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1 Chapter 23. Diplostomiasis (*Diplostomum spathaceum* and related species)

2

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13

14 **23.1 Introduction**

15

16 *Diplostomum* spp. (Trematoda) are widespread parasites of freshwater and brackish water

17 fishes (Chappell, 1995), and they infect different parts of the fish eyes such as lens, humour

18 and retina (Marcogliese *et al.*, 2001a; Karvonen *et al.*, 2006b; Désilets *et al.*, 2013; Padros *et

19 al.*, 2018). In the ecological literature, species infecting the lens are commonly grouped as a

20 single species, *Diplostomum spathaceum*. However, morphological (Niewiadomska, 1984,

21 1986; Niewiadomska and Kiseliene, 1994) and particularly molecular studies (Niewiadomska

22 and Laskowski, 2002; Locke *et al.*, 2010a; Locke *et al.*, 2010b; Rellstab *et al.*, 2011; Blasco-

23 Costa *et al.*, 2014; Locke *et al.*, 2015) indicated that *Diplostomum* is a species complex with a

24 number of different species infecting specific parts of fish eyes. However, details of life

25 histories and ecological differences of many of the species are not known. Our present focus is

26 on species infecting the lens and causing diplostomiasis. Most of the published literature is on

27 *D. spathaceum* (in the light of the current knowledge possibly including more than one species)

28 or *D. pseudospathaceum* (the species was described by Niewiadomska (1984) and verified by

29 Niewiadomska and Laskowski (2002) using molecular techniques). For simplicity and due to

30 the lack of data, we assume here that effects of all parasite species possibly co-infecting a lens

31 are similar and they respond roughly the same way to climate change.

32

33 The life cycle of lens-infecting *Diplostomum* spp. includes an avian definitive host, a molluscan

34 first intermediate host and fish second intermediate host (Chappell *et al.*, 1994; Karvonen,

35 2012) (Fig. 23.1). Parasites reproduce sexually in the gut of the bird and start producing eggs

36 in three days after establishment (Chappell *et al.*, 1994). Eggs are released into the aquatic

37 environment through bird faeces. They hatch to free-swimming miracidia that are non-feeding

38 and short-lived stages, which infect the molluscan intermediate host. This is typically a snail

39 of the genus *Lymnaea*. Within a snail, each miracidium gives rise to a mother sporocyst, which

40 then replicates asexually to multiple daughter sporocysts. Larval cercariae are formed in the

41 sporocyst through asexual reproduction. Thus, cercariae from a single-miracidial infection in a

42 snail are genetically identical. However, one snail can be infected with multiple miracidia

43 (Rauch *et al.*, 2005; Louhi *et al.*, 2013) and produce cercariae of different genotypes at the

44 same time. Cercariae are released from an infected snail to surrounding water in very high

45 numbers (Lyholt and Buchmann, 1996; Karvonen *et al.*, 2004a). Free-swimming cercariae in

46 the water column do not feed after leaving the snail, but rely on glycogen reserves, which last

47 for approximately 24 hours (Karvonen *et al.*, 2003). Afterwards, cercariae lose their infectivity

48 and die. If a cercaria encounters a fish, it penetrates gills or skin and migrates as a

49 diplostomulum to the eye lens. Details of the route and mechanisms of migration are unknown

50 (Ratanarat-Brockelman, 1974). The migration typically takes place within 24 h, but this

51 depends on the water temperature (Lyholt and Buchmann, 1996). Diplostomulum that fails to

52 complete the migration exhausts its energy reserves and is eliminated by the fish immune

53 system. Those that reach the lens are at least partly protected from the host immune system, as

54 the eye lens is not directly connected to blood circulation of the fish. In the lens, parasites

55 develop to the final larval stages, metacercariae, within a few weeks and afterwards can

56 probably survive in the lens for years. Consequently, numbers of metacercariae in fish tend to

57 increase with time (Marcogliese *et al.*, 2001b). The life cycle is completed when a fish-eating

58 bird consumes an infected fish.

59

60 INSERT FIGURE 23.1 HERE

61 Figure 23.1 Life cycle of *Diplostomum* spp. includes three hosts. Parasites mature in the

62 intestine of a definitive host (1) and release eggs (2) into the aquatic environment with faeces.

63 Eggs hatch into miracidia (3) that seek the first intermediate host, an aquatic snail. Within the

64 snail (4), parasites reproduce asexually producing high numbers of cercariae (5) that are

65 released to water. Cercariae are short-lived and await a fish host. They penetrate the epithelium

66 of the fish, migrate to the eye lenses, and develop to metacercariae (6). The life cycle is

67   completed when the definitive host consumes an infected fish. Reproduced with permission

68   from Karvonen (2012).

69

70   *Diplostomum spathaceum* has been reported from eye lenses of over 100 fish species in Europe

71   (Chappell, 1995), while the global distribution of the *Diplostomum* spp. probably includes

72   many more host species. Thus, the genus is considered one of the most common and abundant

73   parasites of freshwater fishes. Species belonging to Cyprinidae, Percidae, Salmonidae,

74   Coregonidae, Catostomidae, and Gasterosteidae which inhabit littoral waters are commonly

75   infected (Margolis and Arthur, 1979; McDonald and Margolis, 1995; Valtonen and Gibson,

76   1997; Seppälä *et al.*, 2011). *Diplostomum* spp. have also been reported from marine fish species

77   in brackish waters including Gadidae and Pleuronectidae (Buchmann, 1986; Koie, 1999).

78   However, the prevalence and abundance of the infection is typically highly variable and can

79   depend, in addition to fish species, on the geographical location, type of water body, habitat

80   within a water body, season, host age and community structure of all the other hosts in the life

81   cycle (Pennycuick, 1971; Sweeting, 1974; Burrough, 1978; Balling and Pfeiffer, 1997;

82   McKeown and Irwin, 1997; Valtonen and Gibson, 1997; Valtonen *et al.*, 1997; Marcogliese

83   and Compagna, 1999; Marcogliese *et al.*, 2001a; Marcogliese *et al.*, 2001b; Karvonen *et al.*,

84   2004b; Karvonen *et al.*, 2015). For example, locations within a single lake can have different

85   infection levels (Balling and Pfeiffer, 1997), possibly reflecting on factors such as differences

86   in abundance of infected snails. Also, infections in the higher latitudes are seasonal and mainly

87   take place during summer months (McKeown and Irwin, 1997; Marcogliese *et al.*, 2001a;

88   Karvonen *et al.*, 2004b), which results in first infections in eye lenses of young fish and

89   accumulation of infections with fish age.

90

91   **23.2 Diagnosis**

92

93   Metacercariae of *Diplostomum* spp. are soft-bodied, flat, bilateral and round or oval-shaped,

94   with a body length of approximately 0.3-0.4 mm when fully-developed (note that the

95   morphology and size strongly depend on the age of the metacercaria, (see Sweeting (1974)).

96   Infections are clearly visible from a dissected eye lens under a microscope (Fig. 23.2) and

97   identification is straightforward as all parasite species in eye lenses of a fish belong to this same

98   genus. However, identification at species level is notoriously difficult. Different species are

99   morphologically very similar, especially as larval stages (miracidium, cercaria, metacercaria),

100   and their identification requires particular expertise and experience (Niewiadomska, 1986;

101 Niewiadomska and Kiseliene, 1994). More recently, species identification has been aided using

102 molecular techniques (Niewiadomska and Laskowski, 2002; Moszczynska *et al.*, 2009), while

103 these have resulted in a significant increase in the number of *Diplostomum* species (Locke *et*

104 *al.*, 2010b; Blasco-Costa *et al.*, 2014).

105

106 INSERT FIGURE 23.2 HERE

107 Figure 23.2 Three metacercariae of *Diplostomum* sp. in an eye lens of Atlantic salmon. Photo

108 courtesy of Ines Klemme.

109

110 *23.2.1 Parasitic cataracts (diplostomiasis)*

111

112 The most notable sign of infection in an eye lens is cataract formation due to metacercarial

113 movement and metabolism which damage the structure of the lens. If there are many

114 metacercariae, the damage accumulates and can result in the chronic stage of infection, known

115 as diplostomiasis. A severe condition can be observed visually as the eye lens becomes opaque,

116 grey or whitish. In extreme cases, the lens capsule can rupture or the lens becomes dislocated,

117 when the fish host loses its eyesight. Fewer cataracts (e.g. small clouds of granules or thread-

118 like formations (Shariff *et al.*, 1980)) and their early stages following development of the

119 metacercariae can be seen reliably only using a microscope, such as an ophthalmoscope

120 (Karvonen *et al.*, 2004c). This type of infections occur in most of the infected fish species

121 worldwide.

122

123 Cataracts gradually impair the vision of fish and the degree of impairment is linearly related to

124 the number of parasites in the lens (Karvonen *et al.*, 2004c) (Fig. 23.3). In other words, few

125 parasites rarely cause severe cataracts, except in small fishes, although parasites can remain in

126 the lens for years. However, there are no detailed data on long-term dynamics of cataracts

127 recorded from individual fish. Development of cataracts is also related to the size of the fish

128 and, consequently, size of the eye lens. In a small fish, even a low number of parasites can be

129 sufficient to cause severe pathology (Karvonen and Lindström, 2018). Further, recent evidence

130 suggest that fish may also show differences in their ability to tolerate the deleterious effects of

131 the parasites, i.e., the same number of parasites results in different degree of cataract formation

132 (Klemme and Karvonen, 2017). Infection can also decrease the lens size directly (Karvonen

133 and Seppälä, 2008a), but the significance of such effects for visual ability of fish needs further

134 study.

135

136 INSERT FIGURE 23.3 HERE

137 Figure 23.3 Relationship between cataract coverage and the number of *Diplostomum*

138 *pseudospathaceum* in the eye lenses of whitefish (*Coregonus lavaretus*). Data from Karvonen

139 and Seppälä (2008b). Reproduced with permission from Karvonen (2012).

140

141 Infections in the eye lens and the subsequent cataract formation can have significant

142 implications for the well-being of fish. Gradual deterioration of eyesight with increasing

143 infection intensity can cause several physiological and behavioural effects in fish. One notable

144 phenotypic sign of infection is darkening of the fish skin as the light intensity entering the eye

145 decreases. This impairs the cryptic colouration of the fish particularly against a light

146 background (Seppälä *et al.*, 2005a), which can lead to increased detection by predators. Eye

147 infection can also decrease the efficiency of fish to detect and harvest prey items (Crowden and

148 Broom, 1980; Owen *et al.*, 1993), which can result in decreased growth (Karvonen and

149 Seppälä, 2008b). Impaired visual abilities have also a range of other effects that relate to social

150 interactions of fish and susceptibility to avian predation, the latter of which is essential for

151 completion of the parasite life cycle. For example, infection reduces group cohesion of shoaling

152 fish (Seppälä *et al.*, 2008), which can render individual fish detectable by predators. Infected

153 fish may also swim closer to the water surface (Crowden and Broom, 1980), although this

154 evidence is not conclusive (Seppälä *et al.*, 2004). Further, infection and cataracts increase

155 catchability of fish in experiments mimicking predation from fish-eating birds plunging into

156 water from the air. These effects also coincide with the metacercariae becoming fully

157 developed (Seppälä *et al.*, 2004, 2005b), which supports the idea that cataracts can enhance

158 parasite transmission to the definitive hosts.

159

160 *23.2.2 Implications of climate change for the parasite life cycle*

161

162 Parasitism and disease in aquatic animals may increase with climate change, principally in

163 response to rising temperatures that accelerate parasite development (Marcogliese, 2001, 2008;

164 Lõhmus and Björklund, 2015), although general predictions are difficult to make as effects are

165 species specific, context dependent and may vary among hosts (Marcogliese, 2008; Rohr *et al.*,

166 2011; Marcogliese, 2016). The existence of extreme weather events, confounding factors, and

167 non-linear thresholds further complicate matters (Marcogliese, 2008; Rohr *et al.*, 2011; Altizer

168 *et al.*, 2013; Marcogliese, 2016) and some diseases may actually decrease in occurrence with

169   climate change (Lafferty, 2009; Karvonen *et al.*, 2010b). Nevertheless, temperature is
170   considered the most important abiotic factor that influence parasitic platyhelminths in
171   ectothermic hosts, including fish (Chubb, 1979). In parasites with complex life cycles, such as
172   *Diplostomum* spp., temperature affects all free-living life cycle stages as well as those in
173   ectothermic hosts. Higher temperatures are generally expected to lead to faster growth,
174   development and reproduction, earlier transmission and development in the spring, prolonged
175   transmission in the autumn and more generations per year. However, it may also increase
176   mortality rate among parasites in the aquatic environment (Marcogliese, 2001). Thus, it is
177   important to consider the net effects of temperature within the entire parasite life cycle. As
178   there are no data on relationships between temperature and cataracts (diplostomiasis) *per se*,
179   and because cataracts are related to parasite numbers (Fig. 23.3), it is relevant to consider
180   temperature-related factors that control the latter.

181

182   Our current discussion includes the effects of temperature on potential spread of *Diplostomum*
183   spp., followed by temperature effects on production and transmission of the life cycle stages.
184   Throughout, we will explore evidence on temperature effects on *Diplostomum* spp. and other
185   trematode taxa and on general trematode biology, while discussing the net effects of increasing
186   temperature for the parasite life cycle. We will also consider ecological evidence from field
187   studies, and explore effects of temperature on the physiology and the resistance of snails and
188   fish. Besides temperature, we will also focus on other environmental changes that are
189   associated with climate change and likely to either increase or decrease parasite population.
190   Finally, we will discuss the implications of climate warming for parasite prevention strategies
191   in aquaculture.

