

THE RESIDUAL SPRAYING OF DWELLINGS WITH DDT IN THE CONTROL OF MALARIA TRANSMISSION IN PANAMA, WITH SPECIAL REFERENCE TO ANOPHELES ALBIMANUS¹

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INTRODUCTION

Since the discovery of the transmission of human malaria by anopheline mosquitoes, the most widely applied methods of mosquito control, other than naturalistic methods, have been those of draining or larviciding breeding areas and screening dwellings. Another technique which has been long known but less widely applied has been that of destroying adult female mosquitoes found in dwellings. Orenstein (1913) records that as early as 1908 the daily capture of mosquitoes in dwellings was employed as prophylactic measure against malaria in the Canal Zone. The work of Russell and Knipe (1939, 1940, 1941) and Russell, Knipe, and Sitapathy (1943), in India, and the earlier work of De Meillon (1936), in South Africa, has recently served to focus attention on this method. These authors used pyrethrum sprays rather than hand catching.

Where the malaria vector species involved is a domestic one the value of such a method is at once apparent. Since the "seed bed" of malaria is in the human population, the killing of the very mosquitoes which feed on this population is obviously the most direct means of attack on the transmission of malaria. The method is in effect a further refinement of "species control" whereby selective destruction of potentially infected mosquitoes is attained.

In India, Russell, Knipe and Sitapathy dealt with anopheline vector species which were house-resting during the day. Using various pyrethrum sprays, and spraying at intervals of three to ten days, they secured excellent reduction of malaria rates.

In Panama and the Canal Zone, as well as through most of the lowlands of the Caribbean area including the West Indies, *Anopheles albimanus* is considered to be the principal vector of malaria. This species is wholly absent from dwellings during the daylight hours, or at best present in only small numbers. Pyrethrum sprays applied during the daylight hours in the manner of Russell, Knipe and Sitapathy would, therefore, probably meet with only indifferent success in the Caribbean area, although the recent work of Metcalf and Wilson (1945) might indicate that pyrethrins applied in heavy dosages on resting places would have residual effect. The realization of the special properties of DDT when applied

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to surfaces in the manner of a paint has aroused interest in the possibility of a new, simple and inexpensive approach to the control of malaria transmission—for DDT thus applied continues to kill and otherwise affect mosquitoes resting on treated surfaces for a prolonged period.

Even before the advent of DDT, interest in anti-adult spraying had been on the increase (Senior White and Rao, 1944, and Eddey, 1944). It has been superficially evident that the residual properties of DDT would be of particular value in treating dwellings. With the pressure for immediate results for the protection of Allied troops and civilian populations in malarious areas, vast numbers of dwellings were treated with DDT by the armed forces during the latter war years. The evaluation of the effectiveness of treatments of this sort is a difficult process at best. With the rapid movement of the war relatively little detailed information could be gathered.

A number of isolated experiments have been performed, however, and the results of this first work is now forthcoming in the publications of Senior White (1945), Gahan and Lindquist (1945), Gahan, Travis, Morton, and Lindquist (1945), Gahan and Payne (1945), Tarzwell and Stierli (1945), Knowles and Smith (1945), Metcalf, Hess, Smith, Jeffery and Ludwig (1945).

The work of Gahan and Payne (1945) on *Anopheles pseudopunctipennis* in Mexico is the only report of a residual spray experiment on a neotropical anopheline which has come to our attention at the time of writing, although we have visited other such experiments involving *Anopheles albimanus* in Veracruz, Mexico, and have word of work of this sort in progress elsewhere in tropical America.

THE EXPERIMENTAL AREA

In the summer of 1944, a field experiment was set up on the middle Chagres River, Panama, to study in detail the effectiveness of the DDT residual spray treatment of native dwellings in the control of malaria vectors and malaria. The area selected was one in which intensive studies of malaria and *Anopheles* have been carried on by Dr. H. C. Clark and his associates at the Gorgas Memorial Laboratory during the past fifteen years. The results of these various studies have been published over a period of years in this Journal, and in the Annual Reports of the Gorgas Memorial Laboratory.

The village of Gatuncillo was selected for treatment, while Guayabalito and Santa Rosa, adjacent villages, were used as controls. Concurrent observations were recorded for treated and control villages. All three of these settlements are located on the west bank of the Chagres River only a few feet above the water. The river here is ordinarily a sluggish stream with numerous backwaters and lagoons directly across from the villages being studied. (See plate I, figure 1.) Extensive dense mats of *Najas arguta*² provide the principal source of breeding for *Anopheles albimanus*, as well as protection from the numerous surface feeding minnows. In addition to the extensive *A. albimanus* breeding areas along the Chagres River proper, there is additional breeding of this species in small clear areas of tributaries of the Chagres, particularly behind the villages of Santa Rosa and Guayabalito.

² Determined by Prof. R. T. Clausen, Dept. of Botany, Cornell University.

In the past, water lettuce (*Pistia stratiotes*) was abundant along the Chagres and supported breeding of *Anopheles triannulatus*. The lagoons and backwaters

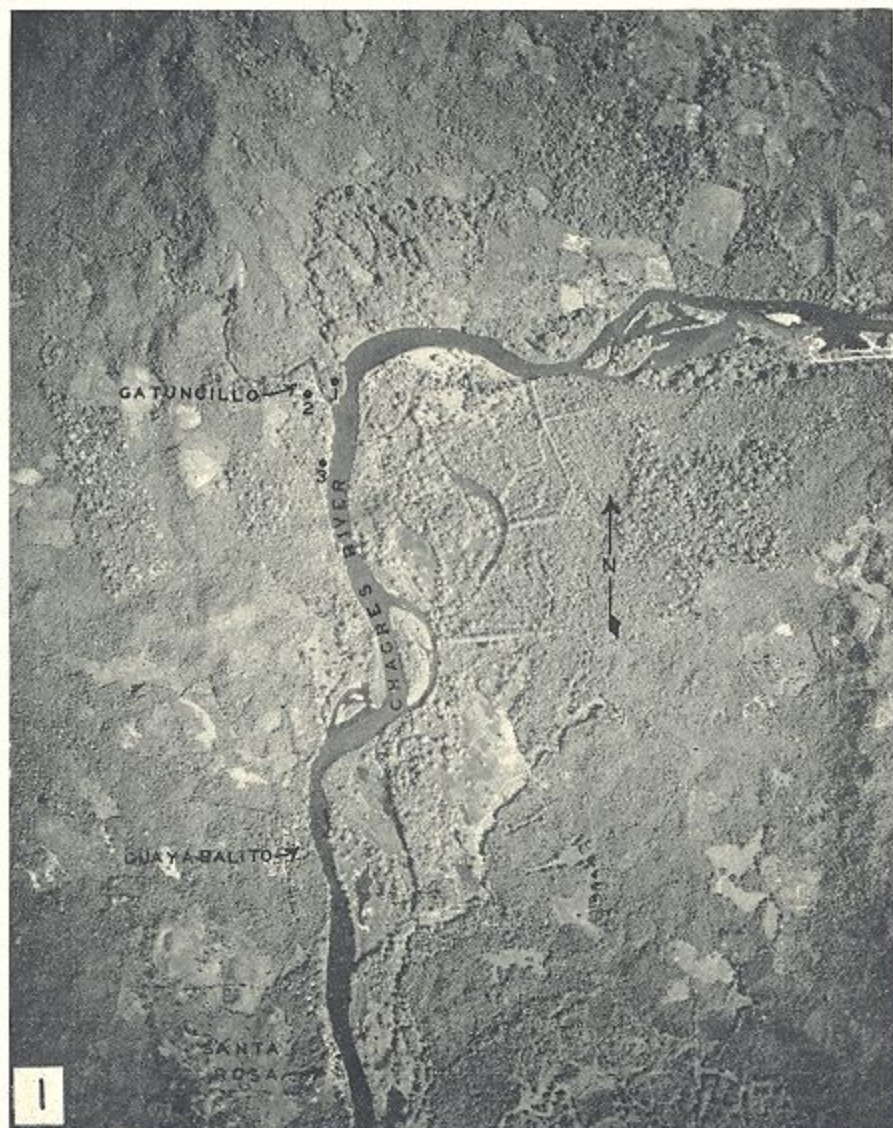


PLATE I, FIG. 1

AERIAL PHOTOGRAPH OF THE MIDDLE CHAGRES RIVER AREA

The treated and control villages on the west bank of the Chagres River are shown as well as the location of the three stable traps in and near Gatuncillo. The extensive breeding area of *Anopheles albimanus* in the backwaters and lagoons of the east side of the river, directly across from the native villages, may be seen.

of this area were artificially created with the formation of Gatun Lake, and the succession of aquatic plants in them has been such that water lettuce is now a minor species and possibly in consequence *A. triannulatus* is now not commonly

encountered. Adult females of *A. albimanus* and *A. triannulatus* may only be differentiated in part. In the present study it has only occasionally been possible to hold female *Anopheles* until oviposition to secure positive identification, based on egg characters. No *A. triannulatus* type eggs were found among house caught mosquitoes. Eight years ago, Rozeboom (1938) found only 0.5 per cent of the house resting *Anopheles* in this area to be *A. triannulatus* (= *A. bachmanni*) although the species was breeding abundantly at that time, indicating that this species "does not care enough about human blood to enter the houses to feed on man at night". In the present study we therefore uniformly regard the *Anopheles* of the *Nyssorhynchus* group as *A. albimanus*.

Anopheles punctimacula, which is also present in the villages studied, breeds in small numbers in small shaded brooks which flow into the Chagres. The fluctuations in numbers of *A. albimanus* and *A. punctimacula* in dwellings and horse-baited stable traps in this area will be considered in a separate paper, but it may be stated here that throughout most of the year *A. albimanus* is dominant, with *A. punctimacula* being taken in significant numbers only at the end of the rainy season (i.e., October, November, and December).

