THE VECTORS OF BRUGIA MALAYI AND B. PAHANGI: A REVIEW OF LITERATURE, WITH SPECIAL REFERENCE TO FILARIASIS DUE TO WUCHERERIA AND BRUGIA SPECIES IN PAKISTAN*

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Since Manson's discovery in 1878 that the mosquito acts as the intermediate host of *Wuchereria bancrofti* the development of some 36 species of filarial nematodes has been studied in their arthropod hosts.

Many different kinds of mosquitoes have been found to transmit the human filaria *W. bancrofti*, and the ability to transmit is not in strict accordance with the systematic classification of the insects. Recent lists of vectors of *Wuchereria* have been given by Manson-Bahr, 1959 and Raghavan, 1956, 1961. A bibliography on the vectors of *Wuchereria* in the South Pacific region has been published by Iyengar, 1956 and by the World Health Organisation Expert Committee on Filariasis, 1962. Edeson and Wilson 1964 gave a list of the confirmed vectors of *W. bancrofti* and *Brugia malayi* in relation to their geographical distribution, listing three species of *Anopheles* and six of *Mansonia* as vectors of *B. malayi*.

Appraisals of recent work on the transmission of filariod nematodes have been made by Kartman, 1957, Lavoipierre, 1958, Hawking and Worms, 1961, and Edeson and Wilson, 1964. Here, therefore, only the role of *Mansonia*, *Anopheles*, and *Aedes* mosquitoes as natural and laboratory vectors of *B. malayi* (sub-periodic and periodic) and *B. pahangi* will be reviewed.

MANSONIA

MALAYA

Brugia pahangi—Edeson, Wharton and Laing, 1960 found that in the swamp forest areas of Malaya, where animals infected with *B. pahangi* occurred, mosquitoes of the genus *Mansonia* were dominant in biting collections, and those of the subgenus *Mansonioides* were the only species found regularly infected with *Brugia* larvae. Since a wide variety of mosquitoes occurred in the region, the authors attempted to feed as many as possible on infected hosts to see which were susceptible to *B. pahangi*. These experiments showed that *M. annulata* was a good host, *M. uniformis* was less efficient, and *M. longipalpis* was a comparatively poor

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laboratory host. *Mansonia* (*Coquillettidia*) *crassipes* also supported development of *B. pahangi* to the mature stage *M.* '*longipalpis*' and *M. annulata* were proved to be the vectors in nature.

Wilson, 1961 suggested that *M. annulata* is the most important natural vector of *B. pahangi* in Malaya with other mosquitoes playing a minor role, and pointed out that *B. pahangi* did not develop nearly so readily in *M.* '*longipalpis*' as did sub-periodic *B. malayi*. Prior to the separation of *M. dives* and *M. bonneae* in dissection results, when they were grouped together as '*longipalpis*' it was thought that both species were vectors of *B. pahangi*. But when the two species were separated, infection experiments by Wharton, 1962 with *B. pahangi* showed that whereas 12-31 per cent of the filarial larvae were stage III in *M. dives* dissected 13-14.5 days after feeding, only four larvae of more than 1,000 were beyond stage I in *M. bonneae*. The author therefore concluded that *M. bonneae* could not be of importance in the transmission of *B. pahangi* in nature. With respect to the other species of *Mansonia*, Wharton, 1962 stated that in *M. annulata* all, or nearly all, *B. pahangi* larvae were stage III by the 11th day, but development was retarded in heavy infections, while in *M. uniformis* an average of 13 per cent were stage III on the 11th day. Feeding experiments with *M. annulifera* and *M. indiana* were inconclusive and showed that only *M. indiana* from Pahang would support the development of larvae to the mature stage and that *M. annulifera* was probably not a host for *B. pahangi*.

Sub-periodic Brugia Malayi—The association between Malayan filariasis due to *B. malayi* and mosquitoes of the genus *Mansonia* was first reported by Brug, 1931, when he was investigating the vectors of *B. malayi* in the Dutch Indies. He found that *M. annulata* and *M. longipalpis* (=*dives/bonneae*) were by far the most common man-biting mosquitoes in the area of his survey. Development of *B. malayi* occurred in these two species only and in two other species belonging to the same genus, *M. uniformis* and *M. annulifera*; the latter two species were however far less abundant in the area. Brug concluded the *M. annulata* and *M. dives/bonneae* were the vectors.

