Large scale patterns of trematode parasitism in a bivalve host: no evidence for a latitudinal gradient in infection levels

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ABSTRACT: Climate is a key driver of parasite transmission and disease dynamics. For trematode parasites, the high temperature sensitivity of transmission between first and second intermediate hosts may lead to higher infection rates with global warming, or spatially with warmer latitudes. However, spatial heterogeneities are common, and local factors are known to play crucial roles in determining infection levels. Using the latitudinal and sea temperature gradient along the New Zealand coastline, we assessed if this temperature sensitivity indeed translates into higher parasite abundance towards lower (i.e. warmer) latitudes in the cockle Austrovenus stutchburyi which serves as second intermediate host for several echinostome trematode species. Seventeen mudflats were sampled, and host densities and infection levels (i.e. metacercariae abundance) were measured for cockles, as well as for whelks and mudsnails (prevalence; first intermediate hosts). No evidence was found for a latitudinal pattern of metacercariae abundance in cockles. Instead, whelk prevalence per site and cockle foot size were found to be the main predictors. This highlights the importance of local factors—in particular, infection levels in first intermediate (i.e. source) hosts. These results indicate that, at least at large spatial scales, the temperature sensitivity of host-parasite systems may be offset by other ecological factors that confer resilience against on-going and predicted climate change.

KEY WORDS: Biogeography · Transmission · Host-parasite interaction · Climate change · Intertidal · Cockles

INTRODUCTION

Climate—in particular, temperature—is an important modulator of the transmission dynamics of parasites, and as a consequence, global climate change may have implications for the epidemiology of infectious diseases (Marcogliese 2001, Mouritsen & Poulin 2002a, Cattadori et al. 2005, Kutz et al. 2005). This is of relevance in all ecosystems, including marine ecosystems, where there have been concerns about an increase in infection levels and potential negative repercussions for affected populations and systems (Harvell et al. 2002, Ward & Lafferty 2004, Poulin & Mouritsen 2006). These concerns stem from the risk of parasite-induced host mortality (e.g. Fredensborg et al. 2004), but also from the fact that parasites play broader ecological roles (Mouritsen & Poulin 2002b, Marcogliese 2004, Wood et al. 2007, Lafferty et al. 2008). Hence, changes to the distribution and abundance of parasites or their vital rates such as reproduction and survival may not only have important implications for affected hosts, but also cascading effects on ecosystem structure and function. Despite the importance of parasites in marine ecosystems and growing concerns about some increasing diseases, our understanding of marine host-parasite systems...
and dynamics remains limited. In particular, knowledge of baseline infection levels are often lacking, thus impeding any attempt to detect changes that may be, or may have been, occurring. Also, determinants of infection levels in natural host populations are often unknown, limiting our ability to identify areas at risk for strong host-parasite interactions such as mortality events.

Trematodes are an important parasite group in coastal ecosystems (Lauckner 1984, Mouritsen & Poulin 2002b). They usually have a complex life cycle involving several members of a community. Typically, their life cycle comprises a definitive host (usually a vertebrate), a first intermediate mollusc host and a second intermediate host (invertebrate or vertebrate). A range of environmental factors are known to influence the transmission of trematodes (Pietrock & Marcogliese 2003, Thieltges et al. 2008). However, in particular the transmission between the first and the second intermediate host is known to be very sensitive to temperature (e.g. Mouritsen & Jensen 1997, Fingerut et al. 2003). Not only is the production of transmission stages (i.e. cercariae) affected by temperature, but also their emergence (Mouritsen 2002), survival (Studer et al. 2010) and infectivity (Thieltges & Rick 2006). Under optimal conditions for transmission, cercariae leave a first intermediate host to infect a second intermediate host. In general, the number of cercariae emerging increases with increasing temperature up to a limit, at the same time, their survival decreases, although this decrease is offset by an increase in infectivity (but see also Morley 2011). These responses are not only important for a parasite’s ability to complete its life cycle, but also in terms of the potential impact of a parasite on affected host populations. For parasites with temperature sensitive transmission processes, such as many trematodes, increasing temperature suggests that these parasites may have higher transmission rates in warmer areas or periods (Poulin 2006, Thieltges et al. 2009d), and therefore that infection levels in hosts (second intermediate hosts in the case of trematodes) may show a latitudinal pattern due to temperature gradients across large spatial scales (Thieltges et al. 2009d).

