

Trematode infection correlates with shell shape and defence morphology in a freshwater snail

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SUMMARY

Parasitism often influences the phenotype of individuals. Many of the resulting changes are due to changes in resource allocation that come with infection. Here we examine the effect of a trematode parasite on the shape and defence morphology of a New Zealand freshwater snail, *Potamopyrgus antipodarum*. The trematode *Microphallus* sp. asexually produces hundreds of metacercarial cysts in the snail. The length, width and 2-dimensional area of each snail were measured. Snails were also assessed for their degree of spininess. Snails were dissected to determine gender, brooding condition and parasitism. Snails infected with *Microphallus* sp. were found to be significantly less spiny than uninfected snails. *Microphallus*-infected snails were also found to have a significantly greater width to length ratio at larger sizes than their uninfected counterparts. These trends could be explained in at least 3 ways. (1) Infection causes the snails to not produce spines and to become wider. (2) Spiny and narrower snails are more likely to die when they become infected. (3) Spiny and narrower snails are more resistant to infection. The changes in phenotype observed are unlikely to be adaptive for either the host or parasite and probably represent physiological by-products of the host-parasite relationship.

Key words: *Potamopyrgus antipodarum*, *Microphallus*, trematode, shell morphology, spines.

INTRODUCTION

Infection by parasites has been shown to influence many aspects of host phenotype including physiology (Thompson, 1990), life-history (Minchella, 1985), behaviour (Moore, 2002), sexually selected traits (Zuk, 1992), and morphology (McCarthy, Fitzpatrick & Irwin, 2004). These effects can change the mean value of a trait, the variance, or both (Poulin & Thomas, 1999). Relatively little work has been done to examine the effect of parasitism on the morphology of molluscs (but see Krist, 1998; McCarthy *et al.* 2004). Changes in morphology can include changes in shell size, shell shape or shell ornamentation.

Most studies examining the effect of parasitism on shell morphology in molluscs have studied the growth rate. Outcomes vary from no effect of parasitism on growth rate (Fernandez & Esch, 1991), to decreased growth rates (Sousa, 1983; Crews & Yoshing, 1989; Krist & Lively, 1998) to increased growth rates (Rothschild & Rothschild, 1939; Mouritsen & Jensen, 1994; Ballabeni, 1995; Krist, 2000), which is often referred to as gigantism. Gigantism has been interpreted in several ways: (1) as a by-product of the parasitic castration where hosts that no longer invest energy in reproduction divert at least some of this energy into increasing the growth rate, (2) as an adaptation of the host to outlast

the parasite (Minchella, 1985), and (3) as an adaptive manipulative effort of the parasite to increase space within the host for parasite growth and reproduction (McCarthy *et al.* 2004). It is also possible that parasitism may affect the shape by altering the rates of growth in one dimension but not another. A change in growth rate in length but not width, or *vice versa*, would change the shape and potentially the volume of the host individual. Such changes have been interpreted as adaptive when the volume of the host increased with parasitism and was related to an increase in parasite reproduction (McCarthy *et al.* 2004).

Parasites may also influence shell ornamentation. Many gastropods produce spiny extensions of the shell as defence or stabilizing structures (Vermeij, 1993). The construction of shell material in molluscs is energetically costly (Brusca & Brusca, 2003). The energetic drain caused by parasitism may leave little energy available for the production of spines.

Here we examine the effect of castrating trematode parasites on two aspects of the morphology of the New Zealand mud snail, *Potamopyrgus antipodarum*. *Potamopyrgus antipodarum* is a small Hydrobiid snail commonly found in New Zealand lakes and streams where it grazes on algae and detritus. It is an intermediate host to a suite of castrating trematode parasites. In Lake Alexandrina on the South Island, the most common parasite by far is *Microphallus* sp., which ranges in infection rate from about 3% to over 50% depending upon time and location within the lake (Jokela & Lively, 1995*a*; Levri unpublished

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data). *Microphallus* has a 2-host life-cycle. Adult worms reproduce sexually in the guts of waterfowl. Eggs are passed out with the faeces and consumed by *Potamopyrgus*. The eggs eventually produce hundreds of metacercarial cysts castrating the snail host. The life-cycle is completed when infected snails are eaten by waterfowl.

Microphallus has been demonstrated to influence the behaviour (Levri & Lively, 1996; Levri, 1998*a,b*; Levri, 1999; Levri & Fisher, 2000) and life-history (Lively, 1987; Jokela & Lively, 1995*b*; Krist & Lively, 1998) of this snail species. The snail varies in maximum length by geographical location (Winterbourn, 1970), and even by depth within a lake (Jokela & Lively, 1995*a*). Populations vary considerably in spine production, both in frequency of individuals with spines and the size of the spines (Winterbourn, 1970). Both environmental and genetic influences have been implicated in determining the degree of spininess in individuals (Winterbourn, 1970). In the shallow waters of Lake Alexandrina spine production is modest compared to other lakes, as the majority of snails do not produce spines or ridges.

