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Contrasting temperature responses in seasonal timing of cercariae shedding by *Rhipidocotyle* trematodes

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Research Article

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Abstract

Global warming is likely to lengthen the seasonal duration of larval release by parasites. We exposed freshwater mussel hosts, *Anodonta anatina*, from 2 high-latitude populations to high, intermediate and low temperatures throughout the annual cercarial shedding period of the sympatric trematodes *Rhipidocotyle fennica* and *R. campanula*, sharing the same transmission pathway. At the individual host level, under warmer conditions, the timing of the cercarial release in both parasite species shifted towards seasonally earlier period while its duration did not change. At the host population level, evidence for the lengthening of larvae shedding period with warming was found for *R. fennica*. *R. campanula* started the cercarial release seasonally clearly earlier, and at a lower temperature, than *R. fennica*. Furthermore, the proportion of mussels shedding cercariae increased, while day-degrees required to start the cercariae shedding decreased in high-temperature treatment in *R. fennica*. In *R. campanula* these effects were not found, suggesting that warming can benefit more *R. fennica*. These results do not completely support the view that climate warming would invariably increase the seasonal duration of larval shedding by parasites, but emphasizes species-specific differences in temperature-dependence and in seasonality of cercarial release.

Introduction

Marked seasonal fluctuation in temperature is a characteristic of high-latitude ecosystems. Such seasonal temperature variation can affect trematode parasites in different ways, including the timing of seasonal production of infective stages (miracidia and cercariae), since their release primarily occurs during the warm summer months in temperate and boreal zones (e.g. Chubb, 1979; Taskinen, 1998*a*; Karvonen *et al.*, 2004). Indeed, most experimental and field studies (e.g. Fingerut *et al.*, 2003; reviewed in Poulin, 2006; Thieltges and Rick, 2006; Studer *et al.*, 2010; Shim *et al.*, 2013; Rosen *et al.*, 2018; Selbach and Poulin, 2020; Vyhlídalová and Soldánová, 2020) have reported increased release of cercariae with a moderate temperature rise. However, in some cases, this effect can be transient (Paull *et al.*, 2015), absent or even negative, most probably because cercariae emergence rates are tended to decrease at the threshold temperatures and drop with temperature rise (Koprivnikar and Poulin, 2009*a*, 2009*b*; Morley and Lewis, 2013). A common expectation is that the predicted climate warming (IPCC, 2014) will increase the seasonal duration of larval release by parasites as a consequence of a longer thermal growing season (longer summer) (Marcogliese, 2001; Harvell *et al.*, 2009; Lõhmus and Björklund, 2015; Prokofiev *et al.*, 2016; Galaktionov, 2017). Such a lengthening of the cercarial release period has been observed in water bodies receiving thermal effluents (e.g. Aho *et al.*, 1982). However, experimental long-term manipulations of temperature conditions over the seasonal cercarial release period are rare. To our knowledge, the only over-season experimental long-term study, by Paull and Johnson (2014), indicates a seasonal shift, rather than lengthening, in the timing of cercariae emergence. This shift to a seasonally clearly earlier occurrence of cercariae emission led to a decrease in parasite transmission and reduced parasite-induced host pathology due to temporal mismatch between cercariae and their target host (Paull and Johnson, 2014). Furthermore, whether such lengthening of the seasonal cercarial shedding period would be a result of a longer cercarial shedding at the individual host level or a result of the seasonal variation between host individuals has remained unexplored. In addition, most experimental studies of cercarial release were conducted at constant – though different – temperatures, while in natural conditions the temperature fluctuates during the cercariae shedding period. Finally, as most of the studies have been performed with snail hosts and marine species, only little is known about temperature effects in freshwater bivalve–trematode associations (however, see Morley and Lewis, 2013; Choo and Taskinen, 2015). This gap demands attention since bivalves are important parts of freshwater ecosystems providing valuable ecosystem services (e.g. Vaughn, 2018), and are very commonly infected by trematodes, frequently with high prevalence of infection (Müller *et al.*, 2015). Bivalves can release a huge amount of cercariae (Taskinen, 1998*a*) which infect many central

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prey fish species and are transmitted to the key predatory fishes of freshwater ecosystems (Taskinen *et al.*, 1991; Cribb *et al.*, 2001). Therefore, a long-term study on temperature effects in a freshwater bivalve–trematode association, with temperature treatments mirroring the natural seasonal fluctuations in temperature conditions should be considered necessary.

Cercarial larvae of trematodes emerge over species-specific temperature conditions both in the laboratory and field studies (e.g. Fingerut *et al.*, 2003). However, interspecific comparisons of cercarial production in varying temperature conditions that can reveal species-specific responses have been utilized quite rarely and mainly focus on short-term temporal variability in cercarial emergence (e.g. diurnal rhythms) rather than on large-scale seasonal patterns (de Montaudouin *et al.*, 2016; Prokofiev *et al.*, 2016; Vyhlídalová and Soldánová, 2020; see, however, field observations by Fingerut *et al.*, 2003; Koprivnikar and Poulin, 2009*a*). In addition, in most previous studies, the influence of temperature on cercariae shedding has been compared between parasite species with different transmission pathways, while species-specific differences in the closely related sympatric parasites sharing the same hosts are rarely studied (Vyhlídalová and Soldánová, 2020). In the present study, we used 2 closely related, sympatric trematodes, *Rhipidocotyle campanula* and *R. fennica*, in their shared first intermediate host, freshwater mussel *Anodonta anatina*. The parasites also have the same second intermediate host, the cyprinid fish *Rutilus rutilus* (Taskinen *et al.*, 1991; Gibson *et al.*, 1992).

In addition, though the importance of studying temperature-dependent cercariae output in high-latitude areas (>60°) has been highlighted earlier (Morley and Lewis, 2013; Studer and Poulin, 2014; Galaktionov, 2017), experimental investigations are still scarce (Prokofiev *et al.*, 2016). They would be timely since impacts of the climate change are predicted to be the most pronounced at high latitude regions. For example, climate models predict an increase in annual temperature from 2 to 7°C by the 2080s compared to a 1961–1990 baseline period in the current study region Finland at 60–70°N (Jylhä *et al.*, 2004), and in the temperate lakes of the northern hemisphere in general (Sharma *et al.*, 2007).

Production of cercariae is an important component of the complex life cycle of trematodes, underpinning transmission to the next host and hence influencing the fitness of the parasite. Cercariae of trematodes are transmitted to a variety of hosts and, in addition to being an infectious agent, they can serve as a valuable food source for many aquatic organisms (Johnson *et al.*, 2010; Orlofske *et al.*, 2012; Mironova *et al.*, 2019, 2020; McKee *et al.*, 2020). Thus, cercariae play an important role in the functioning of aquatic ecosystems (Kuris *et al.*, 2008; Thieltges *et al.*, 2008; Preston *et al.*, 2013). Therefore, the seasonal timing and duration of the cercarial shedding period can affect different trophic levels in aquatic ecosystems by changing parasite burden and food availability.

In the present long-term (5 months) experiment, we investigated the seasonal cercarial shedding traits using 3 different temperature levels reflecting the natural temperature variation over the distribution range of the bivalve host *A. anatina* in the current study region, Finland (60–68°N). We suggest that higher temperature will accelerate parasite development, leading to the earlier seasonal start of cercariae emission. This is based on the general view that parasites receive a competitive advantage over the host under high temperature (e.g. see Lõhmus and Björklund, 2015; Marcogliese, 2016 for the discussion) and on the previous observation of the short-term temperature effect on *R. fennica* cercarial release (Choo and Taskinen, 2015). We were specifically interested in disentangling the seasonal duration of cercarial shedding at the individual host and host population levels. Our hypotheses were that under higher temperatures, mussels will start to emit cercariae of both *Rhipidocotyle* species seasonally earlier, will emit cercariae for a longer period, and need fewer day-degrees to start cercariae shedding than under lower temperatures.

Materials and methods

Study species

The bivalve mollusc host, *A. anatina*, is a common European freshwater mussel with a maximum life span >10 years, age of maturation 2–4 years and maximum length of 12 cm (Taskinen and Valtonen, 1995). *Anodonta anatina* serves as the first intermediate host of the bucephalid trematodes *Rhipidocotyle campanula* and *R. fennica* (Taskinen *et al.*, 1991, 1997; Gibson *et al.*, 1992; Müller *et al.*, 2015). Cercariae are produced asexually by sporocysts located in the gonads of the mussel. The prevalence of infection by *R. campanula* in natural populations is usually less than 10% (Taskinen *et al.*, 1991; Müller *et al.*, 2015), whereas that by *R. fennica* can be up to 50% (Taskinen *et al.*, 1994). Pronounced seasonality in the developmental stages of cercariae, shedding of cercariae, developmental stages of sporocysts and quantity of sporocysts of *Rhipidocotyle* species in *A. anatina* was observed (Taskinen *et al.*, 1994; Taskinen, 1998*a*). Both parasite species have been linked to decreased growth, survival and reproduction of *A. anatina* (Taskinen and Valtonen, 1995; Taskinen, 1998*b*; Jokela *et al.*, 2005; Müller *et al.*, 2015). The second intermediate host of these trematodes is the cyprinid fish *R. rutilus*, in which *R. fennica* metacercariae encyst in the fins and *R. campanula* metacercariae – in the gills (Taskinen *et al.*, 1991; Gibson *et al.*, 1992). The definitive hosts for *R. campanula* are the percid fishes *Perca fluviatilis* and *Sander lucioperca* and the definitive host for *R. fennica* is the esocid fish *Esox lucius* (Taskinen *et al.*, 1991; Gibson *et al.*, 1992).

