

FILARIAL PARASITES OF THE MONKEYS OF PANAMA

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Within recent years several surveys of the blood parasites of the monkeys of Panama made by Dr. Herbert C. Clark, Director of the Gorgas Memorial Laboratory, have revealed that high percentages of monkeys killed in the wild are infected with filarial parasites (Clark 1930, 1931). Eight different species of monkeys have been included in the surveys and representatives of all of them have shown numerous microfilariae in blood smears. In certain of the monkey hosts, large numbers of adult filariae were found regularly in the serous cavities, but in the others, no adult worms were demonstrated, even though large numbers of microfilariae were present in the blood. Subsequently, Faust has discovered that in the marmoset monkey, *Leontocebus geoffroyi*, the adult filariae are located between the large muscles of the back, and recently (1935) he described the parasite as *Tetrapetalonema marmosetae*, n.g., n.sp. in the sub-family, *Setariinae*.

During the summer of 1934, at the Gorgas Memorial Laboratory, the writer, under the auspices of Dr. Clark, undertook another survey of the filarial parasites of Panamanian monkeys with the purpose of ascertaining how many species were present and whether any of them could be identified with *Mansonella ozzardi*, a filarial parasite which is present in about 50 per cent of the Indians in the Darien Province of Panama (McCoy, 1933). Live monkeys such as are usually available for study in the labora-

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tory have almost always been captured when young and reared in captivity. They show very light infections with filariae as compared with mature monkeys which have lived all their life in the jungle. Consequently, to obtain material for this survey it was necessary to procure adult wild monkeys. To this end three trips were made into different regions of the Republic of Panama, and collections of filariae made from 72 representatives of six species of monkeys. Additional material was obtained from 12 captive laboratory monkeys.

INCIDENCE OF FILARIAL PARASITES

The three areas of Panama where wild monkeys were shot and examined were (1) along the Rio Boqueron in the upper Chagres valley, Colon Province, central Panama, (2) in the valley of the Rio Chucunaque, Darien Province, southeastern Panama, and (3) along the Rio La Vaca, Chiriqui Province, southwestern Panama. The yellow titi monkey, *Saimiri orstedii orstedii*, was obtained only in Chiriqui Province. Black spider monkeys, *Ateles dariensis*, and marmosets, *Leontocebus geoffroyi*, were obtained only in Darien Province. Red spider monkeys, *Ateles geoffroyi*, were obtained both in the Chagres valley and Chiriqui Province, while white-faced monkeys, *Cebus capucinus*,² were obtained in all three localities. Five of the six black howler monkeys, *Alouatta palliata inconsonans*, which were examined, came from the Chagres valley; the other was obtained in Darien. Since no significant differences in the incidence of filariae were observed in the monkeys of the same species from different localities, the data from the three collection trips will be considered together (table 1).

All of the wild monkeys examined were shot in the jungle and brought to camp for study within a few hours. Fresh blood preparations in normal saline were made and examined at once

² According to Goldman (1920) two species of white-faced monkeys are found in Panama, *Cebus capucinus capucinus* and *C. capucinus imitator*. The former species occurs in the Chagres valley and Darien, while the latter is found in Chiriqui. The same filarial parasites were found in both varieties and in this paper will be listed simply as occurring in *Cebus capucinus*.

for the presence of living microfilariae. Thick blood films were also prepared to be dehemoglobinized later and stained with Giemsa or Bullard's hematoxylin. In addition, a few cubic centimeters of blood from each monkey were laked with 0.5 per cent acetic acid and subsequently preserved in 5 per cent formalin. Since microfilariae in ordinary thick blood films are usually coiled and consequently difficult to study, it was found advantageous to prepare several additional films in which the microfilariae were induced to straighten out by gently heating the freshly made film for a few seconds over an alcohol flame. Such heat-killed micro-

TABLE 1

Incidence of filarial parasites in wild monkeys in Panama

SPECIES OF MONKEY	NUMBER EXAMINED	TOTAL NUMBER POSITIVE	A. GRACILE	T. ATELENSIS	T. PARYUM	M. PANAMENSIS	M. OBUSA	T. MARMOSELAE
White-face, <i>Cebus capucinus</i>	15	14	14	0	8	6	12	0
Yellow titi, <i>Saimiri orstedii orstedii</i>	6	5	0	0	4	3	2	0
Red spider, <i>Ateles geoffroyi</i>	20	20	20	15	0	0	0	0
Black spider, <i>Ateles dariensis</i>	18	14	9	14	0	0	0	0
Marmoset, <i>Leontocbeus geoffroyi</i>	7	5	0	0	0	0	0	5
Black howler, <i>Alouatta palliata inconsonans</i> ...	6	5*	0	0	0	0	0	0

* These infections consisted of a microfilaria different from the others, but insufficient material was obtained to warrant its description.

filariae stained quite as satisfactorily as those in the ordinary preparations, and were found to measure approximately the same length as those in the laked-blood specimens.

