It was prior to 1882 that Grant Allen, when recounting Dr. McCook's studies of honey ants in Colorado, placed entomologists in two classes—to quote his own words—"one kind, now, let us hope, rapidly verging to extinction, sticks a pin through his specimens, mounts them in a cabinet, gives them systematic names, and then considers he has performed the whole duty of man and a naturalist; the other kind, now, let us hope, growing more usual every day, goes afield to watch the very life of the creatures themselves at home, and tries to learn their habits and customs in their own native haunts."

The Army School of Hygiene, at Aldershot, is favourably situated from this point of view, being sited in a mosquito belt which extends from Ash to Fleet, and in this area it is possible to collect the majority of species. For breeding room and demonstration purposes in winter, the hibernating Theobaldia annulata and Anopheles maculipennis are the mainstays. In this district they are found in stables, rarely in cowsheds, and practically never in association with man. In Sandwich district collections are chiefly drawn from cow-sheds and pig-styes. Both are frequently found in the same stable, T. annulata in some dark corner and A. maculipennis resting on cobwebs. Here surrounded by the warm exhalations of the beasts they remain throughout the winter.

Occasional use is also made of Culex pipiens which is collected in damp situations, dug-outs, boiler houses, cellars and the like. It seems to conform more truly to our ideas of hibernation, seeing that it remains practically without movement for days together, does not search for blood, and in all probability subsists upon the store of nourishment contained in an enlarged fat body.
These three adults can be induced to oviposit in the warm breeding room under the stimulus of banana and blood feeds. They all readily avail themselves of the presence of the rabbit, *T. annulata* and *A. maculipennis* literally gorging themselves, but *C. pipiens* is a more temperate feeder. The former are usually immobilized after a feed, but the latter is particularly active and may only show a slight staining of the abdomen. The eggs hatch out and the larvae are raised under laboratory conditions. These arrangements ensure a supply of adult specimens, anopheline eggs, egg rafts, and large numbers of developing larvae for demonstration purposes.

In regard to the habits of adult *A. maculipennis* and *T. annulata*. In the summer both show marked zoophylic tendencies. The improved brightness and ventilation of present-day barracks may also contribute to this end, but they are often to be found in a particular group of stables where the whitewash is spotless and the ventilation above reproach. They regularly invade barrack rooms and houses, but in the case of grooms' quarters over stables, even if the ventilation of these is not ideal, the majority will be found in the stables below and very few in the quarters overhead.

Grooms living alongside their horses have also frequently stated that as a rule they are only bitten when grooming the horses in the stables.

Certain residents in the district have also remarked that the mosquito nuisance is greater now than in pre-war times. Undoubtedly the psychological effect of articles in the daily press may tend to foster this opinion, but although the War Department land is not being brought under cultivation or used extensively for building purposes, it must not be assumed that details affecting the amelioration of drainage are being neglected. During the last two years considerable progress has been made in this respect and a marked reduction in breeding places has automatically taken place. In this connexion the present-day decrease of the equine garrison should not be overlooked.

The bite of *T. annulata* usually gives rise to severe reaction, occasionally septic infection accompanied by marked ecchymosis. The bite of *A. maculipennis* is of the same type but less severe in character. During the winter both insects feed readily during the day, *T. annulata* being the more constant day feeder. At 4 p.m. during a spell of bright sunshine in March, 1925, one of our men at work near a spinney was surrounded by hundreds of freely biting *T. annulata*.

There is no doubt that certain people are more attractive to mosquitoes than others, and a count of the bag after forays in the woods has revealed that three times as many mosquitoes may be caught on one person as on another. This quality is more usually possessed by the fair, and one particular serjeant was frequently referred to as "the bait."

The home service khaki with its greenish shade is favoured by sylvan species, but whereas they appear to distrust the smoother cloth of the
officer, they readily settle on the rough and stubby surface of the man's serge. This predilection becomes more noticeable in the breeding room, where mosquitoes generally ignore the shaven back of the rabbit and settle on its nose with alacrity.

