CEYLON J. MED. SCI. (D) Vol. XI, Pt. 1 (May 1962)

Some Observations on *Brugia (Brugiella) buckleyi* Dissanaike and Paramananthan, 1961 —A Filarial Worm from the Heart and Blood Vessels of the Ceylon Hare*

by

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(With two Text Figuers & two Plates)

UNTIL recently two types of filarial larvae (microfilariae) were met with in the peripheral blood of man in this country. They were the microfilariae of two different adult filarial worms known as *Wuchereria bancrofti* and *W.malayi*. Fig. 1 shows the main differences between the microfilariae of these two worms. Both have sheaths; but *microfilaria malayi* differs from *microfilaria bancrofti* mainly in the fact that it has two distinct tail nuclei at the tail end and its body nuclei are not discrete but overlapping. In the last few years microfilariae like those of 'malayi' were met with in a number of animals in Malaya and later in Africa (Edeson et al. 1955, Nelson and Heisch, 1957, Poynton and Hodgkin, 1939). The adult

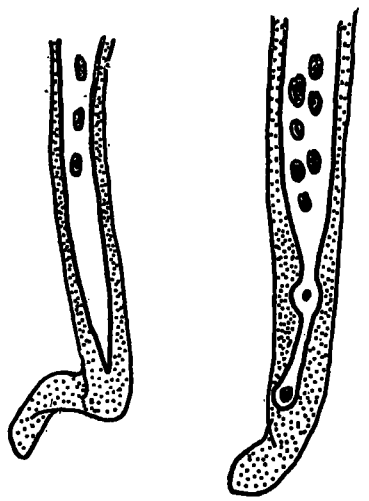

Fig 1

worms of these were carefully described and compared with those of the human form (Buckley and Edeson, 1956, Buckley, Nelson and Heisch, 1958). As a result of these studies,

* Paper read at the 17th Annual Sessions of the Ceylon Association for the Advancement of Science held in Colombo in November, 1961.

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three different species of adult filarial worms of man and animals were found to have microfilariae with the typical tail nuclei. They were named *Wuchereria malayi*, *W.pahangi* and *W.patei*. Quite recently Buckley (1958, 1960) decided that these three species should be separated from the genus *Wuchereria* and created for them a new genus *Brugia*. Apart from the fact that they had the same type of microfilaria, they had other common characters such as the arrangement and number of the papillae and the pattern of the spicules of the males.

We recently reported the finding of microfilariae of the *Brugia* type from a Ceylon Black Naped Hare (*Lepus nigricollis singhala*) (Dissanaike and Paramananthan, 1961). That hare was shot at Mundumurippu in the Northern Province and on that occasion we recovered a few adult worms as well; but from the heart and hepatic vessels and not from the lymphatics. Since then we have found this filarial worm to be quite a common parasite of the hare. The adult worms proved to belong to a new species of *Brugia*, and since the characters were distinct from those of the hitherto described species we created a new sub-genus *Brugiella* (Dissanaike and Paramananthan, 1961) and named it *Brugia (Brugiella) buckleyi*. A detailed description of these worms has already been published (Dissanaike and Paramananthan, 1961). Since then Jayawardena (1961) has reported the finding of *Brugia* type microfilariae from dogs in Ceylon. It will be interesting to know what the adult worms of dogs turn out to be.

It is the purpose of this communication to report some preliminary observations on the incidence and distribution of *Brugia (Brugiella) buckleyi* and the pathological changes it causes in the host.

Brief Description of the Worm

It will be useful at this stage to point out briefly some of the essential features in the morphology of this worm which differentiate it from the other species of *Brugia*.

1. Adult measurements—Much larger than the other three species. More like *W. bancrofti*. For instance, in females the length ranges from 80mm—100mm and the maximum breadth is over 300μ and in the males the range is from 40mm—50mm and the maximum breadth is about 190μ.
2. Head bulb—Not distinct and position behind level of the head papillae.
3. Female—Cuticular bosses at tail end larger than in the others and more like *W. bancrofti*. Oesophagus and vulva relatively much closer to each other.
4. Male—Spicule ratio 2 or less than 2:1. Adanal papillae different in arrangement. The base of the left spicule is not cup-shaped and its tip is not spatulate. Oesophagus relatively short.

