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Has the introduction of brown trout altered disease patterns in native New Zealand fish?

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SUMMARY

1. It is well recognised that non-indigenous species (NIS) can affect native communities via the 'spillover' of introduced parasites. However, two other potentially important processes, the 'spillback' of native parasites from a competent NIS host, where the latter acts as a reservoir leading to amplified infection in native hosts, and the 'dilution' of parasitism by a NIS host acting as a sink for native parasites, have either not been tested or largely overlooked.
2. We surveyed the helminth parasite fauna of native New Zealand fish in Otago streams that varied in the abundance of introduced brown trout *Salmo trutta*, to look for evidence of spillback and/or dilution. Spillover is not an issue in this system, with trout introduced as parasite-free eggs.
3. Seven native parasite species were present across 12 sites; significant inverse relationships with an index of trout abundance (i.e. dilution) were documented for three species infecting the native upland bully *Gobiomorphus breviceps*, and one species infecting the native roundhead galaxias *Galaxias anomalus*.
4. An inverse relationship between bully energy status and infection intensity of one parasite species suggests that parasite dilution could have positive effects on bully populations. Our failure to detect similar relationships for the other parasites does not preclude the possibility that dilution is beneficial to native fish, since parasites may have subtle or unmeasured impacts.
5. The parasite dilution patterns reported are compelling in that they occurred across several native host and parasite species; as such they have important implications for invasion ecology, providing an interesting contrast to the largely negative impacts reported for NIS. Mechanisms potentially responsible for the patterns observed are discussed.

Keywords: dilution effect, disease, introduced species, spillback, trout

Introduction

The loss of native biodiversity due to the introduction of non-indigenous species (NIS) is driven by a number of underlying mechanisms, of which predation and competition are most commonly invoked (Sax, Gaines & Brown, 2002; Clavero & Garcia-Berthou, 2005). However, ecologists and conservation managers have become increasingly aware that NIS can also threaten native species by altering disease dynamics (Daszak, Cunningham & Hyatt, 2000; Prenter *et al*., 2004). This can occur in several ways. First, the introduction and transmission of novel pathogens by NIS, or 'pathogen pollution', can have very serious consequences for native species (Holdich & Reeve, 1991; Roelke-Parker *et al*., 1996; Daszak *et al*., 2000; Tompkins, White & Boots, 2003). Second, NIS (including domestic stock) may act as reservoir hosts

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for 'spillover' infections to sympatric native wildlife, since NIS often attain sufficiently high densities to promote pathogen persistence (Daszak *et al*., 2000). Spillover differs from pathogen pollution because, in the former, the constant presence of an NIS could enhance the abundance of infective stages in native hosts whereas, in the latter, transmission and persistence could occur even if the NIS becomes locally extinct. A third potential mechanism is parasite 'spillback', in which NIS act as reservoirs of native parasites that spill back infection to native hosts (Tompkins & Poulin, 2006; Kelly *et al*., in press). Circumstantial evidence that spillback could pose a risk to native populations comes from observations that, while NIS tend to lose parasites from their region of origin during introduction (Torchin *et al*., 2003; MacLeod *et al*., 2008), they also tend simultaneously to acquire generalist parasites from the local fauna (Dobson & May, 1986; Poulin & Mouillot, 2003; Kelly *et al*., in press).

The risk of spillover or spillback infections to native hosts hinges on the retention or acquisition of parasites by NIS during introduction. However, the direction and strength of any impact on native host populations depends also on further conditions. First, NIS are likely to spillback infection to native species only if they are competent hosts for native parasites; that is, the parasite should be capable of establishment, survival and reproduction in the new host (Kelly *et al*., in press). If not, NIS might have an effect analogous to that of adding incompetent reservoir species to vector-borne disease systems, where models predict that the addition of incompetent hosts could dilute infection in focal hosts; these predictions are partly supported by negative correlations between species richness and infection for some multi-host systems of vector-borne diseases (Norman *et al*., 1999; Keesing, Holt & Ostfeld, 2006). In such cases, NIS might actually benefit native hosts by acting as 'sinks' that remove infective stages from the environment (e.g. Telfer *et al*., 2005; Kopp & Jokela, 2007). A second factor affecting the consequences of shared parasitism on native host populations is the extent to which infection affects host fitness. Although parasites may change host fitness by altering behaviour, reducing somatic condition, growth, fecundity or by causing direct mortality (e.g. Tompkins *et al*., 2002; Smith *et al*., 2008), amplification or dilution of infection may have little consequence for hosts if the infection is benign.

Non-indigenous fish provide useful models for assessing how NIS alter the disease dynamics of native species, because the introduced species can either acquire native parasites or introduce novel parasites, and such introductions are known to be associated with changes in native species demography and distribution (Font, 2003; Gilbert & Granath, 2003; Townsend, 2003). Furthermore, freshwater fish often occur in discrete, well defined habitats, where changes in host infection might be more apparent than in spatially extensive terrestrial systems. Here we investigate whether introduced brown trout (*Salmo trutta* L.) have altered disease patterns in native New Zealand fish. Although the negative impacts of trout on native stream fish populations through predation and competition have been well documented (see Townsend, 2003), we do not know whether they have also altered native host-parasite dynamics. In New Zealand, parasite spillover from trout can be ruled out since the species was introduced as eggs, free of metazoan parasites (Kennedy & Bush, 1994; Poulin & Mouillot, 2003). However, trout now harbour rich faunas of local generalist parasites (11 helminth parasites recorded; Dix, 1968; Hine, Jones & Diggles, 2000), so they may alter patterns of infection in native fish through spillback or dilution.

Here, we first assess the extent of acquisition of native helminth parasites by trout across stream sites of varying native fish communities. Second, we contrast parasitism of native fish species across the same sites, with varying levels of trout abundance, to assess the potential for infection amplification (spillback) or dilution by the invader. Third, we investigate the possible impacts of trout-altered parasitism in native fish, by relating host condition to infection intensity. Finally, we discuss possible mechanisms to account for observed differences in parasitism in native fish under varying levels of abundance of both trout and invertebrate intermediate hosts, and consider the implications for individual fitness of the native fish.

Methods

Fish sampling

The study was conducted in 'trout-invaded' and 'trout-free' stream sites within the Manuherikia and Taieri River catchments of Central Otago, South

Island, New Zealand (Table 1; Fig. 1). Sites were selected *a priori* as trout-free or trout-invaded using the New Zealand freshwater fish database (NZFFD) and from previous observations of native fish and trout distributions in Otago streams (Townsend & Crowl, 1991; Leprieur *et al*., 2006).

All sites were sampled in the austral winter (June–August 2007), except for the Swinburn which was also re-sampled in October. Sampling in winter allowed assessment of parasite accumulation over the previous spring–summer growth period for hosts, and coincided with high or increasing prevalence of infection in invertebrate intermediate hosts of several common fish parasites (see Lagrue & Poulin, 2007). The October sampling was performed in the Swinburn because, although trout were absent at site Q1 in June (Table 1), a few trout were observed at the site in October in a related study. Moreover, in June at this site the adults of an acanthocephalan worm, *Acanthocephalus galaxii* Hine, 1977, were prevalent in the native roundhead galaxias, *Galaxias anomalus* Stokell. Re-sampling allowed us to assess whether *A. galaxii* had been acquired by trout.