Experimental set-up

Altogether 281 *A. anatina* mussels were collected from the River Kuusaankoski (17 May 2011; 62°25′N, 26°00′E) and 290 mussels from the River Haajaistenjoki (22 May 2011; 63°63′N, 26°99′E), Finland. These 2 sampling sites were chosen for comparison of cercariae emergence traits in different mussel populations. They were located quite far from each other (distance > 140 km) and differed by environmental conditions, which allowed the collecting of mussels and parasites of various phenotypes and life stories for the experiment. At the Konnevesi Research Station, University of Jyväskylä, the mussels were individually marked and measured. Average shell length ± S.E. for the River Haajaistenjoki mussels was 61.9 ± 0.6 (range 33.0–92.6 mm), and for the River Kuusaankoski mussels 77.7 ± 0.6 (range 38.8–101.7 mm). There was a significant size difference between mussel populations (estimate ± S.E. = 16.9 ± 1.1, $t = 16.08$, $P < 0.0001$), while the mussels shedding and non-shedding cercariae did not differ in their size (estimate ± S.E. = 1.83 ± 1.3, $t = 1.45$, $P = 0.15$) (Supplementary Fig. S1, Tables S1 and S2). Twelve mussels were infected with both species of parasites and were excluded from the subsequent analyses, except for the analysis of probability of cercariae shedding in different temperature treatments (see section Data analysis). Data about sizes of mussels taking into account parasitic species are presented below.

From the date of collection to 25th June, mussels were kept in the laboratory in 2 tanks (1 population per tank) under flow-through conditions (open tanks that allow a constant flow of new water). The infection status of collected mussels was unknown at this stage (it became clear later during experimental monitoring of cercariae emission), thus infected and uninfected mussels were maintained together. Each 163-l tank was filled

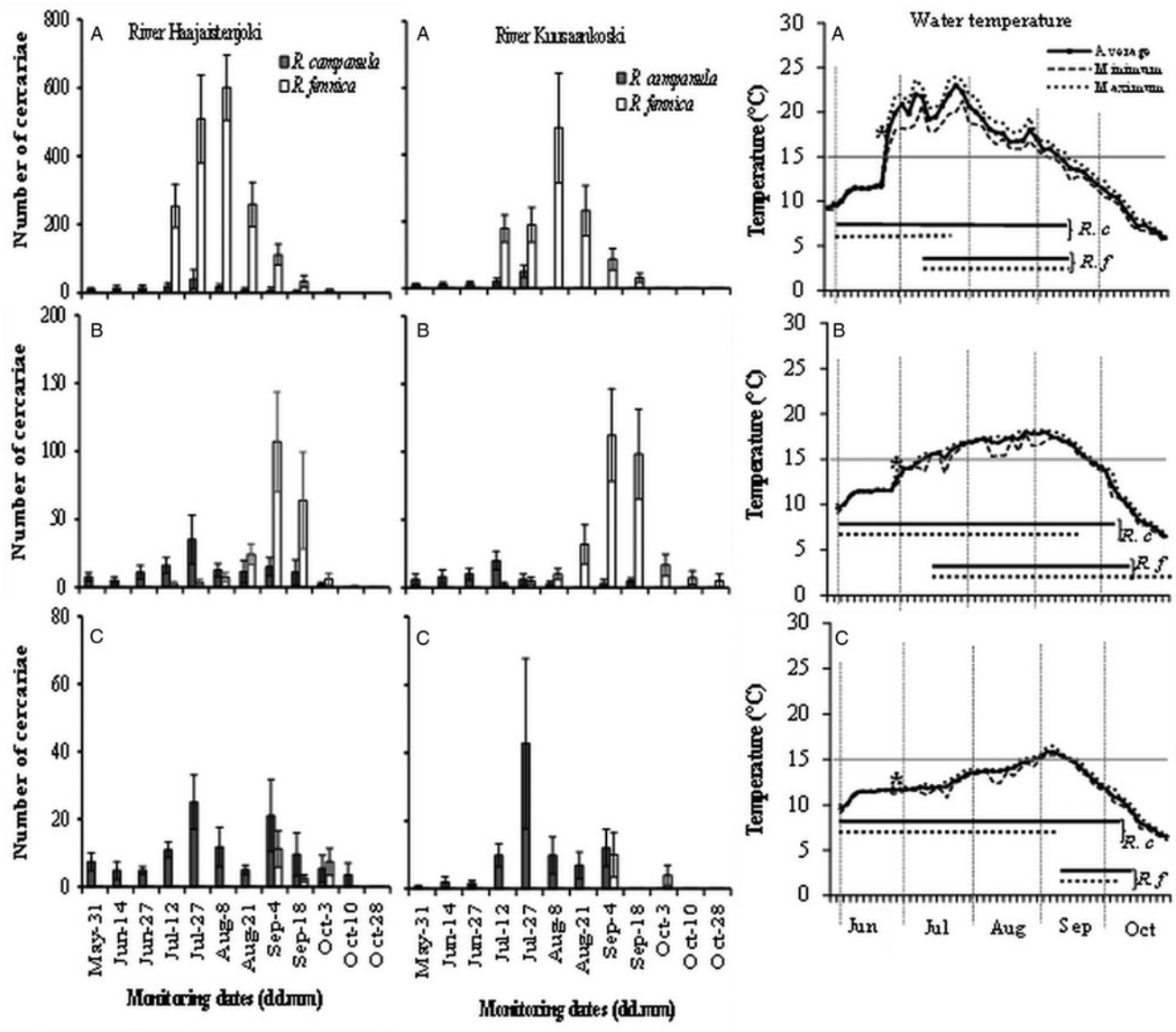

Fig. 1. The daily (mean ± S.E.) cercarial release of *Rhipidocotyle campanula* (*R. c*) and *R. fennica* (*R. f*) from the mussel host *Anodonta anatina,* water temperature profile at mean 3-day intervals and the total duration of cercarial release by mussels from the River Haajaistenjoki (straight horizontal line) and from the River Kuusaankoski (dotted horizontal line), from 31 May to 28 October in the high- (A), intermediate- (B) and low-temperature treatments (C). An asterisk represents the day when the mussels were assigned to the different temperature treatments. Note the different scales on the *y*-axes.

with 5 cm of sand at the bottom and supplied with water from the hypolimnetic zone (9 m depth) of Lake Konnevesi at a rate of up to 10 L min^{-1}. Water temperatures in both tanks were similar throughout this period ranging from 10.5°C on 31 May to 11.7°C on 25 June (Fig. 1).

On 25 June, the mussels were randomly assigned to 1 of the 3 temperature treatments – high, intermediate and low temperature (see below) – with 2 replicate tanks per treatment. Mussels from both populations and from all size groups were distributed evenly to each of the 6 tanks (for mussel numbers per tank, see Table 1). The average water temperatures from 25 June to 28 October, when the experiment was terminated, were 18°C (range 7–24°C), 15°C (range 7–20°C) and 13°C (range 6–18°C) in high, intermediate and low temperatures, respectively. There was no length difference between mussels allocated to temperature treatments (2-way analysis of variance; $F_{2,\ 553} = 0.056$, $P = 0.945$) and no interaction between population and treatment ($F_{2,\ 553} = 0.697$, $P = 0.499$). Average shell length of mussels (±S.E.) in high-temperature tanks was 70.0 ± 1.0 mm (range 33.0–101.7 mm), in intermediate temperature tanks 69.4 ± 1.0 mm (range 37.9–100.0 mm) and in low-temperature tanks 69.8 ± 0.9 mm (range 37.4–101.5 mm). See Table S3 for the treatment per population descriptive statistics.

The water temperature ranges in the different temperature treatments corresponded to the natural water extreme temperature variations currently occurring throughout the distributional area of *A. anatina* in Finland, from 60 to 68°N and represent the maximum summer temperatures varying from about 17 to 24°C, respectively (Kuha *et al.*, 2016). The number of days when the average daily water temperature in the different treatments was ⩾15°C, a measure of the length of the warm/growing season, was 74, 71 and 20 days in high, intermediate and low-temperature treatments, respectively.

