Parasites may contribute to 'magic trait' evolution in the adaptive radiation of three-spined sticklebacks, *Gasterosteus aculeatus* (Gasterosteiformes: Gasterosteidae)

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In the past decade, there has been a new effort to understand the ecology that drives population divergence and speciation. It is well established in theory that speciation is most likely to occur when a trait that is under divergent natural selection in different populations is also used in mate choice. Such traits have been dubbed 'magic traits' (Gavrilets, 2004) and, although there appears to be good evidence that they exist, the ecological mechanisms that underlie their divergence are not well understood. Size at maturity in three-spined sticklebacks is an archetypal example of a magic trait. The present study documents for the first time that differences in body size at maturity in sympatric species pairs of lacustrine three-spined sticklebacks in British Columbia, Canada, are caused by differences in age at maturity. It is also shown that there are differences between the sympatric species in the patterns of infection with a virulent cestode, *Schistocephalus solidus*. Although the evidence is circumstantial, these differences in infection are consistent with the hypothesis that they have contributed to the observed divergence in age and size at maturity in these populations. © 2009 The Linnean Society of London, *Biological Journal of the Linnean Society*, 2009, **96**, 425–433.

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INTRODUCTION

Speciation is greatly facilitated when traits that experience divergent selection between populations also contribute to reproductive isolation (Slatkin, 1982). Such traits have been termed 'magic traits' by Gavrilets (2004: 368). As yet, there are too few good examples of magic traits in animals to allow generalization about which aspects of ecology generate divergent selection or the kind of traits upon which selection acts. For half a century, the idea that competition for resources, and the ensuing evolution of specialized trophic morphology, can account for

divergence has been a central paradigm of evolutionary ecology (Dobzhansky, 1951; Simpson, 1953; Schluter, 2000). There have been several experimental studies where competition has been shown to result in divergent selection (Pacala & Roughgarden, 1985; Taper & Case, 1992; Schluter, 1996) but there is little evidence that such divergence can result in reproductive isolation. Indeed, the kind of traits that allow resource specialization may not be good candidates for magic traits because, generally, they have little obvious link to mate choice; although see also Hawthorne & Via (2001) and Podos (2001). For this and other reasons, in the past decade, evolutionary ecologists have begun to question whether competition and resource specialization are the only mechanisms underlying adaptive radiation and speciation. In particular, there has been interest in the impact of

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natural enemies (Vamosi, 2005). Recent studies have demonstrated a role for predation in magic trait evolution and speciation (Naisbit, Jiggins & Mallet, 2001; Vamosi & Schluter, 2002; Nosil, 2004). Despite this, few studies performed to date have explicitly examined whether parasites could contribute to the adaptive radiation of their hosts (Buckling & Rainey, 2002) and none in natural populations. This is surprising given the ubiquity of parasites in natural systems and their acknowledged ability to cause divergent selection of traits involved in mate choice (Hamilton & Zuk, 1982; Summers *et al.*, 2003).

Body size has a good claim to be a generic magic trait given that it probably diverges more frequently than any other trait during adaptive radiation (Schluter, 2000) and has been linked to reproductive isolation in several systems (Jones *et al.*, 2003; McKinnon *et al.*, 2004; Bolnick, Near & Wainwright, 2006; Richmond & Jockusch, 2007). However, the causes of the divergence have seldom been directly examined (Camp, Marshall & Austin, 2000; Giannasi, Thorpe & Malhotra, 2000). This is surprising given the well researched and documented ecological consequences of variation in body size (Peters, 1983).

The possibility that divergent selection due to natural enemies can drive the evolution of life-history characters such as body size has long been appreciated (Gadgil & Bossert, 1970). Theory leads us to expect that, if certain age or size classes are particularly susceptible to natural enemies, then natural selection should favour those individuals with life histories that minimize this fitness reduction. This principle has perhaps been best explored in the evolutionary interaction between guppies (*Poecilia reticulata*) and their predators (Reznick, Bryga & Endler, 1990). There are few examples, however, in which variation in parasitism between host populations has been suggested to lead to evolved differences in host life-history traits (Hamilton & Poulin, 2001; Koskela, 2002; Fredensborg & Poulin, 2006) despite the fact tha the possibility is well supported theoretically (Agnew, Koella & Michalakis, 2000; Koella & Restif, 2001). If the probability of infection with a virulent parasite increases with age in a host population then selection is likely to favour those individuals that mature early and thus avoid the negative evolutionary effect of the parasite. In organisms with discrete, ecologically prescribed breeding seasons, this could be quite significant because there may be a discontinuous response to such selection. For example, although selection in the absence of natural enemies might favour breeding at the age of 2 years, mortality due to parasitism between the ages of 1 and 2 years may favour earlier breeding and result in breeding at 1 year of age rather than at something just less than 2 years.