On more than one occasion with *A. maculipennis* on the wing and despite the presence of several people, a dog has been singled out, and again the point of attack was the muzzle. This type of surface may be warmer and associated with a superior blood-supply, and as the mosquito usually makes a careful selection of the site of bite and has a particular affinity for babes, in like manner the texture of the skin of a fair person may present greater attractions.

An actively sweating man is not particularly attractive, but during the period of cooling off after exercise a heated man is more likely to suffer from the attentions of these insects than a cool one. This is in keeping with Howlett's observations on the effect of heated surfaces, in his case test-tubes. Another somewhat curious fact has been noticed, that, in the absence of blood, mosquitoes will occasionally gorge themselves with milk, even if water is available.

Although *A. maculipennis* seems to pass the winter solely in the adult stage, with nearly every winter collection of larvae *T. annulata* appears.

As regards breeding places in this district, both *A. maculipennis* and *T. annulata* are to be found in foul water and frequently together. *T. annulata* selects the worst type and has been recovered from drains taking washings from stables and middens. One of the most constant breeding places of *A. maculipennis* is in a roadside ditch at Ash, the end of which has been used as a rubbish tip for years. After the winter rains this drain under a hawthorn hedge is converted into a fairly large expanse of water. In the early summer, amongst the coarse grass along the edge *A. maculipennis* larvae are found in enormous numbers, and towards the centre, where the water is more impure, *T. annulata* can be seen.

Though normally the Basingstoke canal proper can be explored for days with negative results, *A. maculipennis* is to be found in its weedy and boggy offshoots. With the advance of summer these, the normal breeding places, dry up and the contained water becomes extremely foul. The larvae of *T. annulata* persists in these situations until the end, but *A. maculipennis* now takes to breeding in the relatively clear water of the canal. From this it will be seen that the above species frequently do select water which is more impure than some sewage effluents. Details regarding the pH of these will be discussed later.

There is some evidence that the larva of *T. annulata* pupates and emerges during the winter, and on rare occasions a male adult which has a short span of life has been collected.

During the late autumn use is made of *C. pipiens* obtained in the vicinity of Camp Farm sewage works, but as a general rule collections of *T. annulata* and *Theobaldia morsitans* are made and occasionally *Aedes*
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punctor is available. *T. morsitans* is normally found near Mytchett flash in old trenches filled with leaves and also in the overflow from the lake. The water in these places approximates to the acid type favoured by the majority of sylvan mosquitoes, but at the same time *T. morsitans* does not avoid impure water. An unusual association with *Anopheles bifurcatus* in the alkaline foul water of an overgrown roadside channel receiving washings from a country road covered with cow droppings is noteworthy, and it has also been collected in company with *T. annulata* and *A. bifurcatus* in a slightly acid water resulting from washouts from certain dykes.

**Anopheline Larvae (Winter).**

The only species available is *A. bifurcatus* which in Palestine selects the cool and clean water of wells. In Aldershot it has been regularly collected in large numbers in old sewage runnels at Camp Farm sewage works and in stagnant and evil-smelling water of roadside dykes in Ash district. It also resorts to breeding places frequented by *A. maculipennis* during the summer, though the impurity of the water in these is materially reduced by the winter rains. In none of these situations can it be said to be breeding in relatively clean water or in woodland country. The majority of the larvae are in the 3rd instar, although collections made in February and December, 1925, included at least twenty-five per cent. of 2nd instar larvae.

During the winter, 1925-26, a particular marshy area in Ash yielded large numbers of this species in company with *T. morsitans*, and the water possessed a definitely acid reaction.

Owing to the preponderance of *A. maculipennis* larvae the summer breeding places of *A. bifurcatus* have not been identified, but as the majority of the winter breeding places are dry until autumn, alternative breeding places must be common.

