

## Studies on Schistosomiasis in the Mekong Basin

### III. Prevalence of *Schistosoma* Infection among the Inhabitants

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#### Introduction

Since the discovery of the first case of schistosomiasis by vic Dupond in Laotian subjects in 1957, much attention has been focused on the countries in the Mekong Basin. Chaiyaporn *et al.* (1959), Komiya (1961), and Harinasuta *et al.* (1962a, 1962b) reported several cases of schistosomiasis in Thailand. In the years of 1963-1966, Barbier found 5 cases of the disease among the persons from Laos (Barbier & Brumpt, 1969). In Cambodia, a few cases were also discovered by staff of Pasteur Institute (Velimirovic, 1971).

On the other hand, ECAFE plans to undertake extensive irrigation schemes in the Basin. The investigation to determine the presence of schistosomiasis in the areas is considered to be necessary. Because, if the infection were present, adequate measures should be then taken to prevent its further spread from foci of infection.

With a view to elucidate these matters, the present authors, in the capacity of the Short-Term Consultant of World Health

Organization, carried out the epidemiological surveys of schistosomiasis in the Mekong Basin in the years of 1966-1967 and 1968-1969.

The following is a detailed report on the investigation of the prevalence of the disease.

#### Materials and Methods

In selection of areas to be surveyed, the irrigation schemes of ECAFE was taken into consideration. In the 1966-1967 survey, the investigation was carried out at 5 villages, Muong Khong, M. Sene, Ban Na, B. Dong and Khong Nhai, on Khong Island. In the survey of 1968-1969, the areas selected for the survey are as follows (Fig. 1):

Laos: Vientiane District (B. Bo-o, B. Sithan Tay and B. Tha Deua), Pakse District (Pakse City, M. Kao, B. Nakhong, B. Nong Hoi, B. Phack Pheo, B. Mo Pheo, B. Pakson, B. Keng Xane and B. Photak) and Khong District (M. Khong and B. Hatxaykhoun).  
Cambodia: Stung Treng District (B. Bachon, B. Khan Din and B. Khan Memay),  
Kratie District (Kratie City, Phou Samboc,

Regarding the morphology of adult worm of *Schistosoma*, their reservoir, and intermediate hosts, reports have been published in the Japanese Journal of Parasitology, 1970, 20, 24-33, and in the Chinese Journal of Microbiology, 1971, 4, 168-181.