In coastal British Columbia, there are seven lakes that contain, or have until recently contained, coexisting, reproductively isolated populations of two types of three-spined sticklebacks (*Gasterosteus aculeatus* L. complex). The types are generally accepted to comprise good biological species given that they are substantially reproductively isolated with only low levels of hybridization (McPhail, 1992; Gow, Peichel & Taylor, 2006). In each lake, one species, termed 'limnetic', inhabits the pelagic zone and feeds largely on plankton, whereas the other, termed 'benthic', is littoral and feeds in the benthos. Both breed in the littoral zone. The available evidence suggests that the 'species pairs' in these lakes are evolutionarily independent (Taylor & McPhail, 1999; Taylor & McPhail, 2000) and probably evolved as the result of double invasions of both lakes from the sea by a common marine ancestor (McPhail, 1993). Sticklebacks in these populations begin breeding around the beginning of April and most breeding has finished by late June. The timing of breeding is probably associated with the availability of suitable food for fry and the need to grow as much as possible before winter. The consequence is that, for sticklebacks, the distribution of ecologically and evolutionary feasible ages at maturity is not continuous in time. Rather, the adaptive landscape for age at maturity probably has the kind of sharp peaks, at ages 1, 2, and 3 years, etc., that could favour speciation (Schluter, 2000).

The present study shows for the first time that the sympatric species pairs in British Columbia have quite different life histories that result in different age and size at maturity. These differences in life history are consistent with selection caused by differences in patterns of infection by the virulent cestode *Schistocephalus solidus*. Abundant evidence shows that, in mate choice trials in sticklebacks, body size is the most important trait determining compatibility between populations. Choice is positively size assortative (i.e. big fish prefer to mate with big fish and small with small; McKinnon *et al.*, 2004). Body size has diverged greatly in the adaptive radiation of sticklebacks and tends to be correlated with environment (McPhail, 1977). Thus, body size is a magic trait in sticklebacks (Gavrilets, 2004).

MATERIAL AND METHODS

Sticklebacks were sampled from Paxton lake and Priest Lake (hereafter Paxton and Priest) both on Texada Island (49°40′N 124°25′W) in the Georgia Strait, BC, Canada. The lakes are approximately 4 km apart in separate catchments that drain to opppposite sides of the island. Both lakes contain benthic and limnetic sticklebacks. Paxton and Priest were sampled on five separate occasions at

approximately monthly intervals from April to September 2004 (22 April, 25 May, 28 June, 27 July, and 14 September). Fish were caught in minnow traps ('Gee's Minnow Traps', Tackle Factory). Twenty of these were set from the bank or a boat in approximately 20–200 cm of water, to cover a range of microhabitats. These included sites along the edge of emergent vegetation in deep and shallow water, in weed in deep and shallow water, along the sides of fallen logs or large stones, and resting in the open on the benthos. Individual traps commonly caught a range of sizes of sticklebacks of both sexes and both species. This suggests that there is substantial overlap in microhabitat use by species/sex/age classes, at least during the spring and summer. Traps were set in approximately the same locations in each trapping session. Traps were lifted the day after setting and the contents emptied into a bucket of lake water. Approximately 15 fish (mean 13.7, range 5–20) of each species were selected haphazardly from among those in the bucket and were returned alive to the laboratory in sealed aerated cool boxes. Fish were euthanased and dissected fresh within 1 week, or were euthanased and stored in 70% ethanol for dissection at a later date. Fish were dissected and the number of *S. solidus* plerocercoids recorded. For fish caught on and after 28 June, the reproductive condition of individuals was recorded as active (females: any enlarged ovules, males: enlarged testes) or inactive (females: no enlarged ovules, males: undeveloped testes). The presence or absence of perivisceral fat (fat on the intestine) was also recorded.

To establish the length distribution of fish, samples of both species of sticklebacks, from Priest Lake only, had their standard length measured to the nearest 0.5 mm at the lakeside, using a 'stopped' avian wing rule (BTO). Sticklebacks from Paxton were not measured in this way partly because of a desire to reduce intervention in this lake, where there are stickleback conservation concerns, and partly because it is known to be difficult to catch limnetics in Paxton as the summer progresses. Samples for measurement purposes were collected (using the same methods as above) from Priest on 25 May, 4 June, 28 June, 27 July, and 14 September. The aim was to measure at least 100 'female' (see below) stickleback of each species, haphazardly selected from the holding bucket. This was sometimes not possible, especially for limnetics. As individuals were measured, they were classified as obvious males (any red coloration on the face, throat, or belly) and 'females' which comprised all other fish. It is likely that the 'female' sample included some young males in both species. In the sample of fish caught on 4 June, females were classified as gravid (swelling of abdomen not due to *S.*

solidus infection) or not, in order to examine differences between the species in size at reproduction.