192

193   **23.3 Potential spread of *Diplostomum* spp. with increasing water temperature**

194

195   Range shifts of aquatic biota are expected with climate change (Settele *et al.*, 2014). However,
196   given that *Diplostomum* spp. are already widespread in the northern hemisphere (*e.g.*, see Fig.
197   1 in Locke *et al.* (2015)), large range shifts are unlikely. Lens-infecting *Diplostomum* spp. are
198   generalists infecting a range of fish species (Locke *et al.*, 2010a; Locke *et al.*, 2010b; Rellstab
199   *et al.*, 2011; Locke *et al.*, 2015), so changes in fish species composition as a result of fish host
200   range expansion or contraction should not have large effects on the parasite's distribution.
201   However, the host spectrum in any given habitat could change. For example, increasing
202   temperatures are predicted to have significant negative effects on cold-water stenotherms, such

203   as salmonids and coregonids, contracting their range (Marcogliese, 2001; Chen *et al.*, 2016).

204   These high-latitude cold-water stenotherms may experience an increase in their northern range,

205   but a contraction of their southern boundaries with the expansion northward of temperate fishes

206   (Ficke *et al.*, 2007; Settele *et al.*, 2014). In addition, warm- and cool-water fishes may displace

207   native species as they migrate into higher latitudes in the northern hemisphere and lower

208   latitudes in the southern hemisphere (Ficke *et al.*, 2007). Another potential complicating factor

209   is that snail intermediate host populations may be at risk because freshwater molluscs are

210   predicted to be unable to track high rates of climate change (Settele *et al.*, 2014). Their

211   populations are further compromised by invasive species, habitat modification, and

212   contaminants (Settele *et al.*, 2014).

213

214   In addition to range shifts of resident fish and invertebrate species, increasing temperature is

215   expected to lead to the invasion of new and potentially susceptible hosts (Marcogliese, 2001;

216   Altizer *et al.*, 2013), including those for *Diplostomum* spp. For example, there have already

217   been numerous introductions of warm-water fish species into the lower Great Lakes, expanding

218   their distributions northward (Marcogliese, 2001). Further warming should facilitate the

219   introduction and expansion of warm-water invaders (Collingsworth *et al.*, 2017). There are at

220   least two possible outcomes. First, invasive species can facilitate parasite transmission by

221   effectively acting as new susceptible hosts and boost the life cycle completion. Second, new

222   hosts can be resistant to infections and act as sinks of infection while diluting infection risk

223   among the native hosts. In Europe, round gobies (*Neogobius melanostomus*) and bighead

224   gobies (*Ponticola kessleri*) were heavily infected with *Diplostomum* spp. in their introduced

225   range in the Danube River, the Rhine River, and parts of the south-western Baltic Sea,

226   potentially enhancing transmission of *Diplostomum* spp. to piscivorous birds (Ondračková *et*

227   *al.*, 2009; Muhlegger *et al.*, 2010; Francová *et al.*, 2011; Kvach and Winkler, 2011; Ondračková

228   *et al.*, 2015). In contrast, following the introduction of the round goby in the St. Lawrence

229   River, Canada, abundance of *Diplostomum* spp., which at one time was one of the most

230   common fish parasites in that river, declined to extremely low levels within five years or less

231   in yellow perch, *Perca flavescens*, golden shiner (*Notemigonus crysoleucas*) and the spottail

232   shiner, *Notropis hudsonius* (Gendron and Marcogliese, 2017). The authors suggested this was

233   due to gobies acting as incompetent decoy hosts for cercariae and diluting the risk of infection

234   to the native fish. The difference between the capacity of gobies as hosts for *Diplostomum* spp.

235   in Europe and North America may be because invasive gobies in Europe were exposed to

236   widespread European species, while those in North America were exposed to new parasites

237 with which they had no previous experience. This idea is supported by the increase in
238 abundance of *Diplostomum* spp. in round gobies over time (15 years) since their initial invasion
239 into the Great Lakes (Gendron *et al.*, 2012).

240

241 **23.4 Implications of increasing temperature for the parasite life cycle stages**

242

243 *23.4.1 Effects of elevated temperature on life cycle stages*

244

245 Life cycle stages of *Diplostomum* spp. outside the endothermic avian host (miracidia, cercariae,
246 and the larval forms residing in ectothermic snails and fish) are potentially influenced by
247 increasing water temperature. However, it is important to note that in general, physiological
248 tolerance of parasites to temperature not only varies among species, but also among stages of
249 the same species (Chubb, 1979; Marcogliese, 2001). Overall, there are a few experimental
250 studies on effects of temperature on different life cycle stages of *D. spathaceum* and related
251 species (Table 23.1), although much more information is needed to make reliable predictions.
252 For example, swimming velocity of the miracidia hatching from eggs increased at higher
253 temperatures, but the life span declined (Harris, 1986). While the latter result is likely due to
254 faster depletion of the finite glycogen reserves, the net effects on transmission are unknown.
255 Considering these effects alone, an increase in temperature should likely promote the encounter
256 between miracidia and potential snail hosts, but decrease the infective time-period. Further
257 evidence on the snail host has shown that the time to patency decreased and cercarial output
258 increased at higher temperatures (Harris, 1986; Waadu and Chappell, 1991), both of which
259 should promote transmission to the fish host. However, both cercarial activity time and life
260 span were reduced at higher temperatures, which should limit transmission (Harris, 1986; Sous,
261 1992; Lyholt and Buchmann, 1996). Moreover, cercarial penetration and speed of migration to
262 the eyes increased at higher temperatures (Whyte *et al.*, 1988; Lyholt and Buchmann, 1996),
263 but infectivity peaked at the mid-range of the experimental exposure temperatures (Stables and
264 Chappell, 1986b). The latter results suggest that infection success would decrease eventually
265 as temperature increases. To sum up, the contrasting effects of higher temperature would
266 increase parasite reproduction, but decrease longevity and infectivity of the transmission
267 stages. Consequently, it is at present difficult to predict the overall effects of temperature on
268 the parasite transmission success (Fig. 23.4).

269

270   Table 23.1 Experimental studies on effects of temperature on free-living stages of *Diplostomum*

271   *spathaceum* and other species as well as on those stages in gastropod (*Lymnaea* spp.) and

272   rainbow trout (*Oncorhynchus mykiss*). Parasites are *D. spathaceum* unless otherwise indicated.

273

Trait	Temperature	Comment	Reference
Egg hatch	Delayed at 4 °C in *D. phoxini*	At 4 °C, 6 d delay in hatch, but equals rate at 20 °C by 14 d	Harris (1986)
Miracidial swimming velocity	Increases with temperature to a maximum at 25 °C, then declines in *D. phoxini*	Examined velocity between 5 and 40 °C	Harris (1986)
Miracidial life span	Maximum at 4 °C, then declines with increasing temperature in *D. phoxini*	No survival at 40 °C	Harris (1986)
Miracidial infectivity to snails	Declines if exposed at lower temperatures (<14 °C) and switched to 20 °C	Effect lost if snails all exposed at same temperature (20 °C), then switched to lower temperatures	Waadu and Chappell (1991)
Time to patency in snails	Faster at higher temperatures in *D. phoxini*	75h at 10 °C vs. 40 hr at 20 °C	Harris (1986)
	Affected by snail maintenance temperature	Delayed in snails infected at 20 °C if held at 14 °C, not 20-25 °C	Waadu and Chappell (1991)
Cercarial shedding (minimum temperature)	None < 10 °C		Bauer (1959)

	Occurs at 4-6 °C	Stops at 3-5 °C	Lyholt and Buchmann (1996)
	None < 9 °C	Field-based study	Sous (1992)
Cercarial output	Increases with temperature, peaks at 18 °C		Bauer (1959)
	Declines at temperatures < 10 °C	Gradual decline to 5-6 °C, then drops rapidly.	Lyholt and Buchmann (1996)
	Rate of output increases between 10 and 20 °C in *D. phoxini*		Harris (1986)
	Rate of output increases between 10 and 27 °C	<100/hr at 4-14 °C 20-1100/hr at 15-20 °C 100-4700 at 20-27 °C	Sous (1992)
Cercarial activity	None < 9-10 °C	Move to upper waters at 18-22°C	Bauer (1959)
	Peaks at intermediate temperatures in *D. phoxini*	None at 4 °C, maximum at 15 °C, then declines	Harris (1986)
Cercarial life span	Shorter at higher temperatures (e.g., 72 hr at 20 °C vs. 240 hr at 4 °C	Consistent decline between 4 and 25 °C	Harris (1986); Sous (1992); Lyholt and Buchmann (1996)
Cercarial penetration	Increases at higher temperatures	*In vitro* system; occurs as low as 4 °C	Whyte *et al.* (1988)
	Occurs at 7.5 °C		Stables and Chappell (1986b)
Cercarial migration to fish eyes	Faster at higher temperature		Lyholt and Buchmann (1996)

	Inhibited at <10 °C		Stables and Chappell (1986b)
Cercarial establishment in eyes	Highest at ≥ 18 °C	Occurs at 13-16 °C	Bauer (1959)
	Maximum in mid-range (17.5 °C)	No infections at < 10 °C if fish maintained at < 10 °C, but infections obtained at 5 °C if fish maintained at 15 °C	Stables and Chappell (1986b)
	Greater at high temperature (15 °C)	No infections at 5 °C	Lyholt and Buchmann (1996)

274

275 INSERT FIGURE 23.4 HERE

276 Figure 23.4 A dense swarm of cercariae of *Diplostomum pseudospathaceum* released from

277 snail (*Lymnaea stagnalis*). Production and release of cercariae increase significantly from 10

278 to 20 °C. However, cercarial infectivity and lifespan deplete faster at higher temperatures.

279 Photo by Anssi Karvonen.

280

281 An early meta-analysis by Poulin (2006) suggested that cercarial emergence could increase

282 200-fold with a 10°C increase in temperature, prompting the author to suggest climate change

283 could have a huge influence on parasite populations. However, in a subsequent meta-analysis

284 that accounted for the minimum emergence temperature threshold (the temperature where

285 emergence rates decrease to almost zero) and acclimation status of infected molluscs,

286 temperature above a particular threshold actually does not appear to affect cercarial

287 development (Morley and Lewis, 2013). In addition, cercarial emergence from molluscan hosts

288 shows a peaked pattern with temperature, at first increasing within low temperature ranges. It

289 was unaffected within the optimum temperature ranges (thermostability), which correspond to

290 the latitudinal range inhabited, but then declined at higher temperatures (Morley and Lewis,

291 2013). However, there were also geographic strain-specific differences in thermostability

292 within *D. spathaceum* in two lymnaeid species (Morley and Lewis, 2013), which underscores

293 the complexity of making predictions and establishing general rules for *Diplostomum* spp.

294

295   Thermostability over a range equivalent to typical summer temperatures for a particular species

296   also was observed for most trematode species in cercarial mortality and glycogen utilization

297   rate over normal temperature ranges encountered (Morley, 2011). A more recent meta-analysis

298   of over 30 trematode species including *D. spathaceum* demonstrated an optimal temperature

299   for both cercarial output and infectivity, while mortality was directly related to temperature

300   (Studer and Poulin, 2014). Specifically, cercarial mortality and glycogen utilization rate

301   increased linearly with temperature in *D. phoxini* (Morley, 2011). Furthermore, temperature

302   had little influence on miracidial survival and metabolism over normal temperature ranges,

303   suggesting that miracidia are more resistant to temperature changes than cercariae (Morley,

304   2012). Interestingly, there was little correlation in thermal responses between miracidia and

305   cercariae within geographic strains of the same species (Morley, 2012). Using metabolic

306   measures, Morley and Lewis (2015) showed that in general, trematode miracidia and cercariae

307   show increased infectivity with temperature, maximizing over optimal temperature ranges and

308   then declining at higher temperatures. Infectivity of metacercariae to definitive hosts, in

309   contrast, was highest at low temperatures and declined as temperature increased. The overall

310   conclusion is that temperature is not hugely important for the survival and function of

311   trematode free-living transmission stages. Rather, transmission may depend more on thermal

312   effects of climate change on the target hosts, among other factors (Morley and Lewis, 2015).

313   For example, it is possible that any higher production of infective stages with temperature

314   would be compensated for by their higher mortality, resulting in a roughly stable risk of

315   infection to fish regardless of temperature. Under such conditions, factors such as host age, size

316   and physiological state may affect infectivity more than direct effects of temperature on

317   miracidia or cercariae (Morley and Lewis, 2015).

318

319   *23.4.2 Ecological evidence from field studies*

320

321   Comparisons of parasite infections in fish inhabiting areas of elevated temperatures with those

322   under ambient conditions may provide insight into effects of temperature increases at the scale

323   of an entire host-parasite relationship (Marcogliese, 2001, 2008). For example, infection of

324   European perch (*Perca fluviatilis*) by *D. baeri* occurred earlier in Biotest Lake, a semi-enclosed

325   area in the Baltic Sea heated by nuclear power plant thermal effluent, than at an ambient site,

326   and infections accumulated there to a higher degree at an increased prevalence in 1986-87

327   (Höglund and Thulin, 1990). Additionally, fish were presumed to show increased mortality in

328 the heated area due to selective predation on heavily infected hosts. In another example of a

329 similar system, the release of cercariae by infected snails (*Helisoma trivolvis*) and recruitment

330 of metacercariae of the eyefluke *Tylodelphys scheuringi* in the mosquitofish (*Gambusia affinis*)

331 were prolonged into the winter months in a thermally-altered reservoir compared to ambient

332 areas in South Carolina, USA (Aho *et al.*, 1982). Cercarial release also ceased during the

333 warmest months (e.g. July and August), implying an upper thermal limit to this trait, in

334 agreement with Morley and Lewis (2013). The trematode *Ornithodiplostomum ptychocheilus*

335 released cercariae from infected *Physa* sp. year-round in the same thermally altered reservoir,

336 also with the exception of the warmest months (Camp *et al.*, 1982). However, recruitment by

337 mosquitofish did not differ between the thermally altered and ambient areas, showing that there

338 can be distinct different responses to temperature between phylogenetically related parasites in

339 the same fish host at the same sites.

