

Original Article

The effect of ecological context and relatedness on larval cannibalism in a fungus-associated beetle

Corlett W. Wood,^a Hannah E. Marti,^b and Edmund D. Brodie III^a^aMountain Lake Biological Station, Department of Biology, 485 McCormick Road, University of Virginia, Charlottesville, VA 22904, USA and ^bDepartment of Biology, St. Olaf College, 1520 Street, Olaf Avenue, Northfield, MN 55057, USA

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The fitness consequences of extreme competitive interactions such as cannibalism are often governed by the environment because the costs and benefits of such behaviors depend on the ecological and social context of the participants. However, most studies of context-dependent cannibalism are conducted under extreme circumstances or examine only a single environmental context, conditions that are unlikely to exist in natural populations. In this study, we tested the effect of multiple environmental contexts on the frequency of cannibalism in forked fungus beetle larvae (*Bolitotherus cornutus*), which develop in 1 of 3 different fungus species. We paired larvae in laboratory trials and measured the effect of 1) ecological context (the 3 fungi) and 2) the relatedness of the paired larvae on the frequency of cannibalism. We found a strong effect of ecological context on cannibalism: larvae in 1 fungus cannibalized nearly twice as often as larvae in the other two. We did not detect an effect of relatedness on cannibalism in the 1 species of fungus in which trials were conducted. Cannibalism conferred benefits in the form of accelerated growth rates in all measured traits relative to noncannibals. However, contrary to most studies, cannibalism was most common in the highest quality fungus, contradicting the hypothesis that cannibalism occurs in poor environments to compensate for resource deficiencies. We discuss alternative mechanisms that may drive the ecological context dependence of cannibalism in *B. cornutus* and emphasize the importance of studying context-dependent behavior in naturally occurring environments.

Key words: *Bolitotherus cornutus*, context dependence, inclusive fitness, kin selection, resource environment.

INTRODUCTION

The environment in which a behavior is expressed affects its associated costs and benefits. Consequently, behaviors as diverse as foraging activity (MacArthur and Pianka 1966), reproductive investment (Badyaev and Duckworth 2003), altruism (Hamilton 1964), mate choice (Gasparini et al. 2013), and aggression (Tanner and Adler 2009) depend on the ecological or social context in which they occur. Identifying the axes of environmental variation that drive differences in behavior, as well as the phenotypic and fitness consequences of such variable behaviors, is necessary to understand the forces that govern behavioral evolution in heterogeneous environments.

Cannibalism provides an excellent opportunity to explore the context dependence of behavior. It is taxonomically widespread, pervasive in natural populations even in the absence of stress, and has profound fitness consequences that often depend on the

environment in which it occurs (Fox 1975; Rudolf et al. 2010). The propensity for cannibalism is heritable (reviewed in Polis 1981) and has been shown to respond to selection in laboratory populations (Stevens 1989), suggesting that differences in the frequency of cannibalism among environments are potentially adaptive. The conflicting benefits and costs of cannibalism are well documented. Cannibals often develop faster (Chapman et al. 1999; Via 1999), attain larger body size and increased fecundity at maturity, and experience lower mortality than their noncannibalistic counterparts (Church and Sherratt 1996; Vijendravarma et al. 2013), either as a direct result of the nutritional benefits of cannibalism or as an indirect consequence of reduced competition for otherwise shared resources (Fox 1975). These benefits are countered by the costs of injury due to reciprocal violence or disease transmission and the loss of inclusive fitness if cannibals consume relatives such as offspring or siblings (Polis 1981; Collie et al. 2013).

The ecological context in which cannibalism occurs and the relatedness of interacting individuals both influence the relative magnitude of these costs and benefits. Most research on the effect of ecological context on cannibalism focuses on food-restricted

Address correspondence to C.W. Wood. E-mail: cww9fg@virginia.edu.

circumstances. Cannibalism tends to be more common under resource-deficient conditions (Fox 1975; Polis 1981), in which the caloric benefits of cannibalism outweigh the risks to the cannibal. For example, cannibalistic larvae of the fall armyworm developed faster than noncannibals only when raised in a limited food environment (Chapman et al. 1999). Similarly, Neotropical mosquitos that cannibalized other larvae survived longer than those that did not, but only in low-food conditions (Church and Sherratt 1996). These results accord with experimental demonstrations that the propensity for risky foraging behavior increases when organisms are operating at or below the break-even point of their energy budgets (Caraco et al. 1990; Cartar and Dill 1990). Very little data exist on the response of cannibalism to more complex ecological contexts (e.g., alternate food resources); in the few studies that have examined a realistic range of ecological contexts, it is often difficult to disentangle direct effects on cannibalism from other plastic responses to the same environmental differences. For example, because the diet differences that affect cannibalism propensity in spadefoot toad tadpoles also trigger sweeping morphological changes (Pfennig and Murphy 2000), it is difficult to isolate the effects of food resource on cannibalism alone.

The costs of cannibalistic behavior are also influenced by the relatedness of cannibals to their victims. Inclusive fitness costs can constrain the evolution of cannibalism (Rudolf et al. 2010) because in kin-structured populations, individuals engaging in cannibalism are likely to eliminate a relative (Pfennig 1997). Kin recognition and avoidance mitigate this cost in many species (Parsons et al. 2013). Some exhibit a reduced propensity for cannibalism when paired with relatives (Joseph et al. 1999), whereas others preferentially associate with nonrelatives (Pfennig 1999). A canonical example of the latter is the tadpoles of spadefoot toads (*Spea*), which exhibit diet-induced behavioral and morphological polymorphism. The noncannibalistic omnivores school with siblings, whereas the carnivores, which are more cannibalistic, preferentially associate with nonrelatives, ameliorating the inclusive fitness cost that cannibalism would otherwise incur (Pfennig 1992).

