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FILARIASIS STUDIES IN THE REPUBLIC OF THE PHILIPPINES

by

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November 1967

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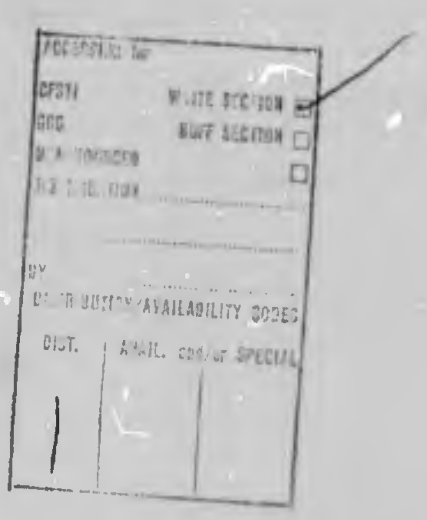

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Abstract

A total of 3,695 persons were examined for microfilaremia with 120 found positive or a prevalence rate of 3.2 per cent. Out of these 120 positives 91 or 76 per cent were W. bancrofti and 29 or 24 per cent were B. malayi filariasis. Tawi-Tawi group of islands particularly Bongao municipality became the second endemic focus for malayan filariasis in the Republic of the Philippines. The first one is in Quezon municipality, Palawan Province which we reported in 1964.

As expected the microfilaremia rates in males was higher than in females in practically all ages and in all municipalities found endemic for the disease. The higher the endemicity of filariasis in an area, the closer the prevalence rates between sexes and the lower the endemicity the higher the prevalence rates of males over females. This condition we have explained as probably due to occupational exposure risks.

Malayan filariasis in Southern Sulu appeared to have been introduced relatively quite recent as compared to that found in Palawan Province. The microfilariae exhibited the nocturnal subperiodic behavior similar to the malayan filariasis found in Palawan.

There is that trend of increasing W. bancrofti microfilaremia rates in islands with large areas of farm land planted with abaca, with Aedes (Finlaya) poecilus as the vector mosquito. However, in areas where abaca is absent filariasis may also be absent or if present the prevalence rate is low. Anopheles minimus flavirostris is the vector in abaca-free areas.

The purple sheath of W. bancrofti microfilariae cannot be attributed to the "brand" of Giemsa stain used, but rather to the solution used in dilution of stain and the washing off of excess stain.

The Log-Probit Regression Line as applied in our data is ideal in comparing intensity of infection by microfilarial counts and would be a nice tool in assessing filariasis control programs.

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INTRODUCTION

In last year's work on filariasis under contract No. 92-557-FEC-38366 the results of which I reported in Report No. J-253-2 we were able to make a relatively extensive blood survey in all the eight municipalities of Jolo island which is the largest and most thickly populated island in the Sulu Archipelago. The overall microfilaremia rate found was 11.4 per cent with higher rates among males than in females and also higher in adults compared to children. The vector was found to be Aedes (Finlaya) poecilus which breed in the collection of water in the leaf axil of abaca plants. We also found that the extent of abaca plantation in the eight municipalities of Jolo island did not show a significant statistical correlation with the filariasis prevalence although the results seemed to show some indication. Finally, examination of blood smears stained with Giemsa according to Wilson, revealed the presence of some W. bancrofti microfilariae, with their sheath stg. purple, a finding quite unusual for this species(1,2)

Sulu province is composed of several islands and islets which are of volcanic and coral origin. The archipelago is located about 118°-20' to 122° West longitude and 4°-40' to 6°-30' North latitude. There are approximately 448 islets with 92 of them larger than one square mile and are inhabited by a vivid colorful people. For convenience sake the province is divided into groups as follows: Jolo group, Pangutaran group, Samales group, Tapul group, Tawi-Tawi group and Sibutu group. The province has a mixture of inhabitants composed of the original indigenous tribes and immigrants coming from Luzon, Visayas and Mindanao. The original groups or tribes are the Tausog, Badjao and Samal, with the Tausogs predominating in Jolo, Pangutaran and Tapul groups while the Samals and Badjaos predominate in the Samales, Tawi-Tawi and Sibutu groups. These three tribes belong to the "Muslim" or Mohammedan population of the Philippines, with an estimated overall number of 1-1/2 million as of 1960 census. The muslims in general inhabit 5 provinces of Mindanao, parts of Palawan and Sulu province. Sulu province has an estimated population of 395,600 and Jolo island alone has about 197,000(3).

The principal industries are farming, fishing, pearl industry and mat weaving. Their main agricultural products are coconut, abaca, root crops, rice and corn. The climate is warm and moist and the rainfall is evenly distributed throughout the year. This province is located outside of the typhoon belt.

The following are the objectives of the present study: (I) Extent and distribution of filariasis in the Southern islands of Sulu Archipelago, (II) attempt to correlate the abaca industry in this province with filariasis prevalence; (III) attempt to determine probable explanation for the difference in staining reaction of the sheath of W. bancrofti microfilariae in Giemsa stain; (IV) attempt to determine the mosquito vector of filariasis in Southern Sulu; (V) attempt to develop serologic test for the diagnosis of filariasis.

I. Extent and distribution of filariasis in the Southern islands of Sulu Province.

Sulu archipelago consists of several islands and islets so that for convenience sake these group of islands are divided into at least six groups as seen in Figure 1. Although it was our desire to cover as many islands as possible in this survey, we were generally prohibited from doing so by the lack of commercial water transportation going to some islands, hence we were unable to include in this survey the Samales group.

Materials and Methods

Blood survey procedures:

The technics employed in the preparation of night blood smears consists of two thick blood films approximately 20 mm^3 each smear on a glass slide per subject. The blood was best and conveniently obtained from the finger. The smears were dried overnight and stained in the morning with dilute Giemsa according to the method of Wilson(1). We employed one or two methods of obtaining blood from the inhabitants depending upon the peace and order situation in the locality. Whenever possible the people were requested thru the "headman" to assemble in certain designated places not earlier than 1900 hours, otherwise we employed the house-to-house blood survey method with the help of a guide and interpreter. Stained blood smears were shipped by air to the Institute of Hygiene and examined under a compound microscope at low magnification. However when in doubt as to the species identification, I shift to high magnification, or even resort to destaining the smears and later restained with Delafield's hematoxylin. Every single blood smear was examined by the senior investigator.

In order to have an idea of the intensity of the microfilaremia, microfilarial count was done on each positive slide and analysis of these counts were done. Also we want to know if the behavior of the microfilariae in this area is any different from other endemic areas in this country and so we made some periodicity studies. Actually we were only interested in the periodicity characteristics of B. malayi microfilariae however since we encountered two mix-infections and another W. bancrofti case we also made periodicity counts on these three cases. This was done by withdrawing blood from a lancet wound on the finger by means of a calibrated pipette specially designed for this purpose. The exact amount of 20 mm^3 was then smeared on a glass slide forming two parallel linear smears of approximately 10 mm^3 per line according to the method of Sasa(4). These were dried overnight and stained with Giemsa in the morning. Blood was taken from the subjects at 2-hour intervals for a total of 24 hours.

We have followed the method used by Turner et al(5) in presenting the actual counts for each subject, which were then converted to percentages of peak counts and these percentages were averaged. These averages were then plotted on the graph.

Results

With Jolo island as the reference point, all other islands and/or municipalities covered in this survey except the municipality of Pangutaran are located south of Jolo island and hence we refer to them in this report as southern Sulu islands.

Distribution and prevalence:

For convenience we have arranged the different municipalities we have surveyed, under their respective group starting from north to south with the names of barrios under each municipality. The distribution and prevalence of filariasis by municipality is shown in Table I. There were nine municipalities covered in this survey and the disease was found endemic in six of them or 67 per cent of the municipalities surveyed.

The prevalence rate of microfilaremia ranged from 0 to 10.25 per cent with the municipality of Balimbing being highest followed consecutively by Pata, Bongao, Siasi, Tapul and Tandu' Bas. The municipalities where filariasis is absent are Pangutaran, Simunul and Sitangkai.

Of the 120 cases found positive for microfilaremia, 91 or 76 per cent were bancroftian filariasis and 29 or 24 per cent were infections due to Brugia malayi. Twenty-three malayan filariasis cases came from Bongao, four from Siasi and two from Tandu' Bas. The municipalities of Bongao and Tandu'Bas belong to the Tawi-Tawi group while Siasi belong to the Tapul group. It appears therefore that Tawi-Tawi group particularly Bongao municipality is the "hot bed" for malayan filariasis in the Sulu archipelago (Fig. 1-b). Incidentally, this becomes the second endemic focus for Brugia malayi infection in the Republic of the Philippines, the first one being in Quezon municipality of Palawan Province.(6)

The overall microfilaremia rate for the nine municipalities of Southern Sulu is only 3.2 per cent as compared to 11.4 per cent for the eight municipalities in Jolo island.(2)

Age and Sex Distribution:

Table II (a-i) and Figures 2-7 present the age and sex distribution of persons examined by municipality. Practically all age groups are represented with the greater bulk of subjects falling below 30 years of age. There were only two positive cases found under 6 years of age. One was a bancroftian filariasis case in a 5-year old boy from barrio Tarawakan, Balimbing municipality, the other was a malayan filariasis case in a 3-year-old boy coming from barrio Masantong, municipality of Bongao. The oldest cases found were two females ages 70 and 75 respectively both from barrio Masantong, Bongao, infected with Brugia malayi filariasis. One will notice, that marked increase in prevalence rates are most evident after the age of 20 years in all the positive municipalities. This same general trend of increase in prevalence after the age of 20 years was likewise observed in Jolo island(2) as well as in Palawan(7).

Table III and Figure 8 present the prevalence rates of filariasis on the total persons surveyed in Southern Sulu, by age and sex. The total microfilaremia rate for males was 4.4 per cent as compared to 1.7 per cent for females with an overall prevalence rate of 3.2 per cent. The prevalence rate for both sexes among children age 1-15 years is only one per cent as against 5 per cent among children in Jolo island(2). As expected, the males showed consistently higher prevalence rates in all ages except in age group 16-20 years. Table IV and Figure 9 are summaries of prevalence rates by municipality and sex. In all the 6 municipalities found endemic for filariasis the males have consistently higher prevalence rates over the females. The highest prevalence rate for both sexes came from the municipality of Balimbing, followed by Pata, Bongao and Siasi with rates of 10.3, 5.1, 3.8, and 3.7 per cent respectively.

Overall endemicity and Proportion of the Infected Females over Infected Males:

In Table V and Figure 10 we tried to determine the relationship of overall endemicity of filariasis to the proportion of females getting infected over those of the males. One can see that the lower the prevalence rate for both sexes (low endemicity) the farther the ratio of prevalence between sexes from 1, and the higher the prevalence rate (high endemicity) the more even the chances of infection occurring in both sexes. The ratio will therefore be close to 1. Maimbung municipality however showed a deviation from the general trend. In here the per cent positive for both sexes was only 6.2 per cent and yet the ratio between sexes was 0.85 which is rather close to 1. The pattern depicted by the points in Figure 10 slopes upward with the scale for prevalence increasing from left to right. This means that the female prevalence tend to approach the male prevalence in areas of high endemicity where there are relatively higher infection risks. As a rough rule, it can be said that the representation of females percentagewise in the total prevalence picture tends to equal that of the males when the per cent positive for both sexes goes beyond 13 per cent.

Intensity of Microfilaremia:

Table VI (a-f) show the frequency distribution of cases by sex and by intensity of microfilaremia for each of the six municipalities found endemic for filariasis. Whereas in Pata, Tapul and Balimbing only bancroftian filariasis was encountered, in Siasi, Tandu' Bas and Bongao both bancroftian and malayan filariasis are found endemic. In both types of filariasis, the highest microfilarial count was always on a male subject. In Balimbing, Table VI (d) out of 44 positive cases, 31 (25 males and 6 females) or 70.4 per cent had microfilarial counts less than 50 per 20 mm^3 of blood while 13 (10 males and 3 females) or 29.5 per cent had counts above 50 per 20 mm^3 of blood. In Bongao, Table VI (f) where both species of filaria are present at approximately equal prevalence rates, one will note that the microfilarial counts were quite low. The highest counts were 43 and 27 per 20 mm^3 of blood for W. bancrofti and B. malayi respectively.

Table VII shows the frequency distribution of cases by sex and by intensity of microfilaremia in Southern islands, Sulu. The highest count of 544

microfilariae (W. bancrofti) was found in a 36 year-old male from Balimbing. The next highest count was also a male with bancroftian filariasis from Balimbing with 390 microfilariae. There were only 7 cases (6 males and 1 female) with bancroftian filariasis having microfilarial counts beyond 100 per 20 mm^3 of blood and none of the malayan filariasis gave counts beyond 30 microfilariae. The highest count for the malayan type was 27 microfilariae per 20 mm^3 of blood on a 26 year-old male from Bongao. This same subject had also microfilariae of W. bancrofti in addition to B. malayi microfilariae.

Tables VIII (a) and VIII (b) show the intensity of microfilaremia per 20 mm^3 of blood by age and sex in Southern Islands, Sulu. For bancroftian filariasis the highest average microfilarial count per positive among males was 67.4 in the 36-40 age group and 90 among females in the 6-10 age group. For the total average microfilarial count per positive the males had 39.1 while the females had 32.4. For malayan filariasis the highest average microfilarial count per positive among males was 14.5 in the 21-25 age group and 14 among females in the 16-20 age group. However, the number of female subjects found positive for B. malayi microfilariae was rather small to be of significant value in the analysis. For the total average microfilarial count per positive the males had 4.6 while the females had 7.8 microfilariae. It appeared from the data that the average microfilarial count per positive among male subjects with bancroftian filariasis is about 8 times higher than those with malayan filariasis, while the comparative figure for females is about 4 times.

Table IX gives the median microfilarial counts per 20 mm^3 of blood of 6 municipalities by sex and species. For bancroftian filariasis we found the median microfilarial counts of 21 and 11 among males and females respectively or a total median microfilarial count of 22 microfilariae for both sexes. For malayan filariasis we found the median microfilarial counts of 4 microfilariae for males and 5 for females or a total median microfilarial count of 3 for the two sexes combined. It was observed here that for both sexes combined the municipality of Balimbing had the highest median microfilarial count followed by Pata, Bongao and Siasi in bancroftian filariasis. One will notice a parallelism between the median microfilarial counts and prevalence rates.

Further Analysis of Microfilaria Counts:

Microfilaria counts of individual cases when determined under more or less uniform conditions could give valuable information on the epidemiology of filariasis. They also provide a useful quantitative index in the presentation and evaluation of the results of a parasitological survey. Sasa(9) et al were the first to recognize that some well-known techniques in statistical analysis could be applied to such observations. One important results is the finding that the distribution of positive cases by density of microfilaria per unit volume (usually, 20 mm^3 or 30 mm^3) of blood is essentially logarithmically normal, which simply means that while the original counts follow a highly skewed distribution on account of the presence of a few subjects with extremely high counts compared to the rest, transformation of the counts to corresponding logarithms converts the asymmetrical distribution to an essentially normal distribution. Graphical tests of the data to this effect may be done

either by plotting the frequencies against the logarithms of the counts and examining the resulting frequency curve for any semblance to a normal distributions; or by plotting the cumulative frequencies on log-probability paper. The latter is decidedly a better test since a log normal distribution plots as a straight line on this special graphing paper, thus reducing the test procedure to a graphical appraisal of linearity. Departures from a linear plot can come about in various ways, and these if present may be used to throw more light on the exact nature of the distribution. Lines so obtained will henceforth be referred to as the log-probit regression line or simply regression line of the comulative distribution of microfilarial counts.

The various islands covered in the survey were arbitrarily classified into two groups depending on the extent of filariasis observed. The first group is composed of those with prevalence ratios of 1 per cent and above, while the second includes all those whose prevalence ratios fall below 10 per cent. This was done in an effort to find out if differences in endemicity levels between localities will be similarly reflected in their microfilarial density patterns. Jolo and Balimbing, with prevalence ratios of 11.45 per cent and 10.26 per cent respectively composed the first group. All the others were included in the second group.

Frequency distribution of the counts for these two groups of localities are given in Tables IX (b) and IX (c). Since differentials in density were expected between males and females, separate sections were constructed for the sexes. The tabulations also include, for the purpose of plotting the probit regression line, the cumulative frequencies and the relative cumulative frequencies in per cent. Since the B. malayi infections did not present a radically different distributions as gleaned from the 29 cases so far uncovered, it was felt that separate treatment for the two species is not warranted, although B. malayi counts might have pulled down the peak of the distributions somewhat.

Figure 11 shows the plot of the probit-regression lines for the groups mentioned above. These lines were fitted to the points by eye. Females as expected tend to have relatively less microfilaria per unit volume of blood than males, although this is not very clear in the case of localities with lower endemicities. However, it is quite reasonable to expect that the same degree of consistency as observed in Jolo and Balimbing would have been detected had there been more positives in the distribution. It is to be noted that in this diagram, higher microfilarial counts correspond to lower regression lines relative to that of lighter microfilarial densities. In line with this then, we can easily conclude from the diagram that the more endemic localities also showed higher counts among positives. Regression lines for male and female positives from Jolo and Balimbing occupy inferior positions relative to the lines for the other localities surveyed. These considerations may be placed on a more quantitative basis as follows: When the lines are parallel or effectively so, as is the case here at least for the greater part of the distributions, a meaningful index for comparing the distributions is the median microfilaria density. This is obtained from each distribution by drawing a line at the 50 per cent level. The point of intersection of this line and the regression is the median microfilaria density for the distribution. This value is considered

more reliable than the arithmetic mean of the microfilaria counts. From Fig. 11, we see that the median microfilarial density for both sexes at the areas of lower endemicity is about 5 per 20 mm^3, compared to around 11 and 17 per 20 mm^3 for females and males respectively, at areas of higher endemicity. This verifies our previous impressions regarding density of infection and endemicity.

One final point regarding the observed distribution is the effectiveness of the logarithmic transformation in producing normality. It is easily seen that we have regression curves rather than regression lines. There is slight concavity upwards, which means that we observed high microfilarial counts at frequencies less than those required by the lognormal distribution. An informal examination of some published data on microfilaria density reveals that this is the usual departure from log-normal distribution that occurs.

Actually we could pursue this thing further by introducing some adjustments for the non-linearity observed. If we let X represent microfilaria count for a given subject, then the variable actually used in the above analysis is Y = log. X. For the type of departure noticed above, the modified logarithmic transformation z = Log(X + K) would be more useful in achieving linearity. K in this expression is an appropriate constant that can be estimated from the regression curves, However, while this transformation may be more successful in achieving linearity and hence normality of the distributions, further complexities will arise in the interpretation of the variable Z.

Comparison of B. malayi average microfilarial counts per 20 mm^3 blood of subjects from Palawan and Sulu.