340

341 Systems with natural elevation in water temperature compared to that in the ambient

342 environment may also provide interesting comparisons on the effect of temperature. Karvonen

343 *et al.* (2013) examined *Diplostomum* spp. in threespine sticklebacks (*Gasterosteus aculeatus*)

344 from two Icelandic lakes that possess natural temperature gradients due to groundwater inflow

345 and geothermic activity. In both lakes, sticklebacks from the warm areas showed a much higher

346 abundance of *D. baeri* than those from cold regions. A second species of *Diplostomum* also

347 had much higher prevalence and abundance in the warm part of one lake compared to the colder

348 part, although it was absent from the second lake (Karvonen *et al.*, 2013).

349

350 **23.5 Population dynamics of the hosts**

351

352 Since fish growth is temperature-dependent, an extended growing season and reduction in

353 overwintering stress could lead to increases in fish productivity in temperate fishes that are

354 currently limited by sub-optimal temperatures for their growth (Ficke *et al.*, 2007). This,

355 however, assumes that temperatures remain within optimal ranges and other conditions are

356 adequate and food is not limiting. However, reproduction could be negatively affected in those

357 fishes requiring low overwintering temperatures for spawning, such as salmonids (Ficke *et al.*,

358 2007). However, the duration of optimal temperatures for growth will likely increase for all

359 thermal guilds of fishes (Collingsworth *et al.*, 2017). Furthermore, recruitment and production

360 of spring and summer spawners can be promoted (Collingsworth *et al.*, 2017). Fish populations

361 also could be negatively affected not only by increased temperatures, but by decreased levels

362   of dissolved oxygen, and changes in contaminant concentrations, disease dynamics, and

363   hydrography, along with any other associated habitat modifications (Ficke *et al.*, 2007;

364   Collingsworth *et al.*, 2017). Besides fish, similar processes could apply also to other hosts in

365   parasite life cycles, such as snails in case of *Diplostomum* spp. Without long-term data or

366   epidemiological modelling, however, it is not possible to predict how these changes could

367   affect levels of *Diplostomum* spp. in fishes. Nevertheless, given that most lens-infecting species

368   of *Diplostomum* are generalists, any decrease in the availability of fish intermediate hosts likely

369   will be offset by increases in others.

370

371   **23.6 Effect of temperature on parasite mortality**

372

373   *Diplostomum* spp. metacercariae are generally well protected within the fish eye lens, both

374   from the host immune attacks and from the external environment, and there are no experimental

375   data showing metacercarial mortalities in fish directly following environmental perturbations.

376   Thus, effects of the ambient environment on the parasite population are more likely to concern

377   the free-living infective stages, miracidia and cercariae, as well as processes related to host

378   physiology and resistance. As discussed earlier, increasing temperature tends to decrease the

379   longevity of the infective stages as their finite energy reserves are exhausted more rapidly in

380   higher water temperatures (Table 23.1). Similarly, temperature could enhance host immune

381   function to prevent parasites migration in host tissues towards the eye (see below). Whether

382   this results in negative net effects on the parasite population given the probable increase in

383   parasite replication with temperature needs elucidation.

384

385   *23.6.1 Effects of climate warming on host physiology and immunological resistance*

386

387   Temperature also controls the physiological functions (e.g. immunity) in the fish hosts, some

388   of which have direct relevance to parasite infections. Early work examining the immune

389   response in fish to *Diplostomum* spp. used the rainbow trout (*Oncorhynchus mykiss*)-*D.*

390   *spathaceum* system, while more recent studies have explored ecological immunology in

391   threespine stickleback. In general, immune responses in the eye lenses of fish are considered

392   weak or non-existent as the lens is not directly connected to blood circulation (Sitjá-Bobadilla,

393   2008). Therefore, the time window for fish to fight off an initial infection is very narrow and

394   consists of the time diplostomules are migrating to the lens, typically within 24 hr from

395   exposure (Chappell *et al.*, 1994; Sitjá-Bobadilla, 2008). Given that not all diplostomules reach

396   the eye in an initial exposure, non-specific immune responses of the fish are likely responsible

397   for partly preventing the infection (Whyte *et al.*, 1991). In rainbow trout, these responses

398   include, for example, activity of the alternative-pathway of the complement cascade (Whyte *et*

399   *al.*, 1988, 1989) as well as macrophages (Whyte *et al.*, 1989; Chappell *et al.*, 1994). Fish also

400   display antibody-mediated specific responses to infection with *Diplostomum* spp. that develop

401   within a few weeks from the first exposure and significantly reduce the number of parasites

402   establishing in subsequent exposures (Stables and Chappell, 1986a; Höglund and Thuvander,

403   1990; Whyte *et al.*, 1990; Karvonen *et al.*, 2005; Karvonen *et al.*, 2010a; Rellstab *et al.*, 2013).

404   In sticklebacks, *in vitro* experiments have demonstrated that head kidney leucocytes (HKL)

405   exhibit a strong respiratory burst when exposed to antigens of *D. pseudospathaceum* (Franke

406   *et al.*, 2014). However, the HKL respiratory burst activity also drops 1.5 days after exposure,

407   implying that phagocytic cell activation is important for the immune response to *D.*

408   *pseudospathaceum* (Scharsack and Kalbe, 2014). These authors suggested that the innate

409   immune response, but not the acquired immune response, was activated to defend against *D.*

410   *pseudospathaceum* in threespine sticklebacks (Scharsack and Kalbe, 2014). There was also

411   evidence supporting parasite genotype-specific innate immune activity in *G. aculeatus* (Haase

412   *et al.*, 2014), while other studies found no evidence of genotype-specificity in the acquired

413   responses (Rellstab *et al.*, 2013; Haase *et al.*, 2016). Further, the immune response in threespine

414   sticklebacks against *Diplostomum* spp. varies among populations and habitats (Scharsack and

415   Kalbe, 2014; Scharsack *et al.*, 2016). For example, fish sympatric with *D. pseudospathaceum*

416   show a stronger innate response against initial infection than those from uninfected populations

417   (Kalbe and Kurtz, 2006).

418

419   Temperature basically affects all physiological functions in ectotherms (Bowden, 2008). In

420   fish, the immune response is stimulated or at least positively correlated with temperature, as

421   shown by lysozyme activity, concentration of circulating IgM, and major histocompatibility

422   complex and cytokine gene expression (Tort *et al.*, 2003; Bowden, 2008; Martin *et al.*, 2010;

423   Uribe *et al.*, 2011). Circulating IgM concentration increases in salmonids when acclimated to

424   19 °C (Uribe *et al.*, 2011). However, it is not known if these processes play a role in defence

425   against *Diplostomum* spp. In contrast, temperature effects on complement activity are

426   inconclusive, effects on haematology vary with cell type, and phagocytosis is not greatly

427   affected (Bowden, 2008; Uribe *et al.*, 2011), processes and functions which do play a role in

428   the immune response against *Diplostomum* spp. In most fish species examined, acquired

429   immune activity and immune gene expression are enhanced while innate immune activity is

430   suppressed at the highest temperatures tested (Dittmar *et al.*, 2014). Nevertheless, one might

431   expect resistance to parasites such as *Diplostomum* spp. to increase with climate change

432   (Scharsack *et al.*, 2016). However, higher temperatures also accelerate parasite growth,

433   development and life cycle completion, and it is not clear whether the host or the parasite

434   benefits more under these circumstances (Scharsack *et al.*, 2016). Again, the above aspects

435   well illustrate the complex nature of temperature effects; they potentially elevate transmission

436   and can result in higher parasite numbers (Fig. 23.5), but also interact with different types of

437   temperature effects on the host as well as with many other ecological and evolutionary factors

438   determining host resistance.

439

440   INSERT FIGURE 23.5 HERE

441   Figure 23.5 Timing and temperature-dependence of transmission of *Diplostomum* spp. to fish

442   in natural conditions. Data show the mean number of new *Diplostomum* spp. infections (±SD)

443   in eye lenses of fish caged in an oligotrophic lake during two-week periods in May-October.

444   The solid line indicates water temperature of the lake. Infections peak naturally in July-August

445   when water temperature exceeds 15 °C. Climate warming could potentially enhance parasite

446   reproduction in the snail intermediate hosts and transmission to fish resulting in higher number

447   of infections within the current window on transmission. Additionally, higher water

448   temperatures in spring and autumn could prolong the optimal infection period from both ends

449   of the range. Reproduced with permission from Karvonen *et al.* (2004b).

450

451   Extreme weather events are also predicted to increase with climate change (Marcogliese,

452   2001). Examination of how host-parasite systems respond to extreme weather such as heat

453   waves and drought may provide insight into how climate change will affect outbreaks of

454   diseases in ecosystems (Hudson *et al.*, 2006; Poulin and Mouritsen, 2006; Morley and Lewis,

455   2014). Studies have shown that acute temperature changes experienced during heat waves can

456   cause immunosuppression in fish (Uribe *et al.*, 2011; Scharsack *et al.*, 2016). Generally, in

457   ectotherms, such changes can suppress various immune functions such as phagocytosis,

458   respiratory burst and antibody production (Martin *et al.*, 2010). Immune function is more

459   efficient if fish are acclimated to higher or varying temperatures (Martin *et al.*, 2010; Scharsack

460   *et al.*, 2016). Indeed, in simulated heat wave experiments, innate and adaptive immune

461   functions were optimal at 13-17 °C compared to 18-24 °C in threespine stickleback (Dittmar

462   *et al.*, 2014). Exposure to a simulated heat wave also caused long-lasting deleterious effects on

463   immune function, but less so if fish were from presumably better-adapted populations (Dittmar

et al., 2014). Sticklebacks maintained in artificial enclosures with the lowest parasite load and an intermediate level of MHC class IIb sequence variation survived best, while those with the highest parasite burdens perished during the 2003 European heat wave. This suggests a link between MHC diversity and fitness (Wegner et al., 2008).

In addition to fish, higher water temperatures can influence resistance of the other poikilothermic intermediate host of Diplostomum spp., the snail. Compared to fish, however, there is little information on the immune response of snails to the infection. It has been established that the susceptibility of Lymnaea stagnalis to D. spathaceum varies with age - young snails being susceptible and older snails more resistant to infection (Chappell et al., 1994). Haemocyte profiles also differ between infected and uninfected snails with haemocytes from infected snails displaying reduced phagocytotic capability, and serum showing lower opsonic and agglutinating abilities (Riley and Chappell, 1992). This is consistent with the suggestion that the initial infection decreases immune function in snails, making them more susceptible to accumulate further infections (Louhi et al., 2013).

Evidence on the effect of temperature on snail immune function comes mostly from parasite systems other than Diplostomum spp. For example, exposure of L. stagnalis to simulated heat waves of 25 °C increased infection success of the trematode Echinoparyphium aconiatum (Leicht and Seppälä, 2014). Exposure of the snails to 23.5-25 °C for more than one week also reduced their haemocyte concentration and phenoloxidase-like activity, an oxidative defence against parasites (Leicht et al., 2013; Leicht et al., 2017; Salo et al., 2017). It is likely that similar processes could influence also infections of Diplostomum spp. in L. stagnalis and in other lymnaeids. Overall, this evidence suggests that increasing temperature could impair the ability of snails to prevent infections, likely resulting in increasing prevalence of Diplostomum spp. and other trematode infections. As these infections typically castrate the host, increased likelihood of parasitism would undoubtedly influence also the snail populations, which again would be reflected in the net effects of temperature on the parasite life cycle.

23.6.2 Net effects of increasing temperature on the parasite life cycle

Overall, there are no detailed studies on the net effects of temperature on transmission and pathology of Diplostomum spp. However, studies on a similar host-parasite system may shed some light on the question of net effects. Ribeiroia ondatrae has a three-host life cycle,

498 infecting snails and birds. One fundamental difference is that amphibian tadpoles are second

499 intermediate hosts, where the parasite causes limb malformations. Nevertheless, the second

500 intermediate host is a freshwater ectothermic vertebrate with pathological consequences.

501 Studies on net effects of temperature on different life history aspects of *R. ondatrae* may

502 provide informative for *Diplostomum* spp. Paull and Johnson (2011) and Paull *et al.* (2012)

503 demonstrated differential effects on different parasite and host life history characteristics (see

504 Marcogliese (2016)). For example, cercarial survival and establishment in the tadpole peaked

505 at low temperatures, while egg development rate, cercarial development rate and cercarial

506 penetration to tadpoles peaked at high temperatures, but metacercarial numbers in the tadpoles

507 were lowest at high temperatures. Growth of snails (*Planorbella trivolvis*), infected or not, and

508 Pacific chorus frog (*Pseudacris regilla*) tadpoles peaked at high temperatures, along with snail

509 fecundity. However, fecundity of infected snails peaked at intermediate temperatures. Their

510 crucial finding was that pathology in the snail in terms of castration and gigantism peaked at

511 high temperatures, but malformations in the tadpoles were maximised at intermediate

512 temperatures and were lowest at high temperatures (Paull and Johnson, 2011; Paull *et al.*,

513 2012). In a year-long mesocosm study, a temperature increase of 3 °C induced snails to release

514 cercariae of *R. ondatrae* nine months earlier than at ambient conditions and increased snail

515 mortality four-fold (Paull and Johnson, 2014). However, infections in bullfrog (*Lithobates*

516 *catesbeianus*) tadpoles peaked two months earlier. In chorus frogs (*Pseudacris triserata*),

517 infections were reduced by half and malformations by two-thirds (Paull and Johnson, 2014).

518 After one year, 92% fewer adult snails were releasing cercariae in the thermally-altered

519 mesocosm compared to the ambient one (Paull and Johnson, 2014). These results suggest that

520 changes in the impact of parasites on their hosts following global warming depend on the

521 timing and temporal overlap of the temperature-driven changes in the host and parasite

522 populations.

