URINE PRECIPITIN REACTION FOR SCHISTOSOMIASIS JAPONICA

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I. INTRODUCTION

In cases of helminthiasis caused by an alimentary canal parasite, such as *Ascaris* or hookworm, the fecal examination is sufficient for its diagnosis. In cases of helminthiasis caused by a tissue parasite, however, the fecal examination cannot be sufficient, as the eggs of the parasite are not always discharged regularly in feces. In acute schistosomiasis japonica it takes about one month to discover the ova, and in chronic cases we cannot always find the eggs in the feces. Accordingly, it is very difficult to evaluate the therapeutic effects of medicine for schistosomiasis. It is not seldom that some patients discharge the eggs in several months after the treatment. Many concentration methods have been studied for the *Schistosoma* eggs, and yet these methods are not perfect. Immunological examinations such as the intradermal test, the precipitin test, the flocculation test or the complement fixation test have been investigated with the ova, cercariae, and the adult antigens by many authors. Okabe et al. (1954) reported that the precipitin reaction with the adult antigens was positive for acute schistosomiasis japonica with dilution as high as 1:6400, but the reaction was lower in chronic cases, only with the dilution of 1:800. Matsuse (1956) in our department reported that the precipitin reaction became positive in rabbits after 4 weeks from the exposure to cercariae.

It is probable that metabolic products of the worms in the blood are discharged sooner or later through the kidney into the urine. The authors intended to catch the metabolic products in the urine by the precipitine reaction and reported the obtained results in the preliminary report (1958). The urine precipitin reaction was examined in experimental animals for the evaluation of the therapeutic effects and for the earlier diagnosis of schistosomiasis japonica.

II. MATERIALS AND METHODS

1. Materials

a. The urine of schistosomiasis patients

The urine discharged early in the morning from a schistosomiasis patient was condensed up to one third with a vacuum pump at 40°C and dialysed with cellophane in running water for 24 hours. In summer, when the urine was left by for several days before the concentration, sodium azide was added to it at a dilution of 0.1%. The condensed urine was diluted with saline solution in a series of twofold dilutions.

b. The urine of schistosome infected rabbits.

The urine was catheterized and condensed up to one third, dialysed for 24 hours, and used as an antigen.

c. Serum of rabbits immunized with the schistosome antigen.

Schistosoma japonicum removed from rabbits were washed with saline solution and dried in a dessicator. Dried worms were put in Coca's solution (0.5% NaCl, 0.5% NaHCO₃ and 0.4% phenol) at a dilution of 1:100, stored in an icebox at 4°C for 24 hours, and extracted. Five ml of the antigen was injected intravenously into a rabbit, seven times every two days. After the last injection the blood was removed when the serum was over a titer of 1:3200. The immune serum was stored in a deepfreeze at -20°C and the amount desired was thawed for use.

2. Methods

The serum was divided into small precipitin tubes and the urine antigens which had been obtained by diluting with saline solution in a series of twofold dilutions (See II-1-a) were overlaid.

Readings were made after 2 hours. (In summer the temperature of the place where the test was incubated was 26°C.) The degree of white ring formation at the interphase was the measure of reading the reaction.

III. RESULTS

1. Acute cases

The urine precipitin reaction was tried and examined on two acute cases. These patients had a chill and fever. Liver tumor, eosinophilia and an increase of red cell sedimentation rate were also observed on them. A fecal examination was not done.

The titer of one of them was 1:4 and the other's 1:2, while the serum precipitin test showed the 1:800 titer in each case.

TABLE 1
The urine precipitin reaction for acute schistosomiasis japonica patients

Name	Age	Sex	1:1	1:2	1:4	1:8	1:16	Cont.	Serum precipitin reaction
K. N.	12	m	+	+	+	—	—	—	1:800
K. K.	66	m	+	+	—	—	—	—	1:800

2. Sub-acute cases

Two sub-acute cases were examined. Both of them complained of a malaise and had a little fever. A fecal examination was not done.

The titer of the one was 1:2 and the other's 1:4.