Despite the wide-ranging phenotypic and fitness consequences of context-dependent cannibalism, however, the effects of ecological context and relatedness on cannibalism are rarely explored in the same system under a realistic range of ecological environments. The paucity of data on the effects of multiple contexts on cannibalism constrains our ability to understand its dynamics in natural populations that inhabit heterogeneous environments. If both social and ecological factors independently affect cannibalism, then in combination they may oppose or reinforce each other, resulting in the consistent underestimation of cannibalism risk in some contexts and overestimation in others. This limitation is compounded by the fact that studies of the ecological context dependence of cannibalism have been confined primarily to oversimplified or extreme circumstances (e.g., food limitation), leaving the natural range of developmental environments largely unexplored (but see Laycock et al. 2006). If ecological environments that natural populations experience influence the frequency and consequences of cannibalism, then the ecological and evolutionary consequences of cannibalistic behavior may be more widespread than we currently appreciate.

In this study, we examined the effect of ecological context (food resource) and relatedness on the frequency of larval cannibalism in *Bolitotherus cornutus*, a beetle that spends its entire larval period on 1 of 3 fungus species. Because larvae are confined to a discrete ecological environment—a fungus fruiting body—throughout

development and are unable to disperse, larval competition is likely intense. Moreover, the risk of cannibalizing siblings in the same fruiting body may alter costs incurred by cannibalistic larvae.

We tested whether the fungal host affected the propensity of *B. cornutus* larvae to cannibalize and whether larvae avoided cannibalizing siblings. To assess differences in environmental quality among the fungi, we first reared larvae individually in each of the 3 fungi and measured larval growth. We then paired these lab-reared larvae and scored the frequency of cannibalism 1) in the 3 fungus species and 2) between sibling and nonsibling larval pairs. Finally, to assess the benefits of cannibalism in the 3 fungi, we compared the growth rate of cannibals with noncannibals and measured the risk of noncannibalistic mortality in the 3 fungi. We hypothesized that cannibalism would be most common in the lowest quality fungus and in nonsibling pairs and that growth benefits of cannibalism would be most pronounced in the lowest quality fungus.

METHODS

Study system

Forked fungus beetles (*B. cornutus*; Coleoptera: Tenebrionidae) develop in the fruiting bodies (“brackets”) of 3 wood-decaying fungi: *Fomes fomentarius*, *Ganoderma applanatum*, and *Ganoderma tsugae*. Females lay eggs singly on the surface of the brackets, and the larvae burrow into the brackets and remain inside with other conspecific larvae and pupae until they eclose as adults several months to over a year later (Liles 1956). Preliminary trials under laboratory conditions demonstrated that *B. cornutus* larvae engage in cannibalism (Wood CW, Marti HE, unpublished data). Cannibalism likely occurs only between larvae; because the eggs are laid on the bracket surface, they are protected from foraging larvae, and their hard frass-like covering protects them from foraging adults. Growth effects associated with cannibalism in the larval stage may have broad fitness consequences later in life: in insects, larger females tend to be more fecund (Bonduriansky 2001), and both body size and thoracic horn length positively affect fitness in *B. cornutus* males (Conner 1988; Formica et al. 2011).

The 3 host fungi provide different social and ecological environments for developing larvae in natural populations. Dissection of field-collected brackets indicates that the potential for larval competition differs among the 3 fungi: in brackets that contained at least 1 larva, larval density was higher in *G. applanatum* (10.07 larvae per bracket, which translates to 0.034 ± 0.031 larvae/cm³) than in either *G. tsugae* (3.95 larvae per bracket, or 0.010 ± 0.010 larvae/cm³) or *F. fomentarius* (2.96 larvae per bracket, or 0.023 ± 0.023 larvae/cm³; del Sol J, Wood CW, unpublished data). The fungi appear to be characterized by distinct chemical compositions, as adult beetles collected from *G. applanatum* and *F. fomentarius* can be distinguished by the chemistry of their defensive secretions (Holliday et al. 2009). Moreover, adults discriminate between the fungi in lab-based feeding choice trials (Heatwole and Heatwole 1968).

Ecological environment and larval growth

One hundred and twenty-seven mating pairs of *B. cornutus* were established from collections on and around Salt Pond Mountain in Giles County, VA, during the summers of 2012 and 2013. Adult pairs were maintained on *G. tsugae* in an incubator on 16:8 light:dark cycle at 23 °C. Water and fungus were provided ad libitum. Pairs were checked for eggs 1–2 times per week and all eggs were collected. Collected eggs were randomly assigned to 1 of the

3 fungi, and each egg was transferred to a 1.5-mL tube filled with pulverized fungus of its assigned species. Several brackets of each species were dried, frozen, and ground in an industrial blender (Blendtec, Orem, UT) to ensure resource homogeneity within each fungus species. These eggs were housed in the same conditions as adults, and water was added and mold removed when necessary. Eggs were checked approximately every other day and hatch date was recorded when an empty egg case was found. Approximately 10 days after a larva hatched, it was weighed to the nearest 0.01 mg using an Ax205 DeltaRange balance (Mettler-Toledo, Columbus, OH).