Distribution of the Parasite

In order to study the distribution of the worm in Ceylon we arranged for hares to be shot from different parts of the Island, and examined them either fresh or after preservation of the organs in 10% formalin. At the beginning of the investigation we observed that the

worms were usually found in adult hares and that the leverets were free of infection. Accordingly, we concentrated on adult hares. We have now examined hares from all the provinces except the Sabaragamuva and the Uva Provinces. 41 out of the 58 hares examined in all were found to harbour either immature or mature adult worms giving a high incidence of 70.7% (**Table 1,** & Fig. 2). 8.6% of them showed infection with another filarial worm *Micipsella* sp. It will be noticed from the table that the largest number examined was from the Western Province in the suburbs of Colombo where we have found that practically every adult hare was positive.

TABLE I

Brugiella Survey

PROVINCE	LOCALITY	HARES EXAMINED	HARES POSITIVE	REMARKS
NORTHERN	Vavunikulam	2	1	2+ve for Micipsella sp.
	Mundumurippu	1	1	
	Omantai	1	nil	
NORTH CENTRAL	Anuradhapura	5	4	One hare had immature worms only.
	Mannampitiya	5	3	
NORTH WESTERN	Moonemalgas-wewa	1	1	Immature worms only.
	Lunuwila	3	3	
WESTERN	Negombo	1	1	
	Avissawella	1	nil	
	Homagama	1	1	
	Kottawa	2	2	
	Battaramulla	1	1	Immature worms only.
	Athurugiriya	4	2	
	Hokandara	11	10	
	Katubedde	3	2	
	Bandaragama	2	1	
CENTRAL	Peradeniya	3	2	
EASTERN	Okande	6	4	2+ve for Micipsella sp.
	Pottuvil	2	nil	
SOUTHERN	Ambalangoda	1	nil	
	Wiraketiya	2	2	1+ve for Micipsella sp.
		58	41	70·7%

Site

Although in the original report we mentioned the occurrence of the adult worms in the heart and hepatic blood vessels, we now find that they are mostly present in the right ventricle and the pulmonary arteries (Figs. A—F). In many instances they were found in large numbers practically filling the cavity of the right ventricle and the lumen of the pulmonary

