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ScienceDirect
Aquaculture 275 (2008) 34–41
Aquaculture
www.elsevier.com/locate/aqua-online

Transmission dynamics of *Discocotyle sagittata* (Monogenea) in farmed rainbow trout interpreted from parasite population age structure

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Received 19 August 2007; received in revised form 23 November 2007; accepted 19 December 2007

Abstract

Water temperature in the Isle of Man, Great Britain, is generally below 10 °C for half the year, from November to April. During 3 consecutive years, samples of rainbow trout, *Oncorhynchus mykiss* were taken at 2 fish farms in May (following 6 months <10 °C) and in November (after a semester >10 °C), to study the populations of the gill fluke *Discocotyle sagittata.* Four distinct types of parasite population structure were found, 2 in May, 2 in November. In the first May scenario, the majority of parasites found were adults, and almost no developing worms occurred: this indicates that no major transmission takes place during the cold season. In the second May scenario, large numbers of freshly-invaded larvae appeared alongside the established mature worms, indicating that intensive transmission can take place when permissive temperatures allow the mass hatching of eggs laid in winter/spring. The first pattern shown by sampling in November was characterised by the co-occurrence of all parasite developmental stages reflecting continuous transmission over several months. A second pattern of infection evident from November samples may indicate that despite recent, intense transmission, some hosts carrying relatively low burdens of adult parasites experienced little or no successful recruitment during preceding periods favourable for transmission. This may provide evidence of differences in susceptibility between hosts. Overall, the 4 contrasting patterns document the effects of temperature as a major factor shaping the population age structure of *D. sagittata*.

Keywords: Monogenea; *Discocotyle sagittata*; Trout; *Oncorhynchus mykiss*; Transmission

1. Introduction

Knowledge of annual variation in disease patterns can inform methods and policies for disease prevention and control (Altizer et al., 2006); this is particularly relevant to aquaculture, because several pathogens of farmed fishes exhibit seasonality: examples include gyrodactylid monogeneans infecting Atlantic salmon *Salmo salar* (Appleby and Mo, 1997) and brown trout *Salmo trutta* (Mo, 1997), the crustacean *Argulus coregoni* parasitizing rainbow trout *Oncorhynchus mykiss* (Hakalahti et al., 2004), and cryptosporidians infecting Spanish gilthead sea bream *Sparus aurata* and European sea bass *Dicentrarchus labrax* (Sitjà-Bobadilla et al., 2005).

Discocotyle sagittata is a gill-fluke infecting salmonids, which has caused lethal epizootics in fish farms located in the Isle of Man (IoM), Great Britain (Gannicott, 1997; Rubio-Godoy and Tinsley, 2008). Being a temperate island, the IoM has contrasting winter and summer environmental conditions which could have a powerful effect on the transmission of *D. sagittata*, a parasite whose reproductive biology is known to be regulated by temperature (Tinsley, 2004). Experimental studies on single worm burdens of *D. sagittata* on rainbow trout showed that egg production was strongly temperature-dependent (Gannicott and Tinsley, 1998): output increased from 1.5 eggs/day at 5 °C to 7 eggs/day at 13 °C and to 12 eggs/day at 20 °C. Since these temperatures approximate to winter, spring/autumn and mid-summer conditions in the IoM, this study was

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doi:10.1016/j.aquaculture.2007.12.023

Table 1
Discocotyle sagittata mean abundance and range in different rainbow trout (*Oncorhynchus mykiss*) year classes

Fish	*n*	Total mean abundance			*Discocotyle sagittata* developmental category								
		All developmental categories			Juvenile			Intermediate			Mature		
		Prevalence	Worms/ host±SE	Range	Worms/ host±SE	Range	% of total (n)	Worms/ host±SE	Range	% of total (n)	Worms/ host±SE	Range	% of total (n)
1+ May	120	70%	3.7±0.52	0–33	0.03±0.01	0–1	0.7 (3)	0.9±0.19	0–12	25.6 (113)	2.7±0.46	0–33	73.7 (325)
2+ May*	70	98%	28.0±2.99	0–160	0.11±0.04	0–1	0.4 (8)	0.6±0.11	0–3	2.0 (40)	27.3±2.96	3–158	97.6 (1912)
Mass-infected 2+	22	100%	159±13.95	60–255	95.1±8.55	36–160	57.4 (2092)	0.3±0.16	0–3	0.1 (5)	70.4±11.84	2–214	42.5 (1548)
0+ November	120	52%	1.1±0.13	0–6	0.15±0.04	0–2	13.3 (18)	0.2±0.05	0–3	20.7 (28)	0.7±0.10	0–5	65.9 (89)
1+ November	109	100%	53.8±6.80	4–489	5.3±0.82	0–50	9.8 (575)	12.8±1.70	0–124	23.8 (1396)	35.8±4.80	1–359	66.4 (3898)
2+ November	60	100%	121.7±14.57	7–506	57.1±7.34	0–318	44.3 (3427)	24.6±3.15	0–105	19.1 (1477)	47.3±5.08	3–175	36.6 (2835)

Abundances are shown for the total parasite population, and for juvenile (1 to 2 pairs of clamps), intermediate (2.5 to 3.5 pairs of clamps) and mature (4 pairs of clamps and adults) parasites. Percentages indicate the contribution of the different developmental categories to the total worm population. Data pooled from 1999–2001. Notes: 0+, 1+ and 3+ refer to fish year (age) classes. *These statistics do not include mass-infected 2+ fish from farm 1 in May 2000, which are presented as a separate case.

designed to assess the influence of the natural fluctuations of temperature on parasite transmission in fish farms, and on parasite population age structure.

D. sagittata is distinctive in that developmental stages in the formation of the opisthaptor (the addition of clamps) provide a guide to parasite age: upon emergence from the egg, oncomiracidia possess a functional pair of sclerotised clamps on their haptors (Owen, 1970), and attached worms subsequently develop additional pairs of clamps (p.c.) until they have grown a total of 4 p.c. present on adults; sexual maturity is marked by the production of eggs visible in the uterus. This enables a detailed analysis of parasite population age structure, which in this study provided a "snapshot" of the sequence of invasions contributing to the parasite burdens of infected fish in the preceding months. At the same time, the record of age cohorts provides a guide to the growth and development of these burdens (and hence their pathogenic effects) over the following months.