Indeed, there is an increasing number of studies investigating latitudinal patterns of parasitism, and in particular trematodes, in marine and other systems (Rohde 1992, Rohde & Heap 1998, Rohde 2002, Poulin & Mouritsen 2003, Poulin & Morand 2004, Thieltges et al. 2009b, Thieltges et al. 2009d). While Poulin & Mouritsen (2003) found no evidence for a latitudinal gradient in trematode infection levels in first intermediate snail hosts, Thieltges et al. (2009d) found latitude to be an important predictor of infection levels and trematode richness in second intermediate amphipod hosts. Their meta-analysis indicated that levels of infection (intensity and abundance) decreased with increasing latitude, consistent with the pattern expected based on the temperature sensitivity of the transmission from first to second intermediate hosts. However, as transmission dynamics are influenced by a complex interplay between biotic as well as abiotic environmental parameters (Thieltges et al. 2008), the importance of local, small scale factors causing heterogeneity and influencing infection levels has also been strongly emphasised (Poulin & Mouritsen 2003, Thieltges & Reise 2007, Thieltges et al. 2009c).

Here, we investigated latitudinal trends of trematode infections in the New Zealand cockle Austrovenus stutchburyi. This suspension-feeding bivalve is endemic to New Zealand, occurs in soft sediment ecosystems where it can reach densities of several hundred individuals per square meter, and has substantial value for commercial and recreational fisheries. The cockle serves as a second intermediate host for a number of parasites (Poulin et al. 2000), with echinostomes, namely Curtuteria spp. (Allison 1979, Leung et al. 2009a) and Acanthoparyphium spp. (Martorelli et al. 2006, Leung et al. 2009a) infecting the feet of cockles. These echinostomes are characterised by a spiny collar surrounding their oral sucker, with different numbers of spines in different species. The echinostomes targeted in this study use birds as definitive hosts (oystercatchers, possibly other species), whelks (e.g. Cominella glandiformis for Curtuteria spp.) and mudsnails (e.g. Zeacumans subcarinatus for Acanthoparyphium spp.) as first intermediate hosts. In terms of their transmission mode and their impact on cockles, the echinostome species are roughly equivalent (Babirat et al. 2004). Cercariae emerge from infected snail hosts (temperature dependent; Allison 1979, Koprivnikar & Poulin 2009) and infect a cockle, encysting as metacercariae in the cockle’s foot. Metacercariae accumulating in a cockle’s foot eventually impair the burrowing ability of the cockle, leaving heavily infected individuals exposed on the sediment surface (Thomas et al. 1998, Mouritsen & Poulin 2005). While this increases the likelihood of avian predation and hence transmission to definitive hosts, the manipulation of the cockle’s burrowing ability also affects the structure and function of the entire benthic community and ecosystem; these parasites are thus considered ecosystem engineers.
This trematode host-parasite system may be a good candidate for evaluating whether the high temperature-sensitivity of transmission that has been measured in laboratory experiments translates into spatial patterns of infections associated with temperature. Spanning about 12° latitude, mean annual sea surface temperatures along the North and South Islands of New Zealand range from about 11°C in the South to 18°C in the North (Figs. 1 & 2) (Chiswell 1994, Uddstrom & Oien 1999). However, temperatures on mudflats tend to be more extreme than temperatures in nearby main water bodies (Studer & Poulin 2012). Therefore, the temperature ranges covered in laboratory experiments — on which our understanding of the temperature effects is mainly based — should be expressed along this latitudinal and hence climatic gradient. The aim of the present study was therefore to investigate levels of cockle parasitism along the latitudinal range of New Zealand, while simultane-