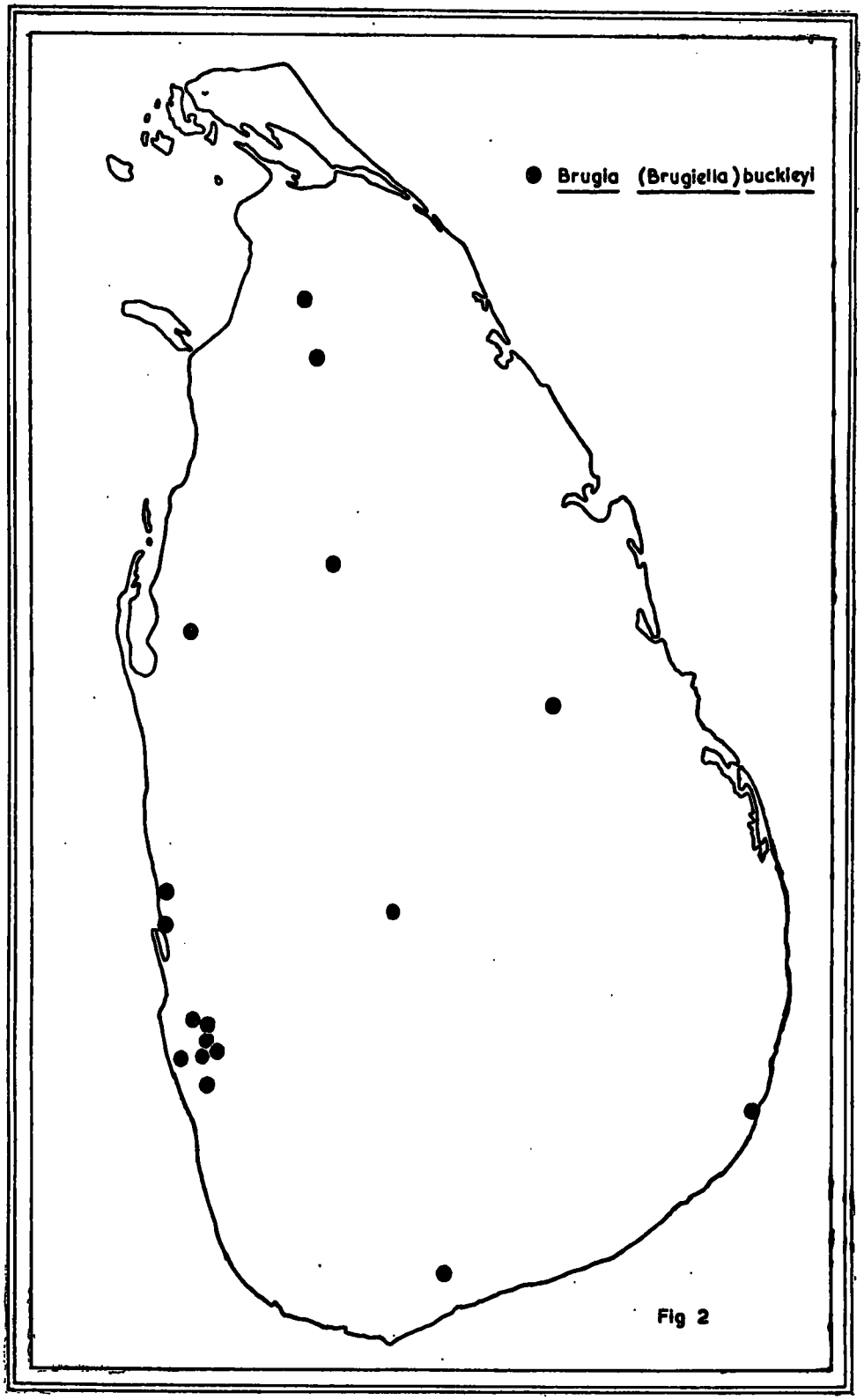

Fig 2

artery. In one animal we recovered over 50 worms from the right ventricle and the pulmonary arteries. On one occasion we found a single worm in the posterior vena cava. One hare had a few worms in the right auricle as well as in the right ventricle.

Most of the hares, even though they harboured a small number of worms, were microfilariae-positive. In several, where the blood was negative, we found immature worms. In a few which showed microfilariae in the blood we were unable to recover the adult worms from the usual sites. It is possible that in these instances the adult worms were in some other sites.

The other filarial worm *Micipsella* sp. was found mainly in the hepatic blood vessels.

Pathology

No attempt has been made to study the detailed pathological changes. We merely describe the obvious gross changes due to the presence of large numbers of adult worms choking the right venticle and the pulmonary arteries. In many of the heavily infested hares the right ventricle was either hypertrophied or dilated and in several that showed worms in the pulmonary artery we found red infarcts in the lungs. Dead microfilariae were seen in these infarcted areas. The pulmonary arteries too showed signs of hypertrophy and also dilatation.

While the presence of the microfilariae did not appear to cause any serious changes we found them quite frequently in sections of the heart and lungs. We have not studied sections of other organs.

Experimental Work

Several attempts were made to obtain a live positive hare for the purpose of mosquito feeding as well as for studies on periodicity of the microfilariae. We have so far not succeeded in trapping a positive hare.

In the meantime we have been trying to introduce the adult worms recovered from a recently-shot hare into a domestic rabbit. Using a wide bore needle (12 gauge) we injected 4 rabbits intracardially with the adult worms in a little physiological saline. Several male and female worms were introduced into each rabbit. None of these animals showed microfilariae in the blood upto date. It is quite likely that the worms reached the left ventricle and as this was not a suitable site for them they eventually died and were absorbed.