The temperature treatments were established as follows. (1) High-temperature tanks were placed in outside shelter and supplied with running water – using a pump – from the littoral zone (<2 m depth) of Lake Konnevesi. (2) Intermediate-temperature tanks were kept indoors and supplied with heated hypolimnetic water pumped from Lake Konnevesi. Water was heated in a separate tank with aquarium heaters before delivery to mussel tanks. (3) Low-temperature tanks were kept indoors and supplied with

Table 1. Total numbers of *A. anatina* mussels (*N*) and numbers of mussels shedding cercariae (N_s; *n* shedding *R. fennica*/*n* shedding *R. campanula*) from the River Haajaistenjoki and the River Kuusaankoski kept in high- (HT), intermediate-(IT) and low-temperature (LT) treatments

			Start date		Stop date		Duration		Start °C		Stop °C	
	N	N_s	*R. fennica*	*R. campanula*	*R. fennica*	*R. campanula*	*R. f.*	*R. c.*	*R. f.*	*R. c.*	*R. f.*	*R. c.*
River Haajaistenjoki												
HT	96	33/8	12 Jul–8 Aug	31 May–27 Jun	27 Jul–18 Sep	14 Jun–18 Sep	10	16	21.5	10.5	16.0	16.0
IT	97	19/11	12 Jul–3 Oct	31 May–4 Sep	4 Sep–14 Oct	12 Jul–3 Oct	14	18	15.5	10.5	9.0	12.0
LT	97	6/20	4 Sep–3 Oct	31 May–8 Aug	18 Sep–14 Oct	14 Jun–3 Oct	6	18	16.5	10.5	11.0	11.0
River Kuusaankoski												
HT	93	37/3	12 Jul–8 Sep	31 May–14 Jun	8 Aug–18 Sep	14 Jun–27 July	10	8	21.5	10.5	16.0	23.0
IT	93	13/6	12 Jul–14 Oct	31 May–12 Jul	4 Sep–28 Oct	12 Jul–18 Sep	16	16	15.5	10.5	8.0	15.7
LT	95	2/8	4 Sep–3 Oct	31 May–8 Aug	4 Sep–3 Oct	12 Jul–4 Sep	4	14	16.5	10.5	11.0	16.5

Start and stop dates represent the range between the earliest and latest observations of cercarial emergence, respectively. Duration indicates the length (weeks) of the cercarial release at the host population level, from the first to the last observation of shedding. Start °C and Stop °C represent the water temperatures (°C) on the dates when the first and last cercariae emerged, respectively.
R. f. and *R.c.* = *Rhipidocotyle fennica* and *R. campanula*, respectively.

hypolimnetic water pumped from Lake Konnevesi. *Anodonta* mussels are filter-feeders utilizing phytoplankton, bacteria and fine organic particles (Jorgensen *et al.*, 1984), thus a continuous flow of lake water was necessary to provide the mussels with food. Due to logistic constraints, differences other than temperature existed between the treatments. Mussels in the high-temperature treatment were subject to a larger daily fluctuation of temperature than those in the intermediate- or low-temperature treatments (Fig. 1), as the littoral water and outdoor tanks were used. In addition, the daily/seasonal profile varied such that the daily water temperature in the high-temperature treatment tanks peaked in late July (24°C), while in the intermediate (19°C) and low-temperature tanks (17°C), it peaked in early September (Fig. 1).

The indoor tanks were illuminated by artificial light with the photoperiod set to correspond with the natural rhythm. The outdoor tanks received natural light but the shelter above the tanks provided effective cover against direct sunlight. However, during the 24 h cercarial release monitoring period, similar artificial light was used for all mussels to provide equal light conditions (see below). Water flow into the holding tanks was adjusted such that it was higher in the intermediate and low temperatures (10 L min^{-1}) than in the high-temperature tanks (5 L min^{-1}). This was to compensate for the probable higher food density in the high-temperature tanks that received littoral water, than the intermediate- and low-temperature tanks that received hypolimnetic water. A submersible temperature logger was placed in 1 replicate tank per treatment to measure water temperature every 4 h from 25 June to 28 October (end of the experiment). Temperature in the high-temperature treatment (outdoor tanks) varied much stronger during the day than in other treatments (Fig. 1). However, results by Roushdy (1984) indicate that cercarial release does not differ between constant and diurnally variable temperatures.

Cercarial release from each mussel was followed over a period of 20 weeks by counting cercariae released per *A. anatina* at roughly 2-week (12–15 days each) intervals between 31 May and 28 October, during a total of 12 monitoring sessions. On each monitoring day, individual mussels were placed in a 4-l transparent plastic box (length 26.5 cm, width 19 cm and height 13.6 cm) filled with 2 L of filtered lake water for 24 h (possible dead mussels were removed at this stage) and then returned to their respective holding tanks. The water temperatures in the monitoring boxes during the 24 h period of cercarial shedding were adjusted to correspond with those in the respective holding tanks and, when necessary, a temperature-controlled room was used. Light conditions during the monitoring were also set to correspond with the natural day length and rhythm because the cercarial release of *Rhipidocotyle* species is diurnal (Taskinen *et al.*, 1991). The chosen 24 h incubation period allows excluding the effects of circadian rhythms, considerably influencing the results on cercariae shedding (Hannon *et al.*, 2017). The number of cercariae in the box was counted visually (at low densities, <20 cercariae), or microscopically from a 50 mL of the mixed subsample (at high densities, >20 cercariae). The only 1 sample was used for cercarial counts since we were interested in estimating cercariae emergence at different temperatures (but not cercariae emergence per mussel per day), so all mussels from a certain temperature treatment at a certain day can be considered a sample, while each individual mussel is a subsample.

The experiment was terminated on 28 October 2011, when cercarial release approached zero in practically all treatments. For *Rhipidocotyle* species, the cercarial shedding in the field and in the laboratory has been reported to occur between late May and early October (Taskinen *et al.*, 1994, 1997; Taskinen, 1998*a*).

Data analysis

Statistical analyses and plots preparation were performed using PASW Statistics 18 and R (R Core Team, 2020). Plots were drawn using ggplot2 (Wickham, 2016), cowplot (Wilke, 2020) and gridExtra packages (Baptiste, 2017), multiple comparisons for GLMs were, when possible, done using multcomp package machinery (Hothorn *et al.*, 2008), while when impossible (e.g. with zero-inflated models) Bonferroni corrections were used.

Fisher test was used to compare the proportion of mussels shedding cercariae in different treatments and independence of infections. In the former case, double-infected mussels were added to both *R. campanula* and *R. fennica* columns. When double-infected mussels were excluded, results were similar (see the Supplementary material). For all other analyses, mussels that did not shed cercariae and double-infected mussels were not included in the statistical analyses. Data from replicate tanks were combined, as prior tests revealed no differences between replicates for any measured variable.

To check whether mussels from different treatments and populations differ in their shedding start date, day-degrees required for the start, stop date and the mean duration of cercarial emergence, we used the following strategy. First, we tried

generalized linear models (GLMs) with the Gaussian error structure and identity link function. We checked the residuals visually on Q-Q plots and using the Shapiro–Wilks test. Ordinary GLMs (hereinafter GLMs) were preferred due to their easier interpretability. When necessary, response variables were log-transformed. If models' assumptions were still violated, we used negative binomial models since in most cases our data were strictly positive with a heavy right tail. In a few cases, we used zero-inflated models to account for high numbers of zero values (Zeileis *et al*., 2008). Finally, several times we had to switch to quasi-Poisson models due to the convergence problems with negative binomial models.

We always started with models including mussel length, mussel origin (River Haajaistenjoki or River Kuusaankoski population) and temperature treatment as predictors. Mussel lengths were centred since zero length is not biologically sensible. We considered it biologically relevant to include mussel size since it is often positively correlated with cercariae emergence rate (e.g. Morley *et al*., 2010). We also included all double interactions, while higher-order interactions were not included due to a lack of *a priori* hypotheses and interpretation problems.

Initial models were simplified using Akaike's information criterion (AIC) (Symonds and Moussalli, 2011). For quasi-Poisson models, which do not have AIC in the output, nested models were compared using the *F*-test.

To compare survival of mussels shedding cercariae in different temperature treatments, we started with a logistic regression, where treatment and mussel's length were predictors (interaction was included), and then simplified it.

To check the influence of temperature on the cercariae emission rate, we calculated mean temperature and mean cercariae emission at each sampling day in each treatment. Under temperatures below 9–11°C, mussels rarely emit cercariae, which was predictable since for many species of trematodes such temperatures are likely to be threshold ones for shedding infective stages (Morley and Lewis, 2013). In the subsequent analysis, we used only non-zero emission data. Models, where zero-emission data were included, are presented in the Supplementary material. Qualitatively, they are similar to non-zero ones; however, we suggest that they mainly reflect a large bulk of points with zero-emission at low temperatures rather than a biologically sensible relationship. One point in *R. campanula* data looked like an obvious outlier (see the Supplementary material) and was excluded from the dataset before the analysis.

We fitted GLMs where temperature and the day of the observation were predictors (interaction included). A seasonality factor (number of days from the beginning of the experiment, which roughly represents the time since the beginning of shedding season) was added since, during the exploratory analysis, we noticed that under similar temperatures, cercariae emissions were lower at the end of shedding period than at the beginning of it. Temperatures were centred since a cercariae emission rate at 0°C has no biological sense, while at mean temperature does have. The response variable (mean shedding rate) was log-transformed.

To check between-species differences in shedding traits, we used a similar analytical strategy; however, instead of the population factor, we added the species factor in our models. We did not include population since our analysis of separate species did not find substantial differences in cercariae shedding traits in mussels from different populations. Since data on the temperature emission start looked very different for different parasitic species and residuals looked 'fishy' in any model we tried, a robust regression was used for the data analysis. 'f.robtest' function from the sfsmisc (Maechler, 2021) package was used to obtain *P* values.

Results

Shedding probability

In total, 178 out of 571 *A. anatina* mussels shed cercariae in the course of observations. Among those, 110 mussels were infected only with *R. fennica* and 56 only with *R. campanula*, whereas 12 were infected with both species (excluded from most of data analyses, see above). The empirical probability of co-infection (0.021) was almost equal to the expected one assuming the independence of infections (0.019) (Fisher's exact test: $P = 0.53$). The proportions of mussels shedding *R. fennica* significantly differed between temperature treatments with higher share at higher temperatures. The general test on 3×2 contingency table and all paired comparisons between treatments were significant even after applying the Bonferroni correction (Fisher's exact test: $P < 0.0002$, Table S4). For *R. campanula*, no significant differences between treatments were found ($P > 0.2$ in all cases, Table S4; Fig. S2). When double infections were included, results were similar (Table S4).