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<td>17 Waikawa Harbour</td>
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Fig. 1. Location of the 17 sampling sites and their grid references (latitude, longitude) in New Zealand

Fig. 2. Mean annual temperatures (°C) at each sampling location’s latitudinal position. Temperature data was extracted from the World Ocean Atlas selecting the grid references closest to the sampling locations.
ously controlling for the effects of relevant local factors. If temperature is an important environmental factor determining levels of parasitism in these second intermediate hosts, then we would expect higher infection levels at lower latitudes, i.e. northern and warmer sites. If no such spatial pattern is detected, then other local ecological factors may be the key drivers of trematode parasitism in Austrovenus stutchburyi. Establishing such baseline data across large spatial scales, identifying determinants of infection levels and hence identifying hotspots of host-parasite interactions are all crucial, especially in the context of global climate change.

MATERIALS AND METHODS

Field sampling

The sampling was conducted on tidal mudflats along the east coast of both North and South islands of New Zealand from late February to early April 2012 (Fig. 1; total of 17 sites). Mudflats were, as much as logistically feasible, not sampled in a latitudinal order, and differences in times between sampling are considered negligible. Sampling locations were chosen to span the maximum latitudinal gradient logistically feasible (approx. 12°C latitude; 46°37’S to 34°31’S). On each mudflat, three 50 m transects were laid out in the mid-upper intertidal area at least 30 m apart and 30 m away from the upper shore (except in Hakahaka Bay: 30 m transects spanning the entire mudflat; Bluff: 50 m transects spanning the entire mudflat). The mid-upper area was targeted, as infection levels in cockles tend to be higher at high shore compared to low shore sites (Poulin et al. 2000, Mouritsen et al. 2003). Transects were laid out perpendicular to the low tide water line (except in Ahuriri estuary: sampling area enclosed by channels on either side). Along each transect, 3 randomly placed quadrats (0.45 × 0.45 m) were sampled (i.e. 9 replicate quadrats per location).

The upper 10 cm of the sediment within a quadrat were sieved (5 mm mesh size) and all cockles Austrovenus stutchburyi, whelks Cominella spp. and mudsnails Zeacumans subcarinatus or Z. lutulentus were collected and stored in separate 2 l plastic containers. All individuals were counted for density estimates. From each quadrat, 10 cockles (min. size 15 mm), 10 whelks (min. size 15 mm) and 10 mudsnails (min. size 10 mm) were haphazardly selected for dissections. In situations when there were insufficient individuals available from the initial sampling, additional specimens of above minimum size were collected haphazardly from the immediate vicinity of a quadrat or from the wider area of the mudflat to ensure, where possible, the collection of a total of 90 cockles, whelks and mudsnails per location. Cockles were measured (shell length and foot length in mm) using callipers, dissected and the foot of each cockle was cut off and put into a 1.5 ml tube filled with 99% ethanol for sample preservation and later assessment of parasite (i.e. metacercariae) abundance in the laboratory (see next subsection). The focus on the cockles’ feet was based on available knowledge on the echinostome species targeted—especially the temperature sensitivity of cercarial emergence from snail first intermediate hosts as well as the important ecological role that these parasites play by affecting the cockles’ burrowing behaviour (see ‘Introduction’). Whelks and mudsnails were measured (shell length in mm) under a dissecting microscope and dissected alive to assess their infection status and to identify, where possible, the trematode species or family. Identification was based on morphological differences between sporocysts/rediae (larval forms infecting a snail, which asexually produces cercariae) as well as morphological and behavioural differences between cercariae (e.g. swimming behaviour) (Schell 1970, Allison 1979, Martorelli et al. 2006). Trematodes were categorised as either ‘echinostomes’ or ‘other’ trematode species (including microphallids, philophthalmids, opisthorchids, and unknown species).