Adult filariae were searched for in the serous cavities and, as far as time would permit, in the rest of the carcass by removing the skin and tearing apart the large muscles. All the worms found were killed in 70 per cent alcohol and later transferred to a solution composed of one-third glycerine, one-third 70 per cent alcohol and one-third normal saline. In this solution the worms remained flexible and cleared sufficiently for study. The worms which were encountered in the connective tissue between the

muscles were thin and thread-like and, therefore, easily confused with nerves, small tendons and shreds of fascia. Consequently, unless the infection was extremely heavy, worms in these locations were easily missed in the type of examination possible in a field laboratory. Dunn (1931) devised a method for recovering adult filariae from the muscles of the marmoset monkey by immersing the skinned carcass in normal saline heated to 38°C. The worms emerged from the tissues and could be recovered uninjured much more easily than by dissection. This method was used successfully on several small monkeys studied in the laboratory, but it was not applicable in the examination of larger monkeys under field conditions.

In staining microfilariae it was found that the excretory cell and G₁ cell were most readily demonstrated with a light Giemsa stain. The nuclei of these cells became a bright blue before other nuclei took up the stain. The time of exposure which permitted their differentiation was determined by trial and error. Fifteen minutes in a 1:30 dilution of Giemsa sufficed for most species.

The incidence of microfilariae in the 72 wild monkeys examined is shown in table 1. With few exceptions, all of these animals were adult. The blood smears of 87 per cent of them contained microfilariae. Seven different types of microfilariae were recognized, four of them representing new species (table 2). Insufficient material was obtained of the form discovered in the black howler monkey to warrant its description, but it too is probably a new species. No adult filariae were found in this host. It will be noted that five of the microfilariae occurred in two or more species of monkey host, and that most of the monkeys harbored more than one species of parasite. Definite size differences, fortunately, made it comparatively easy to distinguish the various types. In order to determine the proper relation between the different types of microfilariae and the adult worms which were found in the same monkey, mature female worms were dissected and an examination made of microfilariae taken directly from the uterus. Thus, the proper microfilariae was assigned to the proper adult worm.

ACANTHOCHEILONEMA GRACILE

Acanthocheilonema gracile, a form originally described by Rudolphi in 1809 from Brazilian monkeys, occurred more abundantly than any of the other species, the adult worms being found mainly in the peritoneal cavity, to a lesser extent in the pleural cavities, and occasionally within the pericardium. Extremely heavy infections were frequently encountered in red spider monkeys in Chiriqui, as many as 254 adult worms being recovered from the peritoneal cavity of a single host. Although the inci-

TABLE 2

Species of microfilariae found in Panamanian monkeys

MICROFILARIA	APPROXIMATE LENGTH	SHEATH	HOSTS
	μ		
<i>Acanthocheilonema gracile</i> (Rudolphi, 1809)	130	Present	White face, red spider and black spider
<i>Tetrapetalonema marmosetae</i> (Faust 1925)	260	Not present	Marmoset
<i>Tetrapetalonema atelensis</i> (new species)	200	Not present	Red spider and black spider
<i>Tetrapetalonema parvum</i> (new species)	180	Not present	White face and yellow titi
<i>Microfilaria panamensis</i> (new species)	270	Not present	White face, yellow titi and night monkey
<i>Microfilaria obtusa</i> (new species)	120	Not present	White face and yellow titi
<i>Microfilaria</i> species	170	Not present	Black howler

dence of infection was just as high among the red spider monkeys in the Chagres valley, the number of worms per monkey was much less. *A. gracile* was also found in 9 of 18 black spider monkeys examined in Darien, but 3 worms was the largest number found in any one animal. Several of the female worms from this host measured over 50 cm. in length, whereas the length of female worms from red spider monkeys ranged between 25 and 30 cm.

A. gracile was the only species of filaria which occurred in both spider monkeys (*Ateles*) and white-faced monkeys (*Cebus*). The specimens found in the white-faced monkeys, however, were

considerably smaller than those from the spider monkeys, the female worms measuring only from 16 to 21 cm. in length. In spite of the fact that most of the female worms found in the white-faced monkeys contained mature microfilariae in the uterus, in only a few instances were microfilariae found in blood smears. This observation will be discussed later.