On 20.8.61 we introduced 2 male and 6 female worms surgically into the peritoneal cavity of a rabbit. This rabbit too remains microfilariae negative.

On several occasions we have collected blood in vials containing a few cc. of sterile saline from positive hares shot around Colombo. These microfilariae remained alive and actively motile for 3-4 weeks when kept in the refrigerator. After this they became vacuolated and sluggish and eventually died.

Discussion

The finding of this *Brugia* type filarial worm in Ceylon hares becomes all the more interesting since the adults are found in the heart and pulmonary arteries rather than in the lymphatics. This is not altogether surprising when we remember that the same situation obtains in the dog filarial worms *Dirofilaria immitis* and *D.repens*. The former worms are found in the right ventricle and pulmonary vessels while the latter are confined to the subcutaneous tissues. *D.immitis* however undergoes its early stages of development in the dog in the subcutaneous tissues and muscles and only later enters the veins to reach its final destination in the right heart. We can assume that a similar course of events takes place in *Brugiella*.

Drawing the analogy from *D.immitis*, once again, we can assume that the chief effects of the worms in the heart of the hare are due to interference with circulation in the right side of the heart. The mechanical blockage due to the presence of a large number of worms can in many cases eventually cause compensatory hypertrophy and enlargement of the right ventricle. In the end passive congestion of the lungs, liver and spleen, as well as ascites may result. According to Smith and Jones (1957) other pathological changes known to occur in *D.immitis* infection are subintimal proliferation of connective tissue in the pulmonary arteries, with diffuse thickening of the subintima; severe endothelial proliferation in small pulmonary arterioles, sometimes with almost complete obliteration of the lumen; and medial hypertrophy of some of the larger pulmonary arterioles. While we have not studied these pathological changes in detail we have sometimes wondered how a heavily infected hare with worms literally clogging the right heart and pulmonary arteries ever survived.

The microfilariae normally do not produce tissue damage and we have seen them in sections of the heart and lungs. It would be interesting, however, to study sections of other organs like the liver, spleen and kidney.

Once we are able to trap a positive live hare, periodicity studies of the microfilariae could be carried out and the transmission to the laboratory rabbit and perhaps to other laboratory animals like rats and guinea pigs should not be difficult. Such infected animals will be invaluable in therapeutic studies of antifilarial drugs. In this connection we would like to repeat a suggestion we made earlier (Dissanaike and Paramananthan, 1961) that it is possible that the adult worms missed by Laing et al. (1960) in the squirrels with *Brugia* type microfilariae were probably in the heart and blood vessels.

The high incidence of infection of hares in most localities in Ceylon makes one wonder what the vector of this parasite would be. On several occasions when we had heavy infection in a hare with microfilariae in the blood none of the numerous ticks found on the host showed any microfilariae or developmental stages in them. The chances are that the tick is not the likely vector. If indeed the vector is a *Mansonia* mosquito, it is possible that the *Brugia* type infective larvae occasionally met with in routine dissections of wild caught *Mansonia* from the suburbs of Colombo (Niles, 1961) are those of worms from the hare. This again suggests that the *Brugia* of the Ceylon hare may play an important role in the aetiology of Tropical Pulmonary Eosinophilia.

Summary

1. The incidence and distribution of *Brugia (Brugiella) buckleyi* has been studied. 41 out of the 58 hares examined (70.7%) were infected.

2. The gross pathological changes observed in the heavily infected hares were hypertrophy and dilatation of the heart and pulmonary arteries and red infarctation of the lungs.

3. Some preliminary attempts at introducing adult worms into the domestic rabbits are described.

Acknowledgements

We are grateful to the following for providing us with the hares examined during this investigation :— Dr. D. V. Arndt, Dr. P. Seneviratna, Messrs. Zacky Deen, E. C. Fernando, P. Kirthisinghe, K. Alagarajah, L. de Alwis, H. Todd, S. Dharmarajah, and S. Nagamuthu.

We are also thankful to Messrs. R. B. Arseculeratne, A. U. Jayaweera and H. E. Bartholomeusz for technical assistance.