As previously stated certain islands of the Southern Sulu became the second endemic focus for malayan filariasis in the Republic of the Philippines. We were curious to find out if malayan filariasis in Sulu was introduced at about the same time as that of Palawan. Table X shows the average microfilarial densities by age, of subjects from Palawan and Sulu. Among positive subjects from Palawan the highest average microfilarial count per positive was 57.5 microfilariae in the 51-55 age group(7), while the highest from Sulu was 14.5 microfilariae in the 21-25 age group. For all ages, the average microfilarial count per positive among Palawan subjects is 27 while Sulu subjects had 5 microfilariae per 20 mm^3 blood.

The forgoing comparison may be somewhat distorted by the presence of extremely high counts which tend to unduly inflate the average number of microfilaria per positive. This is especially important when we consider the fact that the range of the Palawan counts is 1-300 as compared to only 1-29 for Sulu. (See bottom part of Table X.) To minimize the effects of these unusual values in the comparisons, the corresponding median counts were determined for each province. As indicated at the bottom of Table X, the figure 6 and 3 were obtained for Palawan and Sulu respectively. This confirms the general trends shown by the averages per positive, although not in the proportion of 5 to 1 as implied by averages of 27 to 5 microfilaria per case.

Periodicity characteristics of the microfilariae:

From Table XI and Figure 12 it is seen that all the 3 cases of bancroftian filariasis exhibited the typical nocturnal periodicity with practically no microfilariae between 0900 and 1500 hours except for case No. 3 wherein a zero count was seen only at 1100. This finding could be explained by the fact that the peak count was quite high and hence few microfilariae may be seen in the peripheral circulation during the daylight hours.

Microfilarial densities of 15 cases of B. malayi infections are presented in the same table and figure. It is seen here that B. malayi microfilaremia increased in intensity in the early evening and remained at a high level during the night but begin to drop at about 0700 in the morning. Such type of microfilarial behavior is termed nocturnal subperiodic. This same periodicity characteristic was also observed among subjects from Palawan where the vector was found to be Mansonia bonneae(6, 8).

It is evident in the two mixed-infections, (cases No. 3 and 14) that each parasite maintained its own type of periodicity and was not in any way influenced by the presence of the other species of microfilariae.

In the determination of the vector of malayan filariasis in Palawan, we have found Mansonia bonneae as the vector(6), whether or not this finding holds true in Sulu is explored in Part IV.

Discussion

Due to the lack of water transportation going to the Samales group of islands, we were unable to extend our survey to these areas. We had attempted no less than 4 times to go to these islands but we failed. However, from some epidemiological information we gathered among people who have been there we felt that filariasis could not be endemic in these islands.

Now that we have completed our filariasis survey in the southern islands of Sulu, it is quite evident from the results that filariasis as a public health problem seemed to be focused in the 8 municipalities of Jolo island with prevalence rate of 11.4 per cent as compared to 3.24 per cent for the 9 municipalities in southern islands of Sulu.

In areas of low endemicity and hence of lesser infection risks, one would suppose that majority of the infection would be confined to the males since it would be reasonable to presume that the factor of occupation will play a greater role in exposure and transmission. If this were so, then there should be some sort of positive relationship between the overall prevalence rate and any index descriptive of the relative sizes of the contributions that each sex has in the overall rate, like the ratio of prevalence in females to that of the males. This aspect was examined in Table V and Figure 10, which suggest that a moderate but significant relationship exists. Indications are that further observations involving places of different endemicity levels will serve to confirm the narrowing sex differential with increasing risks of infection.

The most significant finding in this survey is the existence of malayan filariasis in 3 municipalities of southern Sulu. Whereas in the island of Jolo, there was not a single case of malayan filariasis found out of 526 positive cases, this recent survey revealed 29 B. malayi cases from 120 microfilaremia positive cases, or 24 per cent of the total positives. Twenty-three of these malayan filariasis cases came from Bongao municipality which we have labeled as the "hot bed" for this species of filariasis in Sulu Province. Barrio Masantong had 7 B. malayi cases out of 40 examined or 17.5 per cent prevalence. There is no more doubt in our minds that B. malayi infection is endemic in this area because of several cases encountered and also the fact that 9 cases were found among children below 15 years of age with no history of having left the place.

There were only two positive cases found under 6 years of age while the majority of cases fall after the age of 20 years. The oldest case was a 75 year-old female from Bongao. As expected, the males showed consistently higher prevalence rates in all ages as compared to the females. This trend was observed in all municipalities found endemic.

The lower the endemicity of filariasis the farther the ratio of prevalence between females over males from 1, and the higher the endemicity the more even the chances of infection between sexes. The latter condition implies that the ratio will be close to 1.

The highest microfilarial count for W. bancrofti was 544 while the highest count for B. malayi was only 27 microfilariae per 20 mm^3. The total average microfilarial count per positive is approximately 35 for W. bancrofti and about 6 microfilariae for B. malayi, or 6 times higher in W. bancrofti filariasis. The total median microfilarial count for W. bancrofti was 22 microfilariae while it was only 3 for B. malayi.

A more ideal method of comparing intensity of infection using microfilarial counts between males and females or between areas of varying endemicity is by the use of the "Log-Probit Regression Line". Whereas the comparison using averages is concerned only with certain points of the distribution the above mentioned method provides a comprehensive comparison of the distributions. This method also can be utilized in the assessment of a control program for filariasis.

Comparison of the average B. malayi microfilarial counts per 20 mm^3 blood of subjects from Palawan surveyed by us about two years ago and this recent survey revealed that Palawan subjects had 27 microfilariae while those from Sulu had only 5 microfilariae. It can be implied here that probably the malayan filariasis in Sulu has only been recently introduced and that this same disease must have been in Palawan for quite some time. These findings are further supported by the absence of scrotal as well as leg enlargement among microfilaremia cases.

Knowledge of the periodicity characteristics of microfilariae will enable one to know the right time to take a blood smear for the diagnosis

of filariasis. It is also important to know microfilarial periodicity in the determination of the probable mosquito vector. Our previous study of malayan filariasis in Palawan revealed that the behavior of the microfilariae was nocturnal subperiodic and we found the mosquito vector to be Mansonia bonneae. In Malaysia(5) in addition to the existence of a subperiodic malayan filariasis, they also have the so-called nocturnal periodic type, transmitted by the Anopheles barbirostris group. The latter type of periodicity has not been observed in Palawan nor in Sulu.

Periodicity studies of the malayan filariasis cases in this survey revealed that the behavior was also nocturnal subperiodic and hence we can surmise that probably the vector could also be Mansonia rather than Anopheles barbirostris group. There were two cases of mixed infection and it is interesting to note that despite the presence of two species of filaria in one individual, each species maintained its own periodicity characteristics.

Summary and Conclusion

Filariasis was found endemic in 6 out of 9 municipalities surveyed in Southern Sulu. A total of 3,695 persons were examined and 120 individuals were found positive or a prevalence rate of 3.2 per cent. Of these 120 positives 91 or 76 per cent were W. bancrofti while 29 or 24 per cent were B. malayi filariasis. The Tawi-Tawi group of islands became the second endemic focus for malayan filariasis in the Republic of the Philippines.

The youngest cases found were under 6 years of age while the oldest were above 70 years of age. The marked increase in prevalence rates was evident after the age of 20 years, and as expected the males showed higher prevalence over the females in practically all age groups and in all municipalities found endemic for filariasis. Of 120 positives 94 were males and 26 were females.

In areas with high endemicity the female prevalence tend to approach the male prevalence because of higher infection risks. The reverse is true in areas of low endemicity where the male prevalence is higher compared to female prevalence, due probably to occupational exposure risk.

The average microfilarial count per positive among male subjects with W. bancrofti is 8 times higher than those with the malayan type, while for females with W. bancrofti is 4 times higher than those with B. malayi.

The Log-Probit Regression Line is ideal for comparing intensity of infection using microfilarial counts between sexes or areas of varying endemicity. Likewise this method is best applied in the assessment of filariasis control programs.

Based on the average microfilarial count per positive plus the absence of physical signs of filariasis, it would seem logical to state that malayan filariasis in Sulu Province is relatively new as compared to that of Palawan Province.

Microfilarial behavior among W. bancrofti cases was nocturnal periodic while among B. malayi cases it was nocturnal subperiodic. Microfilarial periodicity in Sulu is the same as that in Palawan for both species of filaria.

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Table I. Prevalence of Microfilaremia by Municipality, Southern Sulu, 1967.

Municipality	No. Examined	No. Positive	% Positive
Pangutaran Group			
Pangutaran	119	0.00	0.00
Lantung	53	0	0
Simbahan	66	0	0
Jolo Group			
Pata	158	8	5.06
Kayawan	54	2	3.7
Saimbagun	104	6	5.8
Tapul Group			
Siasi	349	13(4)*	3.72
Tausan, Siasi Is.	22	5	22.7
Ambilan, Lapak Is.	76	3(2)	4.0
Lapak Agr. Sch., Lapak Is.	164	3(1)	1.8
Muslim	87	2(1)	2.3
Tapul	309	2	0.64
Kalang, Tapul Is.	225	2	0.9
Gapas, Lugus Is.	84	0	0
Tawi-tawi Group			
Balimbing	429	44	10.25
Malaka	59	2	3.4
Kamagong	63	10	15.9
Tarawakan	83	14	16.9
Agricultural School	60	2	3.3
Malum	41	11	26.8
Kulape	59	4	6.8
Takot-takot	64	1	1.6
Tandu'Bas	602	3(2)	0.50
Sapa	307	0	0
Sallangan	93	1(1)	1.1
Tandu'Bas proper	108	2(1)	1.9
Basbas Isi	94	0	0
Bongao	1320	50(23)	3.78
Lakit-lakit	129	6	4.7
Tubig-basag	69	2	2.9
Pakyas	44	4	9.1
Pancagan	21	0	0
Karungdong	67	2(2)	3.0
Pangasinan	58	0	0

Table ((Cont.)

Municipality	No. Examined	No. Positive	% Positive
Lato-lato	64	0	0
Luuk-Pandan	50	0	0
Tangput	90	0	0
Luuk-Tulay	80	1(1)	1.2
Tubig-Sallangan	52	0	0
Sanga-Sanga	44	0	0
Poblacion	48	0	0
Lapid-lapid	82	4	4.9
Mandulan	154	8(3)	5.19
Malasa	134	4(4)	3.0
Tingol-tingol	94	7(6)	7.4
Masantong	40	12(7)	30.0
Simunul	143	0.00	0.00
Tubig-Indangan	143	0	0
Sibutu Group			
Sitangkai	266	0.00	0.00
Sitangkai poblacion	14	0	0
Tongmaging, Tumindao Is.	53	0	0
Nanukan, Sibutu Is.	126	0	0
Sibutu, Sibutu Is.	73	0	0
T o t a l	3695	120(29)	3.24

* Figures in parenthesis indicate the numbers of B. malayi infections. All others are W. bancrofti.

Table II (a). Filariasis Prevalence Rates, By Age and Sex, Siasi Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	19	---	0	21	---	0	40	---	0
6-10	48	3(3)*	6.3	25	---	0	73	3(3)	4.1
11-15	40	---	0	19	---	0	59	---	0
16-20	24	1	4.2	16	1	6.3	40	2	5.0
21-25	23	1	4.3	5	---	0	28	1	3.6
26-30	17	1	5.9	13	---	0	30	1	3.3
31-35	16	3(1)	18.8	11	---	0	27	3(1)	11.1
36-40	17	1	5.9	7	---	0	24	1	4.2
41-45	14	1	7.1	2	---	0	16	1	6.3
46-50	5	1	20.0	1	---	0	6	1	16.7
51-55	1	---	0	---	---	0	1	---	0
56-60	---	---	0	2	---	0	2	---	0
60 & over	1	---	0	2	---	0	3	---	0
TOTAL	225	12(4)	5.33	124	1	0.81	349	13(4)	3.72

* Figures in parenthesis indicate the number of B. malayi infections. All others are W. bancrofti cases.

Table II (b). Filariasis Prevalence Rates by Age and Sex, Balimbing Municipality, Sulu, 1967.

Age in Year	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	26	1	3.8	26	---	0	52	1	1.9
6-10	63	4	6.3	34	3	8.8	97	7	7.2
11-15	28	1	3.6	16	---	0	44	1	2.3
16-20	47	5	10.6	21	1	4.8	68	6	8.8
21-25	31	6	19.4	12	1	8.3	43	7	16.3
26-30	25	7	28.0	13	1	7.7	38	8	21.1
31-35	13	2	15.4	13	---	0	26	2	7.7
36-40	25	5	20.0	9	1	11.1	34	6	17.6
41-45	6	1	16.7	2	1	50.0	8	2	25.0
46-50	3	1	33.3	1	---	0	4	1	25.0
51-55	2	---	0	4	---	0	6	---	0
56-60	1	1	100.0	2	1	50.0	3	2	66.7
61-65	4	1	25.0	2	---	0	6	1	16.7
66 & over	0	0	0	0	---	0	0	---	0
TOTAL	274	35	12.77	155	9	5.81	429	44	10.25

Table II (c). Filariasis Prevalence Rates by Age and Sex, Tandu'Bas Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	45	---	0	49	---	0	94	---	0
6-10	107	---	0	67	---	0	174	---	0
11-15	70	1	1.4	36	---	0	106	1	0.9
16-20	23	---	0	17	---	0	40	---	0
21-25	13	(1)*	7.7	16	---	0	29	(1)	3.4
26-30	22	(1)	4.5	25	---	0	47	(1)	2.1
31-35	10	---	0	15	---	0	25	---	0
36-40	17	---	0	22	---	0	39	---	0
41-45	13	---	0	6	---	0	19	---	0
46-50	9	---	0	3	---	0	12	---	0
51-55	1	---	0	1	---	0	2	---	0
56-60	2	---	0	3	---	0	5	---	0
61 & over	3	---	0	7	---	0	10	---	0
TOTAL	335	3(2)	0.90	267	---	0	602	3(2)	0.50

* Figures in parenthesis indicate the number of B. malayi infections. All others are W. bancrofti cases.

Table II (d). Filariasis Prevalence Rates by Age and Sex, Tapul Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	24	---	0	26	---	0	50	---	0
6-10	45	---	0	32	---	0	77	---	0
11-15	21	---	0	20	---	0	41	---	0
16-20	11	---	0	8	---	0	19	---	0
21-25	9	---	0	10	---	0	19	---	0
26-30	15	---	0	8	---	0	23	---	0
31-35	5	---	0	17	---	0	22	---	0
36-40	8	1	12.5	17	---	0	25	1	4.0
41-45	9	1	11.1	5	---	0	14	1	7.1
46-50	4	---	0	2	---	0	6	---	0
51-55	2	---	0	1	---	0	3	---	0
56-60	1	---	0	2	---	0	3	---	0
61 & over	5	---	0	2	---	0	7	---	0
TOTAL	159	2	1.26	150	---	0	309	2	0.65

Table II (c). Filariasis Prevalence Rates by Age and Sex, Bongao Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	106	1(1)*	0.94	88	--	0	194	1(1)	0.52
6-10	195	1(1)	0.51	124	1(1)	0.8	319	2(2)	0.63
11-15	115	4(2)	3.47	86	2(1)	2.32	201	6(3)	2.98
16-20	85	1	1.17	56	4(2)	7.14	141	5(2)	3.54
21-25	63	5(1)	7.93	49	1	2.04	112	6(1)	5.35
26-30	46	5(3)	10.8	46	1	2.17	92	6(3)	6.52
31-35	41	6(3)	14.6	36	--	0	77	6(3)	7.79
36-40	34	7(5)	20.5	28	3	10.7	62	10(5)	16.1
41-45	17	3	17.6	19	--	0	36	3	8.33
46-50	23	1	4.34	14	--	0	37	2	5.4
51-55	12	--	0	8	--	0	20	--	0
56-60	6	1(1)	16.6	5	--	0	11	1(1)	9.09
61 & over	10	--	0	8	2(2)	25.0	14	2(2)	14.3
TOTAL	753	35(17)	4.64	567	15(6)	2.65	1320	50(23)	3.78

* Figures in parenthesis indicate B. malayi infections. All others are W. bancrofti cases.

Table II (f). Filariasis Prevalence Rates by Age and Sex, Simunul Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	2	—	0	2	—	0	4	—	0
6-10	48	—	0	32	—	0	80	—	0
11-15	7	—	0	7	—	0	14	—	0
16-20	4	—	0	2	—	0	6	—	0
21-25	1	—	0	—	—	0	1	—	0
26-30	1	—	0	2	—	0	3	—	0
31-35	—	—	0	—	—	0	—	—	0
36-40	7	—	0	5	—	0	12	—	0
41-45	6	—	0	1	—	0	7	—	0
46-50	5	—	0	1	—	0	6	—	0
51-55	4	—	0	1	—	0	5	—	0
56-60	3	—	0	2	—	0	5	—	0
61 & over	—	—	0	—	—	0	—	—	0
TOTAL	88	—	0	55	—	0	143	—	0

Table II (g). Filariasis Prevalence Rates by Age and Sex, Sitangkai Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	15	—	0	12	—	0	27	—	0
6-10	29	—	0	34	—	0	63	—	0
11-15	11	—	0	12	—	0	23	—	0
16-20	14	—	0	17	—	0	31	—	0
21-25	10	—	0	14	—	0	24	—	0
26-30	17	—	0	12	—	0	29	—	0
31-35	6	—	0	11	—	0	17	—	0
36-40	6	—	0	4	—	0	10	—	0
41-45	6	—	0	7	—	0	13	—	0
46-50	4	—	0	7	—	0	11	—	0
51-55	5	—	0	1	—	0	6	—	0
56-60	3	—	0	—	—	0	3	—	0
61 & over	3	—	0	6	—	0	9	—	0
TOTAL	129	—	0	137	—	0	266	—	0

Table II (h). Filariasis Prevalence Rates by Age and Sex, Pata Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	6	—	0	8	—	0	14	—	0
6-10	27	—	0	14	—	0	41	—	0
11-15	17	—	0	3	—	0	20	—	0
16-20	15	1	6.7	2	—	0	17	1	5.9
21-25	9	—	0	4	—	0	13	—	0
26-30	10	—	0	9	—	0	19	—	0
31-35	9	—	0	4	—	0	13	—	0
36-40	6	2	33.3	—	—	0	6	2	33.3
41-45	3	3	100.0	7	1	14.3	10	4	40.0
46-50	—	—	0	—	—	0	—	—	0
51-55	2	—	0	—	—	0	2	—	0
56-60	—	—	0	—	—	0	—	—	0
61 & over	3	1	33.3	—	—	0	3	1	33.3
TOTAL	107	7	6.54	51	1	1.96	158	8	5.06

Table II (i). Filariasis Prevalence Rates by Age and Sex, Pangutaran Municipality, Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	19	--	0	8	--	0	27	--	0
6-10	16	--	0	17	--	0	33	--	0
11-15	7	--	0	4	--	0	11	--	0
16-20	5	--	0	4	--	0	9	--	0
21-25	1	--	0	--	--	0	1	--	0
26-30	7	--	0	9	--	0	16	--	0
31-35	5	--	0	8	--	0	13	--	0
36-40	3	--	0	3	--	0	6	--	0
41-45	1	--	0	--	--	0	1	--	0
46-50	--	--	0	--	--	0	--	--	0
51-55	2	--	0	--	--	0	2	--	0
56-60	--	--	0	--	--	0	--	--	0
61 & over	--	--	0	--	--	0	--	--	0
TOTAL	66	--	0	53	--	0	119	--	0

Table III. Filariasis Prevalence Rates, by Age and Sex, Southern Sulu, 1967.

