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Size as a Factor in the Infection of *Tribolium* spp. (Coleoptera: Tenebrionidae) and *Eleodes* sp. (Coleoptera: Tenebrionidae) by *Hymenolepis diminuta* (Cestoda)

SAROJAM K. MANKAU

California State College, 5500 State College Parkway, San Bernardino, California 92407

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Introduction

Tribolium spp. have been established as a successful intermediate host for the rat tapeworm, *Hymenolepis diminuta*. The tapeworm ova are passed out of the rat host with the fecal matter. When they are ingested by a suitable species of beetle, the eggs hatch in the intestine. The oncospheres then penetrate the intestinal wall and enter the haemocoel where they develop into cysticercoids. Factors affecting the incidence of infection in beetles has been investigated. Role of sex of the beetles in infection has been studied by Mankau *et al.* 1971, where they reported a significantly higher rate of infection in the female beetles compared to the males. A comparative study of the two sexes in three different species of *Tribolium* showed that in *T. confusum* (Duval) and *T. castaneum* (Herbst) a significantly greater percentage of female beetles became infected compared to the males and that the females of those two species harboured significantly greater numbers of cysticercoids per beetle than the males. *T. brevicornis* (Lec) showed no significant difference between the sexes in the percentage of infected beetles or in the number of cysticercoids harboured by each beetle (Mankau 1977). The role of age of beetles on the incidence of *Hymenolepis* infection in *T. confusum* beetles had been reported by Kelly *et al.* (1967) where they observed that old females had a significantly smaller burden of cysticercoids when compared to young or middle-aged females, whereas middle-aged males generally had a higher incidence only where compared with young or old males, concluding that age resistance to *H. diminuta* cysticercoids in *T. confusum* occurs only in the females.

The purpose of the present study was to determine whether or not the differences in size and weight between a pygmy mutant strain and the normal strain of *T. castaneum* has a significant effect on the number of *H. diminuta* cysticercoids harboured by the beetles. Also whether *Eleodes* sp., a commonly found beetle in So. California could serve as an intermediate host for *H. diminuta* and if so, whether its relatively huge size would harbour a proportionate number of cysticercoids per gram of body weight.

Materials and Methods

Sixty beetles of normal strain and sixty of the mutant pigmy strain of *Tribolium castaneum* species were obtained from Dr.

Table 1 Rate of infection and incidence of *H. diminuta* cysticercoids in experimentally infected *T. castaneum* (N), *T. castaneum* (py) and *Eleodes*

Beetle	Mortality	Survivors	Infected	% infection	Total cysticercoids	Cysticercoids (infected)	
						per beetle	per gm body wt.
T, castaneum (N)	2	58	41	71	138	3.4	1.36
T. castaneum (py)	8	52	33	64	95	2.9	2.37
Eleodes	21	9	1	10	2	2.0	
Total	31	119	75	63	235	3.1	

Alex Sokoloff, Director, Tribolium Stock Center, California State College at San Bernardino. Fifteen *Eleodes* sp. beetles were collected from the Riverside-San Bernardino area. The beetles were denied food for three days prior to the infection. They were grouped in petri dishes, each containing ten *Tribolium* beetles/dish. *Eleodes* sp. were separated in groups of five. All beetles were maintained at 17 Celcius.

Hymenolepis diminuta inoculum was obtained by removing tapeworms from an infected white rat (Carolina Biological Supply, Inc.) Gravid proglottids were placed in mammalian ringer's solution and teased apart to liberate the ova. Ova concentration was determined by direct count under a dissecting microscope. A suspension of approximately 200–240 ova per drop was used to infect the beetles by placing a drop of the inoculum on an oatmeal flake 3–4 mm in diameter. The inoculum was absorbed into the oatmeal flake and five such flakes were placed in each of the petri dishes containing the beetles, providing approximately 1,000–1,200 ova per dish. The petri dishes were covered and taped to prevent dessication of the ova. Most of the oatmeal was consumed in the first two days and subsequently the beetles were fed their regulat diet of 95% unbleached wheat flour and 5% brewer's yeast. After seven weeks all the beetles were dissected under a dissecting microscope and the cysticercoids counted.

Results

Thirty-one of the initial 150 beetles died during the experiment. Sixty-three percent of the remaining 119 beetles were infected. The survivors consisted of 58 *T. castaneum* (normal), 52 *T. castaneum* (pigmy) and 9 *Eleodes,* showing 71, 64 and 10 percent infection respectively. (Table I)

Prevalence of *H. diminuta* cysticercoids per infected beetle was highest in *T. castaneum* (N) with 3.4 per beetle, followed by *T. castaneum* (P) with 2.9 per beetle, with *Eleodes* harbouring 2 cysticercoids, both found in the single infected beetle.