Fish were sampled by electrofishing in both riffle and pool sections of one stream reach (70–100 m length) at each study site (Table 1). All fish were preserved in 10% formalin with the number taken per site and catchment (see Table 2) restricted by permitting regulations of the Department of Conservation and New Zealand Fish and Game.

Intermediate host sampling

To determine the presence and relative abundance of invertebrate intermediate hosts of the most common parasites of New Zealand fish, a 3–5 min kick-sample (using a D-frame pole net) was taken at each site, sampling riffle and pool habitats along the same reach as surveyed for fish. Sampling proceeded in an upstream diagonal transect from stream edge to stream edge with greater sampling effort apportioned to the most common habitat types. All samples were preserved in 10% formalin.

Two known intermediate hosts of New Zealand freshwater fish parasites were counted from each kick-sample, the gastropod *Potamopyrgus antipodarum* (Gray, 1853) (intermediate host for the trematodes *Stegodexamene anguillae* Macfarlane, 1951, *Telogaster opisthorchis* Macfarlane, 1945 and *Apatemon* spp.) and the amphipod *Paracalliope fluviatilis* Chilton, 1909 (intermediate host for the acanthocephalan *A. galaxii* and the trematode *Coitocaecum parvum* Crowcroft, 1945). Note that *P. antipodarum* is also an intermediate host for *C. parvum*, although transmission to fish occurs via the amphipod.

Table 1 Site characteristics for the fish parasite survey of streams in the Manuherikia and Taieri catchments in winter 2007

Site/code	Fish community type	Date	Reach width (m)	Reach length (m)	NZ grid E	NZ grid N	Fish abundance		
							Trout	Bully	*Galaxias*
	Manuherikia								
Station Ck.	Sympatric (Bully)	June	2	80	22°61′100	55°83′800	6	25	–
Mata Ck.	Sympatric (Bully)	Aug.	3	80	22°60′800	55°83′640	4	14	–
McCleans	Sympatric (Bully)	Aug.	1.5	80	22°40′415	55°64′317	3	20	–
Becks	Sympatric (Bully)	June	1.5	70	22°52′635	55°73′016	5	20	–
Miller	Sympatric (Bully)	June	1.2	70	22°50′000	55°74′500	10	35	–
Lauder	Sympatric (Bully)	June	4.5	100	22°48′800	55°74′500	2	35	–
Chatto	Sympatric (Bully)	June	1.5	80	22°35′730	55°65′892	1	40	–
Ida	Sympatric (Galaxias)	June	2.5	80	22°54′900	55°66′600	15	–	18
	Taieri								
Hound	Trout-free (Galaxias and bully)	Aug.	1.5	70	22°98′735	55°53′055	–	7	27
Old Hutt	Sympatric (Galaxias)	June	2.5	70	22°94′000	55°68′734	27	–	21
Camp Ck.	Sympatric (Galaxias)	Aug.	1.75	80	22°97′704	55°67′874	1	–	41
Swinburn Q1	Trout-free (Galaxias only)	June	3	80	22°96′900	55°58′700	–	–	28
Swinburn Q1	Sympatric (Galaxias)	Oct.					6	–	65
Swinburn Q2	Sympatric (Galaxias)	Oct.	2.5	80	22°98′300	55°59′400	22	–	40

Sites are either ‘trout-free’ (native fish only) or ‘sympatric’ (i.e. native fish co-occur with trout). Sites in which trout are sympatric with native fish are denoted as having either upland bully (*Gobiomorphus breviceps*) or roundhead galaxias (*Galaxias anomalus*) present but not both species with trout (see also location map in Fig. 1).

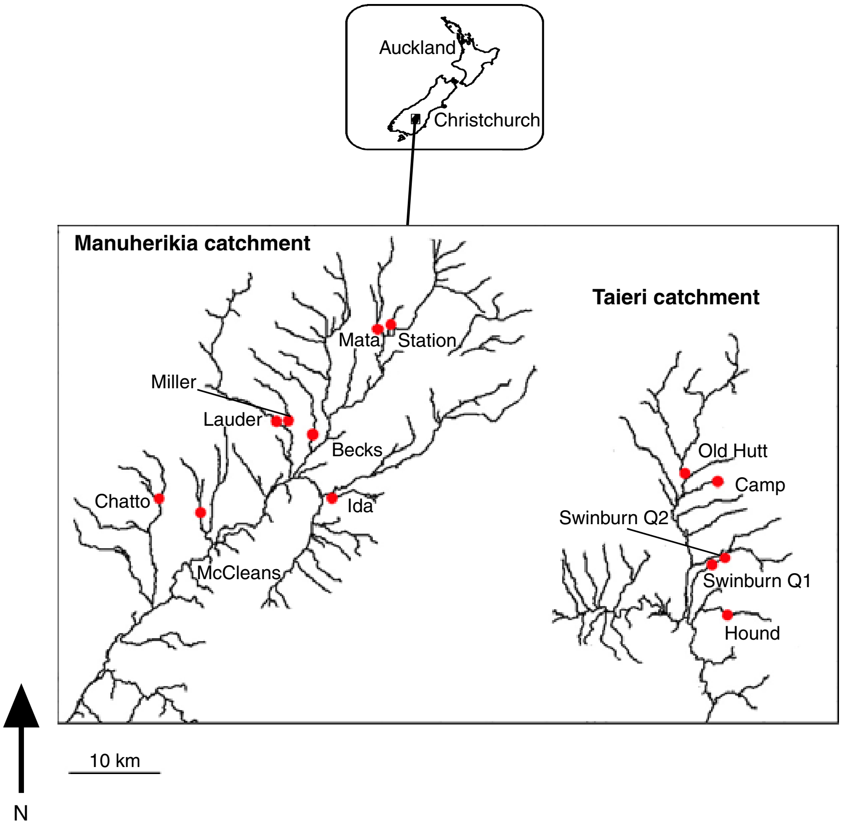

Fig. 1 Map showing locations of stream sites in the study area (see Table 1).

Host autopsies

The length and wet mass (blotted dry with tissue paper) of each fish were recorded, followed by removal of the gut and internal organs and weighing of the liver. Endoparasites were identified and counted by gut dissection, systematic screening of all internal organs and compression of thin pieces of tissue between glass plates. Screening and counting of metacercarial cysts in larger tissue was conducted following a pepsin digestion of host tissue in a water bath (6 g pepsin in 7 mL 10 M HCl + 1 L of distilled water at 40 °C for 4 h; see McFarland, Mouritsen & Poulin, 2003). Parasite identification was based on previous descriptions of the parasite fauna of New Zealand freshwater fish (see MacFarlane, 1945, 1951; Hine, 1977a,b; Holton, 1983, 1984). Infection parameters were expressed as prevalence, mean abundance or mean intensity, according to Bush *et al.* (1997).