TABLE 2
The urine precipitin reaction for sub-acute schistosomiasis japonica patients

Name	Age	Sex	1:1	1:2	1:4	1:8	1:16	Cont.	Serum precipitin reaction
M. I.	12	m	+	+	—	—	—	—	1:400
K. N.	8	m	+	+	+	—	—	—	1:800

3. Chronic cases

102 cases in whose feces schistosome eggs were found by mass-examination were examined. All cases turned out to be positive for the urine precipitin reaction.

TABLE 3
The urine precipitin reaction for chronic schistosomiasis japonica patients

No.	Name	Age	Sex	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	Serum precipitin reaction
1	M. O.	35	m	+	+	—	—	—	—	1 : 1600
2	T. H.	48	m	+	+	+	—	—	—	1 : 1600
3	M. O.	38	m	+	+	+	—	—	—	1 : 1600
4	S. S.	46	m	+	+	+	+	+	—	1 : 800
5	H. S.	19	m	+	+	+	+	—	—	1 : 1600
6	M. A.	33	m	+	+	—	—	—	—	1 : 3200
7	T. A.	39	m	+	+	+	—	—	—	1 : 1600
8	N. M.	53	m	+	+	+	—	—	—	1 : 800
9	N. M.	31	m	+	+	+	—	—	—	1 : 1600
10	T. M.	22	m	+	+	—	—	—	—	1 : 1600
11	K. M.	46	m	+	+	+	—	—	—	1 : 800
12	S. M.	51	m	+	+	+	—	—	—	1 : 1600
13	O. M.	31	f	+	+	+	+	—	—	1 : 800
14	H. A.	16	f	+	+	+	—	—	—	1 : 1600
15	Y. A.	29	f	+	+	+	—	—	—	1 : 800
16	T. T.	31	f	+	+	+	—	—	—	1 : 1600
17	M. A.	34	f	+	+	+	—	—	—	1 : 1600
18	Y. A.	33	f	+	+	+	+	—	—	1 : 1600
19	H. M.	32	f	+	+	+	+	—	—	1 : 3200
20	N. S.	43	f	+	+	+	—	—	—	1 : 800
21	T. M.	20	f	+	+	—	—	—	—	1 : 1600
22	R. I.	54	m	+	+	+	—	—	—	1 : 800
23	T. N.	45	m	+	+	+	—	—	—	1 : 400
24	T. O.	39	m	+	+	+	—	—	—	1 : 800
25	K. O.	23	m	+	+	+	—	—	—	1 : 1600
26	Y. T.	20	m	+	+	+	—	—	—	1 : 800
27	S. E.	32	f	+	+	+	+	—	—	1 : 800
28	Y. T.	33	m	+	+	+	—	—	—	1 : 1600
29	M. M.	31	m	+	+	+	—	—	—	1 : 800
30	H. N.	27	m	+	+	—	—	—	—	1 : 1600
31	E. M.	30	m	+	+	+	—	—	—	1 : 800
32	I. M.	35	m	+	+	+	—	—	—	1 : 800
33	T. M.	38	f	+	+	+	—	—	—	1 : 800
34	H. N.	53	f	+	+	+	+	—	—	1 : 1600
35	K. I.	48	f	+	+	+	+	+	—	1 : 1600
36	M. M.	37	f	+	+	—	—	—	—	1 : 3200
37	S. N.	42	f	+	+	+	—	—	—	1 : 1600
38	M. E.	31	f	+	+	+	—	—	—	1 : 1600
39	M. S.	33	f	+	+	+	—	—	—	1 : 1600
40	M. N.	52	f	+	+	+	+	—	—	1 : 1600
41	K. Y.	35	f	+	+	+	+	—	—	1 : 1600
42	S. O.	24	f	+	+	+	—	—	—	1 : 800
43	Y. O.	43	f	+	+	+	+	—	—	1 : 800
44	T. Y.	35	f	+	+	—	—	—	—	1 : 800
45	M. Y.	36	m	+	+	+	—	—	—	1 : 1600