2. Materials and methods

2.1. Fish

Rainbow trout (*O. mykiss*) samples were obtained on the Isle of Man (IoM), UK, from 2 fish farms (farm 1 in the South-West, farm 2 in the North-East) that have experienced mortality due to *D. sagittata*-induced anaemia (Gannicott,

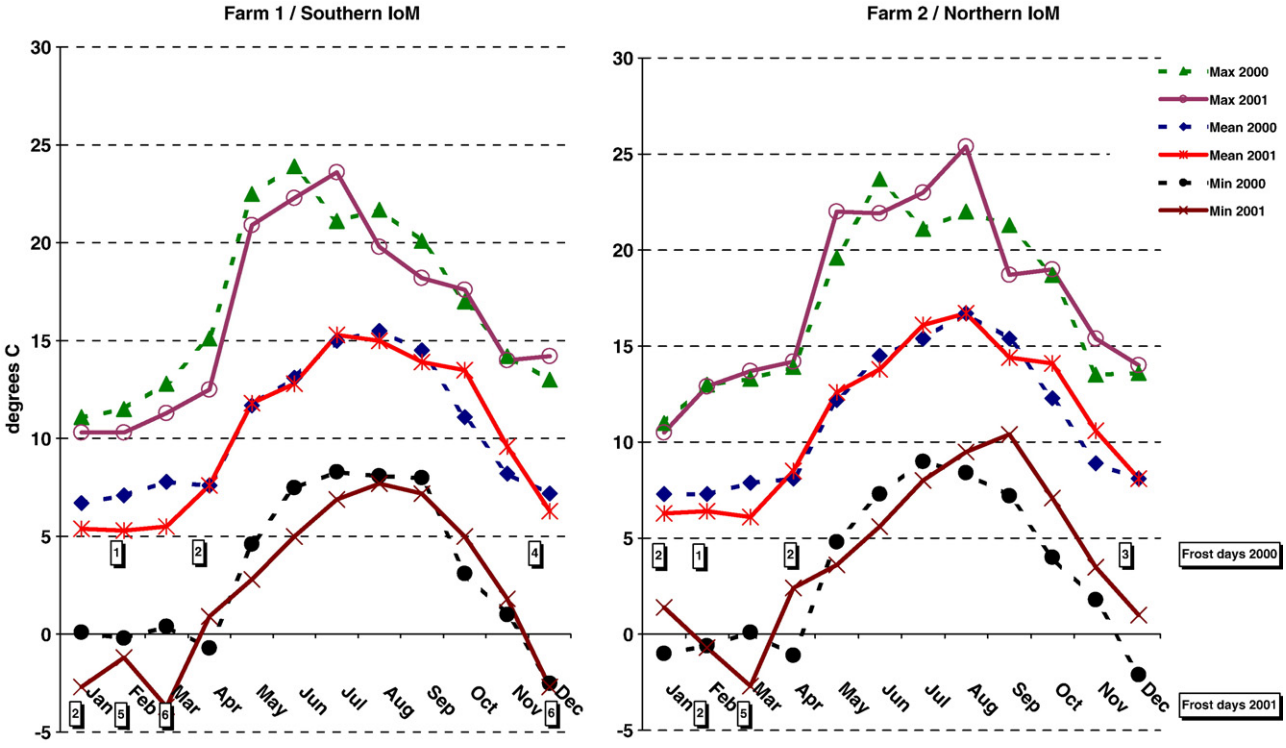

Fig. 1. Air temperature (monthly mean, minima and maxima) in the Isle of Man during the years 2000 and 2001. Southern data from Ronaldsway Airport, Ballasalla; Northern data from Point of Ayre. Weather data kindly provided by and reproduced with permission of the Meteorological Office, Isle of Man Government.