Hodgkin, 1938, 1939, 1940 examined large numbers of *Mansonia* caught in human bait net-traps in Pahang, Perak, Sabak Bernam, Malaya. *M. indiana*, *M. dives/bonneae* and *M. uniformis* were reported as the principal vectors of *B. malayi*. The majority of the infections were found in *M. dives/bonneae*. Hodgkin used five species of *Mansonia* in feeding experiments. In *M. annulata* development of the larvae was more rapid than in *M. uniformis* or *M. dives/bonneae*, but the latter was the more important vector in nature.

In *M. annulifera* a few stage III larvae were found on the 10th day but many larvae were still immature on the 17th day. In *M. indiana* from two localities, development varied considerably.

Wharton, 1962 and Edeson and Wilson, 1964 listed *M. annulata*, *M. bonneae*, *M. dives* and *M. uniformis* as the confirmed vectors of sub-periodic *B. malayi* in Malaya.

Periodic Brugia malayi—Post-war investigations in the coastal rice fields of Kedah and Penang showed that *M. uniformis* and *M. indiana* were among the chief vectors of periodic *B. malayi*; *M. annulifera* was possibly a secondary vector (Reid, Wilson and Ganapathipillai, 1962).

INDIA

Periodic B. malayi—Iyengar, 1938 found that *M. annulifera*, *M. uniformis* and *M. indiana* were natural vectors in Travancore, India. Only *M. annulifera* showed a high incidence of infection and this was also the most abundant mosquito. In the laboratory Iyengar found *M. uniformis* to be a favourable host, since the development of the parasite proceeded much more rapidly and the intensity of infection was much heavier than in *M. annulifera*. Raghavan, Nair & Krishnan, 1958 during their filariasis survey in 1956-1957 of the hill tribe settlements in Quinlon and Trivandrum districts, India, found *M. uniformis* to be the vector of *B. malayi* infections. Nair and Roy, 1958 during the filaria survey of Eriyad Panchayat, incriminated *M. annulifera* as the main vector of *B. malayi* with *M. uniformis* playing a secondary role. Nair, Krishnan and Roy, 1959 on the basis of filaria survey of Edapally Panchayat (Ernakulum district) reported *B. malayi* in all positive human cases but one. The exception was a case with *W. bancrofti* infection. *M. annulifera* was the only vector in the area, with a total larval rate of 12.9 per cent and a mature larval rate of 3.2 per cent. Rajindar Pal *et al.*, 1960 on the basis of their dissection results of *M. annulifera* and *M. uniformis*, collected over a period of 12 months from Ernakulum, Kerala, found that the total larval rate in *M. annulifera* was much higher than in *M. uniformis*. The total larval rate in the former was 27.8 per cent while in the latter it was 5.9 per cent. Joseph Menon and Nair, 1960 reported that the primary vector in *B. malayi*-infected areas of Kerala, was *M. annulifera*; *M. uniformis* was also a good vector but played only a secondary role in transmission because of its small numbers and its preference for cattle blood. Beye and Gurian, 1960 reported that the major vectors of *B. malayi* in India included *M. annulifera*, *M. annulata*, *M. dives/bonneae*, *M. indiana*, *M. indica* and *M. uniformis*.

CEYLON

Carter, 1948 reported that from the known endemic foci of *B. malayi* in North Western and Eastern provinces of Ceylon, infections were found in 15 species of mosquitoes, but a large proportion (about two-thirds) occurred in species of *Mansonia*. Chow, Thewasagayam and Wamback, 1955 confirmed the conclusions of Carter that *Mansonia* mosquitoes were the chief vectors of rural filariasis, and Cooray, 1960 reported that *M. uniformis* and *M. annulifera* were the vectors.

THAILAND

Iyengar, 1953 reported four species of *Mansonia*, *M. annulifera*, *M. uniformis*, *M. indiana* and *M. 'longipalpis'* as vectors of *B. malayi* in the eastern coastal belt of peninsular Thailand. They were found naturally infected with filarial larvae, probably *B. malayi*.

INDONESIA

Klokke, 1961 reporting on filariasis due to *B. malayi* in South Borneo, found that of the four species of *Mansonia* which he examined, namely *M. ochracea*, *M. 'longipalpis'*, *M. annulata*, and *M. uniformis*, mature larvae were present in *M. ochracea*, *M. 'longipalpis'* and *M. annulata*.

PHILLIPINE ISLANDS

Cabrera and Rozeboom, 1964 reported that of the 2,062 mosquitoes collected in Marriong, Kambing and Gungnan, Phillipine Islands, 48 per cent were identified to be *M. dives*. Of these, a total of 0.5 per cent had developing stage II and stage III larvae of *B. malayi*.

