

EFFECTS OF A TAPEWORM PARASITE ON THE COMPETITION OF *TRIBOLIUM* BEETLES

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Abstract. The effect of parasites on host community structure and biodiversity is an important issue of community ecology. Both verbal and mathematical models suggest that host species with higher fitness costs associated with parasitism should bear a competitive disadvantage in the presence of parasites. However, few rigorous empirical tests exist. This study examined the effects of a tapeworm parasite (*Hymenolepis diminuta*) on competition between two flour beetle species (*Tribolium confusum* and *T. castaneum*). The flour beetles are the intermediate host of this rat tapeworm parasite. Previous studies demonstrated that *T. castaneum* is usually the superior competitor in competition with *T. confusum* in parasite-free environments. However, because *T. castaneum* is more susceptible to parasitism and shows higher fitness costs when infected than *T. confusum*, we expected that *T. castaneum* would bear a competitive disadvantage in the presence of the parasite. In contrast, we found that tapeworm infection significantly increased the likelihood of *T. castaneum* winning the competition, reduced the time needed for *T. castaneum* to win, and increased *T. castaneum* population density compared to parasite-free populations. Therefore, our results suggest that the tapeworm infection confers some advantage to *T. castaneum* and are in contrast to the expectation based on mathematical studies and verbal arguments. Disagreements between experimental results and the hypothesized predictions probably reflect the effect of parasite-induced changes in intraguild predation. Our results suggest that theories on parasite effects of competition need to consider the complex nature of host–parasite associations, including the effect of parasites on host behaviors.

Key words: coexistence; *Hymenolepis diminuta*; intraguild predation; parasite; *Tribolium*.

INTRODUCTION

The effect of parasites on host community structure and biodiversity is an important issue of community ecology. Both verbal (e.g., Barbehenn 1969, Anderson 1972, Saunders 1973, Holmes 1979, Price et al. 1986, 1988, Jaenike 1992) and mathematical (e.g., Holt and Pickering 1985, Bowers and Begon 1991, Begon et al. 1992, Begon and Bowers 1994, 1995, Yan 1996) models of the effect of parasitism on host competition agree in the prediction that host species with higher fitness costs associated with parasitism should bear a disadvantage in competition. For example, Holt and Pickering (1985) show that the mode of parasite transmission and pathogenicity may have significant consequences for the coexistence or exclusion of two sympatric host species in the absence of any other regulatory factor. The general model of Yan (1996) extended the classic Lotka–Volterra competition model and concluded that parasitism can affect host coexistence, and the outcome of host competition depends on host susceptibility to the parasite, the parasite's pathogenicity, and other parameters determining host pop-

ulation dynamics. Some field data, although not designed to test this specific prediction, suggest that parasite-mediated competition is taking place in nature (e.g., Feener 1981, Freeland 1983, Settle and Wilson 1990, Grosholz 1992, Schall 1992).

We present here the results of a laboratory experiment on the impact of a tapeworm parasite, *Hymenolepis diminuta*, on the competition between two species of *Tribolium* beetle. The beetle community involves two types of interspecific interactions (Park 1962): (1) exploitative competition for shared resource (e.g., food and space); and (2) interspecific predation in an age-structured community—predation of mobile life stages (adults and larvae) on nonmobile life stages (eggs and pupae; Park et al. 1965, Stevens and Mertz 1985). The tapeworm parasite has a negative effect on host reproductive success that may affect the competition coefficients of the beetle (Yan 1997). Park and his colleagues (e.g., Park 1948, 1957, Leslie et al. 1968) demonstrated that *T. castaneum* is usually a superior competitor to *T. confusum*, probably because *T. castaneum* is more predatory intraspecifically and interspecifically than *T. confusum*. Craig (1986) showed that *T. castaneum* adults (genetic strain c+) preferentially eat *T. confusum* (strain b+) eggs when the eggs of both species are present in the flour medium. He further demonstrated that *T. castaneum* adults prefer larger eggs, and *T. confusum* eggs are larger than *T. castaneum*

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eggs. Predicting the competitive outcome between the two beetle species is complicated because the competition may be influenced by demographic stochasticity (e.g., stochastic fluctuations in birth and death rates), and genetic stochasticity (e.g., genetic drift resulting from small numbers of founders; see Park 1948, Park et al. 1964, Dawson and Lerner 1966, Dawson 1970, Mertz et al. 1976, Goodnight and Craig 1996).

The study by Park (1948) on the effect of a protozoan parasite (*Adelina tribolii*) on the competitive outcome of two flour beetle species has been a classical example in the textbooks. His study has inspired much of the subsequent interest in parasite-mediated competition, but the reversal of the competitive outcome by the protozoan parasite is not surprising because the parasite is more infectious to *T. castaneum* and often lethal to immature beetles. *Tribolium castaneum* may simply be eliminated by the parasite. In this study we considered a different situation: the effect of a parasite with low to intermediate virulence on interspecific competition. We also addressed the role of stochastic vs. deterministic forces in the outcome of competition by varying starting densities of the competitors. This study did not intend to test a particular mathematical theory, but was designed to test the general hypothesis that a more susceptible host species, which shows higher pathogenicity associated with parasitism, should bear a disadvantage in competition with the less susceptible host species.

