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Persistence of host and parasite populations subject to experimental size-selective removal

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Abstract Predators have the potential to limit the spread of pathogens not only by selecting infected prey but also by shaping prey demographics. We tested this idea with an epidemiological experiment in which we simulated variable levels of size-selective predation on zooplankton hosts and monitored the persistence of host and parasite populations. In the absence of simulated predation, the virulent protozoan *Caullerya mesnili* frequently drove its host *Daphnia galeata* to extinction. Uninfected control populations showed lower extinction rates and higher average densities than infected populations in the absence of simulated predation (all of the latter went extinct or remained infected). With a weak removal rate of the largest hosts, the proportion of populations in which the parasite drove the host to extinction decreased, while the number of populations in which the host persisted and the parasite went extinct increased. Host-parasite coexistence was also observed in some cases. With intermediate levels of removal, most of the parasite populations went extinct, while the host populations persisted. With an even higher removal rate, *Daphnia* were driven to extinction as well. Thus, variation in one factor, size-selective mortality, resulted in four different patterns of population dynamics. Our results highlight the potential role of predation in shaping the epidemiology and community structure of host-parasite systems.

Keywords *Caullerya mesnili* · *Daphnia galeata* · Experimental epidemiology · Extinction · Size-selective predation

Introduction

The role of predators (Holt 1984; Zaret 1980a) and parasites (Anderson and May 1986; Bonsall and Hassell 1997; Holt and Pickering 1985; Park 1948) in constraining or promoting coexistence of competing species has been established both theoretically and empirically. Most studies have concentrated on the effect of one antagonist on a pair of victims competing for similar resources, or on apparent competition mediated by the antagonist. Only recently has the importance of simultaneous interactions between two antagonists and their mutual target (prey for the predator, host for the parasite) in structuring natural communities been recognized in ecological and parasitological literature (Decaestecker et al. 2002; Hall et al. 2005a; Hudson and Greenman 1998; Ostfeld and Holt 2004; Packer et al. 2003).

Culling of infected animals from herds of domestic animals or populations of endangered animals has long been used as a means of removing the perceived source of infection and controlling the spread of an infectious disease among the animals in concern (Anderson et al. 1981; Barlow 1996). Mathematical models developed to calculate the level of culling necessary to reduce the pool of susceptible hosts below the threshold for pathogen establishment assume that culling is done randomly, irrespective of the infection status of the animals (Barlow 1996). Pathogens, however, are rarely distributed randomly in their host populations, but tend to aggregate in certain hosts which, depending on the host-parasite system, could be the youngest/smallest or the oldest/largest individuals (Hudson and Dobson 1995). Predators, as well, seldom select their prey randomly (Brooks and Dodson 1965; Zaret 1980b) and in many cases parasitized individuals have been shown to be more vulnerable

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to predation than individuals in the population in general (Hudson et al. 1992). Therefore, except for cases in which the predator is included in the life cycle of the pathogen, predators have the potential to constrain the occurrence and distribution of pathogens by disproportionately removing infected individuals from the host populations (Packer et al. 2003).

Recent studies have shown that microparasites are common in natural *Daphnia* populations (see review in Ebert 2005). A few empirical studies have suggested that the levels of parasitism might be lower in populations with fish predation than those without predation (Duffy et al. 2005; Ebert et al. 1997; Willey et al. 1993). In *Daphnia*, the largest individuals have the highest probability of being infected because of their high filtering rates (and thus uptake of infective stages of parasites) and accumulation of parasites with age (Mangin et al. 1995; Stirnadel and Ebert 1997). It has therefore been speculated that, in populations with high adult mortality, parasite prevalence might be reduced (Ebert et al. 1997). For zooplankton, including *Daphnia*, it has been shown that fish preferentially prey on larger individuals (Brooks and Dodson 1965). For example, in Lake Constance, whitefish and perch feed preferentially on large individuals of *D. galeata* and *D. hyalina*, even when juveniles are more abundant (Stich 1989). Furthermore, parasites might increase the risk of predation for planktonic organisms (Willey et al. 1990, 1993).