No.	Name	Age	Sex	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	Serum precipitin reaction
46	Y. S.	60	m	+	+	+	—	—	—	1 : 1600
47	I. Y.	57	m	+	+	+	—	—	—	1 : 800
48	Y. W.	41	m	+	+	—	—	—	—	1 : 3200
49	K. H.	42	m	+	+	—	—	—	—	1 : 3200
50	H. S.	48	m	+	+	+	—	—	—	1 : 1600
51	S. Y.	44	m	+	+	+	+	—	—	1 : 3200
52	S. I.	61	m	+	+	+	—	—	—	1 : 800
53	I. K.	36	m	+	+	+	—	—	—	1 : 3200
54	K. N.	17	m	+	+	+	+	—	—	
55	T. K.	20	m	+	+	—	—	—	—	1 : 3200
56	K. H.	37	f	+	+	+	—	—	—	1 : 3200
57	H. T.	43	m	+	+	+	—	—	—	1 : 800
58	Y. T.	39	m	+	+	+	—	—	—	1 : 1600
59	K. K.	30	m	+	+	+	+	+	—	1 : 800
60	H. H.	55	m	+	+	—	—	—	—	1 : 800
61	H. H.	27	m	+	+	+	—	—	—	1 : 800
62	Y. M.	28	m	+	+	+	—	—	—	1 : 800
63	H. U.	48	m	+	+	+	+	—	—	1 : 400
64	T. O.	12	m	+	+	+	—	—	—	1 : 1600
65	K. H.	11	m	+	+	+	+	—	—	1 : 800
66	H. K.	12	m	+	+	+	—	—	—	1 : 1600
67	T. K.	12	m	+	+	+	—	—	—	1 : 1600
68	H. M.	11	m	+	+	—	—	—	—	1 : 800
69	H. Y.	10	f	+	+	+	—	—	—	1 : 800
70	E. H.	12	f	+	+	—	—	—	—	1 : 800
71	I. I.	10	f	+	+	+	—	—	—	1 : 1600
72	H. I.	12	f	+	+	+	+	—	—	1 : 1600
73	Y. O.	10	f	+	+	+	—	—	—	1 : 1600
74	J. O.	12	f	+	+	—	—	—	—	1 : 800
75	S. Y.	12	f	+	—	—	—	—	—	1 : 800
76	Y. W.	12	m	+	+	+	—	—	—	1 : 800
77	T. K.	11	m	+	+	—	—	—	—	1 : 1600
78	Y. H.	12	f	+	+	—	—	—	—	1 : 1600
79	K. H.	12	f	+	+	+	—	—	—	1 : 800
80	N. O.	9	m	+	+	+	—	—	—	1 : 1600
81	J. O.	12	m	+	+	+	—	—	—	1 : 3200
82	T. K.	12	m	+	+	+	+	—	—	1 : 1600
83	S. T.	11	m	+	—	—	—	—	—	1 : 200
84	T. S.	10	f	+	+	+	—	—	—	1 : 800
85	S. Y.	11	m	+	+	—	—	—	—	1 : 1600
86	T. H.	11	m	+	+	+	—	—	—	1 : 800
87	Y. K.	9	m	+	+	—	—	—	—	1 : 1600
88	M. N.	7	m	+	+	+	—	—	—	
89	T. S.	8	m	+	—	—	—	—	—	1 : 800
90	H. Y.	8	m	+	+	+	—	—	—	
91	Y. N.	9	m	+	+	+	—	—	—	
92	M. O.	9	m	+	+	+	—	—	—	
93	S. Y.	9	m	+	+	+	+	—	—	
94	Y. K.	9	m	+	+	+	—	—	—	
95	H. S.	10	m	+	+	—	—	—	—	1 : 800

96	Y. Y.	10	m	+	+	+	+	-	-	1 : 800
97	K. Y.	10	m	+	+	-	-	-	-	
98	Y. Y.	11	m	+	+	-	-	-	-	
99	S. K.	11	m	+	+	-	-	-	-	
100	R. Y.	11	f	+	+	+	-	-	-	
101	E. S.	11	f	+	+	-	-	-	-	1 : 800
102	K. Y.	11	m	+	+	+	+	-	-	

TABLE 4

The titer of urine and the distribution of the cases

Titer of Urine	Cases	%
1 : 1	3	2.9
1 : 2	23	22.6
1 : 4	55	53.9
1 : 8	18	17.6
1 : 16	3	2.9
Total	102	

More than the half of the examined showed a titer of 1 : 4.

4. Normal persons

The urine precipitin reactions of ten normal persons were examined with *Schistosoma* immune serum, but all were negative.

5. Group reactions

a. The urine of *Schistosoma mansoni* infected rabbits.