Parasite pathogenicity and host susceptibility are important parameters for parasite effect on host competitive outcome. In our system, these parameters are known. *Tribolium castaneum* is more susceptible to infection with the tapeworm than *T. confusum* (Yan and Norman 1995). That is, a higher proportion of *T. castaneum* become infected (higher prevalence), and infected *T. castaneum* contains more parasites (higher infection intensity). Parasite-induced reduction in beetle fitness (reproductive success) is significantly higher for *T. castaneum* than for *T. confusum*, probably due to a higher level of susceptibility to the parasite in *T. castaneum* (Yan 1997). Further analysis on the fitness components of *T. castaneum* revealed that the parasite infection reduced female fecundity and male sperm precedence, and increased adult cannibalism of eggs (Yan and Stevens 1995). Based on the above information we predicted that *T. castaneum* should bear a disadvantage in competition with *T. confusum* in the presence of the tapeworm parasite.

The culture conditions used in this experiment always lead to the competitive exclusion of one of the two species (Goodnight and Craig 1996). Thus, we considered competition to be resolved when only one species (the "winning" species) is left and one species (the "losing" species) has gone extinct. We measured three community-level parameters of these competitive interactions: (1) the outcome of competition (which species wins); (2) the time to extinction of the losing

species; and (3) the profile of the census history. Our hypothesis predicts that the presence of the tapeworm parasite would result in increased probability of *T. confusum* winning, shorter time to extinction for *T. castaneum*, and increased population density of *T. confusum*.

MATERIALS AND METHODS

Beetles and parasites.—*Tribolium* beetles in nature are intermediate hosts of *H. diminuta* and the parasite is readily maintained in the laboratory (Arai 1980, Keymer 1981). The adult parasite lives in the small intestine of rats, and produces eggs that pass out with the host's feces. After parasite eggs are ingested by the proper intermediate hosts (a wide variety of insects including beetles; Arai 1980), they hatch and develop into cysticercoids. Cysticercoids are infective to mammals only when the infected intermediate hosts are eaten by mammals. They are not horizontally or vertically transmissible from beetle to beetle. Thus, in the laboratory, the infection can be controlled and monitored. The natural environment of the beetles, clumps of raw or processed grain, can be closely mimicked in the laboratory.

Tribolium confusum (strain b+-black) and *T. castaneum* (strain cSM-red) were used in this study. The b+ strain has been maintained in laboratory culture since the 1930s (Park 1948). Strain cSM originated by combining several laboratory strains in the 1970s (Wade 1977). Both of these are standard laboratory strains that have been used extensively in ecological and evolutionary studies (e.g., Park 1948, 1954, Park et al. 1964, Wade 1977, 1985, Stevens 1989, Wade and Goodnight 1991, Goodnight and Craig 1996). Wild-type flour beetles are red. Strain b+-black is homozygous for a black body color marker that facilitates distinguishing the two species in the experimental communities (Wade 1985, Pray et al. 1994). The black mutation was discovered segregating in the b+ stock population. A mutant stock was derived by selecting over 200 black individuals from the original stock (Wade 1977). The effect of the black body color marker has not been estimated in *T. confusum*; however estimates for a similar mutant phenotype in *T. castaneum* indicate it is either neutral (Sokoloff and Slatis 1960) or has a small (5–10%) negative fitness effect (Rich and Bell 1979). Since we are interested in the effect of a tapeworm on the outcome of competition using infected and uninfected populations, fitness effects of the black mutation should not affect our conclusion. The beetles used in the following experiments came from stock cultures that have not been exposed to the tapeworm since they have been cultured in laboratory, ~200 generations for b+ and ~70 generations for cSM.

Fresh rat feces mixed with *H. diminuta* eggs were obtained from Carolina Biological Supply Company, Burlington, North Carolina, USA. Carolina has maintained rats infected with *H. diminuta* for the past 25

yr. The beetles were exposed to the tapeworm eggs immediately after we received the culture (~24 h after the rat feces were collected). All beetles were infected simultaneously using the randomized design described below.

Method of infection.—Twenty beetles were placed in a 2.5 cm diameter \times 8 cm height (8-dram), shell vial and starved for a week before they were exposed to 0.3 g of infected rat feces mixed with 0.2 g of distilled water on a 2 \times 6.5 mm strip of filter paper for 24 h. The infected rat feces were stirred to increase homogeneity before being applied to the filter paper and we randomized the order in which the filter paper was added to the vials to diminish the effects of uneven distribution of tapeworm eggs (Yan and Norman 1995). Microscopy examination found that ~1000 parasite eggs could be extracted from this amount of infected rat feces. Control beetles were also starved and fed with 0.3 g of uninfected rat feces mixed in 0.2 g distilled water on filter paper for 24 h to control for the effect of rat feces. The filter paper was then removed and 8 g of flour medium were added to each vial. This protocol was used to infect all beetles in appropriate treatments at every generation.

Experimental design.—The founder density treatments were factorial combinations of 0, 4, 8, and 16 adults for *T. castaneum*, and 0, 16, 64, and 128 adults for *T. confusum*, the sex ratio being equal. We started with a higher density of *T. confusum* because *T. castaneum* is usually a superior competitor (Park 1948), and we wanted the parasite-free competition to be close to equal. We maintained two treatments: infected treatment and uninfected controls. In the infected treatment adults of both species were infected; in the controls neither species was infected. We maintained 10 replicates per density-infection treatment, thus the experiment consisted of 300 populations ([6 single-species density treatments + 9 two-species density treatments] \times 2 infection treatments \times 10 replicates = 300 populations). A total of 2240 virgin *T. castaneum* beetles were used to set up the experiment: (4 + 8 + 16) for one replicate of each density \times 10 replicates \times 2 infection treatments = 560 beetles for single-species cultures, and (4 + 8 + 16) \times 10 replicates \times 2 treatments \times 3 combinations = 1680 beetles for two-species cultures. Similarly, a total of 16 640 virgin *T. confusum* beetles were employed.