STATISTICAL ANALYSIS

To assess whether one or more age class was present among the fish that were measured from Priest, the goodness of fit of length frequency data to a single or double normal distribution model was assessed using the Distribution procedure in GENSTAT, release 8.1 (VSN International). The difference in deviance between fits for double and single normal distributions was compared to chi squared with two degree of freedom (for the additional mean and variance parameters).

Log-linear models (GENSTAT) were used to analyse count (including prevalence) data. Log-linear models provide a flexible method for the analysis of multidimensional contingency table data. For example, with a three-dimensional table, the modelling process begins by fitting a fully specified model with the main effects of a, b, and c and all their two and three-way interactions (where a, b, and c are the dimensions of the table each with two or more discrete classes; e.g. lake, species, and season). This model is necessarily a perfect fit to the data. Simpler models are then sought by dropping interactions (most complex first) and comparing the change in deviance to chi squared with appropriate degrees of freedom. If interactions are significant then this indicates that the dimensions involved in the interaction are not independent and the interaction should be retained in the model.

RESULTS

BODY SIZE DISTRIBUTIONS

Length frequency distributions for Priest benthics showed some suggestion of bimodality in most of the samples collected (Fig. 1) and they were significantly better fitted by a double normal than a single normal distribution in three of the five samples (Table 1). The histograms provided in Figure 1 are consistent with a 2+ year age class having been present early in the year, but having gradually died out as the 1+ year age class strengthened. Fish of the new cohort (0+) began to appear in July and were present in relatively large proportion by September. By contrast, the length frequency distribution for limnetics in Priest appeared to consist of mainly 1+ fish and showed no sign of bimodality (Fig. 1, Table 1), except in September when a 0+ age class was apparent. The double normal distribution model for limnetics for June failed to converge (Table 1). However, the good fit of the single normal distibution model (deviance = 10.44, d.f. = 3) indicates little scope for improvement in fit by
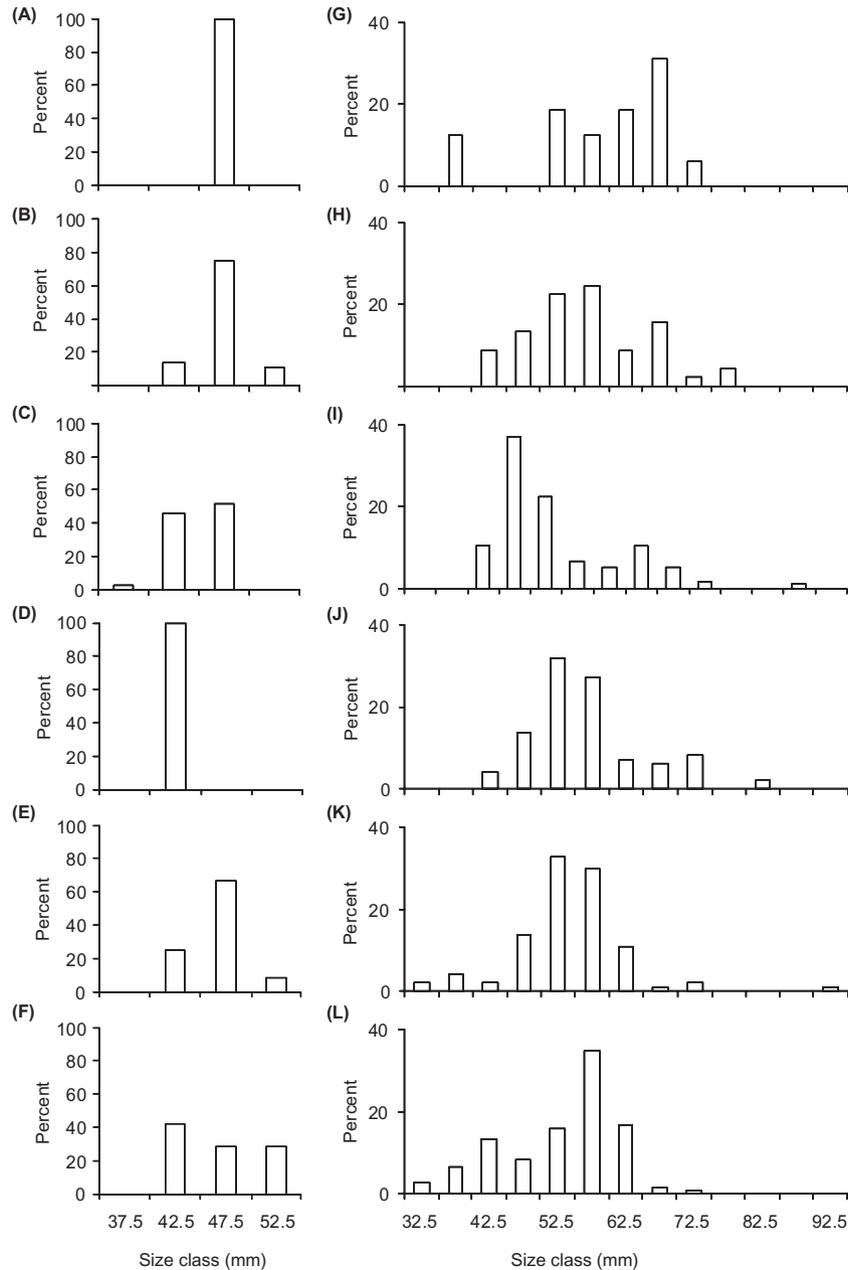
Figure 1. The length frequency distributions of 'female' (see text) three-spined sticklebacks sampled from Priest Lake for six time points in 2004. A–F, limnetics. G–L, benthics. The six time points were (with sample sizes for limnetics and benthics respectively): 22 April (8, 16) (A, G); 25 May (64, 45) (B, H); 4 June (37, 116) (C, I); 28 June (10, 100) (D, J); 27 July (12, 100) (E, K); 14 September (7, 157) (F, L). Note that the scale of the *y*-axes in (A) to (F) is different from that in (G) to (L).