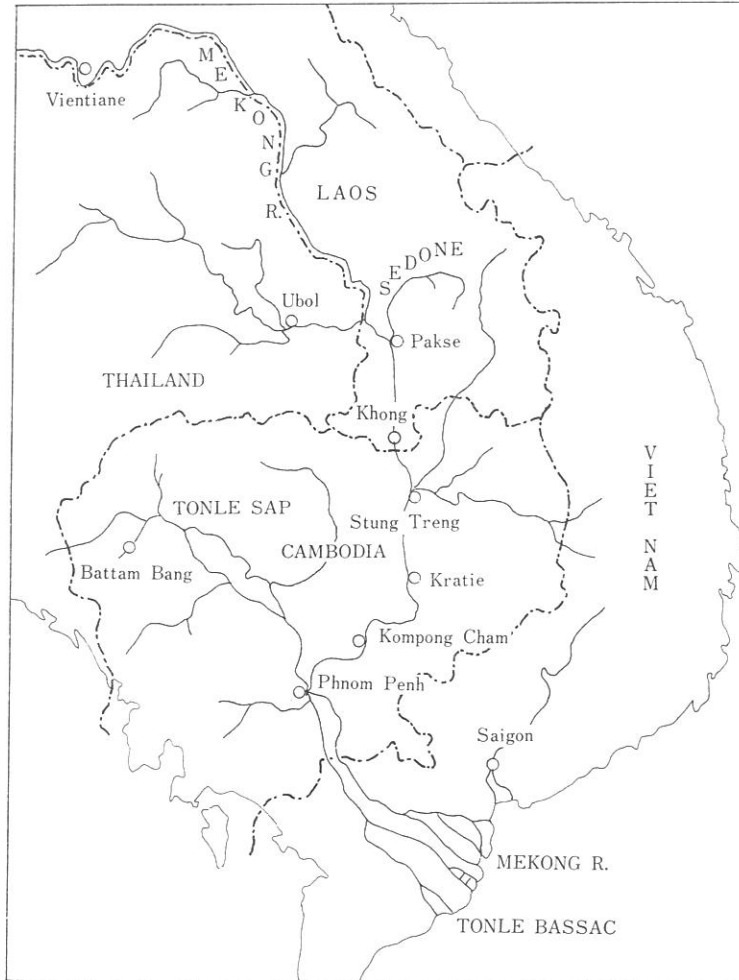


Fig. 1 General map of the Mekong Basin.

and Ph. Roka Kandal), Kompong Cham District (K. Cham City, Ph. Ro Ang and Ph. Kaphen) and Bassac-Lower Mekong District (Ph. Sethbo, Ph. Chhoung Leap, Ph. Prek Ambel, Ph. Thang Thin, Ph. Sampan, Ph. Sam Rong Tham, Ph. Dey Dos and Ph. Sam Rong).

In the survey areas, primary-schoolchildren were principally chosen as the object of the examination.

For skin test, the antigen applied is an acid-soluble fraction ( $6 \mu\text{g}/\text{ml}$  of total protein), prepared from adult worms of *Schistosoma japonicum* by Veronal Buffered Saline (VBS) Method. At the forearm of the individual,

$0.02 \text{ ml}$  of the antigen was intradermally injected. The reaction was recorded 15 minutes afterward as each average of two rectangular crossed diameters of both erythema and swelling. A swelling larger than  $9 \text{ mm}$  and/or an erythema larger than  $20 \text{ mm}$  was recognized as positive, and a swelling of  $7\text{--}9.5 \text{ mm}$  and/or an erythema of  $15\text{--}19.5 \text{ mm}$  judged as doubtful.

Stools of those who showed a positive or doubtful reaction by the skin test were examined by direct smear method, because the equipment to carry out the examination by egg concentration method was not fully available. Two to 5 smears were examined

for each fecal specimen, and in most cases, 50-200 mg of the stool was used for the examination.

### Results

The results are shown in Tables 1-3.

#### Vientiane District :

Ban Bo-o, B. Sithan Tay and B. Tha Deua along the Mekong River, were selected for the survey. A total of 935 persons were tested by intradermal skin reaction. Number and percentages of the positive, doubtful and negative were 154 (16.5%), 20 (2.1%) and 761 (81.4%) respectively.

Stool examination was made on 121 persons who had been positive or doubtful by the skin test. And, *Schistosoma* eggs were detected in none of the cases.

#### Pakse District :

For the survey, Pakse City, Muong Kao, B. Nakhong, B. Nong Hoi, B. Phack Pheo and B. Mo Phou located along the Mekong River, and B. Pakson, B. Keng Xane and B. Photak along Se Done were selected. The result of skin test of 1,478 inhabitants was as follows: 200 (13.5%) positive, 20 (1.4%) doubtful and 1,258 (85.1%) negative.

Stool examination was performed on 203 persons. No schistosomiasis were found

from the native. However, 3 students in Lycée Collège were positive for the ova of *Schistosoma*.

#### Khong District :

In the 1966-1967 survey, M. Khong, M. Sene, B. Na, B. Done and Khong Nhai were selected. The results of the skin tests of 1,012 inhabitants were 184 (18.2%) positive, 319 (31.5%) doubtful and 509 (50.3%) negative.

A total of 547 inhabitants who received skin test, were examined by direct smear method, and 47 cases (8.59%) were found to be positive for ova of *Schistosoma*. The highest percentage of positive cases, 26.4%, was found in Muong Khong and the lowest, 0.65%, in Khong Nhai.

In the 1968-1969 survey, a total of 223 persons (71 Khong Primary-schoolchildren, 52 Khong Island inhabitants and 100 Hatxaykhoun Primary-schoolchildren) were examined by skin test. Positive reaction appeared in 62 (82.7%), 45 (86.5%) and 72 (72%) persons respectively.