All statistical analyses were performed in R 3.0.1 (R Core Team 2013). We used deviation coding (“contr.sum” in R) for unordered categorical variables. To test for an effect of the fungus environment on larval growth, we compared the mass of 10-day-old larvae reared individually in the 3 fungi. Our model (executed using the function `lmer` in the `lme4` package) included log-transformed larval mass as the dependent variable, fungus as a fixed effect, and family and the family \times fungus interaction as random effects. We log-transformed larval mass because the residuals of untransformed data were nonnormally distributed and violated the assumption of homogeneity of variance. We tested the significance of the fungus effect with a likelihood-ratio test using the `drop1` function. To test for the presence of a significant genotype-by-environment interaction for growth, we used a likelihood-ratio test to compare the full model with a reduced model in which the family \times fungus interaction was omitted.

Ecological environment and cannibalism

To test the effect of the fungus environment on the frequency of cannibalism, larvae from the above experiment were then assigned to 1 of 6 treatment groups. Three of these groups were cannibalism trials, in which each larva was paired with one other unrelated larva (from a different mating pair of adults) that had been reared on the same fungus ($N = 35$ larval pairs for each fungus). The remaining 3 groups were controls to allow us to measure growth benefits of cannibalism and the rate of noncannibalistic mortality. These larvae were housed unpaired on the fungus on which they had been reared ($N = 35$ for *F. fomentarius* and *G. applanatum*; $N = 38$ for *G. tsugae*). The paired larvae in the cannibalism trials were size matched within 1 mg whenever possible because size asymmetries between competitors can determine the outcome of cannibalistic interactions (Petersen et al. 2010). We chose to size match larvae to avoid confounding the effects of size differences and fungus environment on cannibalism frequency. Because the fungi had a strong effect on the mean and variance in larval size (see Results), if we had not size matched the larvae, the average size difference would have been much larger in *G. tsugae* (in which variance in larval size was large) than in *F. fomentarius* (in which variance in larval size was small), rendering it difficult to isolate the influence of the fungus environment on cannibalism.

All trials were housed in clear 1.5-mL tubes with a hole in each lid for ventilation, and water was added to each tube as needed throughout the experiment. Tubes with paired larvae were filled with 0.050–0.070 g of dried ground fungus, lightly packed into 0.5 mL volume. Control tubes were filled with 0.025–0.035 g of dried ground fungus, lightly packed into 0.25 mL volume. Larval densities in the experiment were equal across fungus types and higher than those generally observed in the field to increase the potential for interactions among larvae.

Paired trials were checked daily for 14 days, or until cannibalism occurred, or until both larvae died from other causes. Although the larval stage can last a year in *B. cornutus* (Liles 1956), we chose this experimental duration because most cannibalism occurred in the first 2–3 days in all fungi (see Results), suggesting that the initial encounter was disproportionately responsible for determining whether a pair would cannibalize. We scored a trial as cannibalism whenever we found 1 larva that was either partially or completely consumed. The remains of cannibalized larvae could be distinguished from molted exoskeletons by the presence of soft tissue inside the exoskeleton or head capsule of the cannibalized larva. The severity of cannibalism ranged from a single wound to consumption of nearly the entire larva. Larvae that were found dead but not wounded were scored as noncannibalistic deaths. Control groups were checked daily for the first 3 days to replicate the disturbance to the paired larvae, and after the third day were measured on the same schedule as the larvae that cannibalized their partners (see Cannibalism and Growth Rate in Methods).

We used a generalized linear model (the `lmer` function in R) with a binomial error distribution and logit link to test for differences in the probability of cannibalism among fungi. In this model, trial outcome was the dependent variable and fungus species was an independent variable. To control for any effect of initial larval size or the size difference between paired larvae on the probability of cannibalism, we included the mass difference between larvae and average initial mass of each pair as covariates. We performed a likelihood-ratio chi-square test using the `drop1` function to test for significance of the fungus effect. We used a Kruskal–Wallis rank sum test to test for an effect of the fungus environment on the latency to cannibalism (measured in days) because the number of days until cannibalism occurred was nonnormally distributed. We used Fisher’s Exact tests to test for differences in the frequency of noncannibalistic death among fungi, after excluding the trials in which cannibalism had occurred. We performed these tests separately for paired and control larvae, which were unpaired and raised individually. We tested for differences in the latency to noncannibalistic death among fungi using Kruskal–Wallis rank sum tests.

Relatedness and cannibalism

To test the effect of relatedness on cannibalism, larvae were paired with a sibling (a larva from the same mating pair of adults; $N = 35$ larval pairs). These trials were conducted in *G. tsugae*. The frequency of cannibalism in this group was compared with the frequency of cannibalism in the pairs of unrelated larvae raised in *G. tsugae* from the ecological environment experiment ($N = 35$ pairs; see Ecological Environment and Cannibalism in Methods). Sibling trials were conducted only in *G. tsugae* because logistical constraints prevented us from raising sufficient numbers of larvae in each of the 3 fungi. The sibling pairs may have included some half-siblings because wild-caught females could have previously mated. These trials were conducted under the same experimental protocol as the ecological environment trials described above.