Laboratory analysis

To determine the number of metacercariae in each cockle foot, the foot was rinsed in distilled water and then transferred into tubes filled with 1.5 ml of pepsin solution (6 g pepsin powder and 7 ml conc. HCl in 1000 ml Milli-Q water). Tubes containing individual cockle feet were incubated at 40°C for 2 to 3 h to allow digestion of the cockle tissue. After incubation, metacercariae were rinsed by removing the pepsin solution and adding distilled water 3 times, and then transferred back into 99% ethanol for sample preservation. Subsequently, metacercariae were counted under a dissecting microscope. Species differentiation for metacercariae based on morphology (i.e. not considering cryptic species) would require the careful examination of individual cysts under high magnification for a count of the collar spines of each larval worm; this was not done as part of the present study.
Statistical analyses

Generalised linear mixed effect models (GLMM) were used to account for the nested structure of the sampling design. As a random factor, we included quadrat nested within transect, nested within site. All continuous variables were centred. Error structures were chosen to best account for the type of data analysed (see below). Models were fit to the data using maximum likelihood methods, and models were compared using Akaike’s Information Criterion (AIC; Burnham & Anderson 2002). All analyses were done using R 2.15.1 (R Development Core Team; www.R-project.org).

Metacercariae abundance in cockles, i.e. number of parasites per cockle foot including uninfected cockles, was analysed using the glmmADMB package (Skaug et al. 2012) and a negative binomial error structure. This analysis included only data on cockles collected from within quadrats (i.e. no cockles collected additionally were included). Two sets of models were assessed. The first set included density of infected whelks and mudsnails per quadrat as predictor variables. Model 1 included latitude as a predictor; Model 2, sea surface temperatures; Model 3, neither latitude nor temperature. The second set included the total density of whelks and mudsnails (i.e. infected and uninfected snails combined) per quadrat and their respective prevalence per site as predictor variables. Model 4 included latitude as a predictor; Model 5, sea surface temperatures; Model 6, neither latitude nor temperature. Temperature data used in the models were retrieved as mean annual temperatures over 1/4 degree-fields from the World Ocean Atlas (www.nodc.noaa.gov/OC5/WOA09/pr_woa09.html); values from the grid references closest to the sampling locations were extracted. All models also included cockle density and length of each cockle foot as predictor variables. Models were compared using AIC values. A GLMM (Gaussian error structure) was used to assess the relationship between latitude and cockle shell length (log transformed). Separate GLMMs (fitted with a Poisson error structure) were run to assess the effect of latitude on the density of cockles, whelks, echinostome-infected whelks, mudsnails, echinostome-infected mudsnails, and metacercariae per quadrat. Metacercariae per quadrat was calculated based on the mean parasite abundance per cockle in a quadrat times the density of cockles in that quadrat. Separate GLMMs with a binomial error structure were used to assess the effect of latitude and shell length on the echinostome infection status of whelks or mudsnails. These analyses were done using the package lme4 (Bates & Maechler 2009).

RESULTS

Infected cockles were found throughout the entire range covered by the study (Fig. 3A). Prevalence (the percentage of infected individuals) ranged from 29.2% (Waikawa Harbour) to 100% (several locations), and metacercariae abundance from a minimum range of 0 to 2 (Waikawa Harbour) to a maximum range of 1 to 2260 (Lower Portobello Bay) metacercara-
Metacercariae abundance in cockles

Of the models compared, the model with density of snail hosts and their prevalence per site (as opposed to the density of infected snail hosts) and excluding temperature and latitude as a predictor emerged as the best model (i.e. the model with the lowest AIC; Table 1). For the abundance of metacercariae per cockle foot, whelk prevalence per site and cockle foot length emerged as the only important predictors (Table 2) (the same results apply to analyses with infection status of cockles as response variable, i.e. prevalence; results not shown). Latitude was also not a strong predictor for the shell length of dissected cockles, and the model was not significantly different from the respective null model ($\lambda^2 = 0.554$, $p = 0.457$). Cockle shell and foot size correlated linearly ($y = -0.904 + 0.459x; r^2 = 0.780$) (see Fig. S1 in the Supplement).

