected were densely shaded. In the shaded pools, however, they were always found in comparatively inconsiderable numbers. Dixa is more abundant in shaded than in unshaded water.

The general character of those bodies of water in which Anopheles were found breeding most abundantly is a matter of especial interest. Examination of the table of localities will show that all the Anopheles pools possessed one character in common. They were accumulations of water in ditches or pools in the intervale of the Androscoggin or in meadows bordering smaller streams. Such ditches were in some cases spring-fed, but in most cases they simply intercepted the ground water on its way to the river. In this respect they are quite homologous to the so-called filter-basins excavated near the bank of a stream for the purpose of capturing the ground water flowing towards the river. Many municipal water-supplies in the eastern part of the United States are derived from such filter-basins, which often furnish water of quite exceptional purity. The ditches in the intervale of the Androscoggin resemble in all essential respects these filter-basins. It is well-known that the storage of such ground-waters for public water supplies involves peculiar difficulties, in that waters which, like these, are rich in nitrate, afford highly favorable conditions for the development of microscopic algæ, when exposed to sunlight and air, and if undesirable forms get a foothold in such a water disagreeable odors and tastes are frequently produced. The amazing multiplication of microscopic forms in ground-waters of this class is conclusive proof of the superior character of such water as a pabulum and breeding-place for algal life. It is precisely upon these microscopic organisms that the well-being of Anopheles depends. The Anopheles larvæ, as is well known, are surface feeders and derive their sustenance from the plankton. In this respect they differ from most Culex larvæ, which are able to browse over the bottom and get their food in large part from the bottom ooze as well as from the floating life or plankton. It is no accident, therefore, that malaria has been observed to prevail especially along river bottoms and sea coasts. These are the very localities, at the bases of water-sheds, that are most likely to afford an opportunity for the ground-water to come into contact with light and air, and whenever this happens, Anopheles larvæ are provided with suitable and abundant food. Accumulations of surface water, on the other hand, are less apt to furnish proper pabulum, at least in temperate regions, and are available as breeding-places only for the larvæ of those genera of mosquitoes that are able to range more freely in their search for food than Anopheles larvæ.

It need hardly be pointed out that not only the ditches and trenches in river intervales and meadows that fill with ground-water are favorable breeding-places for Anopheles, but also pools made by damming the flow from a spring or other ground-water source. This is unquestionably one reason why the excavations for railways, sewer-pipes, and water-pipes, which disturb the natural drainage channels and lead to the formation of quiet pools of spring (ground) water, are frequently followed by outbreaks of malaria. Pools formed of a sewage-polluted river water in which vitrification has occurred fall into the same category with other nitrate-rich water.

Influence of Temperature. — As has been already pointed out, pupation and emergence from the pupa, and presumably larval development likewise, take place more rapidly at a moderately high temperature than at a low. Pupal development, at least, is perfectly normal at a temperature of 26–32° C. At the same time, a marvellous degree of adaptability to low temperature is shown. One large larva of A. punctipennis pupated normally in the ice-box at 8° C., and a normal imago emerged six days later, the temperature of the water never ranging higher than 13° C. during this period and averaging about 10°. My last collection of Anopheles larvæ in the field was made on September 21, when the temperature of the air was 13.5° and that of the water 15°. At this time and for some days previously no Culex larvæ were found in this locality. In the whole region, in fact, there was a striking absence of Culex larvæ. In localities Shelburne (4) and (5), where early in the season there had been a great abundance of Culex larvæ (C. stimulans and C. pungens respectively), not a single Culex larva could be found on September 8, or again on September 19.

This falling off in the number of Culex larvæ at the approach of cold weather is in marked contrast to the persistence of numerous Anopheles larvæ in at least one of the localities under observation (p. 11).

Enemies. — Although no systematic studies were made, two aquatic forms were observed to prey upon Anopheles larvæ under natural conditions. On several occasions Anopheles larvæ were seen struggling in the grasp of these enemies immediately after the dip had been made and when the catch was first examined. These two enemies were the larval form of Dytiscus and a species — perhaps several — of Notonecta. Large Anopheles larvæ, also, would not infrequently devour their own kind in the aquaria, but were not observed to do this under more normal conditions.

A. Punctipennis. — Most of the statements in this paper refer only to the larvæ and pupæ of A. punctipennis. The larva of A. punctipennis can be most easily distinguished from the larva of A. maculipennis by the pigment markings on the dorsal aspect of the head. An excellent figure of A. maculipennis is given by Nuttall and Shipley (op. cit., Plate II., Fig. 4), but I have not been able to find anywhere a sufficiently detailed drawing of the A. punctipennis larva. The accompanying figure (Plate I., Fig. I.) may consequently be useful in enabling observers to separate the two species by larval characteristics.

A. Maculipennis. — I am unable to offer any explanation for the relative scarcity of A. maculipennis in this locality. Only three individuals of A. maculipennis were raised from larvæ and pupæ captured during the season (all from Shelburne (1)), as against thirty individuals of A. punctipennis from the same pools. No adults of this species were captured, as compared with six specimens of A. punctipennis. Curiously enough, A. punctipennis has not been recorded before from the State of New Hampshire, but the species A. maculipennis, which was so much less abundant in Shelburne and Gorham in 1901, has been recorded from the neighboring town of Berlin. (Howard, op. cit., p. 40.)