a double normal distribution model. This supports the idea that these data come from a single normal distribution.

In the sample of fish caught on 4 June, the length distribution of gravid benthics was significantly shifted towards longer fish compared with benthics that were not gravid; compare Fig. 1(I) and Fig. 2 (Kolmogorov–Smirnov, $\chi^2 = 36.2$, d.f. = 2, $P < 0.001$). There was no such difference between gravid and nongravid limnetics; compare Fig. 1(C) and Fig. 2 (Kolmogorov–Smirnov, $\chi^2 = 3.33$, d.f. = 2, $P = 0.19$). Gravid benthics were also longer than gravid limnetics (Kolmogorov–Smirnov, $\chi^2 = 28.9$, d.f. = 2, $P < 0.001$; Fig. 2).

Table 1. Fit of double or single normal distributions to length data for female sticklebacks of both species in Priest Lake

Species	Date	Double normal		Single normal		
		Deviance	d.f.	Deviance	d.f.	P
Benthic	25 May	1.84	2	2.17	4	0.85
	4 June	3.81	6	45.3	8	< 0.0001
	28 June	10.1	5	29.1	7	< 0.0001
	27 July	–		5.36	7	–
	14 September	23.8	8	70.1	10	< 0.0001
Limnetic	25 May	9.88	3	10.37	5	0.78
	4 June	–		10.44	3	

Dashes indicate nonconvergence of models. *P*-values are for the difference in deviance between the single and double normal models compared to chi squared, contingent on the difference in degrees of freedom between the models. A significant *P*-value indicates that a double normal was a better fit than a single normal.
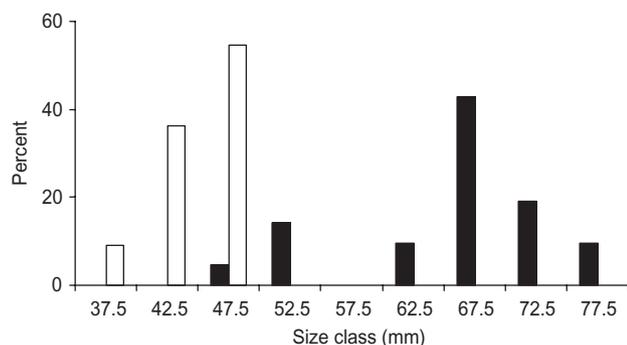
Figure 2. The length frequency distribution of gravid female limnetics ($N = 11$, open histogram) and benthics ($N = 21$, solid histogram) in the sample measured on 4 June 2004 at the height of the breeding season.

