MALARIA NOTES FOR THE ASSAM VALLEY

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The Assam Valley of the Northeastern Frontier, India, was of primary importance during World War II and it may well be important again. For almost three years during 1943, '44, and '45, when invasion was first threatened, the Seventh Malaria Survey Unit was active throughout the Valley gathering data which helped tremendously with the control of malaria in one of the most malarious areas of the world

Much survey work had been done prior to 1943 by both the British and Indians but it was quite some time before the Americans could put the knowledge available to practical use. General information was helpful but specific recommendations were, for the most part, a matter of trial and error, and it is certain that if the job had to be done again it would not be done in quite the same way. The region was and still is one where the problems are numerous, complex and extremely difficult.

The plain in Assam is a country with a rich anopheline fauna. During the monsoon period, May through November, the climate is extremely hot and very humid, making ideal conditions for the very prolific breeding of mosquitoes. Although the maximum daytime temperature varies between 90° F. and 100°F. throughout the entire year, the minimum night-time temperature has a much greater variation, between 40° F. and 78° F. It seems that the hot, humid nights, in addition to the heavy rains, really represent the essential difference between the summer and winter. In other words, the winters are dry with cold nights, the summers wet with warm nights. The days are always warm and pleasant.

Plains and hills make up the land form, and intensive cultivation is common practice. Rice grows everywhere in the water and serves as the chief article of diet. The outcroppings of better soils are utilized as tea gardens. This is the cash crop of the region, others are for subsistence.

The natural vegetation is of the tropical rain-forest variety with areas of lighter tropical forest species. The forests, in spite of the presence of countless land leeches which attach themselves to one's legs, are beautiful and truly a biologist's paradise. Striking sights are the hundreds of butterflies hovering along the foot paths, among the trees and vines, as are the beautiful colors and patterns of those butterflies attracted to the droppings of elephants and monkeys. A single sweep of the net over some of the manure very often yields the collector a half-dozen species of spectacular specimens.

In addition to the things which are considered beautiful there are others which are not. The area is highly malarious with a spleen rate which varies between 50 percent and 85 percent in the villages. Laboratory records in the tea garden hospitals indicate a parasite index of close to 50 percent in many localities. And, in spite of all control efforts during World War II, the number of primary attacks of malaria among Americans was quite high. It is apparent, then, that preventive and control measures for malaria and its vector are of significant importance throughout the Assam Valley.

Immediately upon arrival in the Valley, if it be during the monsoon period, one can see that water, both standing and running, is in abundance everywhere but is confined mostly to river beds during the dry season. A greater variety of mosquito species was taken during the dry season but a larger number of specimens were collected when water covered much of the land. Since such larval habitats as lakes, ponds and rivers were rather abundant in contrast to collecting sites such as open wells and tree holes, these sites were visited more often and consequently their typical larvae represented the greater pro-

portion of the total specimens collected. The habitats discussed are arranged in order of their importance as breeding sites of the vector species, *Anopheles minimus*.

Although many Buffalo Wallows. attempts were made to collect anopheline larvae from these places, none was ever found. The wallows are holes where buffaloes do must of their loafing. The animals are owned by the natives and are used for pulling carts, plowing rice fields, and doing other types of work which beasts of burden do. In the evenings they are unharnessed and left to roam about the fields. After the buffaloes eat as much as they can hold, they wander off to their favorite spots which may be a deep borrow pit, a pond, or an abandoned rice field—any place where there is water and much warm, slimy mud for them to wallow in. Whether or not manure is deposited in the water is unknown but the animals undoubtedly urinated in it (from the odor) and washed off much of their body sweat.

Man-Made Containers. Frequent investigations were made of the many fire barrels and other similar water containers throughout the Valley but none were ever found to be serving as anopheline breeding places. It is quite possible that Anopheles kochi should have been found breeding in this type of habitat. The protozoan Euglena sp. often was observed moving in great clouds near the surface of the water in the barrels, and many interesting larvae of other species of aquatic insects such as Dystiscus and the larvae of Aedes albopicus and Megarhinus sp. could be seen swimming about.

