[Jpn. J. Parasitol., Vol. 37, No. 2, 104–107, April, 1988]

Lack of Cross Immunity between a Tissue Phase of *Hymenolepis nana* and a Lumen Phase of *H. diminuta* in Rats

AKIRA ITO, SHIRO KANO, ATSUSHI HIOKI, SHIRO KASUYA AND HIROSHI OHTOMO

(Received for publication; December 23, 1987)

Abstract

Prior infection with a single or 10 cysticercoids of *Hymenolepis diminuta* did not evoke any cross immunity to a challenge with oncospheres of *H. nana* in Rowett strain rats. There was no difference in the number of cysticercoids of *H. nana* in the intestinal tissue recovered in rats harboring a single adult of *H. diminuta* and those without prior infection, and also in both congenitally hypothymic nude and phenotypically normal rats harboring 10 adults of *H. diminuta*. Prior infection with either a single dose or repeated doses of *H. nana* oncospheres did not affect the establishment or fecundity of adult *H. diminuta*. A tissue phase of *H. nana* at prior infection did not affect a lumen phase of *H. diminuta*. Reciprocally, a lumen phase of *H. diminuta* did not affect a tissue phase of *H. nana*, either. Therefore, there is no cross immunity assessed by decrease in the number and/or fecundity of the two species of hymenolepidid cestodes, between a tissue phase of *H. nana* and a lumen phase of *H. diminuta* in rats.

Key words: *Hymenolepis nana*, *H. diminuta*, cross immunity, lumen phase, rats, nude rats

Introduction

It has become valid that (1) immunogenicity of the mouse tapeworm *Hymenolepis nana* differs between the tissue and lumen phases in the permissive mouse host and (2) the lumen phase of the rat tapeworm *H. diminuta*, which has no tissue phase, is highly immunogenic not only in the nonpermissive mouse host but also the permissive rat host (reviewed by Hopkins, 1980, Andreassen, 1981, Williams, 1982, Rickard, 1983, Ito and Smyth, 1987).

Most recently, Ito and Onitake (1987) found that BALB/c mice initially infected with *H. diminuta* become highly resistant to a lumen phase of *H. nana* resulting in complete failure in developing adult *H. nana*, and those mice initially infected with *H. nana* and experienced a lumen phase of this parasite become highly resistant to a lumen phase of *H. diminuta*.

The rat, in general, is an unnatural definitive host of *H. nana*, since there is no lumenal adult development of *H. nana* in rats given either oncospheres or cysticercoids (Ito and Smyth, 1987). However, when oncospheres of *H. nana* were given into naive rats, some of them develop into cysticercoids in the intestinal tissues (Shorb, 1933. Heyneman, 1962, Weinmann, 1964, 1966, Ito, 1982, 1983, Ito and Kamiyama, 1984, 1987). Therefore, the rat is susceptible to a tissue phase, but simultaneously highly resistant to a lumen phase of this parasite (Ito and Smyth, 1987). In contrast, the rat is the natural definitive host of *H. diminuta*. Cysticercoids of *H. diminuta* inoculated to rats develop into mature tapeworms and survive for a long period in the lumen without any tissue phase (Hopkins, 1980, Ito and Smyth, 1987). Thus, oncospheres of *H. nana* infect rats and develop into tissue cysticercoids but thereafter no lumenal development occurs, whereas cysticercoids of *H. diminuta* infect rats and develop

Department of Parasitology, Gifu University School of Medicine, Gifu 500, Japan

This work was supported in part by the grant provided to Ito by The Ichiro Kanehara Foundation 1987.

伊藤　亮　加納至朗　日置敦巳　粕谷志郎
大友弘士（岐阜大学医学部寄生虫学教室）

into mature tapeworms in the lumen without any tissue phase.

In the present work, we have examined whether immunity evoked by the lumen phase of *H. diminuta* affects the tissue phase of *H. nana* challenge infection in rats, and whether immunity evoked by prior tissue phase of *H. nana* affects the lumen phase of *H. diminuta* in rats.

Materials and Methods

Parasites

Gravid proglottids and oncospheres of *Hymenolepis nana* and *H. diminuta* were prepared from donor mice and rats experimentally inoculated with cysticercoids of these cestodes, respectively. Cysticercoids of these two species of hymenolepidid cestodes were prepared from flour beetles, *Tribolium confusum* and *Tenebrio molitor* experimentally ingested with gravid proglottids of these cestodes, respectively (Ito and Onitake, 1987).

Rats

Rowett strain rats (euthymic rnu/+ and hypothymic nude rnu/rnu), 5-7 weeks old males and females were used (Ito and Kamiyama, 1984, 1987). Throughout the text the word rat means the euthymic rat except that described nude rat.