In the late spring the adult mosquito is frequently found in houses in Ash district. It apparently enters these for shelter, and complaints as regards biting are unusual. During the summer it is rarely found indoors, and stables which abound with *A. maculipennis* occasionally yield an odd one. It will readily attack man in the breeding room, and has been caught in the woods in early summer in the act of biting. The same has been noted with *Anopheles plumbeus*, which is very infrequently met in this district. The bite of *A. bifurcatus* gives rise to little irritation and reaction. In early spring it is quite a common occurrence for the male to accompany the female indoors, whereas in the case of *A. maculipennis* this condition only obtains with the very early broods, and later in the year only here and there a male is encountered.

**Culicine Larvae (Spring).**

Collections can be made within 200 yards of the school under the trees and low bushes shading the weedy offshoots and numerous depressions along the banks of the Basingstoke canal, and during the last three winters...
these places have been subjected to a close scrutiny. In these situations the water becomes acid in type in the presence of decomposing leaves, and *T. morsitans* is commonly located, followed as a rule by broods of *A. punctor*.

In the winters of 1923-24 and 1924-25 *A. punctor* larvae were not found, although they subsequently bred out in the spring; but in 1925-26 both *A. punctor* and *T. morsitans* were found in association throughout the winter. Up to date one brood of *A. punctor* has only been noted in the course of the year, and this is in agreement with the observations made by Dr. H. G. Dyer in North America. It can hardly be pleaded that this condition is due to drought, particularly in the case of the summer 1925, and also in order to induce second broods, some of the breeding places were kept filled with water from the canal during such periods, and still no second brood resulted. In this connexion no information is available regarding the time that *A. punctor* ova take to reach maturity, and as the canal water is alkaline in the summer, it may lack a very necessary stimulus which an acid water imparts. On one occasion, when a second brood was suspected, it was finally decided that it was a delayed first-brood, and that the breeding place was one which had not been filled with water since the previous spring owing to improved drainage of the area.

At Puckridge Hill a record of collections has been kept, and from one small sylvan pool during the winter and early spring the following succession of larvae has been obtained: *T. morsitans*, *A. punctor*, *Aedes annulipes*, *Aedes cinereus*, *Aedes geniculatus*, and *Aedes rusticus*.

I am informed by F. W. Edwards, Esq., that he has never encountered *A. annulipes* in such a situation, and that it usually breeds among rushes.

It is of interest to record that *A. geniculatus*, normally a tree-hole breeder often found in company with *A. plumbeus*, has in the course of the last three seasons been found in association with *A. cinereus*, a river-haunting mosquito, and also with several other species of sylvan larvæ in a pool of water in the ground; while, on the banks of the same pool, adult *Aedes maculatus*, *A. punctor*, *A. annulipes*, *A. rusticus*, and *A. geniculatus* have been caught in fair numbers.

In the spring of 1925 a collection of larvæ made from one of the offshoots of the canal occluded by yellow iris proved to be *A. punctor* and *A. cinereus*, and the water possessed a definitely alkaline reaction.

The warm evenings of early summer in Aldershot and district are frequently rendered unbearable by the attack of adults of the *Aedes* series, and although the bites do not result in severe lesions, they have a peculiarly irritating character. *A. cinereus* does not appear to be strongly attracted to man, and is collected with difficulty, its bright chestnut colouring making it conspicuous when on the wing. The other members of the series bite freely throughout the day under the shade of the trees, and are especially active between the hours of 6 and 8 p.m.

It has been stated that, with the exception of *Aedes caspius*, these
mosquitoes do not enter houses, but *A. punctor* has frequently been observed biting indoors, and on one occasion *A. annulipes* was collected in the act of biting a member of my household standing directly under a twenty candle-power electric light.

It has been generally observed that *A. maculipennis*, *T. annulata*, and even on occasions *A. bifurcatus*, make more or less open attacks during the evening indoors, gorge themselves with blood, and are discovered next day on the walls and ceilings. On the other hand, the *Aëdes* series are more difficult to detect, and usually make their attacks under cover of darkness or beneath chairs and tables. They then leave the rooms and make for their haunts in the woods, and this accounts for the difficulty in procuring naturally fed specimens.