Densities

Average density of cockles in our sampling locations ranged from 64.2 (Waikawa Harbour) to 1001.4 m$^{-2}$ (Ahuriri estuary) (Fig. 4A). Cockle density showed an increase towards lower latitudes, i.e. northern sites (Table 2). The average density of whelks ranged from 1.6 (Robinsons Bay) to 39.5 m$^{-2}$ (Matarangi) (Fig. 4B). Whelk density increased towards lower latitudes (Table 2). However, no trend was found for the density of echinostome infected whelks, and the model including latitude was not significantly different from the null model ($\lambda^2 = 0.004$, $p = 0.953$). Mudsnail density in locations where mudsnails were found ranged from 0.5 (Lamlash Bay) to 186.6 m$^{-2}$ (Matarangi) (Fig. 4C). Mudsnail density also increased towards lower latitudes (Table 2), but the density of infected mudsnails did not, with the model including latitude not being significantly different from the null model ($\lambda^2 = 2.536$, $p = 0.111$). The density of metacercariae per quadrat was also not affected by latitude, with the model including latitude not being significantly different from the null model ($\lambda^2 = 0.187$, $p = 0.666$).
Studer et al.: Cockle parasitism along latitudinal gradient

**Echinostome prevalence of first intermediate snail hosts**

The infection status of whelks was significantly affected by latitude and shell length (Table 2), with whelks having higher infection levels towards higher latitudes and with increasing shell length. For mudsnails, the model including latitude and shell length was not significantly different from the null model ($\lambda^2 = 2.549$, $p = 0.636$).

**DISCUSSION**

To explore whether the temperature sensitivity of parasite transmission translates into a large scale biogeographic pattern, the present study assessed the relationship between climate (i.e. latitude as a proxy for temperature) and levels of trematode parasitism in the New Zealand cockle *Austrovenus stutchburyi*. No latitudinal gradient was found for metacercarial infection levels in cockles collected from the mid-upper intertidal zone, for which shell size did not vary along the latitudinal gradient covered. Instead, prevalence in first intermediate whelk hosts and cockle foot size were identified as the main predictors of metacercariae abundance in cockles. Echinostome prevalence in whelks increased towards higher (i.e. colder) latitudes, while the density of whelks increased towards lower (i.e. warmer) latitudes. These opposite trends resulted in the density of infected whelks not showing a latitudinal pattern. On the other hand, neither echinostome prevalence in mudsnails, nor density of infected mudsnails showed a latitudinal pattern. Latitudinal trends were, however, found for the density of cockles, whelks and mudsnails, with densities for all hosts increasing towards warmer latitudes.

If temperature was a main driver of infection levels in cockles, then based on the fact that the density of infected first intermediate snail hosts did not differ across latitude, cockles should have been more infected at lower, warmer latitudes. This was clearly not observed in the present study, where sites with the highest infection levels in cockles were found at higher, colder latitudes (see also below). This highlights the importance of other local ecological factors for determining infection levels in *Austrovenus stutchburyi* (see also Mouritsen et al. 2003, Poulin & Mouritsen 2004 for small-scale, within mudflat patterns). Our results are in agreement with previous studies showing a relationship between abundance and infection levels in source hosts and infection levels in target hosts (Bustnes & Galaktionov 1999, Smith 2001, Hechinger & Lafferty 2005, Fredensborg et al. 2006, Byers et al. 2008), and hence the input of infective stages into a particular system. These studies mainly focussed on the abundance of definitive hosts (usually birds) and infection levels in first intermediate snail hosts. Although the link between bird abundance and infection levels in intermediate hosts is not directly considered here, this interrelation could be strong. Our results provide evidence for this link extending to a second intermediate host (see also Thieltges 2007, Thieltges & Reise 2007, de...
As the complex life cycle of trematode parasites requires all host types to be present in order to complete the cycle, the link between the abundance of definitive hosts, and first and second intermediate hosts is fundamental.