523

524 To conclude, the above examples highlight the need to understand the net effects of temperature

525 increases on parasite transmission in general and on *Diplostomum* spp. in particular (Altizer *et*

526 *al.*, 2013; Marcogliese, 2016). It seems clear that elevated temperatures will influence both

527 parasites and hosts, patterns that could show contrasting effects on parasite prevalence and

528 abundance. Untangling these relationships requires rigorous experimental approaches in

529 laboratory and under field conditions. Due to multiple underlying factors, interpreting the

530 overall effect of climate warming also emphasises the importance of long-term time-series data

531 on parasite population dynamics. Such data are not available for most systems, but would be

532   invaluable as they capture the outcome of the entire process within a host-parasite interaction.
533   Furthermore, the current evidence on temperature effects needs to be interpreted with caution,
534   as they may not have accounted for acclimation of both hosts and parasites, in addition to
535   infected hosts, or variation in temperature (Morley and Lewis, 2013; Raffel *et al.*, 2013; Rohr
536   *et al.*, 2013; Raffel *et al.*, 2015; Altman *et al.*, 2016). This is important, as organisms generally
537   acclimate their performance after a temperature shift, which could change the interpretation of
538   the temperature effects. Moreover, natural temperatures are rarely constant but variable and
539   even a short-term variation in temperature will change the outcome of a host-parasite
540   interaction (Paaijmans *et al.*, 2010; Raffel *et al.*, 2013). Indeed, variation in temperature is
541   expected to increase with climate warming (Jiménez Cisneros *et al.*, 2014; IPCC, 2018), which
542   emphasises the importance of incorporating temperature dynamics into studies on disease
543   occurrence. Undoubtedly, short-term temperature variation plays an important role in
544   epidemics of directly transmitted pathogens. However, implications of the temperature
545   variation for macroparasites with complex life cycles, such as *Diplostomum* spp., may be
546   challenging and difficult to predict.

547

548   **23.7 Other associated consequences of climate change**

549

550   Freshwater ecosystems can be expected to undergo numerous changes aside from increased
551   temperature associated with climate change. These include changes in precipitation, salinity
552   eutrophication, acidification, hydrology and water levels, reduced ice cover, habitat loss,
553   fragmentation, pollution, ultraviolet (UV) radiation, and invasive species (Marcogliese, 2001,
554   2008, 2016), all of which could affect the distribution and abundance of *Diplostomum* spp.
555   (Table 23.2). There is no general unidirectional effect of these environmental changes; they
556   can lead to parasite population increases or declines, emphasizing the importance of
557   confounding factors and context dependency (Rohr *et al.*, 2011; Altizer *et al.*, 2013).

558

559   Table 23.2 Putative effects of environmental or biological changes associated with climate
560   change derived from Table I in Marcogliese (2008) on populations of *Diplostomum* spp. in
561   fishes based on information in Marcogliese (2001, 2004, 2005, 2008), Marcogliese *et al.* (2010)
562   and Tully *et al.* (2019).

563

Environmental or biological change	General response of *Diplostomum* spp.	Putative cause
Species introductions with change in host range	Population increase or decrease	Introduction of host species should increase generalist and specialist *Diplostomum* species of introduced hosts, but decrease specialist *Diplostomum* species of native hosts at risk
Loss of habitat due to temperature	Population decline	Applies to specialist species whose hosts lose habitat
Reduced flow rates	Population increase	Retention of free-living infective stages, increased infectivity of fish, promotion of snail habitat
Eutrophication	Population increase	Promotes parasites, which use snails as intermediate hosts and birds as definitive hosts
Increased stratification	Population decline	Reduction in snail habitat due to seasonal anoxia in bottom waters
Reduced ice cover	Population increase	Promotes transmission of *Diplostomum* spp. to avian definitive hosts over longer period
Increased acidification in headwater streams	Population decline	Reduced survival of snail intermediate hosts sensitive to acidification
Decreased acidification in lakes	Population increase	Promotes survival of snail intermediate hosts
Increased ultraviolet (UV) radiation	Population decline	Mortality of free-living infective stages
Decrease in salinity due to increased precipitation	Population increase	Increase in available habitat due to lower salinity
Rise in sea level	Population decline	Loss of habitat due to saltwater intrusion
Increased concentration of contaminants	Population decline	Combined effects of contaminants and *Diplostomum* spp. infection reduces

		fish health; transmission to fish reduced through effects on cercariae
Socioeconomic adaptation (dam construction)	Population increase	Replacement of lotic conditions with still or slow-moving waters (see altered hydrology above)
(modifying water withdrawal or delivery)	Population decrease	Increased stream flow

564

565 There are some examples of impacts of these factors on *Diplostomum* spp. that may be
566 illuminating. For example, abundance of *Diplostomum* spp. in mudpuppies (*Necturus*
567 *maculosus*) in the St. Lawrence River was highest in a regulated fluvial lake with stable water
568 levels compared to two other fluvial lakes where levels fluctuated (Marcogliese *et al.*, 2000).
569 Experimental studies also demonstrated that transmission of *D. spathaceum* to rainbow trout
570 was greatly reduced at higher flow rates, with a ten-fold increase in flow rate decreasing
571 infections thirty-fold (Stables and Chappell, 1986b). Abundance of a similar parasite,
572 *Posthodiplostomum minimum*, also increased under low-flow conditions, but was severely
573 reduced under high flow conditions associated with snowmelt in *Fundulus zebrinus* in the
574 Platte River, Nebraska (Janovy *et al.*, 1997). Further, infection of eye flukes in the snail
575 *Lymnaea peregra* plummeted in a reservoir after it was filled to maximum volume (Moody and
576 Gaten, 1982). Thus, regulation of water bodies and flow rates, coupled with declining water
577 levels, should potentially serve to increase infection levels of *Diplostomum* spp. Indeed,
578 reservoir construction is considered a means of helping to mitigate or adapt to effects of climate
579 change on streams, rivers and wetlands (Jiménez Cisneros *et al.*, 2014; Muller, 2019).

580

581 Contaminant concentration may increase under low water conditions and climate change
582 (Johnson *et al.*, 2009; Jiménez Cisneros *et al.*, 2014; Landis *et al.*, 2014; Morley and Lewis,
583 2014). Lethal and sub-lethal effects of combined exposure of animals to both parasites and
584 contaminants can be greater than the effects of either stressor alone (Marcogliese and Pietrock,
585 2011). For example, the combined exposure to municipal, agricultural and industrial pollution
586 and infection with *Diplostomum* spp. increased oxidative stress in yellow perch, *Perca*
587 *flavescens* (Marcogliese *et al.*, 2010). Moreover, exposure to increasing temperature

588 concurrently with another stressor may negatively impact an organism's health, leading to
589 population declines in ectotherms (Rohr and Palmer, 2013). Survival of naturally infected
590 snails (*L. stagnalis* and *L. peregra*) was reduced when exposed to cadmium compared to
591 controls (Morley *et al.*, 2003a). Free-living stages of a parasite also are sensitive to
592 environmental contaminants (Morley *et al.*, 2003c; Pietrock and Marcogliese, 2003). Exposure
593 of cercariae of *Diplostomum* spp. to cadmium, chromium, mercury and sediment extracts from
594 the polluted Oder River reduced their life span (Pietrock *et al.*, 2001; Pietrock *et al.*, 2002a;
595 Pietrock *et al.*, 2002b). Exposure to mixtures of cadmium and zinc, however, increased survival
596 in *D. spathaceum* (Morley *et al.*, 2001, 2002). Notably, cercarial activity of *D. spathaceum* was
597 reduced following exposure to zinc, cadmium, and zinc-cadmium mixture at all concentrations
598 tested and were vulnerable during the period of maximal cercarial infectivity (Morley *et al.*,
599 2003b). Infectivity of cercariae of both *Posthodiplostomum minimum* and *Ornithodiplostomum*
600 *ptychocheilus* to fathead minnows (*Pimephales promelas*) was reduced following exposure to
601 cadmium (Pietrock and Goater, 2005). Climate change can also increase the toxicity of
602 chemical contaminants as well as their uptake and an animal's susceptibility (Schiedek *et al.*,
603 2007; Noyes *et al.*, 2009; Hooper *et al.*, 2013; Stahl *et al.*, 2013). Furthermore, exposure to
604 contaminants may decrease an organism's thermal tolerance to increasing temperature (Noyes
605 *et al.*, 2009), as well interact with other climate-associated stressors (Moe *et al.*, 2013). Thus,
606 any increase in contaminants may decrease *Diplostomum* spp. infections in fish and snail
607 intermediate hosts.
608
609 Nutrient pollution is also expected to increase in fresh waters (Ficke *et al.*, 2007; Jiménez
610 Cisneros *et al.*, 2014; Collingsworth *et al.*, 2017), which can have significant implications for
611 parasitism. For example, occurrence of limb malformations and abundance of *R. ondatrae* in
612 anurans in agricultural wetlands was associated with eutrophication through effects on snail
613 species composition and biomass (Johnson and Chase, 2004; Johnson *et al.*, 2007).
614 Eutrophication combined with high temperatures leads to more frequent blooms of harmful
615 algal blooms (HABs) of cyanobacteria (Paerl *et al.*, 2011; Moe *et al.*, 2013; Jiménez Cisneros
616 *et al.*, 2014). HABs produce toxins and hypoxic conditions, detrimental to aquatic life (Moe *et*
617 *al.*, 2013). Interestingly, exposure to low concentrations of the cyanobacterial toxin
618 microcystin-LR (MC-LR) increased infection intensities of larval trematodes in leopard frogs,
619 *Rana pipiens* (Milotic *et al.*, 2018). While it did not affect growth or survival, exposure of the
620 snail *L. stagnalis*, the intermediate host for *Diplostomum* spp., reduced fecundity of adult snails
621 (Gérard *et al.*, 2005). Therefore, eutrophication associated with climate change may promote

622 infections of *Diplostomum* spp. in fish, but if allowed to progress, resulting in anoxia and the

623 proliferations of HABs, infections may decrease (see also Budria (2017)). Similar effects of

624 more frequent and widespread hypoxia in the benthos is expected in several water bodies

625 because of longer periods of stratification during summer (Ficke *et al.*, 2007; Collingsworth *et*

626 *al.*, 2017). Such developments would also negatively affect populations of snail and fish

627 intermediate hosts of *Diplostomum* spp. in deeper waters (Table 23.2).

628

629 Exposure to ultraviolet (UV) radiation in freshwater ecosystems is expected to increase due to

630 enhanced penetration under certain conditions, and it may be most problematic in clear, shallow

631 waters (see Marcogliese (2001)). UV is harmful to invertebrates, including parasites, whose

632 free-living stages such as cercariae are sensitive to environmental stressors (Pietrock and

633 Marcogliese, 2003). While exposure to UV radiation may negatively affect free-living stages

634 of *Diplostomum* spp., it also is immunosuppressive in fish. Exposure of rainbow trout to UV

635 radiation led to increased numbers of *D. spathaceum* compared to controls, presumably

636 because of reduced resistance (Markkula *et al.*, 2007). Exposure to UV is expected to increase

637 in streams with climate change following reduced discharge, lower stream depth, and reduced

638 dissolved organic carbon (Clements *et al.*, 2008; Moe *et al.*, 2013). However, conflicting

639 effects on parasites and hosts make any predictions problematic.

640

641 Other abiotic parameters that may be affected by climate may also negatively or positively

642 impact the immune response in fish (Uribe *et al.*, 2011). For example, an increase in hypoxia

643 decreased the respiratory burst activity of macrophages and lowered the level of circulating

644 antibodies. In contrast, elevated salinity increased lytic enzyme activity, macrophage

645 respiratory burst activity, HKL phagocytic activity, plasma lysozyme concentration and

646 circulating IgM (Bowden, 2008; Uribe *et al.*, 2011). Effects of pH on immune response, on the

647 other hand, have provided conflicting results (Bowden, 2008; Uribe *et al.*, 2011). Temperature

648 stress combined with contaminants such as nickel and chlorine also causes immunosuppression

649 in fishes, including reduced spleen cellularity, erythrocyte and leukocyte counts, and increased

650 superoxide production (Prophete *et al.*, 2006; Verma *et al.*, 2007).

651

652 **23.8 Control and prevention of *Diplostomum* spp.**

653

654 The lens-infecting *Diplostomum* species also occur in pond-aquaculture as all the necessary

655 hosts of the parasite are commonly present. For example, fish farms typically attract fish-eating

656   birds, the definitive hosts of *Diplostomum* spp., to feed. Earth ponds with vegetation used in

657   rearing aquaculture fish also provide favourable habitats for snail intermediate hosts that

658   become readily infected following parasite output from birds attracted to the ponds. Prevalence

659   of infection in the snails can be high, which results in high infection also in fish (Stables and

660   Chappell, 1986c; Field and Irwin, 1994; Karvonen *et al.*, 2006a). Parasite cercariae can also be

661   brought into a facility with incoming water from upstream water bodies, but this is considered

662   not as significant source of infection in fish compared to transmission occurring within the

663   facility (Field and Irwin, 1994; Karvonen *et al.*, 2006a). High numbers of metacercariae and

664   resulting pathology in the eyes of fish may become a problem if they reduce the desired growth

665   in fish intended for market. Similarly, infected fish for stocking to support natural fish

666   populations may have lower success in the wild, although detailed data on the effects of

667   *Diplostomum* spp. infections in natural fish populations are not available.