WATER-HOLDING PLANTS. The dense forests contain many unusual sites in which mosquitoes were found breeding, such as holes in the trunks of trees, reservoirs of water within bamboo stems, and certain plants such as orchids which sometimes hold small quantities of water, but no anopheline larvae were collected from these places. Even the half-eaten fruits of *Diospyros birmanica*, the elephant apple, lying about the ground fill up with water and serve as the breeding place of

many species of mosquitoes. Within the bamboo baris (thick growths—sometimes an acre or more in area) where cuttings are made for commercial uses, it is almost impossible to walk during the day or night because of the large numbers of blood-thirsty Acdes albopictus and Culex fatiguns breeding in the cut stems.

Since these open spaces within the patches of bamboo are protected from view by over-flying planes they serve as storage areas for drums of gasoline and other items, and because there is much traffic in and out by military personnel it is necessary to take measures to control the mosquitoes. These habitats were never considered as serious producers of malaria vectors.

RING WELLS. These are vertical cement pipes, 4 to 7 feet in diameter, completely open on top, with the surface of the water anywhere from 10 to 30 feet below the surface of the ground. They were used almost constantly by the Indians, but in spite of those disturbances, mosquito larvae were commonly found in the muddy water. Quite often the entire inside of the well was covered with ferns, and frogs and toads could sometimes be seen swimming about in the water. The only two anophelines collected from the water of open wells were A. kochi and A. vagus.

CULTIVATED RICE FIBLDS. As the rains begin in the spring, open-field pools are plowed, rice is planted, and intense cultivation begins. In these places, where there was constant activity, no A. minimus larvae were ever taken, but A. hyrcanus and A. philippinensis were found in large numbers. The rice fields as such were not a serious problem.

Borrow Pits. In order to build roads and other structures which would remain above the water level throughout the year it was necessary to remove dirt from areas close by, thereby leaving holes which would fill with rain water during the mon soon. In these places, mosquitoes would breed in large numbers. Wagon ruts and roadside puddles were so very similar in all respects that they were considered as part of the same type of habitat. In all of

these sites, the water was usually shallow (r to 2 feet in depth) and quite muddy. Both shoreline and submerged aquatic vegetation were scarce. Unicellular algae, however, were extremely abundant in the water in spite of its being muddy. A. hyrcanus and A. vagus were the most common anophelines taken but specimens of A. minimus larvae were found in very small numbers.

Forest Pools. In the Assam Flood Plain, the forests contained many different types of mosquito habitats including flowing streams, both temporary and permanent, stream-bed pools, and typical emporary forest pools. The latter vary in size from 10 square feet to several hundred square feet and closely resemble American woodland pools. The water was usually ea-colored with an abundance of organic natter in solution. Aquatic insects were lways quite numerous. Eleven species of anopheline larvae were collected in hese places with A. barbirostris, A. barbiostris var. ahomi and A. hyrcanus the hree dominant species. These places ould be considered potentially dangerous ince quite a few larvae of the vector speies were taken from such dark, cool pools.

STREAM-BED POOLS. The dry season asts from November until May with a veek or two of rain occurring in Decemer or January. The streams or pools radually dry out after the rains stop so hat by the first of December, everything almost dry. The "out-of-season" rains etard this complete drying-out so that the treams continue to flow, but very slowly, ntil February. From then until the rains egin once more, 90 percent of the water missing. Only the large rivers remain owing but their volume is greatly reuced. The true ponds and lakes of the rea remain but are greatly reduced in In the beds of the smaller streams, atic pools of water are all that are left f the stream, and it was in these places hat the greatest variety of anophelines as found. A. hyrcanus dominated but . minimus was abundant enough so that some attention had to be paid to these places by control units. In fact, it was recommended that they be treated heavily so that the pools could not serve as nurseries to hold the insects over until the rains would begin once more.

