Public Health Reports

Vol. 63 • JULY 30, 1948 • *No. 31*

Printed With the Approval of the Bureau of the Budget as Required by Rule 42 of the Joint Committee on Printing

Transfer of Malarial Parasites between Blood Films during Mass Staining Procedures[1]

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Although earlier workers had mentioned staining more than one thick blood film at a time, Barber and Komp in 1924 (*1*) were the first to outline a practical routine for staining large numbers of slides simultaneously. Their method, with only minor changes, and other mass staining procedures are used for present-day surveys, in clinical laboratories, and in malarial research studies. A comprehensive discussion of the current application of the mass staining techniques and a review of the history of staining procedures for malarial parasites are given in the excellent manual by Wilcox (*2*).

Recently, several hundred thick blood films were made from a malarial therapy patient with a heavy infection of *Plasmodium falciparum*. When these films were examined after having been stained en masse, indisputable forms of *P. malariae* and *P. vivax* were observed. At first, it was believed that the patient from whom the slides were obtained simply had a mixed infection with the three species. However, more careful examination of the slides revealed that of 553 slides studied, 42 showed *P. vivax* parasites, 28 showed *P. malariae* parasites, and 5 showed both *P. vivax* and *P. malariae* parasites in addition to the numerous *P. falciparum* parasites. By questioning the technicians who did the staining, it was learned that the *P. falciparum* slides had been stained along with 1,100 other slides in two large pans. The 1,100 slides consisted of 650 thick films of a high *P. vivax* infection and 450 thick films of a heavy *P. malariae* infection, obtained from two other patients receiving malarial therapy. The exact distribution of the three types of blood films in the two pans is not known, but all slides in a given package were from a single patient. Since it did not appear logical that the therapy patients had a triple

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infection, the only alternative explanation of the facts observed was that parasites must have transferred from one slide to another during the staining procedure.

In line with the research program of this laboratory to evaluate diagnostic techniques for parasitic diseases (*3*, *4*), it was decided to test under more controlled conditions, whether, and to what extent, transfer of blood cells and malarial parasites may take place between thick films which are stained together by mass procedures. Exactly the same staining procedure was employed as had been used by the technicians who stained the slides described above. The procedure was not developed in this laboratory but had been introduced by two experienced technicians who had transferred from a laboratory engaged in extensive surveys of malaria. Although the slides were wrapped as described by Barber and Komp (*5*), the staining procedure was similar but not identical with that reported by Komp (*6*). However, it was decided to test the procedure which was used by the technicians since the same procedure, or one with only minor modifications, is used in a number of laboratories when many slides have to be stained for malaria.

Methods

In all of the following experiments, the techniques used for the cleaning of the slides and the preparation and staining of the blood films were essentially the same. Customary laboratory procedures were used throughout; however, for clarification, the details will be presented.

Cleaning of Microscope Slides.—Only new, 1- by 3-inch microscope slides of high quality were used in the experiments. The slides were immersed in 95 percent ethyl alcohol, dried, and polished with lint-free cloths. They were then protected from dust and used within 2 or 3 days. When performed by experienced technicians, this procedure has been found to be satisfactory for the cleaning of small groups of slides.

Preparation of Blood Films.—Thick blood films were used in all of the experiments. In the first four experiments thin films were also prepared on the same slides. The human blood was obtained by pricking a finger after it had been thoroughly cleaned with alcohol and dried. After the smearing of the thin films, the thick films were prepared by the circular technique without losing contact with the drop of blood and without touching the patient's finger to the slide (*2*). After the films had been prepared, the slides were inserted with the film side down in upright slide boxes and allowed to dry for 24 to 48 hours.

Grouping slides for mass staining.—After the films were thoroughly dry, the slides were labeled with a wax pencil and were bound to-

gether in groups of 25 or more, according to the technique of Barber and Komp (*5*). This method involves the inserting of cardboard squares between adjacent slides at the ends opposite to the thick films, the slides being held together in packages by rubber bands or string. The arrangement of the slides within the packages varied in different experiments and will be described with the experimental data.

Staining.—When thin films were used, they were fixed by dipping in absolute methyl alcohol. After packaging, the slides were placed in dishes which varied in size according to the number of packages to be stained together. A solution of Giemsa stain (1:50 dilution in buffered water, pH 7.0–7.2) was poured into the dish until it rose to a level adequate to cover the films.[2]

After 45 minutes, the stain was poured off and buffered water was introduced to the same level. The thick films were washed for 3 minutes. If thin films were present, after one minute enough water was poured off, to lower the level to just above the thick blood films. The thick blood films were then washed for two additional minutes after which all of the water was poured off. The groups of slides were then removed from the dish and placed upright on paper toweling to air-dry.

Examination of Normal Films.—In this paper the term "normal films" refers to films prepared with human blood taken from individuals who had no previous histories of malaria and whose blood showed no parasites. In the first experiment, which involved the use of avian blood, each normal film was examined with the 16-millimeter objective which gave a sufficient magnification to reveal the presence of avian erythrocytic nuclei. In the next three experiments the entire thick film of each normal slide was examined systematically under the oil-immersion objective for the presence of malarial parasites. A complete examination of a thick film under oil-immersion required from 2 to 4 hours, depending upon the size of the film. Therefore, in the last four experiments, the normal films were examined until indisputable malarial parasites were found or, if no parasites were found, until the entire film had been examined. The examinations were made by four experienced technicians who confirmed each other's observations.[3]

[2] There are three obvious advantages to this procedure over the practice of placing the packages of slides in a container already filled with the diluted stain. It allows (1) more accurate timing of the staining, (2) less handling of the individual slide packages, and (3) careful regulation of the level to which the stain rises within the container. Because of these advantages, this method is frequently employed by diagnostic aboratories.

[3] The authors acknowledge the technical assistance of Mrs. Ursula Benefield, Mrs. Kitty Pelham, Mrs. Frances Bartlett, and Mrs. Margaret Warren, and the cooperation of the Malaria Investigations Offices of the National Institute of Health in Columbia, S. C., and Milledgeville, Ga., where all of the malarial blood films used in this study were obtained.

Examination of Positive Films.—The term "positive films" refers to blood films obtained from malarial therapy patients or, in one experiment, from a pigeon. After staining, the positive films were examined with the aid of a 12-power hand lens for evidence of loss of blood from the thick films. It has not been possible to ascertain definitely how the blood is lost from the thick films. The only visible loss of blood resulted, apparently, from what we have termed "flaking." In such instances, there were areas void of blood, within the limits of the thick blood films, thus exposing the surfaces of the glass slides. The complete loss of a thick blood film did not occur in any of the experiments. Usually, the flaking did not extend more than one or two millimeters inward from the margins of the thick films. The greatest amount of flaking observed constituted a ring approximately two millimeters in width extending completely around the thick film just within the outer margin. In grading the extent of flaking (table 1) this amount was given a value of 4 plus. If the ring of flaking extended three-fourths of the way around the film, it was graded 3 plus; half-way around, 2 plus; and any flaking one-quarter of a ring or less, 1 plus.

TABLE 1.—*The relative amount of flaking (visible blood loss) which occurred in the thick blood films prepared with avian blood (Experiment 1), with human malarial blood (Experiments 2, 3, and 4), and with human blood collected on two surveys (Survey 1 and 2)*

	Degree of flaking [1]				
	−	+	++	+++	++++
Experiment 1	2	23	10	9	6
Experiment 2	24	42	23	10	1
Experiment 3	36	28	17	10	8
Experiment 4	1	33	34	19	12
Survey 1	12	40	18	16	14
Survey 2	0	0	3	26	71

[1] See text for method of grading.

Experimental Data

Experiment 1.—In staining groups of human thick blood films, it is possible to observe the transfer of cells from one film to another only when these cells happen to contain malarial parasites. Inasmuch as it was desirable to know the extent of transfer of blood between slides, an experiment was designed utilizing avian blood. Since avian erythrocytes are nucleated, their presence on a human blood film can readily be detected. In the thick films thus contaminated, only the nuclei can be seen due to the hemolysis of the avian cells during the staining process. The avian erythrocytic nuclei can

easily be distinguished from the usual constituents of human thick blood films.

Fifty combination thick-and-thin films were prepared with human blood. Fifty similar preparations were made with blood from the leg vein of a pigeon. The slides were arranged in packages so that those bearing normal (human) blood films were alternated with slides bearing avian blood films. In preparing each package, all of the slides were faced to the left except the slide to the extreme left which was turned so the film side faced to the right, or inward (fig. 1 A). This practice prevents any film of the group from being exposed to scratching or scraping off during the staining procedure (*6*). The four

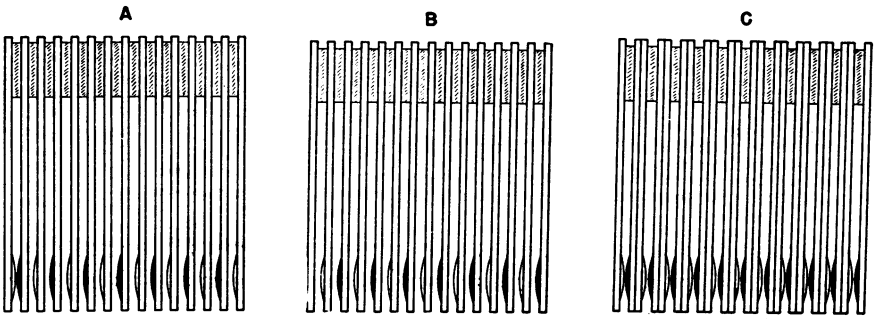

FIGURE 1—Arrangement of slides in packages with positive slides shaded.
A. End film turned inward for protection. One normal film opposes a positive film. Experiments 1, 2, 3, and 4.
B. Blank slide at end for protection. No opposing films.
C. Slides back to back. All films opposing. Experiments 6 and 7.

packages were stained, as outlined above, together in a single glass dish. After they were stained, 48 of the 50 avian films showed evidence of blood loss (table 1). In most instances the amount of visible flaking was minute and in no case did it constitute more than a tenth of the film. In examining the human thick blood films, only contaminated areas with four or more avian erythrocytic nuclei were considered.

Results.—Thirty-five of the 50 human slides had avian erythrocytic nuclei within the thick blood films (table 2). In all, 48 of the 50 human slides were contaminated within or adjacent to the thick blood films. From 1 to over 25 areas of contamination were found on the human thick blood films; the number of avian erythrocytic nuclei in each area varied from 4 to over 100. All of the human slides examined on the reverse side were heavily contaminated with avian erythrocytic nuclei.