Age in Years	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
1-5	262	2(1)*	0.76	240	--	0	502	2(1)	0.39
6-10	578	8(4)	1.38	379	4(1)	1.05	957	12(5)	1.25
11-15	316	6(2)	1.89	203	2(1)	0.98	519	8(3)	1.54
16-20	228	8	3.50	143	6(2)	4.19	371	14(2)	3.77
21-25	160	13(2)	8.12	110	2	1.81	270	15(2)	5.55
26-30	160	14(4)	8.75	137	2	1.45	297	16(4)	5.38
31-35	105	11(4)	10.47	115	--	0	220	11(4)	5.00
36-40	123	16(5)	13.0	95	4	4.25	218	20(5)	9.17
41-45	75	9	12.0	49	2	4.08	124	11	8.87
46-50	53	3	5.66	29	1	3.44	82	4	4.87
51-55	31	--	0	16	--	0	47	--	0
56-60	16	2(1)	12.5	16	1	6.25	32	3(1)	9.37
61 & over	29	2	6.89	27	2(2)	7.40	56	4(2)	7.14
TOTAL	2136	94(23)	4.40	1559	26(6)	1.66	3695	120(29)	3.24

*. Figures in parenthesis indicate the number of B. malayi infections. All others are W. bancrofti cases.

Table IV. Filariasis Prevalence Rates by Municipality and Sex, Southern Sulu, 1967.

Municipality	Males			Females			Both Sexes		
	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.	No. Exam.	No. Pos.	% Pos.
Pangutaran	66	—	0	53	—	0	119	—	0
Pata	107	7	6.54	51	1	1.96	158	8	5.06
Tapul	159	2	1.26	150	—	0	309	2	0.65
Siasi	225	12(4)*	5.33	124	1	0.81	349	13(4)	3.72
Tandu'Bas	335	3(2)	0.90	267	—	0	602	3(2)	0.50
Balimbing	274	35	12.77	155	9	5.81	429	44	10.26
Bongao	753	35(17)	4.64	567	15(6)	2.64	1320	50(23)	3.78
Simunul	88	—	0	55	—	0	143	—	0
Sitangkai	129	—	0	137	—	0	266	—	0
TOTAL	2136	94(23)	4.40	1559	26(6)	1.66	3695	120(29)	3.24

* Figures in parenthesis indicate the number of B. malayi infections. All others are W. bancrofti cases.

Table V. Proportion Positive for Filaria and the Ratio (R) of Prevalence Rates Between Sexes in Municipalities Surveyed, Sulu, 1967.

Municipality*	% Positive Both Sexes	Female Prevalence Rate / Male Prevalence Rate
Pangutaran	0	--
Pata	5.06	0.30
Taoul	0.65	0
Siasi	3.72	0.15
Tandu' Bas	0.50	0
Balimbing	10.26	0.45
Bongao	3.78	0.57
Simunul	0	--
Sitangkai	0	--
Jolo	3.37	0.37
Taglibi (Patikul)	17.26	0.52
Indanan	16.72	1.06
Bilaan (Talipao)	13.67	0.84
Panamao (Seit)	16.01	0.65
Luuk (Camp Andres)	17.20	0.92
Maimbung	6.16	0.85
Parang	4.19	0.43

* Data for the last 8 municipalities were taken from Cabrera et al: Filariasis Survey in Jolo, Sulu, Acta Med. Phil. 3:1966.

Table VI (a). Intensity of W. bancrofti Microfilaremia by Sex, Pata, Sulu, 1967.

Mf. Count per 20 mm^3 Blood	Male	Female	Total
1-5	-	-	-
6-10	2	1	3
11-25	1	-	1
26-50	2	-	2
51-100	1	-	1
101-125	1	-	1
TOTAL	7	1	8

Note: Highest microfilarial count: 41 year-old male, Mf:135.

Table VI (b). Intensity of Microfilaremia by Sex, Siasi, Sulu, 1967.

Mf. Count Per 20 mm^3 Blood	Male		Female		Total	
	W.b.	B.m.	W.b.	B.m.	W.b.	B.m.
1-5	5	3	-	-	5	3
6-10	1	-	-	-	1	-
11-25	-	1	-	-	-	1
26-50	-	-	1	-	1	-
51-100	1	-	-	-	1	-
101-116	1	-	-	-	1	-
TOTAL	8	4	1	-	9	4

Note: Highest microfilarial count for W. bancrofti: 35 year-old male, Mf:116.
Highest microfilarial count for B. malayi: 7 year-old male, Mf:13.

Table VI (c). Intensity of W. bancrofti Microfilaremia, Tapul, Sulu, 1967.

Mf. Count Per 20 mm^3 Blood	Male	Female	Total
1-5	-	-	-
6-10	-	-	-
11-25	2	-	2
TOTAL	2	-	2

Note: Highest microfilarial count: 45 year-old male, Mf:13.

Table VI (d). Intensity of W. bancrofti Microfilaremia, Balimbing, Sulu, 1967.

Mf. Count Per 20 mm^3 Blood	Male	Female	Total
1-5	9	1	10
6-10	6	2	8
11-25	4	-	4
26-50	6	3	9
51-100	6	2	8
101-200	2	-	2
201-300	-	1	1
301-400	1	-	1
401-544	1	-	1
TOTAL	35	9	44

Note: Highest microfilarial count: 36-year-old male, Mf:544.

Table VI (e). Intensity of Microfilaremia, Tandu' Bas, Sulu, 1967.

Mf. Count Per 20 mm^3 Blood	Male		Female		Total	
	W.b.	B.m.	W.b.	B.m.	W.b.	B.m.
1-5	-	2	-	-	-	2
6-10	1	-	-	-	1	-
11-25	-	-	-	-	-	-
TOTAL	1	2	-	-	1	2

Note: Highest microfilarial count for W. bancrofti: 12 year-old male, Mf:8.
Highest microfilarial count for B. malayi: 23 year-old male, Mf:2.

Table VI (f). Intensity of Microfilaremia, Bongao, Sulu, 1967.

Mf. Count Per 20 mm^3 Blood	Male		Female		Total	
	W.b.	B.m.	W.b.	B.m.	W.b.	B.m.
1-5	6	13	4	3	10	16
6-10	4	3	2	1	6	4
11-25	4	-	2	2	6	2
26-43	4	1	1	-	5	1
TOTAL	18	17	9	6	27	23

Note: Two mixed infection were found and tallied according to species and microfilarial count.

B.m. at 6-10 and 26-50 age groups.
W.b. at 11-20 and 26-50 age groups.

Highest microfilarial count for W. bancrofti: 25 year-old male, Mf:43.
Highest microfilarial count for B. malayi: 25 year-old male, Mf:27.

Table VII. Intensity of Microfilaremia by Sex, Southern Islands, Sulu, 1967.

Mf. Count Per 20 mm³ Blood	Male		Female		Total	
	W.b.	B.m.	W.b.	B.m.	W.b.	B.m.
1-5	20	18	5	3	25	21
6-10	14	3	5	1	19	4
11-25	11	1	2	2	13	3
26-50	12	1	5	-	17	1
51-100	8	-	2	-	10	-
101-200	4	-	-	-	4	-
201-300	-	-	1	-	1	-
301-400	1	-	-	-	1	-
401-544	1	-	-	-	1	-
TOTAL	71	23	20	6	91	29
	94		26		120(29)	

Note: Highest microfilarial count for W. bancrofti: 36 year-old male, Mf:544 from Balimbing.

Highest microfilarial count for B. malayi: 25 year-old male, Mf:27 from Bongao.

Table VIII (a). Intensity of W. bancrofti Microfilaremia per 20 mm³ Blood by Age and Sex, Southern Islands, Sulu, 1967.

Age Group	Male				Female			
	No. Exam.	No. Pos.	Total Mf. Count	Ave. Mf. Count Per Pos.	No. Exam.	No. Pos.	Total Mf. Count	Ave. Mf. Count Per Pos.
1-5	262	1	3	3.0	240	-	-	-
6-10	578	4	247	61.7	379	3	270	90.0
11-15	316	4	52	13.0	203	1	1	1.0
16-20	228	8	177	22.1	143	4	67	16.7
21-25	160	11	736	66.9	110	2	50	25.0
26-30	160	10	87	8.7	137	2	85	42.5
31-35	105	7	350	50.0	115	-	-	-
36-40	123	11	742	67.4	95	4	86	21.5
41-45	75	9	312	34.6	49	2	52	26.0
46-50	53	3	50	16.6	29	1	6	6.0
51-55	31	-	-	-	16	-	-	-
56-60	16	1	6	6.0	16	1	31	31.0
61 & over	29	2	11	5.5	27	-	-	-
TOTAL	2136	71	2773	39.05	1559	20	648	32.4

Table VIII (b). Intensity of B. malayi Microfilaremia per 20 mm^3 Blood by Age and Sex, Southern Islands, Sulu, 1967.

Age Group	Male				Female			
	No. Exam.	No. Pos.	Total Mf. Count	Ave. Mf. Count Per Pos.	No. Exam.	No. Pos.	Total Mf. Count	Ave. Mf. Count Per Pos.
1-5	262	1	4	4.0	240	-	-	-
6-10	578	4	29	7.2	379	1	2	2.0
11-15	316	2	3	1.5	203	1	14	14.0
16-20	228	-	-	-	143	2	28	14.0
21-25	160	2	29	14.5	110	-	-	-
26-30	160	4	8	2.0	137	-	-	-
31-35	105	4	16	4.0	115	-	-	-
36-40	123	5	8	1.6	95	-	-	-
41-45	75	-	-	-	49	-	-	-
46-50	53	-	-	-	29	-	-	-
51-55	31	-	-	-	16	-	-	-
56-60	16	1	8	8.0	16	-	-	-
61 & over	29	-	-	-	27	2	3	1.5
TOTAL	2136	23	105	4.56	1559	6	47	7.8

Table IX(a)Median Microfilarial Count per 20 mm^3 Blood by Sex and Species, Southern Islands, Sulu, 1967.

Municipality	Male		Female		Total	
	W.b.	B.m.	W.b.	B.m.	W.b.	B.m.
Bongao	10	4	9	5	9	4
Balimbing	11	neg	49	neg	25	neg
Tandu' Bas	(a)	(a_1)	neg	neg	neg	neg
Tapul	(b)	neg	neg	neg	neg	neg
Siasi	4	(c)	(c_1)	neg	5	neg
Pata	16	neg	neg	neg	16	neg
TOTAL	21	4	11	5	22	3

Note:

(a) -- One case of W. bancrofti in a male subject, with 8 mf. count.
(a_1) -- Two cases of B. malayi from 2 male subjects, with microfilarial counts of 1 and 2 respectively.
(b) -- Two cases of W. bancrofti found in 2 males with microfilarial counts of 13 and 21 respectively.
(c) -- Four cases of B. malayi, all from males, with microfilarial counts of 13, 1, 4 and 4 respectively.
(c_1) -- One W. bancrofti from a female, with microfilarial count of 27.

Table IX (b). Distribution of Microfilaria Positive Cases by Density per 20 mm^3 Blood Sample and Sex, Jolo and Balimbing, Sulu, 1967.

Mf. Count	Males			Females		
	Number	Cum. Freq.	Cum. %	Number	Cum. Freq.	Cum. %
1	23	23	6.80	21	21	9.21
2	15	38	11.24	17	38	16.67
3	18	56	16.57	7	45	19.74
4	17	73	21.60	11	56	24.56
5	15	88	26.04	12	68	29.82
6	7	95	28.11	13	81	35.53
7	12	107	31.66	6	87	38.16
8	9	116	34.32	4	91	39.91
9	8	124	36.69	5	96	42.11
10	5	129	38.17	7	103	45.18
11-20	57	186	55.03	41	144	63.16
21-30	22	208	61.54	20	164	71.93
31-40	37	245	72.49	19	183	80.26
41-50	16	261	77.22	9	192	84.21
51-60	16	277	81.95	11	203	89.04
61-70	11	288	85.21	4	207	90.79
71-80	9	297	87.87	4	211	92.54
81-90	12	309	91.42	1	212	92.98
91-100	6	315	93.20	-	212	92.98
101-200	18	333	98.52	15	227	99.56
201-300	2	335	99.11	1	228	100.00
301-400	2	337	99.70			
401-500	-	337	99.70			
501-600	1	338	100.00			
TOTAL	338			228		

Table IX (c). Distribution of Microfilaria Positive Cases by Density per 20 mm^3 Blood Sample and Sex, Southern Sulu (excluding Balimbing), 1967.

	Males			Females		
Mf. Count	Number	Cu. Freq.	Cum. %	Number	Cum. Freq.	Cum. %
1	12	12	19.35	4	4	22.22
2	5	17	27.42	3	7	38.89
3	7	24	38.71	-	7	38.89
4	4	28	45.16	1	8	44.44
5	2	30	48.39	-	8	44.44
6	5	35	56.45	1	9	50.00
7	4	39	62.90	2	11	61.11
8	4	43	69.35	1	12	66.67
9	-	43	69.35	-	12	66.67
10	-	43	69.35	-	12	66.67
11-20	7	50	80.65	4	16	88.89
21-30	3	53	85.48	1	17	94.44
31-40	3	56	90.32	1	18	100.00
41-50	2	58	93.55	-	-	-0.00
51-60	-	58	93.55	-	-	0.00
61-70	1	59	95.16	-	-	0.00
71-80	1	60	96.77	-	-	0.00
81-90	-	60	96.77	-	-	0.00
91-100	-	60	96.77	-	-	-0.00
101-200	2	62	100.00	-	-	0.00
TOTAL	62			18		

Table X. Average Microfilarial Densities for B. malayi by Age, Palawan and Sulu, 1965 and 1967.

Age in Years	Palawan* Number Positive	Palawan* Ave. Mf. Count/Pos.	Sulu Number Positive	Sulu Ave. Mf. Count/Pos.
Less than 1	--	0.0	—	0.0
1-5	24	13.3	1	4.0
6-10	37	17.1	5	6.2
11-15	17	33.1	3	5.6
16-20	18	41.7	2	14.0
21-25	18	6.1	2	14.5
26-30	22	43.2	4	2.0
31-35	13	39.5	4	4.0
36-40	14	39.5	5	1.6
41-45	9	21.0	—	0.0
46-50	4	15.8	--	0.0
51-55	4	57.5	--	0.0
56-60	1	4.0	1	8.0
61 & over	3	2.3	2	1.5
TOTAL	184	26.6	29	5.2
Microfilaremia Rate:	$\frac{184}{3726}$ X 100 = 4.9		$\frac{29}{3695}$ X 100 = 0.78	
Range of Mf. Counts/20 mm^3	1-800		1-29	
Median of Mf. Counts/20 mm^3	6		3	

* Data were taken from Cabrera et al: Bancroftian and Malayan Filariasis in Palawan. Extent and Distribution, Acta Med. Phil., 3:1966.

Table XI. Microfilarial Counts in 20 mm^3 Blood of 16 Microfilaremia Cases, Bongao, Sulu, 1967.

Case No.	Species	TIME 0700	0900	1100	1300	1500	1700	1900	2100	2300	0100	0300	0500
1.	B.m.	12	5	1	3	6	4	16	10	11	9	8	14
2.	B.m.	0	0	0	0	0	1	0	4	4	1	3	1
3.	B.m.	32	21	21	13	9	19	27	41	42	58	35	38
	W.b.	25	1	0	2	1	2	43	90	111	58	95	60
4.	B.m.	2	0	0	1	1	1	3	4	2	2	8	9
5.	B.m.	0	0	0	0	0	4	3	8	6	7	5	5
6.	B.m.	2	2	2	1	0	2	1	1	1	1	2	5
7.	B.m.	0	0	0	0	0	3	3	5	4	6	2	3
8.	B.m.	12	9	1	6	4	5	11	8	19	16	-	15
9.	B.m.	21	16	11	9	13	43	31	43	41	63	60	-
10.	B.m.	2	2	3	4	1	3	12	14	9	6	5	5
11.	B.m.	5	1	0	0	0	0	0	3	2	6	2	2
12.	B.m.	2	1	1	2	3	6	2	6	6	3	10	6
13.	B.m.	3	2	0	0	0	2	4	6	7	4	8	8
14.	B.m.	10	3	2	5	5	9	9	6	12	10	17	8
	W.b.	1	0	0	0	1	12	17	26	26	31	45	22
15.	B.m.	4	3	0	0	0	1	9	2	6	4	5	2
16.	W.b.	1	1	0	0	0	3	15	27	49	69	27	27
Mean:													
15 Cases	B.m.	36.5	19.8	9.9	13.1	11.5	33.3	46.9	62.3	64.8	61.4	66.4	62.0
3 Cases	W.b.	8.7	0.8	0.0	0.6	1.0	10.7	32.8	59.4	76.3	73.7	74.9	47.4

Note: A dash (-) means the smear was not taken.

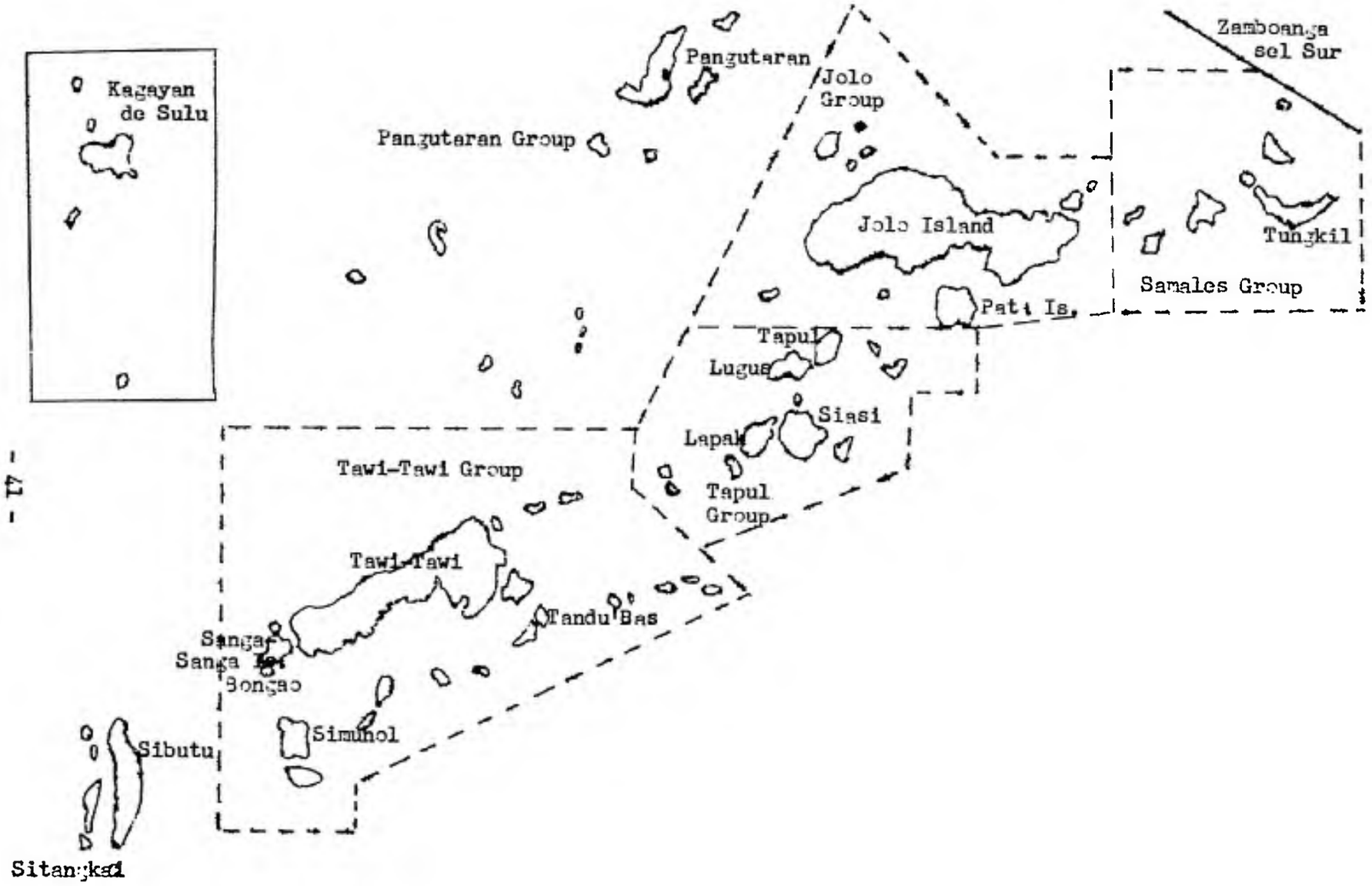


Figure 1-a. Map of Sulu Archipelago.