Cercarial release at the total host population level

At the host population level, the total cercarial emergence period (from the first to the last observation of emerged cercariae) of *R. fennica* ranged from 12 July to 28 October, whereas that of *R. campanula* ranged from 31 May to 3 October (Table 1; Fig. 1). The total period of cercarial shedding by *R. fennica* lasted for 10 weeks in the high, and 14–16 weeks in the intermediate, but for only 4–6 weeks in the low-temperature treatment (Table 1; Fig. 1). The total period of cercarial shedding by *R. campanula* in the low-temperature treatment was clearly longer than that of *R. fennica*, as it ranged from 14 to 18 weeks (Table 1). In other temperatures, differences in shedding duration between the parasites were not consistent (Table 1). At host population level, duration of cercarial shedding in *R. campanula* decreased, rather than increased, by temperature (Table 1). Water temperature at the time of the first emergence of *R. fennica* cercariae varied from 16.5 to 21.5°C, which was clearly higher than 10.5°C observed for *R. campanula* (Table 1; Fig. 1). The exact timing of the cercariae emergence for each mussel shedding cercariae is given in Figs S7 and S8.

Seasonal cercarial release with respect to temperature

Quantitatively the peak cercarial release by *R. fennica* co-occurred with the seasonal thermal maximum, but that of *R. campanula* clearly outran it (Fig. 1). Cercarial shedding by *R. fennica* increased substantially at temperatures above 15°C (Fig. 2A and B; Fig. S3) but high numbers of *R. campanula* cercariae were released as soon as the temperature exceeded 10°C (Fig. 2C and D; Fig. S3). For *R. fennica* a 'raw' relationship between temperature and cercariae production (Fig. 2A) shows abnormally low values of the cercariae emission at intermediate temperatures. These values can be explained by the fact that such temperatures could be met twice in a shedding season: in spring and in autumn. The GLM showed that temperature had a strong positive influence on the number of released cercariae (Table 2). The main effect of the season (i.e. day from the start of the experiment) was also positive at the average temperature (Table 2). However, significant interaction suggests that with time the influence of temperature decreased. The amount of variance explained by the model was remarkably high ($R^2 = 0.83$). The residual plot (Fig. 2B) showed that after accounting for the season, the relationship between temperature and cercariae production in *R. fennica* became almost linear. A similar model (Table S6) for *R. campanula* did not explain data better than the intercept-only model.

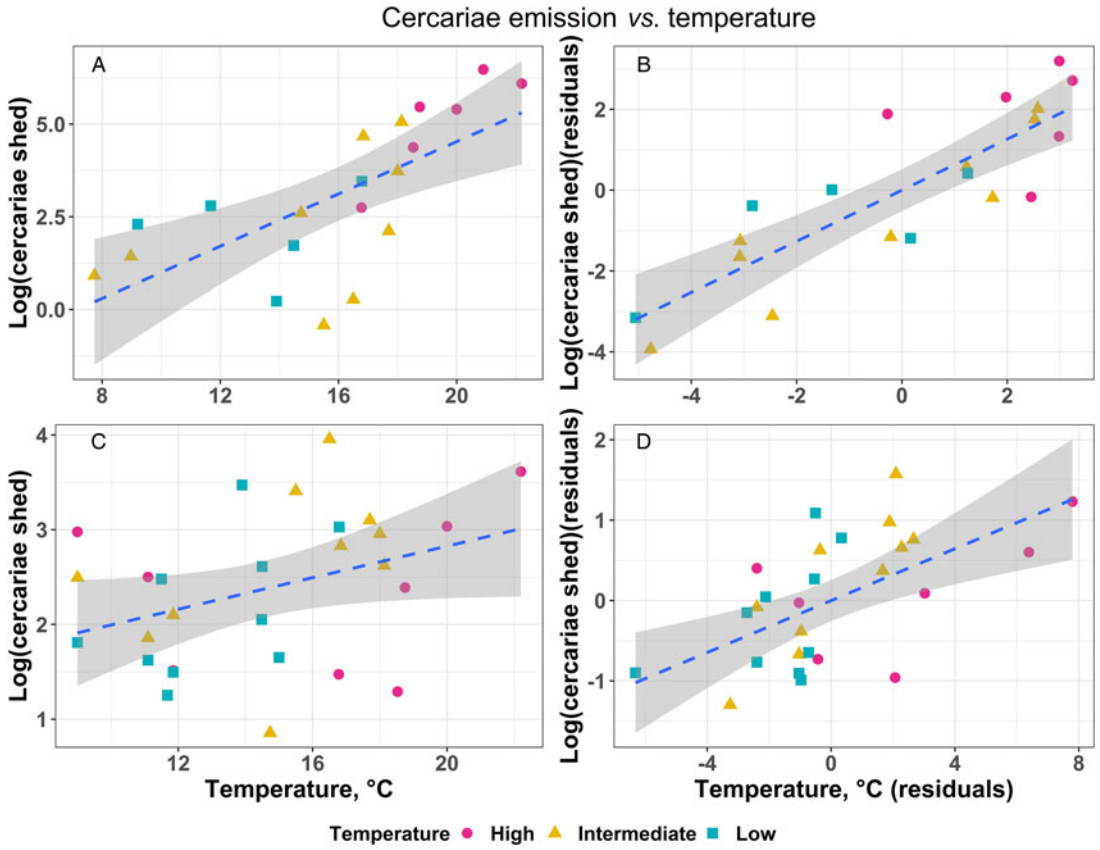

Fig. 2. Relationship between mean temperature during each 3-week monitoring period and release of *Rhipidocotyle fennica* (A, B) and *R. campanula* (C, D) cercariae by *Anodonta anatina* mussels (2 study populations combined). The average cercariae release by mussels from different temperature treatments indicated with dot (high), triangle (intermediate) and square (low temperature). (A, C) 'Raw' data. (B, D) Residual plot accounting for the seasonality.

However, exclusion of 1 outlier value (see Fig. S4B) resulted in a model where both main effects of seasonality and temperature were significant. With the increase in temperature, the emission of *R. campanula* also increased; however, the number of emitted cercariae falls with time (Table 2; Fig. 2C and D). There was no interaction between these 2 predictors ($F_1 = 0.38$, $P = 0.54$). For the similar models including zero values, see the Supplementary material (Tables S5 and S7; Figs S3 and S4A).

Cercarial release at the individual host level at different temperatures

For the sake of brevity, we present only the most important qualitative results here (summarized with statistics in Table 2), while the full statistical models are available in the Supplementary material. At high temperature, emission of *R. fennica* cercariae started (Table S8a; Fig. 3A and B) and ended (Table S8b; Fig. 3C and D) earlier than at low and intermediate temperatures (Table 2). The differences between low and intermediate temperature treatments were not significant (Tables S8a and S8b, respectively) after correcting for the multiple comparisons. Such a shift in the emission start and end did not result in a substantial emission duration change. Interestingly, in larger mussels, *R. fennica* cercariae emission started (Table S8a) and stopped (Table S8b) later than in smaller ones. However, although treatment × length interactions were non-significant, the estimates were negative and it seems that the size effect was reliable only in the high-temperature treatment in case of cercariae shedding start (Table S8a; Fig. 3B). Since many mussels appeared to emit cercariae only once during the observations, the emission duration for them was considered a zero, which demands using the zero-inflated model for emission duration analysis (see above). Although *R. fennica* cercarial release lasted for longer at the host population level in higher temperatures (see above), evidence for this was not sufficient at the individual host level. There was a tendency towards a slightly longer *R. fennica* cercariae emission duration in low-temperature treatment (Table S8c), but this did not hold after the Bonferroni correction. Importantly, temperature treatment had a substantial effect on the number of day-degrees needed to start the *R. fennica* cercariae release, with less day-degrees required under high-temperature treatment compared to intermediate and low ones (Fig. 4; Table S8d). Again, the difference between the 2 lower temperature treatments was not significant (Table S8d). The larger mussels needed more day-degrees to start *R. fennica* cercariae release (Table 2); however, this effect was reliably seen only in the high-temperature treatment.

Twenty-five out of 56 mussels releasing *R. campanula* cercariae had been already emitting them when the experiment started. Therefore, zero-inflated model was used to evaluate the influence of predictors on the emission start date in *R. campanula*. After the stepwise removal of non-significant terms, we found that the intercept-only model is the most parsimonious one. However, the AIC of the model, where the temperature is the only predictor, differs from the AIC of the intercept-only one by less than 2 points and, therefore, can be considered informative (Symonds and Moussalli, 2011). Therefore, we decided to present the results of the above-mentioned model (Table S8e) though they should be treated with great care. Essentially, in lower temperatures, *R. campanula* cercariae emission started later than in high temperature (estimate ± S.E. = 0.70 ± 0.27 and 0.59 ± 0.25, $z = 2.65$ and 2.34, $P = 0.008$ and 0.019 for low and intermediate temperature, respectively). The only predictor left in the model explaining the duration of cercariae emission in *R. campanula* was host length. Larger mussels tended to

Table 2. Comparison of cercarial shedding traits between *Rhipidocotyle* species; 2 populations of the host mussel *Anodonta anatina* combined