The area in which this work is being conducted possesses several particular advantages for an experiment on the DDT residual spray control of malaria. These may be summarized as follows:

1. The area is one which is accessible to the investigator, and yet sufficiently isolated so that the native population moves relatively little. The villages studied are inaccessible except by dugout cayuco or foot trail. Local studies of malaria in populations located on roads tend to be confused by the uncertainty of determining just where infection was incurred since the natives can freely and conveniently move along the road to attend markets, fiestas, etc. In the present situation movements occur but they are less frequent. The Gorgas Memorial Laboratory is equipped with dugout cayucos provided with outboard motors enabling investigators to reach the Chagres River villages, some five and one half miles from Gamboa (the end of the road), in approximately forty-five minutes. Natives lacking motor driven boats require several hours of paddling for this trip and are thereby somewhat deterred from leaving the villages. Information derived from malaria rates is therefore relatively reliable although necessarily subject to interpretation in individual cases.

2. The writer has been singularly fortunate in being able to work at villages where the malaria in the populations has been under continuous study by Dr. H. C. Clark and his associates for the past fifteen years. Malaria rates based on thick film blood examinations are available on a monthly basis for ten years, and on a bimonthly basis for the last five years. We began our experiment therefore with a large backlog of information on malaria in these communities.

3. Conditions are excellent for the continuous production of large numbers of the malaria vector species, thus providing a most severe test of the effectiveness of the control method under study. The level of the Chagres River, in the study area, is maintained by the impounded Gatun Lake, and while there is some fluctuation

tuation in water level, the breeding areas produce mosquitoes throughout the year. While some spraying with copper sulfate to kill aquatic vegetation is done by the Dredging Division of the Panama Canal, through agreement with the Chief Health Officer no larviciding procedures have thus far been employed in the study area. The confusing factor introduced by other means of mosquito control in the same area is largely eliminated and modifications in *Anopheles* abundance may be attributed either to natural causes, or to the techniques employed in the DDT residual spraying of dwellings.

4. The present study provides a severe and critical test of house spraying against adult female mosquitoes, since the species involved, *A. albimanus* and *A. punctimacula*, are not domestic as are, for example, *A. quadrimaculatus* or *A. gambiae*, which rest in human dwellings and domestic animal shelters during the day. While Rozeboom (1941) considers the adult female *A. albimanus* to be "very domestic" he also remarks that they "do not remain long in houses; most of them return to the jungle or to their breeding places soon after feeding or early in the morning".

Thus we are dealing with mosquitoes which seek dwellings not as resting places but as a source of a blood meal. If the residual house spraying method should prove successful against these species, it may be inferred that the technique will be even more effective against other malaria vector species which are house resting by preference.

5. The natives of the study area have been employed and trained as helpers and collectors by various members of the staff of the Gorgas Memorial Laboratory through the years and now constitute a good source of local assistants.

METEOROLOGICAL CONDITIONS

Meteorological information for the Chagres River villages is based on data from the Madden Dam Meteorological Station of the Panama Canal Section of Meteorology and Hydrography. This station is located approximately two and one half miles east of the village of Gatuncillo. Data for the period of the experiment reported here are given in table 1.

Weather conditions, particularly rainfall, are often very local in nature and the data from Madden Dam sometimes do not reflect conditions at the study villages. For example, in the severe storm and flood of December 13-15, 1944, which drastically affected the mosquito population, only 4.88 inches of rainfall were recorded at Madden Dam while 18.67 inches of rain fell at the Candelaria station about five miles away. In this same storm all twenty-four hour rainfall records for the Canal Zone area were broken, with 13.62 inches of rain recorded at Agua Clara. The annual rainfall at Madden Dam based on a forty-six year average is 98.25 inches.

The humidity at the Chagres river villages tends to be higher than that recorded at the Madden Dam Station since the latter is located on a hill above the river, while the villages are situated on the banks of the stream. On the occasions we have made hygro-thermograph records at Gatuncillo in the rainy sea-

son, we found the relative humidity to be virtually 100% throughout the night when the mosquitoes are active. Even in dry season the atmosphere becomes saturated by 10:00 or 11:00 p.m.

The data contained in table 1 will serve, however, to give a general picture of meteorological conditions in the region of the experimental villages.

NATIVE HOUSE CONSTRUCTION AND LIVING CONDITIONS

Construction in the villages of the middle Chagres River is of two sorts. The native huts are provided with steeply sloping palm-thatch roofs and walls of canes which are for the most part vertically arranged and secured with vines or, in some cases, wire (plate II, figures 2 and 4). Other houses are constructed of boards,

TABLE 1
Meteorological information, Madden Dam

	PRECIPITATION (IN INCHES)		NUMBER OF RAINY DAYS	AVERAGE RELATIVE HUMIDITY (IN PER CENT) (8:00 A.M.)	TEMPERATURE (IN DEGREES FAHRENHEIT)		
	Actual	46 Year average			Maximum	Minimum	Mean
Oct. '44.....	22.92		28	93.4	90	69	78.9
Nov.....	10.66		18	91.5	89	66	78.6
Dec.....	6.63		17	87.5	88	68	78.4
Jan. '45.....	.20	.98	6	86.8	89	61	77.6
Feb.....	.02	.59	2	79.6	89	66	79.6
Mar.....	.04	.49	1	78.0	93	67	80.2
April.....	1.05	3.17	9	81.4	95	68	81.1
May.....	12.44	11.25	21	86.1	94	69	79.8
June.....	5.95	11.60	21	91.1	94	70	80.8
July.....	13.28	12.22	25	93.4	90	70	79.6
Aug.....	11.79	12.09	26	95.8	91	69	79.3
Sept.....	14.08	11.35	23	94.6	92	67	78.5
Oct.....	9.44	14.74	22	94.1	90	67	78.3
Nov.....	10.54	13.96	27	94.8	90	68	78.4
Dec.....	8.58	5.81	16	92.2	89	66	77.2

mostly salvaged from abandoned Canal Zone houses, and roofed with sheets of galvanized iron. Various of the houses are intermediate in construction, with thatch roofs and board walls (plate II, figure 3). For the most part, houses consist of a single room with an incomplete partition or two of cane or scrap lumber, and a small extension where cooking is done on an open wood fire. These extensions are usually roofed with several sheets of corrugated galvanized iron. Interior walls and partitions are either bare cane (plate II, figures 6 and 7), or "papered" with pages from magazines, or sheets of newspaper (plate III, figure 8). The natives sleep on rough board beds, or in hammocks slung across the main room. The canes comprising the walls and partitions are only loosely apposed to one another, providing numerous spaces through which mosquitoes can gain entrance and egress. Crude wooden shutters serve to close window openings at night. Glass and screens are nowhere in use.

Other than beds, furniture consists only of an occasional open shelf cupboard, and rough wooden tables, chairs, and benches. Extra clothes are hung from



PLATE II

NATIVE HOUSES AT GATUNCILLO

FIG. 2. Native house showing typical construction of cane walls and palm thatch roof.

FIG. 3. House showing intermediate type of construction with palm thatch roof and rough board walls.

FIG. 4. General view down "street" at Gatuncillo.

FIG. 5. Interior view of roof of palm thatch construction. The impossibility of calculating precise amounts of DDT deposited on surfaces like these is evident.

FIG. 6. Interior view of house with cane walls showing loose apposition of the canes permitting mosquitoes free access to the interior.

FIG. 7. Interior view of house showing crude cupboard and cooking arrangement.

wooden pegs or nails, or stored in chests. As decorations there are occasional framed pictures, or horns of deer. Paint is rarely used.

Houses constructed in the manner here outlined seldom last for more than a few years. Even during the fifteen-month period of the experiment reported here, there has been considerable tearing down of old houses and construction of new ones. The village of Gatuncillo is composed of about twenty-five houses while the control villages combined have about twice as many houses.

There is another type of native construction not employed in the villages used in this experiment, but frequent in the dryer portions of Panama and elsewhere in tropical America. These dwellings are about the same size, but the walls are plastered with mud, or mixtures of mud and dung. Others obtain a solid wall by constructing walls of adobe blocks, cement, or native stone. Construction of the latter types is, however, more common at some elevation (largely above the range of *A. albimanus*), where temperatures are lower and better shelter necessary.

TABLE 2

Mosquito catches in houses at Santa Rosa and Guayabalito from October 1944 through December 1945

	NUMBERS	PER CENT OF TOTAL CATCH
<i>Anopheles albimanus</i>	33,381	62.4
<i>Anopheles punctimacula</i>	1,009	1.9
Total <i>Anopheles</i>	34,390	64.3
<i>Mansonia titillans</i>	16,225	30.3
<i>Mansonia nigricans</i>	2,605	4.9
Other Culicines.....	292	.5
Total Culicines.....	19,122	35.7
Total, all species.....	53,512	100.0

THE MOSQUITO FAUNA

Mosquitoes taken in the houses are principally of four species; *Anopheles albimanus*, *A. punctimacula*, *Mansonia titillans*, and *M. nigricans*. These mosquitoes are almost wholly absent from the houses during daylight hours, appearing at dusk, and departing at or shortly after dawn. Houses in which as many as 500 or 1000 mosquitoes may be taken by collecting throughout the night will have fewer than a dozen, or even no mosquitoes at all during the daylight hours.

A summary of mosquito catches in houses in the untreated villages of Santa Rosa and Guayabalito for the fifteen-month period from October 1944 to December 1945 is given in table 2.

An examination of table 2 shows that of 53,512 house-resting mosquitoes taken throughout the year almost two-thirds were *Anopheles*, mostly *A. albimanus*.

ENTOMOLOGICAL METHODS

While the malaria rate is the ultimate criterion of the effectiveness of the DDT house spraying method, some entomological check on the villages was obviously

desirable, not only to keep a close record on how the vectors were being affected, but also to provide a basis for decision on the time of retreatment. At first the attempt was made to secure evening biting rates on native human subjects, but these were soon found to be too variable to be of value, since the time of flight of the anophelines varies considerably with local meteorological conditions. This technique was therefore soon abandoned. Horse-baited stable traps were found to give a more reliable index of outdoor mosquito activity. The traps used were of the Magoon type with the ingress baffles modified as in the Egyptian trap of Bates (1944). Horses were purchased so that throughout the course of the experiment the same horse might be used in the same trap, thus avoiding any error caused by differences in the attractiveness of individual animals pointed out by Bates (1944).