Experiment 2.—Having demonstrated with avian blood that transfer of portions of the thick film from one slide to another may occur during this mass staining procedure, this and the next two experiments were

TABLE 2.—*Percentage transfer of malarial parasites or avian erythrocytes to normal thick blood films during mass staining procedures*

Experiment	Slide arrangement in packages [1]	Parasitemia (No. parsites per cu. mm. of blood)	Number of slides		Percent normal thick films contaminated with transferred parasites
			Positive	Normal	
1	A	([2])	[2] 50	50	[3] 70
2	A	[4] 7, 500	100	100	8
3	A	[4] 7, 600	100	100	7
4	A	[5] 1, 000, 000	100	100	80
5A	B	[5] 136, 000	50	50	24
5B	B	[5] 136, 000	50	50	48
6A	C	[5] 136, 000	50	50	74
6B	C	[5] 136, 000	50	50	60
7A	C	[5] 136, 000	50	50	90
7B	C	[5] 136, 000	50	50	94
8	([6])	[5] 136, 000	122	100	41

[1] See figure 1.
[2] Avian blood.
[3] Percentage human blood films with transferred avian erythrocytes.
[4] *Plasmodium malariae.*
[5] *Plasmodium falciparum* and rare *Plasmodium malariae.*
[6] Only positive or normal slides in a given package; a blank slide at the end for protection.

performed to determine if a similar transfer occurs when normal thick blood films are stained together with thick films containing human malarial parasites.

One hundred positive slides were made from a malarial patient infected with *Plasmodium malariae.* On the day the films were prepared, the patient had a parasite count of 7,500 per cu. mm. These slides were wrapped along with 100 normal slides in the same manner as outlined in experiment 1 (fig. 1 A) and were stained in the usual way.

After staining, 24 of the positive thick films showed no visible loss of blood, while 76 showed varying degrees of flaking ranging from scarcely any to approximately one-eighth of the entire film (table 1).

Results.—P. malariae parasites were found on eight of the normal thick films (table 2); on another slide, a single parasite was found just outside the thick film. Young and old trophozoites and presegmenters were observed. One thick film had 14 parasites, another had 2, and the remaining 6 had 1 parasite each.

Experiment 3.—One hundred positive slides were prepared from a malarial patient infected with *P. falciparum.* At that time, the patient had a count of 7,600 parasites per cu. mm. These positive slides together with 100 normal slides were wrapped as in figure 1 A and stained.

Upon examination of the positive slides for evidence of flaking, it was found that 36 of the thick films showed no visible loss of blood while the remaining 64 showed varying degrees of flaking (table 1). Although on one positive slide about one-half of the thick film was

lost, in most instances the flaking did not exceed approximately one-tenth of the film.

Results.—Examination of the normal thick films revealed that seven were contaminated with rings of *P. falciparum* (table 2). One of the contaminated films contained 63 parasites; a second had 4 parasites; a third had 2; the other 4 contained 1 parasite each.

Experiment 4.—One hundred positive slides were prepared from a moribund malarial patient infected with *P. falciparum*. The parasite count of the patient on that day was nearly 1,000,000 per cu. mm. On the same day, 100 normal slides were made. The 2 groups were stained together as in the previous experiments (fig. 1 A).

All but one of the positive thick films showed evidence of flaking (table 1). There was considerably more flaking of these films than was observed in Experiments 2 and 3. In some instances, as much as a third of the film was lost.

Results.—*P. falciparum* parasites were found on 80 of the normal thick films (table 2); on 6 others, parasites were found just outside the thick films. Thirteen normal slides showed no parasites within or adjacent to the thick films, but incomplete examinations revealed from 1 to 200 parasites in the thin films. On only one slide were no parasites observed. Twenty-two of the 80 contaminated thick films showed 1 to 10 parasites; 11 films, from 11 to 20 parasites; 13 films, from 21 to 50 parasites; 13 films, from 50 to 100 parasites; 11 films, from 100 to 200 parasites; 6 films, from 200 to 500 parasites; and 4 films, over 500 parasites. On some contaminated slides, parasites were scattered generally over the entire thick film and both rings and gametocytes were observed. Several of the normal slides were examined on the reverse surface from the blood films and in every instance parasites were observed.

Experiment 5.—Because of the manner in which the slides were wrapped in Experiments 2, 3, and 4, one normal film in each package directly opposed the neighboring parasitized film (fig. 1 A). In the analysis of the results of these experiments, it was noted that the normal opposing films showed a significantly higher rate of contamination than the normal nonopposing films in the same experiments (50 and 30 percent, respectively). Since in these experiments the number of opposing films (24) was low compared to the number of nonopposing films (276), experiments 5 and 6 were designed to test this point further. In the first, the slides were so arranged that there were no opposing films; in the second, each normal film directly opposed a positive film. In both experiments, the blood used for the positive slides was obtained from a single patient with a mixed infection of malaria. On the day the blood was taken, there was a count of approximately 136,000 parasites per cu. mm. There were many

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rings of *P. falciparum* and rare *P. malariae* organisms in various stages. Normal thick films were prepared as in previous experiments.

One hundred normal and 100 positive films were wrapped together in packages of 25 slides each in such a way that no normal film directly opposed a positive film. This necessitated the use of a blank slide at one end of each package for protection of the exposed film instead of turning the end film inward (fig. 1 B). In order to obtain duplicate observations, the packages were divided into two sets, A and B, of four packages each. The two sets were stained separately in glass dishes.

Results.—Malarial parasites were observed in 24 and 48 percent, respectively, of the normal blood films in sets A and B (table 2).

Experiment 6.—One hundred normal and 100 positive films were wrapped together in packages of 25 slides each in such a way that each normal film directly opposed a positive film (fig. 1 C). As in Experiment 5, these packages were divided into two sets, A and B, which were stained separately.

Results.—Examination revealed parasites on 74 and 60 percent, respectively, of the normal films in sets A and B (table 2).

Experiment 7.—In order more thoroughly to lake old thick films which have been allowed to dry for a week or more, dehemoglobinization prior to staining is sometimes used. The present experiment was designed to test the effect of this procedure upon the rate of transfer of parasites.

One hundred normal and 100 positive films (prepared at the same time and from the same patient as those in Experiments 5 and 6) were wrapped together in packages of 25 slides each so that every normal film directly opposed a positive film (fig. 1 C). These slides were divided into two sets, A and B. Prior to staining, the dried films were dehemoglobinized with buffered water (pH 7.2) for 10 minutes. Following dehemoglobinzation, the slides were rinsed with buffered water, each set was placed in a dish, and staining was performed as described previously.

Results.—A high rate of transfer occurred in both sets. Ninety percent of the normal films in set A and 94 percent of the normal films in set B showed parasites (table 2). These results should be compared with those obtained in Experiment 6 in which the experimental set-up was the same but without prior dehemoglobinization.

Experiment 8.—It has been pointed out in the original observations leading to the present study that there must have been transfer of malarial parasites from one package of slides to another during the staining procedure since all of the blood films in a single package had been prepared from a given individual. Although all of the above experiments demonstrated that transfer of blood occurs between

slides during mass staining procedures, they have not indicated whether the transferred parasites came from slides within the same package or from another package. The following experiment was designed to test the possibility of transfer between slides located in different packages.

One hundred twenty-two thick blood films were made from the same patient and on the same occasion as those used in the preceding three experiments. One hundred normal thick blood films were prepared at the same time. These slides were wrapped in packages with all of the blood films facing in the same direction and a blank slide placed at the end of each package for protection. Four packages contained only parasitized blood films while three contained the normal blood films. Each package contained from 31 to 34 slides. The packages were placed side by side in a rectangular staining dish alternating the two types of bloods so that a positive package was at each end of the dish (fig. 2). The slides were then stained in the manner described previously.

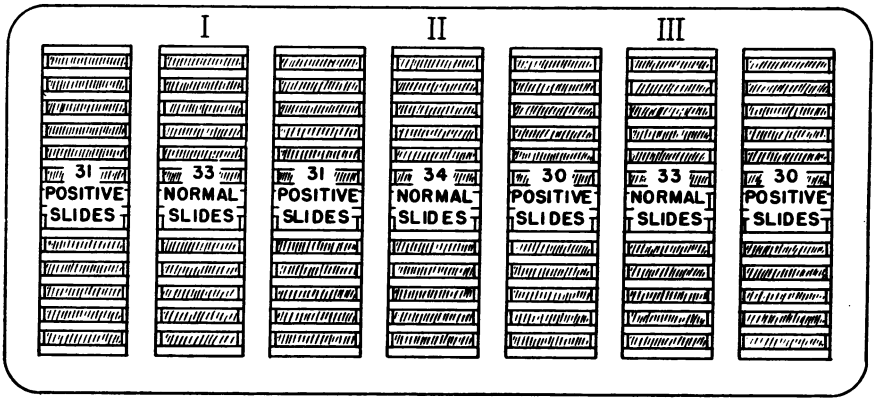

FIGURE 2—Top view of staining dish showing orientation of packages of slides in Experiment 8.

Results.—Forty-one percent of the normal blood films contained malarial parasites (table 2). The three packages of normal blood films (I, II, and III in fig. 2) had 20, 11, and 14 films, respectively, that showed transferred parasites.

Discussion

After Ross developed a thick blood film technique for the diagnosis of malaria, a number of the early investigators pointed out that loss of blood may occur during staining (*7*, *8*, *9*). However, in a rather comprehensive survey of the literature, no reports have been found regarding the ultimate fate of those portions of the thick films that become separated from the slide. Furthermore, there is no indication

in the literature that the problem has been studied, and if such transfer of cells to other films has been observed, no mention of it has been encountered. From the above experiments it is evident that when thick blood films are stained en masse according to the procedure tested, there is a possibility that blood of thick films may flake or slough off and adhere to other films stained in the same dish.

In all eight of the above experiments transfer of blood occurred between slides stained together by a mass procedure. In the seven experiments utilizing human parasitized blood, the rate of transfer to normal thick films ranged from 7 to 94 percent (table 2). The actual transfer of blood cells between slides was undoubtedly even greater since only those cells which were parasitized could be recognized during the examination of the normal thick films. That more transfer of blood actually occurred was apparent with the finding of blood elements and parasites on areas of the slides other than the thick films and, in many cases, on the reverse sides of the slides. This fact is also evident from the high rate of transfer observed in Experiment 1 in which transferred avian blood on the human blood films could be recognized by the erythrocytic nuclei.

The occurrence of lower rates of transfer in Experiment 5 with no opposing films than in Experiment 6 with all opposing films indicates that the normal thick films which directly oppose parasitized films are more likely to receive transferred parasites. The construction of some staining dishes and racks permits the placing of slides back-to-back so that each of the specimens to be stained directly opposes another. In view of the increased possibility of contamination under such circumstances, the authors believe the use of this type of equipment is not advisable for staining malarial blood films. Furthermore, in wrapping together groups of slides according to the Barber-Komp technique, the use of a blank slide at the end of the package to protect an otherwise exposed film is recommended instead of turning the last slide, film inward.

Although it is advisable to stain thick blood films within 3 days after their preparation, it is sometimes necessary for a laboratory to stain films which are much older. Because old films are sometimes inadequately laked by the staining solution, frequently they are dehemoglobinized prior to staining by being placed in buffered water for 10 minutes. The results obtained in Experiment 7 indicate that prior dehemoglobinization may increase the possibility of transfer of parasites, since a higher rate of contamination was obtained and more visible loss of blood was observed than in the comparable Experiment 6. However, it should be pointed out that the films used in Experiment 7 had dried for only 48 hours and, therefore, may not have demonstrated what actually would have occurred had they been old films.

The fact that blood can be transferred from one package to another during mass staining procedures was demonstrated by the original observations which led to the present study. This was confirmed by the results of Experiment 8. The rate of transfer is comparable to the rates obtained in Experiment 5 in which the experimental set-up was similar, except that the positive and normal blood films were alternated within individual packages.