LAKES AND PONDS. A rice field, in fallow, is quite unlike one which is under cultivation. It seems to resemble an openfield pool which is more advanced in its seral development. The filling-in with aquatic vegetation causes it to acquire the characteristics of a pond or lake. Organic matter seems to be plentiful in the ponds and lakes indicated by the abundance of protozoa such as Vorticella, and Volvox. crustacea such as Daphnia and Cyclops, and other indicators such as fresh-water sponges and dense aquatic vegetation including many algae and the bladderwort, Utricularia. Anopheles hyrcanus and A. annularis were most abundant but A. minimus was found in significant numbers.

OPEN-FIELD Pools. These pools of rain water in the open fields are sometimes created by streams which overflow during heavy showers or by streams diverted from their course by the presence of barriers such as masses of floating hyacinths or logs. Sometimes they are purposely created by Indians for the growing of rice. Bunds, the earth mounds (15-24 inches in height), which are constructed by the Indians for diverting the normal flow of water for the purpose of impounding an acre or two of shallow water, are extremely abundant throughout the Vallev. These pools change rapidly in appearance, becoming more and more pondlike which suggests that in reality they are very early stages in a seral development. Possibly because of a movement of water, A. minimus larvae were found in these pools but were absent in the cultivated rice fields. A. philippinensis was lacking but, as usual, A. hyrcanus was most numerous.

The next three habitats described are those which produced the greatest number of A. minimus, the vector species. It was in these running waters where most of the serious control work was carried out.

Forest Streams. Due to the decreased amount of light which falls on the surface of the water and its relatively low temperature, aquatic plants are not nearly so abundant as they are in openfield streams. Forest streams have rocky bottoms, large patches of gray sand, clear cold water, mayflies, no floating vegetation such as duckweeds, hyacinths, or shell flowers, and with stream species of fish darting about. A. burbirostris var. ahomi was most common but 20 percent of the total were A. minimus.

OPEN-FIELD STREAMS. Possibly due to the warmth of the water and the long sunny hours of the tropics, these streams are filled with aquatic vegetation. The water is extremely clear and the depth rarely greater than 5 feet. Vallisneria and the Potamogetons predominated. The common duckweeds cover much of the water surface especially where the current is retarded. The fish do not have spindle-shaped bodies which are ordinarily characteristic of flowing-water species, but instead are relatively big and bulky, and all are very brightly colored. Damselflies and dragonflies seemed to be more abundant than the mayflies. fourth of the larvae collected Anopheles pallidus and about a fifth were of the species A. minimus.

RIVERS. The rivers of the Assam Valley are wide, shallow, and sluggish. Usually the water is so muddy that aquatic plants are entirely absent. Sandbars occur frequently. Wherever shoreline vegetation grows thickly and falls over into the water, anopheline larvae can be found. Water hyacinths, which are produced in tremendous numbers high in the watershed and wash down into the main channel, serve as a shelter for the flowingwater species of anophelines. Wherever these conditions do not exist, the action of the water and sand tend to interfere with the production of mosquitoes. The limiting factor seems to be a physical one since other conditions are optimum for the growth of larvae. Unicellular algae are plentiful enough so that food is no problem for the larvae. By clearing away all living plants and brush from the banks of the rivers close to military sites, malaria-control units can expose the larvae to the scouring action of the current and to predators.

A. minimus, A. annularis, and A. aconitus are the three anophelines most commonly found in rivers. If, along the course of the river, there is a lake or pone which is periodically flushed out, this semistagnant body of water usually produces large numbers of A. annularis Rivers with their islands of floating hyacinths produced more anopheline larvae than any of the others with A. minimum making up one-fourth of those produced

During the monsoon when the malaris rate is at its peak, larvae of A. minimum may seem to be almost absent; actually however, breeding may be substantially in creased but the larvae more difficult to secure because of the high water.