We used a generalized linear model with a binomial error distribution and logit link to test for differences in the probability of cannibalism between sibling and nonsibling pairs; this model was identical to the model used to test for differences among fungi, except that a relatedness fixed effect replaced the fungus fixed effect. We used Kruskal–Wallis rank sum tests to test for differences in the latency to cannibalism between sibling and nonsibling pairs. We used Fisher’s Exact tests to test for differences in the frequency of noncannibalistic death between sibling and nonsibling pairs

(after excluding the trials in which cannibalism had occurred) and tested for differences in the latency to noncannibalistic death using Kruskal–Wallis rank sum tests.

Cannibalism and growth rate

To test for growth benefits of cannibalism, we examined whether cannibals (larvae that consumed one other larva) grew significantly faster than control larvae, which never had the opportunity to cannibalize. This analysis included all paired larvae that cannibalized, including the sibling pairs raised on *G. tsugae*. Control larvae were weighed and an image was taken with a microscope-mounted camera at the start of the experiment and again 7–12 days after they were placed in their control tubes; cannibals were weighed and an image was taken on the day that they cannibalized and again 7–12 days later. The variance in the number of days between initial and final measurements was due to the fact that imaging the larvae was time consuming, so we were unable to process all larvae exactly 10 days after they cannibalized. However, this is unlikely to have affected our results because we controlled for the number of days between the initial and final measurements in our statistical analysis (see below) and because the number of days between initial and final measurements was random with respect to fungus treatment.

We measured 3 traits from the microscope images using ImageJ (Abramoff et al. 2004): larval length, head capsule width, and mandible length. Length was measured from the base of the head capsule to the end of the larva, using a segmented line to incorporate curvature. Head capsule width was measured as the straight-line distance between the antennal insertions, and mandible width was measured as the straight-line distance from the base of the mandible to the tip. Because sclerotized structures like the head capsule and mandibles grow most rapidly following progression from one developmental stage (“instar”) to the next (Daly 1985), accelerated growth in these traits may indicate an effect of cannibalism on the rate of larval development.

We calculated growth rates for the 4 traits (mass, length, head capsule width, and mandible length) for both controls and cannibals by subtracting each larva’s initial measurement from its final measurement and dividing by the number of days separating the 2 measurements. We used Anova to test for an effect of the fungus environment, cannibalism, and their interaction on larval growth and ran a separate model for each of the 4 traits. In these models, growth rate was the dependent variable and fungus, cannibalism, the fungus × cannibalism interaction, and initial size were included as fixed effects. We conducted significance tests for all fixed effects using the Anova function in the car package.

RESULTS

Ecological environment and larval growth

The fungus environment had a significant effect on the mass of 10-day-old larvae (Figure 1; degrees of freedom [df] = 2, likelihood-ratio $\chi^2 = 164.430$, $P < 0.001$). This effect was driven by larvae raised on *F. fomentarius*, which were smaller than those reared on either *G. applanatum* or *G. tsugae*. We found no evidence for a genotype-by-environment interaction for larval growth (family × fungus interaction: df = 1, likelihood-ratio $\chi^2 = 0.321$, $P = 0.571$).

Ecological environment and cannibalism

The probability of cannibalism differed significantly among fungus environments (Figure 2 and Table 1) but was not affected by the

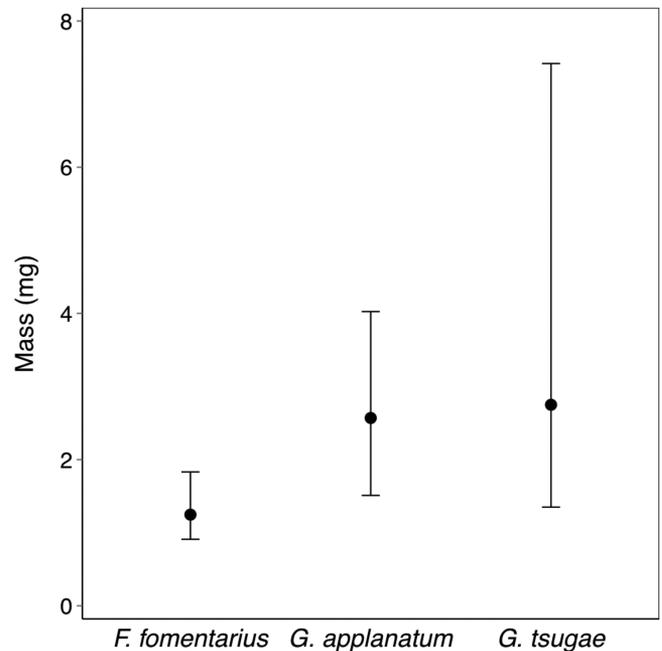


Figure 1

The fungus environment affects the mass of 10-day-old larvae. Error bars are 95% confidence intervals.

average initial size of the paired larvae or by their size difference (Table 1). There was no significant effect of the fungus environment on the number of days until cannibalism occurred although there was a trend toward a longer latency to cannibalism in *G. applanatum* (df = 2, Kruskal–Wallis $\chi^2 = 5.010$, $P = 0.082$). This pattern was driven by larvae in *G. applanatum*, which continued to cannibalize for several days after the start of the experiment, whereas *F. fomentarius* and *G. tsugae* rarely cannibalized after the second day. In all 3 fungi, the highest rates of cannibalism occurred in the first 2 days of the experiment. The fact that most cannibalism took place at the start of the experiment suggests that larvae assess whether to cannibalize the first time they encounter another larva (and that in the small test arenas the time until first encounter was short). The rapid decline in cannibalism after the first few days of the experiment further suggests that a larva that does not cannibalize after the first encounter is unlikely to cannibalize in the future.