As far as the writer is aware, the microfilaria of *A. gracile* has never been described. The body is comparatively short and stocky averaging about 130μ by 4.5μ in size (see plate 1, fig. 5). The posterior end is sharply pointed and the body nuclei extend

TABLE 3

Measurements of microfilariae in stained smears

The locations of the critical levels are given as the percentage distance from the anterior end. All figures are averages of 50 specimens except those for *T. mamosetae*, which are taken from Faust (1935), and represent averages of 100 specimens.

MICROFILARIA	LENGTH	FIRST BODY NU- CLEUS	NERVE RING	EXCRE- TORY PORE	G ₁ CELL	ANAL PORE
	μ					
<i>Tetrapetalonema mamosetae</i>	299	1.6	23.2	32.1	73.7	84.3
<i>Tetrapetalonema atelensis</i>	205	2.3	24.8	34.0	76.4	86.9
<i>Tetrapetalonema parvum</i>	187	1.2	23.3	32.7	71.4	82.1
<i>Microfilaria panamensis</i>	263	2.2	24.7	34.0	76.0	86.7
<i>Microfilaria obtusa</i>	120	1.4	24.0	36.0	80.8	90.8
<i>Acanthocheilonema gracile</i>	130	1.5	23.4	36.0	71.0	84.3

to the extreme tip. The locations of the nerve ring and other critical points in relation to the body length are given in table 3. The microfilaria possesses a delicate sheath which usually extends about one-fourth the body length posterior to the tip of the tail, and protrudes anteriorly a somewhat smaller distance. This sheath was observed with difficulty in living specimens and much more easily in smears stained with Bullard's hematoxylin. In ordinary Giemsa stained smears the sheath was not visible.

According to the generic definition given by Yorke and Maplestone (1926, page 425), members of the genus, *Acanthocheilo-*

nema,³ should possess microfilariae without sheaths. The finding that the microfilaria of *A. gracile* possesses a sheath necessitates a corresponding modification of the generic definition. As far as is known, the larva of all other members of the genus are unsheathed.

The pathological effects of heavy infections with *A. gracile* were noted by Clark (1931) and his findings were confirmed in the present survey. When large numbers of worms occurred in the peritoneal cavity, a marked fibrous peritonitis with adhesions between the mesenteries and organs was usually present. The same type of reaction was observed, in rarer instances, when parasites were found in the pleural and pericardial cavities. In none of the monkeys examined, however, was the reaction deemed sufficient to impair seriously the health of the animal. In one red spider monkey a single full-grown female filaria was found in the pericardial cavity. The worm was cut open, but no microfilariae or developing eggs were seen in the uterus, indicating that the worm had never been fertilized. This observation suggests that the worm had developed to sexual maturity in this location. In one other monkey, a single partly grown female worm was found in the pericardial cavity.

TWO NEW SPECIES OF TETRAPETALONEMA

The genus, *Tetrapetalonema*, was established by Faust (1935) for the new species of filaria, *Tetrapetalonema marmosetae*, which he described from the back muscles of the marmoset monkey, *Leontocebus geoffroyi*. This genus belongs in the sub-family, *Setariinae*, and differs from *Acanthocheilonema* chiefly by the presence in the female of two pairs of lappets near the caudal extremity instead of only one. In the present survey, two new species of *Tetrapetalonema* very similar to *T. marmosetae* were found and will be described. One occurred in red and black spider monkeys, *Ateles geoffroyi* and *A. dariensis*, and has been

³ The name *Dipetalonema* Diesing, 1861, has been revived by Yorke and Maplestone (1926) to apply to this genus. The writer agrees with the reasons given by Baylis (1929, page 208) for retaining *Acanthocheilonema* Cobbold, 1870, as the more valid name.

named *Tetrapetalonema atelensis*. The other was found in the white-faced monkey, *Cebus capucinus*, and in the yellow titi, *Saimiri orstedii orstedii*, and has been given the name *Tetrapetalonema parvum*. In the two latter hosts another microfilaria was found which is similar to the microfilariae of the three known species of *Tetrapetalonema* and probably belongs to an additional species in this genus. The adult forms were not found, however, and this larva will be described under the name, *Microfilaria panamensis*.

The adult forms of *T. atelensis* were found beneath the fascia of the large muscles of the back, particularly in the region of the scapula. In contrast, the adults of *T. parvum* occurred in the connective tissue between the muscles and to a lesser extent underneath the skin. The region ventral to the scapula appeared to be the most favored location for both species. Several coiled specimens of *T. parvum* were found within definite connective tissue capsules, but for the most part the worms appeared free and in tissue devoid of inflammatory reaction.