The aim of the current study was to test the effect of size-selective removal on parasite load and persistence in experimental *Daphnia* populations. In the absence of predation, the highly virulent parasite *Caullerya mesnili* frequently drives experimental populations of its host *D. galeata* to extinction (Bittner et al. 2002). Therefore, we predicted that without predation, or with a small removal rate, the experimental host population and the associated parasite population would be driven to extinction through parasite-induced mortality. Increased removal of the largest hosts carrying the highest load of parasite transmission stages would reduce the rate of parasite spread and might again promote coexistence of the two antagonists. However, an even higher removal rate of larger *Daphnia* would lead to parasite extinction because the production rate of parasite transmission stages would fall below the level needed to maintain the epidemic, i.e., the basic reproductive rate of the parasite, R_0 , would fall below 1 (Anderson and May 1981). Beyond a certain level of removal, we expected extinction of the host population again because host mortality would exceed natality. Variability in population size is an important factor predicting extinction risk, especially for small populations (Pimm et al. 1988). Parasites have been shown to decrease host population density (Anderson 1979) and increase variability (Ebert et al. 2000a). On the other hand, predation may decrease variability, by reducing synchrony in reproduction (Bengtsson and Milbrink 1995). Therefore, we expected *Daphnia* population variability to increase with a decrease in removal intensity of large infected hosts. Thus, we conducted an epidemiolog-

ical experiment with variable levels of host removal to test whether different levels of removal would result in different patterns of host and/or parasite persistence.

Materials and methods

The host-parasite system

We used the planktonic crustacean *Daphnia galeata* Sars 1863 (Crustacea: Cladocera) which is an obligatory host of the parasite *C. mesnili* (Chatton 1907). *D. galeata* is a freshwater species often found in large European bodies of water (Flössner 2000). It reproduces by cyclic parthenogenesis, and thus can be maintained in the laboratory in clonal cultures. The endoparasite *C. mesnili* infects the gut epithelium of *Daphnia*. *C. mesnili* is classified as a Haplosporidium, but its classification is controversial (Bittner et al. 2002). It is transmitted horizontally between hosts via infective stages released into the water by either living or dead hosts. These are then ingested by the next host (Bittner et al. 2002). The parasite forms large ($10 \times 8 \mu\text{m}$) egg-shaped structures that are clustered in groups of 8–20 in the gut epithelium. A cluster is up to $100 \mu\text{m}$ in size and is clearly visible in living hosts observed with a dissecting microscope. Under laboratory conditions, the clusters are first seen 6 days after exposure, and the number of clusters increases until host death, about 18 days post exposure (Bittner et al. 2002). *C. mesnili* grows exponentially in *D. galeata* and after a few reproductive events it castrates the host and increases host mortality.

The *Daphnia* clone used in the experiment originated from Lake Constance in Germany. It was collected in 1997 and has since been maintained in the laboratory. The *C. mesnili* strain used in the experiment was isolated from *Daphnia* collected from Lake Constance in 1998 and propagated in the laboratory in the *Daphnia* clone mentioned above.

We cultured monoclonal *D. galeata* populations in glass jars filled with artificial freshwater medium, ADaM (Klüttgen et al. 1994) modified according to Ebert et al. (1998). We also established and maintained mass cultures of *D. galeata* infected with *C. mesnili*. Prior to the experiment the infected cultures were screened with a stereo microscope and infected females were collected to be used as donor animals.

We began the experiment by distributing uninfected *Daphnia* evenly into 64 populations of 200 animals each in 400-ml jars so that the age and size distribution was as similar as possible among all populations. We fed the animals daily with suspension of the green algae *Scenedesmus gracilis* grown in chemostats with a ration of 40 million cells per jar per day. We kept the cultures in a growth chamber at 20°C with 16 h light/8 h dark period, and randomized the position of the jars weekly.

Eight of the populations were used as uninfected controls, while the rest of the populations were infected

by adding 20 infected adult donor females within a net enclosure into each jar. The net (250 μm) prevented the infected and uninfected *Daphnia* from mixing, but allowed algae and parasite transmission stages to pass through. The nets were shaken every day to maximize the passage of transmission stages into the culture. To the eight uninfected control populations, we added uninfected *Daphnia* in the net enclosures. All donor animals were removed after 8 days. The experiment was set up in two blocks, starting 1 week apart. Half of the replicates in each treatment (net mesh size) belonged to each block. The blocks were always treated on the same day, but the second block was terminated 1 week later than the first.