Stool examination was performed on 52 Khong Primary-schoolchildren, 20 Khong Island inhabitants and 73 Hatxaykhoun Primary-schoolchildren. Among them, 22 (59.5%), 7 (35%) and 20 (27.4%) were posi-

Table 1 Results of intradermal skin test using *Schistosoma japonicum* antigen and stool examination (direct smear method) for *Schistosoma* of the inhabitants in Khong District, Laos in 1966-1967 survey.

District	City-Town Village (Object)	Intradermal skin test				Stool examination	
		Number Tested	Number Positive (%)	Number Doubtful (%)	Number Negative (%)	Number Examined	Number Positive (%)
KHONG	Muong Khong (Primary school)	360	88 (24.4)	182 (50.6)	90 (25.0)	136	36 (26.5)
	Ban Dong (Inhabitants)	124	5 (2.4)	23 (18.6)	98 (79.0)	46	1 (2.2)
	Ban Na (Inhabitants)	111	28 (25.2)	27 (24.3)	56 (50.5)	67	4 (6.0)
	Muong Sene (Primary school)	243	50 (20.5)	65 (26.8)	128 (52.7)	144	5 (3.5)
	Khong Nhai (Primary school)	174	15 (8.6)	22 (12.6)	137 (78.8)	154	1 (0.7)
	Total	1,012	184 (18.8)	319 (31.5)	509 (50.3)	547	47 (8.6)

Table 2 Results of intradermal skin test using *Schistosoma japonicum* antigen and stool examination (direct smear method) for *Schistosoma* of the inhabitants in three districts in Laos in 1968-1969 survey.

District	City-Town Village (Object)	Intradermal skin test				Stool examination		
		Number Tested	Number Positive (%)	Number Doubtful (%)	Number Negative (%)	Number Examined	Number Positive (%)	
VIENTIANE	Ban Bo-o (Primary school)	423	52 (12.3)	8 (1.9)	363 (85.8)	38	0 (0)	
	B. Bo-o (Inhabitants)	82	12 (14.6)	1 (1.2)	69 (84.2)	10	0 (0)	
	B. Sithan Tay (Primary school)	218	67 (30.7)	4 (1.8)	147 (67.5)	52	0 (0)	
	Tha Deua (Primary school)	212	23 (10.8)	7 (3.3)	182 (85.9)	21	0 (0)	
	Total	935	154 (16.5)	20 (2.1)	761 (81.4)	121	0 (0)	
PAKSE	Se Done	Pakse (Lycée College)	592	117 (19.8)	6 (1.0)	469 (79.2)	118	3 (2.5)
		Pakse (Inhabitants)	105	19 (18.1)	3 (2.9)	83 (79.0)	16	0 (0)
		Muong Kao (Primary school)	168	21 (12.5)	1 (0.6)	146 (86.9)	19	0 (0)
		B. Nakhong (Primary school)	140	16 (11.4)	2 (1.4)	122 (87.2)	7	0 (0)
		B. Nong Hoi (Primary school)	69	12 (17.4)	3 (4.3)	54 (78.3)	15	0 (0)
	Mekong	Phack Pheo (Inhabitants)	112	3 (2.7)	0 (0)	109 (97.3)	2	0 (0)
		B. Mo Phou (Primary school)	126	7 (5.6)	3 (2.4)	116 (92.0)	10	0 (0)
		B. Pakson (Primary school)	98	4 (4.1)	1 (1.0)	93 (94.9)	5	0 (0)
		B. Keng Xane (Primary school)	34	1 (2.9)	0 (0)	33 (97.1)	1	0 (0)
		B. Photak (Primary school)	34	0 (0)	1 (2.9)	33 (97.1)	0	—
Total	1,478	200 (13.5)	20 (1.4)	1,258 (85.1)	203	0 (0)		
KHONG	M. Khong (Primary school)	71	61 (85.9)	1 (1.4)	9 (12.7)	56	22 (39.3) (34.3)*	
	M. Khong (Inhabitants)	52	45 (86.5)	0 (0)	7 (13.5)	20	7 (35.0) (28.0)*	
	B. Hatxaykhoun (Primary school)	100	72 (72.)	3 (3.)	25 (25.)	73	20 (27.4) (20.6)*	
	Total	223	178 (79.8)	4 (1.8)	41 (18.4)	149	49 (32.9) (26.3)*	

\* Corrected percentage of *Schistosoma* infection for the total number in each group.