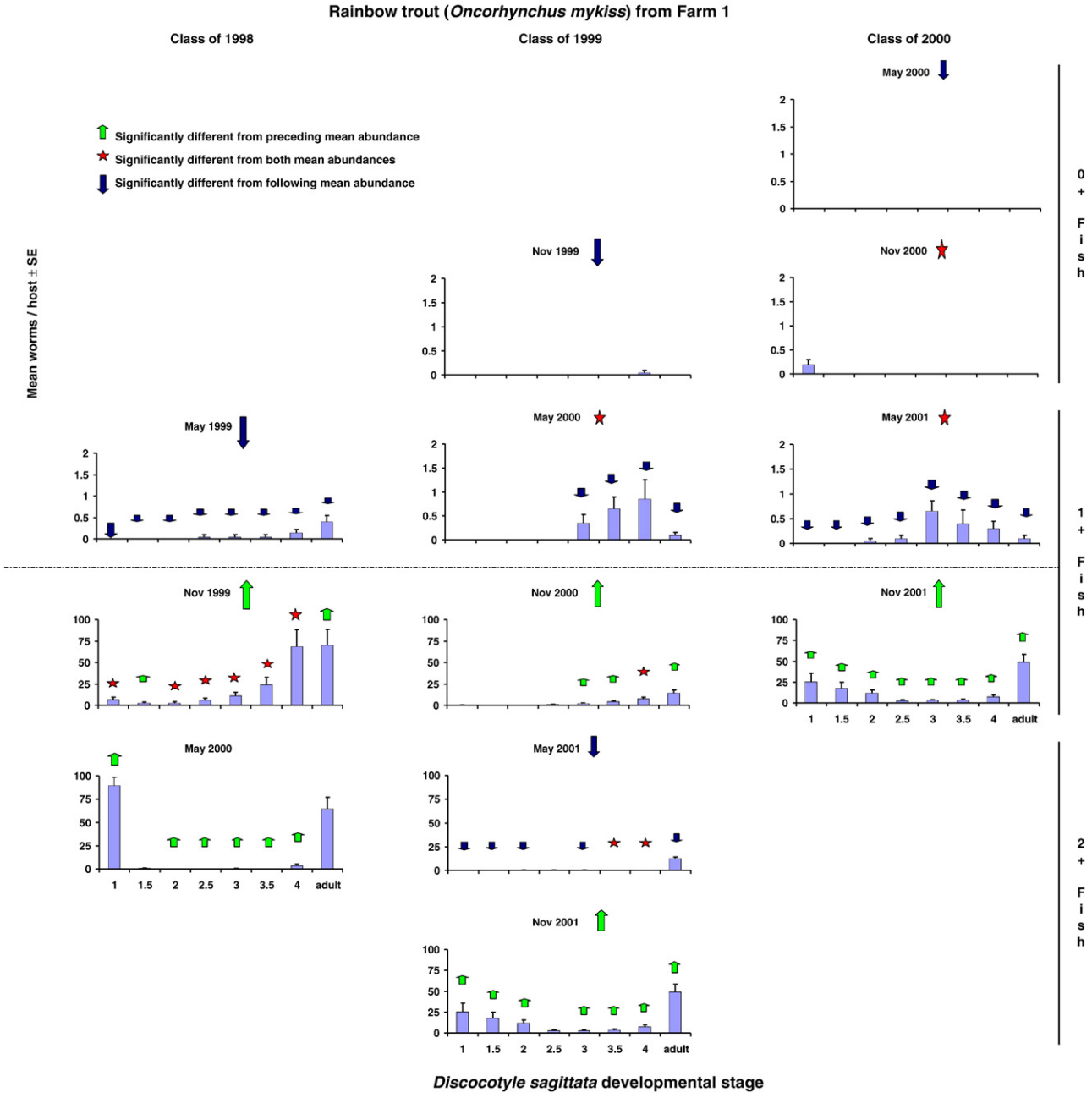

Fig. 2. Mean *Discocotyle sagittata* abundance±SE for individual developmental cohorts from different rainbow trout (*Oncorhynchus mykiss*) year classes in farm 1. Arrow/stars above the bars indicate statistically-significant differences when contrasted with the same developmental cohort in previous/subsequent samplings; arrows next to the sample date refer to statistics based on the overall mean parasite abundance. Horizontal dotted line indicates a significant increase in the abundance axis (*Y* axis) of the graphs.

1997; Rubio-Godoy and Tinsley, 2008). Most samples had $n=20$, except: 0+ fish in May, where $n=30$; class of 1999 fish from farm 1, where November 1999 samples had $n=10$ and May 2000 had $n=22$; class of 2000 fish from farm 1, where November 2001 $n=19$; and class of 1998 fish from farm 2, where November 2000 $n=10$. Farms were visited in early summer (mid-late May) and late autumn (late November) in 1999, 2000 and 2001, during general fish health inspections carried out for the Department of Agriculture, Fisheries and Forestry (IoM Government). The May and November sampling dates were designed to give maximum information on the annual transmission of *D. sagittata*, and considered unpublished data obtained in the same farms during studies with higher sampling frequency (Gannicott, 1997). Trout were anaesthetised terminally with MS222 and dissected for general bacteriological/parasitological tests. Fish-of-the-year (0+), one-year-old (1+) and two-year-old (2+) trout were measured (fork length) and weighed, and their gill arches were removed and preserved in 10% formalin for later microscopic analysis. Data obtained from 10 class of 1997 2+ fish in May 1999 and from 40 class of 2001 0+ fish in November 2001 are not described in detail, but are included in the pooled results presented in Table 1.

2.2. Study sites

Farm 1 utilizes mud-bottomed ponds, farm 2 concrete-lined tanks. They receive water from unconnected river systems. Water circulates from tanks with younger fish to those containing older animals. Fish-of-the-year (0+ fish) were kept in raceways, which receive river water directly, from hatching in winter until mid- to late summer, when they were transferred downstream to ponds