Our results therefore depart from the general expectation solely based on laboratory-based temperature effects on transmission processes (e.g. Poulin 2006, Studer et al. 2010) and also from the latitudinal trend described in a meta-analysis for amphipod hosts (across ~20° latitude) (Thieltges et al. 2009d). However, these contrasting patterns may be reconciled by reconsidering the spatial scale at which the temperature-sensitivity of trematode transmission affects infection levels. Almost all studies of the effects of temperature on the production, survival and infectivity of cercarial transmission stages involve experiments on single populations performed under simplified conditions (e.g. Thieltges & Rick 2006, Studer et al. 2010). They generally show that all else being equal, higher temperatures (up to some maximum) lead to higher net transmission. In natural conditions, however, all else is not equal: at higher temperatures, the feeding activity of non-host cercarial predators may increase, the target hosts may shift their microhabitat choice to seek cooler temperatures and coincidentally avoid cercariae, etc.

Although the net effect of warming may still be that trematode transmission is enhanced, this cannot apply equally to different geographic localities, which will vary in terms of community composition (e.g. abundance of cercarial predators, alternative hosts or non-hosts) and physical features (e.g. availability of microhabitat refuges, sediment composition, mudflat topography). In addition, geographical variation, possibly due to genetic differences, has been observed in how temperature affects cercarial production under controlled conditions (Koprivnikar & Poulin 2009), adding yet another layer of complexity to the search for latitudinal gradients in infection levels in second intermediate hosts. Thus, the temperature-sensitivity of trematode transmission may result in local increases in infection risk as a consequence of climate warming, but may not result in a latitudinal gradient in infection levels; such effects may be offset by other important ecological factors that are not related with latitude.

The contrasting patterns may also be reconciled by considering the temperature range of laboratory experiments compared to that experienced by organisms in nature. Although temperature levels chosen in experiments are usually within the range of temperatures experienced in nature for a given host-parasite system (e.g. Mouritsen 2002, Thieltges & Rick 2006), they often do not include upper temperature levels beyond the optimum range. Temperatures above the optimum are, however, important when considering the full thermal environment experienced by the organisms in their actual habitat (Studer et al. 2010, Studer & Poulin 2012), as an overall negative net effect on transmission can be expected. Mean annual sea surface water temperatures across New Zealand range from about 11°C to 18°C; our sampling sites on the North Island have an annual mean >15°C, whereas those on the South Island are <15°C. Temperatures on the mudflats where transmission occurs are more variable and extreme than in nearby water bodies (Studer & Poulin 2012) (e.g. Lower Portobello Bay mudflat [Site 14; 45° 49’ S] mean 11.6°C, max. 26.5°C, min. 3.0°C compared to the adjacent main water body [Otago harbour] mean 11.6°C, max. 18.1°C, min. 6.5°C). For example, Allison (1979) assessed the emergence of Curtuteria australis cercariae from infected whelk hosts at 15°C and 23 to 25°C, finding an optimum around 25°C, but noted that temperatures on the mudflat of origin (Avon-Heathcote estuary, South Island, New Zealand; 43°33’S) ranged from 3 to 33°C. Hence, the high inherent temperature variability of intertidal systems coupled with our limited understanding of the thermal biology of the species involved—especially in terms of their actual optimum temperature ranges—in addition to potential effects of local adaptations, further complicate our understanding of large scale patterns in trematode infection levels. Additionally, it is possible that due to this variability, the 12° latitude covered and the ~7°C difference in mean annual sea surface temperature may simply not be large enough to result in notable differences across the latitudinal gradient investigated here.

The present results are also not in full agreement with Thieltges & Reise (2007), in which the density of first intermediate snail hosts was the strongest determinant of infection levels in the European cockle Cerastoderma edule. The authors argued that density of first intermediate hosts should roughly correlate with the number actually infected. In our study, the density of whelks and mudsnails increased towards warmer latitudes, while echinostome infection levels only showed a latitudinal pattern for whelks, which offset the effect of whelk density. Hence, our results indicate that density of hosts and their infection levels should be considered separately. The 2 studies are, however, in agreement with regards to infection levels increasing with cockle size (but see
Montaudouin et al. 2009). In the present study, no
presence of trematodes found in the present study.