We follow the protocol of Goodnight and Craig (1996) and maintained discrete-generation populations. Beetles were placed in a 2.5 cm diameter \times 8 cm height (8-dram) shell vial containing 8 g of standard flour medium (95% by mass fine sifted whole wheat flour and 5% dried powdered brewer's yeast). The vials were maintained in a dark incubator regulated at 29°C and the adults were allowed to oviposit for 7 d. After 7 d the adults were removed and discarded. After 56 d the number of adults of each species in each vial was counted. During this period beetle eggs would develop into

adults, but as a result of adult cannibalism of eggs, no further population growth occurred. *Tribolium confusum* eggs normally take ~6 wk to develop into mature adults under our experimental conditions, and *T. castaneum* has a slightly shorter developmental time. Identification of beetle species was based on adult body color phenotype, and species identity was further confirmed by microscopic examination. All adult beetles in the infected treatment were exposed to the parasites every generation using the protocol described above, and a fresh batch of parasite eggs was used for each generation. All surviving adults were then transferred into new glass vials with fresh flour medium to start a new generation. The experiment was terminated after we observed no host coexistence in all populations (nine generations). At generations 0 (founders), 2, and 5, we dissected 39–56 beetles from several two-species cultures for each species to determine infection intensity and prevalence.

Data analysis.—The data were analyzed in four ways. First, we analyzed between-species variation in infection intensity and prevalence (infection intensity and prevalence are defined in the *Introduction*), using the Wilcoxon rank sum test (SAS 1994) and the *G* test (Sokal and Rohlf 1981), respectively. We previously determined that among-vial variation in infection intensity is negligible, compared to the between-species variation (Yan and Norman 1995). Therefore, infection intensity data were pooled for various vials. Second, we analyzed the outcome of competition using log-likelihood analysis. Third, we used univariate analysis of variance to examine the time until one of the two species went extinct (time to extinction). Finally, the population dynamics of single-species cultures and two-species cultures were analyzed using population profile analysis (Harris 1985).

Identity of the winning species.—The experiment was terminated when one species had gone extinct in each of the two-species cultures, and the winning species in a particular population were identified. The identity of the winning species was analyzed using log-likelihood analysis, fitting a log-linear logistic response to the complete model. We calculated the χ^2 statistics and probabilities for the main factors and their interactions using likelihood ratio tests (Sokal and Rohlf 1981, Goodnight and Craig 1996). Significance indicates that the probability of the species winning is affected by the independent factors: infection and initial densities of *T. confusum* and *T. castaneum*.

Time to extinction.—We defined the time to extinction to be the first census in which there were no members of one of the two species. The univariate analysis of variance, with infection and initial densities of *T. confusum* and *T. castaneum* as main factors, was used to examine the effects of these factors on the time to extinction. Winning species was included in the analysis as a covariate and was nested within infection.

Population dynamics.—Population dynamic data

TABLE 1. Infection intensities and prevalences of the beetles used in the competition experiments.

Generation	Sex†	<i>T. castaneum</i>			<i>T. confusum</i>		
		N‡	Prevalence (%)	Mean intensity§ (SD)	N‡	Prevalence (%)	Mean intensity§ (SD)
0	Male	43	69.77	4.17 (3.70)	39	46.15	1.89 (1.60)
	Female	40	62.50	3.72 (2.56)	42	47.62	1.40 (0.94)
2		52	42.31	2.27 (1.52)	52	28.85	1.47 (0.52)
5		51	37.25	1.84 (1.21)	56	21.43	1.42 (0.67)

† Beetles were not sexed after generation 1. Infection data were pooled for both sexes.

‡ N is the number of beetles dissected.

§ Units are number of tapeworm parasites.

were analyzed using profile analysis (Harris 1985). We tested one possibility: are there differences between treatments in the mean densities of the beetles (level hypothesis). For two-species cultures, we tested the level hypothesis using multivariate analysis of variance (MANOVA) with population densities of the two beetles as dependent variables, and with infection, initial densities of the two beetle species and the interactions as independent variables. Vials were included in the analysis as a covariate and nested within infection and initial densities. Censuses were not included in the model, and we did not perform separate MANOVA for each census, thus this analysis examined the effect of various factors on the average population densities of each species over various censuses. If one species went extinct at census *i*, a zero density was scored for this and all subsequent censuses, and used for the MANOVA. Significances of the mean-square variance and covariance matrix deviation from zero were tested using the critical values of the greatest characteristic root (GCR) distribution of the matrix (Harris 1985: 444).

For single-species *T. confusum* cultures, significant number of lineages went extinct simply because the larvae failed to develop into pupae under overcrowded conditions, particularly for cultures of high initial densities. For example, half of populations with initial densities of 64 and 128 beetles went extinct at generation 2, irrespective of population infection status. For populations with 16 *T. confusum* beetles as initial densities, 9 out of 20 populations went extinct at generation 4. *Tribolium confusum* single-species population dynamics are reported for populations with 16 beetles as initial densities. Population profile analyses of the single-species cultures were performed in the same manner as those of the two-species cultures, except that univariate analysis of variance was used. The populations that went extinct due to overcrowded conditions were not included in the analysis.