PATTERNS OF PREVALENCE OF *S. SOLIDUS*

There was a very large and significant difference in prevalence of *S. solidus* infection between the stickleback species and this did not differ between the lakes. Indeed, prevalence was zero in benthics in both lakes and high in limnetics in both lakes (log-linear model incorporating lake, species and infection status: lake × species × infection status, $\chi^2 = 0.00$, d.f. = 1, $P = 0.96$; species × infection status, $\chi^2 = 109.6$, d.f. = 1, $P < 0.0001$; Fig. 3). There was a difference between the lakes in the seasonal pattern of infection in limnetics. In both lakes, the prevalence started low but, whereas in Paxton it climbed through the summer to reach 100% by September, in Priest it stayed at approximately 40% (log-linear model, for limnetics only; Fig. 3) incorporating lake, month, and infection status: lake × month × infection status; $\chi^2 = 2.39$, d.f. = 4, $P = 0.048$). However, the overall difference in prevalence between the lakes was not quite significant (in the same log-linear model: lake × infection status; $\chi^2 = 3.67$, d.f. = 1, $P = 0.056$).
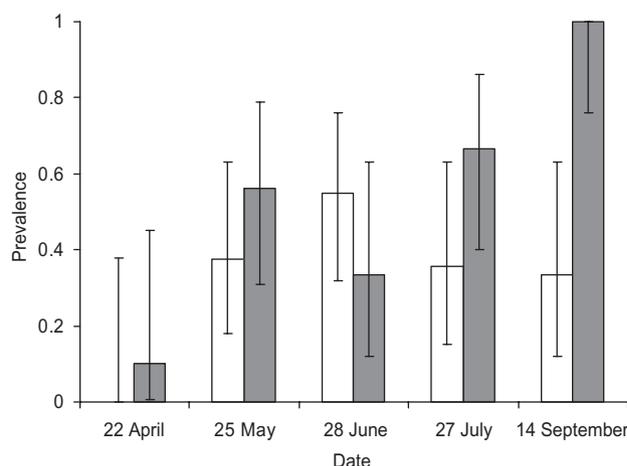
Figure 3. Seasonal change in the prevalence (mean ± 95% confidence interval) of *Schistocephalus solidus* in limnetics in Priest (open) and Paxton (shaded). Sample sizes: Priest, 7, 16, 20, 14, and 14; Paxton, 10, 16, 12, 15, and 12.

To look at the prevalence of *S. solidus* in different age classes, limnetics were assigned to the 0+ or 1+ age class on the basis of standard length (0 + if less than 40 mm). Among the 0+ fish (which were caught only in July and September), the prevalence of *S. solidus* was 19 of 26 in Paxton and zero of three in Priest.

PERIVISCERAL FAT AND *S. SOLIDUS*

Benthics were much more likely to have perivisceral fat than were limnetics and this pattern did not differ between lakes (log-linear model incorporating lake, species, and presence of fat: lake × species × fat, $\chi^2 = 0.68$, d.f. = 1, $P = 0.41$; species × fat, $\chi^2 = 99.2$, d.f. = 1, $P < 0.0001$; Table 2). Infected limnetics were significantly less likely to have perivisceral fat than uninfected fish, although this was largely the

Table 2. Proportions of benthic and limnetic sticklebacks from Paxton and Priest Lakes that had perivisceral fat (around the intestine) and that were in reproductive condition in late summer (June to September samples combined)

Lake	Paxton				Priest			
		Limnetic				Limnetic		
Species	Benthic	Overall	Uninfected	Infected	Benthic	Overall	Uninfected	Infected
Sample size	39	37	13	24	49	39	20	19
Proportion with perivisceral fat	0.79 (0.64, 0.90)	0.11 (0.05, 0.27)	0.08 (0.00, 0.34)	0.13 (0.03, 0.31)	0.94 (0.83, 0.98)	0.18 (0.09, 0.34)	0.35 (0.17, 0.58)	0.00 (0.00, 0.18)
Proportion in reproductive condition	0.33 (0.22, 0.51)	0.38 (0.24, 0.55)	0.77 (0.48, 0.93)	0.17 (0.06, 0.37)	0.35 (0.23, 0.49)	0.90 (0.76, 0.97)	0.90 (0.68, 0.98)	0.89 (0.68, 0.98)

Means for limnetics are broken down according to whether fish were infected with *Schistocephalus solidus*. Data are the mean ± 95% confidence interval.

result of a difference in Priest (log-linear model incorporating lake, infection status, and presence of fat: lake × infection status × fat, $\chi^2 = 6.96$, d.f. = 1, $P = 0.008$; infection status × fat, $\chi^2 = 4.53$, d.f. = 1, $P = 0.03$; Table 2).

REPRODUCTIVE CONDITION AND *S. SOLIDUS*

Uninfected limnetics were much more likely to be in reproductive condition than were (uninfected) benthics and this pattern did not differ between lakes (log-linear model incorporating lake, species, and reproductive condition: lake × species × reproductive condition, $\chi^2_1 = 0.03$, $P = 0.85$; species × reproductive condition, $\chi^2_1 = 21.3$, $P < 0.001$; Table 2).

Limnetics infected with *S. solidus* were less likely to be in reproductive condition, although this was largely due to a difference in Paxton fish (log-linear model incorporating lake, infection status and reproductive condition: lake × infection status × reproductive condition, $\chi^2 = 9.9$, d.f. = 1, $P = 0.002$; infection status × reproductive condition, $\chi^2 = 5.13$, d.f. = 1, $P = 0.024$; Table 2).