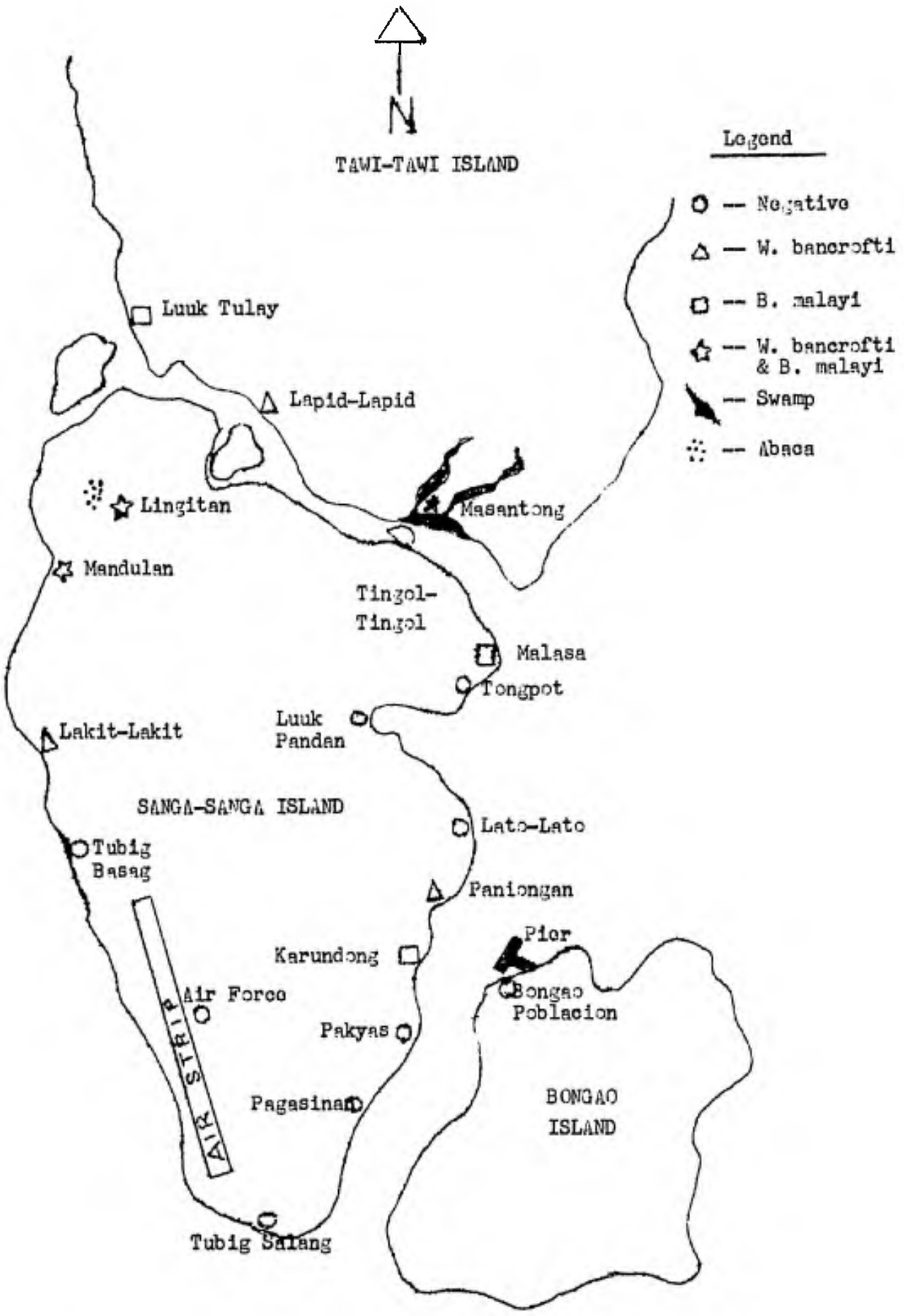


Figure 1-b . Endemicity of Filariasis in the Municipality of Bongao, Sulu, 1967.

Note: All barrios indicated in the map fall under Bongao Municipality.

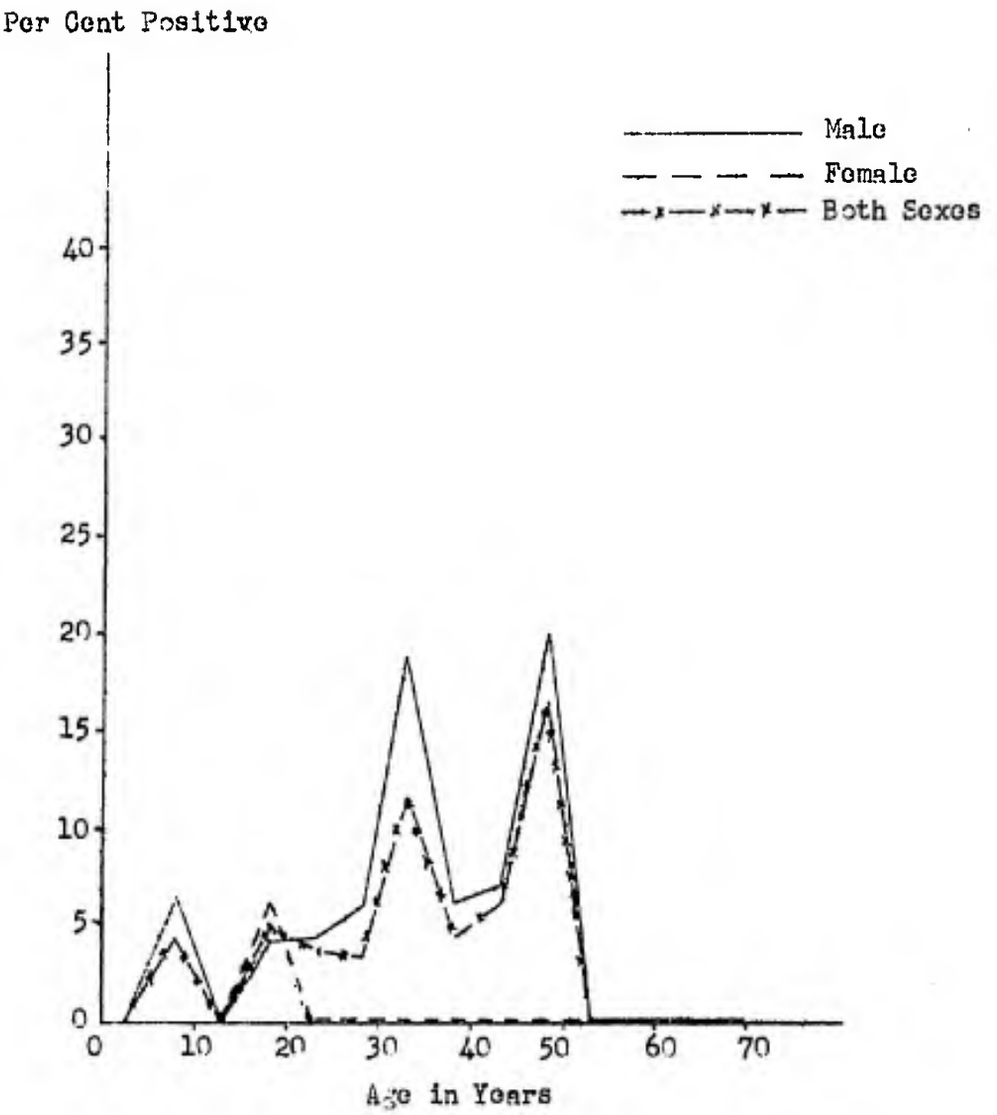


Figure 2. . Filariasis Prevalence Rates, by Age and Sex, Siasi Municipality, Sulu, 1967.

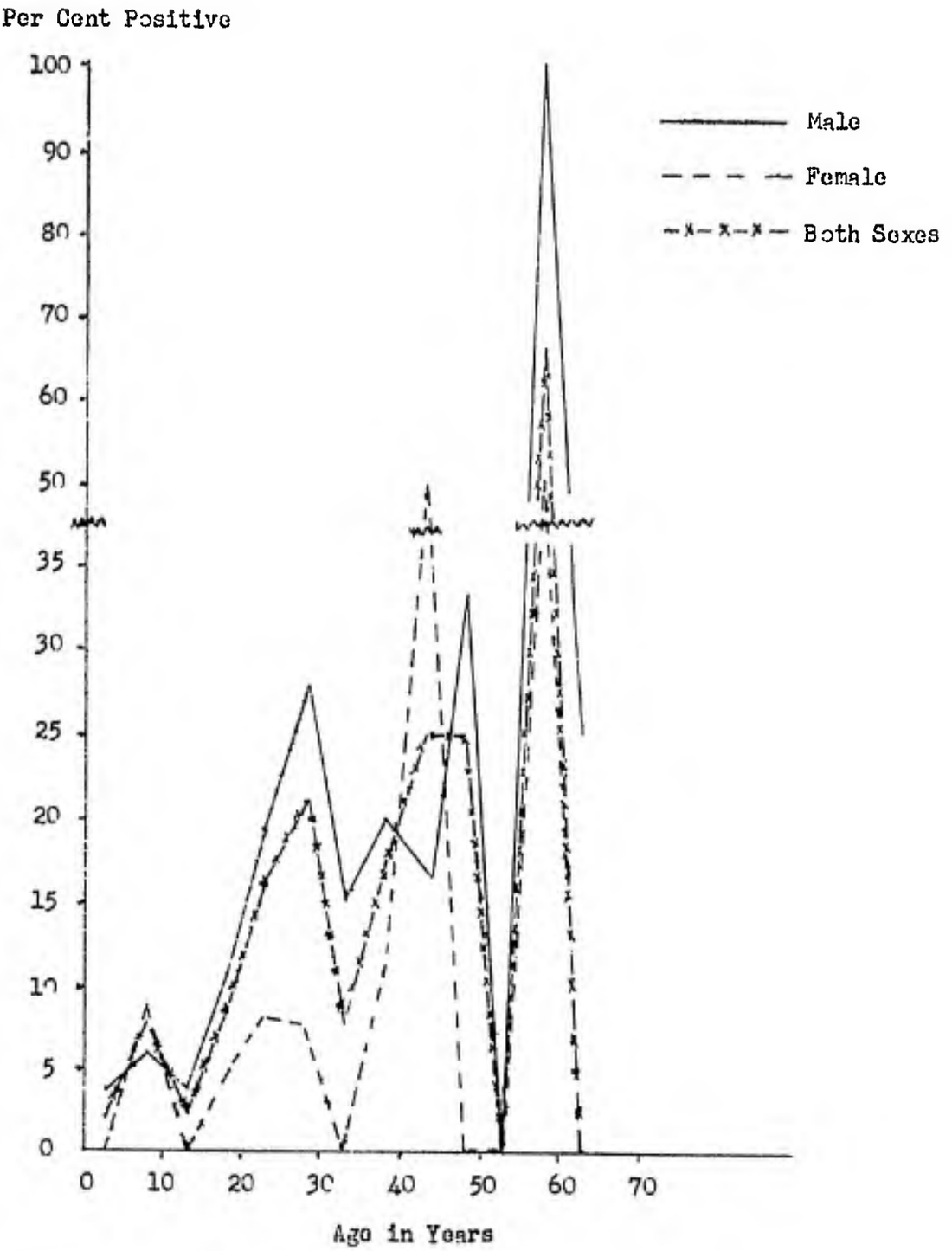


Figure 3 . Filariasis Prevalence Rates By Age and Sex, Balimbing Municipality, Sulu, 1967.

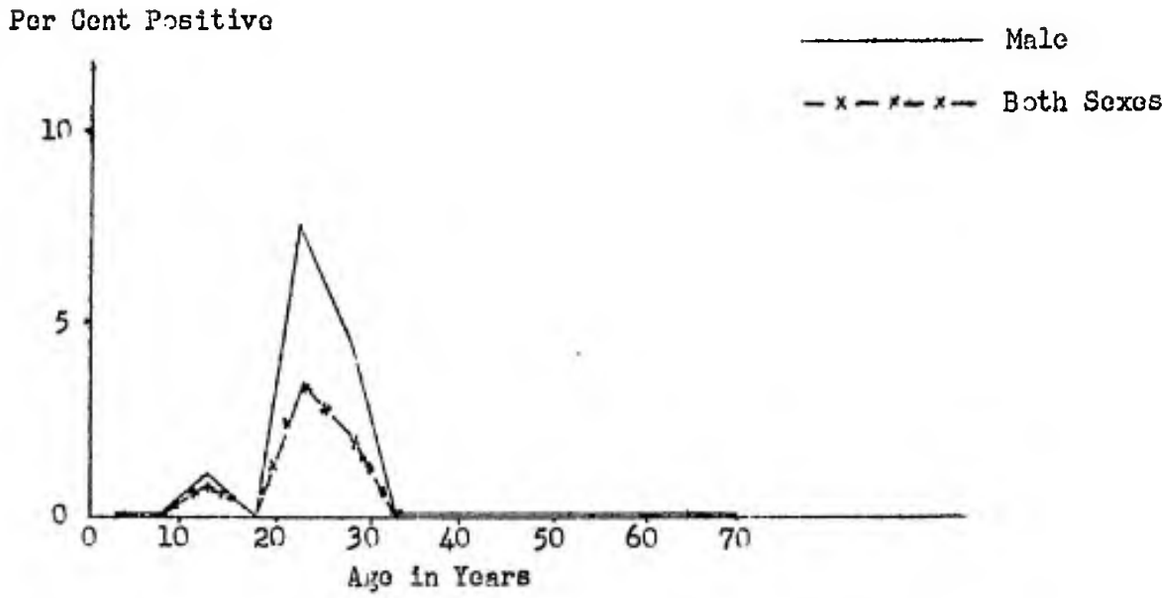


Figure 4 . Filariasis Prevalence Rates by Age and Sex, Tandu' Bas Municipality, Sulu, 1967.

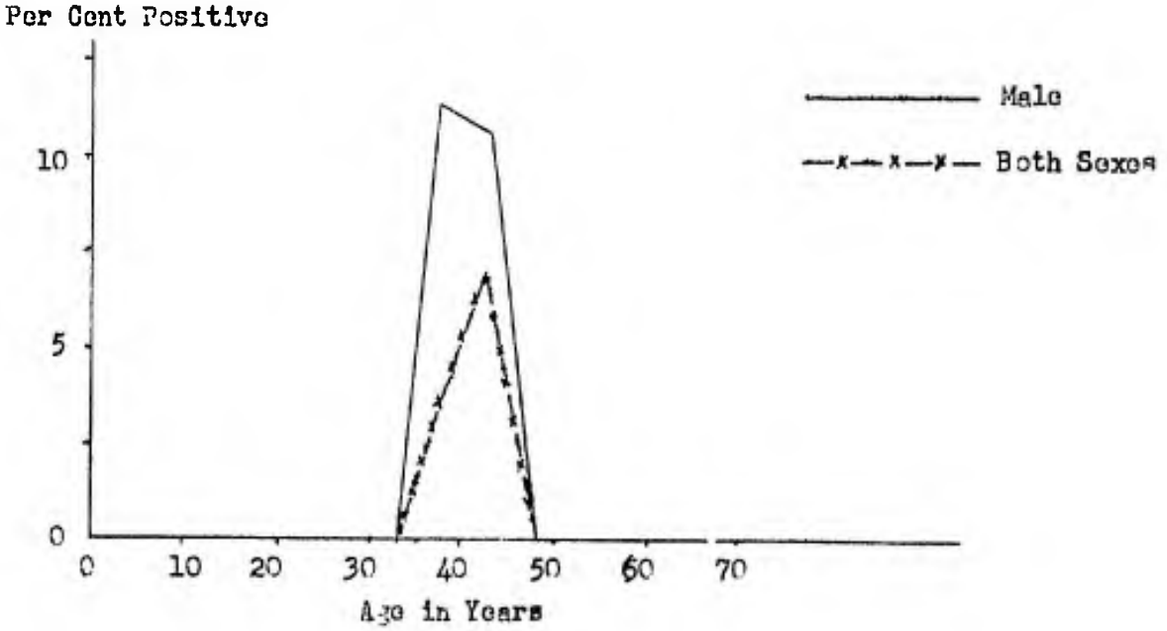


Figure 5. . Filariasis Prevalence Rates by Age and Sex, Tapul Municipality, Sulu, 1967