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EXPLANATION OF FIGURES

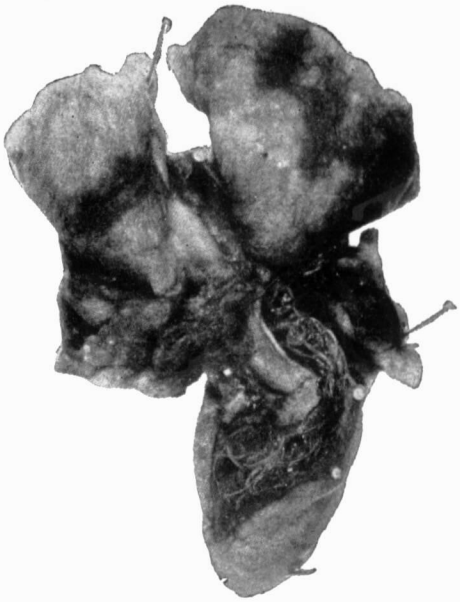
PLATE I.

Figure A—Heart of a hare showing worms in the right ventricle (X 1½).
Figures B & C—Heart and lungs of two hares showing worms in the right ventricle and base of the pulmonary artery (X 1).
Figure D—Heart and lungs of a hare dissected to show the worms in the right ventricle, pulmonary artery and its branches in the lungs (X 1).

PLATE II.

Figure E—Section of a pulmonary artery in the lung showing a number of adult worms (X 29).
Figure F—Portion of the above under a higher magnification (X 400).

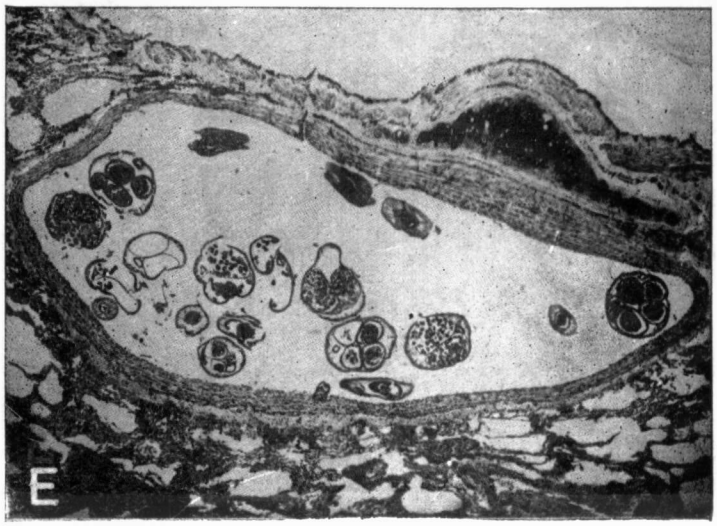
A



B

C

D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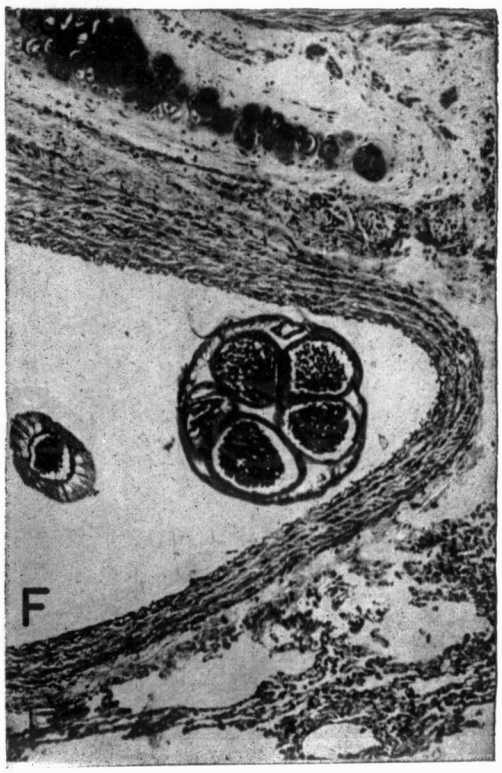
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