DISCUSSION

It is clear from the results obtained in the present study that there are substantial differences in life history between benthics and limnetics. The former often live to their third summer (2+ age class) and females, at least, delay much of their reproduction until they are 2 years old. The fact that benthics are much more likely to accumulate perivisceral fat and much less likely to be in reproductive condition when caught is consistent with this life history. Limnetics, in contrast, seldom or never reach their third summer in the wild but reproduce when they are 1 year old. There is little variation in length among limnetics and this is consistent with a short intense pulse of reproduction early in summer. Indeed, it becomes difficult to catch limnetics after early to mid June and substantial die offs are sometimes seen at this time (A. D. C. MacColl, pers. observ.; M. E. Arnegard & J. Courchesne, pers. comm.). This suggests that the life history of wild limnetics approaches semelparity, consistent with the fact that limnetics are much more likely to be in reproductive condition and seldom accumulate perivisceral fat. Despite all this, limnetics raised from eggs in the laboratory or caught in the wild early in the season and moved to the laboraory, often live and grow through their second winter and into a third summer (A. D. C. MacColl, pers. observ.; D. Schluter, pers. comm.; T. H. Vines, pers. comm.). Limnetics that are captured in the wild later in the season are commonly infected with *S. solidus* and their survival is poor after transfer to the laboratory

(S. Desroches, pers. comm.). This indicates that environmental factors are at least partly responsible for the failure of limnetics to live longer in the wild and that *S. solidus* is one such factor (see below). The importance of environment in post-reproductive survival of limnetics does not imply that there is no evolved difference in life history between the species because probably all limnetics breed at the end of their first year, whereas few benthics do. Age at maturity in three-spined sticklebacks is known to have a genetic component (McPhail, 1977).

The difference in age at maturity between benthics and limnetics is a major proximate cause of the difference in size at breeding. Given the important role of size assortative mating in causing prezygotic reproductive isolation in three-spined sticklebacks (McKinnon *et al.*, 2004), it is highly likely that the difference in life histories reported in the present study has contributed to the reproductive isolation between these species.

It is also clear from the results of the present study that there is a very large difference between benthics and limnetics in the levels of infection by the cestode *S. solidus*. The prevalence of *S. solidus* is low or zero in limnetics early in the year but rises to high levels as the summer progresses. This pattern of infection has been consistent across several years (A. D. C. MacColl, pers. observ.; S. M. Vamosi, pers. comm.; S. Desroches, pers. comm.) and has been documented in other populations of sticklebacks in coastal British Columbia (McPhail & Peacock, 1983). By contrast, *S. solidus* infections in benthics are very rare throughout the year and this is also consistent across years (personal observations over 4 years). Given what is already known about the pathological and selective impact of *S. solidus* infection (Arme & Owen, 1967; Tierney, Huntingford & Crompton, 1996; Barber & Svensson, 2003), this difference is so substantial that it almost certainly gives rise to selective pressures that favour divergent life histories for limnetics and benthics. Apart from the associations between *S. solidus* infection and reproductive condition and the absence of perivisceral fat recorded in the present study, it is likely that many of the limnetics that are infected by *S. solidus* die as a direct or indirect consequence of infection (e.g. higher risk of predation and starvation; Pascoe & Mattey, 1977; Giles, 1987). This is supported by the observation that the prevalence levels in limnetics are consistently low at the start of the year despite the fact that many 0+ fish are infected by the end of their first summer, in Paxton at least. By contrast, benthics, by invading the littoral zone and (presumably) not feeding on the copepod intermediate hosts of *S. solidus*, have evaded this selection pressure. All else being equal, the patterns of infection documented in the present study should favour early onset of, and heavy investment in, reproduction by limnetics, but investment in growth and maintenance by benthics.

The results reported in the present study are strongly consistent with the idea that infection by *S.solidus* has favoured the evolution of an annual, almost semelparous, life history in limnetics and/or that escape from infection has favoured the evolution of a slower life history in benthics. The most parsimonious interpretation depends on the likely ancestral state. The best available information shows that individuals in the wild live for more than a single year in the majority of populations whether lacustrine or anadromous (Baker, 1994). Information is lacking about the life history of marine sticklebacks, although it is likely from their size at maturity that they are multi-annual (Baker, 1994). This supports the idea that the annual life history of limnetics (and numerous other freshwater populations) is an evolutionarily derived state.

Although it is highly likely from the evidence provided by the present study that differences in *S. solidus* infection are contributing to the maintenance of the status quo in terms of age and size at maturity, the data are circumstantial and do not necessarily mean that *S. solidus* is the ultimate cause of the difference in life histories. Body size is a fundamentally important ecological trait and its variation can have diverse causes and consequences (Peters, 1983; Roff, 2002). There are at least two other factors that might curtail limnetic survival during their second winter and so might have contributed to the evolution of the observed differences in age at maturity. The first is predation: sticklebacks in both Paxton and Priest coexist with only one other fish species, the cutthroat trout *Onchorhynchus clarki*. It is likely that limnetics are more severely affected by predation given that they spend most of their time, outside the breeding season, in the pelagic zone (Vamosi & Schluter, 2002; Vamosi & Schluter, 2004) and differences in predation between lakes have previously been implicated in life-history divergence between allopatric populations of sticklebacks in British Columbian lakes (McPhail, 1977). The second factor is insufficient food resources. It is possible that there is insufficient food available in the pelagic zone to maintain larger 1+ limnetics through a second winter. This is not consistent with the observation that benthics rather than limnetics are more likely to accumulate internal fat stores, unless the lack of food is severe and sustained. It is also possible that there is an interaction between some combination of parasitism, predation, and resource availability.