The fungus environment affected the frequency of noncannibalistic mortality in the paired trials in which cannibalism did not occur: mortality was high in *G. tsugae* and low in *G. applanatum* (Figure 2; Fisher’s Exact test, $P = 0.029$). Mortality rates in the control larvae were not significantly different among the 3 fungi (Figure 2; Fisher’s Exact test, $P = 0.103$), although more control larvae died in *G. tsugae*, similar to the pattern observed among paired larvae. There was no effect of the fungus environment on latency to death (df = 2, Kruskal–Wallis $\chi^2 = 1.425$, $P = 0.491$).

Relatedness and cannibalism

Relatedness did not affect the probability of cannibalism (Figure 2 and Table 1). The size difference of the paired larvae also had no effect on the probability of cannibalism although the probability of cannibalism increased with the pair’s average initial size (Table 1). There was no difference in the latency to cannibalism between sibling and nonsibling pairs (df = 1, Kruskal–Wallis $\chi^2 = 0.488$,

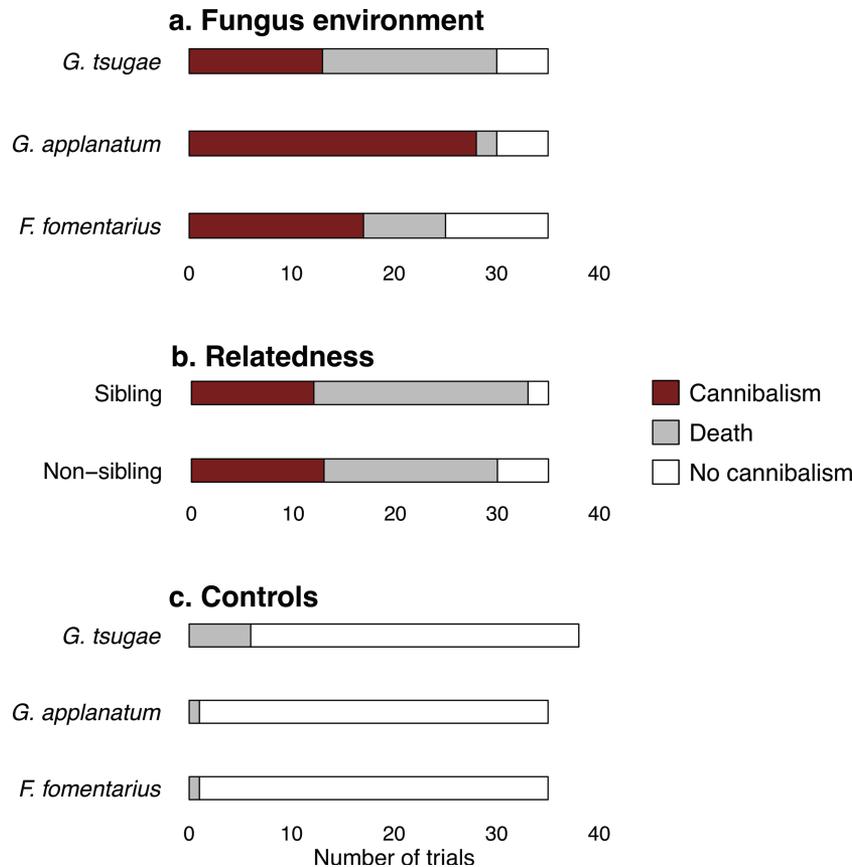


Figure 2

The frequency of cannibalism and noncannibalistic death in different (a) fungus environments, (b) between siblings and nonsiblings, and (c) in the controls. The probability of cannibalism differed significantly among fungi but not between siblings and nonsiblings. There was no significant effect of the fungus environment on mortality in the control larvae.

Table 1

Likelihood-ratio tests for the effect of the fungus environment and relatedness on the probability of cannibalism

Model	Effect	df	Likelihood-ratio χ^2	<i>P</i>
Fungus environment	Fungus	2	15.780	0.001
	Initial size	1	0.349	0.555
	Size difference	1	0.247	0.627
Relatedness	Relatedness	1	0.277	0.782
	Initial size	1	2.092	0.036
	Size difference	1	-0.309	0.757

$P = 0.485$), as would be expected if siblings refrained from cannibalism longer than nonsiblings.

Neither noncannibalistic mortality (Fisher's Exact test, $P = 0.243$) nor the latency to death ($df = 1$, Kruskal-Wallis $\chi^2 = 0.440$, $P = 0.501$) differed between the sibling and nonsibling pairs. Bold text indicates significance ($P < 0.05$).

Cannibalism and growth rate

Cannibals grew at a faster rate than control larvae for all 4 measured traits (Figure 3 and Table 2). Growth rates in all traits were significantly different among fungi, but we found no strong evidence for a cannibalism \times fungus interaction for growth rate (Table 2). The cannibalism \times

fungus interaction was not significant for mass, head capsule width, or mandible length and was only marginally significant for larval length.