	R. fennica (RF)	*R. campanula* (RC)	Statistics (estimate ± s.e.)
Host population level			
n of shedding mussels	110	56	
Shedding period, days	108 (12 Jun–28 Oct)	125 (31 May–03 Oct)	
Shedding duration (H)[a], weeks	10	8–16	
Shedding duration (I)[a], weeks	14–16	16–18	
Shedding duration (L)[a], weeks	4–6	14–18	
Seasonal peak shedding	During peak T °C	Less tied to T °C	
N of released cercariae *vs* T °C	Positive correlation	Positive correlation	GLM (RF): 1.23 ± 0.20, $z = 5.99$, $P = 0.00002$; GLM (RC): 0.162 ± 0.045, $z = 3.60$, $P = 0.001$
N of released cercariae *vs* time[b]	Positive correlation	Negative correlation[c]	GLM (RF): 0.043 ± 0.01, $z = 4.54$, $P = 0.0003$; GLM (RC): −0.012 ± 0.004, $z = -2.96$, $P = 0.007$
N of released cercariae: T °C × time interaction	Temperature effect decreases with time	Interaction not significant	GLM (RF): −0.006 ± 0.002, $z = -3.14$, $P = 0.006$ GLM (RC) (nested models comparison): $F_1 = 0.38$, $P = 0.54$
Host individual level			
Emission start (time/T °C)	Later (mainly >15–20°C)	Earlier (about 10–12° C)	Zero-inflated model: 1.04 ± 0.10, $z = 10.01$, $P < 0.0001$, Table S9d Robust linear model: 10.13 ± 0.44, $F = 570.47$, $P < 0.0001$, Table S9d
Emission start (effect of T °C)	Earlier at higher T °C	Earlier at higher T °C	See Tables S8a and S8e for all details
Proportion of shedding mussels	Higher at high T °C	No effect	Fisher's exact test: $P < 0.0002$, Table S4
Emission start (effect of host size)	Later for large mussels[d]	No effect	GLM with log(DV) (RF): 0.011 ± 0.003, $z = 3.70$, $P = 0.0003$, Table S8a
Emission end (time/temperature)	Later (at similar T °C)	Earlier (at similar T °C)	GLM: 32.6 ± 4.6, $t = 7.1$, $P < 0.0001$. Table S9b/ns, Table S9f
Emission end (effect of T °C)	Earlier at higher T °C	Earlier at higher T °C	See Table S8b, for all details
Emission end (effect of host size)	Later for large mussels	Earlier for large mussels	GLM (RF): 0.34 ± 0.14, $t = 2.50$, $P = 0.014$; GLM (RC): −0.58 ± 0.27, $t = -2.13$, $P = 0.038$, Tables S8b and S8f
Emission duration	Shorter	Longer	Zero-inflated GLM: −0.28 ± 0.0021, $z = 0.47$, $P < 0.0001$, Table S9c
Emission duration (effect of T °C)	Absent	Absent	
Emission duration (host size effect)	No effect	Negative	GLM (RC): −0.66 ± 0.33, $t = -2.02$, $P = 0.048$
Emission start (day-degrees)	More	Less	GLM with log(DV): 1.58 ± 0.10, $t = 15.7$, $P < 0.0001$
E. start (day-degrees, effect of T °C)	Less needed at higher T	No effect	See Tables S8d and S8g for all treatment comparisons

[a]H, I, L = high-, intermediate- and low-temperature treatments, respectively.
[b]Time = days from the beginning of experiment.
[c]Exclusion of 1 outlier value (see Fig. S4B) resulted in a model where effects of seasonality were significant.
[d]This relationship did not hold for intermediate temperature (estimate ± s.e. = −0.011 ± 0.002, $z = -5.16$, $P < 0.0001$, Table S9a).

release cercariae for a shorter time though the relationship was only marginally significant ($P = 0.048$). This is probably because the larger mussels stopped cercariae emission earlier than smaller ones (Table S8f). Similar to *R. fennica*, emission of *R. campanula* cercariae tended to stop later under intermediate and low temperatures; however, only the difference between low- and high-temperature treatments was statistically significant (Table S8f). A substantial share of mussels started to shed *R. campanula* cercariae simultaneously already in the beginning of the experiment, thus having the same number of day-degrees (115) to start shedding. To account for it, we fitted a zero-inflated model using 115 day-degrees as a zero value to explain variation in day-degrees required for start of *R. campanula* cercariae shedding. After simplification, the model contained only the host population as a predictor, while no significant effects were found (Table S8g).

Survival of the mussels through the experiment was 64.2% (see Figs S5, S7 and S8). The mortality of mussels shedding cercariae was significantly higher in the high-temperature treatment, while in other 2 treatments mortality was equal (Table S8h). More detailed survival analysis will be published elsewhere.

Cercarial release at the individual host level and differences between parasite species

There was a significant difference between the parasite species with respect to the 5 cercarial shedding traits studied: start date, water temperature and number of day-degrees at the start, stop date and duration of cercarial release (Table S9). However, the temperature effect on these traits was generally similar for both parasite species. First of all, the release of cercariae by *R. fennica*

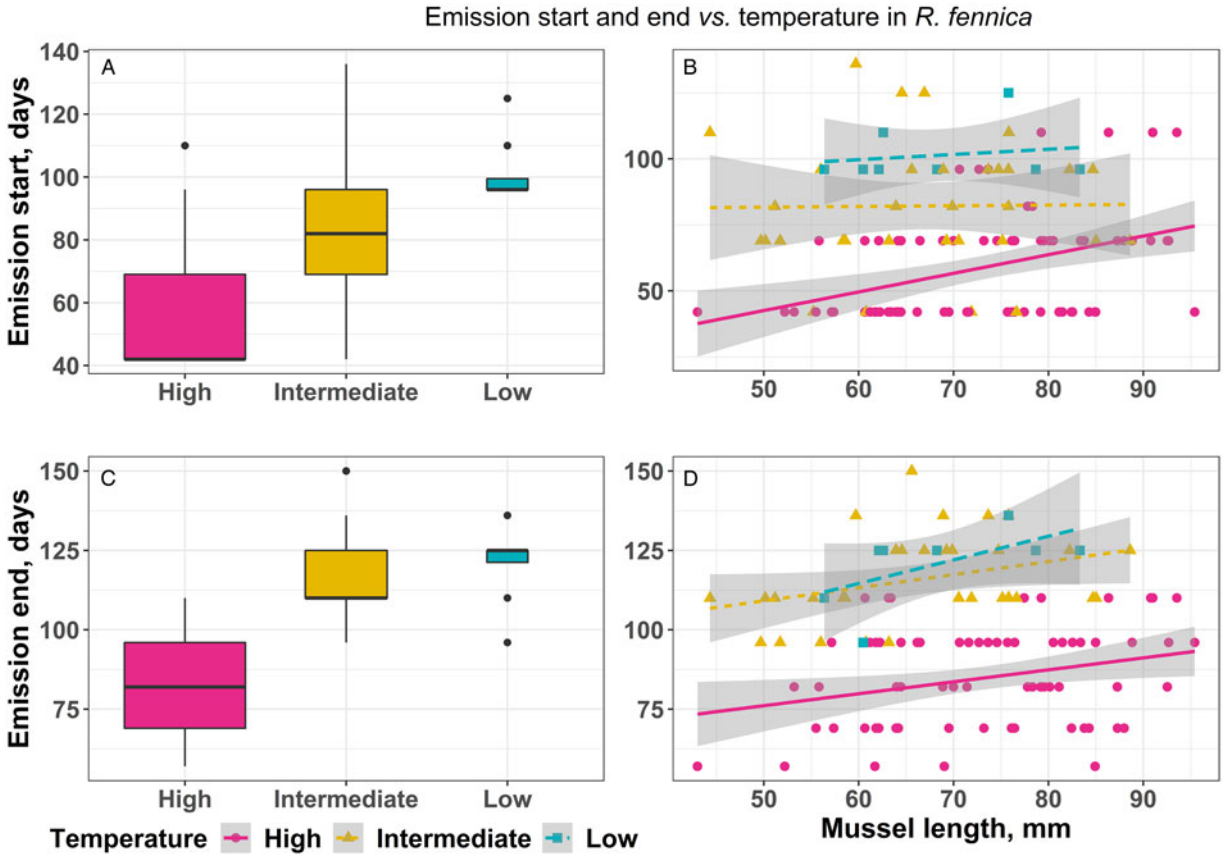

Fig. 3. (A, C) Boxplots for number of days needed to start (A) and stop (C) cercariae release in *Rhipidocotyle fennica* under different temperature treatments (2 study populations combined). Width of box reflects the sample size, height of box denotes limits of upper and lower quartiles, horizontal line is the median of vales, whiskers mark the highest and lowest values within the 1.5 × interquartile range and dots indicate values outside that range. (B, D) Respective plots accounting for size of the mussel host, *Anodonta anatina*, where high temperature marked with dots and continuous line, intermediate temperature with triangles and dotted line, and low temperature with squares and broken line.

started seasonally significantly later than that by *R. campanula* (Table S9a) with the treatment-specific difference varying from 42 to 87 days. In fact, this difference is likely to be even more pronounced since many *R. campanula*-infected mussels had already started cercariae emission at the beginning of the experiment. Lower temperatures led to later seasonal start of cercariae emission in both species (Table S9a). *Rhipidocotyle fennica* stopped cercariae release generally later than *R. campanula*, but lower temperature caused a later stop of cercariae release in both species (Table S9b). Interestingly, the interaction between seasonal timing – and partly also the duration – of cercariae release and the host's length differed in the 2 parasite species. Larger mussels infected with *R. campanula* ended release earlier (leading to a negative trend between host size and duration of cercariae emission; Table S9c), while those infected with *R. fennica* both started and ended cercariae release later than the smaller ones, at least in high temperature (leading to mainly positive trend between host size and duration of cercariae emission; Table S9c). The