To avoid the curious "catching-out" phenomenon reported by Gabaldon, Lopez, and Ocho-Palacios (1940), traps were never operated on successive nights. A schedule of trapping on Monday and Thursday nights of each week was set up and has been adhered to throughout the course of the experiment. Furthermore, at no time have the traps been moved from their original locations. By these precautions we hope to have established a high degree of reliability in the horse trap catches, permitting valid comparisons of catches over an extended period.

At first, three traps were set up: one across the Gatuncillo River about three hundred feet north-east of the village of Gatuncillo, a second in the center of the village, and a third in the forest on the bank of the Chagres River about nine hundred feet south of the village. These traps commenced operation on October 10, 1944. Later three additional traps of the same sort were set up. One in the village of Guayabalito 1.3 miles south of Gatuncillo, a second about 0.4 miles west of Guayabalito, behind a low ridge along the Chagres River, and a third at Juan Mina, 3.2 miles south of Gatuncillo on the east bank of the Chagres River. The locations of the first three traps are indicated in plate I, figure 1. Mosquitoes in stable traps were collected with a suction tube and chloroformed for subsequent identification and counting in the laboratory.

Several methods of collecting mosquitoes in houses were used before a satisfactory technique was evolved. At first chloroform tubes were used. These proved satisfactory for catching resting mosquitoes, but since DDT has a delayed action it was obviously desirable to collect the mosquitoes alive and hold them under favorable conditions to observe subsequent mortality. Catching tubes consisting of ten mm. glass tubing, in one foot lengths, with bolting cloth covering one end, and an attached rubber tube about two feet in length proved satisfactory for this purpose. As holding cages for the mosquitoes, we at first used glass lamp chimneys, with their ends covered with bolting cloth. If these were kept in a dry atmosphere the mosquitoes died; if kept in a moist atmosphere, slight changes in temperature would produce a condensation of moisture on the walls of the chimneys. On alighting, mosquitoes would adhere to the walls and perish. It was suggested by Col. W. H. W. Komp that holding cages of wire mesh or a similar material would overcome this difficulty. Cages of wire mesh were too fragile in the hands of native collectors, and for transportation between the field and laboratory. A series of cages were therefore constructed of per-

forated sheet bronze, four and a half inches in diameter and six inches tall. These were provided with a number eighteen wire screen bottom for convenience in examining the contents of the cage, and a bronze removable cover with a hole three-fourths of an inch in diameter. Through the hole in the cover mosquitoes could be introduced, by blowing, from the catching tubes. The removable feature of the cover was a convenience in cleaning the cages.

For transporting the cages of mosquitoes from the river villages to the laboratory we had constructed carrying boxes 9" x 11" x 22". These boxes contained eight compartments in the bottom of each of which a petri dish with moist cotton was placed. The holding cages were thus carried in separate compartments, each over moist cotton, and in an atmosphere of saturated humidity. All catches of house resting mosquitoes were handled in the same fashion, the mortality among the mosquitoes from the untreated villages serving as a control for that in the mosquitoes from the treated village. Catches were held in these cages for twenty-four hours, at which time the dead mosquitoes were removed, identified and counted. The surviving mosquitoes were then chloroformed and examined.

The catches of mosquitoes from houses were not only identified and counted, but also sorted as to whether or not they were blood engorged. Cards (5" x 8") were printed on which the information for each house, for each night's catch, was recorded. From these cards it is possible to tell at a glance the numbers of mosquitoes of each species which are engorged or unengorged, and the twenty-four hour survival. Summary cards for each house and each village were also routinely prepared at the end of each month.

House catches made during the daylight hours would in no way reflect the nature of the anopheline population visiting these dwellings. We therefore established a routine of collecting in houses after dusk, during the evenings, and mornings before dawn. Native boys were employed who collected in the house in which they were resident. It was not possible to establish an exact time period during which the boys collected, but each was instructed to catch all the mosquitoes resting in his dwelling between dusk and bedtime. In practice this was the period from about 6:30 p.m. to 9:00 p.m. In the morning the boys collected approximately between the hours of 5:30 and 6:30 a.m. No premium was placed on the numbers of mosquitoes collected, boys receiving a fixed sum for each night's collection no matter how large or small. Thus was avoided the temptation of the natives to secure mosquitoes from other sources than their own houses. On the whole this method has worked out satisfactorily. One native who persisted in collecting mosquitoes off tethered animals, when his house catches were low, was discharged.

While the interior of dwellings are exceedingly irregular and resting mosquitoes are in consequence not easy to see, the native boys quickly become adept collectors and a comparison of the catches from different houses on the same night shows a good correlation.

DDT VILLAGE TREATMENT

While it was at first thought necessary to spray a belt of vegetation around the village to be treated, a preliminary study of the habits of *A. albimanus* in-

indicated that spraying of the houses alone might be sufficient. A comparison of catches of mosquitoes taken in dwellings in the evening with those taken just before dawn showed a higher percentage of engorgement in the morning-caught mosquitoes. It was thus indicated that mosquitoes which have fed tend to rest on the conveniently nearby house surfaces for a period, before flying off to a more agreeable microclimate elsewhere for the daylight hours. This behavior characteristic of *A. albimanus* is of critical importance in the consideration of the effectiveness of the DDT house spraying technique in the control of malaria transmission. The "seed bed" of malaria is in the village dwellers and the engorged mosquitoes resting in the dwellings are, in consequence, those potentially infected. By selectively killing those mosquitoes which have fed on the occupants of the houses we may hope to control or terminate malaria transmission without necessarily exterminating, or even seriously modifying the mosquito population of an area.

It was early appreciated that the line of reasoning upon which the success of this method of using DDT rests is dependent on two basic premises. First, it was considered necessary that the human population be a relatively domestic one; i.e., a population which is for the most part indoors during the period of anopheline activity. The natives of the area here studied do not wholly meet this requirement. In the evening they are often abroad in the village, particularly during fiestas. The men are sometimes absent from the village when they remain overnight in shelters on the hillsides some distance from the village where they cultivate upland rice and corn. Data which will be presented below, however, tend to show that this difficulty is not insurmountable. The second premise is that the blood-engorged mosquitoes found in the dwellings are actually those which fed on humans and not mosquitoes which have fed elsewhere on animals other than humans and merely sought shelter in the dwellings. We have been able to check this point by precipitin test analysis of the blood-meals of house-resting mosquitoes.

Preliminary studies of the relation of *A. albimanus* to native dwellings, of the sort described above, showed that in large measure the mosquitoes approaching dwellings rest on the outer wall of the houses, and walk between the canes to the interior. Frequently, too, they enter by the space between the thatch roof and the walls. When houses of cane and thatch were provided with window traps it was found that approximately ninety per cent of the mosquitoes in a dwelling entered by some means other than the doors or windows. Further evidence that most mosquitoes find ingress to the dwellings by some means other than the doors and windows is found in the fact that at night the natives close the doors and shutters to their houses. Nevertheless hundreds of mosquitoes may be collected in these houses at this time.

It was therefore decided to spray the exteriors as well as the interiors of all dwellings so that the opportunity for mosquitoes to contact DDT might be enhanced. Inside the dwellings not only walls, but also any other possible resting places of mosquitoes, as the undersides of chairs, tables, cupboards, beds, and false ceilings were also sprayed. At the time of the first spraying, equipment was not available to reach the underside of the tall peaked roofs, but in subsequent

treatments these surfaces were also sprayed. Outside the houses the undersides of the overhanging eaves were sprayed as well as the walls. In addition to the human habitations all outhouses and animal shelters, such as chicken coops, were also sprayed inside and out.

A statement of the precise rate of application of DDT in terms of milligrams per square foot would be desirable, but is not feasible in view of the type of house sprayed in this experiment. While the plane surface area of walls, partitions, and roofs may be easily calculated, the rounded surface of the canes of which most walls and partitions are constructed (plate II, figure 6), and the multiple surfaces presented by the undersides of the palm thatch roofs (plate II, figure 5) and eaves makes it virtually impossible to establish the actual surface area treated with DDT. A more useful statement of the rate of application of DDT is the mean volume of DDT solution applied per house. Treating houses as outlined above, including the inner and outer walls, interior partitions, false ceilings, undersides of peaked roofs, eaves, and the undersides of furniture, we find that we use approximately three gallons of solution per house. A relatively uniform rate of application of the DDT solution was obtained by training spray operators to wet surfaces without permitting the solution to run. Close supervision of the work is necessary to secure uniform results. Since the construction of these houses is similar, both in terms of construction materials and size, throughout the Caribbean lowlands, this figure for the amount of material used should have wide application in this area.

Where house construction is more substantial, with the cane walls plastered with mud, or with walls of adobe, stone, or cement, the amount of DDT solution necessary per house is materially reduced since spraying inside alone should suffice, for mosquitoes do not gain entrance through the walls as in the circumstances of the present experiment. The relative effectiveness of DDT on surfaces of this sort is yet to be determined in the field.

EQUIPMENT FOR VILLAGE SPRAYING

The material used for the spraying was five per cent technical grade DDT in kerosene which was prepared from Army stock DDT and locally obtained kerosene. Various methods of applying this solution were tried. The work was mostly done with a small portable air-compressor such as is used with the paint-spray equipment for camouflaging jungle positions. This consists of a three-quarter horse-power air-cooled gasoline engine which operates a small compressor capable of delivering pressure of sixty pounds per square inch. This unit is easily carried by two men. An air line runs from the compressor to a liquid pressure tank of five gallons capacity. Attached to this tank are two hoses, one carrying liquid, the other air. These are then attached to a pistol-type spray gun which delivers a fan spray which is excellent for the purpose of applying an even coat of DDT solution. (See plate III, figures 9 and 10.) In practice, however, this equipment was found to be not wholly satisfactory. The two hoses from the liquid tank to the sprayer were cumbersome and permitted too restricted an area of movement by the spray operator, since the hoses were but thirty feet long.



PLATE III

NATIVE HOUSES AND METHODS OF APPLYING DDT

FIG. 8. Rough board partition "papered" with magazine covers and pages.

FIG. 9. Apparatus used in first spraying of Gatuncillo. In the foreground is the portable unit consisting of a $\frac{1}{2}$ horsepower air-cooled motor and air compressor. The five gallon tank containing the DDT solution is beside the figure at the left, the operator at the rear.