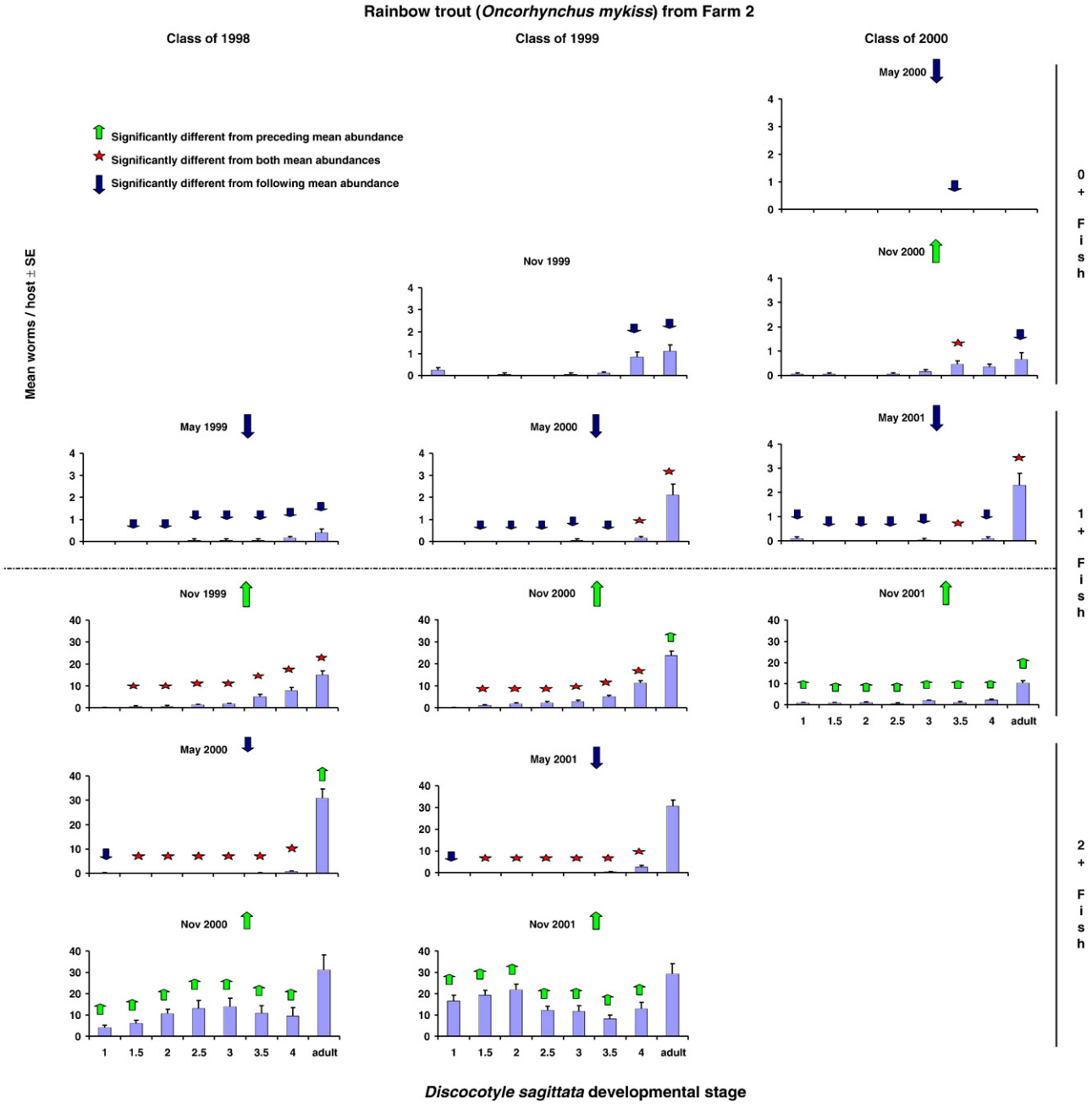

Fig. 3. Mean *Discocotyle sagittata* abundance ± SE for individual developmental cohorts from different rainbow trout (*Oncorhynchus mykiss*) year classes in farm 2. Arrow/stars above the bars indicate statistically-significant differences when contrasted with the same developmental cohort in previous/subsequent samplings; arrows next to the sample date refer to statistics based on the overall mean parasite abundance. Horizontal dotted line indicates a significant increase in the abundance axis (*Y* axis) of the graphs.

previously occupied by 1+ fish, which were in turn moved to ponds previously containing 2+ fish, etc. No fallow period took place between the removal of old fish and the transfer of younger animals. In both farms, fish year classes were kept separate throughout the study. However, during transfer of fish, individuals from the same age class kept in different ponds were mixed. Neither farm applied specific treatments against *D. sagittata* for the duration of this study.

2.3. Parasites

Individual preserved gill arches were examined under the dissecting microscope. The number of parasites per gill arch (right and left) was recorded, and their developmental stages were determined based on the number of pairs of clamps (p.c.) present on the haptor: values ranged from 1 for freshly-hatched worms with 1 p.c. to 4.5 for sexually mature parasites with 4 p.c. and eggs *in utero* (Gannicott, 1997; Rubio-Godoy and Tinsley, 2002). Parasites were assigned to 3 developmental categories: juvenile worms had 1 to 2 p.c., intermediate stages 2.5 to 3.5 p.c., and mature worms included parasites with 4 p.c. and egg-laying adults.

2.4. Weather information

Farm water temperature records were incomplete, so air temperature was used as a proxy. Air temperatures (monthly means, maxima and minima) were determined in the south of the IoM at Ronaldsway Airport, Ballasalla, and in the north at Point of Ayre, and provided by the Meteorological Office, IoM Government.

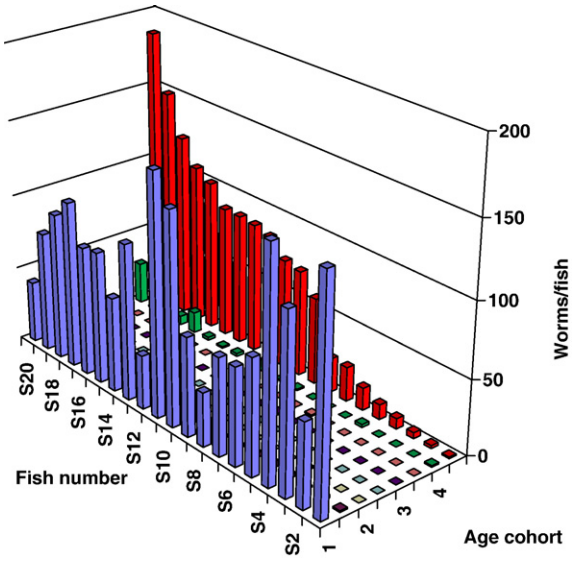

Fig. 4. Developmental stage distribution of *Discocotyle sagittata* on individual 2+ rainbow trout (*Oncorhynchus mykiss*) sampled from Farm 1 in May 2000. Individual fish received a sequential number based on the number of adult worms recovered.

2.5. Statistical analysis

Data were analysed with SPSS 10.0 for Macintosh. Parasite burdens reported represent the total mean worms/host ± SE recorded, since no significant difference was found between infection levels in the right and left gill arches (data not shown). Parasitological terms used follow the recommendations of Bush et al. (1997). Raw data were $\log_{10}(1+x)$ transformed prior to comparisons with ANOVA (General Linear Models) and significant differences between groups were detected with Tukey's honestly significant difference (HSD) test; the significance level was set at $P<0.05$. Pearson's correlations were used to analyse the relationship between fish length and worms/host, and between numbers of parasites in different developmental categories.

3. Results

IoM mean air temperatures are generally above 10 °C for half the year, from May to October (Fig. 1). On average, the south IoM is colder than the north, as shown by monthly mean air temperatures determined at Ballasalla and the Point of Ayre, respectively. However, in winter 1999–2000, weather conditions in the south were milder than in the north. Air temperatures are elevated above water temperatures in spring but water temperatures remain higher for a longer period in autumn: therefore, early summer (May) and late autumn (November) sampling roughly corresponded to time points when water temperature reached 10 °C on, respectively, an up- or downward trend.