We assessed the effect of *Microphallus* on the snail's shape (e.g. length to width ratio) and defence morphology (spine production). *Potamopyrgus* produces spines primarily composed of the periostracum layer of the shell, which is largely protein and thus would be energetically costly to produce (Winterbourn, 1970). The snail also shows substantial variation in shape between locations (Winterbourn, 1970). This variation in shape leads to differences in overall shell volume at a given length. Asexual reproduction by *Microphallus* within the snail results in hundreds of metacercarial cysts. Theoretically, this reproduction may be limited by shell volume, which has been found in other microphallid-infected gastropods (McCarthy *et al.* 2004).

MATERIALS AND METHODS

Snails were collected from shallow-water habitats (<1 m) in Lake Alexandrina, South Island, New Zealand using dip nets in December of 2002. The snails were preserved in 70% ethanol and returned to the lab for measurement and dissection. Prior to dissection a digital photograph was taken of each snail using a SPOT Insight Digital camera. For the photograph, each snail was oriented in the same way. All were placed with the shell opening facing up. Each snail was measured using SPOT Insight software. Three measurements were taken for each snail (Fig. 1): length from one end to the other, the width of the most recently grown whorl (whorl 1), and the 2-dimensional area of the entire snail from the digital image.

Each snail was then assessed for the degree of spininess. We subjectively categorized each snail

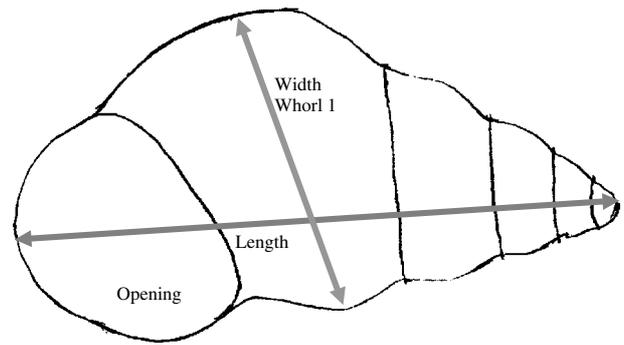


Fig. 1. Drawing of a snail showing the various measurements taken to examine shape. The area of the snail was measured by tracing the outline of the entire 2-dimensional image.

prior to dissection using a scale from 0 to 3 (Fig. 2). A score of 0 indicated no spines or ridges. A score of 1 was given to snails with a ridge, a 2 was given to snails with short spines, and a 3 was given to snails with long spines. The spininess score was given based on assessment of the most recently grown whorl of the snail (whorl 1 in Fig. 1). This was done because, in some snails, the spininess changed as the snail grew.

Each snail was then dissected and gender, brooding condition, infection and type of parasite was noted for each snail. Parasites were identified in part using the descriptions provided by Winterbourn (1974). The snails were separated into 5 mutually exclusive classes including uninfected non-brooding females, uninfected brooding females, uninfected males, *Microphallus*-infected snails, and snails infected with parasites other than *Microphallus*. Snails infected with *Microphallus* and other parasites were rare and were eliminated from the study.

The effect of parasitism on shell shape was analysed using ANCOVA with the length of the snail used as a covariate, width of the first whorl and the square root of the area of the snail used as dependent variables, snail class as an independent variable, and a Type I sum of squares. The various classes of snails were compared in a pair-wise manner. A Levene's test for homogeneity of variance test was performed for each comparison to test for violations of this assumption of ANCOVA. Each pair of classes was first compared by testing for significant differences in the slopes of the regression lines produced by each class. This was done by examination of the interaction between snail class and length. A significantly different slope indicates a significant difference in shapes between the two classes. If there was no significant difference in slopes between the two classes (or the test yielded marginally significant results [$0.01 < P < 0.10$]), a second test was performed without using the snail class by length interaction term in the model. Here a significant effect of class would indicate a significant vertical shift in the regression