In our experiments the only visible loss of blood from the positive films resulted from what we have considered as flaking. The amount and extent of the flaking varied in the different experiments. Many factors probably contribute to this loss of blood; for instance, the cleanliness of the slide, the thickness of the film, and the length of time the film is dried. In these experiments the films were dried for 24 to 48 hours, generally considered a satisfactory length of time. They were also of recommended thickness or a little thinner than the usual survey films. As mentioned in the section on Methods, we feel confident that the slides were adequately cleaned.

In order to ascertain if the amount of flaking observed in our experiments was unduly great as compared to survey slides, 200 survey slides, chosen at random, were examined for evidence of loss of blood. One group of 100 survey slides was cleaned in a State health laboratory. After the preparation of the blood films, they were sent to our laboratory for staining and examination. Table 1 shows that as much flaking occurred in these slides (survey 1) as in the slides of Experiment 4, which exhibited the greatest amount in the present series. Another group of 100 slides was obtained from a 1943 survey in one of the Southern States. These slides were cleaned and stained by another laboratory according to recommended procedures. At least twice as much flaking was observed in these slides (survey 2, table 1) as occurred in Experiment 4. These comparisons would indicate that blood loss may be a common occurrence in thick blood films obtained on surveys and stained by mass procedures.

Various precautions have been suggested by early investigators to reduce the loss from thick blood films during staining. In the same year that Ross advocated this type of film, Ruge (*7*) offered a modification of the technique specifically to prevent loss of part or all of the thick films from the slide. He combined fixation with dehemoglobinization by immersing the dried film for a few minutes in a formalin-acetic acid mixture before staining with a "diluted Manson's solution, or according to Romanovsky." James (*8*) objected to Ross' original method because of the loss of the blood films and introduced a modification utilizing acidified ethyl alcohol as a dehemoglobinizing and fixing agent. Taylor (*9*) cautioned against certain washing procedures in a modified James' method, because of "a danger of

washing cells off." Barber and Komp (*5*) also noted loss of the blood film in their staining technique and insisted upon absolutely clean slides as a preventive.

Inasmuch as the loss of blood from thick films during staining procedures is apparently difficult to prevent, the practical significance of the transfer of malarial parasites to otherwise normal blood films will depend, to a great extent, upon the ability of the technician to recognize transferred parasites as contaminants. During the first examination of the normal films in the experiments reported in this paper, there were very few instances where it was possible to ascertain that the parasites were associated with flakes that had transferred from other films. A definite search for flakes was made with both the dissecting and the compound microscopes in 100 areas containing transferred parasites on 67 slides. In none of these areas was it possible to differentiate visible flakes of donor blood associated with the contaminating parasites.

These observations, together with the frequent occurrence of individual parasites on blank areas of the slide, unassociated with any blood elements, indicate either that individual blood cells with their parasites must slough off the surface of thick films or, if portions of the films do come off as flakes, these may disintegrate before contaminating other films. This lack of association of contaminating parasites with visible flakes is extremely important in regard to the ability of technicians to recognize the false positives. In the present series of experiments it was not at all evident in the greatest number of cases that the parasites had come from other films. The transposed parasites and the blood elements of the recipient film occupied the same focal plane and in most instances the parasites appeared to be completely free of accompanying blood elements.

In further consideration of this point the authors were interested to know if technicians without previous knowledge of the present experiments would recognize the experimental slides as false positives. For this purpose, 17 normal films with from 1 to over 500 contaminating *P. falciparum* parasites were used. These slides were given to six technicians currently responsible for the diagnosis of malaria at separate laboratories in the United States. They were told only that the blood films had been stained by a mass procedure used in malarial surveys and were asked to examine the slides and indicate whether or not a positive diagnosis of malaria would be reported on each slide. In every instance when parasites were encountered, a positive report was recorded by all six technicians. In their remarks concerning the diagnoses no reference was made to any unusual appearance observed in the parasites. In order to make certain that this point was not overlooked, each person was questioned individually concerning the

appearances of the parasites after the reports had been handed in. None of the technicians had noted any peculiarities and all were startled to learn that the parasites had transferred from other blood films and that each of the slides was a false positive.

Assuming from this experience that technicians may not be able to recognize transferred parasites as contaminants, the probability of the occurrence of such false positives will vary with both the incidence of infection and the level of parasitemia in the population surveyed. The incidence of infection will determine the number of true positives from which blood loss may occur and the level of parasitemia will determine the probability that the transferred blood will contain malarial parasites. In surveys the importance of false positives may depend upon the number of individuals examined. With large numbers of slides, the presence of a few false positives would probably not change significantly the observed rate of infection; in small surveys, relatively few false positives might change markedly the observed rate.[4]

In areas of the world where malaria is highly endemic, false positives resulting from transferred blood conceivably may give an erroneous picture of the incidence. Furthermore, in research studies and clinical laboratories in which blood films with high levels of parasitemia may be stained together with normal films, contamination may be an important source of error.

Since survey slides are given only a routine examination limited to a certain number of microscopic fields or minutes, the possibility of encountering transferred parasites on normal blood films is of course considerably reduced. This possibility will be related to the number of contaminated areas and parasites on the normal films. In Experiments 2, 3, and 4, the complete examination of the normal films revealed that the extent of contamination ranged from one parasite well over 500 parasites, distributed over the entire film. Of course, there is no way of predicting whether or not the microscopist will find the transferred parasites in a routine examination. For example, there were instances when the technicians examining these experimental slides happened to focus down directly upon the only contaminated areas on the films; in other cases, parasites were not found until the film had been almost completely examined.

Regardless of the staining technique employed, it is generally accepted that the examination of thick blood films is not an infallible technique for diagnosing malaria, although it is probably the best laboratory method now available. Factors that will influence the recognition of positive films include the staining solutions, lighting

[4] The latter has been demonstrated by this laboratory in a controlled field survey conducted in Puerto Rico, the results of which will be presented in a later paper.

and optical equipment, the training and experience of the technicians, and the amount of time devoted to the examination of each film.

The present study indicates another factor that should be added to the list of influencing circumstances when mass staining techniques are used. When a number of slides are stained together in a dish, the possibility exists that malarial parasites will transfer from parasitized films to normal films and that the transposed parasites cannot be distinguished in all or even many instances from other parasites.

Nevertheless, in spite of these limitations, there will always be occasions when expediency will dictate the necessity of utilizing the most practical method of handling large numbers of slides. Under these circumstances, until improvements in techniques are developed, the present methods of mass staining may have to be employed in surveys. When this is necessary, it must be realized that the results obtained may be inaccurate due to the errors resulting from the transfer of parasites as well as from the other technical defects referred to above. Furthermore, in experimental research on malaria and in the diagnosis of a suspected malarial patient, it would seem to be extremely important to stain slides individually and thus eliminate false positives resulting from transferred parasites.

Summary

Eight experiments are reported in which it is demonstrated that blood can transfer between slides during mass staining procedures. The extent of this transfer and some of the factors involved are discussed. If the blood which transfers to normal films happens to contain malarial parasites, falsely positive reports of malaria may result in survey, clinical, and research examinations.

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Combined Typhus-Malaria Control Residual Spray Operations With Five Percent DDT Emulsion[1]

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Crews employed to apply DDT dust in murine typhus control operations follow a normal procedure of placing liberal patches of the insecticide along exposed rat runways. When this procedure is followed in homes, housewives frequently express dissatisfaction at having the unsightly patches of white powder on their floors and often refuse to allow them to remain long enough to be fully effective in controlling rat fleas.

As no such serious objection has arisen to residual spraying with a water emulsion of DDT, xylene, *Triton X–155*[2] for the control of malaria mosquitoes, a study was initiated in March 1947 to test the feasibility of employing routine residual spraying with DDT for the control of rat ectoparasites. Further, it was desired to determine if residual spraying for malaria control and dusting for murine typhus control might be modified and combined into one operation in the interests of economy and efficiency. After a preliminary survey was made to find a suitable area for the work, Lowndes County, Ga., was selected because it was one of the few remaining areas in the Southeast where DDT had not been used either for malaria or typhus control.

Procedure

Extensive trapping in the county indicated that the largest rat infestations occurred in rural areas, that the rat population consisted primarily of the species *Rattus rattus*, and that the ectoparasites normally associated with rats were present in significant numbers. Of the farm premises trapped, 40 having the largest rat populations were chosen for the study. From this group there were selected, by the use of random numbers, 25 premises to be treated, and 15 to remain as untreated checks. It was planned to conduct the study throughout the total effective duration of a single treatment and to determine the ectoparasite indices[3] on rats taken before treatment, immediately after treatment and at approximately 5-week intervals thereafter. Ectoparasites were collected from the rats by a process

[1] From the Communicable Disease Center, Technical Development Division (Savannah, Ga.), Bureau of State Services.

[2] A product of Rohm and Haas Co., Philadelphia, Pa. The trade names are carried as a means of identifying products under discussion, and do not represent endorsement of the products by the Public Health Service.

[3] The term "index" as used in this paper refers to the mean number of ectoparasites per rat.

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of combing and beating as described by Ludwig and Nicholson (*1*). Only live rats were considered in determining the ectoparasite indices.

A power sprayer and Loftstrand [4] air-pressure hand sprayers were used for the application of the insecticide which was a water emulsion of DDT, xylene, *Triton X–155* concentrate containing 35 percent DDT. Dilution of the above concentrate was at the rate of 1 to 6 to form a 5 percent DDT emulsion. An attempt was made to apply this emulsion to wall surfaces and floors at the rate of 4 ml. or 200 mg. of DDT per square foot. The entrances to rat burrows and harborages were treated more heavily, almost to the point of saturation, as far into these places as conditions would permit. A nozzle producing a fan-shaped spray was used at all times.

At each premise the dwelling was sprayed in the same manner as DDT is usually applied for adult mosquito control, i. e., to the walls and ceilings of each room. In addition a two-foot strip of floor adjacent to the walls was treated. The spray was directed into and around rat holes and all enclosed spaces opening into the living quarters which seemed likely to provide rat harborage. The attic was also treated.

Outbuildings showing any evidence of rat infestation were sprayed, care being taken to avoid contamination of stored animal feed. These buildings were treated in a similar manner to the homes with emphasis placed on spraying rat harborage and rat runways on overhead beams as well as those on floors. Whenever possible the entire floor area of outbuildings was sprayed. Burrows under buildings were searched for and treated.

The types of outbuildings to which the insecticide was most frequently applied were feed houses, barns, tobacco-curing sheds, stables, and granaries. On the average, 3.9 man-hours of labor and 0.3 gallon of emulsion concentrate were required for each premise.

Pretreatment trapping on the 40 premises was done approximately 12 days prior to the mean date of treatment which was April 5. Live rats taken from the check farms at that time numbered 79; while 129 were taken from farms later to be treated. Fleas were not abundant. The mouse flea, *Leptopsylla segnis* (Schönherr), was predominant, being most numerous in March and April. *Xenopsylla cheopis* (Rothschild), the oriental rat flea, was scarce but increased sharply during the late summer. Mites, especially *Liponyssus bacoti* (Hirst), the tropical rat mite, were numerous.[4] The rat louse, *Polyplax spinulosa* (Burmeister), was common although not abundant. The average dates [5] of trapping and the numbers of rats caught in this and subsequent trapping periods are shown in table 1.