It is frequently stated that adult *C. pipiens* may give rise to complaints. The annexes of the majority of houses and barracks in this district harbour these insects in considerable numbers during the winter, and even in the spring prior to sallying forth they are generally regarded as a non-biting species. If numbers of *C. pipiens* are examined it is a rarity to find a specimen showing evidence of recent blood feeds, and in common with sylvan species of mosquito when fed it appears to spend its time outdoors.

On rare occasions this species has been caught in the act of biting, but many of the mosquitoes captured during the evening indoors, which at first sight were thought to be *C. pipiens*, have proved on further examination to be *A. punctor*. The hibernating *C. pipiens*, when brought into the breeding room, will take frequent small feeds from the rabbit and is easily induced to oviposit, and therefore for class purposes is useful.

In regard to breeding places, most collections of water serve and particularly water-butts and the like which contain the decomposing debris from gutters, etc. These waters are alkaline in character, and up to the present larvae of *C. pipiens* outside the laboratory have never been taken from an acid type of water. In the laboratory *C. pipiens*, in the absence of other facilities, will oviposit in acid types of water containing natural silt, and although the eggs hatch the larva fail to grow and gradually die off.

*Feeding Habits.*—The question arises as to whether in common with other insects and animals a relationship may not exist between the siting of breeding places and the feeding grounds. The most constant breeding pools of *A. maculipennis* are within easy flight of stables, but in times of drought the canal and other unusual situations are utilized. Our most dependable breeding places of *A. bifurcatus* are within easy range of cow-sheds or fields with cows in pasture.

The *Aëdes* series, as mentioned previously, favour sylvan pools, but here again the most favoured spots are in the immediate vicinity of bathing places, i.e., Mytchett and No. 2 bathing place, and bathing and boating parties suffer severely from their attentions. At the same time it is impossible to rule out of court the natural denizens, birds, rabbits, stoats and foxes, etc., with which these woods abound.
C. H. H. Harold

Precipitin Reaction of Mosquitoes.

It was thought that the results of these might afford useful supplementary information regarding the propensities of the various species, and antisera were prepared as follows:

1. Horse v. rabbit.
2. Man v. rabbit.
3. Rabbit v. fowl.
4. Pigeon v. rabbit.

1. In this case the rabbit received six intraperitoneal injections of from five to ten cubic centimetres of horse-serum, and the rabbit bled between the eighteenth and twenty-first day.
2. As in (1), using human-serum.
3. Whole citrated blood of the rabbit administered intravenously in accordance with Sutherland's methods.
4. As in (1), using whole citrated blood of the pigeon.

Sutherland's technique was also adopted for the performance of the reactions.

In this respect the field of examination is unfortunately incomplete, because in spite of prolonged searches naturally fed sylvan culicine mosquitoes were not encountered. It would of course have been an easy matter to collect these off one of my own staff sitting out in the woods or to have fed them artificially off man or rabbit in the breeding room. This also applies to A. bifurcatus, but at the same time both A. bifurcatus and A. plumbeus have been collected attempting to bite during the day under the shade of the trees.

In the case of certain specimens a little difficulty was experienced in reading the results, owing to the fact that the mosquito extract was opalescent. This condition was readily amenable to centrifugalization or sedimentation after trituration.

The only naturally fed mosquitoes available were A. maculipennis, T. annulata and C. pipiens, in other words, the domestic species, but although the readings of the reactions give useful and interesting indications, the number carried is far too small upon which to base any final opinion.

C. pipiens.—Over twenty specimens collected around barracks showing no signs of blood feeds were examined, and they gave entirely negative results. Those showing signs of definite blood feeds gave the following reactions: Human, strongly positive, one. Rabbit, strongly positive, one—positive, one; trace, one. Six specimens did not react with any of the type sera.

In the light of laboratory experiences the rabbit reactions are of interest. The number of negatives indicates that the variety of type sera used should be increased.