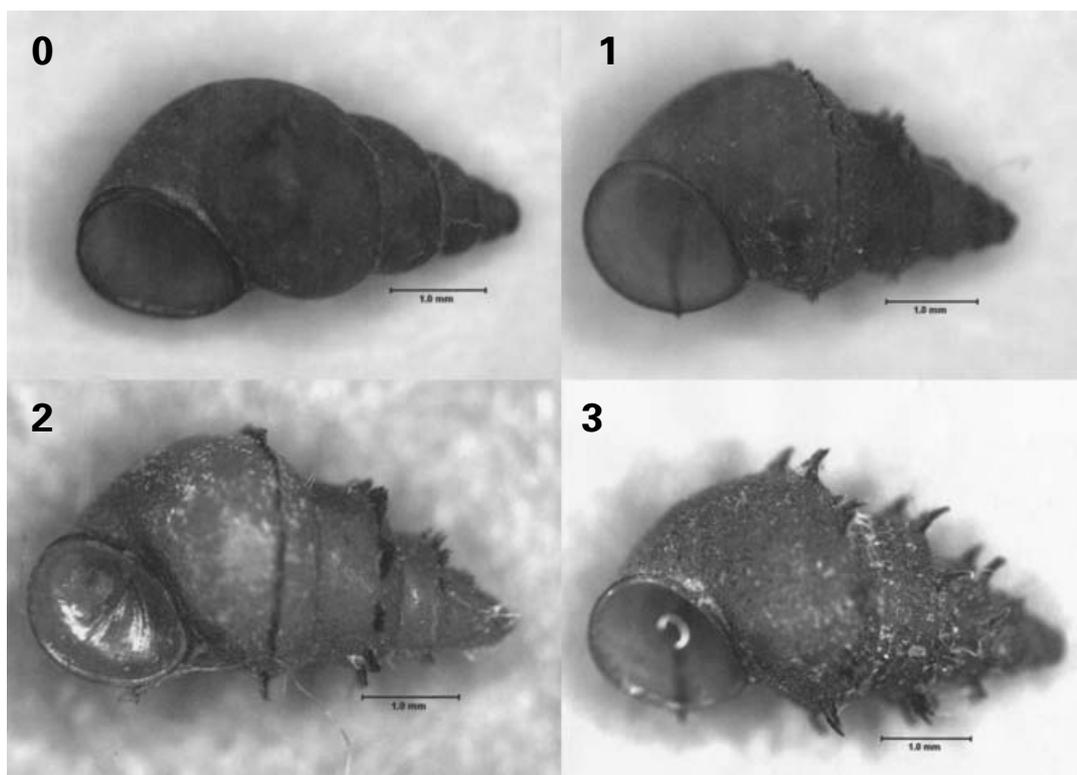


Fig. 2. Photographs of typical snails representing each of the four spininess categories.

lines between the two classes and would also indicate a significant difference in shape between the two groups.

The effect of parasitism on spininess was examined by using log-linear analyses comparing the proportion of spiny individuals in each snail class using pair-wise comparisons. In each class of snails, snails between 3 and 3.9 mm in length were compared to snails greater than 3.9 mm in length to determine if size and/or age influenced defence structures. Since this population of snails has relatively few snails with long spines (score of 3), snails with scores of 2 and 3 were grouped together.

RESULTS

The effect of parasitism on shape

A total of 375 snails were measured and dissected for this part of the study, of which 96 were infected by *Microphallus*, and 15 were infected by other castrating trematodes. These other infections included *Telogaster opistorchis*, *Furcouscercaria*, *Gymnocephalous*, and one or two undescribed species of monostomes (listed in Winterbourn, 1974). An ANCOVA with length as a covariate and the width of the first (most recently grown) whorl as an independent variable was used to compare various groups of snails. Since it is known that the probability of infection increases with age (length) (Jokela

& Lively, 1995a), we were concerned that if the relationship between length and width (or square root of the area) was not precisely linear, then differences between groups may be an artifact of differences in length distribution. Thus, the statistical analyses were run under two different conditions. First, only snails greater than 3.0 mm for males and snails greater than 3.7 mm for females (females are on average larger than males) were used in this analysis. Then all snails of all lengths were used in the analysis. Since the results did not differ between the two analyses in every case but one, we present most of the data using all lengths of snails. The only exception is in comparing uninfected non-brooding female snails to uninfected brooding female snails. In this case, the analysis using snails of all sizes did not meet the assumption of equality of variances, but using sizes greater than 3.0 mm and less than 4.5 mm did meet the assumption. Detailed results from the statistical analysis of shape can be found in the Appendix.

Uninfected brooding females were found to be significantly longer than uninfected non-brooding females ($P < 0.001$), uninfected females were found to be significantly longer than uninfected males ($P < 0.001$), and infected snails were found to be significantly longer than uninfected snails ($P < 0.001$). No difference in shape was found between brooding and non-brooding females (see Tables 1 and 2) thus brooding and non-brooding females were grouped together in subsequent analyses. Uninfected

Table 1. Results of ANCOVA using length as a covariate and width of whorl 1 as an independent variable

Classes compared	R ²	Slope		Vertical shift	
		F	P	F	P
Uninfected non-brooding females <i>vs</i> Uninfected brooding females	0.58 0.54	0.010	=0.92	2.353	=0.13
Uninfected females <i>vs</i> Uninfected males	0.79 0.67	1.779	=0.18	33.536	<0.001
Uninfected females <i>vs</i> <i>Microphallus</i> -infected females	0.79 0.76	6.775	=0.01	344.8	<0.001
Uninfected males <i>vs</i> <i>Microphallus</i> -infected males	0.67 0.82	0.577	=0.45	0.461	=0.503
Uninfected females <i>vs</i> Female snails infected with other parasites	0.79 0.89	4.179	=0.42	0.082	=0.775
<i>Microphallus</i> -infected females <i>vs</i> Female snails infected with other parasites	0.76 0.89	0.714	=0.40	0.994	=0.321

Table 2. Results of ANCOVA using length as a covariate and the square root of the 2-dimensional area as an independent variable

(Vertical shift was not tested for uninfected females compared to *Microphallus*-infected snails because of the strongly significant differences in slopes.)