NOTE UPON OTHER SPECIES OF MOSQUITOES.

The mosquito fauna of Shelburne is quite large. Seven species in all have been collected — C. stimulans, C. impiger, C. triseriatus, C. pungens, A. punctipennis, A. maculipennis, Aedes sp.

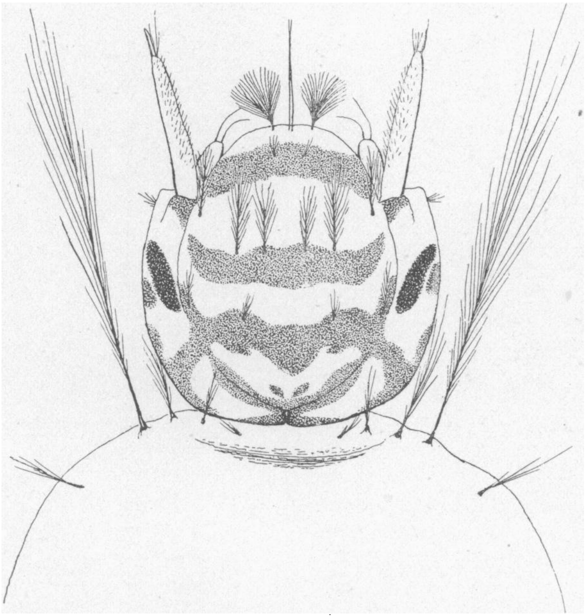
C. Stimulans. — The mosquito most conspicuous through its aggressiveness during the season of 1901 was C. stimulans. Especially in the woods, this species was always ready to attack in large numbers, from the beginning of the season to the end. Of the mosquitoes captured in the act of attacking man it was numerically predominant. This species was found breeding in large numbers in the pools in rock hollows along a mountain brook, and also, but less abundantly, in small pools and ditches in the intervale. The larvæ are usually large, and hang nearly or quite vertically from the surface. They can remain below the surface without rising for as long as eight minutes. The pupal stage lasts from three to four days at 20 to 25° C. Fourteen females and ten males were hatched from larvæ and pupæ collected between July 8th and August 1st.

C. Triseriatus. — This species was quite abundant about the house early in the summer, and after some search their breeding-place was discovered in a pool of water that had gathered in a hollow in an elm tree about three feet above the ground. The water was a rich leaf infusion and had a deep brown color. A male and a female adult were captured in this hollow, and a number of larvæ and pupæ were collected on July 10th. Sixteen females and three males were hatched. The average duration of the pupal stage was four to six days at 20 to 25° C. The house was supplied with mosquitoes mainly from this tree, and the application of a few drops of kerosene to the pool diminished the number of this species invading the house and prevented all development in the tree during the remainder of the summer.

C. Impiger. — The adults of this species were occasionally collected during the early part of the season, but were encountered less frequently during the latter part. Very large larvæ and pupæ were found in a pool in deep woods on a mountain side about 2,200 feet above sea-level. They did not bear transportation well and only one male was hatched. Many of the individual mosquitoes captured in the early summer were infected with a small red mite kindly examined for me by Dr. L. O. Howard, who writes me that it is the immature stage and has not been bred, so that the adult is not known with certainty. It is at present known by the name of *Acarus culicis*.

C. Pungens. — This species was not often captured in the neighborhood of the house, but was sometimes met with along the road and was found breeding in large numbers in several localities. It was found with Anopheles in locality Shelburne (6); four pupæ taken here gave rise to two males and two females. The breathing-tube in this species is proportionately much longer than the breathing-tube of the three other species of Culex observed in Shelburne. The head is larger in proportion to the body and the larva itself smaller than the other species. As Howard has stated, this species does not remain long under water, — another respect in which it differs from the other species which I had under observation.

Aedes sp. — This species appear to differ from Aedes fuscus, according to the published figures (Howard, Mosquitoes, McClure, Phillips & Co.). It was found breeding in pitcher-plant leaves growing around a small pond at the edge of the intervale and also in pitcher-plants on the margin of a small mountain lake 2,500 feet above sea-level. Not very numerous when first found (July 12th), but later in the season (July 27th–August 5th) nearly every leaf contained a pure culture of the larvæ. Three pupæ were found on July 27th. The larvæ are very translucent and very different in character from any other mosquito larvæ that I have seen figured. Both larvæ and pupæ are able to remain under water much longer than the young of other mosquitoes. The larvæ can apparently stay below the surface indefinitely; they certainly so remain for hours at a time. The specific gravity of the pupæ seems somewhat greater than that of other pupæ that I have observed. I have timed one pupa below the surface of the water for five minutes. Six females have been hatched. The larvæ are more hardy than those of any other species I have had under observation. They bear transportation perfectly and will remain alive for months (as long as one hundred and sixty-two days at 20° C.) without any other care than that of occasionally adding water to the tumbler to prevent drying up. The pupal stage lasts five to eight days at 20–25° C. The larvæ are negatively heliotropic to a marked degree.



JORDAN. ANOPHELES PUNCTIPENNIS.