[4] A product of the Loftstrand Co., Rockville, Md.

[5] These dates are weighted average dates of trapping determined by multiplying each day of the month by the number of live rats caught on that date. The results are added and the total divided by the total number of live rats caught. The nearest whole number is the average trapping date for the period.

TABLE 1.—*Results of DDT residual spraying for rat flea control, Mar. 19–Oct. 10, 1947* [1]

Group	Number of days after treatment	Mean trapping date	Number of live rats examined	*Xenopsylla cheopis*		*Leptopsylla segnis*		Total fleas [2]	
				Number	Index	Number	Index	Number	Index
Treated	Pretreatment	Mar. 25	129	267	2.1	1,244	9.6	1,518	11.8
Untreated		Mar. 24	79	14	0.2	365	4.6	382	4.8
Treated	7–15	Apr. 16	67	2	0.03	18	0.3	22	0.3
Untreated		Apr. 17	46	17	0.4	358	7.8	389	8.4
Treated	41–50	May 22	65	1	0.02	1	0.02	67	1.0
Untreated		May 22	42	74	1.8	70	1.7	157	3.7
Treated	76–86	June 25	44	5	0.1	1	0.02	9	0.2
Untreated		June 26	39	135	3.5	19	0.5	303	7.8
Treated	121–127	Aug. 6	42	0	0.0	0	0.0	42	1.0
Untreated		Aug. 6	23	251	10.9	8	0.3	266	11.6
Treated	181–189	Oct. 9	47	19	0.4	14	0.3	38	0.8
Untreated		Oct. 8	29	11	0.4	15	0.5	29	1.0

[1] The mean date of treatment was April 5, 1947.
[2] Other species of fleas encountered were primarily *Echidnophaga gallinacea* (Westwood), and occasionally *Rhopalopsyllus gwyni* C. Fox.

Results

The degree of control attained against rat fleas in this field investigation by the use of 5 percent DDT in an emulsion was very satisfactory, both initially and over an extended period of at least 4 months. The mouse flea, *Leptopsylla segnis*, which was near the expected seasonal peak of abundance at the time of treatment (figure 1), was quickly reduced from an average of 9.6 fleas per rat to 0.3 flea per rat. On the other hand, an initially small number of oriental rat fleas, *Xenopsylla cheopis*, was prevented from developing significantly on the treated premises as compared with the normal midsummer development which occurred on check farms (figure 2). Data for the entire season are presented in table 1.

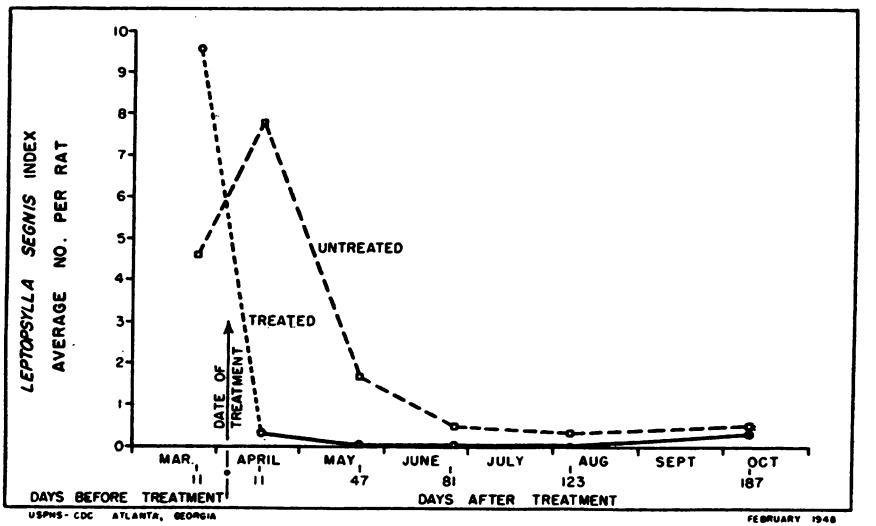

Figure 1. Control of *Leptopsylla segnis*, the mouse flea, with 5 percent DDT emulsion applied as a residual spray on rural premises.

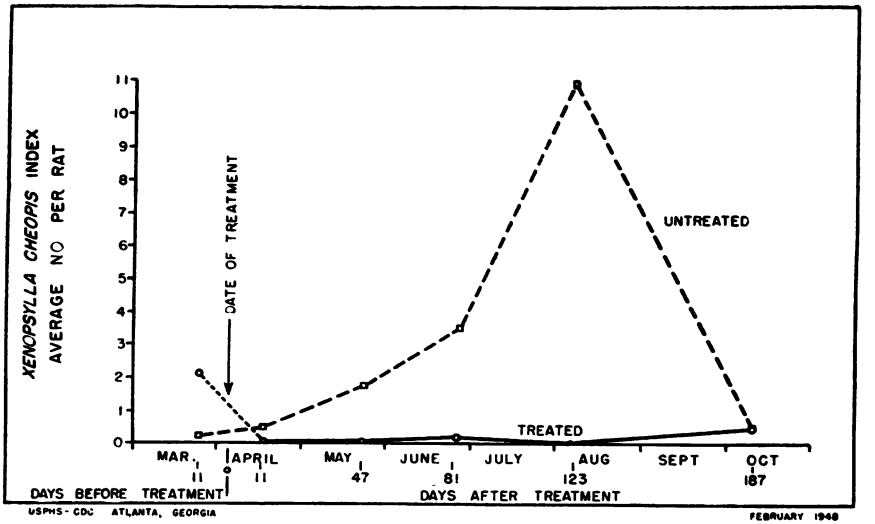

Figure 2. Control of *Xenopsylla cheopis*, the oriental rat flea, with 5 percent DDT emulsion applied as a residual spray on rural premises.

During the course of the study a total of four species of fleas was encountered. In addition to the two species previously discussed, *Echidnophaga gallinacea* (Westwood), the sticktight flea, and *Rhopalopsyllus gwyni* C. Fox were present, but were of minor importance. The former species constituted 9.2 percent of the total fleas taken, and the latter species, 0.5 percent. Figure 3 illustrates the over-all effectiveness of the control achieved against these four species of fleas in comparison with the indices from untreated premises. Two peaks occur on the curve representing the flea indices from untreated

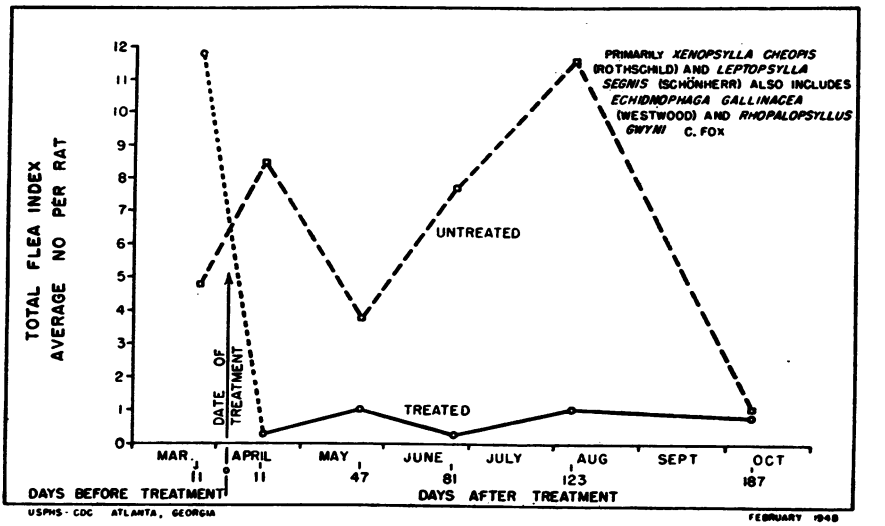

Figure 3. Control of rat fleas with 5 percent DDT emulsion applied as a residual spray on rural premises.

farms. The first of these peaks is due primarily to the normal seasonal abundance of *Leptopsylla segnis*, while the mid-summer peak is due primarily to the presence of *Xenopsylla cheopis*.

A second method of indicating the results of treatment is the comparison of the percentages of rats from check premises bearing one or more fleas with those from treated farms. The results thus obtained are illustrated in figure 4. Prior to spraying, the percentages of rats bearing fleas were high from both the group of farms selected as checks and those to be treated—58 and 84 percent, respectively. Following application of the insecticide, the percentage from

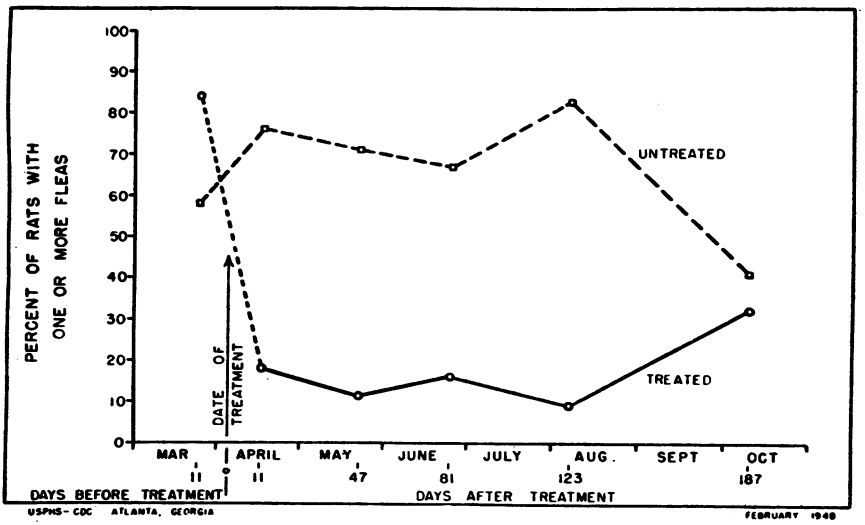

Figure 4. Results of application of 5 percent DDT emulsion to rural premises expressed in percent of rats bearing fleas.

treated farms decreased drastically; while a slight increase occurred in the untreated group. Thereafter, for a period of approximately 4 months, the difference remained roughly the same, indicating continued satisfactory control of rat fleas. However, between the sampling period in August and that in October, 4 to 6 months following treatment, the percentages of rats bearing fleas converged sharply, thus indicating the end of effective control.

Of importance in this field investigation was the determination of the effect of a residual application of 5 percent DDT emulsion on rat ectoparasitic species other than fleas. A total of 19,806 mites was combed from the rats during the course of the study, 98.7 percent of these being the tropical rat mite, *Liponyssus bacoti* (Hirst). Other mite species encountered were *Atricholaelaps glasgowi* (Ewing), *Echinolaelaps echidninus* (Berlese), *Atricholaelaps* sp., *Cheyletus* sp., *Uropoda* sp., and *Cosmolaelaps* sp. Only one species of rat louse, *Polyplax spinulosa* (Burmeister), was encountered. This insect and

the mite, *L. bacoti*, were common throughout the duration of the study. Data pertinent to the seasonal abundance of these species and the effect of the treatment are presented in table 2.