RESULTS

Beetle infection.—The infection intensity and prevalence varied among generations and between species (see Table 1). Both infection intensity and prevalence for *T. castaneum* were significantly greater than for *T. confusum* at generation 0 (Wilcoxon test for intensity,

$\chi^2 = 19.01$, $df = 1$, $P < 0.001$; *G* test for prevalence, $\chi^2 = 6.29$, $df = 1$, $P < 0.05$), but not at generations 2 and 5. Comparisons between species for each generation are independent, so a combined probability test (Sokal and Rohlf 1981) was used and showed that *T. castaneum* exhibited significantly higher susceptibility to the parasite than *T. confusum* ($\chi^2 = 16.89$, $df = 2$, $P < 0.01$ for prevalence; $\chi^2 = 33.25$, $df = 2$, $P < 0.01$ for intensity). Comparison of beetle susceptibility to the parasite among generations was not appropriate because different batches of parasite eggs were used for each generation.

Identity of the winning species.—*Tribolium castaneum* was the superior competitor in our discrete-generation cultures (Tables 2 and 3 for statistical analysis). For example, when the initial densities of the two species were the same (culture 3 of Table 2), *T. castaneum* won the competition in all 10 vials for both infected and uninfected treatments. Tapeworm infection had an overall highly significant effect on the identity of the winning species (Tables 2 and 3). Infection significantly increased the chance of *T. castaneum* winning the competition in various combinations of founder densities (Table 2). Initial densities also had a significant effect on the winning species (Tables 2 and 3). For example, in an uninfected culture with an initial density of 16 *T. confusum* and 8 *T. castaneum* (culture 2 of Table 2), *T. confusum* did not win in any of the 10 vials. When the initial density of *T. confusum* increased to 128 (culture 8 of Table 2), *T. confusum* won 8 out of 9 vials. The interaction between infection and initial densities of *T. confusum* and *T. castaneum* significantly affected the outcome of competition. For example, in uninfected cultures an increase in *T. confusum* initial density usually increased the chances of *T. confusum* winning, but when both species were infected, increases in *T. confusum* density had less of an effect (Table 2).

Time to extinction.—The initial densities affected not only the identity of the winning species, but also the mean time to extinction (Tables 2 and 4). Generally, the mean time to extinction of *T. confusum* decreased with increasing *T. castaneum* initial density for a given *T. confusum* initial density.

ANOVA reveals that infection did not have a sig-

TABLE 2. Summary of the results of competition experiment.

Culture	<i>T. confusum</i> initial density†	<i>T. castaneum</i> initial density†	<i>T. confusum</i> as winner		<i>T. castaneum</i> as winner	
			Probability	Mean extinction time for <i>T. castaneum</i> (SD)‡	Probability	Mean extinction time for <i>T. confusum</i> (SD)‡
1. Uninfected	16	4	0.10	3.00 (...)	0.90	7.22 (1.202)
2. Infected	16	4	0.33	5.33 (1.155)	0.66	6.67 (1.506)
3. Uninfected	16	8	0.00	...	1.00	4.90 (0.738)
4. Infected	16	8	0.00	...	1.00	4.20 (1.033)
5. Uninfected	16	16	0.00	...	1.00	4.10 (0.738)
6. Infected	16	16	0.00	...	1.00	3.60 (0.699)
7. Uninfected	64	4	0.40	1.75 (0.957)	0.60	4.50 (2.950)
8. Infected	64	4	0.10	6.00 (...)	0.90	5.44 (1.509)
9. Uninfected	64	8	0.90	4.00 (1.732)	0.10	2.00 (...)
10. Infected	64	8	0.00	...	1.00	4.70 (1.160)
11. Uninfected	64	16	0.50	3.20 (1.304)	0.50	3.20 (0.837)
12. Infected	64	16	0.00	...	1.00	3.50 (0.707)
13. Uninfected	128	4	0.90	1.89 (0.782)	0.10	7.00 (...)
14. Infected	128	4	0.20	4.00 (0.000)	0.80	4.12 (3.044)
15. Uninfected	128	8	0.89	2.88 (1.356)	0.12	6.00 (...)
16. Infected	128	8	0.10	4.00 (...)	0.90	3.33 (0.707)
17. Uninfected	128	16	0.44	3.25 (2.062)	0.56	6.20 (...)
18. Infected	128	16	0.00	...	1.00	3.10 (0.316)

† Initial density = no. individuals per culture.

‡ Mean extinction time was measured by the number of censuses when one species went extinct in a two-species culture.

§ Ellipses indicate that the parameters could not be calculated.

nificant overall effect on the time to extinction; however, the interaction between infection and *T. confusum* initial density was significant (Table 4). ANOVA orthogonal contrasts revealed that infection significantly reduced the time to extinction of *T. confusum* for six cultures (cultures 13–18; Table 2). Parasitism significantly reduced the time for *T. castaneum* to win the competition (7.01 censuses for uninfected controls vs. 6.37 for infected cultures; ANOVA orthogonal contrasts, $t = 2.958$, $df = 1$, $P < 0.001$).