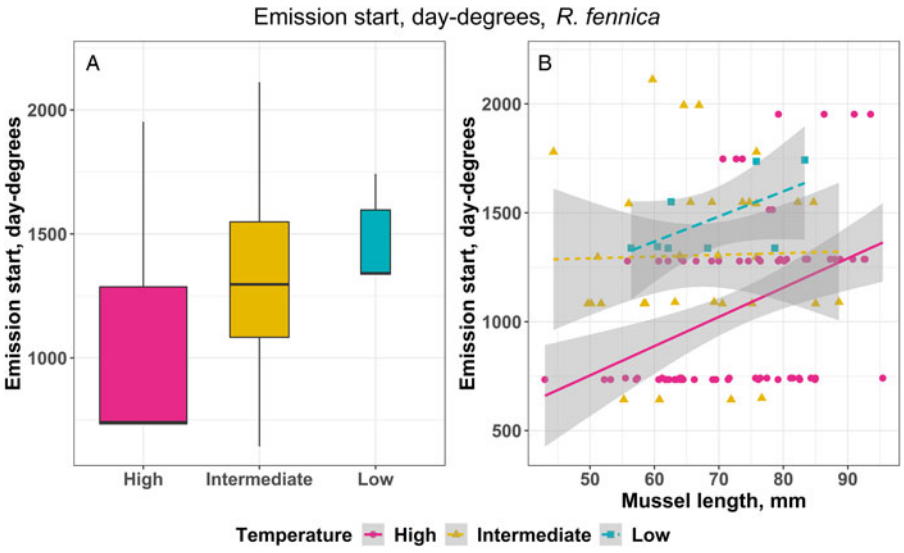

Fig. 4. Sum of day-degrees needed to start release of *Rhipidocotyle fennica* cercariae by the mussel host *Anodonta anatina* (2 study populations combined); boxplot for different temperature treatments (A) and respective plot accounting for size of host (B). For explanation of boxplot details and plot symbols, see Fig. 3.

total duration of cercarial release was shorter for *R. fennica* than for *R. campanula* (Table S9c) and this difference is likely to be underestimated taking into account that a large share of *R. campanula*-infected mussels had already started cercariae shedding in the beginning of the experiment.

The water temperature at the start of the seasonal release of cercarial was higher for *R. fennica* (15–20°C) and lower for *R. campanula* (10–12°C) (Fig. S6; Table S9d); however, this effect became less pronounced under intermediate and low temperatures. For both species, the number of day-degrees needed to start cercariae shedding was larger in intermediate- and low-temperature treatments comparing to the high-temperature one (Table S9e; Fig. S6). Finally, *R. fennica* needed much more day-degrees to start the cercariae release but there were no interspecific differences in the temperatures at which cercariae release stopped in any temperature treatments (Fig. S6; Table S9f).

Discussion

Ongoing and predicted increases in global temperatures and in duration of the growing season will have important implications for many host–parasite systems, e.g. changes in the timing of parasite life-cycle stages (Marcogliese, 2001; Mouritsen and Poulin, 2002; Kutz *et al*., 2005; MacDonald *et al*., 2021). A common expectation is that the seasonal larval release by parasites will start earlier and become more prolonged as a consequence of increased thermal growing season (Marcogliese, 2001; Harvell *et al*., 2009; Nikolaev *et al*., 2020).

The present study on the variation in the seasonal cercarial shedding patterns of 2 sympatric parasites gives mixed evidence both for and against the aforementioned hypothesis. At the individual host level, high temperature caused a marked shift in the cercariae release period towards earlier start and end of cercariae emission, but – against the expectation – did not result in a longer seasonal duration of cercariae release within an individual host. Such a shift can have important consequences in natural habitats leading to a temporal mismatch in occurrence between hosts and parasites (Lõhmus and Björklund, 2015; Cohen *et al*., 2017; Gehman *et al*., 2018), thus, reducing the infection load (Paull and Johnson, 2014; McDevitt-Galles *et al*., 2020). However, the total period of cercarial shedding by *R. fennica* at the host population level, from the first to the last observation of emergence, was much longer in high and intermediate-temperature treatments compared with the low-temperature one – as expected by the general hypothesis. Such a population-level effect was not observed in *R. campanula* probably because about a half of *R. campanula*-infected mussels had already started shedding cercariae when the experiment begun. In addition, though the temperature did not influence the duration of cercariae release at the individual level, it positively correlated with the numbers of produced cercariae in both species, thus, potentially giving the parasite an advantage in the competition with a host for the host's resources. Importantly, under similar temperatures, fewer cercariae were emitted in the end compared with the beginning of the emission season, which is likely to be a result of exhaustion of the host and/or parasite. Therefore, the present results support the view that climate warming would increase the duration of larval shedding, and lengthen the transmission period by parasites, but that such lengthening is produced by increased variation between host individuals, rather than due to a lengthened individual shedding period.

The results also indicated that even closely related, sympatric parasite species that share the same transmission pathway can respond differently to temperature change (see also Selbach and Poulin, 2020). High variability in cercariae shedding patterns (e.g. daily rhythms) have been found previously both within a single trematode genus (Vyhlídalová and Soldánová, 2020) and species (Théron and Combes, 1988; Riley and Uglem, 1995; Le Clec'h *et al*., 2021). *Rhipidocotyle fennica* brought forward the start of seasonal cercarial release and started cercarial release with lower day-degrees in the high-temperature treatment, but for *R. campanula* this was not found. Furthermore, *R. campanula* clearly started the seasonal cercarial release earlier, at a lower temperature than *R. fennica* (10 and 15°C, respectively), with fewer day-degrees, stopped the seasonal cercarial release earlier, and had a markedly longer total seasonal duration of cercarial emission. The peak release of *R. fennica* cercariae occurred during the warmest weeks in concordance with field observations (Taskinen *et al*., 1994), and also the proportion of mussels shedding *R. fennica* cercariae was higher in higher temperature treatments, while in *R. campanula* this phenomenon was not found. Based on these observations, one might predict that the projected longer summers and higher temperatures associated with climate warming (Tietäväinen *et al*., 2010; Ruosteenoja *et al*., 2011) would benefit especially *R. fennica*, which requires higher temperatures (as well as more day-degrees) to start cercariae release. On the other hand, it can be predicted that *R. campanula* could thrive better than *R. fennica* in colder, more northern, short-summer environments, where the early onset of cercarial release is presumably advantageous.

The earlier start of the seasonal cercarial release by *R. campanula* is difficult to explain by the transmission dynamics, as the 2 species share the same current (the bivalve *A. anatina*) and next (the fish *R. rutilus*) host in their life cycles (Taskinen *et al*., 1991; Gibson *et al*., 1992). It is also worth noting that both *R. fennica* and *R. campanula* are specific only to *A. anatina* as their first intermediate host in the study area (Taskinen *et al*., 1991; Gibson *et al*., 1992). The definitive hosts of *R. fennica* and *R. campanula* are the predatory fishes northern pike (*E. lucius*) and perch/pikeperch (*P. fluviatilis*/*S. lucioperca*), respectively (Taskinen *et al*., 1991; Gibson *et al*., 1992). Thus, it is also possible that the timing of cercarial shedding could be an adaptation to increase transmission to the final hosts, such as the differential seasonal feeding of the final hosts on roach that we are not aware of. However, it is difficult to believe that the earlier start of cercarial release by *R. campanula* could be an adaptation only to northern conditions (although it might facilitate occurrence there) because both *R. campanula* and *R. fennica* occur as far south as Ukraine (Taskinen *et al*., 1991; Petkevičiūtė *et al*., 2014; Stunžėnas *et al*., 2014; Müller *et al*., 2015).

We propose that the mechanism enabling the early onset of cercarial release by *R. campanula* is that they have their cercarial production machinery 'on standby' throughout the year (Taskinen *et al*., 1994). Fully developed, mature cercariae are found in *R. campanula* sporocysts in high proportions in all seasons, readily available for shedding when a suitable temperature is attained (Taskinen *et al*., 1994). In *R. fennica*, mature, ready-to-emerge cercariae are only found during the cercarial shedding period (Taskinen *et al*., 1994). This probably means that it takes a relatively long time for *R. fennica* to respond to increasing water temperature in terms of cercarial production, as the growth of sporocyst starts from practically zero in spring (Taskinen *et al*., 1994). Cercarial release by *Rhipidocotyle* spp. can also be triggered outside the natural shedding period by transfering infected mussels to high temperature in the laboratory, but also in that case the time needed for *R. campanula* to start shedding cercariae is much shorter than for *R. fennica* (Taskinen *et al*., 1991).

Although our results about the effect of temperature on shedding start and proportion of mussels shedding cercariae predict that warming is likely to benefit *R. fennica* more than *R. campanula*, the infection success of parasites may also depend on cercarial output, their survival and infectivity. Temperature effects on

these traits can partly compensate for each other, sometimes resulting in similar transmission efficiency at different temperatures (Poulin, 2006). However, it is impossible to account for all these effects in our study.

Two studied parasitic species also differed in their relationships to the host size. Larger *R. fennica*-infected mussels tended to start cercariae release later and shed them for a longer time, while in *R. campanula* no clear relationships were seen. Such a difference hints again that even closely related sympatric parasitic species infecting the same host can differ in strategies of host exploitation. Surprisingly, there were almost no differences in cercariae shedding traits in parasites from different populations, which suggests that our study comprises a reasonable amount of generality.