T. annulata.—Some were collected in houses and others in stables, and
only those with definite signs of blood feeds were submitted to examination. Human, strongly positive, one; positive, two; trace, one. Horse, strongly positive, two; positive, three. Rabbit, positive, one. Pigeon, strongly positive, two—one of these also gave a trace of reaction with human antiserum.

This species in captivity attacks the rabbit with avidity.

A. maculipennis.—Collected in houses and also in stables; well-fed specimens were selected at random.

(1) Mr. Shute’s collection from stables:

Human, positive, three. Horse, positive, two. Rabbit, positive, one.

A number of specimens known to have fed on man gave definite human reactions and are not included.

(2) Aldershot collection:

Human, positive, ten; trace, two. Horse, positive, seven; trace, two. Rabbit, positive, two; trace, three. Pigeon, positive, one. Human positives gave trace reactions with horse, two; rabbit, one. Horse positives gave trace, two human; and trace, two rabbit—one specimen gave positive horse, and trace human and rabbit.

These results indicate that these mosquitoes are somewhat catholic in their tastes. The majority were collected in a district which abounds in wild life, although there also exists a large preponderance of human beings over domestic animals. In the case of double reactions it is possible that one or other of these may be non-specific, but the small amount of fluid available did not permit of the putting up of additional tubes, as is suggested by Sutherland.

**The Hydrogen-ion Concentration of Waters.**

For some time past evidence has been accruing under the artificial conditions imposed by the breeding room pointing to the fact that this is a matter of minor importance to the developing larva and that the prime factor is food. This view was expressed during a visit paid by Dr. A. Balfour to the school three winters ago, and as a direct outcome systematic outdoor observations have been carried out during the last two years. A series of experiments in this connexion were also carried out in the laboratory in March, 1925.

Captive A. maculipennis can be readily induced to oviposit in water with a low pH and healthy young larvae result. If to the water crushed *Myosotis palustris* is added in suitable quantities, a mild septic action is set up leading to an opalescent coloration of the water, the larvae grow to perfection, the weed is slowly lysed and an increase in the pH of the water occurs.

If no further attention is paid to the tank the contained water becomes blackish, develops a higher pH value and nitrites appear. This condition is not wholly suited to the development of the larva, but if further additions
of weed are made and the mild septic action maintained the tank can be successfully employed for some weeks. Eventually a point is reached where the water becomes black, very offensive, the larvae do not thrive in it and it becomes necessary to discontinue using it. A new tank is best started by seeding into the fresh water a little of the black water from a discarded one. The addition of an excess of weed at any time induces a marked septic action accompanied by a heavy scum formation which brings about the death of the larvae.

It should be noted that the above adverse conditions appear comparable with the state of affairs which arise in the natural anopheline breeding places during times of heat and drought, and it is possible that in tropical and sub-tropical regions there may be a connexion between the rapid development of septic action and the selection of cleaner types of water.

Having regard to the biological action occurring in waters and sewages, apart from the presence of protozoal and algal life, the breaking down of colloid, production of ammonia and nitrification are also dependant upon the contained bacterial flora, and superficially the conditions conducive to optimum growth of the anopheline larva in the laboratory and the changes occurring in certain natural waters appear to be similar.

Rudolphs, who is a strong advocate of the importance of food, lays great stress upon the presence of protozoal life and mentions the presence of aquatic fauna such as cyclops in some of the water samples examined by him.

It must be admitted that the presence of daphnia and cyclops is indicative of the availability of food, but in many instances it has been observed that waters containing cyclops are not conducive to optimum growth.