Population dynamics of cultures.—The population dynamics of single-species cultures are shown in Fig. 1. For *T. castaneum*, populations seemed to approach an equilibrium density after four generations (Fig. 1A). We found that neither initial density nor infection showed significant effects on the mean population sizes for either species (Table 5). We further examined the possibility that parasite infection might affect population densities at a particular generation, by reanalyzing the data using ANOVA with infection and initial densities as main factors for each generation. We found that beetle density in the infected treatment was sig-

nificantly higher than the uninfected control on generation 2 ($F = 43.6$, $df = 1$, 53 , $P < 0.001$), but significantly lower on generation 3 ($F = 55.6$, $df = 1$, 52 , $P < 0.001$; see Fig. 1A). Beetle initial density showed a significant effect only on generation 1 ($F = 3.6$, $df = 1$, 53 , $P = 0.04$). Neither factor was significant after generation 4. For *T. confusum*, beetle density in the infected treatment was significantly higher than the uninfected control on generations 1, 5, and 7, but significantly lower on generation 3 (Fig. 1B)

The population dynamics of infected and uninfected populations of the two-species cultures with various initial densities are shown in Fig. 2. The mean population densities of *T. castaneum* in the uninfected two-species cultures were significantly smaller than those of single-species cultures (ANOVA, $F = 68.29$, $df = 1$, 711 , $P < 0.001$; Figs. 1 and 2), confirming that competition existed in the two-species cultures.

Profile analysis of the population dynamics of the two-species cultures revealed a significant effect of beetle initial densities on the population mean densities (level hypothesis), but the overall effect of parasite

TABLE 3. Log-likelihood analysis of the outcome of competition.

Source of variation	df	χ^2	P
Infection	1	13.795	<0.001
<i>T. confusum</i> initial density	2	44.438	<0.001
Infection \times <i>T. confusum</i> initial density	2	83.042	<0.001
<i>T. castaneum</i> initial density	2	64.239	<0.001
Infection \times <i>T. castaneum</i> initial density	2	0.000	>0.05
<i>T. confusum</i> initial density \times <i>T. castaneum</i> initial density	4	21.910	<0.001
Infection \times <i>T. confusum</i> initial density \times <i>T. castaneum</i> initial density	4	16.252	<0.001

TABLE 4. Analysis of variance of the time to extinction.

Source of variation	df	MS	F	P
Infection	1	2.277	1.256	>0.05
<i>T. confusum</i> initial density	2	232.316	128.172	<0.001
Infection \times <i>T. confusum</i> initial density	2	39.855	21.989	<0.001
<i>T. castaneum</i> initial density	2	338.998	187.026	<0.001
Infection \times <i>T. castaneum</i> initial density	2	0.427	0.236	>0.05
<i>T. confusum</i> initial density \times <i>T. castaneum</i> initial density	4	126.989	70.062	<0.001
Infection \times <i>T. confusum</i> initial density \times <i>T. castaneum</i> initial density	4	26.283	14.501	<0.001
Winner [infection]†	2	198.290	109.399	<0.001
Error	1432	1.813		

Note: Data were not transformed.

† Infection was nested within the winning species.

infection was not significant (Table 6). Instead, we found significant interaction between infection and host initial densities. This result suggests that the statistical significance for the effect of infection on population mean densities may depend on the combination of host initial densities. We tested this possibility using MANOVA orthogonal contrasts (SAS 1994), and found that tapeworm infection significantly affected the population dynamics of the two beetle species in six of the nine treatments (cultures 7–18 in Table 2, $P < 0.001$ for all six comparisons; see Fig. 2D–I). Moreover, parasitism influenced the populations of the two species in fundamentally different ways. Parasitism significantly lowered the mean population size of *T. confusum*, but the mean population sizes of infected *T. castaneum* populations were significantly increased (Fig. 2). These results demonstrate that the tapeworm infection provided an advantage to *T. castaneum* in competition with *T. confusum*.

In summary, these results show that the tapeworm infection affected the competition between the two *Tribolium* species. Such effects were reflected in the identity of the winning species, the mean extinction time, and the population mean densities. The tapeworm infection significantly increased the chances of *T. castaneum* winning the competition, especially in combinations with high *T. confusum* initial densities relative to *T. castaneum*. The extinction time of *T. confusum* was reduced by parasitism. Thus, *T. confusum* was more likely to lose the competition, and go extinct more rapidly in the presence of the parasite. In the two-species cultures, parasitism significantly decreased the mean population size of *T. confusum*, but increased the mean population densities of *T. castaneum*. The results suggest that the tapeworm infection placed *T. castaneum* at an advantage in competition with *T. confusum* in our discrete-generation cultures.

DISCUSSION

The notion that parasites might influence host competition has long intrigued ecologists (Haldane 1949, Freeland 1983, Price et al. 1986, 1988, Schall 1992). These authors presented verbal models and supporting

examples that suggest the way a parasite might alter the outcome of competition. For example, Schall (1992) found that the malarial parasite *Plasmodium azurophilum* infects two *Anolis* lizards on the Caribbean island of St. Martin. One anole is widespread throughout the island and is thought to be the superior competitor, the other is found only in a limited area of the island. The superior competitor is much more likely to be infected by the parasite. Schall found that the two anoles coexist only where the parasite is present. Mathematical models support predictions of such verbal arguments (Holt and Pickering 1985, Yan 1996).

The objective of our study was to examine the influence of a parasite on host competitive outcome, using two *Tribolium* species in the laboratory. *Tribolium castaneum* is known to be the superior competitor when matched with *T. confusum*, although demographic stochasticity significantly influences the outcome of competition (Park 1948, 1954, 1957, Mertz et al. 1976). Also, *T. castaneum* is more susceptible to infection with the tapeworm parasite, and harbors a higher parasite density when infected than *T. confusum* (Yan and Norman 1995). Infected *T. castaneum* exhibits a more severe reduction in reproductive success than *T. confusum* (Yan 1997). Therefore, the outcome of competition between the two beetle species could be affected by the tapeworm parasite: the formerly inferior competitor may be maintained in two species cultures, or may even drive down the density of the formerly superior species.