Classes compared	R ²	Slope		Vertical shift	
		F	P	F	P
Uninfected non-brooding females <i>vs</i> Uninfected brooding females	0.80 0.79	0.575	=0.45	0.220	=0.64
Uninfected females <i>vs</i> Uninfected males	0.93 0.92	1.625	=0.20	14.550	<0.001
Uninfected females <i>vs</i> <i>Microphallus</i> -infected females	0.93 0.92	15.51	<0.001		
Uninfected males <i>vs</i> <i>Microphallus</i> -infected males	0.93 0.96	0.052	=0.82	1.216	=0.28
Uninfected females <i>vs</i> Female snails infected with other parasites	0.92 0.92	1.904	=0.17	0.389	=0.53
<i>Microphallus</i> -infected females <i>vs</i> Female snails infected with other parasites	0.92 0.92	0.056	=0.81	1.987	=0.162

female snails were found to have a significantly greater width per unit length than uninfected males (Table 1). Because of this, infected and uninfected males and females were not grouped in subsequent analyses. *Microphallus*-infected female snails were found to have significantly different slopes from uninfected female snails (Tables 1 and 2). On average, infected snails had a greater width to length ratio as length increased (Fig. 3A). *Microphallus*-infected males were not significantly different from uninfected males (Tables 1 and 2). However, there was a very low sample size of infected males (7). Low sample sizes of snails infected by other castrating trematodes limited our ability to detect differences between them and other classes. However, the regression line of snails infected with other parasites was very similar to *Microphallus*-infected snails (Fig. 3B). An analysis with length as the covariate and the square-root of the 2-dimensional area yielded similar results to the analysis utilizing the width of the first whorl (Table 2).

The effect of parasitism on defence morphology

A total of 2574 snails were scored and dissected for this part of the study, of which 1129 were infected by *Microphallus*, and 154 were infected by other castrating trematodes.

No effect of length was found when comparing large to small individuals in any snail class ($P > 0.33$ in all cases). Thus, large and small individuals were grouped together for subsequent analyses. The uninfected classes (non-brooding females, brooding females, and males) showed no differences in their proportion of spiny individuals (at least $P > 0.24$ in all cases). Significant differences were found between all infected snails and all uninfected snails (Table 3; Fig. 4), as well as between *Microphallus*-infected snails and all uninfected snail classes ($P < 0.01$ in all cases). In general, there were fewer spiny individuals found in the infected classes. No difference was found between *Microphallus*-infected snails and snails infected with other castrating trematodes, but

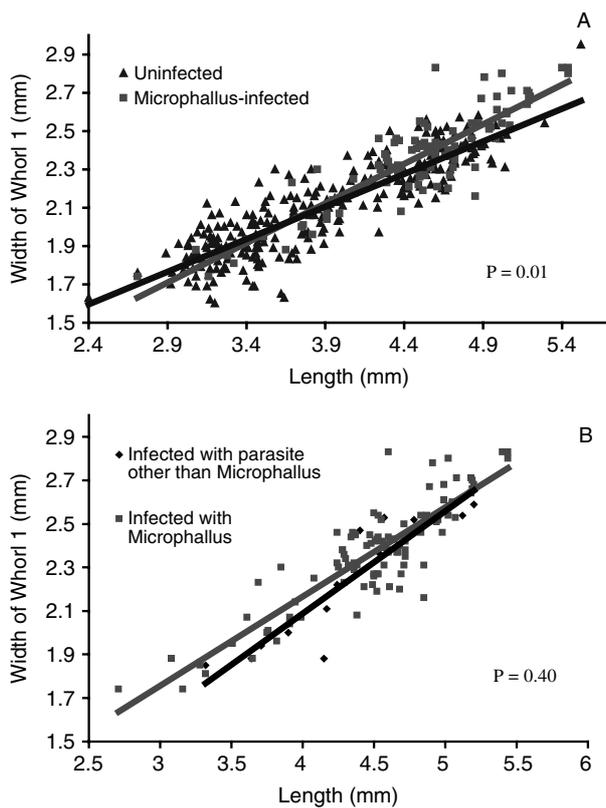


Fig. 3. The relationship between length and width of whorl 1 in *Microphallus*-infected and uninfected snails (A) and *Microphallus*-infected snails and snails that are infected with parasites other than *Microphallus* (B). *Microphallus*-infected snails show a significantly different shape than uninfected snails. *P*-values on the graphs represent the tests performed to determine differences in slopes.

no difference was also found between snails infected with trematodes other than *Microphallus* and uninfected snails. The sample size of snails infected with parasites other than *Microphallus* was limited here, however.