Two rabbits were infected with 50 cercariae of *Schistosoma mansoni* got from *Australorbis glabratus*. The difference of the urine precipitin reaction was examined before and after the infection. Before the infection the reaction of the two rabbits was negative, and in 59 and 63 days after the infection it turned out to be positive respectively, but their titers were very low.

TABLE 5

The urine precipitin reaction between the urine obtained from Schistosoma mansoni infected rabbits and Schistosoma japonicum immunized rabbit serum

No.	Days after infection	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
1	Before infection	-	-	-	-	-	-
	7 days	-	-	-	-	-	-
	21 days	-	-	-	-	-	-
	35 days	-	-	-	-	-	-
	49 days	-	-	-	-	-	-
	63 days	+	-	-	-	-	-
	77 days	±	-	-	-	-	-
2	Before infection	-	-	-	-	-	-
	3 days	-	-	-	-	-	-
	17 days	-	-	-	-	-	-
	31 days	-	-	-	-	-	-
	45 days	-	-	-	-	-	-
	59 days	+	±	-	-	-	-
	73 days	+	-	-	-	-	-

b. Paragonimiasis patients

Four cases out of 11 paragonimiasis patients were positive at a titer of 1:1. Group reaction was observed, but their titer was low.

c. Clonorchiasis patients

The urine precipitine reactions of five cases of clonorchiasis were examined, but all were negative.

IV. NATURE OF THE URINE ANTIGEN

1. The influence of temperature at the urine concentration

The urine titer was examined at various temperatures during the concentration. The reaction did not appear when the temperature was 80°C and higher.

TABLE 6

The influence of temperature on the antigen at the urine concentration

Temperature	Minutes	No. 1					No. 2					No. 3				
		1:1	1:2	1:4	1:8	Cont.	1:1	1:2	1:4	1:8	Cont.	1:1	1:2	1:4	1:8	Cont.
40°C	30min.	+	+	+	-	-	+	+	+	-	-	+	+	+	-	-
50°C	25min.	+	+	+	-	-	+	+	+	-	-	+	+	+	-	-
60°C	15min.	+	+	+	-	-	+	+	-	-	-	+	+	+	-	-
70°C	10min.	+	+	-	-	-	+	+	-	-	-	+	-	-	-	-
80°C	7min.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
90°C	3min.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
100°C	3min.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

2. Preservation of the urine antigen

The urine was putrescible in summer. Sometimes the next day the urine was found decomposed. Even if it was stored in a refrigerator, it could be used for only two days.

When the sodium azide was added to it at a dilution of 0.1%, at the room temperature 12-19°C, the titer of urine had not changed for six days after the urination, but on the 9th day the titer of urine dropped and on the 12th day one of three samples was negative.

TABLE 7

Preservation of urine at 12°-19°C

a) The urine containing 0.1% NaN₃.

Days after urination	No. 1					No. 2					No. 3				
	1:1	1:2	1:4	1:8	Cont.	1:1	1:2	1:4	1:8	Cont.	1:1	1:2	1:4	1:8	Cont.
0	+	+	+	-	-	+	+	+	-	-	+	+	+	-	-
3	+	+	+	-	-	+	+	+	-	-	+	+	+	-	-
6	+	+	-	-	-	+	+	+	-	-	+	+	+	-	-
9	+	+	-	-	-	+	+	-	-	-	+	+	-	-	-
12	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-

b) The urine without NaN_3 .

Days after urination	No. 1						No. 2						No. 3					
	1 : 1	1 : 2	1 : 4	1 : 8	Cont.		1 : 1	1 : 2	1 : 4	1 : 8	Cont.		1 : 1	1 : 2	1 : 4	1 : 8	Cont.	
0	+	+	+	-	-		+	+	+	-	-		+	+	+	-	-	
3	opacitas						+	+	+	-	-		+	+	+	-	-	
6	opacitas						opacitas						opacitas					

3. The cultured medium

Four pairs of *Schistosoma japonicum* were removed from a rabbit and were cultured, two with saline solution and the other two with Ringer's solution in Carrel flasks. The cultured medium was used as an antigen for the precipitin test. The titer of the medium was 1:1 after 5 hours cultivation and 1:2 after 10 hours cultivation in 0.85% NaCl solution. It was 1:2 after 12 hours cultivation and the same after 24 hours in Ringer's solution containing 0.5% glucose.