668

669   It is likely that problems associated with *Diplostomum* spp. in aquaculture are also likely to

670   increase. These may be through increased rate of parasite replication, prolonged period of

671   parasite transmission and metacercarial development, or impaired ability of cold-water species

672   such as salmonids to resist the infection (Hakalahti *et al.*, 2006). Such effects may be

673   manifested as longer and later outbreaks of the disease in the autumn (Fig. 23.5), thus

674   necessitating extra control measures. Control of *Diplostomum* spp. infections, however, can be

675   challenging as there is no effective treatment of the infection in fish and immunizing fish

676   against the infection provides only partial protection against later infections (Höglund and

677   Thuvander, 1990; Karvonen *et al.*, 2005). Studies have shown that immunization alone does

678   not protect fish from the deleterious effects of infection and other means of defence, such as

679   behavioural avoidance of cercariae, may be needed to complement any immune-mediated

680   response (Karvonen *et al.*, 2004b; Karvonen *et al.*, 2010a). However, such behavioural

681   avoidance is often impossible in the confined space of aquaculture tanks and ponds. In addition,

682   other types of control measures such as treatment or filtering of water are not feasible because

683   of large water volumes and the continuous output of parasite cercariae during summer months.

684   Removal or chemical eradication of snails from the rearing ponds is generally considered as

685   the only viable option to control and prevent the infections in fish (Stables and Chappell, 1986c;

686   Field and Irwin, 1994), although this can be system-specific and depends on the magnitude of

687   cercarial input from upstream water bodies. Nevertheless, eradication of snails as the main

688   preventative method should work equally well even with increasing water temperatures,

689   although positive effect of temperature on reproduction of snails may necessitate more frequent

690 use of the eradication protocols. However, constructing the tanks and ponds in a way that limits
691 establishment of vegetation and snail populations should help in longer-term prevention of
692 infections.

693

694 **23.9 Conclusions**

695

696 The current evidence on the relationships between climate warming and infections of
697 *Diplostomum* spp. strongly highlight the difficulty of determining the net effects on the
698 complex parasite life cycle. This is because most, if not all, of the life cycle stages in the aquatic
699 environment likely respond to temperature by increasing parasite replication and infectivity.
700 However, elevated temperature will likely result in increased mortality of the infective stages,
701 and possibly accompanied by higher resistance in the fish hosts. Increasing temperature also
702 acts in concert with many other interrelated environmental changes such as alteration in
703 hydrology, increasing eutrophication, pollution and UV-radiation, loss of habitats and higher
704 risk of invasive species. All these factors working in concert illustrates the magnitude and scope
705 of environmental effects on *Diplostomum* spp., and on many other host-parasite systems
706 covered in this book. The high number of variables emphasises the importance of long-term
707 time-series studies, which would adequately provide the influence of all related factors.
708 Pinpointing the importance of individual factors, on the other hand, requires rigorous
709 experimental approaches supported by mathematical models on parasite dynamics with
710 changing temperature. One area of experimental research needed concerns the effect of
711 temperature and the other related factors (Table 23.2) on the severity of *Diplostomum* spp.-
712 induced pathology in fish, which has received relatively little attention. Alongside the effects
713 on the parasite life cycle, it is one of the key factors determining the impact of the parasite on
714 fish populations in nature and in units of intensive aquaculture.

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23.10 References

Aho, J.M., Camp, J.W. and Esch, G.W. (1982) Long-term studies on the population biology of *Diplostomulum scheuringi* in a thermally altered reservoir. *Journal of Parasitology,* 68, 695-708. doi:10.2307/3280931.

Altizer, S., Ostfeld, R.S., Johnson, P.T.J., Kutz, S. and Harvell, C.D. (2013) Climate change and infectious diseases: From evidence to a predictive framework. *Science,* 341, 514-519. doi:10.1126/science.1239401.

Altman, K.A., Paull, S.H., Johnson, P.T.J., Golembieski, M.N., Stephens, J.P., LaFonte, B.E. and Raffel, T.R. (2016) Host and parasite thermal acclimation responses depend on the stage of infection. *Journal of Animal Ecology,* 85, 1014-1024. doi:10.1111/1365-2656.12510.

Balling, T.E. and Pfeiffer, W. (1997) Location-dependent infection of fish parasites in Lake Constance. *Journal of Fish Biology,* 51, 1025-1032.

Bauer, O.N. (1959) Parasites of freshwater fish and the biological basis for their control (Translated from Russian, Israel Program for Scientific Translations, Jerusalem, 1962). *Bulletin of the State Scientifc Research Institute of Lake and River Fisheries,* 49.

Blasco-Costa, I., Faltýnková, A., Georgieva, S., Skirnisson, K., Scholz, T. and Kostadinova, A. (2014) Fish pathogens near the Arctic Circle: molecular, morphological and ecological evidence for unexpected diversity of *Diplostomum* (Digenea: diplostomidae) in Iceland. *International Journal for Parasitology,* 44, 703-715. doi:10.1016/j.ijpara.2014.04.009.

Bowden, T.J. (2008) Modulation of the immune system of fish by their environment. *Fish & Shellfish Immunology,* 25, 373-383. doi:10.1016/j.fsi.2008.03.017.

Buchmann, K. (1986) Prevalence and intensity of infection of *Cryptocotyle lingua* (Creplin) and *Diplostomum spathaceum* (Rudolphi) - parasitic metacercariae of Baltic cod (*Gadus morhua* L). *Nordisk Veterinaer Medicin,* 38, 303-307.

Budria, A. (2017) Beyond troubled waters: the influence of eutrophication on host-parasite interactions. *Functional Ecology,* 31, 1348-1358. doi:10.1111/1365-2435.12880.

Burrough, R.J. (1978) Population biology of two species of eyefluke, *Diplostomum spathaceum* and *Tylodelphys clavata*, in roach and rudd. *Journal of Fish Biology,* 13, 19-32. doi:10.1111/j.1095-8649.1978.tb03409.x.

758 Camp, J.W., Aho, J.M. and Esch, G.W. (1982) A long-term study on various aspects of the
759   population biology of *Ornithodiplostomum ptychocheilus* in a South-Carolina cooling
760   reservoir. *Journal of Parasitology,* 68, 709-718. doi:10.2307/3280932.

761 Chappell, L.H., Hardie, L.J. and Secombes, C.J. (1994) Diplostomiasis: the disease and host-
762   parasite interactions. *In:* Pike, A.W. and Lewis, J.W. (eds.) *Parasitic diseases of fish.*
763   Samara Publishing Limited, Dyfed, pp. 59-86.

764 Chappell, L.H. (1995) The biology of diplostomatid eyeflukes of fishes. *Journal of*
765   *Helminthology,* 69, 97-102.

766 Chen, Y.S., Todd, A.S., Murphy, M.H. and Lomnicky, G. (2016) Anticipated water quality
767   changes in response to climate change and potential consequences for inland fishes.
768   *Fisheries,* 41, 413-416. doi:10.1080/03632415.2016.1182509.

769 Chubb, J.C. (1979) Seasonal occurrences of helminths in freshwater fishes. Part II.
770   Trematoda. *Advances in Parasitology,* 17, 141-313.

771 Clements, W.H., Brooks, M.L., Kashian, D.R. and Zuellig, R.E. (2008) Changes in dissolved
772   organic material determine exposure of stream benthic communities to UV-B
773   radiation and heavy metals: implications for climate change. *Global Change Biology,*
774   14, 2201-2214. doi:10.1111/j.1365-2486.2008.01632.x.

775 Collingsworth, P.D., Bunnell, D.B., Murray, M.W., Kao, Y.C., Feiner, Z.S., Claramunt,
776   R.M., Lofgren, B.M., Hook, T.O. and Ludsin, S.A. (2017) Climate change as a long-
777   term stressor for the fisheries of the Laurentian Great Lakes of North America.
778   *Reviews in Fish Biology and Fisheries,* 27, 363-391. doi:10.1007/s11160-017-9480-3.

779 Crowden, A.E. and Broom, D.M. (1980) Effects of the eyefluke, *Diplostomum spathaceum*,
780   on the behavior of dace (*Leuciscus leuciscus*). *Animal Behaviour,* 28, 287-294.
781   doi:10.1016/s0003-3472(80)80031-5.

782 Désilets, H.D., Locke, S.A., McLaughlin, J.D. and Marcogliese, D.J. (2013) Community
783   structure of *Diplostomum* spp. (Digenea: Diplostomidae) in eyes of fish: Main
784   determinants and potential interspecific interactions. *International Journal for*
785   *Parasitology,* 43, 929-939. doi:10.1016/j.ijpara.2013.07.002.

786 Dittmar, J., Janssen, H., Kuske, A., Kurtz, J. and Scharsack, J.P. (2014) Heat and immunity:
787   an experimental heat wave alters immune functions in three-spined sticklebacks
788   (*Gasterosteus aculeatus*). *Journal of Animal Ecology,* 83, 744-757. doi:10.1111/1365-
789   2656.12175.

790   Ficke, A.D., Myrick, C.A. and Hansen, L.J. (2007) Potential impacts of global climate
791   change on freshwater fisheries. *Reviews in Fish Biology and Fisheries,* 17, 581-613.
792   doi:10.1007/s11160-007-9059-5.

793   Field, J.S. and Irwin, S.W.B. (1994) The epidemiology, treatment and control of
794   diplostomiasis on a fish farm in Northern Ireland. *In:* Pike, A.W. and Lewis, J.W.
795   (eds.) *Parasitic diseases of fish.* Samara Publishing Limited, Dyfed, pp. 87-100.

796   Francová, K., Ondračková, M., Polacik, M. and Jurajda, P. (2011) Parasite fauna of native
797   and non-native populations of *Neogobius melanostomus* (Pallas, 1814) (Gobiidae) in
798   the longitudinal profile of the Danube River. *Journal of Applied Ichthyology,* 27, 879-
799   886. doi:10.1111/j.1439-0426.2010.01582.x.

800   Franke, F., Rahn, A.K., Dittmar, J., Erin, N., Rieger, J.K., Haase, D., Samonte-Padilla, I.E.,
801   Lange, J., Jakobsen, P.J., Hermida, M.*, et al.* (2014) In vitro leukocyte response of
802   three-spined sticklebacks (*Gasterosteus aculeatus*) to helminth parasite antigens. *Fish
803   & Shellfish Immunology,* 36, 130-140. doi:10.1016/j.fsi.2013.10.019.

804   Gendron, A.D., Marcogliese, D.J. and Thomas, M. (2012) Invasive species are less
805   parasitized than native competitors, but for how long? The case of the round goby in
806   the Great Lakes-St. Lawrence Basin. *Biological Invasions,* 14, 367-384.
807   doi:10.1007/s10530-011-0083-y.

808   Gendron, A.D. and Marcogliese, D.J. (2017) Enigmatic decline of a common fish parasite
809   (*Diplostomum* spp.) in the St. Lawrence River: Evidence for a dilution effect induced
810   by the invasive round goby. *International Journal for Parasitology-Parasites and
811   Wildlife,* 6, 402-411. doi:10.1016/j.ijppaw.2017.04.002.

812   Gérard, C., Brient, L. and Le Rouzic, B. (2005) Variation in the response of juvenile and
813   adult gastropods (*Lymnaea stagnalis*) to cyanobacterial toxin (microcystin-LR).
814   *Environmental Toxicology,* 20, 592-596. doi:10.1002/tox.20147.

815   Haase, D., Rieger, J.K., Witten, A., Stoll, M., Bornberg-Bauer, E., Kalbe, M. and Reusch,
816   T.B.H. (2014) Specific gene expression responses to parasite genotypes reveal
817   redundancy of innate immunity in vertebrates. *PLoS One,* 9.
818   doi:10.1371/journal.pone.0108001.

819   Haase, D., Rieger, J.K., Witten, A., Stoll, M., Bornberg-Bauer, E., Kalbe, M. and Reusch,
820   T.B.H. (2016) Immunity comes first: The effect of parasite genotypes on adaptive
821   immunity and immunization in three-spined sticklebacks. *Developmental and
822   Comparative Immunology,* 54, 137-144. doi:10.1016/j.dci.2015.09.008.

823 Hakalahti, T., Karvonen, A. and Valtonen, E.T. (2006) Climate warming and disease risks in

824   temperate regions - *Argulus coregoni* and *Diplostomum spathaceum* as case studies.

825   *Journal of Helminthology,* 80, 93-98. doi:10.1079/joh2006351.

826 Harris, A.L. (1986) Larval trematode infections of the freshwater snail *Lymnaea peregra*

827   (Muller). M. Phil. Thesis. Queen Mary & Westfield College, University of London.

828 Hooper, M.J., Ankley, G.T., Cristol, D.A., Maryoung, L.A., Noyes, P.D. and Pinkerton, K.E.

829   (2013) Interactions between chemical and climate stressors: A role for mechanistic

830   toxicology in assessing climate change risks. *Environmental Toxicology and*

831   *Chemistry,* 32, 32-48. doi:10.1002/etc.2043.

832 Hudson, P.J., Cattadori, M., Boag, B. and Dobson, A.P. (2006) Climate disruption and

833   parasite-host dynamics: patterns and processes associated with warming and the

834   frequency of extreme climatic events. *Journal of Helminthology,* 80, 175-182.

835   doi:10.1079/joh2006357.

836 Höglund, J. and Thulin, J. (1990) The epidemiology of the metacercariae of *Diplostomum*

837   *baeri* and *Diplostomum spathaceum* in perch (*Perca fluviatilis*) from the warm water

838   effluent of a nuclear power station. *Journal of Helminthology,* 64, 139-150.

839 Höglund, J. and Thuvander, A. (1990) Indications of non-specific protective mechanisms in

840   rainbow trout *Oncorhynchus mykiss* with diplostomosis. *Diseases of Aquatic*

841   *Organisms,* 8, 91-97.

