ANGIOSTRONGYLUS INFECTIONS IN RATS AND SNAILS IN NORTHEAST THAILAND

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Abstract. A survey of *Angiostrongylus* infections in rats and snails was carried out in the provinces of Ubon Ratchathani, Udon Thani, Kalasin, Chaiyaphum and Khon Kaen in northeast Thailand. Only two *Rattus norvegicus* (3.8%) and one *Bandicota indica* (1.4%) out of 151 *R. rattus*, 52 *R. norvegicus* and 69 *B. indica* examined were infected with adult lung worms. All worms recovered were *A. cantonensis*. Prevalence of infection in 423 *Pila polita* was 0.9% while all of 77 *P. ampullacea* were negative for larvae. In contrast to this 36.4% of 500 *Achatina fulica* harbored L_3 of *Angiostrongylus* (with variations of between 29% and 46% in the five provinces). The average infection intensity in *A. fulica* was 13.6 L_3 (1 to 441). Experimental infection of Wistar rats with L_3 (isolated from *A. fulica* resulted in a recovery rate of 48.3% of adult worms of which 91.7% and 8.3% were identified as *A. cantonensis* and *A. malaysiensis*, respectively. This is the first proven finding of *A. malaysiensis* in northeast Thailand.

INTRODUCTION

Among the 20 species of the nematode genus *Angiostrongylus* described so far (Bhaibulaya, 1991), three are known to occur in Thailand. The most important is *A. cantonensis*, the larvae of which are the causative agents of human eosinophilic meningoencephalitis. The distribution of this species covers all parts of the country. This is different with the other two representatives of the genus in Thailand: *A. malaysiensis*, a sibling species of *A. cantonensis*, was found only in the central and western regions (Bhaibulaya and Techasoponmani, 1972; Jeradit, 1977), whereas *A. siamensis* is known only from Nakhon Nayok Province (Kamiya *et al*, 1979; Ohbayashi *et al*, 1979, 1983; Ohbayashi, 1988).

The main definitive hosts of all three species are Muridae of the genera *Rattus* and *Bandicota*. The adult worms live either in the lung arteries and right ventricle (*A. cantonensis, A. malaysiensis*) or are located in the mesenteric arteries (*A. siamensis*) of their hosts. Several species of fresh water and land snails serve as intermediate hosts for *A. cantonensis* and *A. malaysiensis* with *Achatina fulica* as the most important for the maintenance of the parasites' life cycles and with apple snails of the genus *Pila* as the most important sources of human infection. The intermediate hosts of *A. siamensis* are not known yet.

The northeast of Thailand shows the highest incidence of human eosinophilic meningoencephalitis within the country (Punyagupta *et al*, 1970). Results on *A. cantonensis* infections of definitive and/or intermediate hosts are available for 15 of the former 17 provinces (Harinasuta *et al*, 1965, 1970; Crook *et al*, 1968; Punyagupta *et al*, 1970; Jeradit, 1977; Panha, 1988), however nothing is yet known about the role *A. fulica* may play there in the epidemiology of the parasite and whether or not *A. malaysiensis* and *A. siamensis* also occur in this region. To close this deficit a field survey was carried out in five provinces of the northeast.

MATERIALS AND METHODS

The following five provinces were chosen randomly: Ubon Ratchathani, Udon Thani, Kalasin, Chaiyaphum and Khon Kaen. In each of these provinces 100 *Pila* spp 100 *A. fulica* and about 50 to 60 rodents were sampled. Snails were digested by pepsin-HCI and *Angiostrongylus* larvae (L_3) suspended in 0.85% NaCI and counted. Rats were dissected, their lung arteries, right ventricle and mesenteric arteries inspected, adult worms removed and identified under the microscope. Identification of L_3 isolated from *A. fulica*, was carried out by infecting five Wistar rats each for isolates from the five provinces by oral application (stomach tube) of 20 larvae per rat. Dissection of rats was performed six weeks after infection using the same procedures as with the naturally infected rodents.

RESULTS

Infection in rats

Altogether 272 rats (151 *Rattus rattus*, 52 *R. norvegicus*, 69 *Bandicota indica*) were examined for *Angiostrongylus* infections (Table 1). The prevalence was very low (Fig 1) as only 2 *R. norvegicus* (3.8%) and 1 *B.indica* (1.4%) carried adult worms, all being *A.cantonensis*. The same is true for the infection intensity. One *R. norvegicus* from Ubon Ratchathani harbored 4 males and 4 females of the parasite, another from Khon Kaen 2 males and 1 female, while in the single *B. indica* from Udon Thani only one female worm was found.

Infection in snails

Of 500 *Pila* snails examined only 4 out of 423 *P. polita* (0.9%) contained third-stage larvae (L_3), whereas all of 77 *P.ampullacea* were negative for L_3 (Table 2).

In *A. fulica* prevalence was much higher with an average of 36.4%. There were variations between the five provinces (Table 2, Fig 1), prevalence in Khon Kaen (29%) being significantly lower ($p<0.05$) if compared to Chaiyaphum (46%) and Ubon Ratchathani (44%). The infection intensity in *A. fulica* varied between 1 and 441 L_3 with an average of 13.6 larvae per snail ($m_g = 0.78$). There was no direct correlation between size of the snail and infection intensity. The number of larvae found correlated well with the number of snails in the respective size class ($r = 0.945$; Fig 2). There is no evidence of older snails accumulating higher numbers of L_3.