TABLE 8

The precipitin reaction between the cultured medium of *Schistosoma japonicum* and *Schistosoma japonicum* immunized rabbit serum

a) 0.85% NaCl solution							b) Ringer's solution containing 0.5% glucose						
Hours	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	Hours	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
5 hours	+	-	-	-	-	-	12 hours	+	+	-	-	-	-
10 hours	+	+	-	-	-	-	24 hours	+	+	-	-	-	-

V. ASSESSMENT OF CURE

1. TWSb treatment

Five patients of schistosomiasis japonica were injected intramuscularly with antimony-a, a'-potassium succinate (TWSb).

Three male patients were injected with 0.3-0.4g TWSb totaling 2.05-2.28g, and two female patients were injected 0.3-0.4g TWSb totaling 1.5-1.7g during a week. The therapeutic effects had been examined observing the eggs in feces, the titer of urine and serum precipitin reactions of the patients during the course of about one year after the treatment.

The fecal examination, after three days of the treatment, revealed that three of them were negative, but two patients were still positive. After 77 days no eggs were found in the feces of any of them and by the examination after about one year they were still negative.

The urine precipitin reaction was positive in all cases for three days after the treatment. After 43 days the reaction of one of them turned negative, and that of the four were positive at a titer of 1:1. After 78 days the reaction of all proved negative and remained negative after one year. It could be considered to be certain that no recidivation and no reinfection occurred during the year. (It must be added here the fact that there was not found any *Oncomelania*

	Feb.	8	Before treatment	+	+	+	+	+	-	-	-	1 : 3200
	Feb.	18	4 days	-	+	+	±	-	-	-	-	"
	March	30	44 dayt	-	+	-	-	-	-	-	-	"
T. I.	May	3	78 days	-	-	-	-	-	-	-	-	"
	June	10	115 days		-	-	-	-	-	-	-	"
	Aug.	4	170 days		-	-	-	-	-	-	-	1 : 6400
	Sept.	16	213 days	-	-	-	-	-	-	-	-	1 : 3200
	Oct.	7	237 davs	-	-	-	-	-	-	-	-	"
	Jan.	28, 1959	348 days	-	-	-	-	-	-	-	-	"

TABLE 10

Course of the serum precipitin reactions and fecal examinations after treatment

[illegible]

2. *Stibnal* (*Sodium antimony tartrate*)

Twenty chronic patients were treated with *Stibnal* (0.3% sodium antimony tartrate) and followed up with the urine precipitin reaction. 20ml *Stibnal* was injected to each patient 13–50 times respectively. In Japan *Stibnal* is injected 20 times usually. By our experiment, about the cases with 27–50 times injection the reaction was negative, but about the cases injected 26–13 times the reaction was positive with one exception. These results imply that 20 times injection of *Stibnal* was not sufficient to be curative.

TABLE 11
The urine precipitin reaction of treated cases with Stibnal

Name	Age	Sex	Times of injection	Months after treatment	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	1 : 32	Cont.
T. O.	44	m	50	2	—	—	—	—	—	—	—
T. I.	42	m	42	3	—	—	—	—	—	—	—
M. N.	26	m	40	13	—	—	—	—	—	—	—
M. M.	33	f	36	28	—	—	—	—	—	—	—
Y. S.	34	m	33	8	—	—	—	—	—	—	—
S. T.	44	f	30	5	—	—	—	—	—	—	—
T. O.	24	m	30	3	—	—	—	—	—	—	—
S. T.	34	m	30	8	—	—	—	—	—	—	—
Y. N.	28	m	30	13	—	—	—	—	—	—	—
A. Y.	50	f	27	58	—	—	—	—	—	—	—
S. A.	38	f	26	10	+	+	—	—	—	—	—
T. O.	31	m	25	10	—	—	—	—	—	—	—
H. M.	14	m	25	3	+	—	—	—	—	—	—
S. T.	38	f	25	8	+	+	—	—	—	—	—
Y. M.	14	m	20	9	+	+	+	—	—	—	—
Y. T.	34	m	20	57	—	—	—	—	—	—	—
K. K.	67	m	20	2	+	+	—	—	—	—	—
K. K.	48	m	20	8	+	+	—	—	—	—	—
K. T.	13	m	15	9	+	+	+	—	—	—	—
H. H.	29	f	13	3	+	+	+	—	—	—	—