DISCUSSION

Snail classes could have different shapes in two possible ways. First, the slopes of the regression lines of the classes could be significantly different. This was the case for snails infected with *Microphallus* compared to uninfected females. Here the slope of the line for *Microphallus*-infected snails was significantly greater than the slope of uninfected females, indicating greater width to length and square root area to length ratios. This result is probably best explained by the fact that infected snails were once uninfected. The probability of infection increases with age and older (longer) snails are more likely to have been infected for a longer time than younger (shorter) snails. Thus the smaller infected snails used in the analysis were likely only recently infected. At small sizes there was little difference between

infected and uninfected snails due to the lack of time for the parasite to have any pronounced effect on growth. However, for larger snails, a significant proportion of snails were infected for a longer period of time, and the difference between infected and uninfected snails is more pronounced. The regression lines of *Microphallus*-infected snails and uninfected females intersect at about 3.7 mm in length. Detectable *Microphallus*-infection does not usually become common in snails until a length of about 4.0 mm.

If the slopes of the regression lines were not significantly different, an ANCOVA was performed to test for a vertical shift in the regression lines. Such a shift would indicate a significantly greater width to length ratio (or square root area to length ratio) at all lengths. Such a difference was found between uninfected males and uninfected females. Since some of the differences in slopes were marginally significant, the ANCOVA was performed as a precaution in case the slope differences were simply due to chance.

Here we show that *Potamopyrgus antipodarum* infected with *Microphallus* are shaped differently from and are less likely to produce spines than uninfected snails. These results could be explained in at least three ways. (1) The infection influences the growth pattern and spine production in the snails. (2) Wider and smoother snails are more likely to become infected. The probability of this is reasonable in this system because the snails in Lake Alexandrina (as well as other lakes) live in a mixed population of sexual and asexual individuals, and the frequency of clones varies over time. Some clones tend to be spinier than others, and some clones have shown greater abilities than others to resist infection (Lively & Dybdahl, 2000; Lively, personal communication). (3) Parasitism results in differential mortality due to higher parasite-induced death rates in narrower and spinier snails. Spiny snails are allocating more energy to spine production than smooth snails. When infected, the parasite usurps a certain amount of resources from the snail, and if the snails make spines as well, there may not be enough resources remaining to sustain the snail, thus increasing mortality. With regard to shape, if a certain number of metacercariae are always produced, that number in a wider snail may be able to be sustained, while in a narrow snail that number of metacercariae may stress the snail to a greater degree, increasing mortality. This seems unlikely from an evolutionary standpoint however. It would make more sense for the parasite to adjust the number of metacercariae based on the size of the snail. To produce too many may result in the death of the parasite as well. Although, if a certain number of metacercariae are required to reach a critical threshold population size in the waterfowl gut, selection may act to maintain a high number of metacercariae despite the mortality cost to the snail and parasite.

Table 3. Results of pair-wise log-linear analyses comparing proportion of spiny individuals between snail classes

Classes compared	D.F.	Chi square	P
All uninfected vs All infected	2	31.06	<0.0001
All uninfected vs <i>Microphallus</i> -infected	2	32.98	<0.0001
All uninfected vs Other parasite infected	2	1.52	=0.4676
<i>Microphallus</i> -inf. vs Other parasite inf.	2	2.40	=0.3018

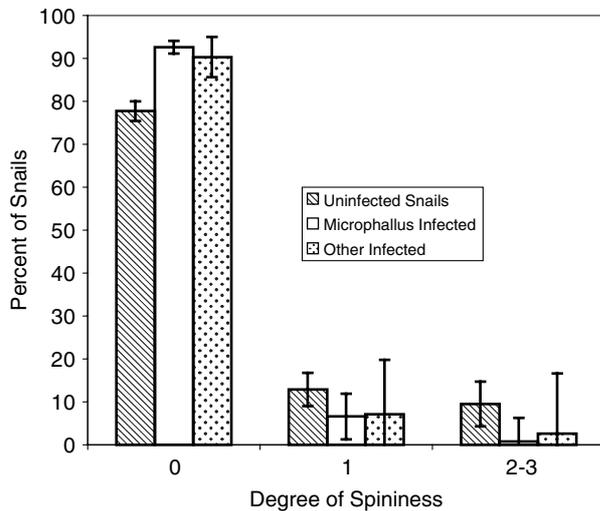


Fig. 4. The proportion of uninfected, *Microphallus*-infected, and snails infected with other parasites within each spininess class. *Microphallus*-infected snails are significantly less spiny than uninfected snails.

Since this study was performed on snails captured and preserved in the field, we cannot differentiate between these alternatives here. Anecdotally, however, with regard to spine production, we noticed that infected snails often had a smooth first (most recently produced) whorl, but would have spiny second or third whorls. This suggests that spininess changed since the time of infection in these individuals. Unfortunately, this was not quantified during this study. We plan future studies to quantify the change in spininess within snails as they age and future experiments utilizing experimental infections to distinguish among the above three hypotheses.

If infection does cause changes in shape and defence morphology in individuals, then these changes could result in reduced fitness consequences for the snails. Spines in this system are hypothesized to reduce the probability of being eaten by predatory fish. The most common fish in Lake Alexandrina, the common bully (*Gobiomorphus cotidianus*), has been shown to eat *Potamopyrgus* in relatively large numbers (Levri, 1998a). *Gobiomorphus* is gape limited and has been shown to consume only snails that are as long or smaller than the width of its mouth (Levri, 1998a). Thus an increase in width of the snail caused by spines or ridges may decrease the probability of predation by this fish. Infection resulting

in reduced spininess may increase the mortality rate of the snails due to predation and also increase the mortality rate of the parasite in the process. Changes in shell shape can influence fitness by changing the ability of the shell to withstand the force of crushing predators and thus increase the likelihood of surviving an attack (Appleton & Palmer, 1988; Krist, 1998, 2002).