Parasitism may affect fish population dynamics strongly without causing mortality, because infection can reduce allocation of energy resources to growth and reproduction and decrease lifetime fitness (Dobson & May, 1987; Tompkins *et al.*, 2002). Therefore, we calculated body condition (K) and the hepatosomatic index (HSI), for each fish, to provide surrogate measures of the energy reserves available for growth and reproduction (Chellappa *et al.*, 1995). HSI was calculated as liver mass/body mass, and the coefficient of body condition K as:

$$K = \frac{100 \times \text{weight (g)}}{\text{length(cm)}^3}$$

(see Lemly & Esch, 1984 and Chellappa *et al.*, 1995). Both indices were calculated using host body and liver mass corrected for parasite mass (e.g. fish body mass – mass of all metacercariae), because parasite cysts often account for a high proportion of fish weight. Mean cyst mass for each parasite species was obtained from estimates of cyst volume. Average cyst volume was calculated from mean length and width measures of 20 cysts per parasite species, using the formula for an ellipsoid:

$$(4/3)\pi L R^2$$

where L is half the length of the cyst and R is half the width of the cyst. Average cyst volume was then

Table 2 Parasites of bully, galaxias and trout in stream sites of the Manuherikia and Taieri catchments

Site/code	*n*	Metacercariae			Adult trematodes				Acanthocephala
		Stegodexamene anguillae	*Telogaster opisthorchis*	*Apatemon* spp.	*T. opisithorchis*	*Coitocaecum parvum*	*Deretrema minutum*	*Deretrema philippae*	*Acanthocephalus galaxii*
Trout									
Station	6	0	0	0	0	0	0	0	0
Mata	7	0	0	0	14.3% 2.4	0	0	0	0
McCleans	3	0	33.3% 0.3	0	0	0	0	0	0
Becks	5	0	0	0	0	0	0	0	0
Miller	4	0	0	0	0	0	0	0	0
Lauder	1	0	0	0	0	0	0	0	0
Chatto	1	0	0	0	0	0	0	0	0
Ida	4	0	25% 0.75	0	0	0	0	0	0
Old Hut	10	0	0	0	0	0	0	0	0
Swin. Q1 Oct.	6	0	0	0	0	0	0	0	83.3% 10.4
Swin. Q2 Oct.	14	0	0	0	0	0	0	0	85.7% 9
Across site mean		0	5.3% 0.1	0	1.3% 0.2	0	0	0	15.35% 1.75
Bully									
Station	9	0	22% 0.33	88% 17.9	0	11.1% 0.22	0	0	0
Mata	5	100% 1.2	40% 1.4	60% 16.2	0	80% 3.4	0	0	0
McCleans	8	100% 35.6	100% 37. 5	100% 225	0	100% 14	0	0	0
Becks	10	60% 8.1	90% 14.8	100% 125	0	0	0	0	0
Miller	9	66.6% 2.8	88.8% 11.5	100% 237	0	11.1% 0.22	0	0	0
Lauder	8	62.5% 1.6	0	25% 0.5	0	0	0	0	0
Chatto	13	84.6% 30.1	84.6% 23	84.6% 3.6	0	83.3% 3.9	0	0	0
Hound	3	0	100% 176	100% 29.3	0	0	0	0	0
Across site mean		59.2% 9.9	65.6% 33.3	69.7% 81.8	0	35.7% 2.7	0	0	0
Galaxias									
Ida	6	100% 8.2	33.3% 7.3	0	0	0	33.3% 0.8	16.6% 0.33	0
Hound	4	100% 27.7	100% 138	0	0	25% 0.5	50% 1	0	0
Old Hut	10	100% 39.3	100% 21.5	10% 0.1	0	0	33.3% 7.4	22.2% 0.22	11.1% 0.11
Camp Ck.	6	100% 24.5	100% 105.2	16.6% 1.6	0	0	50% 25.3	16.6% 0.17	0
Swin. Q1 June	10	90% 7.3	40% 3.1	0	0	0	90% 3.7	40% 1	40% 1.1
Q1 Oct.	27	100% 11.2	92.6% 4.4	0	0	0	96.3% 6	66.6% 1.9	40.7% 2.7
Q2 Oct.	20	100% 38.8	75% 2.6	0	0	0	85% 2.3	40% 0.75	80% 9
Across site mean		98.5% 22.4	77.3% 40.3	3.8% 0.24	0	3.6% 0.07	62.5% 6.6	28.8% 0.62	24.4% 1.8

Parameters of infection for parasite species/life stage and site are given as prevalence and mean abundance respectively.

converted to mass assuming that cyst density was equal to that of distilled water (i.e. 1 μL = 1 mg).

Data analyses

Fish length–mass relationships were analysed in STATISTICA 6.0 (Stat Soft, Tulsa, OK, U.S.A.) using linear regression ($\log_{10}$ transformed). To identify those factors accounting for variation in infection intensity across sites, we constructed generalised linear mixed models (GLMMs) using the open-source software *R* Package (R Development Core Team 2008; version 2.7.1). Patterns of infection in native fish could change with increasing fish size, since many fish accumulate parasites with age (Poulin, 2000), and the size structure of native fish might also be altered by the presence of trout (McIntosh, Crowl & Townsend, 1994); this was investigated by analysing the infection intensity of each parasite with respect to fish size (mass and length), pooling data across sites (but with sites of zero infection excluded).

To investigate whether trout alter disease patterns in native fish, separate GLMMs were constructed using intensity of infection [$\log_{10}$ or $\log_{10}(x + 1)$ transformed] of each parasite species as the dependent variable. Predictor variables for each GLMM analysis were:

1 An index of trout abundance, namely the ratio of the number of trout to the number of native fish caught per site. This index was used as a coarse predictor of infection in native fish under the following assumptions: if trout are incompetent hosts, an increase in the ratio could dilute infection in native fish whereas, if trout are competent hosts, an increase in the ratio could amplify infection in native fish.

2 Total fish abundance (trout + native species); this was used as a second predictor variable since the number of available hosts might also influence *per capita* infection intensity (see Fauchald *et al*., 2007); all fish species were included in the calculation of abundance since we assumed, under a null hypothesis, that infection of trout and native fish was equally likely.

3 Relative abundance of the intermediate host from which the fish acquire infections was the third predictor variable; any differences in fish infection across sites could reasonably be ascribed to changes in intermediate host abundance and the supply of infective stages (e.g. Thieltges & Reise, 2007).