3. *Animal experiments*

a. TWSb treatment

i. rabbits

TWSb was injected intramuscularly, 8mg/kg a day, to *Schistosoma* infected rabbits for five days and its effects observed for 61 days. The urine

TABLE 12
The urine precipitin reaction of treated rabbits with TWSb (40mg/kg)

Days after treatment	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
Before treat.		+	+	—	—	—	—		+	+	+	—	—	—
16 days		+	+	—	—	—	—		+	+	+	—	—	—
28 days	9	+	+	—	—	—	—	10	+	+	—	—	—	—
46 days		+	+	+	—	—	—		+	+	—	—	—	—
61 days		+	+	—	—	—	—		+	+	—	—	—	—

precipitin reaction did not turn out to be negative and some worms were found by autopsy.

ii. dogs

TWSb was injected intramuscularly, 8mg/kg to the dogs infected with 150 cercariae of *Schistosoma japonicum*. After 105 days from the treatment the reaction changed to negative while the control dogs showed positive reactions. By autopsy there no *Schistosoma* was found in the treated dogs, while in the control dogs 29 and 55 worms were found respectively.

TABLE 13

Follow-up with fecal examination and urine precipitin reaction after the treatment of an infected dog with TWSb (40mg/kg in 5 days)

Days after infection	Days after treatment	Treated dog					eggs	Control dog No. 1					eggs	Control dog No. 2					eggs
		1 : 1	1 : 2	1 : 4	1 : 8			1 : 1	1 : 2	1 : 4	1 : 8			1 : 1	1 : 2	1 : 4	1 : 8		
41 days	Before	+	+	+	-		+	+	+	+	-		+	+	+	+	-		+
49	7 days	+	+	+	-		+	+	+	+	-		+	+	+	+	-		+
56	14	+	+	-	-		-	+	+	-	-		+	+	+	+	+		+
63	21	+	+	-	-		+	+	+	+	-		+	+	+	-	-		+
70	28	+	+	-	-		-	+	+	+	-		+	+	+	-	-		+
77	35	+	+	-	-		-	+	+	+	+		+	+	+	-	-		+
91	49	+	-	-	-		-	+	+	+	-		+	+	+	+	-		+
105	63	+	-	-	-		-	+	+	-	-		+	+	+	-	-		+
119	77	±	-	-	-		-	+	+	+	-		+	+	+	+	-		+
133	91	±	-	-	-		-	+	+	+	-		+	+	+	+	+		+
147	105	-	-	-	-		-	+	+	+	-		+	+	+	-	-		+
154	112	Autopsy (-)						male 14 female 15						male 28 female 27					

*Deformed egg

b. Stibnal treatment

Stibnal was injected intravenously, 0.5ml/kg, 0.8ml/kg, and 1.0ml/kg a day, to *Schistosoma* infected rabbits for twenty days and every other week after the treatment the urine precipitin reaction was tried on them and examined.

i. Stibnal 1.0ml/kg for 20 days

No. 1 became negative on the 4th week and No. 2 on the 5th week. The next week the rabbits were dissected and no worms were found.

ii. Stibnal 0.8ml/kg for 20 days

No. 3 did not change to be negative and was found to be still infected with worms (♂ 19 and ♀ 19). No. 4 became negative on the 4th week and on the 5th week, when it was dessected, two degenerated male worms were found in its liver by perfusion.

iii. Stibnal 0.5ml/kg for 20 days

Both No. 5 and No. 6 did not become negative and were found to be still infected with worms.