Burdens of *D. sagittata*, categorised into juvenile, intermediate and mature stages, were recorded for the different age classes of rainbow trout in early summer and late autumn; Table 1 shows data pooled across the 3 years 1999–2001, which illustrate some general trends: In May, 74–98% of parasites recovered were mature, and typically, in most samples, no or few juveniles were found (but see below). In November, 0+ and 1+ trout harboured worm populations containing ca. 66% mature individuals; 2+ fish carried 44% juvenile and 37% mature parasites. Overall, in May samples, a significant negative correlation ($R^2=0.058$; $P=0.008$) was found in 1+ fish between size (length) and worms/host; no significant correlation was found for 2+ fish. In November, significant positive correlations of fish size/worm burden were found for 0+ fish ($R^2=0.187$; $P<0.0001$) and 1+ fish ($R^2=0.098$; $P=0.0009$); no significant correlation was detected in 2+ fish.

Figs. 2 and 3 show the mean abundance for individual *D. sagittata* developmental cohorts, determined for all fish sampled. From these, two distinct types of parasite population age structure were found in May samples. The first and most commonly recorded is exemplified by 2+ *O. mykiss* collected in farm 2 in May 2000 (Fig. 3), which primarily contained mature worms. In this case, all fish carried adult parasites, with a mean abundance ± SE of 30.9 ± 3.65 worms/host. About half the fish harboured 1 or 2 worms with 4 p.c. (mean ± SE 0.8 ± 0.19 worms/host) and 3 out of 20 carried 1 worm with 3.5 p.c. (mean ± SE 0.2 ± 0.82 worms/host). Two out of 20 fish had each 1 worm with 1 p.c. No parasites with 1.5 to 3 p.c. were found. The second type of parasite population age structure found in May is illustrated by the sample of 22 2+ fish collected in farm 1 in May 2000 (Table 1, Fig. 4). This sample shows the same distinct pattern described previously, with hosts carrying different numbers of adult worms and very little transmission represented in the middle part of the range of developmental stages: overall, there is 1 worm with 2 p.c., 0 with 2.5 p.c., 4 with 3 p.c., 3 with 3.5 p.c., 74 with 4 p.c., and 1474 adult worms. Alongside these older burdens, very recent invasions (worms with 1 p.c.) affect 100% of the sample with 36–160 (mean ± SE 93.6 ± 8.49) worms/host. In addition, 14 fish carried 1–6 (mean ± SE 1.5 ± 0.35) worms/host at the 1.5 p.c. stage, representing slightly earlier transmission in the weeks preceding sampling. No significant correlation was found between the numbers of mature and juvenile parasites present on individual fish; for instance, fish S1 concurrently harboured 153 juvenile and 2 mature worms, fish S15 had 88 juvenile and 90 mature worms, and fish S22, 41 juvenile and 214 mature worms.

In both farms, infection levels increased significantly over the warmer part of the year, between May and November. The rate of parasite acquisition varied considerably, annually and between fish cohorts, as illustrated by the frequency distribution of worm developmental stages: for instance, in all November 1+ fish in farm 2 (Fig. 3), the frequency distribution of parasite developmental stages was skewed towards adult worms; occasionally, frequency distributions were bimodal, as found in November 2001 in farm 1, where 1+ class of 2000 and 2+ class of 1999 fish had virtually identical parasite developmental stage frequency

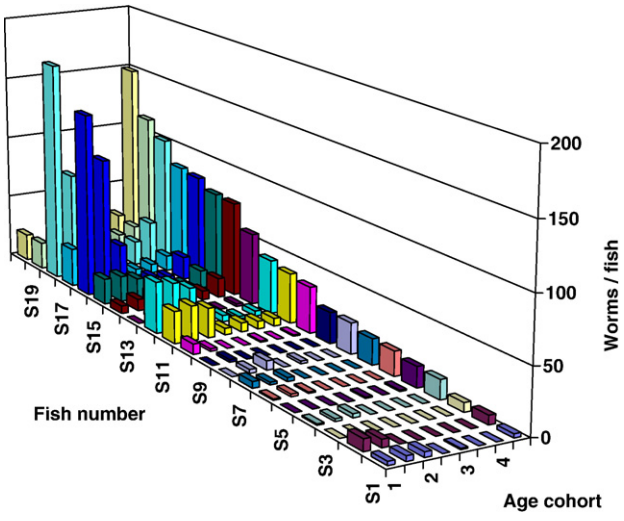

Fig. 5. Developmental stage distribution of *Discocotyle sagittata* on individual 2+ rainbow trout (*Oncorhynchus mykiss*) sampled from Farm 1 in November 2001. Individual fish received a sequential number based on the number of adult worms recovered.

distributions (Fig. 2); finally, all developmental stages were found in 2+ fish sampled in November in farm 2 (Fig. 3).