Table 3 Results of intradermal skin test using *Schistosoma japonicum* antigen and stool examination (direct smear method) for *Schistosoma* of the inhabitants in four districts in Cambodia in 1968-1969 survey.

District	City-Town Village (Object)	Intradermal skin test				Stool examination		
		Number Tested	Number Positive (%)	Number Doubtful (%)	Number Negative (%)	Number Examined	Number Positive (%)	
STUNG TRENG	B. Bachon (Inhabitants)	173	18 (10.4)	2 (1.2)	153 (88.4)	17	1 (5.9)	
	B. Khan Din (Inhabitants)	120	10 (8.3)	3 (2.5)	107 (89.2)	9	0 (0)	
	B. Khan Memay (Inhabitants)	155	33 (21.3)	2 (1.3)	120 (77.4)	29	1 (3.4)	
	Total	448	61 (13.6)	7 (1.6)	380 (84.8)	55	2 (3.6)	
KRATIE	Kratie (Floating village)	151	90 (59.6)	1 (0.7)	60 (39.7)	53	29 (54.7) (32.8)*	
	Phou Samboc (Inhabitants)	212	30 (14.2)	1 (0.5)	181 (85.3)	24	0 (0)	
	Ph. Roka Kandal (Inhabitants)	200	82 (41.0)	0 (0)	118 (59.0)	42	11 (26.2) (10.3)*	
	Total	563	202 (35.9)	2 (0.3)	359 (63.8)	119	40 (33.6) (13.5)*	
KOMPONG CHAM	Kompong Cham (Floating village)	41	5 (12.2)	1 (2.4)	35 (85.4)	6	0 (0)	
	K. Cham (Primary school)	366	9 (2.5)	3 (0.8)	354 (96.7)	11	0 (0)	
	Ph. Ro Ang (Floating village)	22	4 (18.2)	1 (4.5)	17 (77.3)	15	0 (0)	
	Ph. Ro Ang (Inhabitants)	139	13 (9.4)	4 (2.9)	122 (87.7)			
	Ph. Kaphen (Primary school)	93	9 (9.7)	1 (1.1)	83 (89.2)	8	0 (0)	
	Total	661	40 (6.1)	10 (1.5)	611 (92.4)	40	0 (0)	
	BASSAC-LOWER MEKONG	Tonle Bassac	Ph. Prek Ambel (Inhabitants)	126	10 (7.9)	0 (0)	116 (92.1)	7
Ph. Tong Thin (Primary school)			303	11 (3.6)	5 (1.7)	287 (94.7)	13	0 (0)
Ph. Sampan (Primary school)			276	28 (10.1)	4 (1.4)	244 (88.5)	29	0 (0)
Ph. Sethbo (Primary school)			316	33 (10.4)	5 (1.6)	278 (88.0)	32	0 (0)
Ph. Chhoung Leap (Primary school)			295	38 (12.9)	4 (1.4)	253 (85.7)	38	1 (2.6)
Mekong			Ph. Sam Rong Tham (Primary school)	320	27 (8.4)	5 (1.6)	288 (90.0)	31
		Ph. Dey Dos (Primary school)	176	11 (6.2)	1 (0.6)	164 (93.2)	10	0 (0)
		Ph. Sam Rong (Primary school)	283	19 (6.7)	4 (1.4)	260 (91.9)	9	0 (0)
		Total	2,095	177 (8.5)	28 (1.3)	1,890 (90.2)	169	1 (0.6)

\* Corrected percentage of *Schistosoma* infection for the total number in each group.

tive for the ova of *Schistosoma*. Corrected percentage of the infection for the whole number of persons living in each areas are 34.3%, 30.3% and 20.6% respectively.

Stung Treng :

The survey was carried out at B. Bachon, B. Khan Din and B. Khan Memay located along the Mekong River. A total of 448 persons were tested by skin reaction, and the results were 61 (13.1%) positive, 7 (1.6%) doubtful and 380 (85.3%) negative.