Both *T. atelensis* and *T. parvum* are slender, cylindrical, white worms shorter in length than *T. marmosetae*. Their cuticula is smooth and devoid of markings. The comparative sizes of the two sexes in the three species of *Tetrapetalonema* are shown in table 4. Although the females of *T. parvum* are wider at their greatest diameter than those of *T. atelensis*, they taper more at the extremities, so that the diameter of the head end of *T. parvum* is only a little more than half that of the latter species. The head-on view of the anterior ends of the worms of both species is similar to that of *T. marmosetae*, (plate 2, figs. 1 and 7). The mouth is without lips and is surrounded by a rectangular peribuccal plate at each corner of which is a pair of papillae. In the head-on view these papillae lie one above the other and diverge at an angle of about 60°. Dorsal and ventral to the peribuccal plate on the mid-line is a prominent protuberance ending in a small papilla.

The posterior end of the female of *T. parvum* bears two pairs of fleshy lappets located ventrally and slightly subterminally (plate 2, figs. 8 and 9). The two innermost lappets are about

one-half as large as the two outer. In the female of *T. atelensis* the four lappets are of approximately the same size and are more conspicuous than in *T. parvum* (plate 2, figs. 2 and 3). Along the anterior fifth of most specimens of *T. atelensis* are four rounded annular swellings extending about 0.2 mm. along the length of the worm and spaced at more or less regular intervals of from 3 to 4 mm. In the region of the swellings the body is about 25 per cent wider than at other levels. Similar swellings are observed along the anterior fifth of most specimens of *T. marmosetae*, but their location is not as constant. Since no swellings are present in

TABLE 4

Comparative size of the adult forms of the three species of tetrapetalonema
All measurements are in millimeters

	T. MARMOSETAE	T. ATELENSIS	T. PARVUM
Females.....	15 specimens	8 specimens	24 specimens
Range in length.....	71 to 117	49 to 63	28 to 49
Average length.....	86.7	56.4	44.6
Average width.....	0.17	0.15	0.17
Males.....	7 specimens	1 specimen	6 specimens
Range in length.....	31 to 43		16.5 to 19
Average length.....	39.0	32.0	17.6
Average width.....	0.100	0.082	0.073
Length of long spicule.....	0.686	0.771	0.644
Length of short spicule.....	0.179	0.217	0.142
Ratio of long to short.....	3.8:1	3.6:1	4.5:1

T. parvum, this character is of diagnostic importance. As in *T. marmosetae*, the vulvar opening in the females of both *T. atelensis* and *T. parvum* is located ventrally a little less than twice the distance from the nerve ring that the nerve ring is distant from the anterior end.

Only two male specimens of *T. atelensis* were collected, and one of these was a posterior fragment. The posterior end of the male is coiled through 360° and near the tip bears a pair of pointed fleshy lappets located laterally (plate 2, fig. 4). There are six pairs of perianal papillae arranged asymmetrically, the last papilla on the left side being located further caudad than that on

the right side (plate 2, fig. 5). An additional pair of papillae is situated ventrally about half-way between the anal opening and the caudal extremity, and further caudad is another pair lateral in position. The two spicules are unequal in size, the larger one being 3.6 times as long as the smaller one. They are both somewhat longer than the spicules in *T. marmosetae*. No gubernaculum was distinguished. The long spicule is "chitinized" throughout its length and ends in a slight hook with an "unchitinized" dorsal keel (plate 2, fig. 6). The short spicule is only weakly "chitinized." It was not exerted in either of the two specimens examined. Aside from certain small differences in size (table 4) and the fact that the most posterior papillae are single rather than double, the males of *T. atelensis* are almost identical with those of *T. marmosetae*.

The males of *T. parvum* are small and delicate, and present certain definite differences from the males of the other two species of *Tetrapetalonema*. The small spicule is "chitinized" throughout its length. In several specimens it was found exerted together with the large spicule. The shape of the ends of the two spicules is shown in plate 2, figures 13 and 14. No gubernaculum was observed. In addition to a prominent pair of fleshy lappets near the tip of the tail, another much smaller pair is located further anteriorly (plate 2, fig. 11). The presence of this second pair of lappets in the male of *T. parvum* necessitates a corresponding modification of the generic definition given by Faust (1935). Usually five pairs of perianal papillae are present, but their number and arrangement is not constant. Of the 11 specimens available for study, 5 possessed 5 pairs of papillae, 4 had 5 papillae on the left side and only 4 on the right, one had 6 papillae on the left side and 5 on the right, and one had 6 papillae on the left side and only 4 on the right. This last specimen was also abnormal in that one of the fleshy lappets was missing from the left side of the caudal extremity. A study of 11 male specimens of *T. marmosetae* revealed a somewhat similar variation from the 6 pairs of perianal papillae mentioned in Faust's description. Five of the worms had 6 pairs of papillae, while 6 specimens possessed 7 pairs. The typical asymmetric arrangement of the perianal

papillae of *T. parvum* is shown in plate 2, figure 12. The two pairs of papillae near the caudal end are essentially the same as described for *T. atelensis*, except that the posterior lateral papillae occasionally are notched at the end giving them the appearance of being double.