To control for non-independence, and avoid pseudoreplication, each GLMM included 'fish within site' and 'fish body mass' (used as a measure of fish body size, see Results) as random effects (Crawley, 2002). Model fitting involved specification of a maximal model where all terms were included, followed by successive removal of unimportant terms (Crawley, 2002). The importance of each term was assessed using log-likelihood ratio tests that compared models before and after removal of a given term, which resulted in the latter's omission if it was non-significant. Final model selection was based on hierarchical ordering using the Akaike information criterion (AIC), with the best model having the lowest AIC score. If the best model (lowest AIC score) contained a term that was not significant at the $P = 0.05$ level, however, the model with the next lowest AIC and significant term was chosen (Crawley, 2002).

Sites of zero parasite prevalence were excluded from all analyses because we could not determine whether parasite absence was due to historical absence from the site or to environmental factors. To investigate associations between infection and host morbidity, regression analyses were conducted between the intensity of infection (predictor) and body condition and HSI (separate response variables).

Results

Roundhead galaxias, *G. anomalus*, was sympatric with trout in five of the 13 sites, four from the Taieri catchment and one from the Manuherikia catchment (Table 1). The native upland bully, *Gobiomorphus breviceps* Stokell, 1939, was sympatric with trout in seven of the 13 sites, all of which occurred within the Manuherikia catchment. Two stream sites were trout-free; the Hound burn, which was the only site in which bully and galaxiids occurred in sympatry, and Swinburn Q1, where only galaxiids were present in June, but where trout also occurred with *G. anomalus* during re-sampling in October (Table 1).

Parasite surveys

Seven species of parasitic helminths native to New Zealand (six trematodes and an acanthocephalan) were documented in the fish communities of these Otago streams (Table 2).

Roundhead galaxias. Roundhead galaxias were infected with all seven parasite species. The acanthocephalan *A. galaxii* was present in the gut, occurring at low to high prevalence and low to high abundance at three of the six galaxiid sites. Three trematode species were detected as adults and three as larval cysts (Table 2). A single adult *C. parvum* was found, whereas adults of the intestinal trematode *Deretrema minutum* Manter, 1954, reached moderate to high prevalence and abundance in all sites and adults of the gall bladder parasite *Deretrema philippae* Hine, 1977, occurred at low to moderate prevalence and abundance in five of the six sites. Galaxias were rarely infected by *Apatemon* spp. cysts, but were infected at low to high abundance in all sites by cysts of both *S. anguillae* and *T. opisthorchis*.

Upland bully. Bullies were parasitised by four species of trematode (Table 2); *C. parvum* occurred in the intestine, whereas larval cysts of *S. anguillae* and *T. opisthorchis* occurred in the body. Unlike roundhead galaxias, cysts of *Apatemon* spp., which infect birds as the definitive host, were common in bullies. Mean abundance of infection ranged from low to moderate across sites for *S. anguillae*, low to high for *T. opisthorchis* and was highly variable for *Apatemon* spp., which accounted for the highest intensities of infection of any trematode in any host (Table 2).

Brown trout. In contrast to the native fish, parasites of trout were rare except at specific sites. Adult *A. galaxii* were highly prevalent and moderately abundant in trout from the Swinburn Q1 and Q2 sites in October 2007; roundhead galaxias co-occurred in both sites and were also infected by *A. galaxii*. One trout from McClean's and one from Ida Creek were infected by one and three cysts of *T. opisthorchis*, respectively, in contrast to much higher rates of infection in native fish at both sites (Table 2). A single trout from Mata Creek was infected by adult worms of *T. opisthorchis*, despite the high prevalence of this parasite as cysts in native fish sympatric with trout.

Infection of native fish in relation to trout presence

Body mass regressions versus fish length were significant for both roundhead galaxias and bullies ($r = 0.9$ and 0.94, respectively, both $P < 0.0001$), with non-significant Durbin–Watson statistics indicating that linear models were the appropriate fit to the data (Fry, 1993). This validates the use of fish body mass as a random effect in the GLMMs examining relationships with parasite intensity.

Roundhead galaxias. Although models were not fitted to intensity of infection data for *A. galaxii*, *S. anguillae* or *D. minutum*, *T. opisthorchis* intensity was negatively related to intermediate host abundance (snails), whereas *D. philippae* intensity was negatively related to the trout index (Table 3; Fig. 2).

Upland bully. Although intermediate host density (snails) was positively related to intensity for two of the four parasite species infecting bullies, the trout index was negatively related to the intensities of cysts of *S. anguillae* and *T. opisthorchis* (Table 3; Fig. 3a,b). Although a model could not be fitted to the data, a simple linear regression showed that *C. parvum* infection intensity was also inversely related to the trout index ($r = -0.37$, $P < 0.05$; Table 3).

Intensity of infection and host condition

Roundhead galaxias. Hepatosomatic index was the only index related to infection in *G. anomalus*, being positively related to *A. galaxii* and *S. anguillae* intensity (Table 4; Fig. 4a,b).

Upland bully. Bully body condition was positively related to *Apatemon* spp. and *S. anguillae* intensities (Table 4). In contrast, *T. opisthorchis* intensity was negatively related to the HSI, but unrelated to bully body condition (Table 4; Fig. 5).

Brown trout. Lack of infection in trout meant that regression analysis was conducted only for *A. galaxii*, for which intensity was unrelated to indices of fish health (Table 4).

Discussion

Pathogen pollution and spillover are often considered mechanisms by which NIS can alter parasitism of native hosts (Daszak *et al.*, 2000; Power & Mitchell, 2004). While infection dilution and parasite spillback may also be important, infection dilution has received less attention (Keesing *et al.*, 2006) and spillback has not been discussed in the context of invasion ecology

Table 3 Generalised linear mixed models examining the relationships between environmental factors and infection intensity in bully and galaxias

Native host	Parasite species	Candidate models	AIC	d.f.	Variable (s)	Estimate	SE	*P*-value
Bully	*Apatemon* spp.	T.N + P.anti + fish	85.1	7				
		P.anti + fish	83.3	6				
		P.anti	82.5	5	P.anti	0.48	0.1	<0.005
	Stegodexamene anguillae	T.N + P.anti + fish	89.7	7	T.N	−4.88	0.99	<0.05
					P.anti	0.28	0.06	<0.05
					Fish	−2.2	0.64	NS
		T.N + P.anti	95.4	6				
		T.N + fish	97.3	6				
		Fish + P.anti	98.2	6				
	Telogaster opisthorchis	T.N + P.anti + fish	93.3	7				
		T.N + P.anti	93.28	6				
		T.N	94.9	5	T.N	−4.2	1.6	<0.05
	Coitocaecum parvum	Not fitted						
Galaxias	*Acanthocephalus galaxii*	Not fitted						
	S. anguillae	Not fitted						
	T. opisthorchis	T.N + P.anti + fish	36.7	7				
		P.anti + fish	34.7	6				
		P.anti	35.7	5	P.anti	−0.39	0.18	<0.05
	Deretrema minutum	Not fitted						
	Deretrema philippae	T.N + fish	17.67	6				
		T.N	16.1	5	T.N	−0.27	0.07	<0.05

To control for non-independence of data, 'site' and 'fish body weight' were included as random effects. For each response variable, the parameter estimates for the best model are indicated.