Wharton, 1962 summed the role of *Mansonia* mosquitoes as vectors of *Brugia* as follows "the evidence that *Mansonia* are the chief vectors of *B. malayi* and the dominant vectors of *B. pahangi* and *Dirofilaria* infections that occur in the swamp forest of Malaya appears to be quite conclusive....The main facts are indisputable, *M. dives/bonneae* and *M. annulata* are the chief vectors of *B. malayi* in some but not all parts of Indonesia; the same species are responsible for transmission in the swamp forest areas of Malaya, and probably Borneo and Thailand, while *M. uniformis*, *M. annulifera* and *M. indiana* are the main vectors in Ceylon and in India."

AEDES

In the genus *Aedes*, *Ae. togoi* is the only confirmed natural vector of filariasis due to *B. malayi*. *Ae. togoi* is distributed in the northern part of the Oriental region, *e.g.*, in Japan, Korea, China and Taiwan.

Omori, 1962 reviewing the role of mosquitoes in the transmission of Malayan and Bancroftian filariasis in Japan, stated that Malayan filariasis is found in Japan, only on the islet of Hachi-jo-koshima and is transmitted there by *Ae. togoi* and probably by *Culex pipiens pallens*, as the nematode could reach maturity in both mosquitoes. Previous workers had concluded that *Ae. togoi.* was the main vector of Malayan filariasis in Japan (Sasa *et al.*, 1952; Sasa *et al.*, 1959; and Sasa, Hayashi and Tanka, 1960).

Gun, 1960 reported *Ae. togoi* to be a natural host of *B. malayi* and *W. bancrofti* in China; 4.4 per cent of 613 mosquitoes examined in an endemic area of human filariasis contained larvae at various stages of development. Half of the infected mosquitoes contained larvae of *W. bancrofti* and the other half of *B. malayi.* Two per cent of the 613 mosquitoes had mature larvae. Following experimental infection of *Ae. togoi*, 4 out of 21 mosquitoes had mature larvae and one had mature larvae of *B. malayi* and one had immature larvae of *W. bancrofti*; 9 out of 16 mosquitoes had mature larvae of *B. malayi* and 4 had immature larvae.

In Liverpool, Nair 1962 demonstrated *Ae. togoi* to be an efficient laboratory vector of *B. pahangi* and 88 per cent of the mosquitoes tested had become infective. Zaini, 1962 made observations on the same strain of *Ae. togoi* and found that 87 per cent supported development of sub-periodic *B. malayi* to the mature form. Hussain, 1964 (personal communication) stated that about 55 per cent of *Ae. togoi* became infective with sub-periodic *B. malayi.* Experimental infection data of Ahmed, 1965 (under publication) on *Ae. togoi* infected with sub-periodic *B. malayi* and *B. pahangi* indicated that about 25 per cent of *Ae. togoi* became infected with sub-periodic *B. malayi* and about 34 per cent became infected with *B. pahangi.* Ramachandran *et al.*, 1963 have shown that *Ae. togoi* mosquitoes can act as laboratory vectors for the following species of filarial parasites: *B. pahangi*, sub-periodic *B. malayi*, periodic *B. malayi*, Malayan rural strain of *W. bancrofti*, *Dirofilaria immitis*, a *Breinlia* species from the Malayan slow loris, and a *Setaria* sp. from the Malayan mouse deer.

Ae. aegypti is not so far as is known a natural vector of human filariasis but in the laboratory this species has been a convenient experimental vector.

Galliard, 1947 investigated the role of *Ae. aegypti* and *Ae. albopictus* as experimental vectors of *B. malayi* in Tonkin, North Vietnam. *B. malayi* developed completely in a small percentage of mosquitoes. Both species were recorded as new carriers of *B. malayi*, this being the first record of a human filaria developing to the mature stage in *Ae. albopictus.* Edeson, Wharton and Laing, 1960 showed in feeding experiments conducted in Malaya that a small portion of Malayan