Size-selective removal was executed with plankton nets with mesh sizes of 405, 500, 630, 700, 850, 1,000 and 1,200 μm . At the beginning of the experiment, each infected *Daphnia* population was assigned randomly to a treatment (net mesh size, eight populations each). All eight uninfected control populations were assigned to the 1,200- μm net treatment. We did not include uninfected controls for each net size treatment, because our aim was not to compare uninfected and infected populations with similar size-selective mortalities, but to compare the effect of different size-selective mortalities on infected populations. The *Daphnia* populations were poured through the nets once a week starting on day 8. The nets tended to retain the largest animals, but they allowed the smaller individuals to pass through. The 1,200- μm net did not retain any animals. Both the animals retained in the nets and the animals that went through the nets were flushed separately on 250- μm plankton nets. Animals were then checked under a stereomicroscope in a thin film of medium. The animals were counted and checked for visible signs of infection. The animals were measured for body length (from the top of the head to the base of the tail spine) in weeks 3 and 11, and the numbers of parasite clusters in the guts were counted in weeks 2, 6, 10, 11, 12 and 14–21. The animals that had passed through the nets were carefully flushed back to fresh 400-ml jars filled with fresh medium. Cetyl alcohol, which reduces the water surface tension, was added to the top of each jar in order to prevent animals from clinging to the surface. The experiment was terminated at week 21, which equates to about 10–12 *Daphnia* generations. The remaining populations were checked for the presence of the parasite 1 and 3 weeks later.

In a separate experiment, the capture efficiencies (Fig. 1) of the nets used for size-selective removal were determined. A large population of *Daphnia* was poured through each net and, for each *Daphnia* size class, the proportion of individuals retained by each net mesh size was determined. The procedure was repeated 4–5 times for each net mesh size. The populations used for determining the capture efficiencies of the nets did not contain animals larger than 2.0 mm, but in the experimental populations there were a few 2.1–2.3 mm long *Daphnia*.

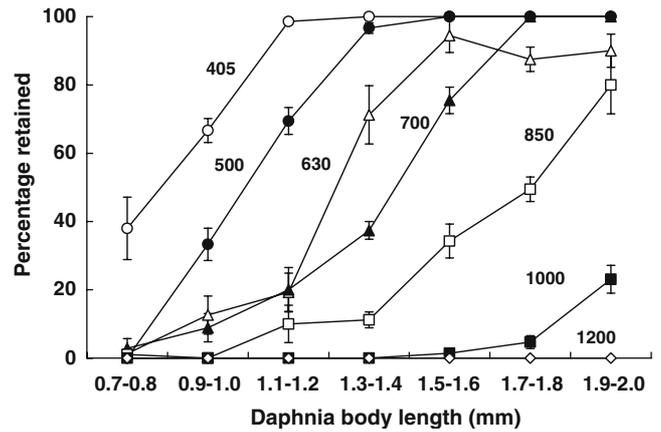


Fig. 1 The capture efficiencies of the nets used to cull the *Daphnia galeata* populations. The markers denote mean \pm SE of 4–5 replicates each. Note that the 1,200- μm mesh did not retain any *Daphnia*.

Data analysis

The time series of each replicate did not result in an equal number of data points because the persistence of host and parasite populations differed between experimental populations. When calculating the densities of the *Daphnia* and parasite populations, we first calculated the mean of each replicate over time either until extinction or until the end of the experiment. Then, we averaged these means across the replicates for each net mesh size. These values were then compared statistically. We chose two parameters, the standard deviation (SD) and the coefficient of variation in density ($\text{CV} = \text{SD} / \text{mean}$) to describe variability in *Daphnia* population density. The SD and CV values were calculated first across time for each replicate and then averaged across replicates for each treatment. Since some of the host populations lost the parasite, we calculated two different means for the prevalence of the parasite. The first mean included all populations regardless whether they contained parasites or not. The second mean included only populations which were visibly infected. Thus, the data points for a certain population were excluded after the last observation when a visible infection was recorded in that population.