AIC, Akaike information criterion; T.N, Trout index; P. anti = *Potamopyrgus antipodarum*; fish, total fish abundance.

(Kelly *et al.*, in press). Each mechanism is dependent primarily on parasite origin and host competence for the parasite (e.g. de Castro & Bolker, 2005; Keesing *et al.*, 2006; Kelly *et al.*, in press). In Otago stream fish communities, two native helminth parasites were observed to infect brown trout: *A. galaxii* in the intestine and *T. opisthorchis* as body cysts and adult worms. This raises the possibility that infection of native fish could either be decreased by the dilution of native parasites by the introduced trout or increased by spillback of native parasites from trout. The relationships between the ratio trout : native fish and native fish infection levels lend support to the hypothesis that introduced brown trout are diluting infection by some native parasites in the native freshwater fish, because significant inverse relationships were observed between intensity of infection and increasing trout presence (higher ratio) for three parasites in bully (two using the GLMMs, and one using simpled linear regression) and one in roundhead galaxias. On the other hand, the results lend no support to the 'parasite spillback' hypothesis, because there was no sign that an increase in the ratio was associated with amplification of infection in native fish. This is not surprising in the case of *T. opisthorchis*, which occurred only in low numbers in trout. In contrast, *A. galaxii* occurred at similar or higher prevalence and intensity in trout as compared to sympatric roundhead galaxias, and could thus potentially spillback infection since adult worms commonly infect fish of the genus *Galaxias* at high intensity (Hine, 1977a,b). Hence, an investigation of *A. galaxii* fitness when infecting the different host species is required to elucidate why spillback was not observed.

Although ecologists have recognised that increasing species diversity could reduce disease risk (e.g. Keesing *et al.*, 2006), only two studies have addressed the phenomenon in an invasion context. Field patterns

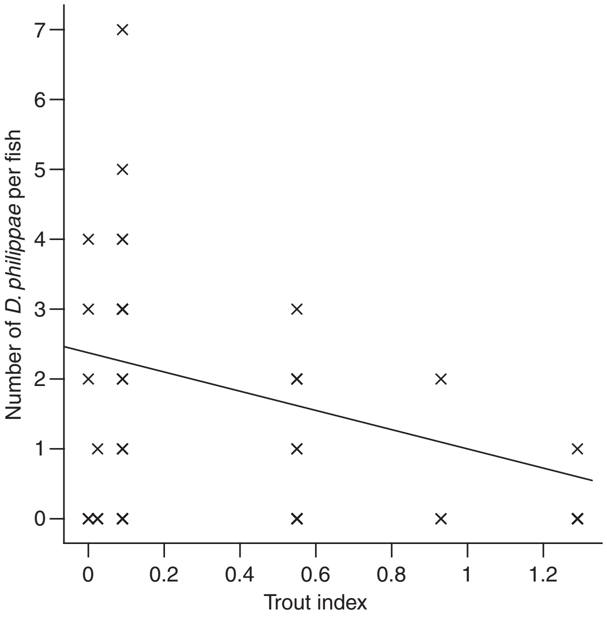

Fig. 2 Relationship between infection intensity of *Deretrema philippae* in *Galaxias anomalus*, and the index of trout abundance. The line represents the linear relationship predicted by regression.

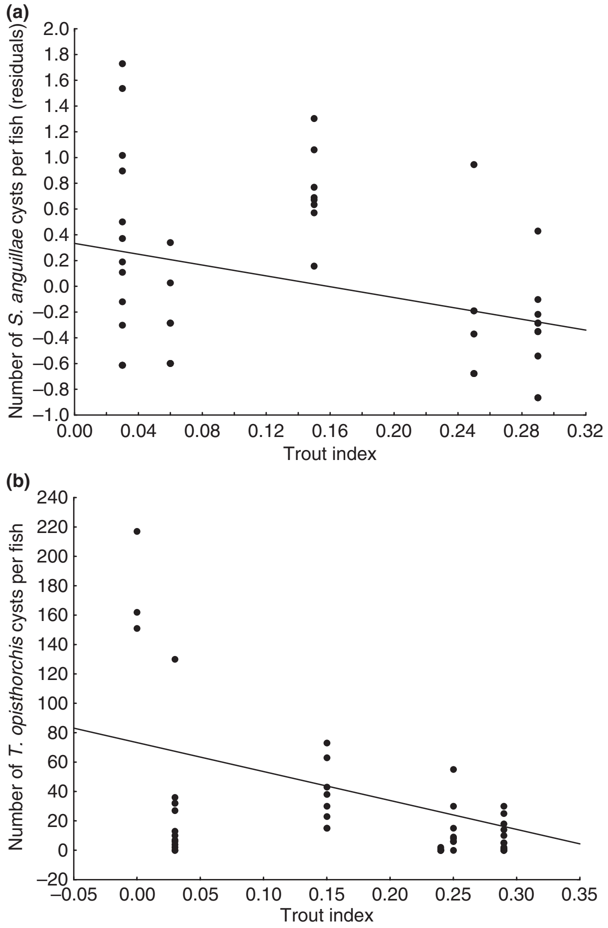

Fig. 3 Relationship between infection intensity of (a) *Stegodexamene anguillae* and (b) *Telogaster opisthorchis* cysts in *Gobiomorphus breviceps* and the index of trout abundance. Note that in (a) the *y*-axis represents partial residuals and thus accounts for the independent effects of *Potamopyrgus antipodarum* (see Table 3).

showed a negative relationship between the density of introduced bank voles *Clethrionomys glareolus* Schreber and infection by the bacterium *Bartonella* in the native wood mouse *Apodemus sylvaticus* L. (Telfer *et al.*, 2005). As an incompetent host for the parasite, but a competent one for the parasites' flea vector, the vole is hypothesised to dilute infection by disrupting mouse – flea encounter rates (Telfer *et al.*, 2005). Only a single study has claimed invader-mediated dilution of infection in a macroparasite system like ours, but without evidence for dilution in the field. In laboratory microcosms, Kopp & Jokela (2007) showed lower prevalence of cysts of the native trematode *Microphallus* spp. in the native gastropod *P. antipodarum* when sympatric with the incompetent, introduced gastropod *Lymnea stagnalis* (L.). Dilution was hypothesised to be caused by indiscriminate feeding by the invader on trematode eggs, thereby reducing egg – native host encounter rates (Kopp & Jokela, 2007). The pattern of dilution observed in our Otago stream fish is remarkable in that it occurred across multiple native hosts and parasite species with complex life-cycles. In a conceptual model that assessed the relationship between species diversity and disease risk, Keesing *et al.* (2006) proposed several mechanisms that could account for dilution of infectious diseases. While they argued that the model was generally applicable, additional mechanisms were overlooked by their omission of macroparasite systems. We propose several hypotheses that could account for the observed patterns of infection dilution in native stream fish, noting their dependence on parasite mode of transmission.