TABLE 2.—*Results of DDT residual spraying on rat ectoparasites other than fleas, Mar. 19–Oct. 10, 1947* [1]

Group	Number of days after treatment	Mean trapping date	Number of live rats examined	*Liponyssus bacoti*			All mites [2]		*Polyplax spinulosa*	
				Range per rat	Number	Index	Number	Index	Number	Index
Treated	Pretreatment.	Mar. 25	129	0–708	4,161	32.2	4,179	32.4	2,124	16.5
Untreated		Mar. 24	79	0–908	2,140	27.1	2,146	27.2	1,294	16.4
Treated	7–15	Apr. 16	67	0–468	2,026	30.2	2,044	30.5	424	6.3
Untreated		Apr. 17	46	0–641	4,346	94.5	4,384	95.3	511	11.1
Treated	41–50	May 22	65	0–532	1,315	20.2	1,323	20.4	523	8.0
Untreated		May 22	42	1–703	3,459	82.4	3,476	82.8	276	6.6
Treated	76–86	June 25	44	0–47	174	3.95	176	4.0	483	11.0
Untreated		June 26	39	0–248	1,186	30.4	1,231	31.6	258	6.6
Treated	121–127	Aug. 6	42	0–10	14	0.3	28	0.7	405	9.6
Untreated		Aug. 6	23	0–205	518	22.5	523	22.9	195	8.5
Treated	181–189	Oct. 9	47	0–65	133	2.8	157	3.3	721	15.3
Untreated		Oct. 8	29	0–42	80	2.8	139	4.8	362	12.5

[1] The mean date of treatment was April 5, 1947.

[2] A minor proportion, 1.3 percent, of the total number of mites taken from the rats were of the following: *Atricholaelaps glasgowi* (Ewing), *Echinolaelaps echidninus* (Berlese), *Atricholaelaps* sp., *Cheyletus* sp., *Uropoda* sp., *Cosmolaelaps* sp., and unidentified specimens including immature and damaged individuals.

The differences in *L. bacoti* index trends from treated and check premises are presented in figure 5, and a comparison is made in figure 6 on the basis of the percent of rats found bearing one or more mites of this species. It is evident that partial control, at least initially, resulted from the application of 5 percent DDT as a residual spray. This control was apparently sufficient to prevent the sharp increase in the rate of infestation of rats which occurred on the un-

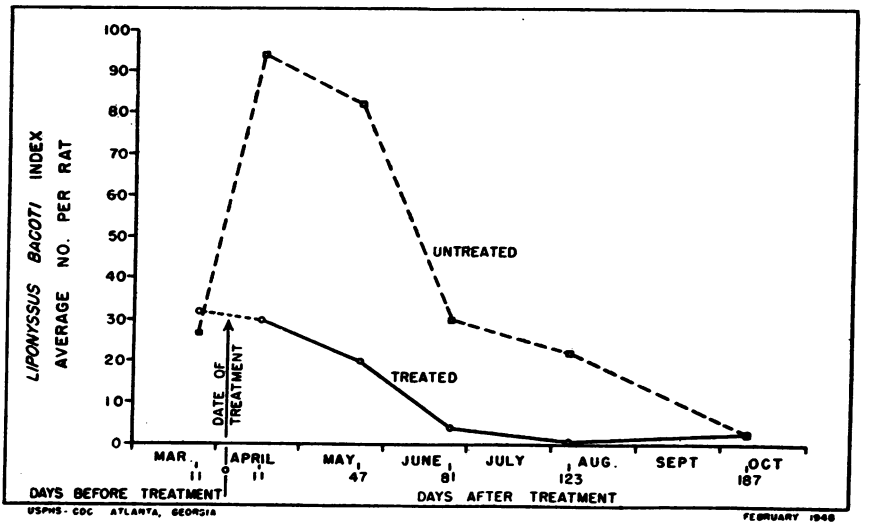

Figure 5. Control of *Liponyssus bacoti*, the tropical rat mite, with 5 percent DDT emulsion applied as a residual spray on rural premises.

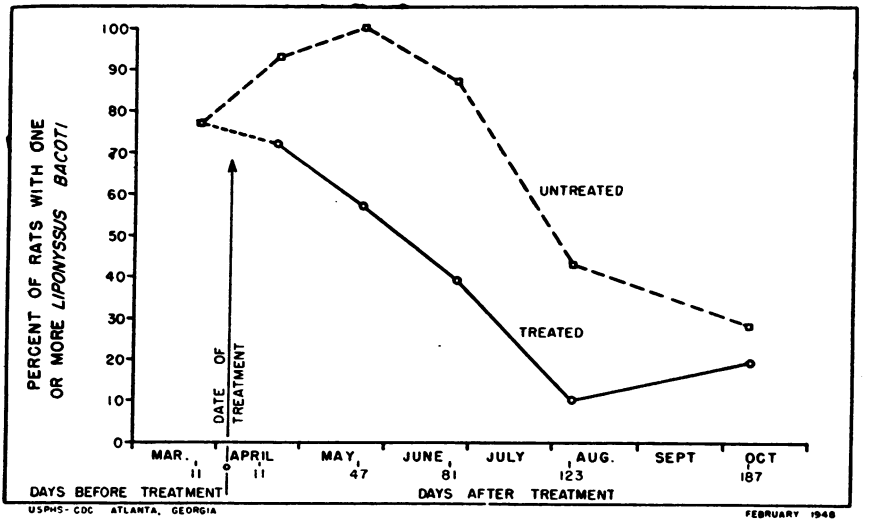

Figure 6. Results of application of 5 percent DDT emulsion to rural premises expressed in percent of rats bearing the tropical rat mite, *Liponyssus bacoti.*

treated premises. The duration of effective control could not be determined with certainty because of the seasonal reduction in numbers which this species normally exhibits during the summer. Following spraying, however, the index in treated premises failed to rise again and did not approach that from the check group of farms until 4 to 6 months later.

No significant effect on *Polyplax spinulosa*, the rat louse, resulted from the treatment. A graph comparing treated with untreated premises is given in figure 7.

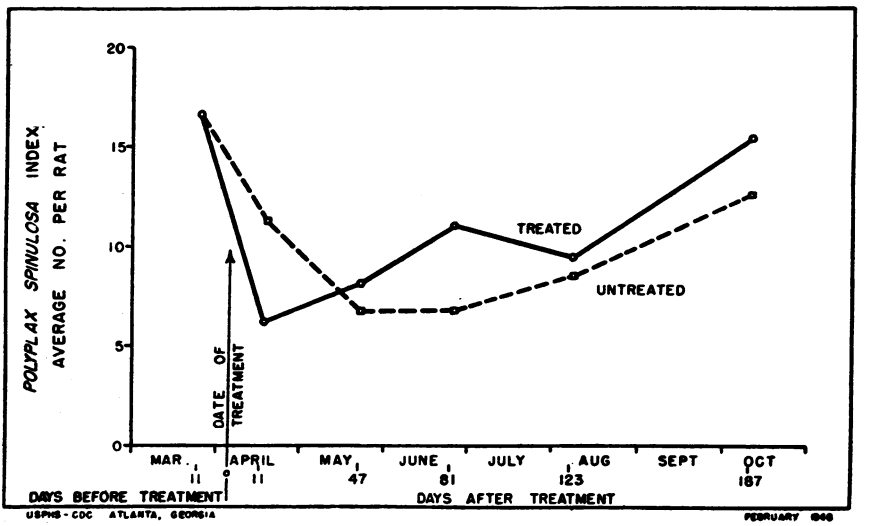

Figure 7. Effect of 5 percent DDT emulsion applied as a residual spray to rural premises upon the rat louse, *Polyplax spinulosa.*

Discussion

From the data gathered during this field investigation in which 25 farm premises were treated, it is apparent that 5 percent DDT applied as a residual spray produced satisfactory control of the oriental rat flea, *Xenopsylla cheopis*, and the mouse flea, *Leptopsylla segnis*, for a minimum period of 4 months. These were the only two species of fleas present in significant numbers. It was demonstrated (fig. 1) that an initially high infestation of *L. segnis* could be reduced to a satisfactorily low level by a single application of the insecticide made early in the season. It was further demonstrated (fig. 2) that this single early application was sufficient to prevent the development of an infestation of *X. cheopis* of significant size several months later when this species is normally expected to increase greatly in abundance.

The tropical rat mite, *Liponyssus bacoti*, was affected to the degree that normal infestation development was arrested (figs. 5 and 6) at a time when rapid increase normally was to be expected. It is believed by the investigators that the climbing habits of this species may have an important bearing on the results achieved with this method of DDT application since the insecticide was present on vertical surfaces where it could be contacted by climbing mites. This success against *L. bacoti* suggests the possibility that DDT residual spray might also be of some value in controlling the rickettsialpox mite, *Allodermanyssus sanguineus* (Hirst), a species said to have similar climbing habits.

In spite of the fact that an emulsion of 5 percent DDT, under the conditions of this field investigation, gave satisfactory immediate and residual control of rat fleas, it does not always appear practical from the standpoint of labor costs to employ the technique of residual spraying alone for the control of rat ectoparasites. In the above described investigation an average of 3.9 man-hours of labor was required to spray the home and rat-infested outbuildings of each farm. This figure does not compare favorably with the average of 0.96 of a man-hour established for spraying homes in the Extended Malaria Control Program of the Public Health Service in 1945 (*2*), nor with the average of 0.26 of a man-hour of labor required to dust the average premises in the Murine Typhus Control Program in Georgia (*3*).

It is believed that the use of a combination dusting-spraying technique is the logical procedure from both the standpoint of economy of application and that of achieving the desired results. Spray could be applied in situations where dust would be obnoxious, particularly in homes. In the attics of most homes, however, and in most outbuildings the use of dust would be permissible and desirable since it would be quicker and easier to apply. For the treatment of rat

harborage, such as enclosed spaces between walls in the homes and elsewhere, and in rat burrows, dust would be preferable since better dispersion can be obtained with it under these conditions than with spray.

Controlled field experiments conducted at this laboratory have indicated that it is necessary to apply the DDT spray directly to rat harborage before consistently good control of *X. cheopis* can be expected. Some reported failures to obtain satisfactory control by spraying may be due to under-emphasis of this factor.

Summary

A field investigation is reported employing a water emulsion of DDT (5 percent), xylene, *Triton X–155* as a residual spray applied at the approximate rate of 200 mg. of DDT per square foot of surface resulting in the attainment of successful control of certain rat ectoparasites.

It is suggested that a combination DDT residual spraying and dusting technique for the control of rat ectoparasites may be practical not only from the standpoint of efficiency in operation but also from the viewpoint of securing cooperation from the housewife. In the event of the combination of such applications for the control of rat ectoparasites and mosquitoes by the same crew, a 5 percent DDT spray would be applied to the walls and ceilings of the rooms of homes and to a narrow strip of floor adjacent to the walls as well as to all other places where dust would be unsightly. Attics and enclosed spaces between walls, particularly those into which rat holes lead, would be treated with DDT dust. All outbuildings would be treated with dust except those where dust would be objectionable, or a hazard to livestock.