FIG. 10. The pistol-type spray gun used with this apparatus is shown here. Two hoses from the insecticide tank may be seen; that at the right delivers the insecticide, that at the left compressed air. A wet fan-type spray is used.

FIG. 11. For the second spraying a modified technique was used. A single hose from the insecticide tank is provided with the nozzle from a knapsack sprayer. This is a less cumbersome arrangement.

FIG. 12. In a later spraying hand-pumped cylindrical knapsack sprayers were used. The fan-type spray can be seen.

FIG. 13. The systematic coverage is accomplished by having the operator move slowly back and forth along the wall holding the spray nozzle at a uniform distance from the surface. A wet treated surface can be seen at the right and on the lower part of the wall section which the operator is spraying.

For a subsequent treatment the equipment was modified by substituting for the pistol-type spray-gun requiring two hoses, the disk-type spray nozzles removed from knapsack sprayers. These were provided with a number sixty wire gauge aperture which gave a wet spray satisfactory for this work. These sprayers required only a single hose, which simplified the work for the operator. (See plate III, figure 11.) To speed up the spraying, two and even three liquid lines could be run from the same tank, since only ten or fifteen pounds of pressure was necessary on each line to secure a satisfactory spray, and the compressor was capable of delivering as much as sixty pounds pressure per square inch. With this type of sprayer, hose extensions could be added so that each operator might have one hundred or one hundred and fifty feet of hose, thus allowing him great freedom of movement.

The fault with this arrangement lay in that a five gallon liquid tank contained sufficient liquid for only about fifteen minutes spraying and too frequent interruptions were necessary to refill the tank. We later improvised a liquid tank from a grease barrel which would hold fifteen gallons of liquid and this proved satisfactory.

Nevertheless it was considered desirable to try the village spraying with manually pumped cylindrical sprayers since labor costs are relatively low and the difficulty of effecting repairs to mechanical equipment in remote places is great. The simplest sort of spray equipment which will perform the work required is the most desirable under the conditions encountered in this area and Central America generally. In a field trial it was found that manually pumped cylindrical sprayers, of three or four gallons capacity, provided with a nozzle delivering a fan spray were excellent. The sprayers used were regular items of Army issue. (See plate III, figures 12 and 13.)

In the present experiment no attempt was made to develop ideal equipment since facilities for this sort of work were limited, and the village involved was not large. With the equipment here outlined we were able to treat the entire village in two days using two spray operators and an assistant.

Operators were supplied with respirators and heavy rubber gauntlet gloves, but these were frequently discarded since they were too uncomfortable for operators working in this tropical climate. Nevertheless, no untoward effects were observed in operators with the exception that one person using a spray gun which leaked as he extended his arm to spray high portions of walls sustained a kerosene burn on his arm and right side of his torso. This cleared up in a few days with no sequelae.

A more extended account of equipment for work of this sort may be found in the paper by Stierli, Simmons, and Tarzwell (1945).

ENTOMOLOGICAL RESULTS

House Catches

Mosquito abundance. The fluctuations in house catches of *Anopheles* and the principal factors influencing them are shown in Chart I and table 3. While the

catches were made semi-weekly (each Monday and Thursday night), in this and succeeding charts and tables the average for each week is shown to simplify presentation of the large volume of data available. Points of interest may be enumerated as follows:

1. In the pretreatment month, October 1944, the numbers of *Anopheles* in both the village to be treated and the control villages showed weekly fluctuations, but

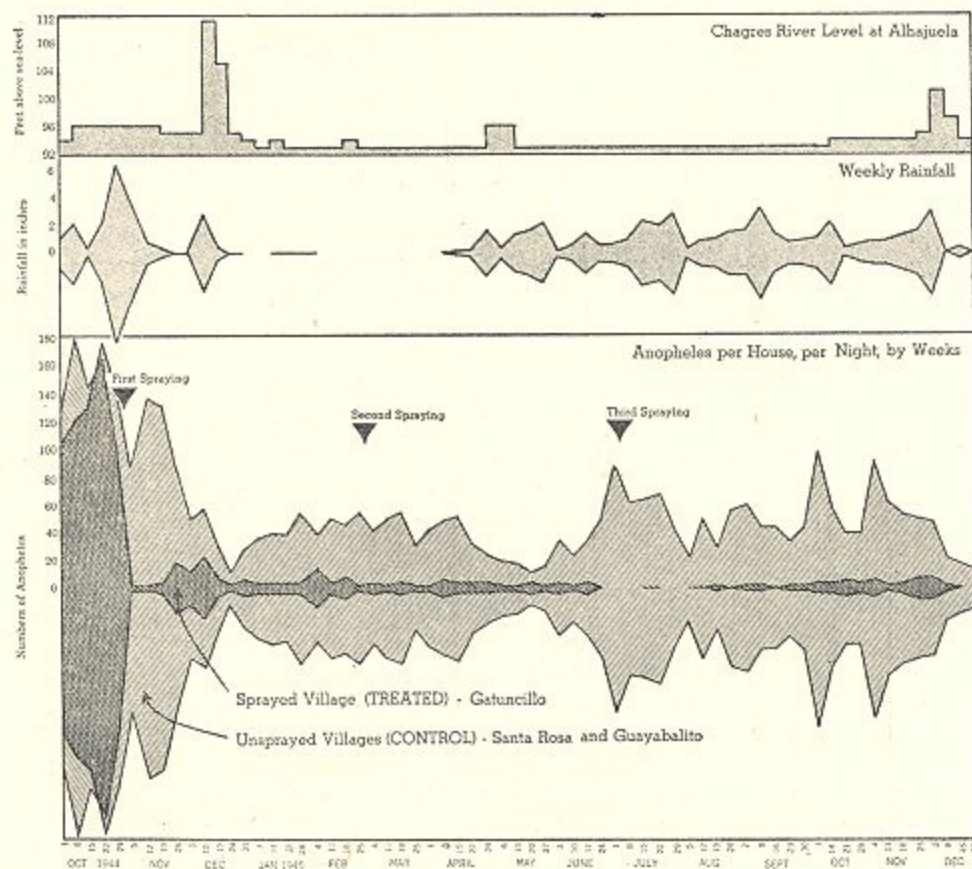


CHART I. CHART OF ANOPHELINE ABUNDANCE IN HOUSES OF TREATED AND CONTROL VILLAGES CORRELATED WITH RAINFALL AND RIVER LEVELS, MIDDLE CHAGRES RIVER, PANAMA

maintained a high level of abundance (from 99 to 178 *Anopheles* per house, per night). At this time the rainfall was relatively high.

2. With the first spraying of Gatuncillo, which took from the 31st of October to the 2nd of November, there was a dramatic drop in the numbers of *Anopheles* in the treated village, the average number of *Anopheles* per house, per night, during the ensuing three weeks being from 1.8 to 4.5. A gradual rise during the succeeding three weeks to 21.5 in the week of the 10th of December followed. During this six week period there is a decline in the numbers of *Anopheles* in the control villages associated with the drop in rainfall and the beginning of dry

TABLE 3

Anopheline abundance in houses of treated and control villages correlated with rainfall and river levels

WEEK OF	AVERAGE NUMBER OF ANOPHELES PER NIGHT		PRECIP. AT MADDEN DAM (IN INCHES)	MAX. RIVER LEVEL (IN FEET)	WEEK OF	AVERAGE NUMBER OF ANOPHELES PER NIGHT		PRECIP. AT MADDEN DAM (IN INCHES)	MAX. RIVER LEVEL (IN FEET)
	Control	Treated				Control	Treated		
Oct. 1, '44	102.3	128.5	2.12	94	May 6	19.5	1.5	.71	96
8	119.7	178.5	4.14	96	13	17.7	2.0	2.54	93
15	129.0	144.3	.68	96	20	10.9	3.3	3.20	93
22	174.9	163.0	4.37	96	27	15.2	1.3	4.24	93
29	142.2	99.0	12.55	96	June 3	33.0	3.5	.33	93
First DDT Treatment					10	23.2	1.3	.99	93
Nov. 5	87.3	1.8	7.51	96	17	34.6	4.0	2.69	93
12	135.2	2.3	1.55	96	24	50.3	0.5	1.27	93
19	130.3	4.5	.56	95	Third DDT Treatment				
26	85.9	18.0	.12	95	July 1	87.8	0.0	1.44	93
Dec. 3	49.9	12.3	.06	95	8	62.5	0.0	1.95	93
10	55.3	21.5	5.57	111	15	63.6	0.8	4.52	93
17	29.9	5.8	.96	105	22	67.5	0.3	4.07	93
24	11.2	1.3	.02	95	29	42.4	0.0	5.58	93
31	27.5	6.5	.03	94	Aug. 5	23.1	0.3	.80	93
Jan. 7, '45	35.9	4.3	.00	93	12	50.2	0.5	1.83	93
14	39.0	3.3	.01	94	19	29.4	1.5	1.96	93
21	37.9	4.3	.15	93	26	55.0	0.8	3.06	93
28	53.1	4.5	.02	93	Sept. 2	60.5	2.8	3.21	93
Feb. 4	37.5	13.5	.01	93	9	44.8	2.5	6.36	93
11	49.5	4.0	.00	93	16	43.8	0.5	2.79	93
18	45.0	7.3	.00	94	23	34.8	1.3	1.50	93
25	54.4	2.0	.00	93	30	43.4	1.8	1.87	93
Second DDT Treatment					Oct. 7	98.7	3.8	2.19	93
Mar. 4	40.4	3.5	.00	93	14	56.7	4.8	4.33	93
11	49.2	2.3	.00	93	21	60.5	6.3	.62	94
18	53.9	3.0	.04	93	28	59.6	4.0	1.19	94
25	30.6	1.3	.00	93	Nov. 4	91.9	5.0	1.66	94
April 1	42.4	1.0	.00	93	11	62.8	1.3	3.66	94
8	47.3	6.8	.09	93	18	53.7	3.5	2.39	94
15	52.3	4.0	.46	93	25	49.3	7.3	3.16	95
22	32.1	4.5	.47	93	Dec. 2	47.0	7.0	6.02	103
29	23.7	3.8	3.14	96	9	21.9	1.0	.02	97
					16	17.4	0.8	.85	94
					23	13.2	3.0	.11	93

season conditions. The week of the 10th of December the anopheline abundance in the control villages had about reached the usual dry season level. (Compare the portion of the chart for the dry season months of February and March.)