TABLE 14

The urine precipitin reaction of the treated rabbits with Stibnal

a) Stibnal 1.0ml/kg for 20 days

Days after treatment	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
Before treat.		+	+	+	-	-	-		+	+	+	+	-	-
1 week		+	+	+	-	-	-		+	+	+	-	-	-
2 weeks	1	+	+	-	-	-	-	2	+	+	+	-	-	-
3 weeks		+	-	-	-	-	-		+	+	-	-	-	-
4 weeks		-	-	-	-	-	-		+	-	-	-	-	-
5 weeks		-	-	-	-	-	-		-	-	-	-	-	-

b) Stibnal 0.8ml/kg for 20 days

Days after treatment	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
Before treat.		+	+	+	-	-	-		+	+	-	-	-	-
1 week		+	+	+	+	-	-		+	+	+	-	-	-
2 weeks	3	+	+	-	-	-	-	4	+	+	-	-	-	-
3 weeks		+	+	-	-	-	-		+	+	-	-	-	-
4 weeks		+	+	-	-	-	-		-	-	-	-	-	-
5 weeks		+	+	-	-	-	-		-	-	-	-	-	-

c) Stibnal 0.5ml/kg for 20 days

Days after treatment	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
Before treat.		+	+	+	-	-	-		+	+	+	+	-	-
1 week		+	+	-	-	-	-		+	+	-	-	-	-
2 weeks	5	+	+	+	-	-	-	6	+	+	-	-	-	-
3 weeks		+	+	-	-	-	-		+	+	-	-	-	-
4 weeks		+	+	-	-	-	-		+	+	-	-	-	-
5 weeks		+	+	-	-	-	-		+	+	+	-	-	-

VI. EARLY DIAGNOSIS

It was showed in the previous part (III-1) that the acute cases of schistosomiasis japonica reacted to the urine precipitin reaction. Animal experiments

TABLE 15

The urine precipitin reaction of infected rabbits

No.	Days after infection	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.	No.	1 : 1	1 : 2	1 : 4	1 : 8	1 : 16	Cont.
	Before inf.	-	-	-	-	-	-		-	-	-	-	-	-
	3 days	±	-	-	-	-	-		+	-	-	-	-	-
	6 days	+	+	-	-	-	-		+	+	-	-	-	-
1	9 days	+	+	-	-	-	-	4	+	+	-	-	-	-
	12 days	+	+	-	-	-	-		+	+	-	-	-	-
	15 days	+	+	+	-	-	-		+	+	+	-	-	-
	18 days	+	+	+	-	-	-		+	+	-	-	-	-
	21 days	+	+	-	-	-	-		+	+	+	-	-	-

2	Before inf.	—	—	—	—	—	—	5	—	—	—	—	—	—
	3 days	+	—	—	—	—	—		—	—	—	—	—	—
	6 days	+	+	—	—	—	—		+	—	—	—	—	—
	9 days	+	+	+	—	—	—		+	+	+	—	—	—
	12 days	+	+	—	—	—	—		+	+	—	—	—	—
	15 days	+	+	+	—	—	—		+	+	+	+	—	—
	18 days	+	+	+	—	—	—		+	+	—	—	—	—
	21 days	+	+	+	—	—	—		+	+	+	+	—	—
3	Before inf.	—	—	—	—	—	—	6	—	—	—	—	—	—
	3 days	±	—	—	—	—	—		±	—	—	—	—	—
	6 days	+	—	—	—	—	—		+	—	—	—	—	—
	9 days	+	+	+	—	—	—		+	+	—	—	—	—
	12 days	+	+	+	—	—	—		+	+	+	—	—	—
	15 days	+	+	—	—	—	—		+	+	+	—	—	—
	18 days	+	+	+	+	—	—		+	+	+	—	—	—
	21 days	+	+	+	—	—	—		+	+	—	—	—	—

will lead us to presume when the reaction turns to be positive after the infection.

Two out of six rabbits, which had been infected with about one hundred cercariae, showed a titer of 1:2 and the others a titer of 1:1 on the 6th day. On the 9th day three of them became positive at a titer of 1:4 and others 1:2.

VII. SUMMARY

The acute, sub-acute and chronic patients of schistosomiasis japonica reacted to the urine precipitin reaction. The antigen in urine is easy to change by heat and putrefaction. It is not clear what substance participates in this phenomenon, but it is presumable that the substance may be thermolabile and nondialyzable.

The urine precipitin reaction may be used for the assessment of cure. Animal experiments showed the fact that after the reaction had become negative, the worm was not found any more.

This reaction may be used for early diagnosis for schistosomiasis. One of the acute cases showed a positive reaction to this reaction test within one month after the infection. In the animal experiment the reaction changed to be positive as early as on the first week after the infection.

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