ACKNOWLEDGMENT

The writers wish to express their appreciation to Dr. S. W. Simmons, Chief of the Technical Development Division, and to Dr. C. M. Tarzwell, Head of the Rodent and Ectoparasite Control Branch, T. D. D., for their aid in planning this field investigation; and to Roy J. Boston, Director, Typhus Control Service, Division of Preventable Diseases, Georgia Department of Public Health, and Dr. G. T. Crozier, Commissioner of Health, Lowndes County, Ga. and his staff for their cooperation in facilitating the field work.

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(*1*) Ludwig, Russel G. and Nicholson, H. Page: The control of rat ectoparasites with DDT. Pub. Health Rep. **62**: 1 (1947).
(*2*) Annual Report, Malaria Control in War Areas, 1945–46. Federal Security Agency, Public Health Service.
(*3*) Personal correspondence, Roy J. Boston, Director Typhus Control Service, Division of Preventable Diseases, Georgia Department of Public Health.

INCIDENCE OF DISEASE

No health department, State or local, can effectively prevent or control disease without knowledge of when, where, and under what conditions cases are occurring

UNITED STATES

REPORTS FROM STATES FOR WEEK ENDED JULY 10, 1948

Summary

Beginning with this week the list of diseases to be reported weekly by the State health officers by telegraph has been reduced from 20 to 15 diseases, which are as follows: Anthrax, diphtheria, infectious encephalitis, influenza, meningococcal meningitis, pneumonia, poliomyelitis, rabies in animals, Rocky Mountain spotted fever, scarlet fever, smallpox, tularemia, typhoid fever, paratyphoid fever, and whooping cough. Of these diseases, current data for 10, included in the following table, are to be published in PUBLIC HEALTH REPORTS. Data for all 15, together with comparable figures for the preceding year and the 5-year medians will be included in the Health Officers' Weekly Statement, a duplicated report issued weekly. After this week the weekly city reports will no longer be published in the Public Health Reports but will be issued as a part of the Health Officers' Weekly Statement. (For revision of morbidity reports see PUBLIC HEALTH REPORTS for June 18, 1948, pp. 822–23.)

The incidence of poliomyelitis increased from 362 cases reported last week to 513 for the current week, as compared with 311 for the corresponding week of 1946 and a 5-year median of 245. The increase is accounted for chiefly in the reports of 6 States, as follows (last week's figures in parentheses): North Carolina 131 (92), California 92 (74), Texas 89 (67), Oklahoma 21 (3), Iowa 19 (0), and Virginia 17 (2). Since March 20, the approximate average date of seasonal low incidence, 2,533 cases have been reported. as compared with 1,696 for the corresponding period of 1946 and a 5-year median of 1,027.

Other morbidity reports for the week show no significant changes.

Deaths recorded during the week in 93 large cities in the United States totaled 8,453, as compared with 8,924 last week, 8,915 and 8,770, respectively, for the corresponding weeks of 1947 and 1946, and a 3-year (1945–47) median of 8,770. The total for the year to date is 267,925, as compared with 267,608 for the corresponding period last year. Infant deaths totaled 610, as compared with 641 last week and a 3-year median of 742. The cumulative figure is 18,868, as compared with 21,372 for the same period last year.

Telegraphic case reports from State health officers for week ended July 10, 1948

[Leaders indicate that no cases were reported]

Division and State	Diphtheria	Encephalitis, infectious	Measles	Meningitis, meningococcus	Poliomyelitis	Rocky Mt. spotted fever	Scarlet fever	Tularemia	Typhoid; paratyphoid fever	Whooping cough
NEW ENGLAND										
Maine			45	1			6		1	13
New Hampshire			15				2			4
Vermont			20		1		4			14
Massachusetts	5	1	872	1	2		93		4	28
Rhode Island			8				1			5
Connecticut		1	69	1	2		3			2
MIDDLE ATLANTIC										
New York	7		1,283	1	11	2	[b] 52		2	67
New Jersey	3		1,192	2	[1] 4	1	16			26
Pennsylvania	4		638	2	5		70		6	58
EAST NORTH CENTRAL										
Ohio	1		272		15	1	79		1	40
Indiana	7	1	[1] 78		3		11		3	12
Illinois	2		136	1	12	2	19		2	23
Michigan [a]	3		648	1	1		24			27
Wisconsin			969	1	4		19	1	1	50
WEST NORTH CENTRAL										
Minnesota			40				11			3
Iowa			39	1	19		6			11
Missouri	4		64	2	2	1	[b] 5	2	1	7
North Dakota			10							5
South Dakota		3	4		4					
Nebraska			35		11		7		1	2
Kansas	4		11		6		3			8
SOUTH ATLANTIC										
Delaware			9		5					1
Maryland [a]	5		417			3	[b] 8			17
District of Columbia			38	1	3				2	4
Virginia	4		146	2	17	3	3	1	2	52
West Virginia	1		2	3		1	[b] 7		6	23
North Carolina	3		26	2	130	3	13		1	54
South Carolina	1		58	1	4			1	1	78
Georgia	1		13		5	1	7	1	10	6
Florida	2		52	2	4		5			11
EAST SOUTH CENTRAL										
Kentucky	3		49		5		2		4	6
Tennessee	2	1	31		6		9	1	2	38
Alabama	4		6	2	3	1	9		4	42
Mississippi [a]	1		5	2	3		4		5	3
WEST SOUTH CENTRAL										
Arkansas	1		28	3	[1] 4		1	14	5	25
Louisiana			[1] 6	1	7				2	2
Oklahoma			[1] 33	1	21		7	1	3	33
Texas	13		380	2	89		11	2	16	160
MOUNTAIN										
Montana			8				5			2
Idaho		1	29		2		[b] 1			4
Wyoming			11		1		1			2
Colorado	3		341		2		15		1	32
New Mexico	1		16		2		2			10
Arizona			70		3		3			7
Utah [a]	1		147				1	1		12
Nevada			1							1
PACIFIC										
Washington	4		179		1		13		1	8
Oregon			206			1	4			38
California	6		863	3	92		46		3	31
Total	96	8	9,618	39	511	20	608	25	90	1,107
Median, 1943–47	138	9	4,299	109	245	18	964	18	129	2,351
Year to date, 27 weeks	4,610	239	526,914	1,957	[1] 2,879	217	53,569	539	1,560	51,223
Median, 1943–47	6,297	243	517,735	5,528	1,329	193	93,132	499	1,871	67,443
	(27th)		(35th)	(37th)	(11th)		(32d)		(11th)	(39th)
Seasonal low week ends	July 10		Sept. 4	Sept. 18	Mar. 20		Aug. 14		Mar. 20	Oct. 2
Since seasonal low week	10,968		561,860	2,739	[1] 2,533		76,108		1,087	82,489
Median, 1943–47	15,307		555,748	7,980	1,027		131,453		1,247	89,490

[a] Period ended earlier than Saturday.

[b] Including cases reported as streptococcal infections and septic sore throat.

[c] Including cases reported separately as paratyphoid fever and salmonella infection, as follows: Massachuestts (salmonella infection), 2; Indiana 1; Virginia 1; West Virginia 1; Georgia 7; Arkansas 2; Colorado 1; Washington 1; California 2.

[1] Delayed report (included in cumulative totals only): Poliomyelitis, Arkansas, week ended June 12, 1 case. Territory of Hawaii: Rabies 0; measles 3; meningitis 1; typhoid fever 1; whooping cough 9. Week ended July 3, additional, 1 case endemic typhus fever.

WEEKLY REPORTS FROM CITIES*

City reports for week ended July 3, 1948

This table lists the reports from 88 cities of more than 10,000 population distributed throughout the United States, and represents a cross section of the current urban incidence of the diseases included in the table.

Division, State, and City	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
NEW ENGLAND												
Maine:												
Portland	0	0	------	0	-------	0	0	0	1	0	0	------
New Hampshire:												
Concord	0	0	------	0	11	0	1	0	0	0	0	------
Vermont:												
Barre	0	0	------	------	------	0	0	0	0	0	0	------
Massachusetts:												
Boston	6	0	------	0	116	2	7	0	63	0	0	6
Fall River	0	0	------	0	90	0	1	0	0	0	0	3
Springfield	0	0	------	------	20	0	------	0	1	0	0	------
Worcester	0	0	------	------	48	0	------	0	12	0	0	2
Rhode Island:												
Providence	0	0	------	0	13	0	1	0	2	0	0	------
Connecticut:												
Bridgeport	0	0	------	------	1	0	------	0	2	0	0	------
Hartford	0	0	------	0	11	0	1	0	1	0	0	------
New Haven	0	0	------	0	8	0	3	0	2	0	0	------
MIDDLE ATLANTIC												
New York:												
Buffalo	0	0	------	0	60	0	3	0	2	0	0	1
New York	7	0	3	0	419	4	40	7	46	0	1	26
Rochester	0	0	------	0	4	0	2	0	0	0	0	1
Syracuse	0	0	------	0	24	0	1	0	0	0	0	17
New Jersey:												
Camden	0	0	------	------	0	0	------	0	2	0	0	------
Newark	0	0	------	0	281	1	3	1	4	0	0	3
Trenton	0	0	------	------	4	0	------	0	0	0	0	------
Pennsylvania:												
Philadelphia	3	0	------	0	228	0	11	1	9	0	2	8
Pittsburgh	0	1	------	0	6	0	7	0	23	0	0	5
Reading	0	0	------	0	1	0	1	0	1	0	0	------
EAST NORTH CENTRAL												
Ohio:												
Cincinnati	0	0	------	0	40	0	8	0	6	0	0	1
Cleveland	0	0	------	------	33	0	------	10	20	0	0	1
Columbus	0	0	------	------	1	0	------	0	2	0	0	3
Indiana:												
Fort Wayne	0	(1)	(1)	------	2	0	------	0	0	0	1	------
Indianapolis	1	0	------	------	24	0	------	1	3	0	0	6
South Bend	0	0	------	------	1	0	------	0	0	0	0	------
Terre Haute	0	0	------	0	-------	0	1	0	0	0	0	------
Illinois:												
Chicago	0	0	1	------	72	2	------	0	21	0	0	9
Michigan:												
Detroit	1	5	------	0	427	1	10	1	20	0	0	13
Flint	0	0	------	------	25	0	------	0	1	0	0	------
Grand Rapids	0	0	------	------	2	0	------	0	0	0	0	8
Wisconsin:												
Kenosha	0	0	------	------	12	0	------	0	0	0	0	------
Milwaukee	0	0	------	0	358	0	3	0	6	0	0	3
Racine	0	0	------	0	19	0	0	0	3	0	0	4
Superior	0	0	------	------	8	0	------	0	0	0	0	------
WEST NORTH CENTRAL												
Minnesota:												
Duluth	0	0	------	1	13	0	0	0	1	0	0	------
Minneapolis	0	0	------	0	5	0	2	0	5	0	0	------
St. Paul	0	0	------	0	2	0	3	0	1	0	0	1
Missouri:												
Kansas City	0	0	1	------	22	0	------	1	0	0	0	------
St. Joseph	0	0	------	0	1	0	0	0	0	0	0	------
St. Louis	0	0	------	------	6	0	------	0	0	0	0	2

* In some instances the figures include nonresident cases.
[1] Diseases not reportable.