Ae. aegypti supported development of *B. pahangi* to the mature stage, Ramachandran, Edeson and Kershaw, 1960 showed that the Liverpool strain of *Ae. aegypti* was a moderately susceptible host of both the periodic and sub-periodic forms of *B. malayi*; about 30 per cent of the mosquitoes became infective after being fed on an infected cat. Macdonald, 1962 *a* therefore conducted experiments to improve the infection rates in mosquitoes by selective breeding. By rearing the offsprings of individuals which were infective, the infection rate of the first filial generation was raised to more than 90 per cent. This rate was maintained in the second and third filial generations, again by selective breeding from selected individual mosquitoes. This strain was maintained through 15 generations, with a mean infective rate of 84.8 per cent. A series of crosses and backcrosses was made between this highly susceptible strain of *Ae. aegypti* and naturally refractory strains from Rangoon, Malaya and Trinidad. The offsprings of the crosses were tested for susceptibility to infection. The results showed that susceptibility of *Ae. aegypti* to sub-periodic *B. malayi* is controlled by a sex linked recessive gene (Macdonald, 1962 *b*). Macdonald and Ramachandran (1965) showed that the selected strain of *Ae. aegypti* susceptible to infection with sub-periodic *B. malayi* was equally susceptible to infection with *B. pahangi*, periodic *B. malayi*, periodic *W. bancrofti* (Malayan rural strain) and sub-periodic *W. bancrofti* (Fijian strain). A selected strain refractory to infection with sub-periodic *B. malayi* proved refractory also to the other filariod species. They also tested the F2 hybrid offspring, from reciprocal crosses between the parent stocks, and from the results concluded that the gene controlling susceptibility to sub-periodic *B. malayi* also controlled susceptibility to the other *Brugia* and *Wuchereria* species tested. The gene had no influence on the susceptibility of the mosquitoes to infection with *Dirofilaria immitis* and *D. repens*.

ANOPHELES

B. pahangi—Anopheles mosquitoes have not been incriminated as vectors of *B. pahangi* in nature but some species have proved good laboratory hosts. Edeson, Wharton and Laing, 1960, showed in the laboratory that *A. barbirostris* and *A. umbrosus* supported development of *B. pahangi* to the infective stage. Wharton, 1962 fed *A. barbirostris*, *A. campestris* and *A. donaldi* on *B. pahangi* carriers. All three species supported development of *B. pahangi* but none was a good host, though *A. barbirostris* and *A. donaldi* appeared to be more susceptible than *A. campestris*. Schacher, 1961 tested eight species of mosquitoes including *A. quadrimaculatus* and *A. bradleyi*. Both the latter proved susceptible, although *A. bradleyi* was a good host it proved difficult to maintain in the laboratory.

CHINA

B. malayi—Grundi, 1945 compiled a list of the *Anopheles* concerned with the transmission of disease in man, and listed *A. barbirostris* as the vector of filariasis in China. Feng, 1934, 1935, 1936; Senoo and Lincome, 1951, Wu, 1959 and Piao *et al.*, 1959 have stated that *A. hyrcanus* var. *sinensis* is the chief transmitter of Malayan filariasis in China.

Feng and Ma, 1957 considered *A. hyrcanus* var. *sinensis* to be the main species responsible for transmitting *B. malayi* in nature and proposed the theory that Malayan filariasis in the hilly regions of South China was probably due to the presence of a race of *A. hyrcanus* var. *sinensis* peculiar to that region. Feng, Ma and Liu, 1958 and Kug-Chien-Chang *et al.*, 1959 confirmed this theory and reported that *A. hyrcanus* var. *sinensis* comprised at least of two forms, which were morphologically distinguishable from each other in the egg stage. The first or wide form had wide decked eggs, the adults being referred to as the large race, and the second or narrow form had narrow decked eggs, the adults being referred to as the small race. In morphology the former closely resembled or was identical to *A. hyrcanus* var. *sinensis*, while the latter was similar to *A. lesteri*. Complete development of the filariae took place in both the large and the small race of *A. hyrcanus* var. *sinensis* but Feng, Ma and Liu, 1958 were of the opinion that as the small race was anthropophilic and the large race zoophilic, therefore the narrow form or small race was chiefly responsible for Malayan filariasis in the mountainous districts of South China. Kung-Chien-Chang *et al.*, 1959 suggested that the wide form or large race was probably an important vector of *B. malayi* in the areas around the lakes and rivers of South China. Experimental infections of both forms demonstrated that both were susceptible to *B. malayi*, the filarial larvae developing to the mature stage in each. Edeson and Wilson, 1964 listed both *A. hyrcanus* var. *sinensis* and *A. lesteri* as the confirmed vectors of *B. malayi* in China.