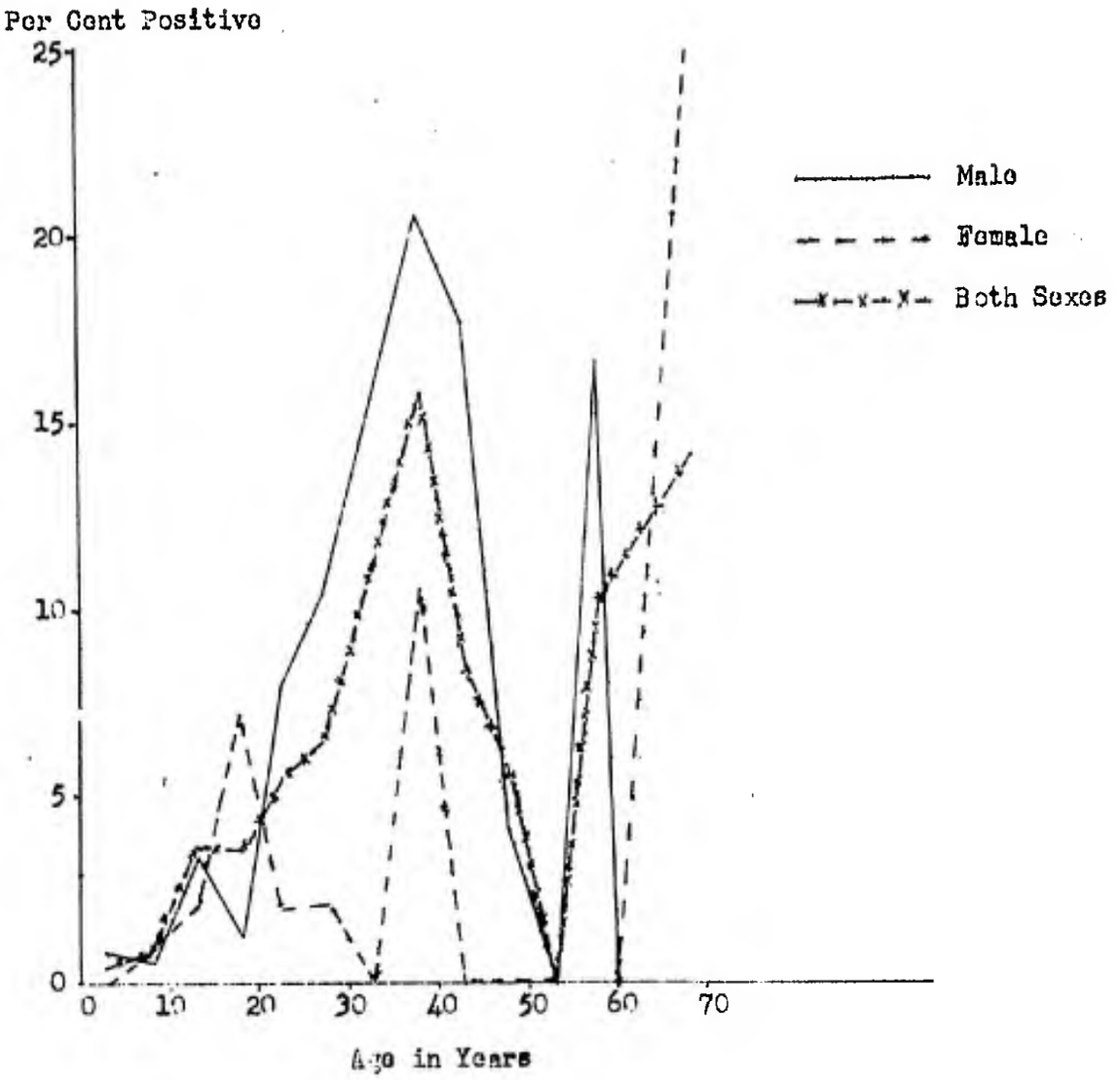


Figure 6 . Filariasis Prevalence by Age and Sex, Bongao Municipality, Sulu, 1967.

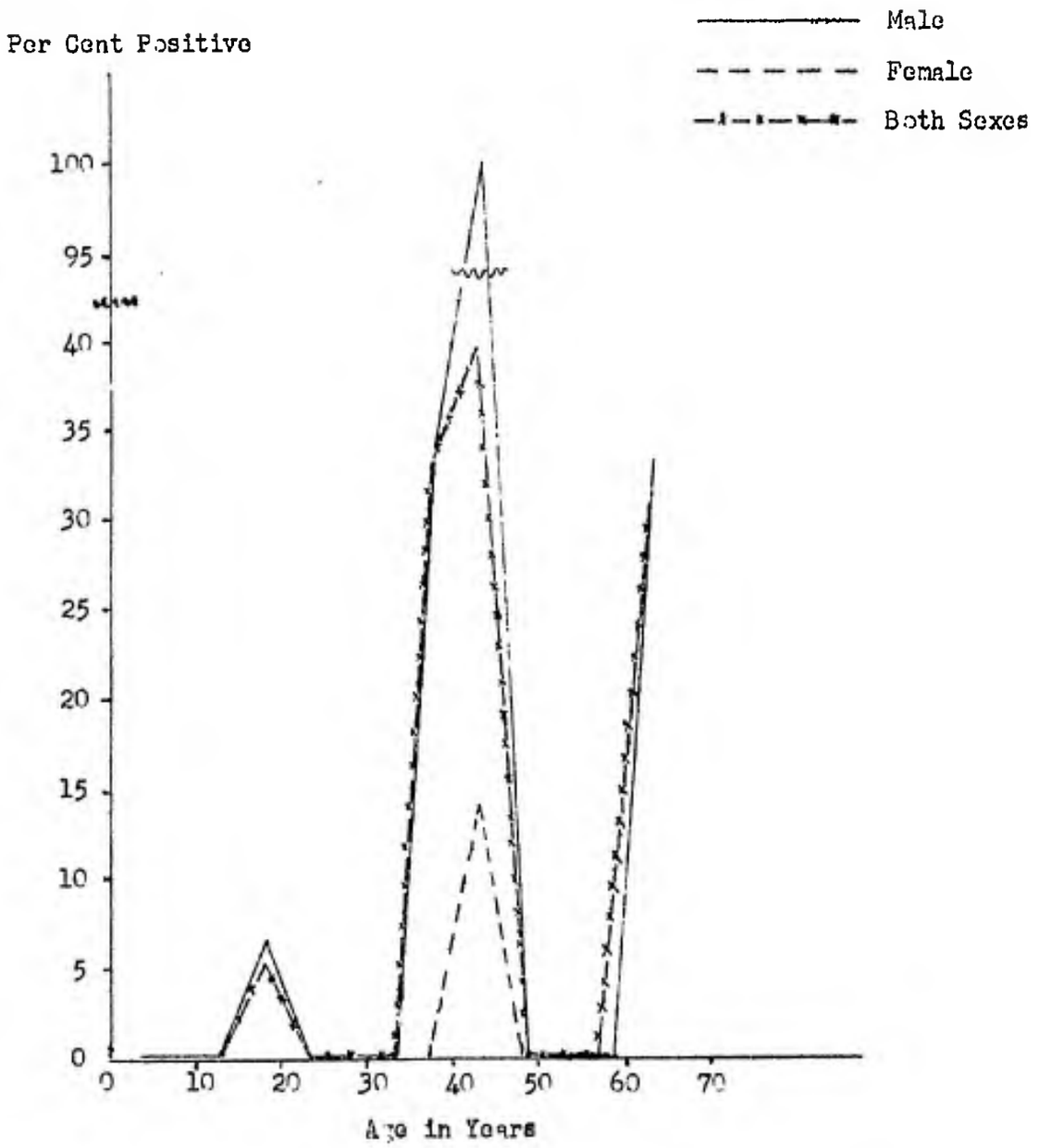


Figure 7. . Filariasis Prevalence Rates by Age and Sex, Pata Municipality, Sulu, 1967

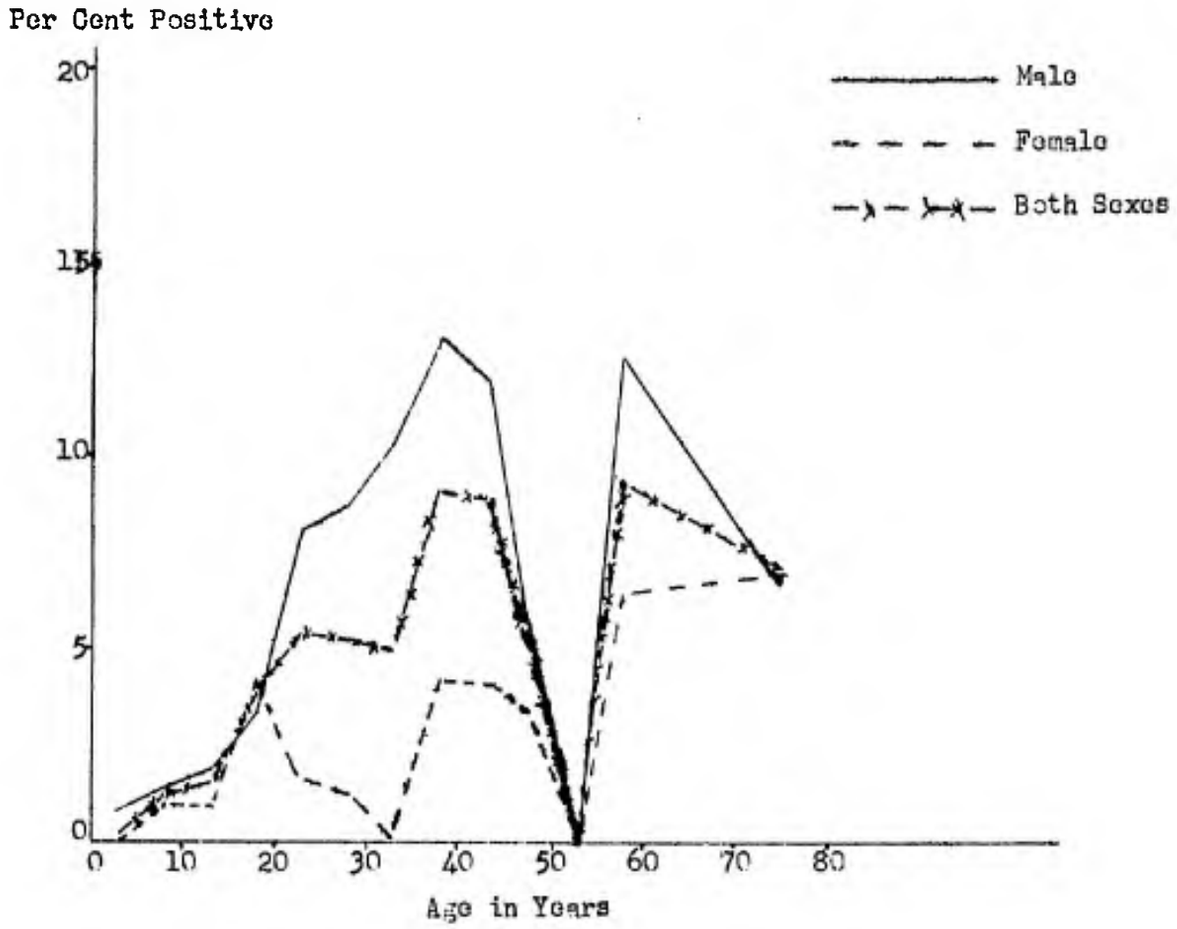


Figure 8. Filariasis Prevalence Rates by Age and Sex, Southern Sulu, 1967.

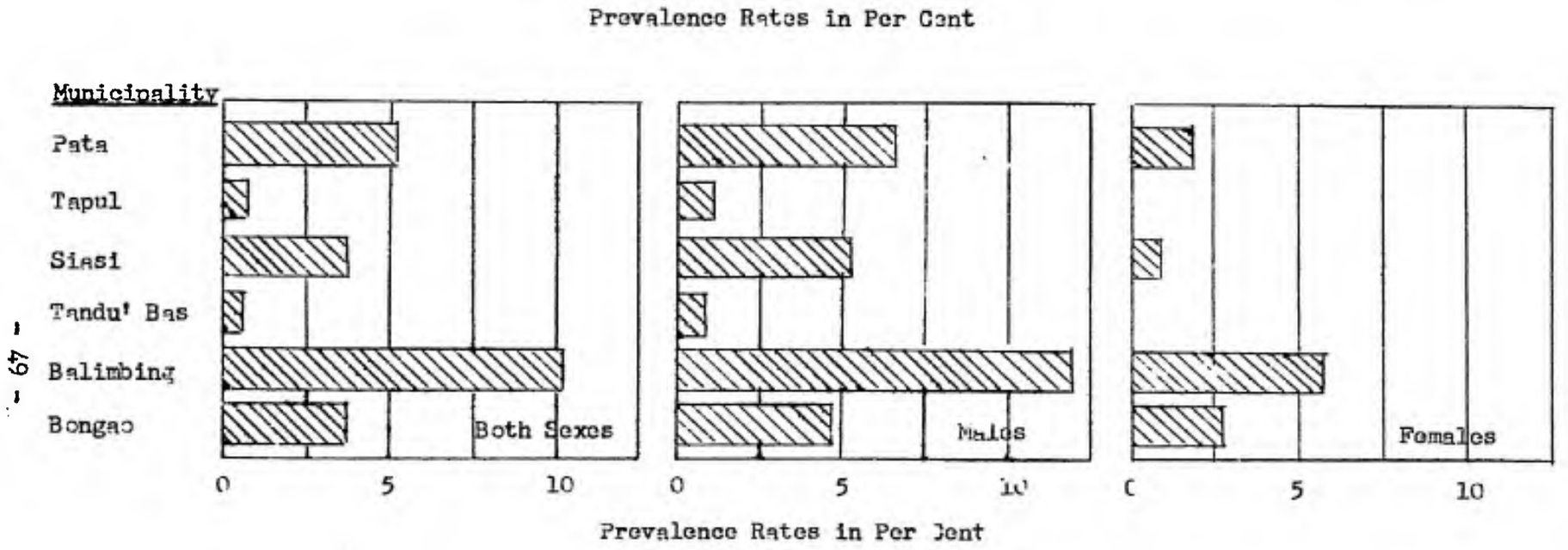


Figure 9 . W. bancrofti Prevalence Rates of 6 Municipalities, by Sex, Sulu Island, Sulu, 1967.

Figure 10. Relationship Between Proportion of Positives (Both Sexes) and the Ratio, R = % Positive, Females/% Positive, Males.

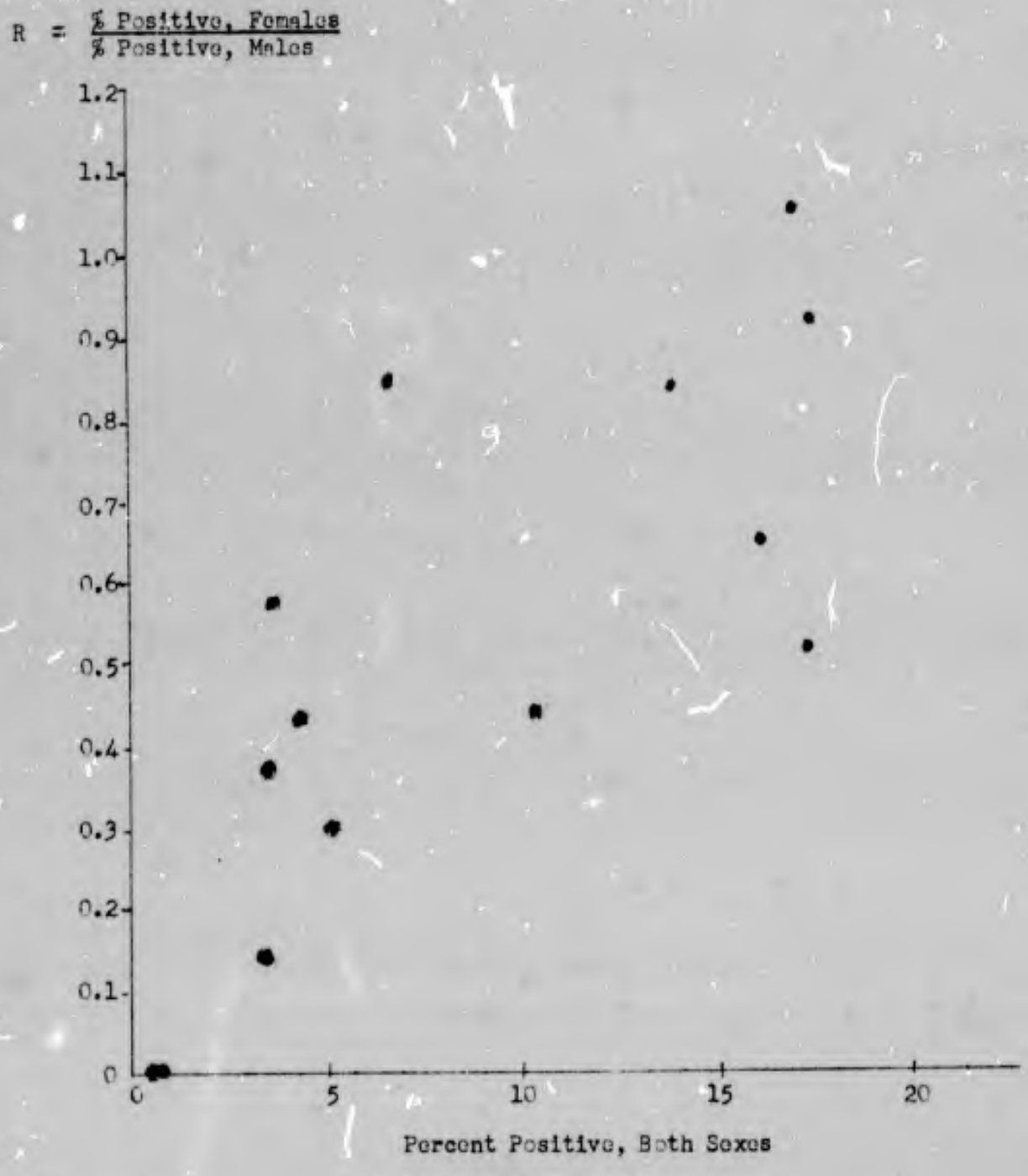
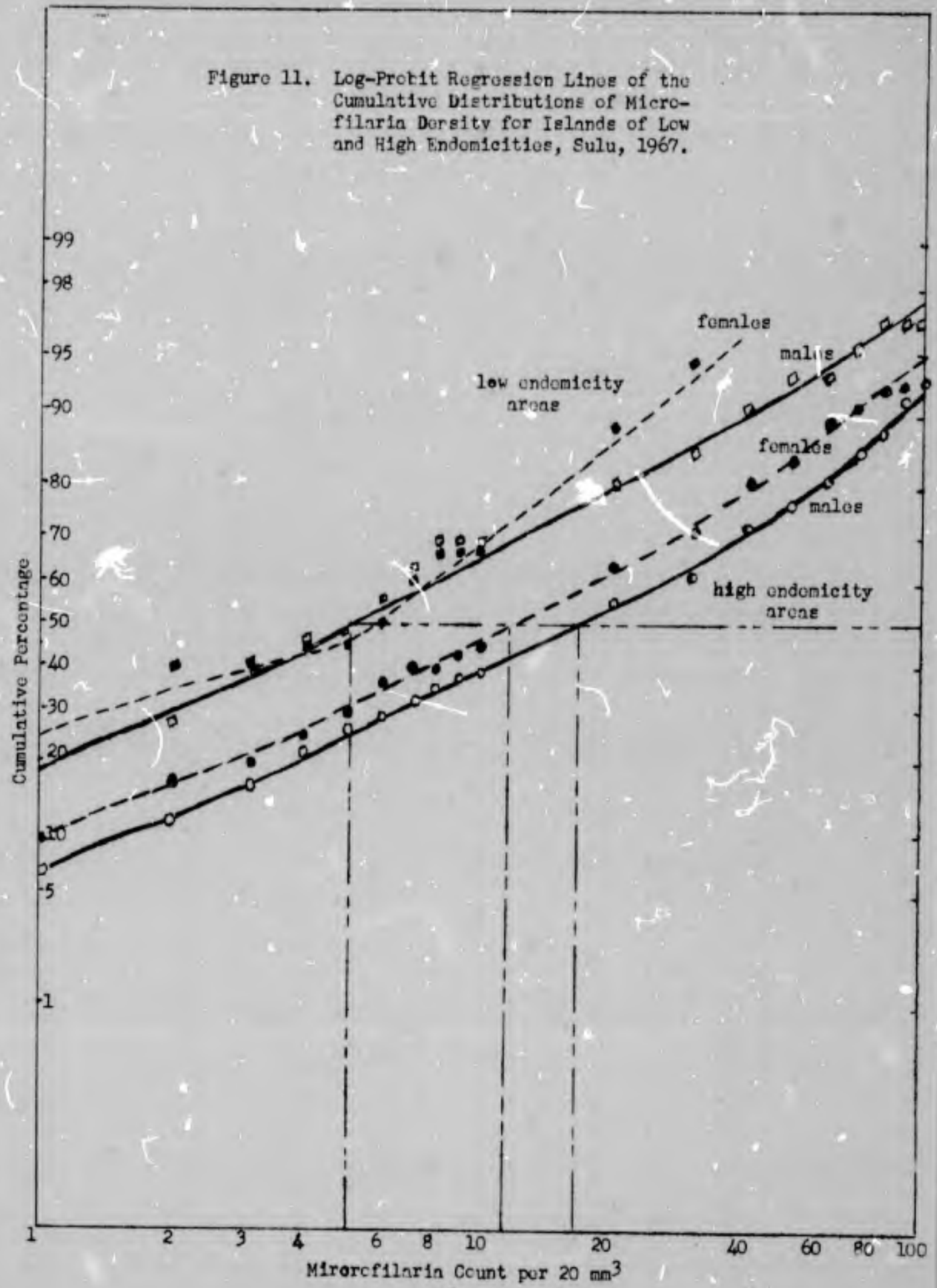


Figure 11. Log-Probit Regression Lines of the Cumulative Distributions of Microfilaria Density for Islands of Low and High Endemicities, Sulu, 1967.