It is important to notice that, although we explained the obtained results by temperature and seasonality effects, there were differences between the treatments also in terms of water flow and water source (littoral *vs* hypolimnetic), light conditions, temperature fluctuation and seasonal temperature profile. Whereas the high-temperature tanks were kept in an outdoor shelter and were subject to a diurnal temperature fluctuation and natural light, the intermediate- and low-temperature tanks were kept in an indoor tank hall and illuminated with artificial light. However, the photoperiod was equal in all treatments and corresponded to the natural rhythm. In addition, the cercarial release was shown to be similar at constant and diurnally variable temperatures (Roushdy, 1984). According to Choo and Taskinen (2015), a short-term (1 h) temperature increase triggers cercariae emission of *R. fennica* from *A. anodonta*, but on the other hand a similar decrease in temperature results in an equivalent decrease in cercariae release. Therefore, the higher daily fluctuation of temperature in the high-temperature treatment probably did not influence the net daily cercariae release. In addition, the monitoring of cercariae release was performed at constant temperature (no variation within a day) in all temperature treatments. Importantly, the mussels in high-temperature treatment supplied with littoral water could receive more food than those in colder treatments, supplied with water from the lake hypolimnion, although we aimed at compensating this by doubling the water flow in the cold- and intermediate-temperature treatments (see section Methods).

Since food deprivation of host can constrain cercariae release (e.g. Seppälä *et al.*, 2015), higher cercariae output in high-temperature treatment obtained in our study could be, e.g. a result of higher food availability for mussel host. However, our data on mussel mortality and weight change during the experiment (i.e. higher mortality in high-temperature treatment, no increase in mussel weight) do not indicate the importance of nutritional differences for the interpretation of the results. Even though we cannot completely rule out confounding factors other than temperature, we do not believe that the difference in water and light source, or temperature fluctuation, could explain the observed contrasting responses in the seasonal cercarial release by *R. fennica* and *R. campanula* between the temperature treatments. New infections of mussels during the experiment, *via* miracidia from unfiltered lake water, were unlikely due to the seasonal maturing of *Rhipidocotyle* trematodes in late autumn (Taskinen *et al.*, 1991). Thus, the present results should reliably indicate temperature responses in the seasonal timing of cercarial shedding by *R. fennica* and *R. campanula.*

Previous studies investigating the seasonal dynamics of trematode cercarial release include field observations showing a significant increase in cercarial emergence during summer months (Taskinen *et al.*, 1994; Taskinen, 1998*a*; Fingerut *et al.*, 2003), a longer seasonal shedding period in water bodies receiving thermal effluents (Aho *et al.*, 1982) and experimental evidence on the role of temperature in controlling daily cercariae output (Koprivnikar and Poulin, 2009*a*; Vyhlídalová and Soldánová, 2020), the start and the duration of cercariae emergence (Taskinen *et al.*, 1991; Fingerut *et al.*, 2003; Paull and Johnson, 2014; Prokofiev *et al.*, 2016). Long-term experimental studies like the present one are still scarce (Paull and Johnson, 2014).

The results of this study partly support the idea that climate warming would increase the seasonal duration of larval shedding by parasites, but emphasize species-specific differences in the seasonal cercarial release and transmission with respect to warming (Marcogliese, 2001; Harvell *et al.*, 2009). Research on the geographic distribution of the species is needed to determine whether the observed temperature differences in cercarial shedding traits affect the current distribution and relative abundance of *Rhipidocotyle* species at the northern boundary of their occurrence. Due to the contrasting species-specific temperature-dependence, the *A. anatina–Rhipidocotyle* spp. association offers a unique system to study the effects of the ongoing and predicted climate warming on host–parasite relationships at high latitudes.

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Data. Data can be requested from the authors.

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Conflict of interest. None.

Ethical standards. None.

References

Aho JM, Camp JW and Esch GW (1982) Long-term studies on the population biology of *Diplostomulum scheuringi* in a thermally altered reservoir. *Journal of Parasitology* **68**, 695–708.

Baptiste A (2017) gridExtra: miscellaneous functions for 'Grid' graphics. R package version 2.3. Available at https://CRAN.R-project.org/package=gridExtra.

Choo JM and Taskinen J (2015) Effect of short-term temperature change on cercarial release by *Rhipidocotyle fennica* (Trematoda, Bucephalidae) from the freshwater bivalve host, *Anodonta anatina*. *Ecological Parasitology and Immunology* **4**, 235932.

Chubb JC (1979) Seasonal occurrence of helminths in freshwater fishes. Part II. Trematoda. *Advances in Parasitology* **17**, 141–313.

Cohen JM, Venesky MD, Sauer EL, Civitello DJ, McMahon TA, Roznik EA and Rohr JR (2017) The thermal mismatch hypothesis explains host susceptibility to an emerging infectious disease. *Ecology Letters* **20**, 184–193.

Cribb TH, Bray RA and Littlewood TJ (2001) The nature and evolution of the association among digeneans, molluscs and fishes. *International Journal for Parasitology* **31**, 997–1011.

De Montaudouin X, Blanchet H, Desclaux-Marchand C, Lavesque N and Bachelet G (2016) Cockle infection by *Himasthla quissetensis* – I. From cercariae emergence to metacercariae infection. *Journal of Sea Research* **113**, 99–107.

Fingerut JT, Zimmer CA and Zimmer RK (2003) Patterns and processes of larval emergence in an estuarine parasite system. *Biological Bulletin* **205**, 110–120.

Galaktionov KV (2017) Patterns and processes influencing helminth parasites of Arctic coastal communities during climate change. *Journal of Helminthology* **91**, 387–408.

Gehman AM, Hall RJ and Byers JE (2018) Host and parasite thermal ecology jointly determine the effect of climate warming on epidemic dynamics. *PNAS* **115**, 744–749.

Gibson DI, Taskinen J and Valtonen ET (1992) Studies on bucephalid digeneans parasitising molluscs and fishes in Finland. II. The description of *Rhipidocotyle fennica n. sp*. and its discrimination by principal components analysis. *Systematic Parasitology* **23**, 67–79.

Hannon E, Calhoun D, Chadalawada S and Johnson P (2017) Circadian rhythms of trematode parasites: applying mixed models to test underlying patterns. *Parasitology* **145**, 1–9.

Harvell D, Altizer S, Cattadori IM, Harrington L and Weil E (2009) Climate change and wildlife diseases: when does the host matter the most? *Ecology* **90**, 912–920.

Hothorn T, Bretz F and Westfall P (2008) Simultaneous inference in general parametric models. *Biometrical Journal* **50**, 346–363.

IPCC (2014) *Climate Change 2014: Synthesis Report. Contribution of Working Groups I, II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change*. [Core Writing Team, Pachauri RK and Meyer LA (eds.)]. Geneva, Switzerland: IPCC.

Johnson PT, Dobson A, Lafferty KD, Marcogliese DJ, Orlofske SA, Poulin R and Thieltges DW (2010) When parasites become prey: ecological and epidemiological significance of eating parasites. *Trends in Ecology and Evolution* **25**, 362–371.

Jokela J, Taskinen J, Mutikainen P and Kopp K (2005) Virulence of parasites in hosts under environmental stress: experiments with anoxia and starvation. *OIKOS* **108**, 156–164.

Jorgensen CB, Kiorboe T, Mohlenberg F and Riisgard HU (1984) Ciliary and mucus-net filter feeding, with special reference to fluid mechanical characteristics. *Marine Ecology Progress Series* **15**, 283–292.

Jylhä K, Tuomenvirta H and Ruosteenoja K (2004) Climate change projections for Finland during the 21st century. *Boreal Environmental Research* **9**, 127–152.

Karvonen A, Seppälä O and Valtonen ET (2004) Parasite resistance and avoidance behaviour in preventing eye fluke infections in fish. *Parasitology* **129**, 159–164.

Koprivnikar J and Poulin R (2009*a*) Interspecific and intraspecific variation in cercariae release. *Journal of Parasitology* **95**, 14–19.

Koprivnikar J and Poulin R (2009*b*) Effects of temperature, salinity, and water level on the emergence of marine cercariae. *Parasitology Research* **105**, 957–965.

Kuha J, Arvola L, Hanson PC, Huotari J, Huttula T, Juntunen J, Järvinen M, Kallio K, Ketola M, Kuoppamäki K, Lepistö A, Lohila A, Paavola R, Vuorenmaa J, Winslow L and Karjalainen J (2016) Response of boreal lakes to episodic weather-induced events. *Inland Waters* **6**, 523–534.

Kuris AM, Hechinger RF, Shaw JC, Whitney KL, Aguirre-Macedo L, Boch CA, Dobson AP, Dunham EJ, Fredensborg BL, Huspeni TC, Lorda J, Mababa L, Mancini FT, Mora AB, Pickering M, Talhouk NL, Torchin ME and Lafferty R (2008) Ecosystem energetic implications of parasite and free-living biomass in three estuaries. *Nature* **454**, 515–518.

Kutz SJ, Hoberg EP, Polley L and Jenkins EJ (2005) Global warming is changing the dynamics of Arctic host-parasite systems. *Proceedings of the Royal Society Biological Sciences* **272**, 2571–2576.

Le Clec'h W, Chevalier FD, McDew-White M, Menon V, Arya GA and Anderson T (2021) Genetic architecture of transmission stage production and virulence in schistosome parasites. *Virulence* **12**, 1508–1526.

Lõhmus M and Björklund M (2015) Climate change: what will it do to fish–parasite interactions? *Biological Journal of the Linnean Society* **116**, 397–411.

MacDonald H, Akçay E and Brisson D (2021) The role of host phenology for parasite transmission. *Theoretical Ecology* **14**, 1–21.