Latitudinal distribution (de Montaudouin et al. 2009,
2003), but may not necessarily be older. In terms of
temperature, it is expected that cockles in the North
grow faster than in the South (Marsden & Adkins 2010). This would indicate that, due to the lack of a
size difference in the cockles dissected in this study
along the latitudinal gradient covered, there might
indeed be an age difference, with cockles in the
South being older. This may help explain why some
of the highest metacercariae levels were found in the
South. Indeed, exposure time rather than size has
been shown to be of importance in determining
infection levels in C. edule, in which mean infection
levels strongly increased with age suggesting tempo-
ral exposure rather than size to be the main determi-
nant of infection levels (Thieltges 2008). In addition,
the fact that metacercariae density (density of cock-
les times average number of metacercariae in cock-
les in a particular quadrat) did not show a latitudinal
pattern (see ‘Results’) despite the density of cockles
increasing towards North (Fig. 4A), indicates that
fewer cockles tend to harbour more metacercariae in
mid-upper shore areas in the South (Fig. 3A). Over-
all, these patterns may therefore be best explained
by a combination of prevalence of infection in whelks
as well as the age of cockles (between sites) and size
of cockles (within sites).

_Cerastoderma edule_ along the Northeast Atlantic
coast were found to be infected with trematodes at all
sites (de Montaudouin et al. 2009), similar to the om-
nipresence of trematodes found in the present study.
Different cockle populations, however, harboured
different parasite communities; with some species
having a restricted range and others showing a large
latitudinal distribution (de Montaudouin et al. 2009,
Thieltges et al. 2009a). In particular, the latitudinal
distribution of first intermediate hosts was suggested
to be important for understanding these patterns (de
Montaudouin et al. 2009). In the present study, no
species differentiation was possible based on the
morphology of the metacercarial cysts in cockle feet.
However, future molecular analysis of the metacerc-
ariae collected may elucidate the species composition
of the trematodes affecting _Austrovenus stutchburyi_
and uncover the presence of other species occurring along the latitudinal gradient of the New Zealand coastline (see Leung et al. 2009a). Moreover,
only foot tissue was assessed in our study, and hence
it remains unknown to what extent other trematode
(or parasite) species were present in the cockle
tissues (e.g. _Gymnophallus_ sp., Leung et al. 2009b).

Latitudinal patterns may exist for individual trema-
tode species, even though none were detected for all
echinostome species combined infecting cockle feet.

Another potential issue with our data stems from
the fact that the same sampling protocol was applied
for mudflats differing substantially in size, topogra-
phy and sediment composition. Moreover, as cockle
density is not uniform across a mudflat, and infec-
tions in hosts are usually patchy, it is possible that our
sampling design (e.g. number of replicates, quadrat
size) was not adequate in some places to provide rep-
resentative data. That cockles are an important tar-
get for commercial and recreational fisheries should,
on the other hand, not be of major concern here, as
cockles in the mid-upper intertidal areas are usually
not targeted.

In conclusion, despite the known temperature sen-
sitivity of trematode transmission, no latitudinal or
temperature-related pattern was identified for meta-
cercariae abundance in second intermediate cockle
hosts. Instead, infection levels in first intermediate
whelk hosts and cockle size were found to be the
main predictors. This probably stems from the fun-
damental relationship between infection levels in
first intermediate (source) hosts and second interme-
diate (target) hosts, and supports the importance of
other local ecological factors in determining levels of
parasitism as opposed to large scale climatic factors.

In the context of climate change, how host species
respond to altered conditions in terms of their abun-
dance and distribution will therefore be of particular
relevance. This may have important repercussions
for parasite species and their ability to complete
their life cycle, and hence infection levels in hosts.
Due to the complexity of ecological systems, antici-
pating how marine host-parasite systems may re-
spond to climate change still remains a major chal-
lenge. This study, however, provides baseline data
on key species in intertidal systems, and hence
should be useful for assessing potential temporal
and/or spatial changes.

Cerastoderma edule
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