We first examined the effect of parasitism on the population dynamics of single-species cultures. We found that *T. castaneum* population density at equilibrium (approximately after generation 4) was not significantly affected by either beetle initial densities or parasite infection. A large number of *T. confusum* single-species populations, particularly the populations with high initial densities, went extinct. Population extinction was simply a result of beetle larval overcrowding, and was not a result of parasite infection. That is, a larger number of larvae appeared in the culture, but these larvae failed to develop into adults. Interestingly,

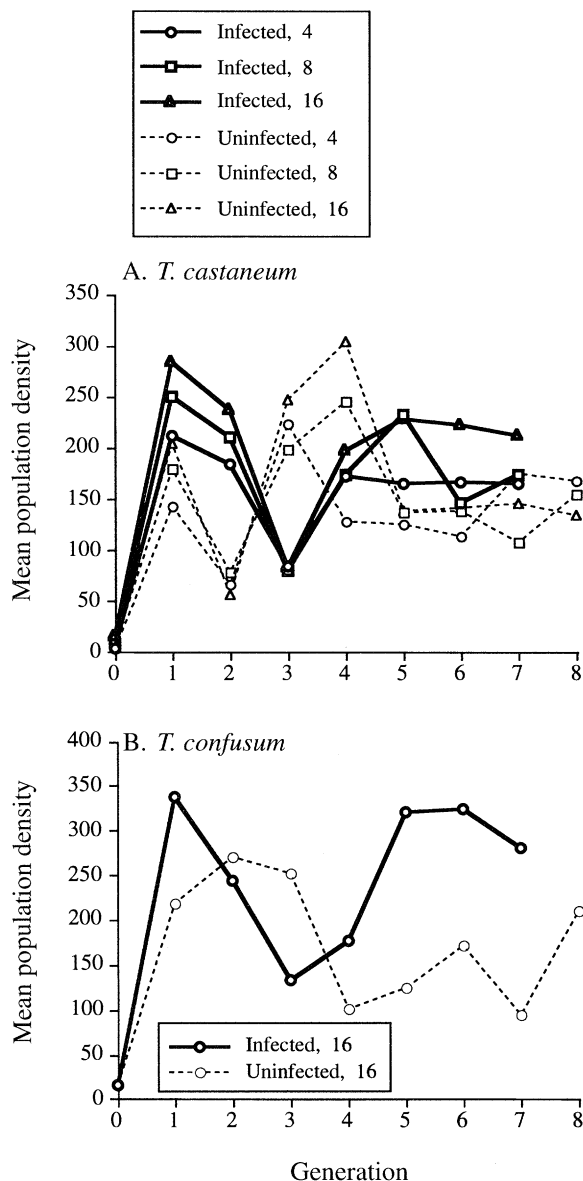


FIG. 1. Population dynamics of the single-species cultures. Discrete-generation populations were maintained for both infected treatments and uninfected controls. (A) *Tribolium castaneum* populations with 4, 8, and 16 adults as founders. (B) *T. confusum* populations with 16 adults as founders. The sex ratio of the founders was always 1:1.

infected *T. confusum* populations seem to maintain higher population density than the uninfected control, perhaps because parasite infection reduced beetle egg production, and thus more adults could emerge by reducing intraspecific competition among larvae. In two-species cultures, we did not observe the phenomenon of larval crowding. The presence of *T. castaneum* probably severely decreased the productivity of *T. confusum* females, or mating success of *T. confusum* males (Sokoloff 1974).

We then confirmed that *T. castaneum* was the superior competitor in our experimental system. Even when the initial densities of *T. confusum* were as much as eightfold higher than *T. castaneum*, *T. castaneum* had significantly higher probability of winning the competition. When the initial densities of both species were equal, *T. confusum* never won the competition. Next, the assays of susceptibility revealed that the superior competitor, *T. castaneum*, did have higher prevalence and mean infection intensity than the inferior competitor, *T. confusum*. The outcome of the experiments can now be examined to test the predictions leading from previous verbal and mathematical models about the effects of parasite on host competition. Specifically, *T. castaneum* is expected to bear disadvantage in competition with *T. confusum* in the presence of the parasite.

The experiments demonstrated that infection with the tapeworm affected competition between the two *Tribolium* species. Such effects were reflected in significant main effects and/or interaction terms in the analyses of the identity of the winning species, the mean extinction time and the population dynamics. Tapeworm infection significantly increased the chances of *T. castaneum* winning the competition. The parasitism reduced the mean extinction time of *T. confusum*, and reduced the time for *T. castaneum* to win the competition. Thus, *T. confusum* was more likely to lose the competition, and go extinct more rapidly in the presence of the parasite. In two-species cultures when both hosts were infected with the parasite, *T. castaneum* maintained significantly higher population density than the uninfected populations, but the mean densities of infected *T. confusum* populations were significantly lower than the uninfected. We conclude that in the discrete-generation, two-species cultures, the tapeworm infection placed *T. castaneum* at an additional advantage in competing with *T. confusum*.