In this study, uninfected females were found to be significantly longer than uninfected males, uninfected brooding females were found to be significantly longer than uninfected non-brooding females, and infected snails were found to be significantly longer than uninfected snails. All of these results are consistent with the results of previous studies utilizing this system (Jokela & Lively, 1995b; Levri & Lively, 1996). Since females brood their offspring in a brood chamber, there is likely to be a reproductive advantage for larger females. Larger females have on average larger broods (Levri, unpublished data). As females get larger, a greater proportion of them can be found brooding, thus increasing the mean length of brooding females compared to non-brooding females (Levri, unpublished data). The reason for a greater length of infected snails is less straightforward. Infected snails grow at a slower rate than uninfected snails, at least when infected as juveniles (Krist & Lively, 1998). The most likely explanation is simply that the probability of having encountered a parasite increases with age. Thus older (longer) snails have a greater probability of being infected (Jokela & Lively, 1995a).

It cannot be determined for either shape or spininess whether the effects are *Microphallus*-specific or not. In both cases, snails infected with parasites other than *Microphallus* were not significantly different from *Microphallus*-infected snails and uninfected snails. The lack of differences here are likely due to the limited sample sizes of snails infected with other parasites. However, visually, snails infected with other parasites appear more similar to *Microphallus*-infected snails than to uninfected snails. This suggests, inconclusively, that parasitism, in general, results in changes in shape and defence morphology. If true, this would make explanations for the changes due to adaptations on the part of *Microphallus* less likely.

McCarthy *et al.* (2004) found that *Littorina saxatilis* infected by *Microphallus piriformes* were shaped

differently than uninfected snails. They concluded that the effect of the parasite was adaptive in that it increased the volume of infected snails resulting in increased space for the asexual production of metacercariae. Results here are a bit different. McCarthy *et al.* (2004) found that parasitism resulted in snails with a greater length to width ratio, while here we found that infected snails have a lower length to width ratio. Also, Krist & Lively (1998) found that juvenile *Potamopyrgus antipodarum* infected with *Microphallus* grew at a significantly slower rate than uninfected snails. Coupled with these results, it appears that the parasite may decrease the rate of growth in length to a greater degree than it influences the growth in width. This appears to make the adaptive manipulation of shape by the parasite in this system less likely. However, it is possible that given reduced growth rate caused by parasitism, the parasite may be making the best of a bad situation by increasing the growth in width and thus volume.

In conclusion, parasitism in *Potamopyrgus* is related to shell shape and defence morphology. Infected snails show a lower probability of being spiny and tend to be wider for a given length as length increases. The mechanism that results in this relationship cannot be determined from this study.