During 1943, '44, and '45, the following anopheline larvae were collected: Anopheles hyrcanus var. nigerrimus Giles, Annuluris Van der Wulp, A. aconitu Donitz, A. minimus Theobald, A. barbir ostris Van der Wulp, A. philippinensi Ludlow, A. barbirostris var. ahomi Chauchury, A. vagus Donitz, A. pallidus Theobald, A. kochi Donitz, A. naculatus Theobald, A. aitkeni var. bengalensis Puri, Astephensi Liston, A. culicifacies Giles, Ainsulaeflorum Swellengrebel, A. subpictu Grassi, A. gigas var. simlensis James, Aleucosphyrus Donitz,

Adults only were collected of the following species: A. umbrosis Theobald, Avaruna Iyengar, A. fluviatilis Theobald A. tessellatus Theobald, A. jamesi Theobald, A. jeyporiensis James.

During World War II when much suvey work was being done throughout the Assam Valley, the majority (78 percent of the A. annularis adults collected we taken from cow sheds whereas most of the adults of A. minimus (88 percent) we taken in the native quarters. It was also observed that in spite of the fact that minimus and A. aconitus were breeding.

in about equal numbers close to coolie lines where collections were made, ten times more A. minimus adults were taken in the sleeping quarters than A. aconitus. A. minimus did not confine itself, however, to the sleeping areas of the huts. They were found in the other parts of the houses but no stratification was ever definitely observed. In all, 14 species were taken in the native huts with about 97 percent of the total catch consisting of six species.

In the tea garden coolie lines, the malaria rate seemed to be inversely proportional to the number of A. minimus, a fact which suggested that some other species might be transmitting the plasmodia. A closer study revealed that this paradox was brought about by the differences in basha construction. huts which consisted of one or two large, well ventilated rooms were much more sanitary, comfortable, and attractive than those having several small, dark, poorly ventilated rooms and they also accommodated mosquitoes much more satisfactorily. From night to night, insects in search of blood found it an easy matter to enter these dwellings and leave at will, whereas entry was more difficult in the others and escape was almost impossible. Therefore, during the day there would be an accumulation of the mosquitoes in the dark corners of the huts, and at night, quite logically, persons sleeping in the better-ventilated huts were bitten more often than were those sleeping in the other huts. This was observed to be true during several night-time investigations of the lines.

On several occasions, adults of A. minimus were collected at night on trains coming into the controlled areas. However, most of the adults flew into the camps, assisted possibly by the prevailing winds. This point requires further study.

Regardless of how A. minimus got into the controlled areas, during the day-time they were found not in the barracks, latrines, or other buildings but hiding in the dark crevices of tree trunks close to the ground. At night they were out on

the screens of the barracks trying to get in to feed.

A. vagus was not found in large numbers feeding on humans. A. annularis preferred cow and goat blood to that of humans. In contrast, A. hyrcanus, A. aconitus, A. philippinensis, and A. minimus were found feeding in coolie huts at the source of infection, and were also found feeding in the quarters of uninfected persons. However, A. minimus, which was known to be infected much more than any other species in Assam, definitely dominated the mosquito population of coolie huts between the hours of midnight and daylight. Some comparison can be made by referring to Table 1.

The information given did not, by itself, answer the question as to how American soldiers became infected with malaria and what could be done about it. By questioning new hospital patients and other Americans and also by closely studying the customs of the local people and the construction of their homes and villages much more was learned.

For example, many imported native laborers were housed in poorly constructed buildings close by American barracks. No effort was made to prevent the breeding of mosquitoes or their biting the natives at night in these areas. Since they were sometimes within the flight range of the mosquito, the source of infection was close by at all times.

Discipline was usually good about the barracks area of American camps but men on the job late at night sometimes became careless. This was especially evident about landing strips and among railroad operating units. These are just two examples which, possibly, are typical. Many areas seemed to be "saturated" with infected A. minimus so that no matter where a person worked or slept, if he exposed himself at night, he became infected.

Although mosquito bars were commonly used by American personnel, the comforter (or blanket) which was com-

TABLE 1 - Activities of adult anonhelines.