During the last two years mosquito larvae have been taken from water with the following ranges of pH:

<table>
<thead>
<tr>
<th>Species</th>
<th>pH Range</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>An. annulipes</td>
<td>4.7 to 4.9</td>
<td></td>
</tr>
<tr>
<td>An. maculatus</td>
<td></td>
<td></td>
</tr>
<tr>
<td>An. geniculata</td>
<td>4.4, 6.6 to 7.5</td>
<td>Almost invariably the lower ranges</td>
</tr>
<tr>
<td>An. punctor</td>
<td>4.4, 6.0 to 7.4</td>
<td>Normally the lower ranges</td>
</tr>
<tr>
<td>An. cinereus</td>
<td>6.0, 7.2 to 7.8</td>
<td>Normally the higher ranges</td>
</tr>
<tr>
<td>T. morsitans</td>
<td>6.8 to 7.8</td>
<td>Has never been recovered from typical sylvan acid water</td>
</tr>
<tr>
<td>An. maculipennis</td>
<td>6.0, 6.8 to 8.0</td>
<td></td>
</tr>
<tr>
<td>T. annulatus</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Gases in Solution.—In this district typical An. bifurcatus and An. maculipennis waters give a dissolved oxygen figure of 0.517 to 0.876 parts per 100,000, and during times of drought this is still further reduced. Sylvan waters (Aedes and Theobaldia morio) have a dissolved oxygen figure of 0.814 to 0.870 and a CO₂ figure of 3.9 to 6.0 parts per 100,000.

The above anopheline waters have a CO₂ content of from 0.25 to 1.1 parts per 100,000, and up to the present time larvae have not been recovered from waters containing CO₂ in excess of 1.8 parts per 100,000.

If normal An. bifurcatus and Aedes waters are supercharged with CO₂
in a Sparklet bottle and the larve returned to their respective waters, both become moribund, but the recovery of *A. punctor* is much more rapid than *A. bifurcatus*. *A. punctor* is normal and markedly active on the following day, but *A. bifurcatus* is still lethargic and does not readily respond to direct stimulation.

The Feeding Larve.—*A. bifurcatus* in Aldershot is frequently found in a brownish or greenish-tinged water with a slightly offensive odour containing much matter in suspension. In such water the larva spends the major portion of the day feeding at ease on the surface. Not so the *Aedes* series which select the clear water of pools lined with rotting leaves and twigs, from which a light red deposit separates out on standing. Under these conditions they spend long periods cruising about below the surface rubbing their mouth-parts along the rootlets and submerged plants and weeds.

In common with certain other species, the *A. punctor* larva with its long retractile gills in a water containing little colloid, etc., in suspension, is anatomically adapted for long periods of submersion. In the case of the anopheline larva the gills are papilliform, more or less rudimentary, and the water selected by them often contains low percentages of dissolved oxygen, e.g., some of the samples are virtually sewage effluent. Hence, it would appear that the anopheline larva may be largely dependant upon food in suspension.

The following simple experiments were performed in order to discover if any evidence could be adduced in support of these views:—

**Experiment I.**

Samples of water were taken from a typical *Aedes* breeding place in an unfrequented part of the wood, and also from one of our usual *A. bifurcatus* breeding places. The *Aedes* sample had a pH of 4.9, and the *A. bifurcatus* a pH of 7.5. On plating on gelatine it was seen that the *A. bifurcatus* water yielded at least five times as many colonies as the *Aedes* water, and that the majority of organisms in the anopheline water were liquefactors of gelatine and gave rise to putrefactive odours.

The appearance of the litmus lactose agar plates was still more striking; with 0.5 and 0.1 of a cubic centimetre of *A. bifurcatus* water the plate was covered with an innumerable mass of colonies, many of these being lactose fermenters, whereas plates from the *Aedes* water showed two bacterial colonies only and a single mould which rapidly obscured the plate.

Further platings with the acid type of water:—

1 c.c. 0.5 1 red colony (Gram + coccus) 3 mould colonies

After stirring up the bottom of the pool:—

1 c.c. 0.5 Extensive growth of Gram-negative bacillus

No growth
It has also been noted, in this Aëdes favoured district, that many of the typical acid type waters do not contain beech leaves, and that moulds and fungi are a frequent source of trouble when carrying out platings and experiments in connexion with water purification. The water supply of the school, a local surface water, has a pH frequently lower than 5·6, and a ferruginous growth and deposit in the pipes has recently necessitated the relaying of new mains. The fact that a reddish deposit separates out from our normal Aëdes water has already been commented on, and the association of moulds and fungi in nature with other organic acids such as oxalic, succinic and citric, is of interest.