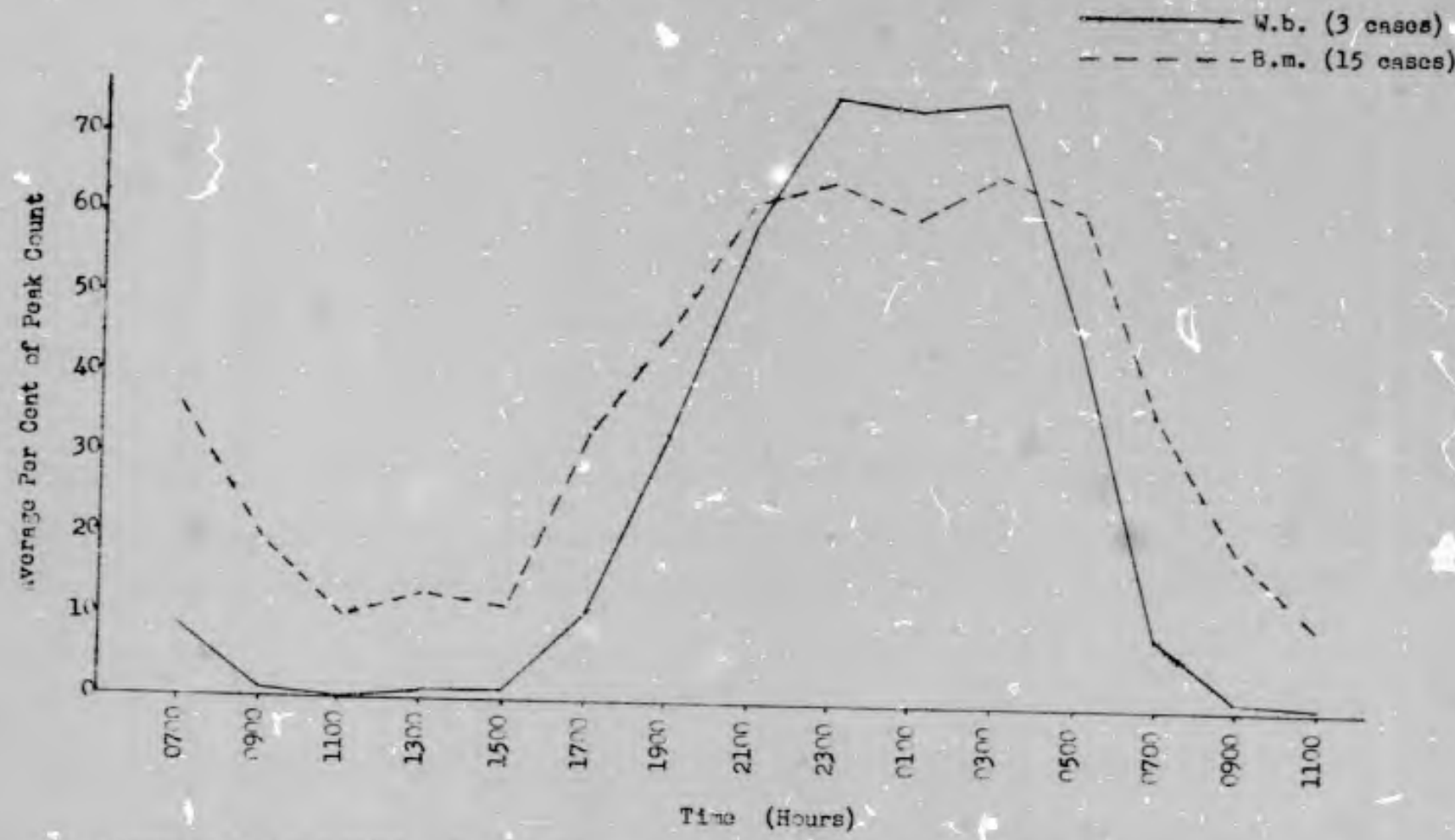


Figure 12. Periodicity of Microfilaremia of 15 B. malayi and 3 W. bancrofti Cases, Bongao, Sulu, 1967.

II. Correlation of abaca industry with filariasis prevalence.

An earlier observation on the correlation of presence of abaca with filariasis was made by Rozeboom and Cabrera (1956) where they found that in 13 abaca planted provinces,10 were also positive for filariasis as compared to 23 non-abaca planted provinces with only 3 localities positive for filariasis(1). In 2 of these 3 non-abaca planted localities, the vector of filariasis was found to be Anopheles minimus flavirostris whose breeding preference are clear, flowing mountain streams(2,3). In the Bicol regions particularly Sorsogon province and more recently in Jolo island, the vector was found to be Aedes (Finlaya) poecilus which breed in the water collected in the axils of abaca plants(4).

Our findings in all the eight municipalities of Jolo island showed no statistical significant correlation between extent of abaca plantation and the magnitude of filariasis prevalence. We have attempted to explain this negative finding to the closeness between municipalities and the overlapping of abaca plantations among municipalities(5). If these explanations were valid then we expect to obtain a significant correlation between abaca plantation and filariasis prevalence by individual islands rather than by municipalities within the same island. With this method, the factor of distance between municipalities and overlapping of abaca plantations among adjacent municipalities were eliminated.

Materials and Methods

The materials used here are the same blood survey results used in Part I of this report plus the data obtained last year for Jolo island(5). Members of our survey team were requested to observe as well as inquire from the inhabitants for the presence or absence of abaca in all islands surveyed. In addition, we referred to the report on the land area planted with abaca prepared by the Provincial Agriculturist, Jolo, Sulu, 1962 and the report submitted by the Bureau of Census and Statistics(6).

Results

Because of the well-established association of the principal filarial vector with abaca plantations, an extension of the analysis was made to see if this will be reflected in the prevalence rates found in the survey. As one gets familiar with the different islands of the Sulu Archipelago, one sees that extent of abaca planting varies from one island to another. There are municipalities with no abaca plantation at all. Is the variation consistent with the levels of endemicity observed in the islands surveyed? Available data seem to point that it does. Table XII and Figures 13 and 14 provide documentations for this epidemiological fact — depicting somewhat parallel trends in microfilaremia and extent of farm land utilization for abaca.

Discussion

Although there is that trend of increasing microfilaremia rates with large areas of farm land planted with abaca, one may not quite agree in the case of Balimbing. This municipality which falls under the grouping of 1-10 hectares with abaca per 1000 hectares of farm area, has a prevalence rate of 10.26 per cent as against 11.45 per cent for Jolo island. However, this prevalence figure for Jolo is the total for the 8 municipalities whereas that of Balimbing is for a single municipality only. There were only 3 municipalities in Jolo island with prevalence rates ranging from 3.4 to 6.2 per cent, the rest of the municipalities had prevalence rates ranging from 13.7 to 17.3 per cent(5). It is very evident though that in localities where abaca is not grown, filariasis is also absent or if present, the prevalence rate is quite low. Then there is also that possibility, as in the case of Tandu' Bas, wherein families commute from Tandu' Bas to north Tawi-Tawi island to work in an agricultural farm and could have gotten the infection in there.

Summary and Conclusion

The data gathered on the prevalence rates of filariasis in the different islands of Sulu Province and the areas of farm lands planted with abaca seemed to establish the trend of increasing microfilaremia rates in islands where large areas of farm land is planted with abaca. In places where abaca is not grown, filariasis is absent or if present, the prevalence rate is relatively low.

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Table XII. Filariasis Prevalence in Islands at Different Levels of Farm Land Utilization for Abaca Planting.

Hectares with Abaca per 1000 Hectares of Available Farm Area

1		1-10		10 & Over	
Tandu' Bas	: 0.50%	Balimbing	: 10.26%	Jolo Island	: 11.45%
Tapul	: 0.65	Pata	: 5.06	Siasi	: 3.72
Pangutaran	: 0.0	Bongao	: 3.78		
Simunul	: 0.0				
Sitangkai	: 0.0				

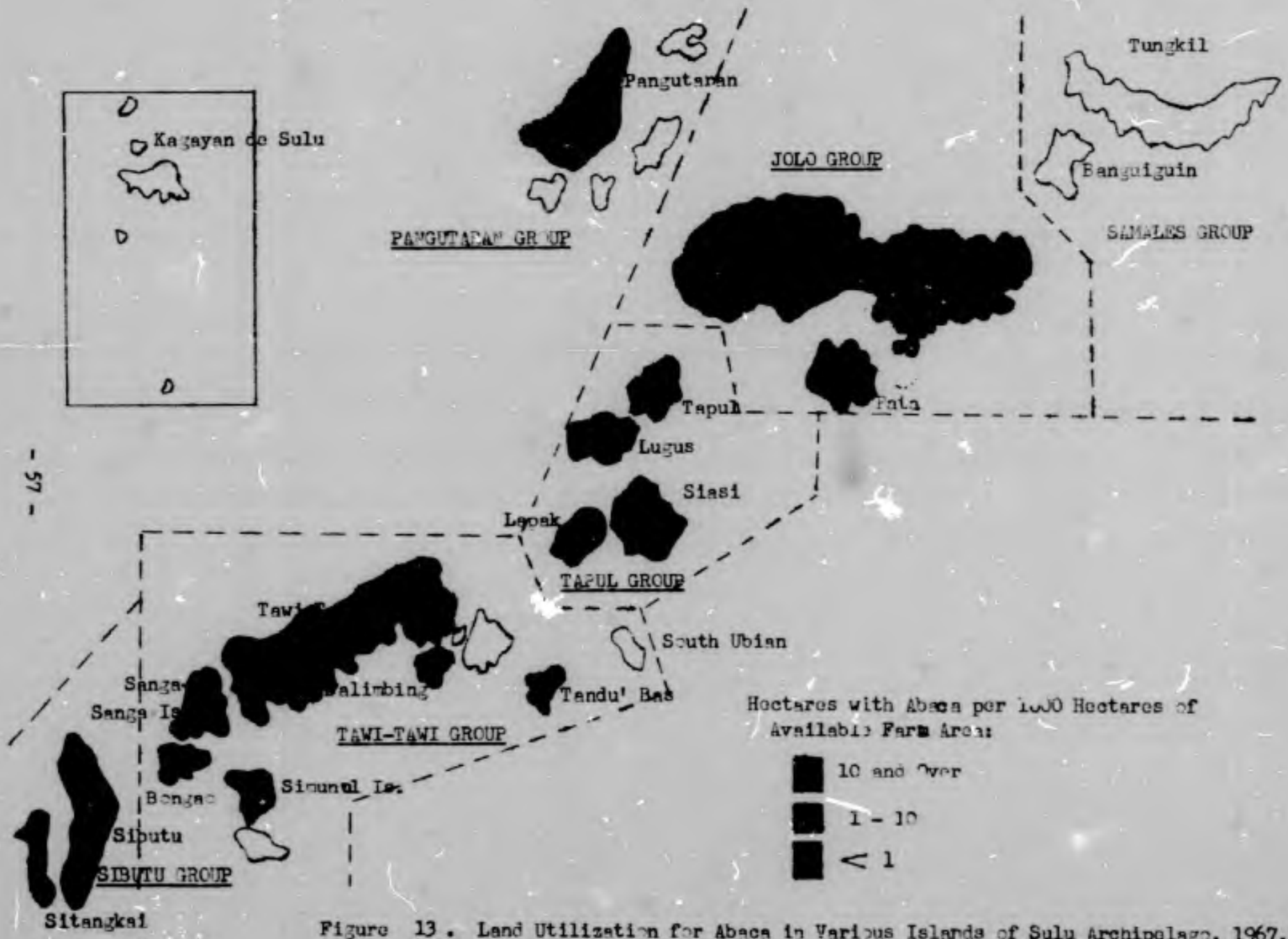


Figure 13. Land Utilization for Abaca in Various Islands of Sulu Archipelago, 1967.

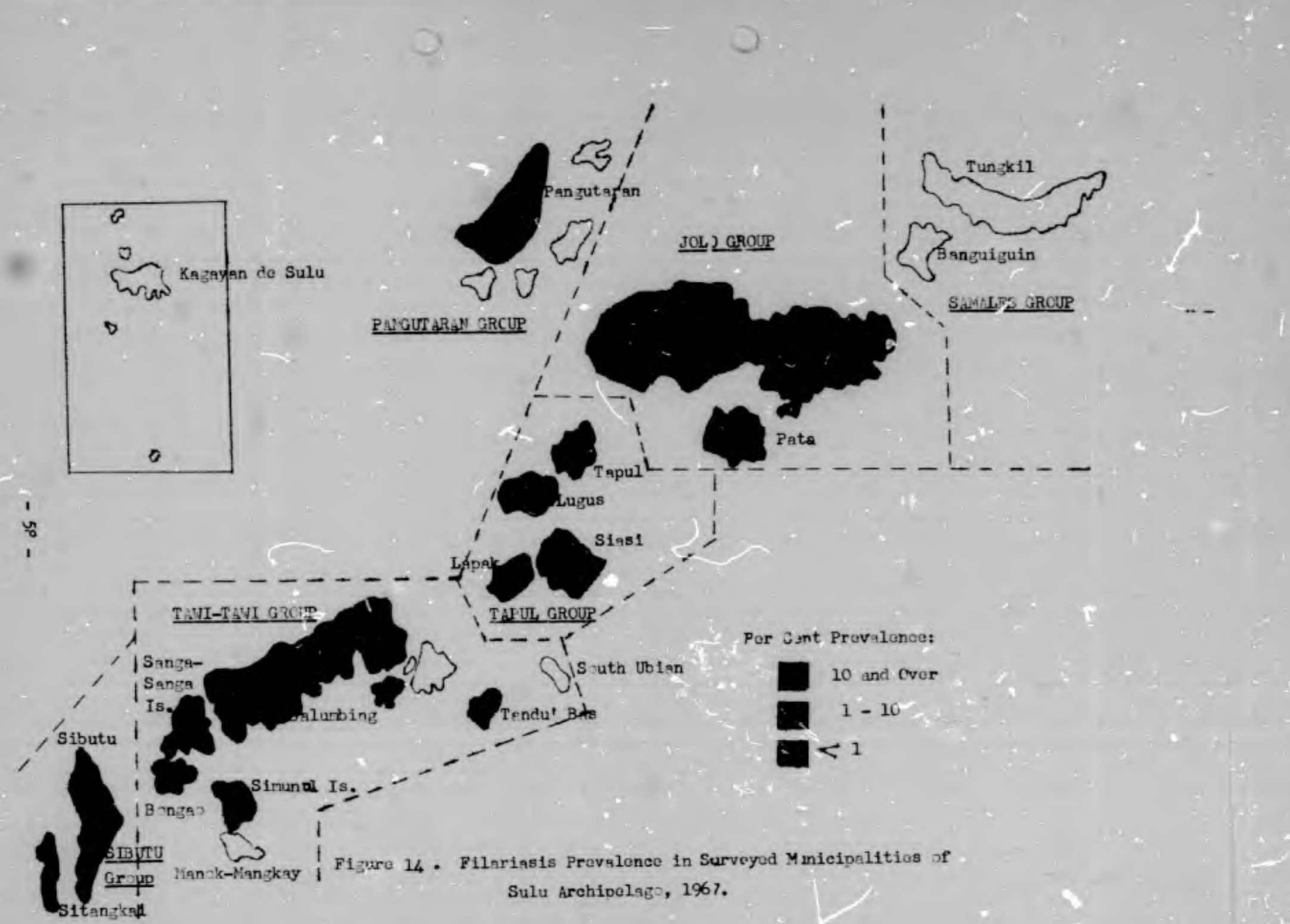


Figure 14. Filariasis Prevalence in Surveyed Municipalities of Sulu Archipelago, 1967.

III. Attempt to explain the unusual appearance of the sheath of W. bancrofti microfilariae when stained with dilute Giemsa.

Ordinarily the sheath of the microfilaria of Wuchereria bancrofti is not apparent or visible when stained with dilute Giemsa. Sometimes the sheath may become visible only on deliberate overstaining with Giemsa stain. The microfilarial sheath of Brugia malayi on the other hand would stain bright pink when stained with Giemsa, a feature used by Wilson in distinguishing B. malayi microfilaria from W. bancrofti(1). There is no doubt that the usefulness of this method of species determination by color contrast as described by Wilson becomes evident when one makes blood surveys in areas where both bancroftian and malayan filariasis are endemic as in the case of West Palawan(7).

If one were to use the color of the sheath alone in distinguishing the microfilariae of W. bancrofti from those of B. malayi especially when using the low power magnification of a compound microscope, the chances of committing an error in the diagnosis becomes great. It is always best to use a combination of color contrasts and morphological differences and better still to resort to Delafield's hematoxylin staining when in doubt of the specific diagnosis.