The first week was excluded from the data for the figures and the analyses, because it reflects the starting conditions. In cases in which the host went extinct, we excluded the last census before the last host had died. For populations sieved through the net mesh sizes 405 and 500 μm , the procedure described above resulted in only one or no data points for each replicate and the treatments were excluded from the statistical analyses for population density, variability and parasite prevalence. In week 2, the number of parasite clusters was determined only from the animals retained in the nets and since the parasite went extinct in all populations sieved with mesh sizes 405–700 μm before the next census for parasite clusters in week 6, we excluded these treatments from the statistical analysis of parasite cluster numbers.

We tested the differences in the mean density and SD and CV in density of the *Daphnia* populations separately among the infected populations and between the uninfected and infected populations sieved with the 1,200- μm mesh size net.

Results

The percentage of the *Daphnia* in each size class retained in the nets decreased as net mesh size increased (Fig. 1). The mean size of *Daphnia* retained in a certain mesh size net (excluding the largest, 1,200 μm , which did not retain any *Daphnia*) increased with the increasing mesh size of the net (Spearman $\rho=0.96$, $n=29$, $P<0.001$).

Host and parasite extinctions

All *Daphnia* populations sieved with mesh size nets smaller than 630 μm went extinct within 4 weeks due to the sieving treatment. All animals in these populations were retained in the nets and removed from the population at the last census (Fig. 2a). Also, 75% of the populations sieved with the 630- μm mesh size net went extinct (Fig. 2a). All host populations sieved with the 700- μm net survived until the end of the experiment (Fig. 2a). The proportion of the host populations that went extinct increased again when the net mesh size increased from 850 to 1,200 μm (Fig. 2a). The proportion of infected host populations sieved with 1,200- μm mesh size net that went extinct (five of eight) was significantly higher than the proportion of uninfected host populations sieved with 1,200- μm mesh size net that went extinct (none of eight) (Fisher's exact test, $P=0.026$).

The parasite went extinct in all host populations that were sieved with mesh sizes 700 μm or smaller (Fig. 2a). In populations sieved with mesh sizes smaller than 630 μm , the parasite went extinct simultaneously with the host, whereas in most populations sieved with mesh size 630 μm the host population persisted several weeks after parasite extinction or until the end of the experiment. Extinctions of the parasite populations only (host did not go extinct) occurred in the populations sieved with 630-, 700-, 850- and 1,000- μm mesh size nets (Fig. 2a). The proportion of simultaneous extinctions of the parasite and host increased with an increase in net mesh size from 850 to 1,200 μm (Fig. 2a). The proportion of infected populations in which the parasite went extinct but the host did not, the host and the parasite went extinct simultaneously, or the host and the parasite coexisted, differed significantly between treatments, even when only the three largest mesh size treatments were included (Likelihood ratio $G=11.15$, $df=4$, $P=0.025$). In those populations among the three largest mesh size treatments in which the host and parasite went extinct simultaneously, parasite prevalence was usually high, but host density was low prior to extinction (Fig. 2b). In contrast, in those populations in which the parasite went