Two contrasting patterns of parasite age structure were identified in November samples, following parasite acquisition over summer/early autumn. The first is exemplified by 2+ fish collected from farm 2 in November 2001 (Fig. 3), where all developmental stages are represented, from freshly-invaded to adult worms. Parasite abundance varied from host to host. For example, the fish with the lowest intensity (37 worms/host), carried 5 worms with 1 p.c.; 7 with 1.5 p.c.; 12 with 2 p.c.; 5 with 2.5 p.c.; 2 with 3 p.c.; 1 with 3.5 p.c.; 1 with 4 p.c.; and 4 adult worms. The fish with the highest intensity (311 worms/host), had the following burdens in each of these developmental categories from 1 p.c. to adults: 38; 32; 40; 31; 44; 22; 39; and 65. This represents a relatively consistent rate of invasion and establishment continuing throughout the preceding warmer months. The second type of parasite population age structure found in November is exemplified by 2+ trout collected at farm 1 in November 2001 (Fig. 5). In this case, half the sample indicates the occurrence of continuous invasion over the months preceding inspection. For instance, fish S11 had a total of 138 worms (with the following intensities in the 1 p.c. to adults range: 25; 27; 23; 5; 7; 6; 6; 39), and fish S19 had 211 worms (burdens in the same developmental categories recorded above: 21; 19; 6; 5; 6; 18; 23; 113). However, despite the evidence of recent, intense transmission within the entire population (e.g., fish S16 had 145 worms with 1 p.c.; fish S18, 174 1 p.c. worms), it is remarkable that there are some individual hosts showing evidence of little or no successful recruitment during appreciable periods of time. Thus, in contrast to fish S11 and S19, which respectively accumulated 19 and 47 worms over the period represented by 3/3.5/4 p.c., fish S1 had acquired only 1 worm with 3 p.c., and fish S2 had acquired none corresponding to these age classes; only 7 parasites in the developmental cohorts from 2.5 to 4 p.c. were found on the S1–S10 hosts. Overall, fish S11–S20 had about 220 worms/host (mean abundance ± SE of 219.9 ± 41.99 worms/host; range 74–506 worms/host), while fish S1–S10 harboured ca. 25 worms/host (24.6 ± 3.61 worms/host; range 7–48 worms/host). A significant positive correlation was found between the numbers of mature and juvenile parasites recovered from individual hosts ($R^2 = 0.28$, $P = 0.017$); the correlation between mature and [juvenile + intermediate] parasites was also significant ($R^2 = 0.32$, $P = 0.009$).

During the colder season (between November and the following May), in most cases, mean parasite abundance did not change significantly — although limited but significant increases took place (see below). In all instances in farm 2, the abundance of adult parasites calculated in May roughly corresponds to the summation of all developmental stages recorded the previous November (Fig. 3). The case of class of 1998 fish in farm 1 (Fig. 2) is interesting, because although the overall mean parasite abundance was not significantly different before and after winter 1999–2000, major worm losses occurred in this period: in November 1999, mean abundances (±SE) of individual developmental stages in the range 1 p.c to adults were as follows: 6.9 (±2.92); 2.6 (±1.27); 3.0 (±1.45); 6.4 (±2.31); 11.2 (±3.99); 24.4 (±7.95); 68.8 (±19.41) and 70.5 (±18.30). In May 2000, mean abundances in the same developmental categories were: 93.6 (±8.49); 1.5 (±0.35); 0.05 (±0.05); 0; 0.2 (±0.14); 0.1 (±0.07); 3.4 (±1.36) and 67.0 (±10.95). Omitting the freshly-invaded worms with 1 p.c. which accounted for over 50% of the parasite population recorded in May, mean parasite abundances differed significantly between samples ($P = 0.004$).

Limited but significant increases in mean parasite abundance occurred over the colder part of the year in 0+ fish in farm 1 (Fig. 2). Both year classes of 1999 and 2000 had almost negligible levels of infection in November: mean abundance was 0.05 ± 0.05 4 p.c. worms/host and 0.2 ± 0.09 1 p.c. worms/host, respectively. Burdens had increased significantly by the following May in both fish cohorts (Fig. 2): class of 1999 fish had a mean parasite abundance of 1.9 ± 0.56 worms/host and class of 2000 fish had 1.6 ± 0.53 worms/host.

4. Discussion

This study demonstrates that transmission of *D. sagittata* in farmed trout is seasonal, with invasion maximised during the warmer part of the year and almost negligible infection over winter. Temperature accounts for this seasonality, as it influences the rate of reproductive processes in *D. sagittata*: at water temperatures <10 °C, *per capita* egg production, development and viability are reduced to very low levels (Gannicott and Tinsley, 1998; Tinsley, 2004). Worm development is also temperature-dependent and inhibited at low temperatures (Gannicott, 1997). Air temperatures in the IoM rise >10 °C in April/May and do not drop below this value until October/November. Considering the different caloric capacity of air and water, it is reasonable to propose that water temperatures lag behind air temperatures and will be >10 °C between May and November. Thus, although in the present study inspections took place only twice a year, the timing of sampling in May and November was designed to coincide with the estimated onset and end of the *D. sagittata* transmission season in the IoM, based on annual average water temperatures which cross the 10 °C threshold in these months. Therefore, the May and November samples give comprehensive data on, respectively: a) the populations of parasites that had persisted throughout the winter when transmission is negligible (i.e., the survival of parasites invading during the previous summer–autumn); and b) the outcome of the transmission season extending from early summer to late autumn (i.e., the combined total of successive invasions accumulating before the cessation of transmission over winter). Unpublished field studies in the same farms with higher sampling frequency than the bi-annual sampling schedule of the present report support this notion (Gannicott, 1997).

Most samples collected in May illustrate the state of *D. sagittata* populations after hosts have spent a semester <10 °C, and confirm the rarity of transmission during winter/spring, reflected in the limited number of worms with 1.5 to 3 p.c. The presence of adult worms in early summer confirms that *D. sagittata* survives from year to year, as reported for wild fish (Chubb, 1977; Valtonen et al., 1990). The few worms recorded with 3.5 and 4 p.c. probably invaded late in the previous season and did not reach sexual maturity over winter–spring. Maturation of worms over the cold season is a possibility, as evidenced by the finding in farm 2 that the abundance of adult parasites in May roughly corresponded to the arithmetical addition of all developmental stages recorded the previous November. This data set also shows the very start of the summer invasion, with the presence of some 1 p.c. worms; e.g., in May 2000, 2 out of 20 2+ fish in farm 2 had one 1 p.c. worm each.