Experimental schedules and Assay of the infections

The experimental schedules are summarized in Tables 1 and 2. After the small intestine of rat was cut open lengthwise in 0.85% NaCl in 25 cm diameter Petri dish, all the worms were removed from the intestine. The number of worms was counted and the worms were rinsed several times with 0.85% NaCl. The fresh worm biomass (the wet worm body weight) was measured after 0.85% NaCl on the worms was blotted out on filter paper. The inner surface of the small intestine was examined for detecting tissue cysticercoids of *H. nana* (Hunninen, 1935).

Results

Experiment 1. Influence of an initial lumen phase of *Hymenolepis diminuta* to a tissue phase of *H. nana* was examined in rats including nude rats initially given one or 10 cysticercoids of *H. diminuta* and challenged with 5,000 oncospheres of *H. nana*. The experimental schedules and the results are summarized in Table 1. All the rats either harboring a single adult tapeworm of *H. diminuta* or none showed similar susceptibility to the challenge with oncospheres

Table 1 Effect of a primary ongoing infection with *Hymenolepis diminuta* against challenge with oncospheres of *H. nana* in congenitally hypothymic and phenotypically normal rats

Initial inoculation with *H. diminuta* (day 0)	Challenge with oncospheres of *H. nana*		*H. diminuta*			*H. nana*	
	dose	day	No. of rats infected	No. of worms Mean±SD	Biomass (mg) Mean±SD	No. of rats infected	No. of tissue cysticercoids Mean±SD
1 cyst†	5×10^3	10	10/10	1.0±0.0	1132±105.2	10/10	56±23.5
1 cyst	0	10	10/10	1.0±0.0	1142±132.6	0/10	0
0 cyst	5×10^3	10	0/10	0	0	10/10	60±30.8
10 cysts	5×10^3	15	5/5	9.0±0.7	2714±210.5	5/5	54±23.6
10 cysts	5×10^3	15	5/5*	9.8±0.5	2618±423.0	5/5	63±23.9
1 cyst	5×10^3	30	10/10	1.0±0.0	1250±157.5	10/10	47±15.8
0 cyst	5×10^3	30	0/10	0	0	10/10	40±18.2

All the rats other than * (given 10 cysts of *H. diminuta*) were normal rats.

* Nude rats.

† cysticercoid.

Table 2 Effect of the tissue phase of *Hymenolepis nana* against the lumen phase of *H. diminuta* in rats

H. nana oncosphere dose (day 0)	Day of *H. diminuta* challenge	16 day old *H. diminuta*	
		No. of rats infected	Worm biomass (mg) Mean±SD
5×10^4	0	5/5	1040± 84.5
0	0	5/5	1005± 65.8
5×10^4	4	14/14	1070±103.8
0	4	14/14	1066± 88.1
5×10^4	15	12/12	1063±147.4
0	15	12/12	1136± 54.1
5×10^4	30	10/10	1083± 95.7
0	30	10/10	1040±110.4
$(5\times10^4)\times5$*	4†	5/5	1102± 95.3
0	4†	5/5	1144±115.8

All the rats were killed 16 days after challenge with a single cysticercoid of *H. diminuta*.

* Inoculated five times every two days.

† 4 days after the last inoculation with oncospheres.

of *H. nana*. Furthermore, there was no difference in the susceptibility to the *H. nana* challenge between immunologically incompetent nude and immunocompetent normal rats (Ito and Kamiyama, 1984) given initially 10 cysticercoids of *H. diminuta*. The tissue cysticercoid recovery rates were approximately 1% of the administered eggs in all the rats either sensitized previously with a single *H. diminuta* or not, and in nude and normal rats sensitized with 10 *H. diminuta*.

Experiment 2. Influence of a tissue phase of *H. nana* to the development of a lumen phase of *H. diminuta* was examined in rats initially given 5×10^4 oncospheres of *H. nana*. The experimental schedules and the results are summarized in Table 2. An initial tissue phase of *H. nana*, which was sufficient to protect rats from the homologous challenge (Ito and Kamiyama, 1984), did not show any influence to the lumen phase of *H. diminuta*. All the rats initially given oncospheres of *H. nana* harbored *H. diminuta*. There was no difference in the worm biomass of *H. diminuta* recovered in rats infected previously with *H. nana* and those without previous infection. Rats given oncospheres five times every two days showed no resistance to the development of *H. diminuta* either.

Discussion

Our present results strongly suggest that there is no cross immunity between *H. nana* and *H. diminuta* in Rowett rats. This contrasts with the recent findings that cross immunity to the lumen phase occurs between the two species of hymenolepidid cestodes in BALB/c mice (Ito and Onitake, 1987). The critical difference in the expression of the cross immunity to the lumen phase of the two species between rats used in the present work and mice in the previous work (Ito and Onitake, 1987) appears to be caused by the different mode of infection with *H. nana* in these host animals. In mice given oncospheres of *H. nana*, both tissue and lumen phases are established, whereas in rats only a tissue phase is established. In the previous study in the mouse model, it was strongly suggested that the cross immunity was lumen phase specific between the two species, since no *H. nana* adults (but not the tissue cysticercoids) were recovered in BALB/c mice previously

infected with *H. diminuta*, and reciprocally, no *H. diminuta* adults in those previously experienced a lumen phase of *H. nana* (Ito and Onitake, 1987).