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REFERENCES

- APPLETON, R. D. & PALMER, A. R. (1988). Water-borne stimuli released by predatory crabs and damaged prey induce more predator-resistant shells in a marine gastropod. *Proceedings of the National Academy of Sciences, USA* **85**, 4387–4391.
- BALLABENI, P. (1995). Parasite-induced gigantism in a snail: a host adaptation? *Functional Ecology* **9**, 887–893.
- BRUSCA, R. C. & BRUSCA, G. J. (2003). *Invertebrates*, 2nd Edn. Sinauer Associates, Inc., Sunderland, Massachusetts.
- CREWS, A. E. & YOSHING, T. P. (1989). *Schistosoma mansoni* – effect of infection on reproduction and gonadal growth in *Biomphalaria glabrata*. *Experimental Parasitology* **68**, 302–334.
- FERNANDEZ, J. & ESCH, G. W. (1991). Effect of parasitism on the growth rate of the pulmonate snail *Helisoma anceps*. *Journal of Parasitology* **77**, 937–944.
- JOKELA, J. & LIVELY, C. M. (1995a). Spatial variation for infection by digenetic trematodes in a population of freshwater snails (*Potamopyrgus antipodarum*). *Oecologia* **103**, 509–517.
- JOKELA, J. & LIVELY, C. M. (1995b). Parasites, sex, and early reproduction in a mixed population of freshwater snails. *Evolution* **49**, 1268–1271.
- KRIST, A. C. (1998). The effects of parasites and predators on the life-history and morphology of freshwater snails. Ph.D. Dissertation. Indiana University, Bloomington, IN, USA.
- KRIST, A. C. (2000). Effect of the digenetic parasite *Proterometra macrostoma* on host morphology in the freshwater snail *Elimia livescens*. *Journal of Parasitology* **86**, 262–267.
- KRIST, A. C. (2002). Crayfish induce a defensive shell shape in a freshwater snail. *Invertebrate Biology* **121**, 235–242.
- KRIST, A. C. & LIVELY, C. M. (1998). Experimental exposure of juvenile snails (*Potamopyrgus antipodarum*) to infection by trematode larvae (*Microphallus* sp.): infectivity, fecundity compensation and growth. *Oecologia* **116**, 575–582.
- LEVRI, E. P. (1998a). The influence of non-host predators on parasite-induced behavioural changes in a freshwater snail. *Oikos* **81**, 531–537.
- LEVRI, E. P. (1998b). Perceived predation risk, parasitism and the foraging behaviour of a freshwater snail. *Canadian Journal of Zoology* **76**, 1878–1884.
- LEVRI, E. P. (1999). Parasite-induced changes in host behaviour of a freshwater snail: manipulation or byproduct of parasitism. *Behavioural Ecology* **10**, 234–241.
- LEVRI, E. P. & FISHER, L. (2000). The effect of a trematode (*Microphallus* sp.) on the response of the freshwater snail, *Potamopyrgus antipodarum* to light and gravity. *Behaviour* **137**, 1141–1151.
- LEVRI, E. P. & LIVELY, C. M. (1996). The effects of size, reproductive condition, and parasitism on the foraging behaviour in a freshwater snail, *Potamopyrgus antipodarum*. *Animal Behaviour* **51**, 891–901.
- LIVELY, C. M. (1987). Evidence from a New Zealand snail for the maintenance of sex by parasitism. *Nature, London* **328**, 519–521.
- LIVELY, C. M. & DYBDAHL, M. F. (2000). Parasite adaptation to locally common host genotypes. *Nature, London* **405**, 679–681.
- MCCARTHY, H. O., FITZPATRICK, S. M. & IRWIN, S. W. B. (2004). Parasite alteration of host shape: a quantitative approach to gigantism helps elucidate evolutionary advantages. *Parasitology* **128**, 7–14.
- MINCHELLA, K. J. (1985). Host life-history variation in response to parasitism. *Parasitology* **90**, 205–216.
- MOORE, J. (2002). *Parasitism and the Behaviour of Animals*. Oxford University Press, Oxford.
- MOURITSEN, K. N. & JENSEN, K. T. (1994). The enigma of gigantism: effect of larval trematodes on growth, fecundity, egestion and locomotion in *Hydrobia ulvae* (Pennant) (Gastropoda: Prosobranchia). *Journal of Experimental Marine Biology and Ecology* **181**, 53–66.
- POULIN, R. & THOMAS, F. (1999). Phenotypic variability induced by parasites: extent and evolutionary implications. *Parasitology Today* **15**, 28–32.
- ROTHSCHILD, A. & ROTHSCHILD, M. (1939). Some observations on the growth of *Peringia ulvae* (Pennant) (1777) in the laboratory. *Novitates Zoologicae* **41**, 240–247.

- SOUSA, W. P. (1983). Host life-history and the effect of parasitic castration on growth: a field study of *Cerithidea californica* Haldeman (Gastropoda: Prosobranchia) and its trematode parasites. *Journal of Experimental Marine Biology and Ecology* **73**, 273–296.
- THOMPSON, S. N. (1990). Physiological alterations during parasitism and their effects on host behaviour. In *Parasitism and Host Behaviour* (ed. Barnard, C. J. & Behnke, J. M.), pp. 193–233. Taylor and Francis, London.
- VERMEIJ, G. J. (1993). *A Natural History of Shells*. Princeton University Press, Princeton, New Jersey.
- WINTERBOURN, M. (1970). The New Zealand species of *Potamopyrgus* (Gastropoda: Hydrobiidae). *Malacologia* **10**, 283–321.
- WINTERBOURN, M. (1974). Larval Trematoda parasitizing the New Zealand species of *Potamopyrgus* (Gastropoda: Hydrobiidae). *Mauri Ora* **2**, 17–30.
- ZUK, M. (1992). The role of parasites in sexual selection: current evidence and future directions. *Advances in the Study of Behaviour* **21**, 39–68.

APPENDIX

Statistical results from pairwise ANCOVA comparing classes of snails with regard to their width of the first whorl and square root of the two-dimensional area using length as a covariate. Results are shown only for tests where there was no significant difference in the slopes between the two classes or if the difference was marginally significant.

Uninfected non-brooding females *vs* Uninfected brooding females

Tests of Between-Subjects Effects

Source	Dependent Variable	Type I Sum of Squares	df	Mean Square	F	P
Corrected Model	width	3.334	2	1.667	129.2	<0.001
	sqrtarea	6.551	2	3.276	410.3	<0.001
Intercept	width	662.4	1	662.4	51339.0	<0.001
	sqrtarea	793.3	1	793.3	99376.4	<0.001
Length	width	3.304	1	3.304	256.0	<0.001
	sqrtarea	6.549	1	6.549	820.4	<0.001
Class	width	0.0303	1	0.0303	2.353	0.127
	sqrtarea	0.0018	1	0.0018	0.220	0.640
Error	width	2.026	157	0.0129		
	sqrtarea	1.253	157	0.0080		
Total	width	667.8	160			
	sqrtarea	801.2	160			
Corrected Total	width	5.360	159			
	sqrtarea	7.805	159			