DISCUSSION

Cannibalism was strongly influenced by ecological context in *B. cornutus* and conferred measurable benefits in the form of accelerated growth rate in cannibals. Cannibalism was nearly twice as common in *G. applanatum* as in the other 2 fungus species. Contrary to most studies on cannibalism across resource environments (King and Dawson 1972; Polis 1981; Wolcott and Wolcott 1984), the highest rate of cannibalism occurred in the highest quality resource. We did not detect an effect of relatedness on cannibalism in this study, but these trials were conducted only in the fungus in which cannibalism was rare. If the propensity to cannibalize siblings depends on the ecological context, results obtained in the other 2 fungi (where cannibalism was more common) may be very different from those obtained in *G. tsugae*, with complex evolutionary ramifications. Finally, because cannibalism was not universal under any of these experimental conditions—and occurred only in the minority of trials in *F. fomentarius* and *G. tsugae*—there may be unmeasured costs of cannibalism in this system that limit its prevalence.

Our results underscore the importance of incorporating the ecological complexity characteristic of natural environments into experimental studies of context dependence. Implicit in the use of

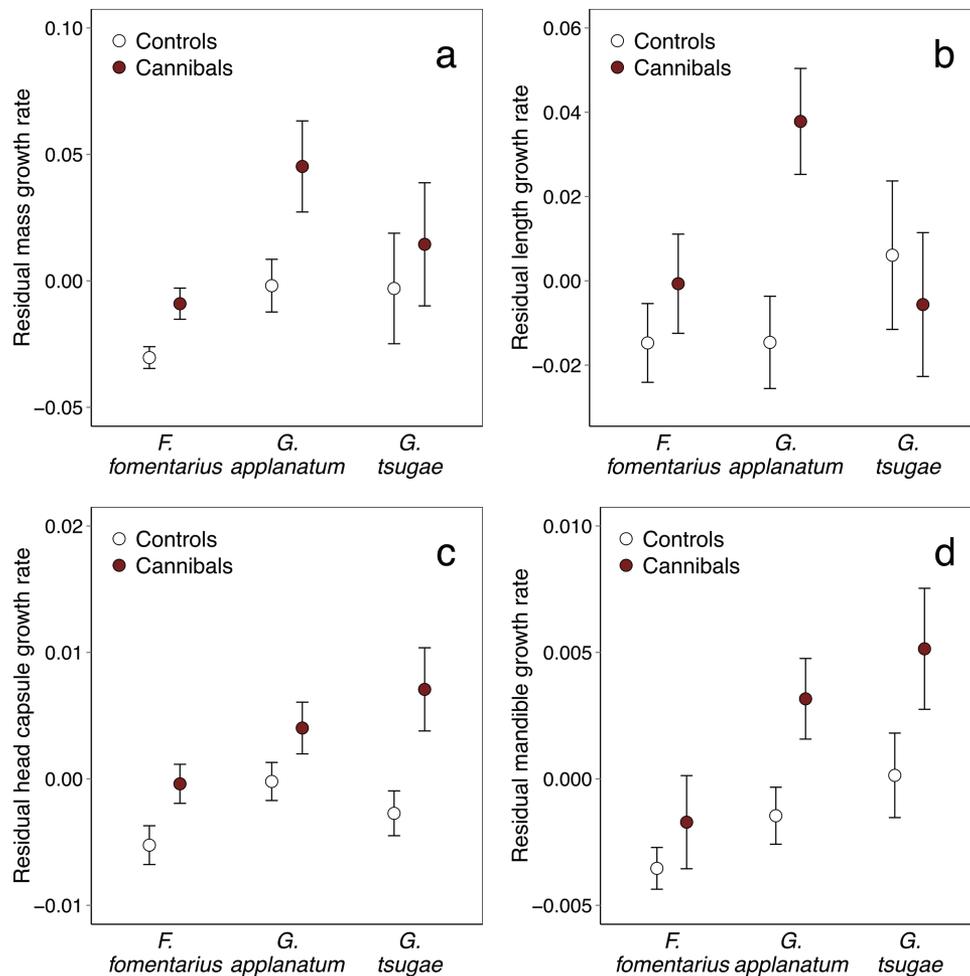


Figure 3

Growth rates in (a) mass, (b) length, (c) head capsule width, and (d) mandible length of control larvae and larvae that cannibalized. The growth rate of cannibals was significantly greater than that of controls for all traits (Table 2). Error bars are standard errors, and values on the y axis are the residuals from the regression of larval growth rate on initial larval size for each trait.

Table 2

Anovas of the effect of the fungus environment, cannibalism, and their interaction on larval growth rate in 4 traits

Trait	Effect	df	F	P
Mass	Cannibalism	1	6.538	0.012
	Fungus	2	4.925	0.008
	Fungus \times cannibalism	2	0.555	0.575
Length	Initial mass	1	0.805	0.371
	Cannibalism	1	5.002	0.027
	Fungus	2	3.109	0.048
	Fungus \times cannibalism	2	3.062	0.050
Head capsule width	Initial length	1	2.316	0.130
	Cannibalism	1	25.116	<0.001
	Fungus	2	15.603	<0.001
	Fungus \times cannibalism	2	2.450	0.090
Mandible length	Initial head capsule width	1	20.409	<0.001
	Cannibalism	1	17.030	<0.001
	Fungus	2	18.637	<0.001
	Fungus \times cannibalism	2	1.698	0.187
	Initial mandible length	1	37.342	<0.001

Bold text indicates significance ($P < 0.05$).

realistic ecological treatments is the recognition that many variables may simultaneously contribute to context dependence of behavior. Because natural environments differ in many dimensions, focusing

on the effect of a single isolated variable (e.g., calorie content) has the potential to lead to inaccurate predictions when extrapolated to natural populations. Therefore, comparing complex ecological environments is crucial to an understanding of the dynamics of context-dependent behaviors like cannibalism in heterogeneous environments.