The microfilariae of *T. atelensis* and *T. parvum* are both much smaller than the microfilaria of *T. marmosetae* (see table 3). All three microfilariae are unsheathed and have a tail tapering to a blunt ending with the body nuclei extending to the extreme tip. In the microfilaria of *T. atelensis* the last tail nucleus is slightly larger than the other nuclei, a fact which in stained specimens gives a knob-like appearance to the caudal tip. The portion of the anterior end of the body which is free of nuclei is almost twice as great in the microfilaria of *T. atelensis* as in the microfilaria of *T. parvum* (plate 1, figs. 1 and 2). There are also a few small differences in the locations of the critical points (table 3). Otherwise the two larvae are very similar.

TETRAPETALONEMA ATELENSIS N. SP.

Partaking of the characters of the genus. Females measuring from 49 to 63 mm. in length with two pairs of fleshy lappets equal in size ventro-lateral to the posterior end. Males approximately 32 mm. long with two copulatory spicules unequal in size, the shorter spicule being only weakly "chitinized"; no gubernaculum distinguished; 6 pairs of perianal papillae, one pair of ventral papillae half-way between the anal opening and the caudal end, and an additional pair of lateral papillae further posterior. Microfilaria approximately 200 μ in length, unsheathed and with the last nucleus in the tail slightly larger than adjacent nuclei. Adults found beneath the fascia of the large muscles in the region of the scapula. Type host: red spider monkey, *Ateles geoffroyi*; additional known host: black spider monkey, *Ateles dariensis*. Type locality: Chiriqui Province, R. de Panama.

TETRAPETALONEMA PARVUM N. SP.

Partaking of the characters of the genus. Females measuring from 38 to 49 mm. in length with two pairs of fleshy lappets ven-

tro-lateral to the posterior end; two innermost lappets about half as large as the two outermost. Males from 16.5 to 19 mm. in length, with two unequal copulatory spicules, the shorter spicule being "chitinized" throughout its length; no gubernaculum distinguished; two pairs of lappets lateral to the caudal end, one pair anterior to the other and much smaller in size; typically 5 pairs of perianal papillae, but sometimes one papilla missing on the right side or an additional one present on the left side; one pair of ventral papillae half way between the anal opening and the caudal end, and an additional lateral pair further posterior. Microfilaria approximately 180μ in length, unsheathed and with a very small portion of the anterior end free of nuclei. Adults found in connective tissue between the large muscles of the back. Type host: white-faced monkey, *Cebus capucinus imitator*; additional known hosts: *C. capucinus capucinus*, and the yellow titi monkey, *Saimiri örstedii örstedii*. Type locality: Chiriqui Province, R. de Panama.

MICROFILARIA PANAMENSIS N. SP.

As already mentioned above, a microfilaria similar to the microfilariae of the three species of *Tetrapetalonema* was found in the blood of the white-face and yellow titi monkeys. It was also found in one specimen of the night monkey, *Aotus zonalis*, examined at the laboratory. This larva never was present in very large numbers and the adult forms were not found. It has been given the name, *Microfilaria panamensis*.

M. panamensis is a large, unsheathed larva measuring about 260μ in length and 5μ in breadth (plate 1, fig. 4). A comparatively large portion (2.2 per cent) of the anterior end is free of nuclei. The relative locations of the critical points are given in table 3 and it will be seen that they are almost identical with those of the microfilaria of *T. atelensis*. *M. panamensis*, however, is readily distinguished from the latter species by the fact that it is almost 40 per cent longer. The tail of *M. panamensis* terminates bluntly and the nuclei extend to the tip. Living specimens of *M. panamensis* in normal saline exhibited fairly rapid progressive locomotion accomplished by vigorous snake-like movements. In

contrast, living specimens of the three *Tetrapetalonema* microfilariae showed relatively little progressive locomotion, their movements being limited mainly to coiling and uncoiling. *M. panamensis* is further distinguished by the fact that specimens in thick blood smears did not take up Giemsa stain as readily as did the other microfilariae studied in this survey. For instance, approximately 40 minutes were required to stain the body nuclei of *M. panamensis* in a 1:30 dilution of Giemsa, whereas 20 minutes in the same dilution were sufficient to stain the microfilariae of *Tetrapetalonema marmosetae* and *T. parvum*.