These results contrast with the predicted outcome by previous models. Although *T. castaneum* is more prone to infection with the tapeworm and suffers more severe pathology, the parasite actually enhanced its ability to exclude *T. confusum*. At the very least, one would expect parasitism should not benefit the superior competitor when the superior competitor is more prone to infection and is most harmed by the parasite. The mechanisms causing this apparent contradiction are not clear. We suggest that parasite-induced behavioral alterations may be, at least in part, responsible for the discrepancy between our experimental results and model predictions. That is, *T. castaneum* wins the competition probably because it is more predacious than *T. confusum* (Park 1954, 1957, Leslie et al. 1968). If parasite infection increases predation of *T. castaneum* adults on eggs, then parasites may confer some advantages to *T. castaneum* in the competition.

Yan et al. (1994) found that the tapeworm parasites affect beetle cannibalism behavior: intraspecific pre-

TABLE 5. Population profile analysis of discrete-generation, single-species cultures.

Source of variation	df	MS	F	P
<i>T. castaneum</i>				
Infection	1	6.25	0.488	>0.05
Initial density	2	9.92	0.774	>0.05
Infection × initial density	2	1.79	0.140	>0.05
Vial (infection, initial density)†	6	8.31	0.643	>0.05
Error	310	12.93		
<i>T. confusum</i> ‡				
Infection	1	34.02	3.77	>0.05
Vial (infection)	2	6.03	0.66	>0.05
Error	86	12.93		

Note: The dependent variable was population density and was square-root transformed. The analysis involved random effects. The denominators for infection, initial density, and interaction between infection and initial density were synthesized by the SAS, JMP statistical program from linear combinations of the appropriate mean squares and had the same expectation as the effect to be tested under the null hypothesis. The degrees of freedom were adjusted for imbalance in the experiment by Satterthwaite's method (see SAS 1994).

† Vial was nested within infection and initial density and was treated as a random effect.

‡ *Tribolium confusum* single-species-culture data with an initial density of 16 beetles were used in this analysis.

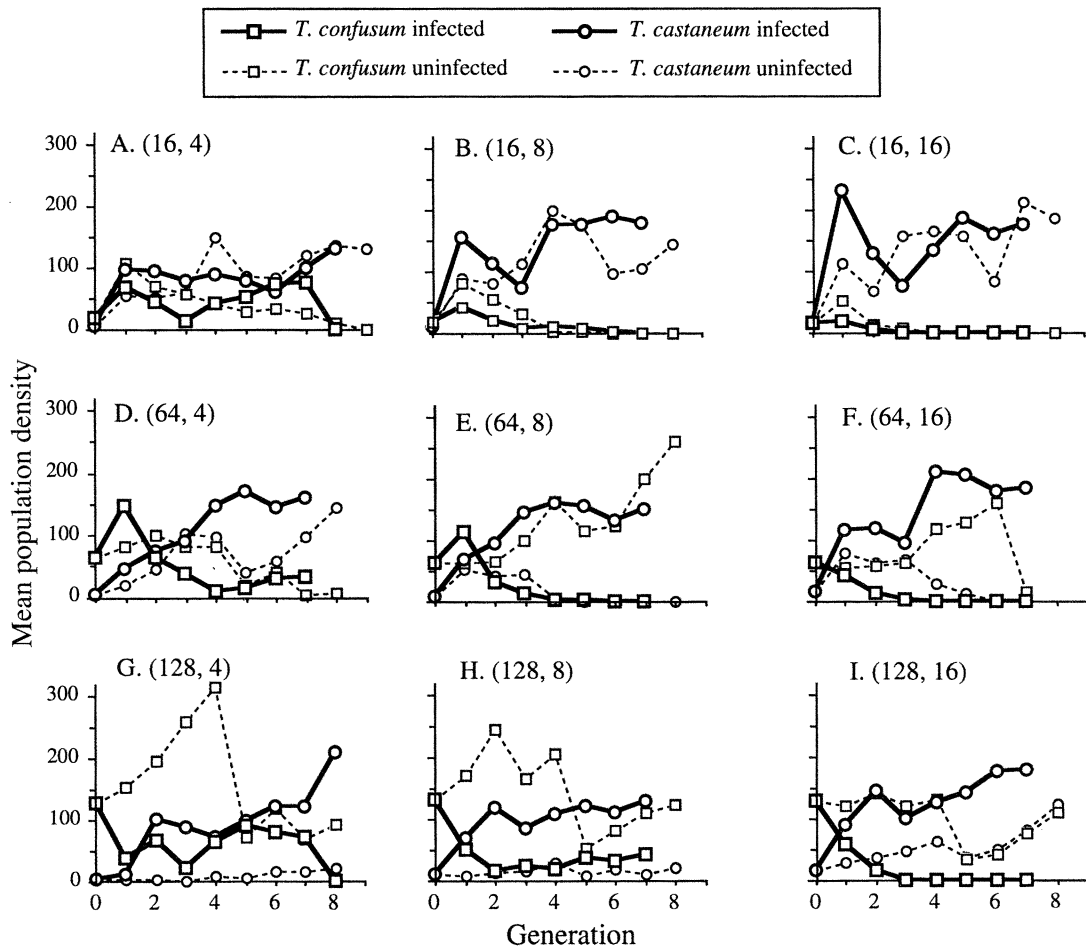


FIG. 2. Population dynamics of the two-species cultures. Discrete-generation populations were maintained for both infected treatments and uninfected controls. The founder densities are indicated (number of *T. confusum*, number of *T. castaneum*). The sex ratio of the founders was always 1:1. If one species went extinct at census i , its density was scored as zero for this and all subsequent censuses. The vertical axis, mean population density of a species, was the average of all replicates, including those that had zero densities of a species.