**EXPERIMENT II.**

In continuation of the above collections of A. puncto* and A. bifurcatus, larvæ were made and carefully removed from their native medium into beakers by means of a ball pipette. The residual water was drained off and the beakers filled up with ordinary tap water. From the natural breeding places the following samples of water were obtained after well stirring up the silt.

A normal A. puncto* water with a pH of 4·9, an abnormal one with a pH of 7·2, and an A. bifurcatus water with a pH of 7·3. Representative groups of larvæ were then placed in dissimilar waters, and also in their natural waters as a control. The vessels were placed in the warm breeding room, and a good layer of silt and debris separated out on standing. The experiment commenced on May 6, 1925, and on May 22, 1925, the condition was as follows:

1. In the Aëdes control water pH 4·9, fifteen dead Aëdes larvæ, others emerged.
2. Aëdes in anopheline water, no deaths, all emerged.
3. Anophelines in abnormal Aëdes water pH 7·2, no deaths, one pupa, six larvæ of poor size, others emerged.
4. Anopheline control, six healthy larvæ remained, three deaths, others emerged.
5. Anophelines in normal Aëdes water pH 4·9, two larvæ remaining, seven deaths, poor specimens emerged.

On June 2, 1925:

3. Three larvæ remaining, others emerged—all emerged eventually.
4. One larva remaining, others emerged—eventually all emerged but development was delayed.
5. All emerged.

The A. puncto* larvæ were in various instars, and the majority of the A. bifurcatus larvæ were in the 3rd instar. The effect of the anopheline water upon A. puncto* was striking; within forty-eight hours sixteen had pupated and the others had attained a good size and were very active. They spent less time below the surface of the water and in every way
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outstripped their brethren in their native medium. The emerging adults were also larger than those from the control water.

As regards *A. bifurcatus*, they did not fare so well; it would appear that they mature more slowly than *A. punctor*, and although no deaths occurred in the abnormal *Aedes* water with a pH of 7.2 they did better in their own type of water. In the normal *Aedes* water pH 4.9, they spent longer periods amidst the debris at the bottom of the vessels and the emerging adults were weedy and similar in appearance to those raised in waters with a deficient food content. If anything the effect of the acid type of water seemed to cause the larvae to develop a little more rapidly than in the control.

On other occasions abnormal conditions have exercised effects which at first sight appear anomalous and do not readily admit of explanation, e.g., larvae transferred to unsuitable waters may either remain in the same stage for very long periods and exhibit a low mortality, or the cycle of development may be appreciably shortened, a heavy mortality may ensue and very poor specimens emerge. The exact line which is followed seems to depend largely upon the instar of the larvae. If they are transferred in the 1st or early 2nd instar the first course is the more usual, but if in more advanced stages the 2nd is more commonly observed. Mosquito larvae bred in stale water in which septic action has subsided have a cycle of development which is extremely prolonged. They are very tenacious of life and *A. bifurcatus* larvae under such conditions, when exposed to higher temperature in the breeding room, have been known to remain in the larval stage for weeks and eventually inferior specimens have bred out. In many instances the transference of well-developed larvae to a different type of water leads to an acceleration in development culminating in rapid pupation, and this response may be regarded as a defensive measure. If the water does not abound in suitable nourishment the curtailment in development is reflected in the inferior physique of the emergent mosquitoes.

On conclusion of the experiment the pH values of the waters were:

Normal *Aedes* water, original pH 4.9 (control); pH 4.7. Note increased acidity.

Normal *Aedes* water pH 4.9, containing *A. bifurcatus*; pH 4.7. Note increased acidity.