Some variation was noticed in the size of specimens of *M. panamensis* from different species of monkey hosts, and to a lesser extent from different hosts of the same species. The following series of the average length of 20 specimens in laked blood samples will suffice to show the extent of this variation: white-face no. 20 = 251μ ; white-face no. 69 = 263μ ; yellow titi no. 57 = 273μ ; yellow titi no. 59 = 273μ ; and night monkey no. 326 = 295μ . The differences between the average lengths in the three different hosts are statistically significant, but are not considered sufficient to indicate species differentiation.

M. panamensis most closely resembles the microfilaria of *T. marmosetae*, and species differentiation would be difficult unless specimens of both were available for comparison. In the present study, 50 specimens of the microfilaria of *T. marmosetae* from laked blood were found to average 256μ in length, and an equal number in stained blood smears averaged 258μ in length. These averages are considerably smaller than the average length of 299μ given in Faust's description of the species. Since, however, in the present study measurements of both *M. panamensis* and the microfilaria of *T. marmosetae* were made under the same conditions, it appears reasonable to conclude that the former species is slightly larger than the latter. In comparison with the microfilaria of *T. marmosetae*, *M. panamensis* exhibits a greater capacity for progressive locomotion in fresh blood smears, and in dried blood films is much less readily stained by Giemsa. Other points of differentiation are the slightly greater portion of the anterior end of *M. panamensis* which is free of nuclei, and also small

differences in the locations of the critical levels (see table 3). In the present survey, *M. panamensis* and the microfilaria of *T. marmosetae* did not occur in the same hosts.

Microfilaria panamensis. Specific diagnosis: unsheathed microfilaria occurring in the blood of monkeys, and measuring from 250μ to 295μ in length. Nucleus-free portion of anterior end relatively large (2.2 per cent of body length). Tail terminates bluntly and nuclei extend to extreme tip. Location of critical levels expressed as per cent of body length from anterior end as follows: nerve ring, 24.7; excretory pore, 34.0; G₁ cell, 76.0; and anal pore, 86.7. Type host: white-faced monkey, *Cebus capucinus imitator*; other known hosts: *C. capucinus capucinus*, the yellow titi monkey, *Saimiri orstedii orstedii*, and the night monkey, *Aotus zonalis*. Type locality: Chiriqui Province, R. de Panama.

MICROFILARIA OBTUSA N. SP.

A microfilaria quite different from *M. panamensis* and the microfilariae of the three species of *Tetrapetalonema* was found in the blood of the white-faced monkey and also of the yellow titi monkey. Since the adult forms were not found, this larva will be described under the name *Microfilaria obtusa*. It is a short, stocky, unsheathed larva measuring about 120μ in length and 4.3μ in breadth. The tail is very short (9 per cent of the body length) and tapers only slightly to a blunt termination. The body nuclei extend to the extreme tip (plate 1, fig. 3). In most stained specimens, at first glance, it is difficult to distinguish the anterior and posterior ends. Seen in fresh blood preparations, *M. obtusa* does not appear to be capable of progressive locomotion, but tumbles about in uncoordinated coiling and uncoiling movements. When *M. obtusa* is killed by heat or by laking the blood in 0.5 per cent acetic acid, the body almost always bends in the middle and assumes an open V-shape. The name of the species was suggested by the fact that the angle of the bend in killed specimens is most often an obtuse angle.

A comparatively short portion of the body (1.4 per cent) at the anterior end is free of nuclei. The remainder of the body is so closely packed with nuclei that it is often difficult to distinguish

the nerve ring and other critical points. The location of the critical points in relation to the body length are given in table 3. The G₁ cell and anal pore are much more posterior in position in *M. obtusa* than in the other microfilariae. Although *M. obtusa* was found in 70 per cent of the specimens of white-faced and yellow titi monkeys which were examined, it was never present in large numbers. In several juvenile white-faced monkeys examined at the laboratory, *M. obtusa* was the only species of microfilaria present.

Microfilaria obtusa. Specific diagnosis: Unsheathed microfilaria occurring in the blood of monkeys and measuring approximately 120 μ in length. Tail very short, only slightly tapered, and ending bluntly with nuclei extending to the extreme tip. Location of critical levels expressed as per cent of body length from anterior end as follows: first nucleus, 1.4; nerve ring, 24.0; excretory pore, 36.0; G₁ cell, 80.8; and anal pore, 90.8. Type host: white-faced monkey, *Cebus capucinus imitator*; other known hosts: *C. capucinus capucinus*, and the yellow titi monkey, *Saimiri orstedii orstedii*. Type locality: Chiriqui Province, R. de Panama.