Ecological environment and larval growth

The 3 fungi appear to be substantially different in quality (Figure 1). The effect of the fungus environment on mass is strong—already pronounced in 10-day-old larvae—and complex. *Fomes fomentarius* is clearly the lowest quality environment. The average mass of 10-day-old larvae reared in the 2 *Ganoderma* species was similar, but variance in both mass and growth rate was higher in *G. tsugae* than in *G. applanatum* (Figures 1 and 3). It is unlikely that this amplified variance reflects higher resource heterogeneity in *G. tsugae* because the fungus was blended and homogenized prior to the experiment to minimize within-fungus species variation. Instead, *B. cornutus* larvae may vary in their ability to metabolize *G. tsugae* to a larger degree than the other 2 fungal hosts. We found no evidence for a genotype-by-environment interaction for larval size; all families performed poorly on *F. fomentarius*.

These observed differences in environmental “quality” likely encompass 3 distinct types of food stress: food limitation (i.e.,

caloric restriction), nutrient limitation (i.e., shortage of an essential nutrient), and food toxicity (i.e., the abundance of defensive compounds). Variance in the abundance of defensive compounds may in fact drive the strong effect of the fungus environment on larval growth in *B. cornutus*, rather than caloric or nutrient content differences among the 3 fungi. Wood-decaying bracket fungi are well defended by chemical volatiles (Jonsell and Nordlander 2004), and negative effects of these compounds may inhibit larval growth in *F. fomentarius* or contribute to the high variance observed in *G. tsugae*. Furthermore, differences in nutritional quality may reflect differences in the invertebrate prey communities within each fungus species, which would have been homogenized along with the brackets in our experiment. The abundance of other invertebrate species appeared to be especially low in *F. fomentarius* relative to the other species in a sample of field-collected brackets (del Sol J, Wood CW, unpublished data).

Ecological environment and cannibalism

Although we documented strong differences in resource quality among the ecological environments, and a large effect of the ecological environment on the frequency of cannibalism, no clear relationship existed between the quality of the ecological environment and the frequency of cannibalism. Cannibalism was twice as common in *G. applanatum* as the other 2 species, contradicting our original hypothesis that the lowest quality environment—*F. fomentarius*—would induce the highest rate of cannibalism. Cannibalism conferred large benefits in the form of accelerated growth rates in all traits, but this effect was not significantly different among environments, contrary to our hypothesis that cannibalism would confer the greatest benefit in the lowest quality environment. This pattern of context dependence may function to compound, rather than minimize, phenotypic differences between individuals developing in low- and high-quality environments.

Three alternative hypotheses may explain why the pattern we observed was opposite to the pattern seen in most other studies (King and Dawson 1972; Polis 1981; Wolcott and Wolcott 1984). First, the high rate of cannibalism in the “best” environment may reflect the ability of individuals in high-quality environments to maintain high activity or aggression levels. If individuals across all environments are equally likely to cannibalize when they encounter another larva, but food- or nutrient-limited individuals are less active, encounter rates—and, as a result, cannibalism—will be elevated in high-quality environments. This pattern has been documented in other cannibalistic systems (Mayntz and Toft 2006). However, this hypothesis does not account for differences in the rate of cannibalism in the 2 *Ganoderma* species, in which larval growth rates were similar.

Second, the deficiencies in the low-quality environment (*F. fomentarius*) in our study may be indicative of a type of food stress that cannot be mitigated through cannibalism. Naturally occurring food resources such as fungi likely differ in multiple dimensions, including calorie content, the availability of essential nutrients, and the abundance of defensive compounds. As a result, it is unsurprising that the pattern of context-dependent cannibalism documented in this study is more complex than that in studies confined to simplified or extreme circumstances that only incorporate a single dimension of food stress (e.g., starvation). If cannibalism does not directly compensate for the deficiencies of a low-quality ecological environment, then in these circumstances, the costs of cannibalism may outweigh its benefits and it should remain infrequent. For example, if the observed differences in larval growth in the

3 fungal environments are due to defensive compounds and not nutrient limitation, then the nutrients gained from cannibalism may not confer any benefit in poor environments like *F. fomentarius*. That the growth benefits of cannibalism were not larger in *F. fomentarius* than in the 2 *Ganoderma* species suggests that whatever benefits are obtained from cannibalism are not directly ameliorating the main source of stress in that environment. However, this hypothesis fails to address the difference in cannibalism frequencies in the 2 *Ganoderma* environments.

Third, the rate of cannibalism may be an indirect consequence of the resource environment, responding to an environmental variable that covaries with it in wild populations. One such variable that may underlie the pattern reported here is conspecific density, which triggers cannibalism in other taxa (Fox 1975), and is one aspect of the social environment that distinguishes *G. applanatum* from the other 2 fungi. Field observations show that the density of *B. cornutus* eggs (Wice E, unpublished data) and larvae (del Sol J, unpublished data) is highest on *G. applanatum* brackets. If these observations reflect consistent differences in density among the 3 environments, *G. applanatum* may trigger cannibalism in developing larvae as a strategy to eliminate abundant competitors. However, because density did not differ among the 3 fungal environments in our experiment, this hypothesis requires that larvae respond to a cue in the fungus itself by facultatively increasing their propensity for cannibalism in *G. applanatum*, rather than responding to direct indicators of larval density (e.g., vibrational cues).