City reports for week ended July 3, 1948—Continued

Division, State, and City	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
WEST NORTH CENTRAL—continued												
North Dakota:												
Fargo	0	0	------	0	1	0	0	0	1	0	0	------
Nebraska:												
Omaha	0	0	------	0	------	0	0	4	0	0	0	------
Kansas:												
Topeka	0	0	------	0	2	0	0	0	1	0	0	2
Wichita	0	0	------	------	2	0	------	2	2	0	0	2
SOUTH ATLANTIC												
Deleware:												
Wilmington	0	0	------	0	5	1	0	3	0	0	0	------
Maryland:												
Baltimore	0	0	1	------	477	0	------	0	4	0	0	4
Cumberland	1	0	------	0	------	0	0	0	1	0	0	------
Frederick	0	0	------	0	------	0	0	0	0	0	0	------
District of Columbia:												
Washington	0	0	------	0	43	0	5	0	4	0	0	1
Virginia:												
Lynchburg	0	0	------	0	3	0	0	0	0	0	0	------
Richmond	0	0	------	0	3	0	3	0	0	0	0	1
Roanoke	0	0	------	0	------	0	0	0	0	0	0	------
West Virginia:												
Charleston	0	0	------	------	------	0	------	0	0	0	0	------
Wheeling	0	0	------	0	------	0	1	0	0	0	0	------
North Carolina:												
Raleigh	0	0	------	0	2	0	------	0	0	0	0	------
Winston Salem	1	0	------	0	------	0	0	3	1	0	0	4
South Carolina:												
Charleston	0	0	------	0	------	0	0	0	0	0	0	4
Georgia:												
Atlanta	0	0	------	0	1	0	3	0	3	0	0	1
Brunswick	0	0	------	0	------	0	0	0	0	0	0	------
Savannah	0	0	------	------	------	1	------	0	0	0	0	------
Florida:												
Tampa	1	-	------	0	1	0	3	0	2	0	0	2
EAST SOUTH CENTRAL												
Tennessee:												
Memphis	0	0	------	0	10	0	8	1	1	0	0	6
Nashville	0	0	------	------	------	0	------	0	0	0	0	2
Alabama:												
Birmingham	0	0	------	------	3	0	------	0	2	0	0	2
Mobile	0	0	------	0	2	1	2	0	4	0	0	------
WEST SOUTH CENTRAL												
Arkansas:												
Little Rock	0	0	------	------	12	0	------	0	1	0	0	3
Louisiana:												
New Orleans	1	0	1	1	4	0	4	0	0	0	0	4
Shreveport	0	0	------	0	------	0	3	3	0	0	0	------
Oklahoma:												
Oklahoma City	0	0	------	0	2	0	0	0	0	0	0	1
Texas:												
Dallas	3	0	------	0	5	1	2	1	0	0	1	1
Galveston	0	0	------	0	1	0	2	6	0	0	0	------
Houston	0	0	------	0	------	0	1	9	1	0	0	------
MOUNTAIN												
Montana:												
Billings	0	0	------	0	------	0	5	0	0	0	0	------
Great Falls	0	0	------	0	3	0	------	0	2	0	0	------
Helena	0	0	------	0	------	0	0	0	0	0	0	------
Missoula	0	0	------	0	2	0	0	0	0	0	0	2
Idaho:												
Boise	0	0	------	------	0	0	------	0	0	0	0	------
Colorado:												
Denver	1	0	------	------	17	0	------	1	5	0	0	5
Pueblo	0	0	------	0	129	0	0	0	0	0	0	------
Utah:												
Salt Lake City	0	0	------	------	65	0	------	0	0	0	0	1

City reports for week ended July 3, 1948—Continued

Division, State, and City	Diphtheria cases	Encephalitis, infectious, cases	Influenza		Measles cases	Meningitis, meningococcus, cases	Pneumonia deaths	Poliomyelitis cases	Scarlet fever cases	Smallpox cases	Typhoid and paratyphoid fever cases	Whooping cough cases
			Cases	Deaths								
PACIFIC												
Washington:												
Seattle	0	0	------	0	94	0	3	1	0	0	0	------
Spokane	0	0	------	0	17	0	3	1	1	0	0	------
Tacoma	0	0	------	------	2	0	------	0	0	0	0	------
California:												
Los Angeles	4	0	4	0	203	0	5	18	7	0	0	5
Sacramento	0	0	------	0	6	0	1	0	1	0	0	------
San Francisco	1	0	1	0	36	0	3	0	1	0	0	1
Total	31	6	12	2	3,571	14	167	75	305	0	5	177
Corresponding week, 1947 [1]	53	------	27	1	1,345	------	197	------	296	0	10	973
Average 1943–47 [1]	48	------	23	[2] 8	[3] 1,513	------	[2] 228	------	452	0	16	791

[1] Exclusive of Oklahoma City. [2] 3-year average, 1945–47. [3] 5-year median, 1943–47.

Rates (annual basis) per 100,000 population, by geographic groups, for the 88 cities in the preceding table (latest available estimated population 34,236,800)

	Diphtheria case rates	Encephalitis, infectious, case rates	Influenza		Measles case rates	Meningitis, meningococcus, case rates	Pneumonia death rates [2]	Poliomyelitis case rates	Scarlet fever case rates	Smallpox case rates	Typhoid and paratyphoid fever case rates	Whooping cough case rates
			Case rates	Death rates [4]								
New England	15.7	0.0	0.0	------	831	5.2	------	0.0	220	0.0	0.0	29
Middle Atlantic	4.6	0.5	1.4	------	475	2.3	------	4.2	40	0.0	1.4	28
East North Central	1.2	3.1	0.6	------	628	1.8	------	7.4	50	0.0	0.6	29
West North Central	0.0	0.0	2.0	------	107	0.0	------	13.9	22	0.0	0.0	14
South Atlantic	5.1	0.0	1.7	------	917	3.4	------	10.3	26	0.0	0.0	29
East South Central	0.0	0.0	0.0	------	89	5.9	------	5.9	41	0.0	0.0	59
West South Central	11.7	0.0	2.9	------	70	2.9	------	55.6	6	0.0	2.9	26
Mountain	7.9	0.0	0.0	------	1,716	0.0	------	7.9	56	0.0	0.0	64
Pacific	7.9	0.0	7.9	------	566	0.0	------	31.6	16	0.0	0.0	9
Total	4.7	0.9	1.8	------	545	2.1	------	11.5	47	0.0	0.8	27

[4] Rate not computed as mortality data were incomplete.

Anthrax.—Cases: Philadelphia 1.
Dysentery, amebic.—Cases: New York 9; Winston-Salem 1; Memphis 13; New Orleans 4; Los Angeles 4.
Dysentery, bacillary.—Cases: Charleston, S. C. 2; Memphis 1; Los Angeles 1.
Tularemia.—Cases: Nashville 1; New Orleans 1.
Typhus fever, endemic.—Cases: Dallas 1.

DEATHS DURING WEEK ENDED JULY 3, 1948

[From the Weekly Mortality Index, issued by the National Office of Vital Statistics]

	Week ended July 3, 1948	Corresponding week, 1947
Data for 93 large cities of the United States:		
Total deaths	8,924	8,053
Median for 3 prior years	8,053	------
Total deaths, first 27 weeks of year	259,469	258,693
Deaths under 1 year of age	641	627
Median for 3 prior years	626	------
Deaths under 1 year of age, first 27 weeks of year	18,258	20,630
Data from industrial insurance companies:		
Policies in force	71,015,454	67,256,797
Number of death claims	11,615	9,442
Death claims per 1,000 policies, in force annual rate	8.6	7.3
Death claims per 1,000 policies, first 27 weeks of year, annual rate	9.8	9.7

PLAGUE INFECTION IN CALIFORNIA AND NEW MEXICO

Plague infection has been reported proved in fleas from rodents collected in California and New Mexico as follows:

CALIFORNIA

Kern County.—A pool of 400 fleas from 50 ground squirrels, *Citellus beecheyi*, taken June 10 on a ranch ½ to 2 miles east of Castair Lake.

NEW MEXICO

Rio Arriba County.—Reported under date of July 6, a pool of 78 fleas from 38 prairie dogs, *Cynomys gunnisoni gunnisoni*, shot on U. S. Government land 1 mile east of Dulce on State Highway No. 17.

FOREIGN REPORTS

CANADA

Provinces—Communicable diseases—Week ended June 19, 1948.—During the week ended June 19, 1948, cases of certain communicable diseases were reported by the Dominion Bureau of Statistics of Canada as follows:

Disease	Prince Edward Island	Nova Scotia	New Brunswick	Quebec	Ontario	Manitoba	Saskatchewan	Alberta	British Columbia	Total
Chickenpox		34		190	561	69	50	34	85	1,023
Diphtheria		1		5		1			2	9
Dysentery, bacillary				1		1				2
German measles		1		38	13			4	11	67
Influenza		23							1	24
Measles			26	433	1,189	32	2	101	92	1,875
Meningitis, meningococcus				4					1	5
Mumps		5		167	185	33	59	40	6	495
Poliomyelitis				4	5				1	10
Scarlet fever		3		75	86	5	1	5		175
Tuberculosis (all forms)		6	4	117	34	12	26	7	27	233
Typhoid and paratyphoid fever			1	14	1			2	1	19
Undulant fever				2	1	1		3	3	10
Venereal diseases:										
Gonorrhea		5	7	100	85	34	22	43	64	360
Syphilis		6	7	79	40	8	6	9	19	174
Other forms									1	1
Whooping cough		62		48	13	2	3	14	3	145

WORLD DISTRIBUTION OF CHOLERA, PLAGUE, SMALLPOX, TYPHUS FEVER, AND YELLOW FEVER

From consular reports, international health organizations, medical officers of the Public Health Service, and other sources. The reports contained in the following tables must not be considered as complete or final as regards either the list of countries included or the figures for the particular countries for which reports are given.

CHOLERA

(Cases)

NOTE.—Since many of the figures in the following tables are from weekly reports, the accumulated totals are for approximate dates.

Place	January–April 1948	May 1948	June 1948—week ended—			
			5	12	19	26
AFRICA						
Egypt	1					
Cairo	1					
ASIA						
Burma	2	16		1	1	
Akyab		2		1	1	
Rangoon		2				
India	37,891	20,450	2,899	2,007		
Ahmadabad	2					
Alleppey	1					
Bombay		[2] 1	[2] 2	[2] 1	[2] 1	
Calcutta [1]	3,532	1,913	273	176	162	168
Cawnpore	22	9	5	19	10	11
Cocanada	2					
Colachel	12					
Cuddalore	12					
Jodhpur			[2] 1			
Kilakarai	21					
Lucknow	12	6	1	2		
Madras	29	14	2	4		10
Nagpur	4					
Negapatam	16					
New Delhi	3	11				
Raj Samand			[3] 5	1		
Tuticorin	16					
Vizagapatam	1					
India (French):						
Chandernagor	21					
Karikal	300					
Pondicherry	59					
Indochina (French):						
Cambodia	972	74	29	33	40	
Cochinchina	444	98	6	11		
Bien Hoa		1				
Chaudoc	2					
Cholon	21	8				
Giadinh	20	3				
Longxuyen	7					
Mytho	34	13		5		
Rachgia	124	8				
Saigon	80	46	1	5		
Laos	[4] 12					
Tonkin	1					
Pakistan	15,379	6,064		1		
Chittagong	30	3		1		
Karachi		2				
Lahore	[5] 1,357	[5] 2,128				
Siam	36	2		2		
Syria	3					

[1] Including imported cases.
[2] Imported.
[3] May 25–June 7.
[4] Deaths.
[5] Lahore City and District.