DISCUSSION

The wild monkeys of Panama harbor a fauna of filarial parasites rich both in number of species represented and percentage of animals infected. In view of the fact that most of the different species of monkeys examined in the present survey live in the same type of environment and often in close proximity to each other, it is interesting to note the host specificity of certain of the species of filaria. In general, the specificity seems to be correlated with the zoölogical relationship of the hosts, but the correlation is not always the same for each species of filaria. For instance, according to the classification of Stiles and Nolan (1929), the white-faced monkey (*Cebus*) and the yellow titi (*Saimiri*) belong in the same sub-family, *Cebinae*, and are more closely related than any other two genera of monkeys included in the present survey. They were found to harbor three species of filaria in common, namely *T. parvum*, *M. panamensis* and *M. obtusa*. On the other hand,

A. gracile occurs regularly in the white-faced monkey, but has never been reported from the yellow titi (present survey and Clark, 1931). *T. atelensis* was found only in the spider monkeys, *Ateles geoffroyi* and *Ateles dariensis*, but *A. gracile* occurred not only in both of the spider monkeys but also in the white-faced monkey. As far as the present survey goes, the black howler monkey, *Alouatta palliata inconsonans*, and the marmoset, *Leontocebus geoffroyi*, appear to harbor distinctive filariae of their own. It is interesting, though, that *T. marmosetae* from the latter host is very closely related to other species of *Tetrapetalonema* occurring in the white-face and spider monkeys.

As has already been mentioned, in spite of the fact that numerous fertile females of *A. gracile* were found in the peritoneal cavities of white-faced monkeys, the microfilariae were only very rarely encountered in the blood smears from this host. In red spider monkeys infected with *A. gracile*, microfilariae could almost always be found in the blood, but frequently their number was much smaller than would be expected from the large number of adult worms harbored. In view of these findings and the fact that the microfilaria of *A. gracile* is sheathed, it seems possible that the microfilarial stage of this species may exhibit a periodicity in its occurrence in the peripheral blood. Since all of the blood smears from the wild monkeys were made during the day, periodicity, if such exists, is nocturnal in character. No examinations of captive monkeys have been made to test this hypothesis.

It is evident that none of the monkey filariae found in Panama can be identified with any of the human species of filariae. From the standpoint of experimentation, it would be desirable to find an animal filarial parasite resembling *Wuchereria bancrofti*, particularly in its pathological effects. These monkey filariae, however, are more closely related to *Acanthocheilonema perstans* and *Mansonella ozzardi*, two human parasites which produce little, if any, pathological effect. As a group, the filarial parasites of Panamanian monkeys offer a number of interesting possibilities for further study, particularly their life cycles. The adult forms of three species of microfilariae have not been found as yet, and the transmitting vectors for all of them are completely unknown.

SUMMARY

Seven different species of filarial parasites were found in the eight species of monkeys which occur in Panama, 87 per cent of the 72 wild monkeys which were examined being infected with one or more species of filaria. *Acanthocheilonema gracile* was the parasite most frequently encountered, the adult worms being located in the serous cavities of the red and black spider monkeys and the white-faced monkey. The microfilaria of this species possessed a sheath; all other microfilariae were unsheathed. Two new species belonging in the genus, *Tetrapetalonema* Faust 1935, have been described. One of these, *T. atelensis*, was found beneath the fascia of the back muscles of the red and black spider monkeys; the other, *T. parvum*, was found in connective tissue between the back muscles of the white-faced and yellow titi monkeys. Two new species of microfilariae, *M. panamensis* and *M. obtusa*, have been described from these two latter hosts. The adult forms of these larvae were not found.

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EXPLANATION OF PLATE 1

All figures $\times 750$. *NR*, nerve ring; *EP*, excretory pore; *G*₁, first germ cell; *AP*, anal pore; *Sh*, sheath.

FIG. 1. Microfilaria of *Tetrapetalonema parvum* (new species).