3. The week of the 10th of December was marked by a heavy rain, widespread over the entire Chagres River Basin, such as often occurs at the end of the wet

season. In this instance, however, the rain was of unusual intensity and duration. The recorded rainfall of 5.57 inches for this week at Madden Dam gives only a slight indication of conditions. Madden Lake rose so rapidly that it became necessary to discharge water from Madden Dam, producing a sixteen foot rise in the Chagres River at Alhajuella (see Chart I). The addition of a heavy inflow of water from streams tributary to the Chagres River below Alhajuella effected a rise of even more than sixteen feet at Gatuncillo.

This flood swept throughout the lowlands along the Chagres, carrying with it great masses of vegetation and even floating islands supporting trees. The decline in the control anopheline population during the latter half of December may reasonably be explained as the consequence of the destruction of so many mosquitoes by the torrential rains, and the sweeping out of daytime resting places as well as breeding mats of vegetation. The decline in numbers of *Anopheles* at Gatuncillo at this time is probably also attributable to this flood and is not a DDT effect.

By the end of January, however, the population at the control villages had been restored and reached a level maintained during the remainder of the dry season. The treated village, on the other hand, showed no such rapid recovery, the numbers of *Anopheles* per house, per night, seldom rising above five. These data imply that following the flood the DDT on surfaces in the treated village was able to prevent recovery of the anophelines to their normal dry season level as established in the control villages.

4. The decision to respray Gatuncillo at the beginning of March, four months after the original spraying, was not influenced by any increase in numbers of mosquitoes, but by the fact that the twenty-four hour survival rate of blood-engorged *Anopheles* was no longer reduced. (This point will be discussed later in this paper.)

5. The decline of the anopheline population in the control villages following the week of the 15th of April is associated with the rains of the beginning of the wet season, and a subsequent three foot rise in the river level at Alhajuella. This decline may be due to a flooding of breeding beds reducing anopheline production. In any event, the low level of anopheline abundance in the treated village is unaffected.

6. Following the third spraying of Gatuncillo at the beginning of July, virtually no *Anopheles* were taken in the house catches at this village despite the fact that this was a time when *Anopheles* were on the increase in the control villages. It will be noted that following each spraying there are progressively fewer mosquitoes in the houses of the treated village. This may well be due to a cumulative effect of the DDT, when the period for respraying is as short as four months. It might be anticipated, therefore, that routine DDT residual sprayings of villages at four-monthly intervals will progressively, over a period of years, produce better and better control of *Anopheles*.

7. With the excellent control effected after the third spraying of Gatuncillo it was decided to permit a six month interval to elapse before the fourth retreatment, to determine whether this interval rather than four months might not be

satisfactory (reducing costs in labor and material by one third for the year): Examination of the chart for the latter part of this period (October, November, and December, 1945) shows a gradual increase of *Anopheles* in the treated village. From these data it appears advisable to retain the four-month period for retreatment, by way of insuring complete control.

8. At the end of the 1945 wet season (October and November) the house collections of *Anopheles* in the control villages did not reach the levels of the same period the previous year. There are unfortunately no data available for com-

TABLE 4

Summary showing per cent of mosquitoes engorged, based on semi-weekly catches totalling 43,878 specimens

	ANOPHELES		CULICINES		ALL MOSQUITOES		
	Control	Treated	Control	Treated	Control	Treated	
Nov. '44	74.3	4.3	57.4	8.7	73.3	5.8	Period following first treatment
Dec.	51.9	3.7	33.6	7.5	46.3	5.6	
Jan. '45	45.0	16.1	33.8	13.1	39.4	13.7	
Feb.	46.7	7.3	45.8	17.9	46.3	13.2	
Mar.	46.8	4.6	32.7	12.1	41.0	7.7	Period following second treatment
April	45.4	11.1	36.3	0.0	40.9	6.3	
May	52.3	12.8	44.0	17.8	48.2	16.0	
June	55.1	2.7	36.9	8.3	47.6	5.5	
July	46.8	0.0	46.1	4.3	46.6	3.7	Period following third treatment
Aug.	42.6	16.7	32.1	5.6	38.8	8.3	
Sept.	31.9	3.6	27.5	8.7	29.9	6.8	
Oct.	27.4	6.8	27.4	3.8	27.4	6.0	
Nov.	31.4	17.6	35.5	10.2	33.3	13.2	
Dec.	48.7	43.9	34.0	46.9	39.9	45.4	
¹ Average Nov. '44-Oct. '45	47.4	7.1	35.4	11.1	43.1	9.4	

¹ These percentages are based on the totals of mosquitoes taken in houses from November 1944 to October 1945. November and December 1945 are excluded since they represent the fifth and sixth months respectively after a treatment period.

parison from years prior to 1944, but annual variations of the magnitude shown in this chart may not be wholly unexpected. It is also possible that native collectors were somewhat less diligent in their work after more than a year of routine collecting, and did not expend so great an effort in house collecting as they did the year previous.

Mosquito engorgement. The reduction in numbers of *Anopheles* in the houses of the treated village, as demonstrated above, indicates a large measure of protection to the village residents. The reduction in numbers of *Anopheles* represents, however, only the first step in the direction of terminating malaria transmission by DDT residual house spraying. In December 1944, several weeks

after the initial village spraying we made the following observation (Trapido, 1944):

"It is also noteworthy that of 135 mosquitoes collected in dwellings since the spraying of the village only four, or 2.9%, have contained blood. It is indicated that mosquitoes entering the treated dwellings and contacting the DDT residue become so activated that they do not feed."

We now have the records on the per cent engorgement in house-caught mosquitoes for fourteen months. These are summarized by months in table 4.

The marked and consistent difference between the per cent engorged in the control and treated villages is at once apparent. If we exclude those periods more than four months after a DDT spraying we find that house-caught *Anopheles* are from 27.4% to 74.3% engorged in control villages, while they are from 0.0% to 16.7% engorged in the treated village. Only in December 1945 did the per cent engorgement in mosquitoes from the treated village approximate that at the control villages. This was the sixth month after treatment, and it seems evident that after so long a period as this the "activating" effect of DDT does not persist. When retreatments with DDT occur at four-month intervals, the per cent engorgement of such *Anopheles* as do enter the dwellings remains at a low level.

If we summarize the percentages for the year November 1944 to October 1945, including three four-month post-treatment periods, we find that of *Anopheles* 47.4% were engorged in the control villages while only 7.1% were engorged in the treated village. In table 4 are also summarized the data for culicine mosquitoes. These data conform in a general way to the results obtained with the anophelines.

Just how the DDT spraying of dwellings serves to reduce the percentage of engorged mosquitoes is a point of much interest. As has been noted earlier in this paper, field observations have shown that most *A. albimanus* alight on the outer surface of the native dwellings and then enter between the canes of the walls or by other crevices, either by walking or by short flights. Once in the dwelling they rest on the walls, ceiling, or furniture, waiting a favorable opportunity to feed. Those mosquitoes not sufficiently affected by the DDT on these surfaces to die, or fly off to die subsequently, may receive sufficient DDT to cause them to lose interest in feeding. This "activating" or "irritating" effect produces an overall reduction in feeding. Effects of this sort are confirmed by the recent observations of Metcalf *et al.* (1945) on *A. quadrimaculatus* in the Tennessee Valley. They made observations in a DDT-treated house and a control house nearby and observed, "Although many mosquitoes entered both buildings, only 4 bites were received by the person in the control house." While there was some increase in the biting rate in the treated house after eleven days, they note that, "An estimated 500 mosquitoes entered the building during a 15-minute period at the break of dawn (5 a.m. to 5:15 a.m.), but only a few of them took blood meals".

This reduction of feeding on the part of such *Anopheles* as may be found in

treated dwellings, due to the "activating" effect of DDT, may be considered the second step in the possible reduction of malaria transmission.

Mosquito survival. There is yet a third way in which the house spraying with DDT serves to reduce malaria transmission. As has been mentioned above we have evidence that blood engorged mosquitoes tend to rest on house surfaces for a period before flying off to seek a diurnal resting place. It is of interest, there-

TABLE 5

Per cent of mosquitoes which are not engorged and survived 24 hours, based on semi-weekly catches totalling 33,538 specimens

	ANOPHELES		CULICINES		ALL MOSQUITOES		
	Control	Treated	Control	Treated	Control	Treated	
¹ Nov. '44							Period following first treatment
¹ Dec.							
Jan. '45	21.1	21.0	32.0	32.4	26.5	30.0	
Feb.	27.0	43.1	32.7	53.7	29.4	49.0	
Mar.	42.1	44.6	58.3	72.7	43.8	54.1	Period following second treatment
April	46.8	61.9	58.1	83.7	52.5	71.4	
May	42.5	59.6	51.5	58.3	46.9	58.8	
June	33.2	89.9	52.4	88.9	41.1	89.0	
July	33.8	0.0	36.5	47.8	34.3	40.7	Period following third treatment
Aug.	30.3	33.3	35.9	75.0	32.3	64.6	
Sept.	34.2	21.4	38.3	36.9	36.0	31.1	
Oct.	48.8	18.9	39.8	38.5	45.9	24.0	
Nov.	37.9	31.1	34.6	46.3	36.6	40.1	
Dec.	25.1	31.7	29.6	35.7	27.8	34.0	
² Average Jan. '45-Oct. '45	36.7	41.7	44.0	49.5	39.6	46.4	

¹ Data on survival rates not available for these months.

² These average percentages are based on the totals of mosquitoes taken in houses from January to October 1945. November and December 1945 are excluded since they represent the fifth and sixth months respectively after a treatment.

fore, to examine the survival rate in engorged and unengorged mosquitoes taken in dwellings, and to compare these rates in treated and control villages.

Data on survival of mosquitoes from both control and treated houses were routinely recorded from January 1945 onward. It was not until then that a standardized procedure for handling living mosquitoes and recording data on survival rates was established. We have recorded, however, (Trapido, 1944) that it was not until the 26th day after the first treatment that even a single mosquito from the treated houses survived for twenty-four hours after capture.