Experimental infection of rats

Of the 25 rats infected experimentally 20 survived until dissection took place six weeks p.i. Altogether 193 adult *Angiostrongylus* (84 males, 109 females) were recovered (recovery rate = 48.25%). Of these, 177 worms were identified as *A. cantonensis* (77 males, 100 females) and 16 as *A. malaysiensis* (7 males, 9 females). The latter species was detected only in rats which infected with larvae isolated from *A. fulica* collected in Khon Kaen. All of these rats showed mixed infections of both *Angiostrongylus* species.

DISCUSSION

The occurrence of *A. malaysiensis* in northeastern Thailand has been established for the first time. *A. malaysiensis* was discovered in Peninsular Malaysia (Bhaibulaya and Cross, 1971), where it has a wide distribution (Lim and Ramachandran, 1979; Lim and Mak, 1983). Later it was also found in western and central Thailand (Bhaibulaya and Techasoponmani, 1972; Jeradit, 1977), Indonesia (Carney and Stafford, 1979) and Japan (Makiya and Sawabe, 1992). One can therefore assume that its distribution includes other parts of Southeast and East Asia. Prior to the discovery of *A. malaysiensis*, adults of the genus *Angiostrongylus* which were found in the lung arteries and the right ventricle of rats and larvae which were isolated from snails were thought to be solely *A. cantonensis*. Most probably this was not the case. Consequently, results published before 1971 have to be revised and the diagnosis "*A. cantonensis*" be replaced by "*A. cantonensis*" and/or "*A. malaysiensis*".

If compared with surveys carried out 25 to 30 years ago (Harinasuta *et al*, 1965, 1970; Crook *et al*, 1968), our study exhibits a much lower prevalence in the definitive hosts. However, infections in apple snails of the genus *Pila* conform roughly to the results of other authors (Harinasuta *et al*, 1965, 1970; Crook *et al* 1968, Punyagupta *et al*, 1970; Jeradit, 1977). Up to 1977 an average of 1.4% of *P. polita* harbored *Angiostrongylus* larvae, while in our own findings the prevalence in this species came to 0.95%.

Table 1

Angiostrongylus cantonensis infection in rodents in five provinces of Northeast Thailand.

Province	*Rattus rattus*		*Rattus norvegicus*		*Bandicota indica*		Total	
	No. exam	No. pos	No. exam	No. pos	No. exam	No. pos	No. exam	No. pos
Ubon Ratchathani	16	0	36	1	0	-	52	1
Udon Thani	45	0	0	-	9	1	54	1
Kalasin	43	0	0	-	18	0	61	0
Chaiyaphum	10	0	0	-	38	0	48	0
Khon Kaen	37	0	16	1	4	0	57	1
Total	151	0	52	2	69	1	272	3

Table 2

Angiostrongylus infection in snails in five provinces of Northeast Thailand.

Province	*Pila polita*		*Pila ampullacea*		*Achatina fulica*	
	No. exam	No. pos	No. exam	No. pos	No. exam	No. pos
Ubon Ratchathani	100	2	0	-	100	44
Udon Thani	100	0	0	-	100	30
Kalasin	100	2	0	-	100	33
Chaiyaphum	54	0	46	0	100	46
Khon Kaen	69	0	31	0	100	29
Total	423	4	77	0	500	182

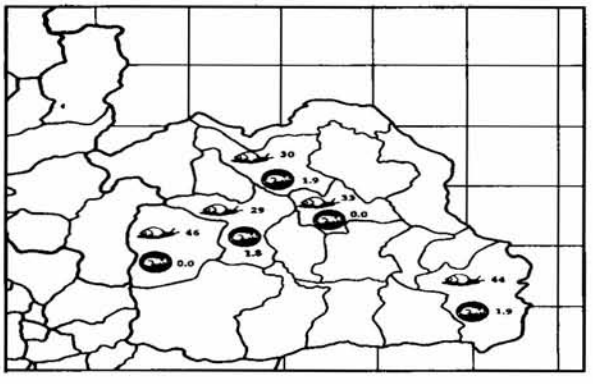

Fig 1 - *Angiostrongylus* infection (%) in rats and snails in five provinces of northeast Thailand.

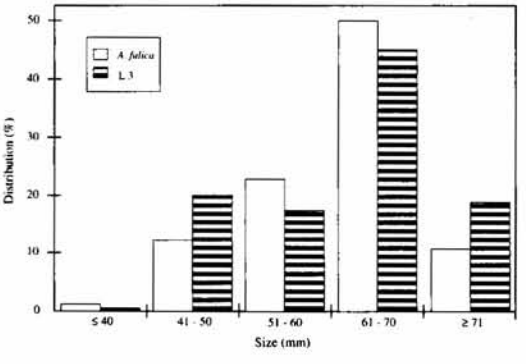

Fig 2 - Percentage distribution of 500 *Achatina fulica* according to size (mm) and of 2,482 *Angiostrongylus* larvae (L3) in these snails.

With regard to *A. fulica,* this snail species has not been examined before in northeast Thailand. Our results now show that since its spread into this region, it has become an important intermediate hast of *Angiostrongylus* with an average prevalence of 36.4%. Furthermore it carries L_3 not only of *A. cantonensis* but also of *A. malaysiensis*. In prevalence it is only surpassed by *Hemiplecta distincta* with 67 out of 137 (= 48.9%) found infected in the five provinces concerned (Panha, 1988). Thus, *A. fulica* and *H. distincta* have to be considered as most important for the maintenance of the life cycle of *Angiostrongylus* in northeast Thailand.

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