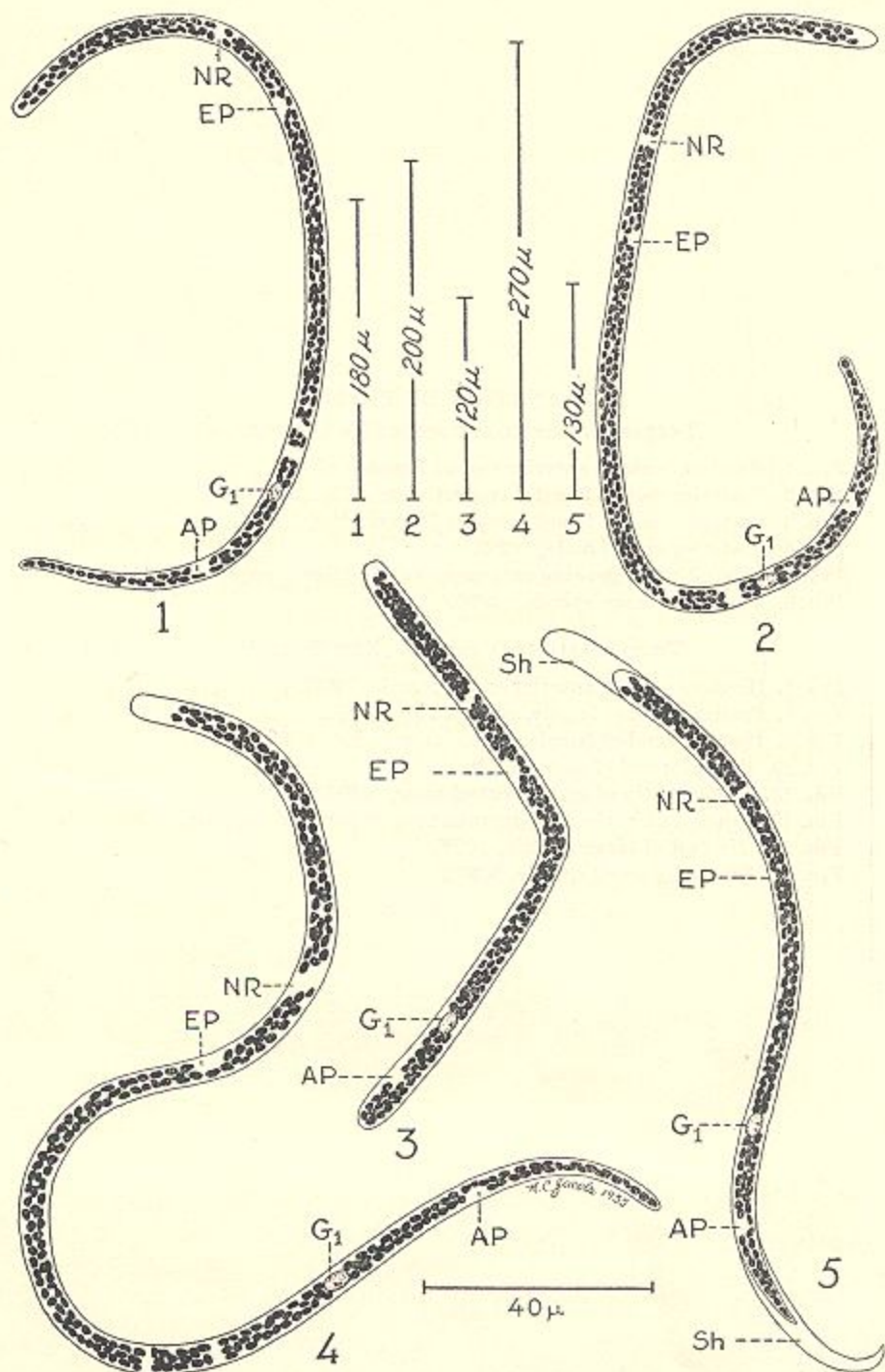
FIG. 2. Microfilaria of *Tetrapetalonema atelensis* (new species).

FIG. 3. *Microfilaria obtusa* (new species).

FIG. 4. *Microfilaria panamensis* (new species).

FIG. 5. Microfilaria of *Acanthocheilonema gracile* (Rudolphi, 1809).

The linear scales at the top of the plate indicate the relative lengths of the different microfilariae.



EXPLANATION OF PLATE 2

TETRAPETALONEMA ATELENSIS (NEW SPECIES)

- FIG. 1. Head-on view of anterior end of female, $\times 640$.
FIG. 2. Posterior end of female, ventral view, $\times 360$.
FIG. 3. Posterior end of female, ventro-lateral view, $\times 360$.
FIG. 4. Posterior end of male, $\times 280$.
FIG. 5. Ventral view showing arrangement of perianal papillae, $\times 360$.
FIG. 6. Tip end of large spicule, $\times 800$.

TETRAPETALONEMA PARVUM (NEW SPECIES)

- FIG. 7. Head-on view of anterior end of female, $\times 640$.
FIG. 8. Posterior end of female, ventral view, $\times 360$.
FIG. 9. Posterior end of female, dorso-lateral view, $\times 360$.
FIG. 10. Posterior end of male, $\times 280$.
FIG. 11. Posterior tip of male, ventral view, $\times 360$.
FIG. 12. Ventral view showing arrangement of perianal papillae, $\times 360$.
FIG. 13. Tip end of large spicule, $\times 800$.
FIG. 14. Tip end of small spicule, $\times 800$.