Maechler M (2021) sfsmisc: Utilities from 'Seminar fuer Statistik' ETH Zurich. R package version 1.1-11. Available at https://CRAN.R-project.org/package=sfsmisc.

Marcogliese DJ (2001) Implications of climate change for parasitism of animals in the aquatic environment. *Canadian Journal of Zoology* **79**, 1331–1352.

Marcogliese DJ (2016) The distribution and abundance of parasites in aquatic ecosystems in a changing climate: more than just temperature. *Integrative and Comparative Biology* **56**, 611–619.

McDevitt-Galles T, Moss W, Calhoun D and Johnson P (2020) Phenological synchrony shapes pathology in host–parasite systems. *Proceedings of the Royal Society B: Biological Sciences* **287**, 20192597.

McKee KM, Koprivnikar J, Johnson PTJ and Arts MT (2020) Parasite infectious stages provide essential fatty acids and lipid-rich resources to freshwater consumers. *Oecologia* **192**, 477–488.

Mironova E, Gopko M, Pasternak A, Mikheev V and Taskinen J (2019) Trematode cercariae as prey for zooplankton: effect on fitness traits of predators. *Parasitology* **146**, 105–111.

Mironova E, Gopko M, Pasternak A, Mikheev V and Taskinen J (2020) Cyclopoids feed selectively on free-living stages of parasites. *Freshwater Biology* **65**, 1450–1459.

Morley NJ and Lewis JW (2013) Thermodynamics of cercarial development and emergence in trematodes. *Parasitology* **140**, 1211–1224.

Morley N, Adam M and Lewis J (2010) The effects of host size and temperature on the emergence of *Echinoparyphium recurvatum* cercariae from *Lymnaea peregra* under natural light conditions. *Journal of Helminthology* **84**, 317–326.

Mouritsen KN and Poulin R (2002) Parasitism, climate oscillations and the structure of natural communities. *OIKOS* **97**, 462–468.

Müller T, Czarnoleski M, Labecka AM, Cichy A, Zając K and Dragosz-Kluska D (2015) Factors affecting trematode infection rates in freshwater mussels. *Hydrobiologia* **742**, 59–70.

Nikolaev K, Levakin I and Galaktionov K (2020) Seasonal dynamics of trematode infection in the first and the second intermediate hosts: a long-term study at the subarctic marine intertidal. *Journal of Sea Research* **164**, 101931.

Orlofske SA, Jadin RC, Preston DL and Johnson PTJ (2012) Parasite transmission in complex communities: predators and alternative hosts alter pathogenic infections in amphibians. *Ecology* **93**, 1247–1253.

Paull SH and Johnson PT (2014) Experimental warming drives a seasonal shift in the timing of host-parasite dynamics with consequences for disease risk. *Ecology Letters* **17**, 445–453.

Paull SH, Raffel TR, LaFonte BE and Johnson PTJ (2015) How temperature shifts affect parasite production: testing the roles of thermal stress and acclimation. *Functional Ecology* **29**, 941–950.

Petkevičiūtė R, Stunžėnas V and Stanevičiūtė G (2014) Differentiation of European freshwater bucephalids (Digenea: Bucephalidae) based on karyotypes and DNA sequences. *Systematic Parasitology* **87**, 199–212.

Poulin R (2006) Global warming and temperature-mediated increases in cercarial emergence in trematode parasites. *Parasitology* **132**, 143–151.

Preston DL, Orlofske SA, Lambden JP and Johnson PTJ (2013) Biomass and productivity of trematode parasites in pond ecosystems. *Journal of Animal Ecology* **82**, 509–517.

Prokofiev V, Galaktionov KV and Levakin IA (2016) Patterns of parasite transmission in polar seas: daily rhythms of cercarial emergence from intertidal snails. *Journal of Sea Research* **113**, 85–98.

R Core Team (2020) *R: A Language and Environment for Statistical Computing*. Vienna, Austria: R Foundation for Statistical Computing. Available at https://www.R-project.org/.

Riley M and Uglem G (1995) *Proterometra macrostoma* (Digenea: Azygiidae): variations in cercarial morphology and physiology. *Parasitology* **110**, 429–436.

Rosen R, Blank S, Akabogu F, Hauschner R, Meneses S, Slater O, Melton A, Tetidrick C and Gosnell W (2018) Effect of temperature on the release of *Proterometra macrostoma* (Trematoda: Digenea) cercariae from their snail intermediate host, *Pleurocera semicarinata* (Gastropoda: Pleuroceridae) at North Elkhorn Creek, Kentucky. *Comparative Parasitology* **85**, 202–207.

Roushdy MZ (1984) The effect of diurnal fluctuating temperature on the development of *Schistosoma haematobium* in *Bulinus truncatus*. *Journal of the Egyptian Society of Parasitology* **14**, 507–514.

Ruosteenoja K, Räisänen J and Pirinen P (2011) Projected changes in thermal seasons and the growing season in Finland. *International Journal of Climatology* **31**, 1473–1487.

Selbach C and Poulin R (2020) Some like it hotter: trematode transmission under changing temperature conditions. *Oecologia* **194**, 745–755.

Seppälä O, Louhi KR, Karvonen A, Rellstab C and Jokela J (2015) Relative reproductive success of co-infecting parasite genotypes under intensified within-host competition. *Infection, Genetics and Evolution: Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases* **36**, 450–455.

Sharma S, Jackson DA, Minns CK and Shuter BJ (2007) Will northern fish populations be in hot water because of climate change? *Global Change Biology* **13**, 2052–2064.

Shim KC, Koprivnikar J and Forbes MR (2013) Variable effects of increased temperature on a trematode parasite and its intertidal hosts. *Journal of Experimental Marine Biology and Ecology* **439**, 61–68.

Studer A and Poulin R (2014) Analysis of trait mean and variability versus temperature in trematode cercariae: is there scope for adaptation to global warming? *International Journal for Parasitology* **44**, 403–413.

Studer A, Thieltges DW and Poulin R (2010) Parasites and global warming: net effects of temperature on an intertidal host–parasite system. *Marine Ecology Progress Series* **415**, 11–22.

Stunžėnas V, Petkevičiūtė R, Stanevičiūtė G and Binkienė R (2014) *Rhipidocotyle fennica* (Digenea: Bucephalidae) from *Anodonta anatina* and pike *Esox lucius* in Lithuania. *Parasitology Research* **113**, 3881–3883.

Symonds MRE and Moussalli A (2011) A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behavioral Ecology and Sociobiology* **65**, 13–21.

Taskinen J (1998*a*) Cercarial production of the trematodes *Rhipidocotyle fennica* kept in the field. *Journal of Parasitology* **84**, 345–349.

Taskinen J (1998*b*) Influence of trematode parasitism on the growth of a bivalve host in the field. *International Journal of Parasitology* **28**, 599–602.

Taskinen J and Valtonen ET (1995) Age-, size-, and sex-specific infection of *Anodonta piscinalis* (Bivalvia: Unionidae) with *Rhipidocotyle fennica* (Digenea: Bucephalidae) and its influence on host reproduction. *Canadian Journal of Zoology* **73**, 887–897.

Taskinen J, Valtonen ET and Gibson DI (1991) Studies on bucephalid digeneans parasitizing molluscs and fishes in Finland I. Ecological data and experimental studies. *Systematic Parasitology* **19**, 81–94.

Taskinen J, Valtonen ET and Mäkelä T (1994) Quantity of sporocysts and seasonality of two *Rhipidocotyle species* (Digenea: Bucephalidae) in *Anodonta piscinalis* (Mollusca: Bivalvia). *International Journal of Parasitology* **24**, 877–886.

Taskinen J, Mäkelä T and Valtonen ET (1997) Exploitation of *Anodonta piscinalis* (Bivalvia) by trematodes: parasite tactics and host longevity. *Annales Zoologici Fennici* **34**, 37–46.

Théron A and Combes C (1988) Genetic analysis of cercarial emergence rhythms of *Schistosoma mansoni*. *Behavior Genetics* **18**, 201–209.

Thieltges DW and Rick J (2006) Effects of temperature on emergence, survival and infectivity of cercariae of the marine trematode *Renicola roscovita* (Digenea: Renicolidae). *Diseases of Aquatic Organisms* **73**, 63–68.

Thieltges DW, de Montaudouin X, Fredensborg B, Jensen KT, Koprivnikar J and Poulin R (2008) Production of marine trematode cercariae: a potentially overlooked path of energy flow in benthic systems. *Marine Ecology Progress Series* **372**, 147–155.

Tietäväinen H, Tuomenvirta H and Venäläinen A (2010) Annual and seasonal mean temperatures in Finland during the last 160 years based on gridded temperature data. *International Journal of Climatology* **30**, 2247–2256.

Vaughn CC (2018) Ecosystem services provided by freshwater mussels. *Hydrobiologia* **810**, 15–27.

Vyhlídalová T and Soldánová M (2020) Species-specific patterns in cercarial emergence of *Diplostomum* spp. from snails *Radix lagotis*. *International Journal for Parasitology* **50**, 1177–1188.

Wickham H (2016) *ggplot2: Elegant Graphics for Data Analysis*. New York, USA: Springer Verlag.

Wilke C (2020) cowplot: Streamlined Plot Theme and Plot Annotations for 'ggplot2'. R package version 1.1.1. Available at https://CRAN.R-project.org/package=cowplot.

Zeileis A, Kleiber C and Jackman S (2008) Regression models for count data in R. *Journal of Statistical Software* **27**, 1–25.