The timing of one of the May samples was fortuitously at the onset of massive transmission once permissive temperatures had been reached. This served to show that hatching of eggs

accumulated on the pond bottoms during spring may produce a mass invasion episode in which 100% prevalence of infection was reached, with a mean of 95.1 juvenile worms/host and a maximum of 160 worms/host. This invasion had taken place 1–2 weeks prior to sampling, as established by the fact that all parasites had 1 p.c. (Gannicott, 1997). These findings confirm similar results from a previous account demonstrating the occurrence under farming conditions of a single pulse of infection (Rubio-Godoy and Tinsley, 2002). The other May samples did not show this mass summer invasion, perhaps because of lower temperatures and/or later hatching of infective stages. An indication that this event may be influenced by local temperature regimes is provided by the May 2000 samples taken at the 2 farms. Even though these samples were taken in consecutive days (23 and 24 May 2000) and water temperatures were similar at the time of inspection (farm 1, 12 °C; farm 2, 11 °C), mass hatching had occurred in the preceding week at farm 1 but not at farm 2. The effects of local temperature patterns in controlling these detailed differences are shown by comparison of temperature records for the 2 farms in this year. Even though the south IoM is on average colder than the north, in winter 1999–2000, weather conditions in the south were milder than in the north and may have favoured the earlier massive hatching of parasite eggs laid during spring 2000. Between January and March, farm 1 (south) experienced air temperature minima close to or above freezing and 1 frost day, and an April minimum of −0.7 °C was recorded; in contrast, farm 2 (north) presented minima ≤0 °C and 3 frost days during the same period, and an April minimum of −1.1 °C. Winter 2000–2001 was colder, with both farms experiencing air temperature minima below freezing and several frost days; this may explain why no mass infection was recorded by the date of sampling in May 2001. Nonetheless, it is reasonable to suggest that massive hatching and infection are features of the *D. sagittata* life cycle. The timing of massive invasion would be advantageous for the parasite, considering that in early summer the fish population contains new, naïve hosts. Moreover, experimental data suggest that fish develop no partial immunity against *D. sagittata* following mass infection, as opposed to gradual infection (Rubio-Godoy and Tinsley, 2004).

As illustrated by the presence of all parasite developmental stages in November, permissive temperatures (>10 °C) during summer and early autumn result in more or less continuous transmission of *D. sagittata*; this confirms the occurrence of trickle infection in farmed trout (Rubio-Godoy and Tinsley, 2002). Parasite burdens found in the present study were heterogeneous among hosts, both in terms of absolute parasite intensity (ranging from 4 to 489 worms/host in 1+ fish, and from 5 to 506 worms/host in 2+ fish) and in the relative abundance of individual parasite developmental stages. This arguably results from stochastic infection events confounded with differences in host susceptibility. It is probable that 2 generations of parasites can appear in a single transmission season, considering that from May onwards, temperatures remain >13 °C for 4 months. Permissive temperatures during this period would enable worms successfully invading in May to reach maturity within 3 months and start producing eggs, which would exhibit high viability and developmental rates and hatch within a month (Gannicott, 1997; Gannicott and Tinsley, 1998). Thus, some of the recently-recruited (1–2 p.c.) worms recorded in November may represent the 2nd parasite generation of that year. In contrast, *D. sagittata* found on wild whitefish (*Coregonus acronius*) in an arctic lake, where the transmission season is shorter, have only 1 generation per year (Valtonen et al., 1990). However, global warming may shorten winter periods and increase the growth potential of parasites and lead to disease outbreaks in both farmed and wild host populations, as proposed for the fish pathogens *A. coregoni* and *Diplostomum spathaceum* (Hakalahti et al., 2006). Extensions of the seasonal window for transmission attributable to climate change have indeed been documented in the Canadian Arctic in nematode infections of muskoxen (Kutz et al., 2005) and sheep (Jenkins et al., 2006).

In November 2001, class of 1999 trout from farm 1 carried quite distinct *D. sagittata* burdens: while a proportion of the fish population had ca. 220 worms/host, another distinct group had ca. 25 worms/host. Farm management practices can result in hosts with different infection levels, when lots of fish with different exposure histories are mixed up; we do not know whether these samples had been thus mixed. However, even if the differences in adult parasite burdens illustrated by these 2+ trout in November 2001 resulted from mixing up of fish lots in the previous months, it is noteworthy that "low-burden" fish had few parasites reflecting recent recruitment, because there was evidence of substantial recent invasions occurring in the pond where they were maintained. Moreover, this dichotomy in infection levels within host populations was noticed several times in November samples in both farms (data not shown). In these cases, parasite age structure in the hosts with low burdens was reminiscent of that assessed early in the transmission season: the majority of worms were adults, and only limited numbers of freshly-attached and developing worms were found. Whilst these individual records give only an indication of possible factors operating, it may be significant that these hosts, demonstrating little parasite survival from a period of otherwise intense transmission in their population, also carry the smallest burdens of adult parasites. This might be a reflection of individual differences in the ability to develop immunity to the parasite, as rainbow trout have been demonstrated to be able to mount immune responses against *D. sagittata* following experimental infection (Rubio-Godoy and Tinsley, 2004) and vaccination with a crude worm extract (Rubio-Godoy et al., 2003).

Significant positive associations were found in November between host length and worm intensities in 0+ and 1+ trout. The abundance of gill-infecting monogeneans has been correlated to host size, where an increase in body length is accompanied by increase in gill surface (Rohde et al., 1995; Barse, 1998). However, 2+ trout in the present report exhibited no significant host size/parasite burden correlation. Further investigations would be needed to assess whether a host age–*D. sagittata* intensity relationship exists, where acquired immunity is assumed to develop as a function of cumulative exposure to parasites, as reported for other pathogens (Cattadori et al., 2005).

We expect that the *D. sagittata* infection scenarios reported here illustrate basic characteristics of the host–parasite interaction and would correspond with infection regimes affecting trout farms at different times of the year: transmission is seasonal, with significant recruitment over summer–autumn and negligible invasion over winter–spring. Although *D. sagittata* infection builds up from year to year in affected farms and can attain lethal levels (Rubio-Godoy and Tinsley, 2008), the fact that transmission is seasonal allows the introduction of measures to decrease parasite recruitment. Thus, if fish can be moved in early summer from the pond where they overwintered, they may avoid exposure to the massive hatching of accumulated parasite eggs. Further reductions in recruitment could be achieved by transferring hosts to clean ponds in mid-summer, and by fallowing previously occupied ponds so that parasite eggs already deposited will hatch without a resident host population to infect. Apart from the empirical evidence supporting possible control strategies for *D. sagittata*, this study provides an example of seasonal parasite transmission mainly driven by one abiotic factor, temperature.