PLAGUE

(Cases)

Place	January–April 1948	May 1948	June 1948—week ended—			
			5	12	19	26
AFRICA						
Belgian Congo	4	6				4
British East Africa:						
Kenya	16					
Tanganyika	240	30				
Madagascar	188	11		[1] 2		
Tamatave		1				
Tananarive	16	9		[1] 2		
Rhodesia, Northern	26					
Union of South Africa	[2] 36					
ASIA						
Burma	[3] 452	20		3		
Mandalay	16	1				
Rangoon	[3] 12	2		1		
China:						
Chekiang Province	11	14				
Wenchow	5	3				
Fukien Province	61	75	2			
Kiangsi Province	16	1		[1] 1		
Kwangtung Province	64	24				
Yunnan Province	65					
India	19,096	653	6	16		
Indochina (French):						
Annam	135					
Cochinchina	40					
Laos	2					
Java	4					
Pakistan		11				
Siam	102		1			
EUROPE						
Portugal: Azores	8					
SOUTH AMERICA						
Argentina:						
Buenos Aires Province	9					
Ecuador	22					
Chimborazo Province	1					
Loja Province	4	10				
Peru:						
Cajamarca Department	8					
Huacho Department	1					
Libertad Department	1					
Lima Department	5					
Venezuela:						
Aragua State		7				
OCEANIA						
Hawaii Territory: Plague-infected rats [4]	5					

[1] June 1–10, 1948.
[2] Includes 4 cases of pneumonic plague.
[3] Includes imported cases.
[4] Plague infection was also reported in Hawaii Territory, under date of February 27, 1948, in a mass inoculation of tissue from 19 rats.

SMALLPOX

(Cases)

(P=present)

Place	January-April 1948	May 1948	June 1948—week ended—			
			5	12	19	26
AFRICA						
Algeria	152	21				
Angola	111	5				
Basutoland	3					
Belgian Congo [1]	920	146	49			
British East Africa:						
Kenya	85	2				
Nyasaland	1,527	533	172	111	74	
Tanganyika	597	42	6			
Uganda	154	26				
Cameroon (French)	3					
Dahomey	197	43			[2] 8	[3] 23
Egypt [4]	336	83	10			3
Eritrea	9					
French Equatorial Africa	10	3				
French Guinea	115	8				
French West Africa: Haute-Volta	385	24		[5] 8		
Gambia	24	3				
Gold Coast	677	133	51			
Ivory Coast	314	176		[5] 3	[2] 19	
Libya	229	25	1			
Mauritania	1					
Morocco (French)	20	8		[5] 4		
Mozambique	24	10				
Nigeria	2,800	133				
Niger Territory	245	48		[5] 31		
Rhodesia:						
Northern	[6] 103	[7] 230				
Southern	383					
Senegal	3	4				
Sierra Leone	125	11				
Sudan (Anglo-Egyptian) [1]	436	462	83	129	52	24
Sudan (French)	16					
Swaziland	4					
Togo (British)	9					
Togo (French)	67	1		[5] 4	[2] 8	
Tunisia	497	10				
Union of South Africa	22	P				
ASIA						
Arabia	[3] 5	2				
British North Borneo	1					
Burma [3]	1,889	393	12	1	13	3
Ceylon [3]	13	2				
China	2,797	522	[8] 50	[8] 32	[8] 31	[8] 28
India	37,000	10,330	1,025	686	[9] 31	[9] 19
India (French)	5	1				
Indochina (French)	1,904	393	170	38	21	
Iran	424	39	4	5		
Iraq	562	82	16	6	2	16
Japan	14	6	1			
Lebanon	57					
Malay States (Federated)	344	40	5	5		
Manchuria	42					
Pakistan	9,317	388	(9)	[9] 4	[9] 3	[9] 9
Palestine	8					
Siam	434	17	4			
Straits Settlements	3	6				
Syria	32	3	2	2	9	
Trans-Jordan	13					
EUROPE						
France	3					
Germany	3					
Portugal	67		1			
Spain	18	1				
Canary Islands	9					
NORTH AMERICA						
Guatemala	2					
Mexico	402	18	5	6	8	6

See footnotes at end of table.

SMALLPOX—Continued

Place	January–April 1948	May 1948	June 1948—week ended— 5	12	19	26
SOUTH AMERICA						
Argentina	9					
Bolivia	31					
Brazil	16					
Chile	4	1				
Colombia	3,216	710	20	18	15	
Ecuador [1]	1,722	285			[10] 6	
Paraguay [1]	412					
Peru	117					
Trinidad		[11] 8				
Venezuela [1]	2,313	211		23	4	2

[1] Includes alastrim.
[2] June 11–20, 1948.
[3] June 21–30, 1948.
[4] Includes imported cases.
[5] June 1–10, 1948.
[6] January–March, 1948.
[7] Mar. 1–May 31, 1948.
[8] Shanghai only.
[9] In ports only.
[10] In Guayaquil only.
[11] Alastrim.

TYPHUS FEVER*

[Cases]

(P = Present)

AFRICA						
Algeria	123	5				
Basutoland	6					
Belgian Congo	102	18	6			
British East Africa:						
Kenya [1]	23	6	2			
Egypt	163	86	1	3	4	
Eritrea	20	6		7		
Gold Coast	2					
Libya	177	137	10	4	23	12
Morocco (French)	59					[2] 2
Morocco (International Zone)		2				
Morocco (Spanish) [1]	2					
Mozambique		2				
Nigeria [1]	3					
Rhodesia (Southern)	1					
Senegal	[3] 1					
Sierra Leone	[3] 5	[3] 1				
Somalia	1					
Tunisia [1]	401	112		[4] 13		
Union of South Africa [1]	179	P	P	P	P	P
ASIA						
Burma	5					
China [1]	56	27	1	[4] 2	1	
Indochina (French) [1]	16	5		5	2	3
Iran [1]	71	16	6	6		
Iraq	79	37	15	5	9	3
Japan	393	44	5	29	15	
Java	3					
Manchuria	25	12				
Pakistan		22				
Palestine [1]	12					
Philippine Islands	[3] 1					
Straits Settlements [1]	6	4				
Syria [1]	15	26				1
Trans-Jordan	30	12	1	3		2
Turkey (see Turkey in Europe).						
EUROPE						
Albania	15					
Bulgaria	544	54	19	12		
Czechoslovakia	6					
France	1			[4] 1		
Germany	5	6		2		
Great Britain:						
England:						
London					[3 5] 1	
Island of Malta [3]	8					

See footnotes at end of table.

TYPHUS FEVER*—Continued

Place	January-April 1948	May 1948	June 1948—week ended 5	12	19	26
EUROPE—continued						
Greece [1] [6]	55	8	1	3	1	4
Hungary	[7] 37	5	1		2	
Italy [1]	42	44		[4] 8		
Sicily	2					
Netherlands	[3] 1					
Poland	154	54				
Portugal—Madeira Islands:						
Funchal	1					
Rumania [1]	20, 421	797	119			
Spain	2	1	1	4		
Turkey	198	26	10	11	5	5
Yugoslavia	391	93	15	8	12	
NORTH AMERICA						
Costa Rica [3]	2					
Cuba [3]	9					
Guatemala	52					
Jamaica [3]	5	2				
Mexico [1]	441	[8] 18	[8] 6	[8] 5	[8] 5	[8] 10
Panama Canal Zone [1]	3					
Puerto Rico [3]	10	9	3		2	
SOUTH AMERICA						
Bolivia	[9] 52	41				
Brazil	71	13	3	3	3	
Chile [1]	[10] 75	[11] 87	[12] 2	[12] 1	[12] 1	
Colombia	1, 112	270				
Curaçao [3]	11	1				
Ecuador [1]	170	56	[13] 3		[13] 3	
Venezuela	52	22			[14] 1	
OCEANIA						
Australia [3]	122	32	9	4		
Hawaii Territory	5					
New Caledonia	1					

*Reports from some areas are probably murine type, while others probably include both murine and louse-born types.

[1] Includes murine type.
[2] June 1–20, 1948.
[3] Murine type.
[4] June 1–10, 1948.
[5] Imported, in crew member of ship from Hong Kong.
[6] Includes suspected cases.
[7] Corrected figure.
[8] In sea- and air ports only.
[9] Includes 9 deaths reported as cases in Cochabamba Department in March 1948.
[10] Jan. 1–Mar. 6, 1948.
[11] Mar. 7–May 29, 1948.
[12] In Valparaiso.
[13] In Guayaquil.
[14] In Maracaibo.

YELLOW FEVER*

[D=deaths]

AFRICA		
Ivory Coast:		
Gagnao	D	1
SOUTH AMERICA		
Colombia:		
Antioquia Department	D	5
Boyaca Department	D	1
Caldas Department	D	3
Cundinamarca Department	D	7
Intendencia of Meta	D	3

*Delayed report: During the months of April and May, *1947*, 5 confirmed cases of yellow fever were reported in Bolivia, distributed as follows: Santa Cruz Department, Nuflo de Chavez 1, Concepcion 1, Cercado 1; La Paz Department, Province of Sud Yungas, Chulumani 1; Province of Nor Yungas, Coroico 1.

EXAMINATION FOR REGULAR CORPS

A competitive examination for appointment in the Regular Corps of the Public Health Service in the grade of assistant surgeon (first lieutenant) and senior assistant surgeon (captain) will be held in October. The written examination will be conducted October 4, 5, and 6 at places convenient to the candidates. The oral examination will be held at various points throughout the country.

All applicants must be at least 21 years of age and citizens of the United States, must present a diploma of graduation from a recognized medical school and satisfactorily pass a physical examination performed by Public Health Service officers.

Physicians beginning internship on July 1, 1948, will be admitted to the examination. Successful candidates will be placed on active duty in the Regular Corps upon completion of internship on July 1, 1949.

Applicants for the grade of assistant surgeon must have had at least 7 years of educational and professional training or experience, exclusive of high school. Applicants for the grade of senior assistant surgeon must have had at least 10 years of educational and professional training or experience, exclusive of high school.

Entrance pay for an assistant surgeon with dependents is $5,011 a year and for senior assistant surgeon with dependents, $5,551 a year. This includes the additional pay of $1,200 for medical officers, as well as subsistence and rental allowance. Provisions are made for promotions at regular intervals up to and including the grade of senior surgeon (lieutenant colonel) and for selection for promotion to grade of medical director (colonel) at $9,751 a year. Retirement is authorized at either completion of 30 years' service or at the age of 64. Full medical care including disability retirement at three-fourths pay is provided.

Application forms may be obtained from Public Health Service Hospitals, District Offices or by writing to the Surgeon General, Public Health Service, Washington 25, D. C.