There is already good evidence that body size is a magic trait in three-spined sticklebacks (Gavrilets, 2004; McKinnon *et al.*, 2004). The results obtained in

the present study show that differences in body size between sympatric stickleback species in British Columbia result from differences in age at maturity and suggest that a parasite may have played an important role in this divergence. Previous studies have demonstrated a role for resource specialization (Hawthorne & Via, 2001) and predation (Naisbit *et al.*, 2001) in the evolution of magic traits. If the conditions that lead to speciation are to be understood properly, there needs to be additional study of the aspects of phenotype that can become magic traits and the ecological situations than can cause divergent selection. The present study indicates a role for parasites, which should be explored further in this and other systems, given what we know about the potential for parasites to drive the evolution of traits that are important in mate choice (Hamilton, 1964; Summers *et al.*, 2003).

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REFERENCES

Agnew P, Koella JC, Michalakis Y. 2000. Host life history responses to parasitism. *Microbes and Infection* **2:** 891–896.

Arme C, Owen RW. 1967. Infections of three-spined stickleback *Gasterosteus aculeatus* L., with plerocercoid larvae of *Schistocephalus solidus* (Muller 1776) with special reference to pathological effects. *Parasitology* **57:** 301–314.

Baker JA. 1994. Life history variation in female threespine stickleback. In: Bell MA, Foster SA, eds. *The evolutionary biology of the threespine stickleback*. Oxford: Oxford University Press, 146–187.

Barber I, Svensson PA. 2003. Effects of experimental *Schistocephalus solidus* infections on growth, morphology and sexual development of female three- spined sticklebacks, *Gasterosteus aculeatus*. *Parasitology* **126:** 359–367.

Bolnick DI, Near TJ, Wainwright PC. 2006. Body size divergence promotes post-zygotic reproductive isolation in centrarchids. *Evolutionary Ecology Research* **8:** 903–913.

Buckling A, Rainey PB. 2002. The role of parasites in sympatric and allopatric host diversification. *Nature* **420:** 496–499.

Camp CD, Marshall JL, Austin RM. 2000. The evolution of adult body size in black-bellied salamanders (*Desmognathus quadramaculatus* complex). *Canadian Journal of Zoology* **78:** 1712–1722.

Dobhzhansky T. 1951. *Genetics and the origin of species*. New York, NY: Columbia University Press.

Fredensborg BL, Poulin R. 2006. Parasitism shaping host life-history evolution: adaptive responses in a marine gastropod to infection by trematodes. *Journal of Animal Ecology* **75:** 44–53.

Gadgil M, Bossert WH. 1970. Life historical consequences of natural selection. *The American Naturalist* **104:** 1–24.

Gavrilets S. 2004. *Fitness landscapes and the origin of species*. Princeton, NJ: Princeton University Press.

Giannasi N, Thorpe RS, Malhotra A. 2000. A phylogenetic analysis of body size evolution in the *Anolis roquet* group (Sauria: Iguanidae): character displacement or size assortment? *Molecular Ecology* **9:** 193–202.

Giles N. 1987. Predation risk and reduced foraging activity in fish: experiments with parasitized and non-parasitized three-spined sticklebacks, *Gasterosteus aculeatus* L. *Journal of Fish Biology* **31:** 37–44.

Gow JL, Peichel CL, Taylor EB. 2006. Contrasting hybridization rates between sympatric three-spined sticklebacks highlight the fragility of reproductive barriers between evolutionarily young species. *Molecular Ecology* **15:** 739–752.

Hamilton WD. 1964. The genetical evolution of social behaviour. *Journal of Theoretical Biology* **7:** 1–52.

Hamilton WD, Zuk M. 1982. Heritable true fitness and bright birds – a role for parasites. *Science* **218:** 384–387.

Hamilton WJ, Poulin R. 2001. Parasitism, water temperature and life history characteristics of the freshwater fish *Gobiomorphus breviceps* Stokell (Eleotridae). *Ecology of Freshwater Fish* **10:** 105–110.

Hawthorne DJ, Via S. 2001. Genetic linkage of ecological specialization and reproductive isolation in pea aphids. *Nature* **412:** 904–907.

Jones AG, Moore GI, Kvarnemo C, Walker D, Avise JC. 2003. Sympatric speciation as a consequence of male pregnancy in seahorses. *Proceedings of the National Academy of Sciences of the United States of America* **100:** 6598–6603.

Koella JC, Restif O. 2001. Coevolution of parasite virulence and host life history. *Ecology Letters* **4:** 207–214.