842 IPCC (2018) Summary for Policymakers. *In:* Masson-Delmotte, V., Zhai, P., Pörtner, H.-O.,

843   Roberts, D., Skea, J., Shukla, P.R., Pirani, A., Moufouma-Okia, W., Péan, C.,

844   Pidcock, R.*, et al.* (eds.) *Global Warming of 1.5°C. An IPCC Special Report on the*

845   *impacts of global warming of 1.5°C above pre-industrial levels and related global*

846   *greenhouse gas emission pathways, in the context of strengthening the global*

847   *response to the threat of climate change, sustainable development, and efforts to*

848   *eradicate poverty.* World Meteorological Organization, Geneva, Switzerland, pp. 32.

849 Janovy, J., Snyder, S.D. and Clopton, R.E. (1997) Evolutionary constraints on population

850   structure: The parasites of *Fundulus zebrinus* (Pisces: Cyprinodontidae) in the South

851   Platte River of Nebraska. *Journal of Parasitology,* 83, 584-592. doi:10.2307/3284228.

852 Jiménez Cisneros, B.E., Oki, T., Arnell, N.W., Benito, G., Cogley, J.G., Döll, P., Jiang, T.

853   and Mwakalila, S.S. (2014) Freshwater resources. *In:* Field, C.B., Barros, V.R.,

854   Dokken, D.J., Mach, K.J., Mastrandrea, M.D., Bilir, T.E., Chatterjee, M., Ebi, K.L.,

855   Estrada, Y.O., Genova, R.C.*, et al.* (eds.) *Climate Change 2014: Impacts, Adaptation,*

856   *and Vulnerability. Part A: Global and Sectoral Aspects. Contribution of Working*

857   *Group II to the Fifth Assessment Report of the Intergovernmental Panel on Climate*

858   *Change.* Cambridge University Press, Cambridge, United Kingdom and New York,

859   New York, USA, pp. 229-269.

860 Johnson, A.C., Acreman, M.C., Dunbar, M.J., Feist, S.W., Giacomello, A.M., Gozlan, R.E.,

861   Hinsley, S.A., Ibbotson, A.T., Jarvie, H.P., Jones, J.I.*, et al.* (2009) The British river

862   of the future: How climate change and human activity might affect two contrasting

863   river ecosystems in England. *Science of the Total Environment,* 407, 4787-4798.

864   doi:10.1016/j.scitotenv.2009.05.018.

865 Johnson, P.T.J. and Chase, J.M. (2004) Parasites in the food web: linking amphibian

866   malformations and aquatic eutrophication. *Ecology Letters,* 7, 521-526.

867   doi:10.1111/j.1461-0248.2004.00610.x.

868 Johnson, P.T.J., Chase, J.M., Dosch, K.L., Hartson, R.B., Gross, J.A., Larson, D.J.,

869   Sutherland, D.R. and Carpenter, S.R. (2007) Aquatic eutrophication promotes

870   pathogenic infection in amphibians. *Proceedings of the National Academy of Sciences*

871   *of the United States of America,* 104, 15781-15786. doi:10.1073/pnas.0707763104.

872 Kalbe, M. and Kurtz, J. (2006) Local differences in immunocompetence reflect resistance of

873   sticklebacks against the eye fluke *Diplostomum pseudospathaceum. Parasitology,*

874   132, 105-116. doi:10.1017/s0031182005008681.

875 Karvonen, A., Paukku, S., Valtonen, E.T. and Hudson, P.J. (2003) Transmission, infectivity

876   and survival of *Diplostomum spathaceum* cercariae. *Parasitology,* 127, 217-224.

877   doi:10.1017/s0031182003003561.

878 Karvonen, A., Kirsi, S., Hudson, P.J. and Valtonen, E.T. (2004a) Patterns of cercarial

879   production from *Diplostomum spathaceum*: terminal investment or bet hedging?

880   *Parasitology,* 129, 87-92. doi:10.1017/s0031182004005281.

881 Karvonen, A., Seppälä, O. and Valtonen, E.T. (2004b) Parasite resistance and avoidance

882   behaviour in preventing eye fluke infections in fish. *Parasitology,* 129, 159-164.

883   doi:10.1017/s0031182004005505.

884 Karvonen, A., Seppälä, O. and Valtonen, E.T. (2004c) Eye fluke-induced cataract formation

885   in fish: quantitative analysis using an ophthalmological microscope. *Parasitology,*

886   129, 473-478. doi:10.1017/s0031182004006006.

887 Karvonen, A., Paukku, S., Seppälä, O. and Valtonen, E.T. (2005) Resistance against eye

888   flukes: naive versus previously infected fish. *Parasitology Research,* 95, 55-59.

889   doi:10.1007/s00436-004-1246-x.

890   Karvonen, A., Savolainen, M., Seppälä, O. and Valtonen, E.T. (2006a) Dynamics of
891   *Diplostomum spathaceum* infection in snail hosts at a fish farm. *Parasitology*
892   *Research,* 99, 341-345. doi:10.1007/s00436-006-0137-8.

893   Karvonen, A., Terho, P., Seppälä, O., Jokela, J. and Valtonen, E.T. (2006b) Ecological
894   divergence of closely related *Diplostomum* (Trematoda) parasites. *Parasitology,* 133,
895   229-235. doi:10.1017/s0031182006000242.

896   Karvonen, A. and Seppälä, O. (2008a) Eye fluke infection and lens size reduction in fish: a
897   quantitative analysis. *Diseases of Aquatic Organisms,* 80, 21-26.
898   doi:10.3354/dao01918.

899   Karvonen, A. and Seppälä, O. (2008b) Effect of eye fluke infection on the growth of
900   whitefish (*Coregonus lavaretus*) - An experimental approach. *Aquaculture,* 279, 6-10.
901   doi:10.1016/j.aquaculture.2008.04.013.

902   Karvonen, A., Halonen, H. and Seppälä, O. (2010a) Priming of host resistance to protect
903   cultured rainbow trout *Oncorhynchus mykiss* against eye flukes and parasite-induced
904   cataracts. *Journal of Fish Biology,* 76, 1508-1515. doi:10.1111/j.1095-
905   8649.2010.02597.x.

906   Karvonen, A., Rintamäki, P., Jokela, J. and Valtonen, E.T. (2010b) Increasing water
907   temperature and disease risks in aquatic systems: climate change increases the risk of
908   some, but not all, diseases. *International Journal for Parasitology,* 40, 1483-1488.
909   doi:10.1016/j.ijpara.2010.04.015.

910   Karvonen, A. (2012) *Diplostomum spathaceum* and Related Species. *In:* Woo, P.T.K. and
911   Buchmann, K. (eds.) *Fish Parasites: Pathobiology and Protection.* CAB
912   International, Oxfordshire, UK, pp. 260-269.

913   Karvonen, A., Kristjansson, B.K., Skulason, S., Lanki, M., Rellstab, C. and Jokela, J. (2013)
914   Water temperature, not fish morph, determines parasite infections of sympatric
915   Icelandic threespine sticklebacks (*Gasterosteus aculeatus*). *Ecology and Evolution,* 3,
916   1507-1517. doi:10.1002/ece3.568.

917   Karvonen, A., Lucek, K., Marques, D.A. and Seehausen, O. (2015) Divergent macroparasite
918   infections in parapatric Swiss lake-stream pairs of threespine stickleback
919   (*Gasterosteus aculeatus*). *PLoS One,* 10. doi:10.1371/journal.pone.0130579.

920   Karvonen, A. and Lindström, K. (2018) Spatiotemporal and gender-specific parasitism in two
921   species of gobiid fish. *Ecology and Evolution,* 8, 6114-6123. doi:10.1002/ece3.4151.

922 Klemme, I. and Karvonen, A. (2017) Vertebrate defense against parasites: Interactions
923   between avoidance, resistance, and tolerance. *Ecology and Evolution,* 7, 561-571.
924   doi:10.1002/ece3.2645.

925 Koie, M. (1999) Metazoan parasites of flounder *Platichthys flesus* (L.) along a transect from
926   the southwestern to the northeastern Baltic Sea. *Ices Journal of Marine Science,* 56,
927   157-163. doi:10.1006/jmsc.1999.0463.

928 Kvach, Y. and Winkler, H.M. (2011) The colonization of the invasive round goby *Neogobius*
929   *melanostomus* by parasites in new localities in the southwestern Baltic Sea.
930   *Parasitology Research,* 109, 769-780. doi:10.1007/s00436-011-2321-8.

931 Lafferty, K.D. (2009) The ecology of climate change and infectious diseases. *Ecology,* 90,
932   888-900. doi:10.1890/08-0079.1.

933 Landis, W.G., Rohr, J.R., Moe, S.J., Balbus, J.M., Clements, W., Fritz, A., Helm, R., Hickey,
934   C., Hooper, M., Stahl, R.G.*, et al.* (2014) Global climate change and contaminants, a
935   call to arms not yet heard? *Integrated Environmental Assessment and Management,*
936   10, 483-484. doi:10.1002/ieam.1568.

937 Leicht, K., Jokela, J. and Seppälä, O. (2013) An experimental heat wave changes immune
938   defense and life history traits in a freshwater snail. *Ecology and Evolution,* 3, 4861-
939   4871. doi:10.1002/ece3.874.

940 Leicht, K. and Seppälä, O. (2014) Infection success of *Echinoparyphium aconiatum*
941   (Trematoda) in its snail host under high temperature: role of host resistance. *Parasites*
942   *& Vectors,* 7, 192. doi:10.1186/1756-3305-7-192.

943 Leicht, K., Seppälä, K. and Seppälä, O. (2017) Potential for adaptation to climate change:
944   family-level variation in fitness-related traits and their responses to heat waves in a
945   snail population. *BMC Evolutionary Biology,* 17, 140. doi:10.1186/s12862-017-0988-
946   x.

947 Locke, S.A., McLaughlin, J.D., Dayanandan, S. and Marcogliese, D.J. (2010a) Diversity and
948   specificity in *Diplostomum* spp. metacercariae in freshwater fishes revealed by
949   cytochrome c oxidase I and internal transcribed spacer sequences. *International*
950   *Journal for Parasitology,* 40, 333-343. doi:10.1016/j.ijpara.2009.08.012.

951 Locke, S.A., McLaughlin, J.D. and Marcogliese, D.J. (2010b) DNA barcodes show cryptic
952   diversity and a potential physiological basis for host specificity among
953   Diplostomoidea (Platyhelminthes: Digenea) parasitizing freshwater fishes in the St.
954   Lawrence River, Canada. *Molecular Ecology,* 19, 2813-2827. doi:10.1111/j.1365-
955   294X.2010.04713.x.

956   Locke, S.A., Al-Nasiri, F.S., Caffara, M., Drago, F., Kalbe, M., Lapierre, A.R., McLaughlin,
957   J.D., Nie, P., Overstreet, R.M., Souza, G.T.R*., et al.* (2015) Diversity, specificity and
958   speciation in larval Diplostomidae (Platyhelminthes: Digenea) in the eyes of
959   freshwater fish, as revealed by DNA barcodes. *International Journal for*
960   *Parasitology,* 45, 841-855. doi:10.1016/j.ijpara.2015.07.001.

961   Lõhmus, M. and Björklund, M. (2015) Climate change: what will it do to fish-parasite
962   interactions? *Biological Journal of the Linnean Society,* 116, 397-411.
963   doi:10.1111/bij.12584.

964   Louhi, K.-R., Karvonen, A., Rellstab, C., Louhi, R. and Jokela, J. (2013) Prevalence of
965   infection as a predictor of multiple genotype infection frequency in parasites with
966   multiple-host life cycle. *Journal of Animal Ecology,* 82, 191-200. doi:10.1111/j.1365-
967   2656.2012.02028.x.

968   Lyholt, H.C.K. and Buchmann, K. (1996) *Diplostomum spathaceum*: Effects of temperature
969   and light on cercarial shedding and infection of rainbow trout. *Diseases of Aquatic*
970   *Organisms,* 25, 169-173. doi:10.3354/dao025169.

971   Marcogliese, D.J. and Compagna, S. (1999) Diplostomatid eye flukes in young-of-the-year
972   and forage fishes in the St. Lawrence River, Quebec. *Journal of Aquatic Animal*
973   *Health,* 11, 275-282. doi:10.1577/1548-8667(1999)011<0275:defiyo>2.0.co;2.

974   Marcogliese, D.J., Rodrigue, J., Ouellet, M. and Champoux, L. (2000) Natural occurrence of
975   *Diplostomum* sp (Digenea : Diplostomatidae) in adult mudpuppies and bullfrog
976   tadpoles from the St. Lawrence River, Quebec. *Comparative Parasitology,* 67, 26-31.

977   Marcogliese, D.J. (2001) Implications of climate change for parasitism of animals in the
978   aquatic environment. *Canadian Journal of Zoology,* 79, 1331-1352. doi:10.1139/cjz-
979   79-8-1331.

980   Marcogliese, D.J., Compagna, S., Bergeron, E. and McLaughlin, J.D. (2001a) Population
981   biology of eyeflukes in fish from a large fluvial ecosystem: the importance of gulls
982   and habitat characteristics. *Canadian Journal of Zoology,* 79, 1102-1113.
983   doi:10.1139/cjz-79-6-1102.

984   Marcogliese, D.J., Dumont, P., Gendron, A.D., Mailhot, Y., Bergeron, E. and McLaughlin,
985   J.D. (2001b) Spatial and temporal variation in abundance of *Diplostomum* spp. in
986   walleye (*Stizostedion vitreum*) and white suckers (*Catostomus commersoni*) from the
987   St. Lawrence River. *Canadian Journal of Zoology,* 79, 355-369. doi:10.1139/cjz-79-
988   3-355.

989   Marcogliese, D.J. (2004) Parasites: small players with crucial roles in the ecological theater.
990   *EcoHealth,* 1, 151-164.

991   Marcogliese, D.J. (2005) Parasites of the superorganism: Are they indicators of ecosystem
992   health? *International Journal for Parasitology,* 35, 705-716.
993   doi:10.1016/j.ijpara.2005.01.015.