We have already outlined under "Entomological Methods" the technique employed to hold mosquitoes under favorable conditions to observe the twenty-four hour survival rate. These observations are summarized by months in

tables 5 and 6. Table 5 shows the per cent of all mosquitoes which were both unengorged and survived twenty-four hours. It is to be noted that, on the whole, for both *Anopheles* and culicines, there is little difference in the percentages recorded from the control and treated houses, although there are considerable monthly fluctuations. The wide range of percentages recorded for *Anopheles* in the "treated" column is due to the fact that these percentages are derived from relatively small numbers of specimens (since the DDT treatment enormously

TABLE 6

Per cent of mosquitoes which are engorged and survived 24 hours, based on semi-weekly catches totalling 33,538 specimens

	ANOPHELES		CULICINES		ALL MOSQUITOES		
	Control	Treated	Control	Treated	Control	Treated	
¹ Nov. '44							Period following first treatment
¹ Dec.							
Jan. '45	40.0	1.2	28.1	3.9	33.6	3.4	
Feb.	39.9	7.3	38.8	10.4	39.4	9.1	
Mar.	42.9	3.1	31.1	12.1	38.0	6.1	Period following second treatment
April	41.5	7.9	35.8	0.0	38.6	4.5	
May	50.1	8.5	43.3	16.7	46.8	13.7	
June	52.4	2.7	35.7	8.3	45.5	5.5	
July	41.2	0.0	38.3	0.0	40.7	0.0	Period following third treatment
Aug.	37.5	0.0	24.9	2.8	33.0	2.1	
Sept.	29.1	3.6	23.0	4.3	26.4	4.1	
Oct.	25.2	0.0	23.2	3.8	24.6	1.0	
Nov.	26.8	10.8	30.9	10.2	28.4	10.4	
Dec.	42.3	43.9	28.5	42.9	34.1	43.3	
² Average Jan. '45–Oct. '45	38.3	4.2	31.3	6.6	35.5	5.7	

¹ Data on survival rates not available for these months.

² These average percentages are based on the totals of mosquitoes taken in houses from January to October 1945. November and December 1945 are excluded since they represent the fifth and sixth months respectively after a treatment.

reduced the overall population). For the period from January to October 1945 there was actually a greater percentage of unengorged mosquitoes surviving twenty-four hours from the treated houses, than from the control houses, although this is probably a chance rather than a significant difference.

The data for the engorged mosquitoes which survived twenty-four hours are quite different (table 6). These data are of particular significance for malaria transmission, since they concern the potentially infected mosquitoes. The following points are noteworthy:

1. The twenty-four hour survival rate in mosquitoes from the control houses was relatively high. Reference to tables 4 and 6 shows that of *Anopheles* col-

lected in the control houses in the four-month post-treatment periods 47.4% were engorged, and 38.3% were both engorged and survived 24 hours. Thus among engorged *Anopheles* taken in untreated houses there was approximately 80% survival; such mortality as occurred being due to a combination of natural causes and the imperfections in our methods of transporting and holding these mosquitoes.

2. The reduced percentages of mosquitoes from the treated houses which were both engorged and survived twenty-four hours are striking. Thus for the period January to October 1945 only 4.2% of anophelines from treated houses were engorged and survived twenty-four hours as compared with 38.3% from the control houses. The comparison of percentages for culicines is approximately the same.

3. A comparison of tables 5 and 6 will also show that even in the treated houses the percentage of *engorged* anophelines surviving twenty-four hours (4.2%) is markedly lower than for *unengorged* anophelines surviving twenty-four hours (41.7%). The fact that engorged *Anopheles* show a decidedly greater mortality than unengorged individuals provides indirect evidence that the engorged *Anopheles* have a longer contact period with DDT. This is a consequence of the behavior characteristic we have already noted, i.e., engorged mosquitoes tend to rest for a period on convenient nearby surfaces before flying off with their heavy blood meal. In effect there is a selective killing of the engorged mosquitoes, a matter of primary significance in understanding the reduced malaria transmission potential.

4. It is of special interest to note the percentages in both tables 5 and 6 for December 1945, the sixth month after the third spraying. We find that DDT effects on engorgement and survival seem wholly wanting, there being no significant differences between control and treated houses. It is thus indicated that six months is too long an interval between retreatments.

5. We have earlier mentioned that the criterion for retreatment at the end of the first four-month period (March 1945) was not an increase in total numbers of anophelines, but the lack of reduction in twenty-four hour survival rate. If we compare, for February and June 1945, the percentages of *Anopheles* from the treated dwellings which are engorged (table 4) with those which are both engorged and survive twenty-four hours (table 6) we find that they are the same. It is thus demonstrated that while in the fourth month after treatment the blood-engorgement is still reduced, there is no twenty-four hour mortality among these engorged *Anopheles*. The lack of "delayed" mortality in the fourth post-treatment month might be taken as the criterion for retreatment at the end of three months. The total reduction of *Anopheles* and the low rate of engorgement in those which are found during the fourth month seem, however, to combine to give adequate protection despite the lack of "delayed" mortality. Beyond this point retreatment is indicated.

Summary. It is now of interest to review the possibilities of malaria transmission in the light of the "three step" control of vectors demonstrated above.

A summary of the progressive effect of each of the three steps is contained in table 7.

The per cent reduction of blood-engorged *Anopheles* for each four-month post-treatment period will be found to be even greater than that recorded under "All *Anopheles*". The blood-engorged *Anopheles* surviving twenty-four hours

TABLE 7

Average number of *Anopheles* per house, per month, based on semi-weekly catches totalling 37,406 specimens

	ALL ANOPHELES				BLOOD-ENGORGED ANOPHELES				BLOOD-ENGORGED ANOPHELES SURVIVING 24 HOURS			
	Control	Treated	Ratio of treated to control	Per cent reduction	Control	Treated	Ratio of treated to control	Per cent reduction	Control	Treated	Ratio of treated to control	Per cent reduction
'Oct. '44	1182.4	1058.0	0.895									
Nov.	952.8	23.0	.024	97.6	1174.5	1.0	0.001	99.9				
Dec.	371.0	109.5	.295	70.5	266.5	4.0	.015	98.5				
Jan. '45	259.0	40.5	.156	84.4	116.6	6.5	.056	94.4	101.0	0.5	0.005	99.5
Feb.	363.4	54.5	.150	85.0	169.6	4.0	.024	97.6	145.0	4.0	.028	97.2
March	375.4	32.5	.087	91.3	175.6	1.5	.009	99.1	161.2	1.0	.006	99.4
April	338.2	31.5	.093	90.7	153.6	3.5	.023	97.7	140.2	2.5	.018	98.2
May	154.0	23.5	.153	84.7	80.6	3.0	.037	96.3	77.2	2.0	.026	97.4
June	302.0	18.5	.061	93.9	166.4	0.5	.003	99.7	158.2	0.5	.003	99.7
July	514.6	2.0	.004	99.6	240.6	0.0	.000	100.0	212.0	0.0	.000	100.0
Aug.	358.4	6.0	.017	98.3	152.6	1.0	.007	99.3	134.4	0.0	.000	100.0
Sept.	367.8	14.0	.038	96.2	117.2	0.5	.004	99.6	107.2	0.5	.047	95.3
Oct.	581.8	37.0	.064	93.6	159.4	2.5	.016	98.4	146.8	0.0	.000	100.0
Nov.	571.4	37.0	.065	93.5	179.6	6.5	.036	96.4	153.0	4.0	.026	97.4
Dec.	185.8	20.5	.110	89.0	90.4	9.0	.100	90.0	78.6	9.0	.103	89.7
Average	407.8	32.1	.079	92.1	231.6	3.1	.013	98.7	134.6	2.0	.015	98.5
Nov. '44-Feb. '45	488.1	56.9	.117	88.3	431.8	3.9	.009	99.1				
Mar.-June	292.1	26.5	.091	90.9	144.1	2.1	.015	98.5	134.2	1.5	.011	98.9
Jul.-Oct.	455.7	14.8	.032	96.8	167.5	1.0	.006	99.4	150.1	0.1	.001	99.9

¹ Pretreatment month.

show yet higher reduction percentages. For the third four-month post-treatment period (July-October, 1945) the reduction of blood-engorged *Anopheles* surviving twenty-four hours reaches the impressive figure of 99.9%. The possibility of malaria transmission in the houses of the treated village during this period becomes exceedingly remote.

Another point emphasized in this table is the progressively increased effectiveness of each successive treatment. If we examine under "All *Anopheles*"

the column for "Per Cent Reduction" we find 88.8% for the first four-month post-treatment period (Nov. '44-Feb. '45), 90.9% for the second period (Mar.-June '45), and 96.8% for the third period (July-Oct. '45). This is probably due to the fact that in each case a residue of DDT remains from the previous treatment, and the fresh treatment adds DDT to that already present.