Stool examinations were performed for 56 persons who showed positive or doubtful reaction in the skin test. Only one person in B. Khan Memay and one in B. Bachon were found positive for the ova of *Schistosoma*.

Kratie District :

A total of 563 persons (212 Ph. Samboc inhabitants, 200 Ph. Roka Kandal inhabitants and 151 Floating villagers) were tested by intradermal skin test. Positive reaction appeared in 30 (14.2%), 82 (41%) and 90 (59.9%) in each area.

Stool examination was performed on 24 Ph. Samboc inhabitants, 42 Ph. Roka Kandal inhabitants and 53 Floating villagers. Among them, 11 (25.2%) and 29 (54.7%) cases were found to be positive for the ova of *Schistosoma* in Ph. Roka Kandal and Floating village respectively. Percentage of the infection for the total number of the inhabitants in each area are 10.3% and 32.8%.

Kompong Cham District :

For the survey, K. Cham City, Ph. Ro Ang and Ph. Kaphen were selected. The results of skin test of 661 inhabitants were 40 (6.5%) positive, 10 (1.7%) doubtful and 611 (91.8%) negative.

Stool examination was performed on 40 persons, and no positive cases were found.  
Bassac-Lower Mekong District :

In the district, Ph. Prek Ambel, Ph. Thong Thin, Ph. Sampan, Ph. Sethbo and Ph. Chhoung Leap located along Tonle Bassac, Ph. Sam Rong Tham, Ph. Dey Dos and Ph. Sam Rong along the Mekong River were selected for the survey. A total of 2,095 persons were tested by intradermal skin test, and the results were 117 (8.6%)

positive, 28 (1.3%) doubtful and 1,890 (90.1%) negative.

Stool examination was performed on 169 persons. Among them, only one pupil, 11 years old, was found to be positive for the ova of *Schistosoma*.

### Discussion

A good number of persons still showed positive skin reaction in areas where no cases of schistosomiasis were found. The present authors assumed that this positive reaction might be "non specific reaction" and/or "cross reaction" caused by infection with other parasites such as *Trichobilharzia* sp., *Orientobilharzia* sp. and *Schistosoma incognitum* and so on. This could be explained by differences in the rate of positivity according to locality and the fact that the majority of the positives complained of dermatitis and itching after going into the Mekong River.

On the other hand, the high degree of correlation between the positive skin reaction and the prevalence of schistosomiasis is obvious. Therefore, intradermal skin test was proved to be an efficient and appropriate way to screen suspected persons of the disease in this basin even if it may accompany "non specific reaction" and/or "cross reaction" in some cases.

No endemic foci of schistosomiasis were found in the Vientiane District.

In Pakse, no ova of *Schistosoma* were found among native-born people. And, 3 persons of Lycée Collège were found positive for the ova. They had, however, been brought-up in the endemic area in South Laos. It is, therefore, difficult to conclude that Pakse District has been contaminated with schistosomiasis. However, the fact that not a few immigrants into a "non-endemic area" from an endemic area have suffered from schistosomiasis, has a very important meaning in epidemiology of the disease. Namely, if there are vector snails capable of transmitting the disease in the area, it could become in due course endemic. It is, therefore, essential to determine whether the potential snail vector are living in the district or not.

On Khong Island, the survey was carried out two times as mentioned before.

In the 1966-1967 survey, the stool examination of 547 persons revealed 47 (8.59%) positive cases. In almost all the endemic areas in Japan and in the Philippines, the infection rate which was obtained by detection of *Schistosoma* eggs in the stool by means of concentration method is less than 10%. Therefore, it is positive that the incidence of infection among the inhabitants in this district is exceedingly high. Local difference in the incidence of the infection was conspicuous. A high rate of the infection was found in schoolchildren in Muong Khong, and, in general, the positive rate of inhabitants in area on the east of the Mekong River was significantly higher than that on the west side.

The result of the examination in 1968-1969 was the same as that of the 1966-1967 survey.

In Stung Treng District, positive reaction for skin test was recognized in low percentage, and only 2 persons were positive by the stool examination. The one in B. Khan Memay was a native, but in B. Bachon the case was an emigrant from Kratie who had just arrived a few days ago.

