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Research Note

Transmission potential of *Wuchereria bancrofti* by *Culex quinquefasciatus* in urban areas of Malaysia

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Abstract. Laboratory strain of the Malaysian *Culex quinquefasciatus* was susceptible to *Wuchereria bancrofti*. Thirty three percent of the *Cx. quinquefasciatus* that fed on *W. bancrofti* patient were infective after 12-14 days. There is a possibility for *W. bancrofti* to occur in the urban areas of the Malaysia in the near future.

Lymphatic filariasis caused by the nematode parasite *Wuchereria bancrofti* (WB), is not a public health problem in Malaysia. In many parts of the world *W. bancrofti* is seen mainly in urban centres and is transmitted by *Culex quinquefasciatus* which breeds in polluted waters (Mak, 1986). Currently more than 120 million people are infected in 83 countries (WHO, 2000). With rapid transportation and transmigration of people it is possible for the parasite to be reintroduced into the country. The Global programme For The Elimination of Lymphatic Filariasis Program (GPELF) hopes to eliminate filariasis world wide by 2020. However, with the influx of migrant workers coming into Malaysia from WB endemic areas we have to be more vigilant in screening these workers.

Here we report recent findings of the susceptibility status of both insecticide susceptible and resistant strain (to malathion) of *Cx. quinquefasciatus* to *W. bancrofti*. A recent case of microfilaria was reported to us from Negeri Sembilan. The patient is a Mayanmar national and the mf count was 4Wb/60ul blood. The patient consented to providing blood for feeding mosquitoes. Fifty insecticide resistant *Cx. quinquefasciatus* and 50 insecticide susceptible strain of the mosquitoes were fed on infected blood. Immediately after feeding 8 mosquitoes were frozen to carry out PCR. All blood fed mosquitoes were maintained for at least 14 days after which they were dissected.

DNA was extracted from patient blood, mosquitoes day 0 and on microfilaria found in the mosquitoes using the Qiagen DNeasy® Kit. Polymerase Chain Reaction (PCR) amplification of *W. bancrofti* Ssp1 repeat was carried out as described by Ramzy *et al.* (1997) with slight modification. Each amplification reaction was performed in a final of volume of 50ul containing 1.5mM $MgCl_2$, 150uM of each deoxynucleotide triphosphate (Biotools®, Madrid, Spain), 10 pmol each of NV-1 and NV-2 oligonucleotide primers and 1.5 U of *Tth* DNA polymerase (Biotools®, Madrid, Spain). Ten microliter of each PCR product was electrophoresed on a 2% agarose gel stained with Gel Star® Nucleic Acid Stain (Cambrex, Rockland, ME).

Table 1. Susceptibility status of *Cx. quinquefasciatus* to *W. bancrofti*

Strain of *Cx. quinquefasciatus*	No. of mosquitoes fed	No. of mosquitoes dissected	No. of mosquitoes with infective larvae (%)	Total L3 larvae
Insecticide resistance strain	24	5	1 (20)	4
Insecticide susceptible strain	29	15	5 (33.33)	18

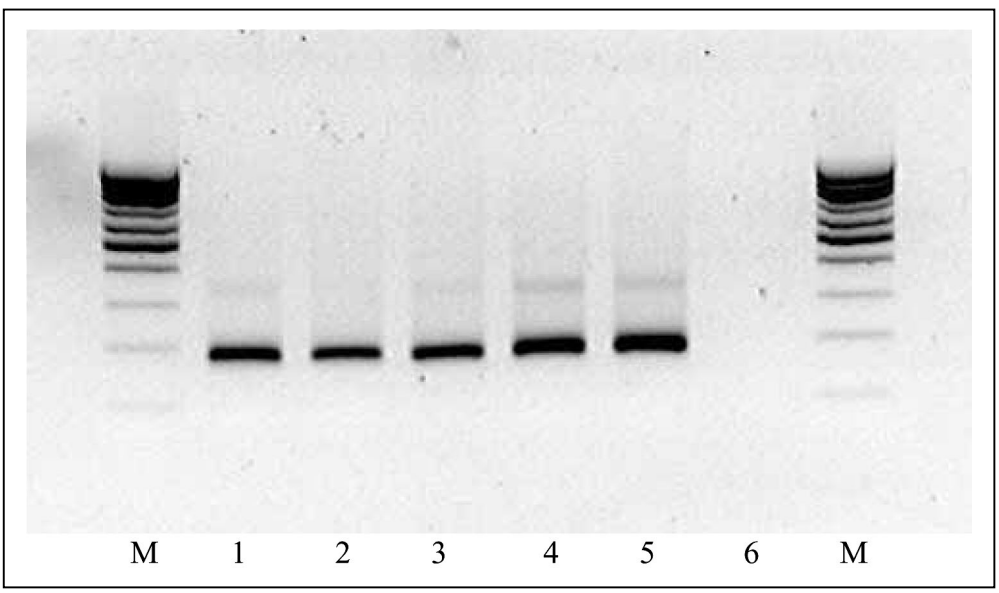

Figure 1. Detection of *W. bancrofti* DNA *Ssp1* by PCR assay. M, a 100-basepair ladder (molecular size maker); Lane 1, positive control; Lane 2, Patient blood; Lane 3, *C. quinquefasciatus* day 0, Lane 4, L3 larva from susceptible strain; Lane 5, L3 larva from resistant strain; Lane 6, negative control.

Of the fifty insecticide resistant *Cx. quinquefasciatus* only 24 were fully blood fed. Five survived until day 12 and only one mosquito was positive for L3 Wb (Table 1). Twenty nine insecticide susceptible *Cx. quinquefasciatus* were fully blood fed and of these 15 survived until day 14. Of these 5 were positive with L3 larvae in the head, giving an infective rate of 33.3 (Table 1). Fig. 1 shows the PCR product from patient and mosquitoes. All samples positive for Wb showed a PCR product of 188bp.

From this brief study it can be said that there is a risk of Wb spreading in the urban areas of Malaysia. According to Mc Carroll *et al.* (2000) insecticide resistant *Cx. quinuqefasciatus* are less likely to transmit filariasis and in their study showed that laboratory strain of

insecticide resistant *Cx. quinquefasciatus* did not produce filarial larvae while 76% of the susceptible strain was positive.

Studies carried out by Ramachandran *et al.* (1964) in an aborigine village about 12 miles away from the city of Kuala Lumpur showed that *W. bancrofti* were present in the aborigine population. Thomas & Ramachandran (1970) also conducted experimental studies with various strains of *Cx. quinquefasciatus* on *W. bancrofti* carrier and found that all strains were capable of supporting development of microfilariae to the infective stage.

Forty years later we still find *Cx. quinquefasciatus* capable of developing the *W. bancrofti* to the infective stage. Our neighbouring country Thailand has reported an influx of more that one million Myanmar immigrants into urban centres of their country and of these 2–5% had patent *W. bancrofti* (Triteeraprapab *et al.*, 2000). Thus there is also fear in Thailand of the re-emergence of urban filariasis. Transmission can occur if there is a close relationship between the host, parasite and vector. Here both the host and vector are present in abundance and population movement across international boundaries will bring in the parasite. Thus we conclude that there may be a potential for an established urban transmission of *W. bancrofti* in this country in the near future. In view of this, the screening of all immigrant workers into this country for filariasis at the entry point is very important to avoid re-emergence of diseases.

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