Materials and Methods

A total of 6 subjects were used in this study. These were previously found positive for W. bancrofti microfilariae whose blood smears were stained with Giemsa (German) diluted with phosphate buffer solution pH 7.2 for one hour and excess stain washed with tap water. Some of the subjects had microfilaria with the sheath stained purple, others the sheath was not visible and the rest had a mixture of these two types of microfilaria.

From each subject we took nine thick blood smears approximately 20 mm^3 in separate glass slides not earlier than 2000 hours. The smears were dried overnight (approximately 10-12 hours), and were stained with three different makes or brands of Giemsa stain (American, German and Japanese) using three different methods of dilution and washing of excess stain after staining. In all the three brands of Giemsa stain we used the proportion of one drop of the stain to one ml. of phosphate buffer solution pH 7.2 and stained for one hour, without previous dehemoglobinization.

The methods of dilution and washing on the three brands of Giemsa stains were as follows: 1. stain diluted with phosphate buffer solution and later washed with tap water; 2. stain diluted with tap water and later washed with tap water; 3. stain diluted with phosphate buffer solution and later washed also with phosphate buffer.

After drying the stained smears, one of us outlined the portion of the smear that stained blue or bluish which is usually the thicker and central portion of each smear with a sharp dissecting needle. Hence the blue portion

of each smear was delimited from the pinkish (usually the thinner and marginal) portion of each smear. The next step was to make counts of those microfilaria with purple sheaths and those without in both zones of each blood smear.

Results and Discussions

To facilitate presentation and analysis, the following symbols are used:

For stains: A - American
G - German
J - Japanese

For different procedure of dilution and washing:
BW - stain diluted with phosphate buffer solution and slide washed with tap water.
WW - stain diluted with tap water and slide washed with tap water.
BB - stain diluted with phosphate buffer solution and slide washed with buffer solution.

For smear thickness: t_1 = thin smear

t_2 = thick smear

Table XIII shows the observations recorded from the experiment. It was decided at the outset of this investigation that the percentage of microfilariae showing visible purplish sheaths to the total number of larvae counted will be used as the observational variable. The problem then was to find out if the factors of types of stain, procedures in dilution and washing, and combinations thereof, influence the appearance or the relative frequency of appearance of purplish sheaths in the slides. If the results show for instance that there is a consistently higher percentage for the A stain over all other stains under similar dilution and washing conditions, then it would be justifiable to assume that this factor (type of stain) is associated with the appearance of the purplish sheath. A preliminary inspection of the data leads one to the following impressions: That if atypical microfilariae are present in a given slide, they are more likely to show upon examination of the thin smear rather than the thick portion of the smear. There are many instances where no microfilariae of this type were detected in the thick smear, yet, relatively high percentages were observed for the corresponding thin smears. Proportions for the thin part of the slides are consistently above those of the thick parts. Not one stain seems to have a monopoly on the appearance of these unusual larvae either, since no marked concentrations could be discerned for any of the three brands studied.

Proceeding to more formal analysis, we may regard the experimental plan as a $3^2 \times 2$ factorial, with 2 factors or "treatments", (stain type and dilution and washing procedures) each at three levels and one at two levels (thick and thin smears). The necessary percentages were computed for each cell in Table XIII. Two difficulties were inherent in the basic data. These

form the major limitations of the report and render the subsequent analysis necessarily approximate.

First, the subjects differed widely in their microfilarial densities, thereby compounding the problem of variance heterogeneity, since the percentages will be based on unequal numbers. The subjects RK and MU in particular showed the extremes here, with the former yielding a count as low as zero and the latter, counts in the neighborhood of 240 microfilariae.

Second, the presence of two zero counts for both typical and atypical microfilariae in one subject (RK). This rendered the corresponding percentages indeterminate.

Variance heterogeneity can be rectified to a satisfactory degree by the angular transformations for percentages, thereby increasing the validity of the usual analysis of variance techniques. However, the effect of this transformation for percentages based on unequal numbers for factorial designs is quite complicated although Cochran[3] indicated that for such experimental arrangements, the usual unweighted analysis of variance is more attractive on practical as well as arithmetical grounds. Regarding the zero counts, two alternatives seem to be workable. First, is to disregard the indeterminate cases altogether. This however, introduces further imbalance of the results. Another procedure which was actually used is based on the observation that total microfilarial count for each treatment combination seem to be at comparable levels within patients. Thus we may with some justification, substitute the average count of the 9 slides coming from each patient, for the zeros. The assumption is that this number might have been detected if a more thorough search had been made.

Table XIV shows the results following the application of angular transformation and corrections for discontinuity suggested by Bartlett[4]. To facilitate analysis Table XV of subtotals was also constructed.

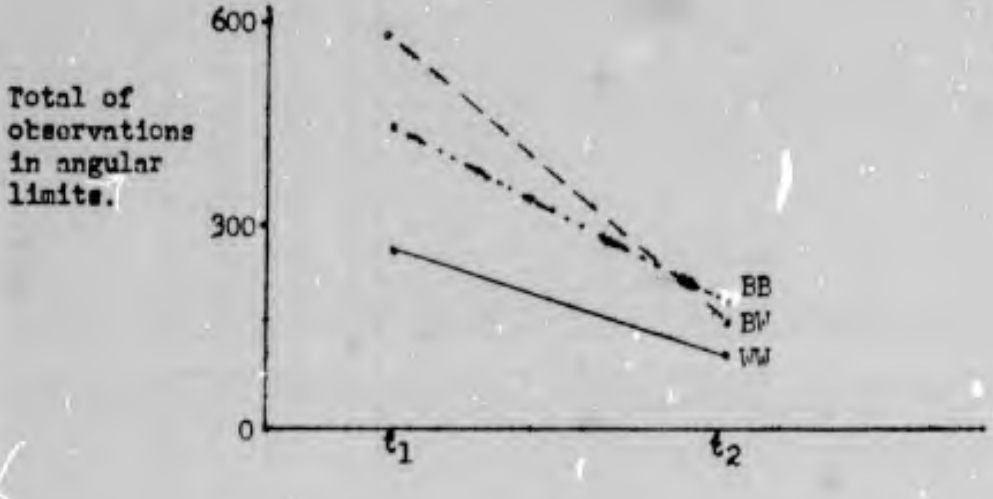
Application of the usual formulas to the transformed values yielded the following ANOVA table;

Source of Variation	DF	SS	MS	F
Replications (patients)	5	1,469.60	293.92	1.26
Treatment combinations	17	14,594.54	858.50	3.68**
Stains	2	1,306.48	653.24	2.80
Dilution and Washing procedure	2	1,988.08	994.04	4.26*
Smear	1	6,647.39	6,647.39	28.48**
Stains X Dil. & Washing procedure	4	2,699.07	674.77	2.89
Stains X Smear	2	156.29	78.14	0.33
Dil. & Wash. Proc. X Smear	2	1,687.22	843.61	3.61*
Stains X dil. & Wash. Proc. X Smear	4	110.01	27.50	0.12
Error	85	19,838.95	233.40	
TOTAL	107	35,903.09		

* Significant at the 5% level.
** Significant at the 1% level.

The F-tests verifies the preliminary impressions above concerning the influence of stains, although there is a mild suggestion of differences between these brands, since an F value of 2.80 is not exactly small. Two main effects and one interaction term turned out to be significant. These are dilution-washing procedures and thickness of smear for the former and dilution-washing X smear interaction for the latter. Thus differences occur between the 3 dilution-washing procedures. Looking back at the original observations, it can be seen that the use of water for both dilution and washing is least favorable for the demonstration of purple-sheathed microfilariae.

There is a definite superiority of thin smears over thick smears, for purposes of detection and possibly species diagnosis. Perhaps, there is nothing surprising about this since the thickness of the smear affects light and color contrasts, Note that the same main effects are involved in the lone interaction term which turned out to be significant. This implies that differences between dilution-washing procedures varies with the thickness of the smear. Alternatively, the differences among the thick and thin portions of the slide in so far as demonstration of atypical microfilariae is concerned, varies with the dilution-washing procedure employed. This is graphically portrayed below, showing that the interaction involves changes in magnitudes of differences between dilution-washing procedures at the two thickness levels. At t_2 there is even an interchange in relative positions of BB and BW.



Summary and Conclusions

The staining experiment was an attempt to study and possibly identify some factors which may have influenced the unusual appearance of purplish sheath of W. bancrofti in stained smears. The results showed, subject to the limitations cited in connection with the basic data, that no significant differences in the appearance of the purplish sheath could be ascribed to the variety of Giemsa staining solutions used (American, German and Japanese). The use of tap water as both stain diluent and for washing of slides appears to be least favorable for the demonstration of this unusual staining reaction. On the other hand, the likelihood of detecting purplish sheaths is highest at the thin sections of smears that were stained with Giemsa diluted with buffer solution and subsequently washed with tap water.

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Table XIII. Proportion* of Microfilariae with Purple Sheaths and Microfilariae with Non-Visible Sheaths Counted in Thin and Thick Smears in Slides Subjected to Various Staining

		S T A I N								
		American			German			Japanese		
Subjects	Smear	WW	BW	BB	WW	BW	BB	WW	BW	BB
RK	t_1	0/4	0/7	0/3	3/8	0/0	5/12	0/0	1/5	1/5
	t_2	0/8	0/15	I/16	0/5	0/7	0/7	0/5	0/13	0/5
MU	t_1	0/9	0/17	0/9	0/8	6/10	15/15	0/14	0/6	0/47
	t_2	0/13	0/54	0/55	0/61	1/4	2/3	0/66	0/102	0/42
TA	t_1	0/9	0/17	0/9	0/8	6/10	15/15	0/14	0/6	0/47
	t_2	0/13	0/54	0/55	0/61	1/4	2/3	0/66	0/102	0/42
SM	t_1	24/28	10/11	7/32	0/13	4/12	3/17	0/24	13/20	1/22
	t_2	1/26	0/9	0/57	0/64	0/71	0/12	0/29	2/13	0/25
SO	t_1	15/66	7/18	3/32	0/59	1/50	1/53	0/66	26/33	0/67
	t_2	0/220	0/86	0/73	0/111	0/126	0/55	0/62	0/60	0/60
SS	t_1	3/9	1/5	1/21	0/20	1/6	1/16	0/22	3/8	0/21
	t_2	0/5	0/34	0/35	0/36	0/44	0/56	0/9	0/38	0/12

* The upper figure is the number of atypical purple-sheathed microfilaria counted while the lower figure refers to the number of typical microfilaria detected upon microscopic examination.

Table XIV. Angular Transformation of Percentage of Atypical Purple-Sheathed Microfilaria.

Subject	Smear	S T A I N								
		American			German			Japanese		
		WW	BW	BB	WW	BW	BB	WW	BW	BB
RK	t_1	14.5	10.9	16.7	37.9	14.5*	40.2	14.5*	26.6	26.6
	t_2	10.2	7.4	7.2	13.0	10.9	10.2	23.0	8.0	13.0
MU	t_1	2.6	16.8	25.5	13.7	70.0	73.0	3.2	30.0	33.2
	t_2	1.6	1.9	7.3	1.5	42.0	37.7	2.2	2.2	5.1
TA	t_1	9.6	7.0	9.6	10.2	51.0	82.8	7.7	11.8	4.2
	t_2	8.0	3.9	3.9	3.7	30.0	54.8	3.5	2.8	4.4
SM	t_1	67.6	72.2	28.0	8.0	35.3	24.9	5.9	54.0	12.2
	t_2	11.2	9.6	3.8	3.6	3.4	8.3	5.3	23.2	5.8
SO	t_1	28.5	38.5	17.9	3.9	8.1	7.9	3.5	62.6	3.5
	t_2	1.9	3.1	3.3	2.7	2.6	3.9	3.7	3.7	3.7
SS	t_1	35.4	26.6	12.6	6.4	24.1	14.5	6.1	37.8	6.2
	t_2	12.9	4.9	4.8	4.8	3.5	3.8	9.6	4.6	8.1

Table XV. - Total of Transformed Values for Various Treatment Combinations.

Stain	Smear	WW	BW	BB	Total WW+BW+BB
A	t_1	158.2	172.0	110.3	440.5
	t_2	45.8	30.8	30.3	106.9
	$t_1 + t_2$	204.0	202.8	140.6	574.4
G	t_1	80.1	203.0	243.3	526.4
	t_2	29.3	92.4	118.7	240.4
	$t_1 + t_2$	109.4	295.4	362.0	766.8
J	t_1	40.9	222.8	85.9	349.6
	t_2	37.3	44.5	40.1	121.9
	$t_1 + t_2$	78.2	267.3	126.0	471.5
TOTAL	t_1	279.2	597.8	439.5	1316.5
	t_2	112.4	167.7	189.1	469.2
	$t_1 + t_2$	391.6	765.5	628.6	1785.7

IV. An attempt to determine the mosquito vector of filariasis in Southern Sulu.

The vector of bancroftian filariasis in areas with plenty of abaca plantations such as Jolo island is Aedes (Finlaya) poecilus[1]. In the municipality of Bongao, although there are no abaca plantations, yet both species of human filaria are endemic. It would be of interest to find out what mosquitoes are transmitting the bancroftian and malayan filariasis in this area.

Our finding of a subperiodic behavior of the microfilariae of B. malayi in this survey have more or less given us some clue that probably the vector would be Mansonia bonneae as we found it to be the vector of malayan filariasis in Palawan[2]. Likewise our previous findings in Bontoc, Mountain Province and in Palawan Province is that Anopheles minimus flavirostris is the vector for bancroftian filariasis in these areas[3,4]. Both places incidentally do not grow abaca for industry. Again we were guided by these findings inasmuch as Bongao did not grow abaca either yet W. bancrafti filariasis is endemic.

Materials and Methods

Mosquito collection and dissection:

In order to determine the most probable mosquito vector of filariasis in these islands particularly in the municipality of Bongao, we collected mosquitoes for dissection. We used a carabao-baited trap in the collection of mosquitoes and in few instances we tried collection off-humans. The trap was set outdoor ideally near a group of houses where a member of the household was found with microfilaremia. The door to the trap was left open during the night baited with a carabao. Very early the next morning the carabao was removed, and the door of the trap was closed. Mosquitoes were gathered by means of a glass-tube aspirator and kept in separate cardboard containers. Mosquitoes were removed from these containers in batches of 12-15, anaesthetized with chloroform and sorted according to species under a stereoscope. After discarding the legs and wings, the head and thorax were teased separately in two drops of saline on a glass slide. We did not bother to dissect the abdomen because all mosquito specimens were fully engorged with blood. The head and thorax were examined carefully for filaria larvae using a compound microscope to detect stage I larvae and stereoscope for detecting stages II and III. For the correct species identification of the larvae, a compound microscope was always used.

Results

A preliminary survey of the area where mosquito trappings were done revealed the absence of abaca plants. The topography of the place seemed very ideal for our malaria vector mosquito, Anopheles minimus flavirostris because of the existence of foothill streams. The place is highly malarious since

several members of the household were found sick with malaria. There were swamps in the vicinity which unfortunately were dry at the time of the survey because it was dry season then.

The results of mosquito collections and dissections are shown in Tables 16 (a) and 16 (b). In Table 16 (a), a total of 4,288 mosquitoes were dissected which were collected from a carabao-baited trap. In two occasions we tried collecting mosquitoes off-human and a total of 90 mosquitoes were caught and dissected, but all were found negative for filaria larvae. The sum of the two sources of mosquito collections gave a total of 4,378 mosquitoes which were all dissected. There were 48 mosquitoes found harboring some filaria larvae, five of which were Anopheles franciscoi, a member of the barbirostris group, 40 Anopheles minimus flavirostris and 3 unidentified Aedes. The filaria larvae found in Anopheles franciscoi and in Aedes (sp)? were definitely not of human origin. Their appearance conformed with the morphological characteristics of Dirofilaria immitis. Out of the 150 Mansonia mosquitoes dissected, 3 were M. bonneae and 78 M. dives but not a single infection was found.

Of 487 Anopheles minimus flavirostris 40 or 8.2 per cent were found harboring larvae. The details of these infections are presented in Table 16 (b). Out of the 40 mosquitoes found harboring filaria larvae, one harbored a larva which is probably that of a Nematode worm. We were unable to identify this larva. Of the remaining 39 mosquitoes, 35 or 90 per cent harbored larvae of W. bancrofti and 3 or 7.7 per cent harbored larvae of B. malayi and one mosquito harbored larvae of Dirofilaria immitis.

Twenty-two of the 39 mosquitoes had stage I of W. bancrofti in the thorax, 8 harbored stage II also from the thorax and 5 mosquitoes harbored stage III of W. bancrofti. Two of these 5 mosquitoes with stage III larvae had the larvae from the head and three from the thorax.

Two of the three mosquitoes harboring B. malayi had stage I from the thorax and one stage II from the thorax.

Discussion

Although the primary aim in the mosquito collection and dissection in this area was intended to determine the probable vector of malayan filariasis more than the determination of the factor of bancroftian filariasis, the results of the mosquito collection pointed to a high density of Anopheles minimus flavirostris and low densities for Aedes poecilus and the Mansonia bonneae, respectively. The presence of swampy areas with some forms of vegetation ideal for Mansonia mosquito breeding did not increase the mosquito density particularly the Mansonia due to the dry season, at the time of survey. Hence, although we suspected either Mansonia bonneae or M. dives as probable vectors of malayan filariasis based on the subperiodic microfilarial behavior, we could not catch enough samples to substantially incriminate the vector species.

Anopheles franciscoi was the most abundant mosquito collected at the time of survey but unfortunately the 5 positive mosquitoes dissected turned out to be larvae of Dirofilaria immitis. This mosquito have been incriminated as the vector of nocturnal periodic malayan filariasis in Malaysia(5).

It is evident from the results of dissection of Anopheles minimus flavirostris, as shown in Table XVI (b) that this mosquito is the vector of bancroftian filariasis in this particular area of Sulu province, where abaca is absent. Such results confirm our previous finding in Bontoc, Mt. Province(3) and in Palawan(4) where abaca was not grown.

There were 7 mosquitoes that harbored a total of 47 stage II larvae of W. bancrofti and 5 mosquitoes that harbored a total of 19 stage III larvae, also of W. bancrofti.

In barrio Masantong and Tingol-tingol both of Bongao municipality where our traps were set, the two species of human filaria are present and hence it did not surprise us a bit to encounter early stages of B. malayi, in Anopheles minimus flavirostris. The fact that we did not find 3rd stage larvae of B. malayi may be interpreted as an indication that this mosquito is not the natural vector for malayan filariasis. The very low intensity of microfilaremia in B. malayi among our subjects could mean that transmission is dependent upon the abundance of rain to enable breeding of Mansonia in the swampy areas previously mentioned above.

Summary and Conclusion

The vector of W. bancrofti filariasis in 2 barrios of Bongao municipality wherein abaca was not grown is Anopheles minimus flavirostris. This finding confirms previous findings in Mt. Province and Palawan Province where abaca was not also grown. There are therefore two species of mosquitoes transmitting bancroftian filariasis in Sulu Province. In areas where abaca was grown the vector is Aedes (Finlaya) poecilus while in areas where abaca was not grown the vector is Anopheles minimus flavirostris.