Acknowledgements

We are grateful to several people: John Ballard and Ken Jervis (Isle of Man) for access to infected fish; Roger Sweeting (Freshwater Biological Association) and the late David McGregor (CEFAS Weymouth) for sharing their fish samples with us; Les and Pam Kneale (Isle of Man) for their support during fieldwork. MRG was supported by postgraduate scholarships from the Mexican National Council for Science and Technology (CONACyT) and Universities UK, and recipient of a post-doctoral fellowship from Fondation Murana, Switzerland.

References

Altizer, S., Dobson, A., Hosseini, P., Hudson, P., Pascual, M., Rohani, P., 2006. Seasonality and the dynamics of infectious diseases. Ecol. Lett. 9, 467–484.

Appleby, C., Mo, T.A., 1997. Population dynamics of *Gyrodactylus salaris* (Monogenea) infecting Atlantic salmon, *Salmo salar*, parr in the river Batnfjordselva, Norway. J. Parasitol. 83, 23–30.

Barse, A.M., 1998. Gill parasites of mummichogs, *Fundulus heteroclitus* (Teleostei: Cyprinodontidae): effects of season, locality, and host sex and size. J. Parasitol. 84, 236–244.

Bush, A.O., Lafferty, K.D., Lotz, J.M., Shostak, A.W., 1997. Parasitology meets ecology on its own terms: Margolis et al revisited. J. Parasitol. 83, 575–583.

Cattadori, I.M., Boag, B., Bjornstad, O.N., Cornell, S.J., Hudson, P.J., 2005. Peak shift and epidemiology in a seasonal host–nematode system. Proc. R. Soc. Lond. Ser. B-Biol. Sci. 272, 1163–1169.

Chubb, J.C., 1977. Seasonal occurrence of helminths in freshwater fishes. Part 1. Monogenea. Adv. Parasitol. 15, 133–199.

Gannicott, A.M., 1997. The biology of *Discocotyle sagittata* (Monogenea) infecting trout. University of Bristol, Bristol, p. 312.

Gannicott, A.M., Tinsley, R.C., 1998. Environmental effects on transmission of *Discocotyle sagittata* (Monogenea): egg production and development. Parasitology 117, 499–504.

Hakalahti, T., Pasternak, A.F., Valtonen, E.T., 2004. Seasonal dynamics of egg laying and egg-laying strategy of the ectoparasite *Argulus coregoni* (Crustacea: Branchiura). Parasitology 128, 655–660.

Hakalahti, T., Karvonen, A., Valtonen, E.T., 2006. Climate warming and disease risks in temperate regions — *Argulus coregoni* and *Diplostomum spathaceum* as case studies. J. Helminthol. 80, 93–98.

Jenkins, E.J., Veitch, A.M., Kutz, S.J., Hoberg, E.P., Polley, L., 2006. Climate change and the epidemiology of protostrongylid nematodes in northern ecosystems: *Parelaphostrongylus adocoilei* and *Protostrongylus stilesi* in Dall's sheep (Ovis d. dalli). Parasitology 132, 387–401.

Kutz, S.J., Hoberg, E.P., Polley, L., Jenkins, E.J., 2005. Global warming is changing the dynamics of Arctic host-parasite systems. Proc. R. Soc. Lond. Ser. B-Biol. Sci. 272, 2571–2576.

Mo, T.A., 1997. Seasonal occurrence of *Gyrodactylus derjavini* (Monogenea) on brown trout, *Salmo trutta*, and Atlantic salmon, *S. salar*, in the Sandvikselva river, Norway. J. Parasitol. 83, 1025–1029.

Owen, I.L., 1970. The oncomiracidium of the monogenean *Discocotyle sagittata*. Parasitology 61, 279–292.

Rohde, K., Hayward, C., Heap, M., 1995. Aspects of the ecology of metazoan ectoparasites of marine fishes. Int. J. Parasitol. 25, 945–970.

Rubio-Godoy, M., Sigh, J., Buchmann, K., Tinsley, R.C., 2003. Immunization of rainbow trout *Oncorhynchus mykiss* against *Discocotyle sagittata* (Monogenea). Dis. Aquat. Org. 55, 23–30.

Rubio-Godoy, M., Tinsley, R.C., 2002. Trickle and single infection with *Discocotyle sagittata* (Monogenea: Polyopisthocotylea): effect of exposure mode on parasite abundance and development. Folia Parasitol 49, 269–278.

Rubio-Godoy, M., Tinsley, R.C., 2004. Immunity in rainbow trout, *Oncorhynchus mykiss*, against the monogenean *Discocotyle sagittata* following primary infection. Parasitol. Res. 92, 367–374.

Rubio-Godoy, M., Tinsley, R.C., 2008. Recruitment and effects of *Discocotyle sagittata* (Monogenea) infection on farmed trout. Aquaculture 274, 15–23.

Sitjà-Bobadilla, A., Padros, F., Aguilera, C., Alvarez-Pellitero, P., 2005. Epidemiology of *Cryptosporidium molnari* in Spanish gilthead sea bream (*Sparus aurata* L.) and European sea bass (*Dicentrarchus labrax* L.) cultures: from hatchery to market size. Appl. Environ. Microbiol. 71, 131–139.

Tinsley, R.C., 2004. Platyhelminth parasite reproduction: some general principles derived from monogeneans. Can. J. Zool. 82, 270–291.

Valtonen, E.T., Prost, M., Rahkonen, R., 1990. Seasonality of two gill monogeneans from two freshwater fish from an oligotrophic lake in northern east Finland. Int. J. Parasitol. 20, 101–107.