Abnormal *Aedes* water pH 7.2, containing *A. bifurcatus*; pH 7.8. Note increased alkalinity.

Normal *A. bifurcatus* water pH 7.3 (control); pH 7.8. Note increased alkalinity.

*A. bifurcatus* water containing *A. punctor*, pH 7.3; pH 7.8. Note alkalinity.

Note.—Typical acid type water becomes increasingly acid and typical anopheline water more alkaline.

The above seems to afford support to the view that as far as the growth of larvae is concerned the important factor is food.
EXPERIMENT III.

As an elaboration of the foregoing, *A. bifurcatus* larvae collected from the usual sources were transferred to tap water with a pH 5·6. In this they remained in a sluggish condition without undergoing further development for fourteen days.

Mixed agar cultures of organisms isolated from *A. bifurcatus* water were emulsified and added to tap water which became opalescent and caused its pH to be raised to 7·6. The pH of the water was now readjusted with acetic acid to its original pH of 5·6 and the larvae transferred to the enriched medium. At the end of three days at breeding room temperature all larvae were lively and well and three had pupated. At the end of eight days one had died, the rest had emerged and all adults were of good size. On conclusion of the experiment the reaction of the water had reverted and indicated a pH of 7·6.

EXPERIMENT IV.

A further experiment was carried out with *A. punctor* larvae. In the previous experiment it is noted that in tap water pH 5·6, *A. bifurcatus* did not grow or develop within fourteen days.

*A. punctor* larvae placed in this water all emerged within twelve days but were poor specimens.

*Aedes detritus*, a salt marsh species, is not found in this district, but *A. punctor* larvae placed in tap-water pH 5·6, which included silt from a normal *Aedes* breeding place and an addition of 0·3 per cent NaCl, grew apace, were vigorous specimens and bred out within twelve days. In the same water, containing 0·3 per cent NaCl but excluding debris, the larvae died one by one and all developed cannibalism and could be seen running their mouth brushes around the bodies of their dear departed. The last larva, a worthy successor of the hero of the "Nancy Brig," died on the twenty-first day, having attained a good size and being in the 4th instar.

It is difficult to provide a satisfactory interpretation of the above. It is known that this particular water gives a high total colony count and apparently the food content is sufficient to sustain life and permit of development. In the presence of excess food (silt) the larvae thrived, and the question arises whether the fatal results are due to deleterious effect of salt upon larvae subsisting upon a minimum of food, or whether in the case of this particular water food factors other than bacteria hold the field and these are inimically affected by the abnormal inclusion of salt.

It may be urged that if in Experiment III the transference of larvae had been delayed they would have eventually developed. This is partially true; a few of this species have been known to survive in this water and after

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1 Note.—We are now using this simple and very satisfactory method for the maintenance of stocks of *A. bifurcatus* larvae for winter demonstrations, but no readjustment of water is made.
delay emerge, but at the same time the response to the beneficent change of environment is too sudden and complete to permit of an alternative interpretation. As a general rule it may be deduced that the *A. punctor* larva develops more rapidly than *A. bifurcatus*, that it is capable of development in the presence of a smaller quantity of bacterial life, and that *A. bifurcatus* can subsist for a longer period without undergoing development, awaiting the advent of more favourable conditions.

Although the above experiments appear reasonable particular attention should be paid to the time of year when they were performed. In mid-winter the response to warmth and suitable food in the breeding room is normally more noticeable in the case of the artificially reared *A. maculipennis* larva than with the imported *A. bifurcatus*. In late winter *A. bifurcatus* outstrips *T. morsitans* and *A. punctor*, but in the spring the positions are reversed.

The impression has therefore been gained that there are other latent agencies at work, and notably an inherent seasonal influence acting in conjunction with certain food factors.

While admitting that the evidence in support is meagre, that the objections to these views are numerous and particularly that they are based on observations made under purely artificial conditions, still on further investigation they may prove very difficult to entirely refute.

*(To be continued.)*