Uninfected females *vs* Uninfected males

Tests of Between-Subjects Effects

Source	Dependent Variable	Type I Sum of Squares	df	Mean Square	F	P
Corrected Model	width	11.76	2	5.878	486.0	<0.001
	sqrtarea	21.33	2	10.663	1559.8	<0.001
Intercept	width	1119.0	1	1119.0	92518.3	<0.001
	sqrtarea	1379.9	1	1379.9	201839.5	<0.001
Length	width	11.35	1	11.35	938.5	<0.001
	sqrtarea	21.23	1	21.23	3105.0	<0.001
Class	width	0.406	1	0.406	33.54	<0.001
	sqrtarea	0.099	1	0.099	14.55	<0.001
Error	width	3.036	251	0.0121		
	sqrtarea	1.716	251	0.0068		
Total	width	1133.8	254			
	sqrtarea	1402.9	254			
Corrected Total	width	14.79	253			
	sqrtarea	23.04	253			

Uninfected females *vs* *Microphallus*-infected females

Tests of Between-Subjects Effects

Source	Dependent Variable	Type I Sum of Squares	df	Mean Square	F	<i>P</i>
Corrected Model	width	18.27	2	9.132	712.0	<0.001
	sqrtarea	34.31	2	17.16	2287.2	<0.001
Intercept	width	1522.8	1	1522.8	118725.3	<0.001
	sqrtarea	1893.3	1	1893.3	252410.2	<0.001
Length	width	13.84	1	13.84	1079.2	<0.001
	sqrtarea	26.48	1	26.48	3530.7	<0.001
Class	width	4.423	1	4.423	344.8	<0.001
	sqrtarea	7.829	1	7.829	1043.8	<0.001
Error	width	4.015	313	0.0128		
	sqrtarea	2.348	313	0.0075		
Total	width	1545.1	316			
	sqrtarea	1929.9	316			
Corrected Total	width	22.28	315			
	sqrtarea	36.66	315			

Uninfected males *vs* *Microphallus*-infected males

Tests of Between-Subjects Effects

Source	Dependent Variable	Type I Sum of Squares	df	Mean Square	F	<i>P</i>
Corrected Model	width	1.228	2	0.614	53.39	<0.001
	sqrtarea	1.765	2	0.883	320.3	<0.001
Intercept	width	131.5	1	131.5	11440.6	<0.001
	sqrtarea	170.5	1	170.5	61860.3	<0.001
Length	width	1.222	1	1.222	106.3	<0.001
	sqrtarea	1.762	1	1.762	636.3	<0.001
Class	width	0.0053	1	0.0053	0.460	0.503
	sqrtarea	0.0034	1	0.0034	1.216	0.279
Error	width	0.356	31	0.0115		
	sqrtarea	0.085	31	0.0028		
Total	width	133.1	34			
	sqrtarea	172.3	34			
Corrected Total	width	1.584	33			
	sqrtarea	1.850	33			

Uninfected females *vs* Females infected with non-microphallid parasites

Tests of Between-Subjects Effects

Source	Dependent Variable	Type I Sum of Squares	df	Mean Square	F	P
Corrected Model	width	11.02	2	5.509	454.1	<0.001
	sqrtarea	21.06	2	10.53	1430.2	<0.001
Intercept	width	1078.2	1	1078.2	88871.8	<0.001
	sqrtarea	1325.0	1	1325.0	179963.9	<0.001
Length	width	11.02	1	11.02	908.0	<0.001
	sqrtarea	21.06	1	21.06	2860.0	<0.001
Class	width	0.00099	1	0.00099	0.082	0.775
	sqrtarea	0.00286	1	0.00286	0.389	0.533
Error	width	2.851	235	0.0121		
	sqrtarea	1.730	235	0.0074		
Total	width	1092.12	238			
	sqrtarea	1347.74	238			
Corrected Total	width	13.87	237			
	sqrtarea	22.79	237			

Microphallus-infected females *vs* Females infected with non-microphallid parasites

Tests of Between-Subjects Effects

Source	Dependent Variable	Type I Sum of Squares	df	Mean Square	F	P
Corrected Model	width	4.667	2	2.333	174.158	<0.001
	sqrtarea	8.173	2	4.087	578.5	<0.001
Intercept	width	567.497	1	567.497	42356.6	<0.001
	sqrtarea	726.388	1	726.388	102831.1	<0.001
Length	width	4.653	1	4.653	347.32	<0.001
	sqrtarea	8.159	1	8.159	1155.1	<0.001
Class	width	0.01332	1	0.0133	0.994	0.321
	sqrtarea	0.01403	1	0.0140	1.987	0.162
Error	width	1.313	98	0.0134		
	sqrtarea	0.692	98	0.0070		
Total	width	573.476	101			
	sqrtarea	735.253	101			
Corrected Total	width	5.980	100			
	sqrtarea	8.866	100			