TABLE 8

Average number of culicines per house, per month, based on semi-weekly catches totalling 20,742 specimens

	ALL CULICINES				BLOOD-ENGORGED CULICINES				BLOOD-ENGORGED CULICINES SURVIVING 24 HOURS			
	Control	Treated	Ratio of treated to control	Per cent reduction	Control	Treated	Ratio of treated to control	Per cent reduction	Control	Treated	Ratio of treated to control	Per cent reduction
¹ Oct. '44	245.6	224.0	0.912									
Nov.	194.4	10.5	.054	94.6	56.0	1.0	0.018	98.2				
Dec.	362.8	98.5	.272	72.8	76.0	8.0	.105	99.5				
Jan. '45	257.0	152.5	.593	40.7	86.8	20.0	.230	77.0	72.2	6.0	0.083	91.7
Feb.	259.6	67.0	.258	74.2	118.8	12.0	.101	99.9	100.6	7.0	.070	93.0
Mar.	264.6	16.5	.062	93.8	86.6	2.0	.023	97.7	82.2	2.0	.024	97.6
Apr.	339.0	24.5	.072	92.8	123.2	0.0	.000	100.0	121.2	0.0	.000	100.0
May	151.8	42.0	.277	72.3	66.8	7.5	.112	88.8	65.8	7.0	.106	99.4
June	210.8	18.0	.085	91.5	77.8	1.5	.019	98.1	75.2	1.5	.020	98.0
July	108.0	11.5	.106	99.4	49.8	0.5	.010	99.0	41.4	0.0	.000	100.0
Aug.	202.4	18.0	.089	91.1	65.0	1.0	.015	98.5	50.4	0.5	.010	99.0
Sept.	300.4	23.0	.077	92.3	82.6	2.0	.024	97.6	69.2	1.0	.014	98.6
Oct.	285.8	13.0	.045	95.5	78.4	0.5	.006	99.4	66.2	0.5	.008	99.2
Nov.	365.8	54.0	.148	85.2	130.0	5.5	.042	96.8	113.0	5.5	.049	95.1
Dec.	276.4	28.0	.101	99.9	94.0	13.0	.138	86.2	78.8	12.0	.152	84.8
Average	255.6	41.2	.161	93.9	85.1	5.3	.062	93.9	78.0	3.6	.046	95.4
Nov. '44-Feb. '45	268.5	82.1	.306	69.4	84.4	10.2	.121	87.9				
Mar.-June	241.6	25.3	.105	99.5	88.6	2.8	.032	96.8	86.1	2.6	.030	97.0
Jul.-Oct.	224.2	16.4	.073	92.7	69.0	1.0	.014	98.6	56.8	0.5	.009	99.1

¹ Pretreatment month.

For purposes of comparison, similar data for culicine mosquitoes are given in table 8. These will be found to confirm, in a general way, the results obtained with anophelines.

Stable Trap Catches

The data from the horse-baited stable traps placed in and around the treated village are of much interest. A full discussion of the relation of anopheline abundance to meteorological and other conditions cannot be given here, but there

are several gross features of importance for this experiment. We can see from table 9 and Chart II plotting the abundance of *Anopheles* that, in the area considered, there are well defined seasonal fluctuations; the period of minimum

TABLE 9

Average nightly catches of Anopheles from horse-baited stable traps at Gatuncillo

WEEK OF	TRAP #1	TRAP #2	TRAP #3	WEEK OF	TRAP #1	TRAP #2	TRAP #3
Oct. 8, '44	384	746	90	May 6	6	15	17
15	402	402	183	13	5	16	5
22	460	860	265	20	12	15	9
29	567	865	192	27	8	20	5
First DDT Treatment				June 3	16	16	3
Nov. 5	126	35	91	10	26	15	18
12	152	47	149	17	34	71	86
19	223	121	164	24	23	52	26
26	199	272	126	Third DDT Treatment			
Dec. 3	170	200	370	July 1	15	33	28
10	133	328	262	8	8	11	50
17	100	23	120	15	13	27	63
24	136	11	88	22	34	52	54
31	80	56	133	29	19	44	39
Jan. 7, '45	34	42	40	Aug. 5	19	17	12
14	64	53	145	12	59	51	108
21	135	79	126	19	168	56	79
28	125	44	106	26	82	55	66
Feb. 4	97	136	180	Sept. 2	74	35	89
11	138	88	104	9	185	99	216
18	212	178	70	16	62	81	58
25	188	No catch	130	23	91	64	74
Second DDT Treatment				30	145	67	141
Mar. 4	42	8	83	Oct. 7	144	166	104
11	109	22	86	14	345	483	171
18	54	20	61	21	245	352	154
25	25	5	22	28	284	678	298
Apr. 1	7	9	39	Nov. 4	229	918	249
8	17	12	21	11	432	652	260
15	26	133	18	18	84	387	157
22	38	36	11	25	148	737	210
29	9	91	21	Dec. 2	189	611	271
				9	78	139	166
				16	121	105	124
				23	78	103	85

numbers being in May, that of maximum numbers being at the end of the wet season, during October and November. Examination of the curves for the last several months of 1945, at a time when no DDT had been applied for a number of months (the last treatment having been at the beginning of July) shows that the

natural drop in numbers occurs during the latter part of November or in December, coincident with the end of the wet season and flood conditions. If we consider then the portions of the curve for the last months of 1944 we note a decided modification of this natural trend. Examining first Gatuncillo stable trap number two, located in the center of the treated village, we note a dramatic drop from a level of 402-865 *Anopheles* per night, in October, to a level of 35-47 per night during the weeks of November 5th and 12th. The drop is obviously a consequence of the DDT house spraying the week of October 29th. This spraying was performed at a time when the *Anopheles* were at their maximum

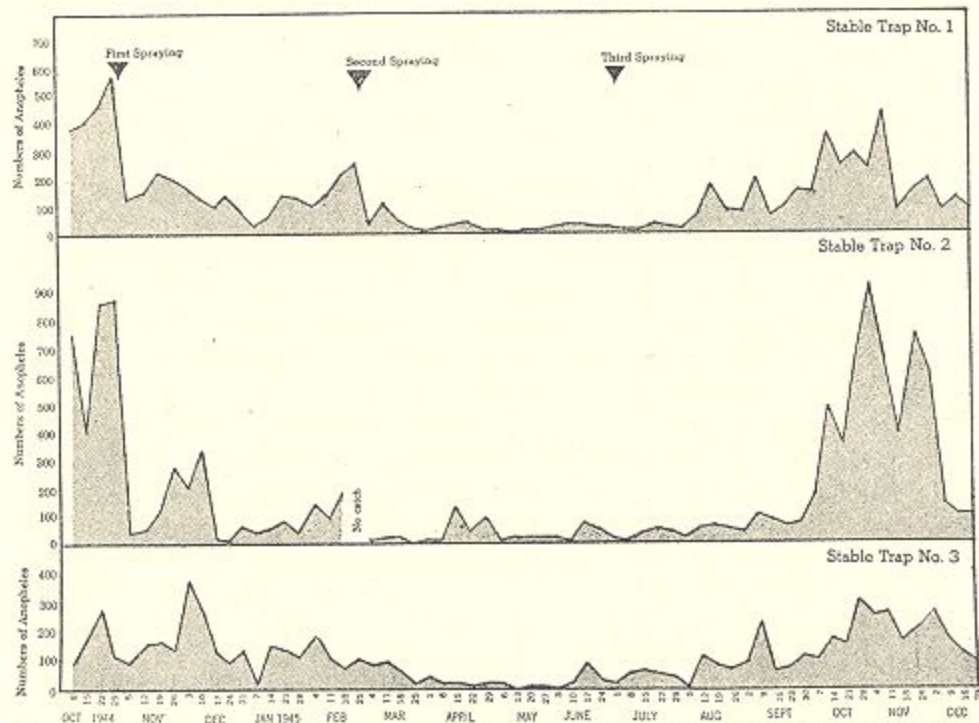


CHART II. CHART SHOWING ANOPHELINE MOSQUITO ABUNDANCE IN THREE STABLE TRAPS AT AND NEAR GATUNCILLO, PANAMA

numbers for the year, but in spite of this heavy population pressure, there was excellent control in the village area for two weeks. By the week of the 10th of December the population had recovered to the point where 329 *Anopheles* per night were taken. At this point the flood intervened, producing a sharp drop the following week. With the almost complete absence of rainfall after this point the population recovered only to the moderate level of abundance characteristic of dry season. There was unfortunately no catch made the week of February 25th, but the drop the following week, coincident with the second house spraying on March 1st and 2nd, is obvious. The drop at this time is more prolonged, persisting for six weeks. This may be due to the accumulative effect of a second spraying, the greater effectiveness of the spraying at a time

when anopheline population levels are small or moderate, or to an undetermined natural phenomenon. The third spraying of Gatuncillo on the 3rd and 4th of July came at a time when the anopheline level was low and little effect is apparent in the chart at this time. The normal expansion of the anopheline population in October and November of 1945 was apparently in no way inhibited by the prior sprayings.

Considering then the anopheline population of the village area outside the houses the following tentative generalizations may be made.

1. Even at times of great anopheline abundance a high degree of area control may be effected for a period of two to three weeks by a spraying of houses alone.

2. During a period of moderate anopheline abundance, effective control for six weeks following house spraying may result.

3. House spraying three months prior to a period of great anopheline abundance is wholly ineffective in preventing the normal increase of *Anopheles* in an area.

Having demonstrated that DDT spraying of houses may produce significant modifications in the anopheline population of the village area outside the houses, as well as in the houses themselves, we may now inquire into the effects of this treatment in the forest surrounding the village. An examination of the chart of anopheline abundance in Gatuncillo stable trap number one provides information on this point. This trap is located approximately three hundred feet north-east of the treated village (See aerial photograph, plate I, figure 1). While this trap does not catch such great numbers of *Anopheles* as trap number two, the general configuration of the chart showing captures in it is the same. The drop in numbers of *Anopheles* the week of November 5th, 1944 corresponds with that in trap number one, but is of lesser magnitude. In the month preceding treatment of Gatuncillo, October 1944, average nightly catches of *Anopheles* were from 384 to 567. For the two weeks following treatment catches dropped to 126 and 152. Again at the time of the second spraying a marked drop from 188 *Anopheles* per trap-night the week of February 25th, 1945 to 42 per trap-night the week of March 4th may be noted. As with trap number two, the mosquito populations at the time of the third spraying, July 3rd and 4th, 1945, were too low for any significant effect to be shown. The high anopheline populations of the following October and November are similar to those which appeared in trap number two.

From these data it may tentatively be stated that not only does house spraying in a village reduce the anophelines of the houses and the village area itself, but also significantly modifies the populations in the adjacent forest up to at least three hundred feet. House spraying alone, therefore, gives some measure of protection not only in the houses, and in the village streets, but in the forest immediately adjacent to the treated village. We may call this "peripheral effect".

The graph showing anopheline catches from stable trap number three, some nine hundred feet south of Gatuncillo (Chart II), may next be examined for further peripheral effects. It will be seen at once that there are no dramatic

drops in catches following the first two sprayings, as may be observed in the graphs for traps one and two. The peripheral effect apparently does not, therefore, extend for distances as great as nine hundred feet.