TABLE 6. Population profile analysis of the two-species cultures.

Source of variation	df	Mean-square covariance	P†
Infection	1	$\begin{bmatrix} 1.84 & -1.73 \\ -1.73 & 1.62 \end{bmatrix}$	>0.05
<i>T. confusum</i> initial density	2	$\begin{bmatrix} 88.56 & -105.66 \\ -105.66 & 177.30 \end{bmatrix}$	<0.001
Infection \times <i>T. confusum</i> initial density	2	$\begin{bmatrix} 142.94 & -130.33 \\ -130.33 & 126.18 \end{bmatrix}$	<0.001
<i>T. castaneum</i> initial density	2	$\begin{bmatrix} 27.17 & -17.86 \\ -17.86 & 13.79 \end{bmatrix}$	>0.05
Infection \times <i>T. castaneum</i> initial density	2	$\begin{bmatrix} 25.46 & -17.57 \\ -17.57 & 13.178 \end{bmatrix}$	>0.05
<i>T. confusum</i> initial density \times <i>T. castaneum</i> initial density	4	$\begin{bmatrix} 16.63 & -33.36 \\ -33.36 & 68.41 \end{bmatrix}$	<0.001
Infection \times <i>T. confusum</i> initial density \times <i>T. castaneum</i> initial density	4	$\begin{bmatrix} 62.66 & -55.07 \\ -55.07 & 50.47 \end{bmatrix}$	<0.001
Vial [infection, <i>T. confusum</i> initial density, <i>T. castaneum</i> initial density]‡	18	$\begin{bmatrix} 59.57 & -45.62 \\ -45.62 & 39.11 \end{bmatrix}$	<0.001
Error	1107	$\begin{bmatrix} 18.65 & -7.99 \\ -7.99 & 13.97 \end{bmatrix}$	

Note: Population densities of the two species were used as dependent variables. Data were square-root transformed. The factor vial was treated as a random effect. The denominators for infection, initial density, and interaction between infection and initial density were synthesized by the SAS, JMP statistical program from linear combinations of the appropriate mean squares and have the same expectation as the effect to be tested under the null hypothesis. The degrees of freedom were adjusted for imbalance in the experiment by Satterthwaite's method (see SAS 1994). Variance estimates for population size of *T. confusum* are in the upper left corner of each matrix, and variance estimates for population size of *T. castaneum* are in the lower right corner of each matrix. Covariances between the traits are in the upper right and lower left corners.

† Significance levels were determined using Roy's maximum-root criterion. Other commonly used criteria gave similar levels of significance.

‡ Vial was nested within infection and initial density and was treated as a random effect.

duction of eggs by *T. castaneum* adults infected with *H. diminuta* was significantly increased by 21.9%, but no change was observed in infected *T. confusum*. Such alterations in cannibalism may be related to flour-surface-seeking behavior. That is, infected *T. castaneum* adults were more likely to be found inside the flour medium and thus more often encounter and eat their own eggs than uninfected adults. One would expect that interspecific egg eating was also increased in infected *T. castaneum* adults. Because uninfected *T. castaneum* adults prefer eating *T. confusum* eggs rather than their own (Craig 1986), *T. confusum* eggs are likely to be the preferred prey by infected *T. castaneum* adults as well. Thus, increases in interspecific predation by infected *T. castaneum* adults would enhance the competition coefficient of this species. If parasite-induced changes in cannibalism is an important mechanism determining the outcome of beetle competition, it is expected that the effect of parasitism should be more evident in continuous two-species cultures than discrete cultures, because adult cannibalism of eggs should be more severe in continuous cultures. It is also expected that infected *T. castaneum* adults would decrease the rate of pupa cannibalism because pupae usually reside on the surface of flour medium. An excellent example that parasites affect host behaviors and subsequently alter host competitive outcome has been doc-

umented by Feener (1981): a parasitic phorid fly *Apocephalus* shifts the competitive balance between the ant species *Pheidole dentata* and *Solenopsis texana* by interfering with the defensive behavior of *P. dentata* soldiers, but not of *Solenopsis texana*. In our host-parasite-competitor system, *T. castaneum* is more susceptible to the tapeworm and shows more pathogenicity than *T. confusum*, thus putting *T. castaneum* at a disadvantage when competing with *T. confusum*. On the other hand, the parasite-induced host behavioral changes may give *T. castaneum* the upper hand. Therefore, our results probably reflect the importance of intraguild predation for the competitive outcome between two species of *Tribolium* beetles.

The differences between experimental results and predictions based on verbal or mathematical models demonstrate that knowledge of parasite virulence differences between host species is insufficient to predict the influence of parasites on the outcome of host competition. The outcome of competition may be influenced by parasite-induced behavioral alteration, particularly when this behavior is an important element of host population ecology and/or host-parasite interaction. We do not believe that this conclusion is unique to this system, but instead that it could well be a common feature of parasite-host biology. Intraguild predation is common in a variety of taxa with age-struc-

tered populations (Polis et al. 1989, Elgar and Crespi 1992). Examples of parasite alteration of host behavior are numerous (Moore and Gotelli 1990). The results suggest that theories about the effects of parasite on host competition need to consider the complex nature of host-parasite associations, including the effect of parasites on host behaviors.

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