MALAYA

Hodgkin, 1938, 1939, 1940 examined 422 specimens of three species of *A. barbirostris*, *A. hyrcanus*, *A. umbrosus* of *Anopheles* caught in human-bait net-traps in Pahang, Perak and Sabak Bernam, Malaya. Of the *A. umbrosus*/*A. letifer* dissected, only one was positive for mature larvae. Hodgkin also fed *A. barbirostris* in the laboratory on a *B. malayi* donor, and found only three per cent infected. Reid, 1950 in experimental infections found 89 per cent of *A. barbirostris* with *B. malayi*. Wharton, 1962 fed *A. barbirostris*, *A. campestris*, and *A. donaldi* on sub-periodic *B. malayi* carriers. With respect to sub-periodic *B. malayi* he found that all three species were poor vectors, and *A. donaldi* was completely refractory.

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However, in the case of periodic *B. malayi* all three species were good hosts. Wharton's results explain the discrepancies between those of Hodgkin and Reid. Reid, Wilson and Ganapathipillai, 1962 reported *A. campestris* as one of the main vectors of periodic *B. malayi* in Malaya.

INDONESIA

Brug, 1939 studied experimental infection of *A. barbirostris* with *B. malayi* in the Celebes, and he found 100 per cent became infected. Kariadi, 1941 found that a variety X of *A. hyrcanus* was an efficient carrier of *B. malayi* in Martapoera in the Dutch Indies.

THAILAND

Iyengar, 1953 reported *A. barbirostris, A. hyrcanus* var. *nigerrimus, A. hyrcanus* var. *sinensis, A. albotaeniatus*, and *A. umbrosus* as vectors of *B. malayi* in Thailand.

Other Countries of Asia

Grundi, 1945 listed *A. barbirostris* as a vector of filariasis in Japan, Polynesian Oceania, and India. Sen, 1956 in epidemiological investigations of the filariasis problem in West Bengal, India, reported for the first time the Indian form of the species of *A. hyrcanus* var. *nigerrimus* as the vector of filariasis; Sen also found *A. philippensis* among the anophelines infected in nature. Niles, 1961 showed in Ceylon that *B. malayi* of man and *Setaria digittata* of cows develop in *A. hyrcanus* var. *nigerrimus*.

Other Species of Mosquitoes.

Edeson, Wharton and Laing, 1960; Wilson, 1961 and Wharton, 1962 have all reported *Armigeres subalbatus* to be a very efficient laboratory host of *B. pahangi*, but not a good host for periodic or sub-periodic *B. malayi*.

In the subgenus *Conquillettidia* of *Mansonia*, Wharton, 1962 from experimental work reported that *M. nigrosignata, M. hodgkini* and *M. ochracea* did not appear to be hosts of *B. pahangi* but *M. crassipes* and *M. aurosquammata* both produced mature larvae and might therefore be vectors in nature.

Edeson, Wharton and Laing, 1960 were able to infect 9 out of 121 (7 per cent) *Culex pipiens fatigans* with *B. pahangi*. Desowitz and Chellapah, 1962 tested a laboratory bred Singapore strain of *C. p. fatigans* for susceptibility to infection with *B. pahangi*. They studied the effects of age and prior non-infecting blood meals on the infection rate of the mosquitoes. They fed groups of mosquitoes of different ages on cats naturally infected with *Brugia* sp. (probably *B. pahangi*). These mosquitoes were dissected 14 days after the infecting feed, and only when

stage III larvae were found in the proboscis was the mosquito regarded as being positive. The average infection rates in the various groups of mosquitoes ranged from 8.5 to 25.4 per cent. Prior non-infecting blood meals did not have an effect on infection rates in younger mosquitoes, but consistently high infection rates were obtained in older *C. p. fatigans* which had been given prior non-infecting blood meals. The infection rates according to the authors were not as high as in *Mansonia* or *Armigeres*, but nevertheless were sufficient to transmit the infection from cat to cat with ease.

Schacher, 1961 tried feeding field-caught *Culex erraticus* and *Orthopodomyia signifera* in New Orleans (U.S.A.) but both refused to take a blood meal from cats.

Filariasis in Pakistan

Foote and Cook, 1959 reported that no information was available on filariasis in West Pakistan. The author has also not come across any published data on filariasis due to *Wuchereria* and *Brugia* species in this wing of the country. No current published data is available on filariasis due to *Wuchereria* and *Brugia* species in East Pakistan. Foote and Cooke, 1959 however recorded that *B. malayi* which causes most of the infections is transmitted by *Mansonia annulifera*, *M. uniformis* and *M. indiana*. All these three species were found to have high natural infection rates.

Sandosham, Wharton, Warren and Eyles, 1962 reported the presence of *B. malayi* microfilariae in the blood of a rehsus monkey from Chittagong (East Pakistan) *Mansonia uniformis* was suspected to be the vector.

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