Koskela T. 2002. Variation in life-history traits among *Urtica dioica* populations with different history in parasitism by the holoparasitic plant *Cuscuta europaea*. *Evolutionary Ecology* **16:** 433–454.

McKinnon JS, Mori S, Blackman BK, David L, Kingsley DM, Jamieson L, Chou J, Schluter D. 2004. Evidence for ecology's role in speciation. *Nature* **429:** 294–298.

McPhail JD. 1977. Inherited interpopulation differences in size at first reproduction in threespine stickleback, *Gasterosteus aculeatus* L. *Heredity* **38:** 53–60.

McPhail JD. 1992. Ecology and evolution of sympatric sticklebacks (*Gasterosteus*) – evidence for a species pair in Paxton lake, Texada island, British Columbia. *Canadian Journal of Zoology* **70:** 361–369.

McPhail JD. 1993. Ecology and evolution of sympatric sticklebacks (*Gasterosteus*): origin of the species pairs. *Canadian Journal of Zoology* **71:** 515–523.

McPhail JD, Peacock SD. 1983. Some effects of the cestode (*Schistocephalus solidus*) on reproduction in the threespine

stickleback (*Gasterosteus aculeatus*), evolutionary aspects of a host parasite interaction. *Canadian Journal of Zoology* **61:** 901–908.

Naisbit RE, Jiggins CD, Mallet J. 2001. Disruptive sexual selection against hybrids contributes to speciation between *Heliconius cydno* and *Heliconius melpomene*. *Proceedings of the Royal Society of London Series B, Biological Sciences* **268:** 1849–1854.

Nosil P. 2004. Reproductive isolation caused by visual predation on migrants between divergent environments. *Proceedings of the Royal Society of London Series B, Biological Sciences* **271:** 1521–1528.

Pacala SW, Roughgarden J. 1985. Population experiments with the *Anolis* lizards of St Maarten and St Eustatius. *Ecology* **66:** 129–141.

Pascoe D, Mattey D. 1977. Dietary stress in parasitised and non-parasitised *Gasterosteus aculeatus* L. *Zeitschrift fur Parasitenkunde* **51:** 179–186.

Peters RH. 1983. *The ecological implications of body size*. Cambridge: Cambridge University Press.

Podos J. 2001. Correlated evolution of morphology and vocal signal structure in Darwin's finches. *Nature* **409:** 185–188.

Reznick DA, Bryga H, Endler JA. 1990. Experimentally induced life-history evolution in a natural population. *Nature* **346:** 357–359.

Richmond JQ, Jockusch EL. 2007. Body size evolution simultaneously creates and collapses species boundaries in a clade of scincid lizards. *Proceedings of the Royal Society of London Series B, Biological Sciences* **274:** 1701–1708.

Roff DA. 2002. *Life history evolution*. Sunderland, MA: Sinauer.

Schluter D. 1996. Ecological causes of adaptive radiation. *The American Naturalist* **148:** S40–S64.

Schluter D. 2000. *The ecology of adaptive radiation*. Oxford: Oxford University Press.

Simpson GG. 1953. *The major features of evolution*. New York, NY: Columbia University Press.

Slatkin M. 1982. Pleiotropy and parapatric speciation. *Evolution* **36:** 263–270.

Summers K, McKeon S, Sellars J, Keusenkothen M, Morris J, Gloeckner D, Pressley C, Price B, Snow H. 2003. Parasitic exploitation as an engine of diversity. *Biological Reviews* **78:** 639–675.

Taper ML, Case TJ. 1992. Models of character displacement and the theoretical robustness of taxon cycles. *Evolution* **46:** 317–333.

Taylor EB, McPhail JD. 2000. Historical contingency and ecological determinism interact to prime speciation in sticklebacks, Gasterosteus. *Proceedings of the Royal Society of London Series B, Biological Sciences* **267:** 2375–2384.

Taylor EB, McPhail JD. 1999. Evolutionary history of an adaptive radiation in species pairs of threespine sticklebacks (Gasterosteus): insights from mitochondrial DNA. *Biological Journal of the Linnean Society* **66:** 271–291.

Tierney JF, Huntingford FA, Crompton DWT. 1996. Body condition and reproductive status in sticklebacks exposed to a single wave of *Schistocephalus solidus* infection. *Journal of Fish Biology* **49:** 483–493.

Vamosi SM. 2005. On the role of enemies in divergence and diversification of prey: a review and synthesis. *Canadian Journal of Zoology* **83:** 894–910.

Vamosi SM, Schluter D. 2002. Impacts of trout predation on fitness of sympatric sticklebacks and their hybrids. *Proceedings of the Royal Society of London Series B, Biological Sciences* **269:** 923–930.

Vamosi SM, Schluter D. 2004. Character shifts in the defensive armor of sympatric sticklebacks. *Evolution* **58:** 376–385.