The present survey confirmed the facts of the previous reports that endemic foci of schistosomiasis are present in the district of Kratie. The infection rate was high among floating villagers in this district. The authors suspected that the infection was caused by their habit of moving from place to place, and that those saucers played a role of diffusion of the disease in this area.

There was no definite evidence that there is endemic focus of the disease in the Kompong Cham District.

In Bassac-Lower Mekong District, only one case of schistosomiasis, eleven years old schoolboy, was found. He emigrated to Ph. Chhoung Leap along Tonle Bassac from Battam Bang in the western part of the country (Fig. 1). It is uncertain whether he infected in Battam Bang or in Bassac. Therefore, a survey in Battam Bang seems to be necessary.

By this survey, the authors observed the existence of the endemic foci of schistosomiasis in the districts of Khong and Kratie. However, data obtained by the survey suggest that there may be several endemic foci in the area covering South Laos and estuary of the Mekong River. Consequently, in order to obtain precise information on the extent of prevalence of schistosomiasis in the Mekong Basin, intensive investigation should be carried out.

The present survey also indicates that the Mekong River is a major site of infection for both human and reservoir host. Attention should be paid to the following points: (1) a great number of cases were discovered among persons who do not work in the rice paddies; (2) many patients suffer from skin itch and dermatitis after entering the water of the Mekong River; (3) snails such as *Pachydrobia pellucida* were found in abundance only near the edge of the river; These snails are suspected to be one of the most likely intermediate hosts (Lo, *et al.*, 1971).

As mentioned in the previous report (Iijima, *et al.*, 1971), the eggs of *Schistosoma* obtained from stool of the inhabitants in the endemic area are of the shape of compressed ellipse (major axis:  $61.7 \pm 4.78 \mu$ , minor axis:  $51.2 \pm 4.30 \mu$ ).

The symptom of schistosomiasis in the surveyed area seemed to be somewhat mild in comparison with that in Japan and in the Philippines. Nevertheless, numerous patients with enlarged liver and spleen were observed in all the endemic areas.

The authors discussed on the difference between the schistosomes in the Mekong Basin and in Japan in the previous report (Iijima, *et al.*, 1971).

### Conclusion

Epidemiological investigations of schistosomiasis were carried out in the several districts in the Mekong Basin in the years of 1966-1967 and 1968-1969, and results obtained are as follows:

1). A new endemic foci of the disease was found in Khong District by the 1966-1967

survey. The incidence of the infection among the inhabitants in this district was exceedingly high.

2). It was confirmed that endemic foci of the disease exist in the district of Kratie. High rate of the infection was found in floating villagers in this district.

3). The fact that a few cases of schistosomiasis were found at outside of the endemic area indicates presence of other foci of the disease in the Basin. Further investigation, therefore, should be carried out.

#### Acknowledgements

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## Mekong 川流域における住血吸虫病の研究

### III. 地域住民に対する感染状況について

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前報で述べたごとく、Mekong 川流域にはかなり広域にわたり住血吸虫病が散発しているが、著者らは WHO の Short-Term Consultant として Vientiane と Phnom-Penh 南部のヴェトナム国境との間の数地区において、1966~1967年、1968~1969年の2回、住民の疫学調査を実施した。

1966~1967年の調査では、南部ラオス、Khong 地区において住民547名中47名(8.6%)の住血吸虫保卵者を検出、これにより Mekong 川流域の最初の流行地が確認された。

1968~1969年の調査では Khong 地区において保虫宿主(イヌ)があきらかにされたほか、カンボジアの

Kratie 地区内に10~33%の感染率を示す相当規模の有病地の存することが確認された。

尚、この回の調査で、ラオスの Pakse、カンボジアの Stung Treng、Phnom-Penh 南部にそれぞれ1~3名の住血吸虫保卵者が検出されたことから、Mekong 川流域には上記2地区のほかにも有病地の存在の可能性があることがうかがえる。殊に Pakse 以南、Mekong 川河口にわたる今後の調査が望まれる。同様、カンボジア西部の Battam Bang からの移住者のうちに1名保卵者が検出されたことから、同地区を含む Tonle Sap 周辺の調査も必要と思惟される。