EFFECTS ON MALARIA

The effect on malaria incidence is the ultimate criterion of the success of the residual spray method. In the area here considered, where malaria is endemic, significant data from the malaria indices may be obtained only after a period of several years. Information which we now have, however, covering the fourteen-month period since the first spraying, is very suggestive of a significant downward trend in malaria incidence in the treated village.

TABLE 10

Comparison of malaria rates at three Chagres River villages, based on the cumulative rate in individuals examined either five or six times during the year

	SANTA ROSA AND GUAYABALITO	GATUNCILLO	DIFFERENCE
¹ 1940-41	45.6%	33.3%	-12.3%
² 1941-42	52.8%	60.9%	+8.1%
³ 1942-43	39.7%	46.7%	+7.0%
³ 1943-44	42.5%	45.5%	+3.0%
	Untreated	Treated	
² 1944-45	51.3%	24.0%	-27.3%
³ 1945	52.0%	14.8%	-37.2%

¹ Annual rate based on the year September to August.

² Annual rate based on the year November to October.

³ Annual rate based on the year January to December.

Malaria indices given here are based on the bimonthly thick blood-film surveys made by Dr. H. C. Clark and his technicians. These surveys are made each February, April, June, August, October and December. For convenience the "year" used by Dr. Clark is from September to August. Since the first spraying of Gatuncillo was performed at the beginning of October 1944, we have broken down the data from the bimonthly surveys and here present the indices for the one-year period November 1944 to October 1945 (table 10). This is the twelve-month period immediately following our first spraying. We have also extracted the data necessary to calculate the indices for the twelve-month period January to December 1945, the year commencing two months after the first spraying. The use of this last mentioned period eliminates positive blood-films occurring in the period immediately following the first spraying, i.e., malaria contracted prior to the first village treatment. The decline of the malaria index at Gatuncillo, in the period following treatment, is striking, the cumulative index being 14.8% for 1945 as compared with 52.0% for the same one-year period at the control villages of Santa Rosa and Guayabalito.

These indices for the 1945 period are unfortunately based on relatively small numbers of individuals, as but twenty-seven persons were examined either five or six times during the year at Gatuncillo, and seventy-nine at Guayabalito and Santa Rosa. While we have survey data from three to four times as many individuals, those persons examined less than five times during the year have been eliminated, since by their absence from the villages at the time of surveys they are disqualified as continuous residents of the houses with which we are concerned.

One difficulty in attempting to establish, by an examination of the parasite index, whether or not malaria transmission has been stopped, arises from the uncertainty as to whether a positive thick film represents a relapse from previously contracted malaria, or a fresh infection. In the villages here studied persons positive on blood thick film examination are treated with a five day course of atabrine of 0.1 gram, three times a day. (See Clark and Komp, 1941, and Annual Reports of the Gorgas Memorial Laboratory.) It has been indicated by McCoy (1945) and others that *falciparum* is less likely to relapse than *vivax*

TABLE 11

Comparison of *falciparum* malaria rates at three Chagres River villages, based on the cumulative rate in individuals examined either five or six times during the year

	SANTA ROSA AND GUAYABALITO	GATUNCILLO	DIFFERENCE
	(Control)	(Treated)	
¹ Nov. 1944–Oct. 1945	39.7%	12.0%	-27.7%
² Jan. 1945–Dec. 1945	39.2%	7.4%	-31.8%

¹ Twelve month period immediately following first spraying.

² Twelve month period commencing two months after first spraying.

following the administration of atabrine. It is therefore of interest to extract the index for *falciparum* malaria alone for the period following the first village spraying. These data are presented in table 11. The difference between the indices in the control and treated villages is even more marked than that noted in the preceding table which considers all forms of malaria together.

A more detailed analysis of the effects of the residual spraying on the malaria in these villages is in order, and will be made by Dr. H. C. Clark at a later date.

COST ESTIMATES

The materials used in the present experiment were DDT and kerosene. Based on the experience of several treatments we find that approximately three gallons of solution are used per house. This represents approximately a pound and a quarter of DDT per house. The cost of DDT has undergone great reduction in the period since this experiment began, when limited quantities for experimental use only were available. Stierli *et al.* (1945) use \$0.64 per pound in calculating costs, but recent information from the Office of the Surgeon General, U. S. Army, indicates that the current price in the United States, less overseas transportation costs, is \$0.46 per pound. The actual cost delivered to the various Caribbean

countries will vary with transportation costs and possible duties. For purposes of our calculations we will use the arbitrary figure of \$0.50 per pound. This should be a fair estimate of the cost of DDT in this area, at the time this report becomes available. Kerosene for this experiment was locally obtained at \$0.07 per gallon from the oil storehouse of the Panama Canal. The cost of materials for the treatment of each house is thus approximately \$0.84: \$0.21 for three gallons of kerosene and \$0.63 for a pound and a quarter of DDT.

To treat twenty-five houses we found it took two laborers and a foreman two days. Laborers received \$1.25 a day, the foreman \$3.33 per day. The total cost of labor to treat twenty-five houses was thus \$11.66 or \$0.47 per house. The combined cost of materials and labor was \$1.31 per house. For the three treatments per year here recommended the cost per house, per year is \$3.93.

There are several factors which influence this cost estimate.

1. The labor cost per house is high, since we treated only one relatively small village, and considerable time was expended in instructing operators how to apply the solution at a uniform rate. The same amount of labor, once proficient, and routinely engaged in work of this sort, could accomplish much more in a day and substantially reduce labor costs.

2. With the cane and thatch construction present in the village, spraying outside as well as inside houses was desirable. Elsewhere in this region where solid wall construction is used it would be necessary to spray only the interiors. This would substantially reduce the costs of both labor and material.

3. As we have noted previously the area of our experiment is one where the most difficult conditions exist since anopheline production proceeds throughout the year, even in the height of the dry season. In western Panama and elsewhere, where dry season conditions of three or four months seriously inhibit *Anopheles* production, it is probable that two treatments per year, strategically timed at the beginning and middle of the rainy season, would provide protection of as high an order as that obtained on the Chagres River with three treatments. The annual cost per house might thus be reduced by one third.

4. The equipment used in this experiment was Army issue and its cost and depreciation is not included in the cost estimate since data of this sort are not available. Were a long-term large-scale program of DDT residual house spraying set up, however, the initial cost of equipment would not add substantially to the per-house cost of the work.

5. In a large-scale program over a considerable area transportation cost would be introduced.

DISCUSSION

Of the non-naturalistic methods, there are two classes of mosquito control for reducing malaria transmission now universally employed. They are spoken of as permanent and temporary. Permanent control consists of such measures as drainage of breeding areas in a radius a mile or so about the area to be protected, and screening dwellings. These are relatively expensive measures but quite justified in areas of concentrated populations, as large towns and cities, particularly where individual incomes are substantial. Temporary measures

such as larviciding are less expensive but must be frequently repeated. In small rural communities and particularly in areas of scattered individual houses the cost of this work is often virtually prohibitive. It is also difficult to supervise work of this sort.

The DDT residual spraying of dwellings essentially meets the needs of this latter situation, providing the population is a relatively domestic one. The tendency to kill selectively the engorged, and hence potentially infected mosquitoes, makes for great efficiency. The technique tends to bring malaria control a step beyond "species control" to "*infected mosquito control*". The relatively low cost of the method makes malaria control possible among the poorer classes, those most needing it, since they now receive little or no protection, and constitute the main "seed bed" of the disease. It is an ironic but fortunate circumstance that DDT residual treatments will work best where the people are poorest. The lower income classes in the American tropics cannot afford and do not use paint on their dwellings. A characteristic of DDT is that it is far more effective, both initially, and in its residual effects, on unpainted surfaces. Since this low income group constitutes the main "seed bed" of malaria, effective control at this level will give added protection to the higher income classes, as the main source of mosquito infection will have been eliminated.

There is a psychological factor, wholly irrelevant to the malaria control picture, but nevertheless of great importance. The rural inhabitant of the American tropics with a meager education is ill equipped to appreciate the nature of malaria transmission, and it is difficult to impress him with the value of such measures as larviciding. DDT residual spraying of dwellings produces not only an immediate and obvious reduction of *Anopheles* in his home, but reduces the annoyance from pest mosquitoes as well (See tables on culicine mosquitoes). In addition, such household pests as cockroaches, ants, bedbugs, and wasps are markedly reduced. It was our experience that while there was at first considerable scepticism and some objection to the first treatment of the experimental village, the inhabitants welcomed retreatment and were cooperative in every way thereafter. A favorable psychological attitude of this sort facilitates operations and provides a body of public opinion which will tend to approve expenditure of public funds for the work.

SUMMARY AND CONCLUSIONS

This paper reports the results of the first fifteen months study of the DDT residual house spray method in the middle Chagres River area of Panama, where the principal malaria vector is *Anopheles albimanus*.

Houses of cane wall and palm-thatch roof construction at the village of Gatuncillo were sprayed, inside and out, with a 5% solution of DDT in kerosene at four-month intervals (excepting one trial period of six months). Entomological observations were made at this treated village and two adjacent villages used as controls.

It is demonstrated that with this technique anophelines visiting the dwellings are affected in three ways:

1. There is a large reduction in numbers of mosquitoes.

2. Among the mosquitoes which are taken in treated dwellings there is a marked reduction in the per cent engorged, since DDT activates the insects and they lose interest in feeding.

3. Among the engorged mosquitoes the twenty-four hour survival rate is low for three months after treatment.

It is thus indicated that the technique tends selectively to reduce the malaria transmission potential by affecting principally those mosquitoes concerned in transmission.

Four months is established as the optimum time for retreatment in the area studied.

It is demonstrated that with each successive treatment the degree of control improves. (For the period following the third treatment the malaria transmission potential is reduced 99.9%.)

Treatment of houses alone was found to produce marked reduction of mosquitoes in the village area outside the houses, and even to some measure in the forest adjacent to the village (for at least three hundred feet), for several weeks.

There is evidence of reduction of malaria in the fourteen-month period following the first treatment.

The cost of the treatment under conditions of this experiment is calculated as \$1.31 per house. Factors affecting the cost of applying the method elsewhere in tropical America are discussed.

The place of this method in relation to other malaria control procedures and to the economy of the area is discussed.

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