In this survey we failed to determine the mosquito vector of B. malayi filariasis because the mosquito survey work was done during the dry season. However, we hgihly suspect either Mansonia bonneae or Mansonia dives as probable vectors based on the microfilarial periodicity and the presence of fresh water swamps which were dry at the time of the survey.

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Table XVI (a). Natural Infections in Mosquitoes, Bongao, Sulu, 1967.

SPECIES	No. Dissected: Carabao Baited	No. Dissected: Off Human	No. Infected	Total No. Dissected
Anopheles franciscoi	2,330	1	5[a]	2331
" kochi	99	2	-	101
" litoralis	4	-	-	4
" manalangi	178	-	-	178
" minimus flavirostris	486	1	40[b]	487
" subpictus indefinitus	23	1	-	24
" tessellatus	57	4	-	61
" vag. limosus	144	-	-	144
Aedes albopictus	7	4	-	11
" lineatopennis	-	5	-	5
" nubiculus	26	26	-	52
" niveus	-	1	-	1
" pampangensis	123	-	-	123
" poecilus	29	6	-	35
" vexans nocturnus	93	11	-	104
" aedes (sp) ?	280	10	3[c]	290
" (Mucidus) aurantius	44	1	-	45
Armigeres baisasi	12	1	-	13
" malayi	31	1	-	32
Mansonia bonneae	3	-	-	3
" dives	76	2	-	78
" ochracea	65	-	-	65
" uniformis	1	3	-	4
Culex bitaeniorynchus	18	-	-	18
" gelidus	28	-	-	28
" summorosus	64	9	-	73
" whitmorei	1	-	-	11
" sinensis	-	1	-	1
(sp) ?	104	-	-	104
TOTAL	4,288	90	48	4,378

a. All are larvae of Dirofilaria immitis.
b. See Table XVI (b) for details.
c. All are larvae of Dirofilaria immitis.

Table XVI (b). Natural Infections with Filaria in *Anopheles minimus flavirostris*, Bongao, Sulu, 1967.

Specimen No.	Larval Stage	Site	No. Larvae Found	Filaria Species
1	1st	thorax	1	W. bancrofti
2	1st	thorax	4	W. bancrofti
3	1st ?	thorax	1	Nematode larva?
4	1st	thorax	1	W. bancrofti
5	1st	thorax	1	W. bancrofti
6	2nd	thorax	19	W. bancrofti
7	1st	thorax	54	B. malayi
8	2nd	thorax	2	W. bancrofti
9	1st	thorax	4	W. bancrofti
10	3rd	head & proboscis	5	W. bancrofti
11	1st	thorax	4	W. bancrofti
12	1st	thorax	15	W. bancrofti
13	3rd	thorax	3	W. bancrofti
14	1st	thorax	3	W. bancrofti
15	1st	thorax	7	W. bancrofti
16	1st	thorax	45	W. bancrofti
17	3rd	head	1	W. bancrofti
18	1st	thorax	3	W. bancrofti
19	1st	thorax	1	W. bancrofti
20	3rd	thorax	5	W. bancrofti
21	1st	thorax	1	W. bancrofti
22	1st	thorax	14	W. bancrofti
23	1st	thorax	17	W. bancrofti
24	2nd	thorax	11	W. bancrofti
25	2nd	thorax	1	W. bancrofti
26	2nd	thorax	3	W. bancrofti
27	1st	thorax	4	W. bancrofti
28	1st	thorax	1	W. bancrofti
29	1st	thorax	10	W. bancrofti
30	3rd	thorax & head	2	D. immitis
31	2nd	thorax	1	W. bancrofti
32	2nd	thorax	10	W. bancrofti
33	3rd	thorax	5	W. bancrofti
34	1st	thorax	2	W. bancrofti
35	1st	thorax	1	W. bancrofti
36	1st	thorax	8	B. malayi
37	2nd	thorax	3	B. malayi
38	1st	thorax	4	W. bancrofti
39	1st	thorax	4	W. bancrofti
40	2nd	thorax	1	W. bancrofti

V. An attempt to develop serologic tests for the diagnosis of filariasis.

It is recognized that microfilariae may not be demonstrated in the blood of human cases where any of the following conditions exists: 1) in case of single sex infection; 2) where living adult males and females are not in the same lymph gland; 3) where there is occlusion of the lymph channels inhabited by fertile females so that microfilariae do not reach the blood circulation; and 4) when the adult filariae are already dead(1). In view of these, indications for serologic tests or lymph gland biopsy often arise. A serologic test is easier to perform than a biopsy and causes less inconvenience to the patient.

Studies were done to determine if it is worthwhile to pursue extensive evaluation of serologic tests using D. immitis antigens for the diagnosis of bancroftian and malayan filariasis since antigens of D. immitis are identical or partially related to those of W. bancrofti(1) and probably to B. malayi. However in the Philippines infections with D. immitis is very common among dogs so that human population maybe sensitized with dirofilaria antigens as a result of bites of mosquitoes. Passive-hemagglutination (HA) test and soluble-antigen-fluorescent antibody (SAFA) tests(2) using antigens of adults and microfilariae of D. immitis were performed on serums of 33 human cases of filariasis and 24 non-infected individuals. By undertaking these observations we hoped to get indications of the proportion of humans who develop serologic response to D. immitis as a result of exposures to bites of mosquitoes harboring the infective larval stage of D. immitis and how the titer of circulating antibodies due to this sensitization compare to that of humans infected with bancroftian or malayan filariasis. If a significant portion of the population have significant titers of circulating antibodies against Dirofilaria antigens, then said antigens cannot be used for serologic diagnosis of bancroftian and malayan filariasis. In such circumstance, purified antigens of W. bancrofti or B. malayi have to be utilized to minimize cross reactions and false positives.

Materials and Methods

Adults and microfilariae of D. immitis were obtained from infected dogs. After recovery from the dogs' heart, the adults were washed in several changes of cold distilled water, lyophilized and homogenized in normal saline. Extraction of soluble antigens was done overnight with constant stirring in a refrigerator. The following morning, the mixture was centrifuged in the cold and the resulting supernate collected and used as antigen. Citrated peripheral blood of infected dogs was centrifuged to separate formed elements from the plasma portion. Red cells were subsequently lyzed in several changes of cold distilled water. After washings in normal saline, the cell debris and microfilariae (a good number of microfilariae are caught in the debris), was incubated in 1:10,000 saponin in a water bath for ten minutes. The mixture was then centrifuged and the sediment washed in saline. After washing, the sediment was teased to free the microfilariae. The microfilariae were separated from cellular debris and fibrin by differential centrifugation. The collected microfilariae were ground in saline and antigens extracted overnight in a refrigerator. The supernate resulting after centrifuging in the cold was collected and used as antigen. Prior to use for hemagglutination tests or for SAFA the

optimal concentration for use of each antigen was determined by block titration.

The procedure of the passive hemagglutination is a modification of the original technic described by Boyden[3] and adopted to a number of parasitic infections[4]. Sheep's red cells were washed in four changes of pH 7.2 phosphate buffered saline and finally suspended to make a 2.5% cell suspension. The cells were tanned by mixing an aliquot of the cell suspension with an equal volume of 1:100,000 dilution of tannic acid. The mixture was placed in an ice cold bath for 15 minutes and periodically stirred. After the tanning the cells were washed and resuspended in normal saline. Sensitization with filarial antigens was accomplished by incubating equal volumes of tanned cell suspension and antigen at optimal dilution with pH 6.4 phosphate buffered saline in a water bath at 37° C for 15 minutes. After sensitization the cells were washed in two changes of saline and finally suspended in pH 7.2 phosphate buffered saline with 0.6% normal rabbit serum.

The hemagglutination test was run in microtiter plates. Serial four-fold dilution of the test serum were made after which a drop of the antigen sensitized cells was added. The plates were vibrated for 10 minutes and allowed to stand for at least two hours before results were read. With every run we indluded known positive and known negative controls.

Extracts of adults and microfilariae diluted with TRIS buffered saline pH 8 were used as antigen for the SAFA tests. Cellulose acetate filter paper discs (Millipore paper) were immersed vertically in the antigen dilution for 15 seconds and allowed to dry. The discs are incubated in 1:4 dilution of the test serum in a tube for 45 minutes after which they were washed in 3 changes of buffered saline. They were then incubated in fluorescein-conjugated rabbit anti-human globulin for 30 minutes and again washed in buffered saline. After drying, the discs are mounted on vinyl plastic electrical tape which has minimal fluorescence. In running the test, a control disc is run with every serum. The control disc is mounted before its corresponding test disc. The tape is placed on the drum of the chromatograph door of a fluorometer to determine objectively the degree of fluorescence. Tests results were read on the dial of a Model III fluorometer (G.K. Turner Assoc.) with a primary filter transmitting 254-420 mu and a sharp cut secondary filter passing greater than 520 mu in combination with a 10% neutral density filter. The dial of the fluorometer was zeroed with the control disc prior to reading the antigen disc. In relation to the activity of the test serums, fluorometer dial readings were interpreted as follows: readings less than 15, non-reactive; 15-19 weakly reactive; and 20-39 reactive; greater than 39, highly reactive[5].

Results and Discussions

Table XVII to XXI present the results of passive-hemagglutination and SAFA tests on the serums of human cases of filariasis and non-infected individuals.

As shown in Tables XVII and XIX, with 2 plus agglutination in a minimal test serum dilution of 1:16 as criterion for positive reaction, 28 of the 33 cases or 84.84% could be considered positive by HA with adult dirofilaria antigens. However 16 of the 24 non-infected individuals, as shown in Tables XVIII and XIX, or 66.66% can also be considered as positives indicating a very large proportion of false positive reactions (referring to bancroftian or malayan filariasis) indicating that D. immitis adult antigens can not be used for diagnosis of human filariasis in the Philippines.

The results of SAFA tests using soluble antigens of adults and microfilariae of D. immitis practically parallel those of passive-hemagglutination. As shown in Tables XX and XXI, although all 33 human cases or 100% show high reactivity by SAFA test using adult dirofilaria antigens; 70.83% of non-infected individuals were positive by the test. With dirofilaria microfilaria antigens, also 70.83% of non-infected humans were also positive by SAFA tests.

The results of both hemagglutination and SAFA tests indicate that although D. immitis antigens can be used in serologic diagnosis of bancroftian filariasis, a very high percent of false positives would be encountered in the Philippines. As mentioned earlier, under such circumstances only purified antigens of W. bancrofti or Brugia malayi can be utilized for serologic diagnosis of human filariasis. Although it is possible to infect animals and recover adult filarid worms in the case of B. malayi, the recovery of adults of W. bancrofti for preparing antigens is extremely difficult. In view of this in so far as W. bancrofti is concerned microfilariae would be easier to obtain as source of antigens. The results of SAFA tests using microfilariae of D. immitis for antigen indicate that microfilariae of W. bancrofti can also be utilized for the test. We were able to obtain very limited amount of W. bancrofti microfilariae which we tried for SAFA test on the serums of the 33 cases of human filariasis. The results are rather inconclusive which are explained as due to our failure to titrate or determine the optimal concentration of the antigen for use in the test since we have very little antigen to work with. However, we feel that the use of W. bancrofti microfilariae for SAFA tests should be further studied.

Summary and Conclusion

Results of passive hemagglutination and SAFA tests on serums of 33 humans with filariasis and 24 non-infected individuals in the Philippines, using antigens of adults and microfilariae of D. immitis indicate as high as 70.83% false positives (in reference to bancroftian filariasis) would be encountered. This indicates that W. bancrofti antigens be utilized for serologic diagnosis of human filariasis in areas where level of endemicity of D. immitis infections in dogs is high. Results of SAFA tests using soluble antigens of D. immitis microfilariae suggests that W. bancrofti microfilariae can be used as antigen for SAFA tests and their use should be studied.

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Table XVII. Results of Hemagglutination Tests and Soluble-Antigen-Fluorescent Antibody Tests on Serums of Human Cases of Filariasis.

Case No.	HA/Adult[@] Dirofilaria Antigen	Soluble-Antigen-Fluorescent-Antibody Technic[@@] Adult Dirofilaria Antigen	Dirofilaria Microfilaria Antigen	W. bancrofti Microfilaria
1.	1:64	highly reactive	highly reactive	highly reactive
2.	1:16	" "	" "	reactive
3.	1:256	" "	" "	highly reactive
4.	neg.	" "	" "	non-reactive
5.	1:256	" "	" "	highly reactive
6.	neg.	" "	" "	" "
7.	neg.	" "	" "	non-reactive
8.	1:64	" "	" "	reactive
9.	1:4	" "	" "	non-reactive
10.	1:64	" "	" "	non-reactive
11.	1:64	" "	" "	reactive
12.	1:64	" "	" "	highly reactive
13.	1:16	" "	" "	" "
14.	1:16	" "	" "	reactive
15.	1:64	" "	reactive	non-reactive
16.	1:16	" "	highly reactive	highly reactive
17.	1:64	" "	" "	" "
18.	1:256	" "	" "	reactive
19.	1:256	" "	" "	highly reactive
20.	1:16	" "	" "	" "
21.	1:64	" "	" "	" "
22.	1:64	" "	" "	" "
23.	1:16	" "	highly reactive	" "
24.	1:64	" "	reactive	weakly reactive
25.	1:64	" "	highly reactive	reactive
26.	1:16	" "	" "	highly reactive
27.	1:16	" "	" "	" "
28.	1:64	" "	" "	" "
29.	1:4	" "	non-reactive	non-reactive
30.	1:256	" "	highly reactive	highly reactive
31.	1:64	" "	" "	reactive
32.	1:16	" "	" "	non-reactive
33.	1:64	" "	" "	non-reactive

Notes: @ represents the maximum serum dilution with 2-plus agglutination
@@ non-reactive - fluorometer dial reading less than 15
weakly reactive - fluorometer dial reading 15 to 19
reactive - fluorometer dial reading 20 to 29
highly reactive - fluorometer dial reading greater than 29.

Table XVIII. Results of Passive-Hemagglutination Test and Soluble-Antigen-Fluorescent-Antibody-Technic on Serums of Humans Not Infected With Filariasis.

Case No.	HA with[@] Adult Dirofilaria Antigen	Soluble-Antigen-Fluorescent-Antibody-Technic[@@]	
		Dirofilaria Adult Ag.	Dirofilaria Micro. Ag.
1.	1:16	highly reactive	reactive
2.	1:4	reactive	non-reactive
3.	1:4	non-reactive	non-reactive
4.	1:4	highly reactive	weakly reactive
5.	1:16	" "	reactive
6.	1:4	" "	weakly reactive
7.	1:16	reactive	non-reactive
8.	1:16	non-reactive	non-reactive
9.	1:64	non-reactive	weakly reactive
10.	1:16	highly reactive	non-reactive
11.	1:16	reactive	non-reactive
12.	1:64	highly reactive	reactive
13.	1:16	" "	non-reactive
14.	1:4	" "	reactive
15.	1:16	reactive	weakly reactive
16.	1:16	highly reactive	reactive
17.	1:4	highly reactive	weakly reactive
18.	1:16	non-reactive	non-reactive
19.	1:16	highly reactive	non-reactive
20.	1:4	highly reactive	reactive
21.	1:64	non-reactive	weakly reactive
22.	1:16	reactive	reactive
23.	neg.	weakly reactive	non-reactive
24.	1:16	non-reactive	weakly reactive

Note: @ represents maximum serum dilution with 2-plus agglutination.
@@ non-reactive - fluorometer dial reading less than 15
weakly reactive - fluorometer dial reading 15-19
reactive - fluorometer dial reading 20-29
highly reactive - fluorometer dial reading greater than 29.

Table XIX. Comparison of Results and Titers of Passive-Hemagglutination Tests on Sera of Cases of Filariasis and Non-infected Humans.

Diagnostic Status	No. Exam.	Highest Dilution of Serum with 2+plus Agglutination: 1/4 or neg. No.	1/4 or neg. %	1/16 No.	1/16 %	1/64 No.	1/64 %	1/256 No.	1/256 %	Total 1/16 or more No.	Total 1/16 or more %
Cases	33	5	15.15	9	27.57	14	42.42	5	15.15	28	84.84
Not-Inf.	24	8	33.33	13	54.5	3	12.5			16	66.66

Note: Any serum with 2-plus agglutination with a dilution of at least 1/16 is considered positive.

Table XX. Results of SAFA Using Adult D. immitis Antigens on Serums of Cases of Filariasis and Non-infected Humans.

Diagnostic Status	No. Exam.	Non-Reactive No.	Non-Reactive %	Weakly Reactive No.	Weakly Reactive %	Reactive No.	Reactive %	Highly-Reactive No.	Highly-Reactive %	Total Reactive & Highly R. No.	Total Reactive & Highly R. %
Cases	33	0	0	0	0	0	0	33	100	33	100
Not-Inf.	24	6	25.5	1	4.5	5	20.83	12	50	17	70.83

Table XXI. Results of SAFA Using D. immitis Microfilaria Antigens on Serums of Cases of Filariasis and Non-infected Humans.

Diagnostic Status	No. Exam.	Non-Reactive No.	Non-Reactive %	Weakly Reactive No.	Weakly Reactive %	Reactive No.	Reactive %	Highly Reactive No.	Highly Reactive %	Total Reactive & Highly R. No.	Total Reactive & Highly R. %
Cases	33	1	3.33	0	0	2	6.66	30	90.9	32	96.66
Not-Inf.	24	6	25	1	4.16	5	20.83	12	50	17	70.83

D-I-S-T-R-I-B-U-T-I-O-N

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13. ABSTRACT

A total of 3,695 persons were examined for microfilaremia with 120 found positive or a prevalence rate of 3.2 per cent. Out of these 120 positives, 91 or 76 per cent were W. bancrofti and 29 or 24 per cent were B. malayi filariasis. Tawi-Tawi group of islands particularly Bongao municipality became the second endemic focus for malayan filariasis in the Republic of the Philippines. The first one is in Quezon municipality, Palawan Province which was reported in 1964.

As expected the microfilaremia rates in males was higher than in females in practically all ages and in all municipalities found endemic for the disease. The higher the endemicity of filariasis in an area, the closer the prevalence rates between sexes and the lower the endemicity the higher the prevalence rates of males over females. This condition we have explained as probably due to occupational exposure risks.

Malayan filariasis in Southern Sulu appeared to have been introduced relatively quite recent as compared to that found in Palawan Province. The microfilariae exhibited the nocturnal subperiodic behavior similar to the malayan filariasis found in Palawan.

There is that trend of increasing W. bancrofti microfilaremia rates in islands with large areas of farm land planted with abaca, with Aedes (Finlaya) poecilus as the vector mosquito. However, in areas where abaca is absent filariasis may also be absent or if present the prevalence rate is low. Anopheles minimus flavirostris is the vector in abaca-free areas.

The purple sheath of W. bancrofti microfilariae cannot be attributed to the "brand" of Giemsa stain used, but rather to the solution used in dilution of stain and the washing off of excess stain.

The Log-Probit Regression Line as applied in our data is ideal in comparing intensity of infection by microfilarial counts and would be a nice tool in assessing filariasis control programs. (Author)

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