	Evening	Late night	Morning
A. minimus	•	Occupied tents and coolie huts, to feed	Dark, cool resting places
1. aconitus	Coolie huts to feed		Dark, cool resting places
1. annularis	Cow sheds to feed	Cow sheds to feed	Dark, cool resting places
1. hyrcanus	Lighted places and coolie huts to feed	f	Dark, cool resting places
1. philippinensis	Lighted places and coolie huts to feed	?	Dark, cool resting places
. vagus	Occupied tents to feed	Occupied tents to feed	Dark, cool resting places

monly used to hold the bottom down was neither heavy enough nor wide enough. A large percentage of the men were sleeping with their nets wrapped close to their bodies.

Much, too, was learned from the Americans who were unlucky enough to become infected. During June and July of 1944, 288 new patients were questioned relative to their activities during the three weeks prior to their admission to the hospital. Nineteen percent of the men had been engaged in combat where very little could be accomplished in the way of preventing infections. Eighteen percent were traveling in uncontrolled areas enroute to their camps and another eighteen percent had duties which took them outside the controlled area every day. Sixteen percent of the men seemed to spend all their time inside the malaria controlled areas and apparently followed all suggestions offered by the control units. However, 29 percent of all the primary infection patients were living in controlled areas but were working at night.

Suggestions for Controlling Malaria

Conditions vary of course throughout the Assam Valley, but, in general, because of the pressure of A. minimus from countless sites where the species is abundant and the engineering difficulties of attempting to confine all of the existing water in a minimum of controlled flowing ditches, the anopheline A. minimus is difficult to control and impossible to eliminate. However, ditches serve to decrease the number of locally-produced mosquitoes. They should be kept entirely free of rubbish and other debris so that they flow properly.

If a similar military operation should again be set up, discipline should be rigidly enforced especially where and when men work during the night. Outlying landing strips and railroad operating stations should be watched. Army personnel should not be permitted at any time to visit certain local villages, where the malaria rate is high. Proper clothing, antimosquito fogs, bed nets, and repellents should be available and properly used.

Bashas (barracks) should be well built with windows covered with mosquito cloth (or screen) in wooden frames, and in good condition.

Either T bars or four corner pieces of bamboo should be used to support mosquito bars. Bamboo bent in the form of an inverted U and placed at either end of the bed should be prohibited since it does not permit proper tucking in of the net. Charpoys (wide wooden beds) should be substituted for cots since they are wider and help to keep the mosquito-bar away from the sleeper's body.

An adjustment should be made at camp sites if at all possible so that only a very minimum number, preferably none, of the men should be required to work outside during the early morning hours (from two until six a.m.). Transient crews at airfields should not be required to guard

their own planes at night.

The movement of human carriers of malaria should be controlled. A department of Indian personnel directed by a member of a malaria control unit (and hiring one Indian interpreter) should be et up. This department should know the ocation of all Indian labor camps and vilages within 2 miles of American camps and should keep records of all Indians employed as bearers, etc. The constant introluction of new labor gangs should be disouraged, bearers should be hired only when absolutely necessary, and a mininum number of persons from outside should be allowed to remain in the area. Those persons remaining should be given as much assistance as possible to prevent being bitten by mosquitoes. Indian labor hospitals, especially, should be properly screened in order to prevent the patients from being bitten by mosquitoes during periods when gametocytes are most numerous in the blood. Native villages should be sprayed daily, and repellent and netting issued. In other words-rather than import a thousand coolies, half of whom have malaria, to be quartered in open frame buildings without nets, in order to dig malaria-control ditches, the same money should be used to house satisfactorily a smaller number of malariafree men.

Malaria survey and control personnel should not be required to drill or stand regular guard duty unless completely separated from other Army units. Commissioned officers should be carefully selected and have a deep appreciation of the job to be done. The enlisted men under them. especially those with proper background and interest should be considered as fellow biologists.

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