Encounter-reduction, where an added host species reduces the probability of encounters between infected and susceptible focal hosts, can reduce disease risk (Keesing *et al.*, 2006). Consistent with this idea, introduced trout may be incompetent physical 'sinks' that reduce bully encounters with cercariae of *T. opistorchis* or *S. anguillae shed by P. antipodarum.* This hypothesised mechanism is analogous to reduced transmission of vector-borne diseases by incompetent

Table 4 Regression analyses examining the relationships between infection intensity and indices of fish health in bully, galaxias and trout

Native host	Parasite species	Variable	d.f.	*t*	*r*	*P*-value
Bully	*Apatemon* spp.	Body condition factor	1,41	4.76	0.59	<0.0001
		HSI	1,41	−0.61	0.095	NS
	Stegodexamene anguillae	Body condition factor	1,34	2.47	0.39	<0.05
		HSI	1,34	0.91	0.15	NS
	Telogaster opisthorchis	Body condition factor	1,36	0.45	0.07	NS
		HSI	1,36	−2.1	−0.34	<0.05
	Coitocaecum parvum	Body condition factor	1,30	0.59	0.1	NS
		HSI	1,30	1.44	0.25	NS
Galaxias	*Acanthocephalus galaxii*	Body condition factor	1,61	−0.73	0.09	NS
		HSI	1,61	3.25	0.38	0.01
	S. anguillae	Body condition factor	1,74	1.25	0.14	NS
		HSI	1,74	6.28	0.59	<0.0001
	T. opisthorchis	Body condition factor	1,74	1.88	0.21	NS
		HSI	1,74	0.92	0.1	NS
	Deretrema minutum	Body condition factor	1,74	0.17	0.019	NS
		HSI	1,74	−0.01	0.002	NS
	Deretrema philippae	Body condition factor	1,74	−1.1	0.12	NS
		HSI	1,74	0.36	0.04	NS
Trout	*A. galaxii*	Body condition factor	1,18	1.27	0.06	NS
		HSI	1,18	0.21	0.02	NS

HSI, hepatosomatic index.

hosts that lure vectors away from focal hosts (Osfield & Keesing, 2000; Telfer *et al*., 2005). Although cysts of *T. opistorchis* and *S. anguillae* were rare or absent in trout sympatric with infected native fish, encounters between trout and cercariae were likely since trout and *P. antipodarum* use both riffle and pool habitats in streams (D. Kelly, pers. obs.). Two additional mechanisms could also account for infection dilution; trout predation on heavily infected native fish, and trout as an alternative prey for shortfin (*Anguilla australis australis* Richardson, 1841) and longfin eels (*A. dieffenbachii* Gray, 1842), both of which would lead to decreased transmission of parasite infective stages. However, whatever the mechanism, the question remains of why the pattern of infection dilution of *T. opistorchis* and *Stegodexamne* observed in bully is not apparent in roundhead galaxias. One possible cause of this disparity could be the differential behavioural responses of bully and galaxias to trout, resulting in changes in the magnitude of cercariae-fish encounters. While impacts of trout on galaxiids have been well documented, evidence of impacts on bullies is lacking (McIntosh *et al*., 1994; McIntosh, 2000). Trout compete more with galaxiids than bullies for habitat and food, displacing galaxiids from favourable riffle microhabitats and reducing coexistence at small spatial scales (Cadwallader, 1975; McIntosh, Townsend & Crowl, 1992; McIntosh *et al*., 1994). Thus, the dilution of *T. opistorchis* and *S. anguillae* infections in bullies, but not galaxias, may reflect lower spatial segregation of trout with bullies, thereby augmenting infection dilution.

A different mechanism is likely to explain infection dilution of *D. philippae* in roundhead galaxias. Although the parasite's invertebrate intermediate host is unknown, transmission will involve fish predation on infected macroinvertebrates, in much the same way as all other species in the genus *Deretrema* (Holton, 1983). Thus, a possible mechanism of infection dilution, not considered by Keesing *et al.* (2006), is that introduced trout disrupt transmission of parasites by decreasing encounter rates between native fish and intermediate hosts. This is plausible because trout reduce diurnal activity and abundance of invertebrates, and competitively exclude galaxiids from riffles where invertebrate drift is abundant (McIntosh *et al*., 1992; Townsend, 2003).

Although modelling studies indicate that infection dilution associated with invasion could be of benefit to native hosts (Heimpel, Neuhauser & Hoogendoorn, 2003), this depends on the effect of parasitism on host fitness. The only field survey indicating infection dilution with invasion (Telfer *et al*., 2005) did not assess host impacts of parasitism, and so the potential benefits of dilution are unknown. However, of the parasites associated with dilution of infection in our Otago streams, only one health index-infection pattern suggests a potential benefit of dilution; the intensity of *T. opistorchis* cysts in bullies was negatively related to the HSI. Since HSI often reflects energy status, which correlates with growth and survival (Chellappa *et al*., 1995), trout dilution of infection may have positive effects on bully fitness. That the majority of health status versus infection intensity relationships were either positive (four comparisons) or non-significant (five out of eight comparisons for bully; eight out of 10 for roundhead galaxias) may be explained as follows: the parasite species were relatively benign (non-significant relationships); hosts in better condition tolerated greater infection or consumed a greater number of infected intermediate hosts (positive relationships; Lafferty, 1992); we failed to detect more subtle effects parasitism (e.g. Lemly & Esch, 1984; Poulin, 1993; Johnson & Dick, 2001).

An important implication of this and similar studies (e.g. Telfer *et al*., 2005; Kopp & Jokela, 2007) is that infection dilution has the potential to redress the balance of the often reported negative impacts of NIS on native biodiversity, illustrated here by the negative correlations between trout abundance and native fish infection observed for multiple parasites across two different host species. In the context of trout invasions, our findings provide a potentially interesting contrast to the largely negative impacts reported to date. In New Zealand streams, trout impacts vary with trout size (see McIntosh, 2000), and it could be predicted that the mechanism and direction of trout-induced disease alteration will vary accordingly. For example, when trout are small, they might actually have positive effects on native fish through disease dilution whereas, as trout size increases, spillback (infection amplification) may occur. For instance, transmission to the adult stage for parasite cysts of *T. opistorchis* and *S. anguillae* in bully and galaxias requires their predation by larger definitive hosts (e.g. native eel), although gape-limitation of the small trout in our study probably precluded this. Future work involving experimental infection challenges is required to identify the mechanisms responsible for observed patterns of dilution, and to give a clear assessment of the potential benefits of reduced parasitism for native fish. Comparison of native host parasitism across systems in which trout vary in their potential for piscivory (i.e. size), may also direct future tests of the role of spillback versus dilution in biological invasions.