994   Marcogliese, D.J. (2008) The impact of climate change on the parasites and infectious
995   diseases of aquatic animals. *Revue Scientifique Et Technique-Office International Des*
996   *Epizooties,* 27, 467-484. doi:10.20506/rst.27.2.1820.

997   Marcogliese, D.J., Dautremepuits, C., Gendron, A.D. and Fournier, M. (2010) Interactions
998   between parasites and pollutants in yellow perch (*Perca flavescens*) in the St.
999   Lawrence River, Canada: implications for resistance and tolerance to parasites.
1000   *Canadian Journal of Zoology,* 88, 247-258. doi:10.1139/z09-140.

1001   Marcogliese, D.J. and Pietrock, M. (2011) Combined effects of parasites and contaminants on
1002   animal health: parasites do matter. *Trends in Parasitology,* 27, 123-130.
1003   doi:10.1016/j.pt.2010.11.002.

1004   Marcogliese, D.J. (2016) The distribution and abundance of parasites in aquatic ecosystems
1005   in a changing climate: More than just temperature. *Integrative and Comparative*
1006   *Biology,* 56, 611-619. doi:10.1093/icb/icw036.

1007   Margolis, L. and Arthur, J.R. (1979) Synopis of the parasites of fishes of Canada. *Bulletin of*
1008   *the Fisheries Research Board of Canada,* 199, 269.

1009   Markkula, S.E., Karvonen, A., Salo, H., Valtonen, E.T. and Jokinen, E.I. (2007) Ultraviolet B
1010   irradiation affects resistance of rainbow trout (*Oncorhynchus mykiss*) against
1011   bacterium *Yersinia ruckeri* and trematode *Diplostomum spathaceum. Photochemistry*
1012   *and Photobiology,* 83, 1263-1269. doi:10.1111/j.1751-1097.2007.00165.x.

1013   Martin, L.B., Hopkins, W.A., Mydlarz, L.D. and Rohr, J.R. (2010) The effects of
1014   anthropogenic global changes on immune functions and disease resistance. *Annals of*
1015   *the New York Academy of Sciences,* 1195, 129-148. doi:10.1111/j.1749-
1016   6632.2010.05454.x.

1017   McDonald, T.E. and Margolis, L. (1995) Synopsis of the parasites of fishes of Canada:
1018   Supplement (1978-1993). *Canadian Special Publication of Fisheries and Aquatic*
1019   *Sciences,* 122, 265.

1020   McKeown, C.A. and Irwin, S.W.B. (1997) Accumulation of *Diplostomum* spp. (Digenea:
1021   Diplostomatidae) metacercariae in the eyes of 0+ and 1+ roach (*Rutilus rutilus*).

1022 *International Journal for Parasitology,* 27, 377-380. doi:10.1016/s0020-
1023 7519(96)00204-4.

1024 Milotic, M., Milotic, D. and Koprivnikar, J. (2018) Exposure to a cyanobacterial toxin
1025 increases larval amphibian susceptibility to parasitism. *Parasitology Research,* 117,
1026 513-520. doi:10.1007/s00436-017-5727-0.

1027 Moe, S.J., De Schamphelaere, K., Clements, W.H., Sorensen, M.T., Van den Brink, P.J. and
1028 Liess, M. (2013) Combined and interactive effects of global climate change and
1029 toxicants on populations and communities. *Environmental Toxicology and Chemistry,*
1030 32, 49-61. doi:10.1002/etc.2045.

1031 Moody, J. and Gaten, E. (1982) The population dynamics of eyeflukes *Diplostomum*
1032 *spathaceum* and *Tylodelphys clavata* (Digenea, Diplostomatidae) in rainbow and
1033 brown trout in Rutland water - 1974-1978. *Hydrobiologia,* 88, 207-209.
1034 doi:10.1007/bf00008315.

1035 Morley, N.J., Crane, M. and Lewis, J.W. (2001) Toxicity of cadmium and zinc to
1036 *Diplostomum spathaceum* (Trematoda : Diplostomidae) cercarial survival.
1037 *International Journal for Parasitology,* 31, 1211-1217. doi:10.1016/s0020-
1038 7519(01)00229-6.

1039 Morley, N.J., Crane, M. and Lewis, J.W. (2002) Toxicity of cadmium and zinc mixtures to
1040 *Diplostomum spathaceum* (Trematoda : Diplostomidae) cercarial survival. *Archives of*
1041 *Environmental Contamination and Toxicology,* 43, 28-33. doi:10.1007/s00244-002-
1042 1244-x.

1043 Morley, N.J., Crane, M. and Lewis, J.W. (2003a) Cadmium toxicity and snail-digenean
1044 interactions in a population of *Lymnaea* spp. *Journal of Helminthology,* 77, 49-55.
1045 doi:10.1079/joh2002148.

1046 Morley, N.J., Crane, M. and Lewis, J.W. (2003b) Toxicity of cadmium and zinc to the
1047 cercarial activity of *Diplostomum spathaceum* (Trematoda : Diplostomidae). *Folia*
1048 *Parasitologica,* 50, 57-60. doi:10.14411/fp.2003.011.

1049 Morley, N.J., Irwin, S.W.B. and Lewis, J.W. (2003c) Pollution toxicity to the transmission of
1050 larval digeneans through their molluscan hosts. *Parasitology,* 126, S5-S26.
1051 doi:10.1017/s0031182003003755.

1052 Morley, N.J. (2011) Thermodynamics of cercarial survival and metabolism in a changing
1053 climate. *Parasitology,* 138, 1442-1452. doi:10.1017/s0031182011001272.

1054 Morley, N.J. (2012) Thermodynamics of miracidial survival and metabolism. *Parasitology,*
1055 139, 1640-1651. doi:10.1017/s0031182012000960.

1056 Morley, N.J. and Lewis, J.W. (2013) Thermodynamics of cercarial development and
1057   emergence in trematodes. *Parasitology,* 140, 1211-1224.
1058   doi:10.1017/s0031182012001783.

1059 Morley, N.J. and Lewis, J.W. (2014) Extreme climatic events and host-pathogen interactions:
1060   The impact of the 1976 drought in the UK. *Ecological Complexity,* 17, 1-19.
1061   doi:10.1016/j.ecocom.2013.12.001.

1062 Morley, N.J. and Lewis, J.W. (2015) Thermodynamics of trematode infectivity. *Parasitology,*
1063   142, 585-597. doi:10.1017/s0031182014001632.

1064 Moszczynska, A., Locke, S.A., McLaughlin, J.D., Marcogliese, D.J. and Crease, T.J. (2009)
1065   Development of primers for the mitochondrial cytochrome c oxidase I gene in
1066   digenetic trematodes (Platyhelminthes) illustrates the challenge of barcoding parasitic
1067   helminths. *Molecular Ecology Resources,* 9, 75-82. doi:10.1111/j.1755-
1068   0998.2009.02634.x.

1069 Muhlegger, J.M., Jirsa, F., Konecny, R. and Frank, C. (2010) Parasites of *Apollonia*
1070   *melanostoma* (Pallas 1814) and *Neogobius kessleri* (Guenther 1861) (Osteichthyes,
1071   Gobiidae) from the Danube River in Austria. *Journal of Helminthology,* 84, 87-92.
1072   doi:10.1017/s0022149x09990095.

1073 Muller, M. (2019) Dams have the power to slow climate change. *Nature,* 566, 315-317.

1074 Niewiadomska, K. (1984) Present status of *Diplostomum spathaceum* (Rudolphi, 1819) and
1075   differentiation of *Diplostomum pseudospathaceum* nom. nov. (Trematoda:
1076   Diplostomatidae). *Systematic Parasitology,* 6, 81-86. doi:10.1007/bf02185515.

1077 Niewiadomska, K. (1986) Verification of the life-cycles of *Diplostomum spathaceum*
1078   (Rudolphi,1819) and *D. pseudospathaceum* Niewiadomska, 1984 (Trematoda:
1079   Diplostomidae). *Systematic Parasitology,* 8, 23-31. doi:10.1007/bf00010306.

1080 Niewiadomska, K. and Kiseliene, V. (1994) *Diplostomum* cercariae (Digenea) in snails from
1081   Lithuania. II. Survey of species. *Acta Parasitologica,* 39, 179-186.

1082 Niewiadomska, K. and Laskowski, Z. (2002) Systematic relationships among six species of
1083   *Diplostomum* Nordmann, 1832 (Digenea) based on morphological and molecular
1084   data. *Acta Parasitologica,* 47, 20-28.

1085 Noyes, P.D., McElwee, M.K., Miller, H.D., Clark, B.W., Van Tiem, L.A., Walcott, K.C.,
1086   Erwin, K.N. and Levin, E.D. (2009) The toxicology of climate change: Environmental
1087   contaminants in a warming world. *Environment International,* 35, 971-986.
1088   doi:10.1016/j.envint.2009.02.006.

1089   Ondračková, M., Davidova, M., Blazek, R., Gelnar, M. and Jurajda, P. (2009) The interaction
1090   between an introduced fish host and local parasite fauna: *Neogobius kessleri* in the
1091   middle Danube River. *Parasitology Research,* 105, 201-208. doi:10.1007/s00436-
1092   009-1384-2.

1093   Ondračková, M., Hudcova, I., Davidova, M., Adamek, Z., Kasny, M. and Jurajda, P. (2015)
1094   Non-native gobies facilitate the transmission of *Bucephalus polymorphus*
1095   (Trematoda). *Parasites & Vectors,* 8. doi:10.1186/s13071-015-0999-7.

1096   Owen, S.F., Barber, I. and Hart, P.J.B. (1993) Low-level infection by eye fluke, *Diplostomum*
1097   spp, affects the vision of 3-spined sticklebacks, *Gasterosteus aculeatus*. *Journal of*
1098   *Fish Biology,* 42, 803-806.

1099   Paaijmans, K.P., Blanford, S., Bell, A.S., Blanford, J.I., Read, A.F. and Thomas, M.B. (2010)
1100   Influence of climate on malaria transmission depends on daily temperature variation.
1101   *Proceedings of the National Academy of Sciences of the United States of America,*
1102   107, 15135-15139. doi:10.1073/pnas.1006422107.

1103   Padros, F., Knudsen, R. and Blasco-Costa, I. (2018) Histopathological characterisation of
1104   retinal lesions associated to *Diplostomum* species (Platyhelminthes: Trematoda)
1105   infection in polymorphic Arctic charr *Salvelinus alpinus*. *International Journal for*
1106   *Parasitology-Parasites and Wildlife,* 7, 68-74. doi:10.1016/j.ijppaw.2018.01.007.

1107   Paerl, H.W., Hall, N.S. and Calandrino, E.S. (2011) Controlling harmful cyanobacterial
1108   blooms in a world experiencing anthropogenic and climatic-induced change. *Science*
1109   *of the Total Environment,* 409, 1739-1745. doi:10.1016/j.scitotenv.2011.02.001.

1110   Paull, S.H. and Johnson, P.T.J. (2011) High temperature enhances host pathology in a snail-
1111   trematode system: possible consequences of climate change for the emergence of
1112   disease. *Freshwater Biology,* 56, 767-778. doi:10.1111/j.1365-2427.2010.02547.x.

1113   Paull, S.H., LaFonte, B.E. and Johnson, P.T.J. (2012) Temperature-driven shifts in a host-
1114   parasite interaction drive nonlinear changes in disease risk. *Global Change Biology,*
1115   18, 3558-3567. doi:10.1111/gcb.12018.

1116   Paull, S.H. and Johnson, P.T.J. (2014) Experimental warming drives a seasonal shift in the
1117   timing of host-parasite dynamics with consequences for disease risk. *Ecology Letters,*
1118   17, 445-453. doi:10.1111/ele.12244.

1119   Pennycuick, L. (1971) Differences in the parasite infections in three-spined sticklebacks
1120   (*Gasterosteus aculeatus* L.) of different sex, age and size. *Parasitology,* 63, 407-418.

1121   Pietrock, M., Meinelt, T., Marcogliese, D.J. and Steinberg, C.E.W. (2001) Influence of
1122   aqueous sediment extracts from the Oder River (Germany/Poland) on survival of

1123   *Diplostomum* sp (Trematoda : Diplostomidae) cercariae. *Archives of Environmental*

1124   *Contamination and Toxicology,* 40, 327-332.

1125 Pietrock, M., Marcogliese, D.J. and McLaughlin, J.D. (2002a) Effects of cadmium upon

1126   longevity of *Diplostomum* sp (Trematoda : Diplostomidae) cercariae. *Chemosphere,*

1127   47, 29-33. doi:10.1016/s0045-6535(01)00283-1.

1128 Pietrock, M., Marcogliese, D.J., Meinelt, T. and McLaughlin, J.D. (2002b) Effects of

1129   mercury and chromium upon longevity of *Diplostomum* sp (Trematoda :

1130   Diplostomidae) cercariae. *Parasitology Research,* 88, 225-229. doi:10.1007/s00436-

1131   001-0529-8.

1132 Pietrock, M. and Marcogliese, D.J. (2003) Free-living endohelminth stages: at the mercy of

1133   environmental conditions. *Trends in Parasitology,* 19, 293-299. doi:10.1016/s1471-

1134   4922(03)00117-x.

1135 Pietrock, M. and Goater, C.P. (2005) Infectivity of *Ornithodiplostomum ptychocheilus* and

1136   *Posthodiplostomum minimum* (Trematoda : Diplostomidae) cercariae following

1137   exposure to cadmium. *Journal of Parasitology,* 91, 854-856. doi:10.1645/ge-473r.1.

1138 Poulin, R. (2006) Global warming and temperature-mediated increases in cercarial

1139   emergence in trematode parasites. *Parasitology,* 132, 143-151.

1140   doi:10.1017/s0031182005008693.