The average number of cysticercoids harboured per infected *T. castaneum* (N) beetle (3.4) is not significantly greater than those found in the mutant, *T. castaneum* (P) (2.9) (Table 1). This is remarkable, since the pigmy mutant weighs only half as much as the normal *T. castaneum* beetle. This therefore indicates that the difference in size and weight between *T. castaneum* (N) and *T. castaneum* (py), has no significant effect on the incidence of *H. diminuta* cysticercoids. The pigmy mutant harboured significantly larger numbers of cysticercoids per gram of body weight (2.4) compared to *T. castaneum* (N) (1.4).

Discussion

In a study (Mankau 1977) using 3 different species of *Tribolium* beetles namely *T.*

castaneum, *T. confusum* and *T. brevicornis*, as intermediate hosts for *Hymenolepis diminuta* it was found that the average number of cysticercoids harboured by *T. castaneum* was greater than in *T. brevicornis* even though the latter species weighed 8–10 times more than *T. castaneum* and *T. confusum*.

Although only one out of the 30 *Eleodes* sp. beetles used in the experiment became infected, it nevertheless is significant. This beetle is commonly found in the fields and backyards of southwestern United States and no doubt serves as a source of food for wild rats and mice. The cysticercoids recovered from this beetle were normal in appearance and development and therefore it is evident that they can serve as effective intermediate hosts for *Hymenolepis diminuta*.

It is possible that the extremely large mouth parts in *Eleodes* sp. is related to the very low infection rate. Voge and Berntzen (1961) in their study of "in vitro" hatching of *H. diminuta* oncospheres have shown that the rupture of the egg shell of the ova is necessary for the successful hatching and continued development of cysticercoids. *H. diminuta* ova contain an oncosphere enclosed by four membranes and an outer egg capsule or "shell". The outermost membrane is very thin and is almost invisible in intact eggs. The second membrane is the vitelline membrane which becomes distended during the hatching process. The embryophore forms the third membrane inside of which is the thin inner membrane. The rupture of the egg capsule is normally caused by the mandibles of the beetle. An interesting case was observed in our laboratory during an experiment, where *T. confusum* were infected with *H. diminuta* ova, and obtained 77% infection with mature infective cysticercoids, one beetle was found to contain approximately 450 ova in its gut, with the egg shell intact, but harboured no cysticercoids. When the mouth parts of the beetle were examined it was found to have a defective mandible, affecting its ability to properly puncture the egg capsule on the oncospheres.

The large mouth parts of the *Eleodes* beetle could have permitted the passage of the majority of the oncospheres without rupture of the membranes thereby preventing subsequent development into cysticercoids. Experiments are currently in progress to test the above hypothesis by introducing artifically hatched oncospheres directly into the hemocoel of *Eleodes* beetles.

Summary

Sixty beetles of normal strain and sixty of the mutant pigmy strain of *Tribolium castaneum* obtained from the Tribolium Stock Center and thirty *Eleodes* sp. collected from the Riverside-San Bernardino area were infected with ova collected from gravid segments of Hymenolepis diminuta from a white rat. Seven weeks after the infection all the beetles were dissected and the cysticercoids were counted. Seventy-one, sixty-four, and ten percent of *T. castaneum* (normal), *T. castaneum* (pigmy) and *Eleodes* sp. respectively, were infected. Even though the normal strain of *T. castaneum* weighs twice as much as the pigmy strain, the number of cysticercoids per beetle harboured by the former (3.4) was not significantly greater than those found in the latter (2.9). The pigmy mutant harboured significantly larger numbers of cysticercoids per gram of body weight (2.4) compared to *T. castaneum* (N) (1.4).

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甲虫 *Tribolium* spp. (Coleoptera: Tenebrionidae) と *Eleodes* sp. (Coleoptera: Tenebrionidae) への縮小条虫の感染
—— 一感染要因としての甲虫虫体の大きさについて ——

Sarojam K. MANKAU
(カリフォルニア州立大学生物学教室)

ラットに寄生していた縮小条虫の受胎節から集めた虫卵を，California 州立大学 Tribolium Stck Center より入手した甲虫 *Tribolium castaneum*（コクヌストモドキ）の野生型60匹と同じく突然変異ピグミー型60匹，並びに Riverside-San Bernardino で採集した甲虫 *Eleodes* sp. 30匹に感染させた．感染後7週目にすべての甲虫を解剖精査し，擬嚢尾虫数を数えた．*T. Castaneum*（野生型）の 71%，*T. castaneum*（ピグミー型）の 64%，*Eleodes* sp. の 10% がそれぞれ感染していた．*T. castaneum* の野生型の体重はピグミー型のそれの2倍あったが，野生型の1匹当りの擬嚢尾虫寄生数（3.4）はピグミー型のそれ（2.9）と有意な差はなかった．ピグミー突然変異型は *T. castaneum*（野生型）（1.4）に比較して有意に多数の体重グラム当りの擬嚢尾虫数（2.4）を保有していた．