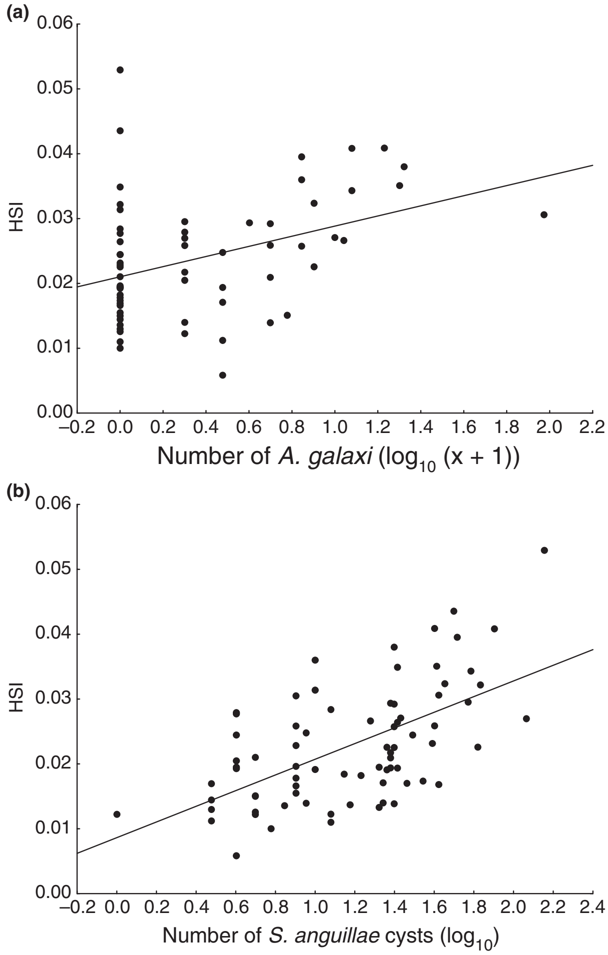

Fig. 4 Relationship between hepatosomatic index (HSI) of *Galaxias anomalus* and (a) the intensity of infection by *Acanthocephalus galaxii* and (b) *Stegodexamene anguillae*.

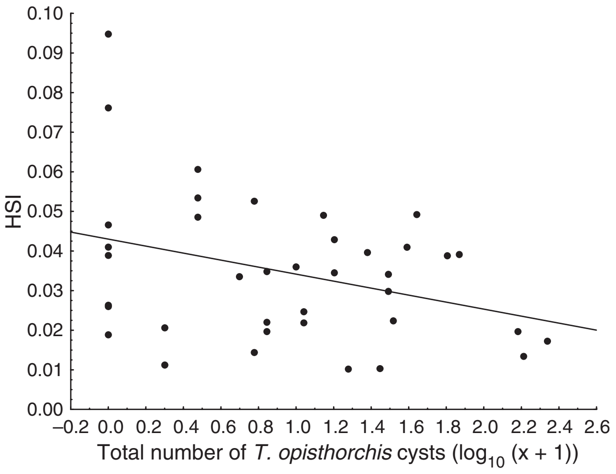

Fig. 5 Relationship between HSI and the intensity of *Gobiomorphus breviceps* infection by *Telogaster opisthorchis* cysts.

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References

Bush A.O., Lafferty K.D., Lotz J.M. & Shostak A.W. (1997) Parasitology meets ecology on its own terms: Margolis *et al*. revisited. *The Journal of Parasitology*, **83**, 575–583.

Cadwallader P.L. (1975) Feeding relationships of galaxiids, bullies, eels and trout in a New-Zealand river. *Australian Journal of Marine and Freshwater Research*, **26**, 299–316.

de Castro F. & Bolker B. (2005) Mechanisms of disease-induced extinction. *Ecology Letters*, **8**, 117–126.

Chellappa S., Huntingford F.A., Strang R.H.C. & Thomson R.Y. (1995) Condition factor and hepatosomatic index as estimates of energy status in male 3-spined stickleback. *Journal of Fish Biology*, **47**, 775–787.

Clavero M. & Garcia-Berthou E. (2005) Invasive species are a leading cause of animal extinctions. *Trends in Ecology and Evolution*, **20**, 110.

Crawley M.J. (2002) *Statistical Computing: An Introduction to Data Analysis Using S-Plus*. John Wiley & Sons Ltd, Chichester.

Daszak P., Cunningham A.A. & Hyatt A.D. (2000) Emerging infectious diseases of wildlife – threats to biodiversity and human health. *Science*, **287**, 443–449.

Dix T.G. (1968) Helminth parasites of brown trout (*Salmo trutta* L.) in Canterbury, New Zealand. *New Zealand Journal of Marine and Freshwater Research*, **2**, 363–374.

Dobson A.P. & May R.M. (1986) Patterns of invasion by pathogens and parasites. In: *The Ecology of Biological Invasions of North America and Hawaii* (Eds H.A. Mooney & J.M. Drake), pp. 58–76. Springer-Verlag, New York.

Dobson A.P. & May R.M. (1987) The effects of parasites on fish populations – theoretical aspects. *International Journal for Parasitology*, **17**, 363–370.

Fauchald P., Rolf Rødven R., Bardsen B.-J., Langeland K., Tveraa T., Yoccoz N.G. & Ims R.A. (2007) Escaping parasitism in the selfish herd: age, size, and density-dependent warble fly infestation in reindeer. *Oikos*, **116**, 491–499.

Font W.F. (2003) The global spread of parasites: what do Hawaiian streams tell us? *BioScience*, **53**, 1061–1067.

Fry J.C. (1993) *Biological Data Analysis: A Practical Approach*. Oxford University Press, Oxford, New York, Tokyo.

Gilbert M.A. & Granath W.O. (2003) Whirling disease of salmonid fish: life cycle, biology, and disease. *Journal of Parasitology*, **89**, 658–667.

Heimpel G.E., Neuhauser C. & Hoogendoorn M. (2003) Effects of parasitoid fecundity and host resistance on indirect interactions among hosts sharing a parasitoid. *Ecology Letters*, **6**, 556–566.

Hine P.M. (1977a) Two new digenean trematodes from New Zealand freshwater fishes. *Journal of the Royal Society of New Zealand*, **7**, 163–170.

Hine P.M. (1977b) *Acanthocephalus galaxii* n.sp. parasitic in *Galaxias maculatus* (Jenyns, 1842) in the Waimeha stream, New Zealand. *Journal of the Royal Society of New Zealand*, **7**, 51–57.

Hine P.M., Jones J.B. & Diggles B.K. (2000) A checklist of the parasites of New Zealand fishes, including previously unpublished records. *NIWA Technical Report*, **75**, 1–94.

Holdich D.M. & Reeve I.D. (1991) Distribution of freshwater crayfish in the British-Isles, with particular reference to crayfish plague, alien introductions and water-quality. *Aquatic Conservation – Marine and Freshwater Ecosystems*, **1**, 139–158.

Holton A.L. (1983) Observations on the life-history of *Deretrema minutum* Manter, 1954 (Digenea, Zoogonidae) in fresh-water crustacean and fish hosts from Canterbury, New-Zealand. *New Zealand Journal of Marine and Freshwater Research*, **17**, 373–376.