Finally, it is important to note that the effect of fungus environment on the frequency of cannibalism is not independent of other sources of mortality that vary among fungi (Figure 2). Noncannibalistic mortality was not attributable to physical injury because we scored all wounded larvae as cannibalism, nor is variation in noncannibalistic mortality among environments an artifact of size matching the larvae, which would have affected all environments equally. Instead, differences among fungus environments in the relative frequency of cannibalistic and noncannibalistic mortality may reflect context-dependent responses to stress imposed by conspecifics. That is to say, in some environments (e.g., *G. applanatum*), larvae respond to elevated stress by attacking and cannibalizing conspecifics, whereas in others (e.g., *G. tsugae*), larvae are equally stressed—and often die as a result—but do not respond by cannibalizing their competitor.

Relatedness and cannibalism

Cannibalistic *B. cornutus* larvae displayed no kin-avoidance behavior, a surprising result given that kin competition tends to be most common when juveniles are confined to discrete, ephemeral resources (Resetarits 1996). Several explanations may account for this result. First, kin-avoidance behavior may only manifest when the risk of cannibalism is high. Our experiment assessed the effect of relatedness on cannibalism in an environment in which cannibalism turned out to be rare (*G. tsugae*). If the effect of relatedness on cannibalism depends on frequency of cannibalism in a given ecological environment—for example, if larvae avoid relatives only when cannibalism risk is very high—then the dynamics of cannibalism in natural populations, in which larvae may encounter siblings in all fungus species, may be significantly more complex.

Second, larval *B. cornutus* may not be capable of kin recognition, in which case cannibals would be unable to discriminate between related and unrelated larvae. Instead, kin avoidance may occur at the maternal level during oviposition site selection. Because *B. cornutus* females lay eggs singly rather than in clutches, females may

spatially separate offspring to minimize kin competition. This pattern could reduce selection for kin recognition in the larval stage because larvae would not likely encounter relatives during development.

Third, the benefits of cannibalism may outweigh the costs of consuming a sibling. We documented strong benefits of cannibalism in *B. cornutus* larvae. The rate of noncannibalistic mortality was significantly higher in the paired trials than in the controls (Figure 2), suggesting that high larval densities are stressful. Cannibalism may be a mechanism to alleviate that stress, counteracting the inclusive fitness cost of consuming a relative (Collie et al. 2013). Cannibals experienced accelerated growth (Figure 3), which is likely doubly beneficial due to direct advantages of larger body size and decreased vulnerability to conspecific predation. It is unlikely that growth rate differences reflect differences in food availability because the control larvae, which were raised in less food than the pairs, did not exhaust available food by the end of the experiment. Accelerated head capsule and mandible (traits that grow primarily between instars) growth in cannibals indicates that cannibalism also shortens larval development time. Such elevated growth rates allow larvae to quickly escape the smallest size classes, when they are likely most vulnerable to cannibalism from larger larvae. This may be especially beneficial in *B. cornutus* because larvae of many age and size classes are often found in the same bracket (Wood C, unpublished data). Taken together, these data suggest that strong benefits of cannibalism could offset the inclusive fitness cost of eliminating a sibling.

Consequences of context-dependent cannibalism in heterogeneous environments

The benefits of cannibalism that we detected in the form of accelerated growth rates, in combination with differences in the frequency of cannibalism among fungi, may have broad ramifications in heterogeneous environments. In *B. cornutus* and other species in which fecundity and mating success depend on body size (Conner 1988; Bonduriansky 2001; Formica et al. 2011), juvenile cannibalism will increase adult fitness if the morphological effects of cannibalism persist into adulthood. When, as in the present study, high-quality environments induce cannibalism that increases growth, this combination should exaggerate the morphological differences between individuals developing in low- and high-quality environments.

As a result, the evolutionary implications of context-dependent juvenile behavior may extend to traits expressed at other life stages. *Bolitotherus cornutus* adults from different environments likely compete directly with each other for mates due to frequent migration among fungi (Wood et al. 2013). Because males possess elaborate sexually selected horns, a class of traits that are used in competition with conspecifics and exhibit heightened nutrient sensitivity (Emlen 1994; Bonduriansky 2007; Emlen et al. 2012), cannibalism expressed in juveniles could exaggerate asymmetries in competitive interactions between adults. Similarly, variation in the risk of cannibalistic mortality, along with differences in fungus quality, may generate selection on female oviposition site choice because females that choose environments that maximize offspring performance and minimize risk will have higher fitness than those that do not (Wolf et al. 1998, 1999; Refsnider and Janzen 2010; Buser et al. 2013).

In this study, we have shown that naturally occurring environmental contexts can influence the expression of extreme competitive behaviors like cannibalism. Furthermore, our data contradict

the results of studies conducted in simplistic ecological conditions, which have found that cannibalism is most common in nutrient-poor environments (Fox 1975; Polis 1981). The incorporation of natural environmental variation into studies of the context-dependent behaviors may be necessary in order to extrapolate experimental inferences to the dynamics of context-dependent behaviors in wild populations.

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