Oncospheres of *H. nana* succeed in developing into tissue cysticercoids in the intestinal wall but fail to develop into lumenal adults in rats. Cysticercoids in the tissue escape to the lumen but are expelled within a few days without any development from excysted juvenile form (Ito, 1982). Thus, it is suggested that these excysted juveniles of *H. nana* can not evoke the immunuity to the lumen phase of *H. diminuta* in rats.

In rats harboring *H. diminuta*, it has been proved that the rats possess the immunity to the homologous challenge (Hopkins, 1980, Andreassen and Hopkins, 1982, Ito and Smyth, 1987) and there is no doubt that some inflammations are evoked by the presence of adult *H. diminuta*. Nevertheless, the fact that oncospheres of *H. nana* could invade the intestinal villi and develop into tissue cysticercoids strongly suggests that immunity and inflammation evoked by the lumen phase of *H. diminuta* have no or little effect against the oncospheral invasion into the intestinal villi and the tissue phase of *H. nana*. It seems to be rational that the lack of the cross immunity between *H. nana* and *H. diminuta* in rats is due to the lack of a lumen phase of *H. nana* in rats. The present results, therefore, are not contradictory to our previous work that the cross immunity evoked by *H. nana* and *H. diminuta* in BALB/c mice is lumen phase specific (Ito and Onitake, 1987).

References

1) Andreassen, J. (1981): Immunity to adult cestodes. Parasitology 82, 153–159.
2) Andreassen, J. and Hopkins, C. A. (1982): Immunologically mediated rejection of *Hymenolepis diminuta* by its normal host, the rat. J. Parasitol., 66, 898–903.
3) Heyneman, D. (1962): Studies on helminth immunity. II. Influence of *Hymenolepis nana* (Cestoda: Hymenolepididae) in dual infections with *H. diminuta* in white mice and rats. Exp. Parasitol., 12, 7–18.
4) Hopkins, C. A. (1980): Immunity and *Hymenolepis diminuta*. In Biology of the Tapeworm *Hymenolepis diminuta* (Arai, H. S. ed.), pp. 551–614, Academic Press, New York.
5) Hunninen, A. V. (1935): A method of demonstrating cysticercoids of *Hymenolepis fraterna* (*H. nana* var. *fraterna* Stiles) in the intestinal villi of mice. J. Parasitol., 21, 124–125.
6) Ito, A. (1982): Induction of adult formation from cysticercoid of *Hymenolepis nana* established in Fischer (F344) rat with cortisone acetate. Jpn. J. Parasitol., 31, 141–146.
7) Ito, A. (1983): *Hymenolepis nana*: Maturation in an immunosuppressed unnatural rat host. Exp. Parasitol., 56, 318–326.
8) Ito, A. and Kamiyama, T. (1984): *Hymenolepis nana*: Worm recovery from congenitally athymic nude and phenotypically normal rats and mice. Exp. Parasitol., 58, 132–137.
9) Ito, A. and Kamiyama, T. (1987): Cortisone-sensitive, innate resistance to *Hymenolepis nana* infection in congenitally athymic nude rats. J. Helminthol., 61, 124–128.
10) Ito, A. and Onitake, K. (1987): *Hymenolepis diminuta* and *H. nana*: Cross immunity against the lumen phase in BALB/c mice. Exp. Parasitol., 64, 48–56.
11) Ito, A. and Smyth, J. D. (1987): Adult cestodes. Immunology of the lumen-dwelling cestode infections. In Immune Responses in Parasitic Infections: Immunology, Immunopathology, and Immunoprophylaxis (Soulsby, E. J. L. ed.), vol. 2, pp. 115–163, CRC Press, Boca Raton, Fl.
12) Rickard, M. D. (1983): Immunity. In Biology of the Eucestoda (Arme, C. and Pappas, P. W. eds.), vol. 2, pp. 539–579. Academic Press, London.
13) Shorb, D. A. (1933): Host-parasite relations of *Hymenolepis fraterna* in the rat and mouse. Am. J. Hyg., 18, 74–113.
14) Weinmann, C. J. (1964): Host resistance to *Hymenolepis nana*. II. Specificity of resistance to reinfection in the direct cycle. Exp. Parasitol., 15, 514–526.
15) Weinmann, C. J. (1966): Immunity mechanisms in cestode infections. In Biology of Parasites (Soulsby, E. J. L. ed.), pp. 301–320. Academic Press, New York.
16) Williams, J. F. (1982): Cestode infections. In Immunology of Parasitic Infections (Cohen, S. and Warren, K. S. eds.), pp. 676–714. Blackwell, Oxford.