Holton A.L. (1984) A redescription of *Coitocaecum parvum* Crowcroft, 1945 (Digenea, Allocreadiidae) from crustacean and fish hosts in Canterbury. *New Zealand Journal of Zoology*, **11**, 1–7.

Johnson M.W. & Dick T.A. (2001) Parasite effects on the survival, growth, and reproductive potential of yellow perch (*Perca flavescens* Mitchill) in Canadian Shield lakes. *Canadian Journal of Zoology*, **79**, 1980–1992.

Keesing F., Holt R.D. & Ostfeld R.S. (2006) Effects of species diversity on disease risk. *Ecology Letters*, **9**, 485–498.

Kelly D.W., Paterson R.A., Townsend C.R., Poulin R. & Tompkins D.M. (in press) Parasite spillback: a neglected concept in invasion ecology? *Ecology*.

Kennedy C.R. & Bush A.O. (1994) The relationship between pattern and scale in parasite communities: a stranger in a strange land. *Parasitology*, **109**, 187–196.

Kopp K. & Jokela J. (2007) Resistant invaders can convey benefits to native species. *Oikos*, **116**, 295–301.

Lafferty K.D. (1992) Foraging on prey that are modified by parasites. *American Naturalist*, **140**, 854.

Lagrue C. & Poulin R. (2007) Life cycle abbreviation in the trematode *Coitocaecum parvum*: can parasites adjust to variable conditions? *Journal of Evolutionary Biology*, **20**, 1189–1195.

Lemly A.D. & Esch G.W. (1984) Effects of the trematode *Uvulifer ambloplitis* on juvenile bluegill sunfish, *Lepomis macrochirus*: ecological implications. *Journal of Parasitology*, **70**, 475–492.

Leprieur F., Hickey M.A., Arbuckle C.J., Closs G.P., Brosse S. & Townsend C.R. (2006) Hydrological disturbance benefits a native fish at the expense of an exotic fish in a New Zealand river catchment. *Journal of Applied Ecology*, **43**, 930–939.

MacFarlane W.V. (1945) The life-cycle of the heterophyoid trematode *Telogaster opisthorchis* n. g., n. sp. *Transactions of the Royal Society of New Zealand*, **75**, 218–230.

MacFarlane W.V. (1951) The life-cycle of *Stegodexamene anguillae* n. g. n. sp., an allocreadid trematode from New Zealand. *Parasitology*, **41**, 1–10.

MacLeod C.J., Paterson A., Tompkins D.M. & Duncan R.P. (2008) Parasites lost: did they miss the boat or drown on arrival? *Kararehe Kino*, **12**, 7–8.

McFarland L.H., Mouritsen K.N. & Poulin R. (2003) From first to second and back to first intermediate host: the unusual transmission route of *Curtuteria australis* (Digenea: Echinostomatidae). *Journal of Parasitology*, **89**, 625–628.

McIntosh A.R. (2000) Habitat- and size-related variations in exotic trout impacts on native galaxiid fishes in New Zealand streams. *Canadian Journal of Fisheries and Aquatic Sciences*, **57**, 2140–2151.

McIntosh A.R., Townsend C.R. & Crowl T.A. (1992) Competition for space between introduced brown trout (*Salmo trutta* L) and a native galaxiid (*Galaxias vulgaris* Stokell) in a New Zealand stream. *Journal of Fish Biology*, **41**, 63–81.

McIntosh A.R., Crowl T.A. & Townsend C.R. (1994) Size-related impacts of introduced brown trout on the distribution of native common river galaxias. *New Zealand Journal of Marine and Freshwater Research*, **28**, 135–144.

Norman R., Bowers R.G., Begon M. & Hudson P.J. (1999) Persistence of tick-borne virus in the presence of multiple host species: tick reservoirs and parasite mediated competition. *Journal of Theoretical Biology*, **200**, 111–118.

Osfield R.S. & Keesing F. (2000) Biodiversity and disease risk: the case of Lyme disease. *Conservation Biology*, **14**, 722–728.

Poulin R. (1993) Age-dependent effects of parasites on anti-predator responses in two New Zealand freshwater fish. *Oecologia*, **96**, 431–438.

Poulin R. (2000) Variation in the intraspecific relationship between fish length and intensity of parasitic infection: biological and statistical causes. *Journal of Fish Biology*, **56**, 123–137.

Poulin R. & Mouillot D. (2003) Host introductions and the geography of parasite taxonomic diversity. *Journal of Biogeography*, **30**, 837–845.

Power A.G. & Mitchell C.E. (2004) Pathogen spillover in disease epidemics. *American Naturalist*, **164**, S79–S89.

Prenter J., MacNeil C., Dick J.T.A. & Dunn A.M. (2004) Roles of parasites in animal invasions. *Trends in Ecology and Evolution*, **19**, 385–390.

R Development Core Team (2008) *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna.

Roelke-Parker M.E., Munson L., Packer C. *et al*. (1996) A canine *distemper* virus epidemic in Serengeti lions (*Panthera leo*). *Nature*, **379**, 441–445.

Sax D.F., Gaines S.D. & Brown J.H. (2002) Species invasions exceed extinctions on islands worldwide: a comparative study of plants and birds. *American Naturalist*, **160**, 766–783.

Smith M.J., White A., Sherratt A., Telfer S., Begon M. & Lambin X. (2008) Disease effects on reproduction can cause population cycles in seasonal environments. *Journal of Animal Ecology*, **77**, 378–389.

Telfer S., Brown K.J., Sekules R., Begon M., Hayden T. & Birtles R. (2005) Disruption of a host–parasite system following the introduction of an exotic host. *Parasitology*, **130**, 661–668.

Thieltges D.W. & Reise K. (2007) Spatial heterogeneity in parasite infections at different spatial scales in an intertidal bivalve. *Oecologia*, **150**, 569–581.

Tompkins D.M. & Poulin R. (2006) Parasites and biological invasions. In: *Biological Invasions in New Zealand* (Eds R.B. Allen & W.G. Lee), pp. 67–86. Springer, Berlin, Heidelberg.

Tompkins D.M., Dobson A.P., Arneberg P. *et al*. (2002) Parasites and host population dynamics. In: *The Ecology of Wildlife Diseases* (Eds P.J. Hudson, A. Rizzoli, B.T. Grenfell, H. Heesterbeek & A.P. Dobson), pp. 45–62. Oxford University Press, Oxford.

Tompkins D.M., White A.R. & Boots M. (2003) Ecological replacement of native red squirrels by invasive greys driven by disease. *Ecology Letters*, **6**, 189–196.

Torchin M.E., Lafferty K.D., Dobson A.P., McKenzie V.J. & Kuris A.M. (2003) Introduced species and their missing parasites. *Nature*, **421**, 628–630.

Townsend C.R. (2003) Individual, population, community, and ecosystem consequences of a fish invader in New Zealand streams. *Conservation Biology*, **47**, 38–47.

Townsend C.R. & Crowl T.A. (1991) Fragmented population structure in a native New Zealand fish: an effect of introduced brown trout? *Oikos*, **61**, 347–354.

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