1141 Poulin, R. and Mouritsen, K.N. (2006) Climate change, parasitism and the structure of

1142   intertidal ecosystems. *Journal of Helminthology,* 80, 183-191.

1143   doi:10.1079/joh2006341.

1144 Prophete, C., Carlson, E.A., Li, Y., Duffy, J., Steinetz, B., Lasano, S. and Zelikoff, J.T.

1145   (2006) Effects of elevated temperature and nickel pollution on the immune status of

1146   Japanese medaka. *Fish & Shellfish Immunology,* 21, 325-334.

1147   doi:10.1016/j.fsi.2005.12.009.

1148 Raffel, T.R., Romansic, J.M., Halstead, N.T., McMahon, T.A., Venesky, M.D. and Rohr, J.R.

1149   (2013) Disease and thermal acclimation in a more variable and unpredictable climate.

1150   *Nature Climate Change,* 3, 146-151. doi:10.1038/nclimate1659.

1151 Raffel, T.R., Halstead, N.T., McMahon, T.A., Davis, A.K. and Rohr, J.R. (2015)

1152   Temperature variability and moisture synergistically interact to exacerbate an

1153   epizootic disease. *Proceedings of the Royal Society B,* 282.

1154   doi:10.1098/rspb.2014.2039.

1155 Ratanarat-Brockelman, C. (1974) Migration of *Diplostomum spathaceum* (Trematoda) in the

1156   fish intermediate host. *Zeitschrift für Parasitenkunde,* 43, 123-134.

1157 Rauch, G., Kalbe, M. and Reusch, T.B.H. (2005) How a complex life cycle can improve a
1158   parasite's sex life. *Journal of Evolutionary Biology,* 18, 1069-1075.
1159   doi:10.1111/j.1420-9101.2005.00895.x.

1160 Rellstab, C., Louhi, K.R., Karvonen, A. and Jokela, J. (2011) Analysis of trematode parasite
1161   communities in fish eye lenses by pyrosequencing of naturally pooled DNA.
1162   *Infection, Genetics and Evolution,* 11, 1276-1286. doi:10.1016/j.meegid.2011.04.018.

1163 Rellstab, C., Karvonen, A., Louhi, K.-R. and Jokela, J. (2013) Genotype-specific vs. cross-
1164   reactive host immunity against a macroparasite. *PLoS One,* 8.
1165   doi:10.1371/journal.pone.0078427.

1166 Riley, E.M. and Chappell, L.H. (1992) Effect of infection with *Diplostomum spathaceum* on
1167   the internal defense system of *Lymnaea stagnalis*. *Journal of Invertebrate Pathology,*
1168   59, 190-196. doi:10.1016/0022-2011(92)90032-y.

1169 Rohr, J.R., Dobson, A.P., Johnson, P.T.J., Kilpatrick, A.M., Paull, S.H., Raffel, T.R., Ruiz-
1170   Moreno, D. and Thomas, M.B. (2011) Frontiers in climate change-disease research.
1171   *Trends in Ecology & Evolution,* 26, 270-277. doi:10.1016/j.tree.2011.03.002.

1172 Rohr, J.R. and Palmer, B.D. (2013) Climate change, multiple stressors, and the decline of
1173   ectotherms. *Conservation Biology,* 27, 741-751. doi:10.1111/cobi.12086.

1174 Rohr, J.R., Raffel, T.R., Blaustein, A.R., Johnson, P.T.J., Paull, S.H. and Young, S. (2013)
1175   Using physiology to understand climate-driven changes in disease and their
1176   implications for conservation. *Conservation Physiology,* 1, cot022.
1177   doi:10.1093/conphys/cot022.

1178 Salo, T., Stamm, C., Burdon, F.J., Räsänen, K. and Seppälä, O. (2017) Resilience to heat
1179   waves in the aquatic snail *Lymnaea stagnalis*: Additive and interactive effects with
1180   micropollutants. *Freshwater Biology,* 62, 1831-1846. doi:10.1111/fwb.12999.

1181 Scharsack, J.P. and Kalbe, M. (2014) Differences in susceptibility and immune responses of
1182   three-spined sticklebacks (*Gasterosteus aculeatus*) from lake and river ecotypes to
1183   sequential infections with the eye fluke *Diplostomum pseudospathaceum*. *Parasites &*
1184   *Vectors,* 7, 109. doi:10.1186/1756-3305-7-109.

1185 Scharsack, J.P., Franke, F., Erin, N.I., Kuske, A., Buscher, J., Stolz, H., Samonte, I.E., Kurtz,
1186   J. and Kalbe, M. (2016) Effects of environmental variation on host-parasite
1187   interaction in three-spined sticklebacks (*Gasterosteus aculeatus*). *Zoology,* 119, 375-
1188   383. doi:10.1016/j.zool.2016.05.008.

1189   Schiedek, D., Sundelin, B., Readman, J.W. and Macdonald, R.W. (2007) Interactions
1190   between climate change and contaminants. *Marine Pollution Bulletin,* 54, 1845-1856.
1191   doi:10.1016/j.marpolbul.2007.09.020.

1192   Seppälä, O., Karvonen, A. and Valtonen, E.T. (2004) Parasite-induced change in host
1193   behaviour and susceptibility to predation in an eye fluke - fish interaction. *Animal*
1194   *Behaviour,* 68, 257-263. doi:10.1016/j.anbehav.2003.10.021.

1195   Seppälä, O., Karvonen, A. and Valtonen, E.T. (2005a) Impaired crypsis of fish infected with
1196   a trophically transmitted parasite. *Animal Behaviour,* 70, 895-900.
1197   doi:10.1016/j.anbehav.2005.01.021.

1198   Seppälä, O., Karvonen, A. and Valtonen, E.T. (2005b) Manipulation of fish host by eye
1199   flukes in relation to cataract formation and parasite infectivity. *Animal Behaviour,* 70,
1200   889-894. doi:10.1016/j.anbehav.2005.01.020.

1201   Seppälä, O., Karvonen, A. and Valtonen, E.T. (2008) Shoaling behaviour of fish under
1202   parasitism and predation risk. *Animal Behaviour,* 75, 145-150.
1203   doi:10.1016/j.anbehav.2007.04.022.

1204   Seppälä, O., Karvonen, A. and Valtonen, E.T. (2011) Eye fluke-induced cataracts in natural
1205   fish populations: is there potential for host manipulation? *Parasitology,* 138, 209-214.
1206   doi:10.1017/s0031182010001228.

1207   Settele, J., Scholes, R., Betts, R., Bunn, S., Leadley, P., Nepstad, D., Overpeck, J.T. and
1208   Taboada, M.A. (2014) Terrestrial and inland water systems. *In:* Field, C.B., Barros,
1209   V.R., Dokken, D.J., Mach, K.J., Mastrandrea, M.D., Bilir, T.E., Chatterjee, M., Ebi,
1210   K.L., Estrada, Y.O., Genova, R.C.*, et al.* (eds.) *Climate Change 2014: Impacts,*
1211   *Adaptation, and Vulnerability. Part A: Global and Sectoral Aspects. Contribution of*
1212   *Working Group II to the Fifth Assessment Report of the Intergovernmental Panel on*
1213   *Climate Change.* Cambridge University Press, Cambridge, United Kingdom and New
1214   York, NY, USA, pp. 271-359.

1215   Shariff, M., Richards, R.H. and Sommerville, C. (1980) The histopathology of acute and
1216   chronic infections of rainbow trout *Salmo gairdneri* Richardson with eye flukes,
1217   *Diplostomum* spp. *Journal of Fish Diseases,* 3, 455-465. doi:10.1111/j.1365-
1218   2761.1980.tb00432.x.

1219   Sitjá-Bobadilla, A. (2008) Living off a fish: A trade-off between parasites and the immune
1220   system. *Fish & Shellfish Immunology,* 25, 358-372. doi:10.1016/j.fsi.2008.03.018.

1221 Sous, S.M. (1992) Influence of abiotic factors on emission and survival of *Diplostomum*
1222   *chromatophorum* (Brown,1931) (Trematoda, Diplostomatidae). *Ecological*
1223   *Parasitology,* 1, 154-159.

1224 Stables, J.N. and Chappell, L.H. (1986a) Putative immune response of rainbow trout, *Salmo*
1225   *gairdneri*, to *Diplostomum spathaceum* infections. *Journal of fish biology,* 29, 115-
1226   122. doi:10.1111/j.1095-8649.1986.tb04931.x.

1227 Stables, J.N. and Chappell, L.H. (1986b) *Diplostomum spathaceum* (Rud 1819) - effects of
1228   physical factors on the infection of rainbow trout (*Salmo gairdneri*) by cercariae.
1229   *Parasitology,* 93, 71-79. doi:10.1017/s0031182000049830.

1230 Stables, J.N. and Chappell, L.H. (1986c) The epidemiology of diplostomiasis in farmed
1231   rainbow trout from north-east Scotland. *Parasitology,* 92, 699-710.

1232 Stahl, R.G., Hooper, M.J., Balbus, J.M., Clements, W., Fritz, A., Gouin, T., Helm, R.,
1233   Hickey, C., Landis, W. and Moe, S.J. (2013) The influence of global climate change
1234   on the scientific foundations and applications of Environmental Toxicology and
1235   Chemistry: Introduction to a SETAC international workshop. *Environmental*
1236   *Toxicology and Chemistry,* 32, 13-19. doi:10.1002/etc.2037.

1237 Studer, A. and Poulin, R. (2014) Analysis of trait mean and variability versus temperature in
1238   trematode cercariae: is there scope for adaptation to global warming? *International*
1239   *Journal for Parasitology,* 44, 403-413. doi:10.1016/j.ijpara.2014.02.006.

1240 Sweeting, R.A. (1974) Investigations into natural and experimental infections of freshwater
1241   fish by common eye-fluke *Diplostomum spathaceum* Rud. *Parasitology,* 69, 291-300.

1242 Tort, L., Balasch, J.C. and Mackenzie, S. (2003) Fish immune system. A crossroads between
1243   innate and adaptive responses. *Inmunologia,* 22, 277-286.

1244 Tully, K., Gedan, K., Epanchin-Niell, R., Strong, A., Bernhardt, E.S., Bendor, T., Mitchell,
1245   M., Kominoski, J., Jordan, T.E., Neubauer, S.C*., et al.* (2019) The invisible flood: The
1246   chemistry, ecology, and social implications of coastal saltwater intrusion. *Bioscience,*
1247   69, 368-378. doi:10.1093/biosci/biz027.

1248 Uribe, C., Folch, H., Enriquez, R. and Moran, G. (2011) Innate and adaptive immunity in
1249   teleost fish: a review. *Veterinarni Medicina,* 56, 486-503.

1250 Waadu, G.D.B. and Chappell, L.H. (1991) Effect of water temperature on the ability of
1251   *Diplostomum spathaceum* miracidia to establish in lymnaeid snails. *Journal of*
1252   *Helminthology,* 65, 179-185. doi:10.1017/s0022149x00010671.

1253 Valtonen, E.T. and Gibson, D.I. (1997) Aspects of the biology of diplostomid metacercarial
1254   (Digenea) populations occurring in fishes in different localities of northern Finland.
1255   *Annales Zoologici Fennici,* 34, 47-59.

1256 Valtonen, E.T., Holmes, J.C. and Koskivaara, M. (1997) Eutrophication, pollution, and
1257   fragmentation: Effects on parasite communities in roach (*Rutilus rutilus*) and perch
1258   (*Perca fluviatilis*) in four lakes in central Finland. *Canadian Journal of Fisheries and*
1259   *Aquatic Sciences,* 54, 572-585.

1260 Wegner, K.M., Kalbe, M., Milinski, M. and Reusch, T.B.H. (2008) Mortality selection during
1261   the 2003 European heat wave in three-spined sticklebacks: effects of parasites and
1262   MHC genotype. *BMC Evolutionary Biology,* 8. doi:10.1186/1471-2148-8-124.

1263 Verma, A.K., Pal, A.K., Manush, S.M., Das, T., Dalvi, R.S., Chandrachoodan, P.P., Ravi,
1264   P.M. and Apte, S.K. (2007) Persistent sub-lethal chlorine exposure augments
1265   temperature induced immunosuppression in *Cyprinus carpio* advanced fingerlings.
1266   *Fish & Shellfish Immunology,* 22, 547-555. doi:10.1016/j.fsi.2006.08.001.

1267 Whyte, S.K., Chappell, L.H. and Secombes, C.J. (1988) In vitro transformation of
1268   *Diplostomum spathaceum* (Digenea) cercariae and short-term maintenance of post-
1269   penetration larvae in vitro. *Journal of Helminthology,* 62, 293-302.
1270   doi:10.1017/s0022149x0001169x.

1271 Whyte, S.K., Chappell, L.H. and Secombes, C.J. (1989) Cytotoxic reactions of rainbow trout,
1272   *Salmo gairdneri* Richardson, macrophages for larvae of the eye fluke *Diplostomum*
1273   *spathaceum* (Digenea). *Journal of Fish Biology,* 35, 333-345. doi:10.1111/j.1095-
1274   8649.1989.tb02986.x.

1275 Whyte, S.K., Chappell, L.H. and Secombes, C.J. (1990) Protection of rainbow trout,
1276   *Oncorhynchus mykiss* (Richardson), against *Diplostomum spathaceum* (Digenea) - the
1277   role of specific antibody and activated macrophages. *Journal of Fish Diseases,* 13,
1278   281-291. doi:10.1111/j.1365-2761.1990.tb00784.x.

1279 Whyte, S.K., Secombes, C.J. and Chappell, L.H. (1991) Studies on the infectivity of
1280   *Diplostomum spathaceum* in rainbow trout (*Oncorhynchus mykiss*). *Journal of*
1281   *Helminthology,* 65, 169-178. doi:10.1017/s0022149x0001066x.

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