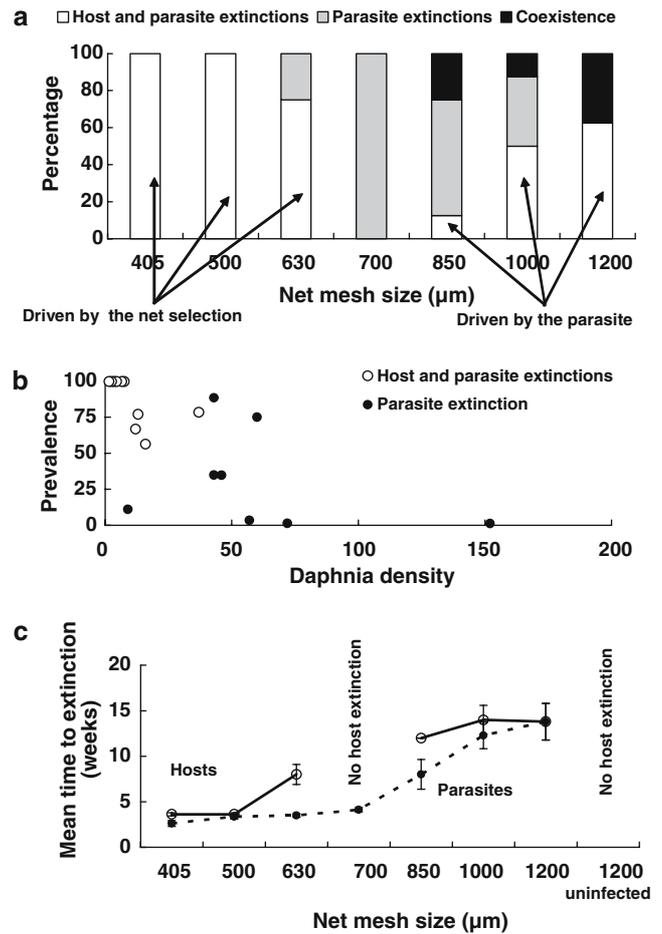


Fig. 2 **a** The percentage of host and parasite populations which went extinct during 21 weeks for each net mesh size. We suggest the host extinctions in the populations sieved with 405- to 630- μm mesh size nets to be due to size-selective mortality brought about with the net selection and in the populations sieved with the 850- to 1,200- μm mesh size nets to be due to the parasite. **b** The relationship between the parasite prevalence and host density 1 week before extinction for those populations in which both the host and the parasite went extinct (*open circles*) and for those populations in which the parasite went extinct (*filled circles*). Data from populations sieved with 850 μm or larger mesh sizes. **c** The mean time to extinction in weeks for each net mesh size for the host (*open circles*) and for the parasite (*filled circles*), for those populations in which extinction occurred. None of the uninfected control populations went extinct

extinct (before the host or when the host survived), prevalence or host density or both were low just prior to extinction (Fig. 2b).

The mean time to extinction was shortest for the populations sieved with the two smallest mesh sizes, for which all host and parasite populations went extinct within 4 weeks (Fig. 2c). In the 700- μm mesh size treatment, none of the *Daphnia* populations went extinct, but all parasite populations did. There was a positive correlation between the mesh size of the net and the mean time to extinction both for the host populations (700- μm mesh size excluded; Spearman's $\rho=0.81$, $n=32$, $P<0.001$) and the parasite populations (Spearman's $\rho=0.87$, $n=50$, $P<0.001$).

Host and parasite population densities

The mean population size of the *Daphnia* retained in the nets decreased (Fig. 3a; Spearman's $\rho = -0.87$, $n = 40$, $P < 0.001$), while the mean population size before sieving (Fig. 3a; Spearman's $\rho = 0.76$, $n = 40$, $P < 0.001$) and after sieving (Fig. 3a; Spearman's $\rho = 0.87$, $n = 40$, $P < 0.001$) increased with the increase in the net mesh size among the infected populations sieved with the 630- μm or larger mesh size nets. The uninfected, non-selected populations were aimed to control for the population dynamics of the infected, non-selected populations. Therefore, we compared the uninfected populations only with the infected populations sieved with the 1,200- μm net. The mean population size of the infected populations before sieving with the 1,200- μm net was significantly smaller than the mean population size of the uninfected populations sieved with the 1,200- μm net (t -test $t = -3.688$, $df = 7.541$, $P = 0.007$). The SD of the density increased with an increase in the net mesh size among the infected populations (Fig. 3b; Spearman's $\rho = 0.92$, $n = 39$, $P < 0.001$). However, the SD of the density did not differ between the infected and the uninfected populations sieved with the 1,200- μm mesh size net (Fig. 3b; t -test $t = 1.439$, $df = 14$, $P = 0.172$). The coefficient of variation in density (CV) was not correlated to the net mesh size among the infected populations (Fig. 3b; Spearman's $\rho = 0.10$, $n = 40$, $P = 0.523$), but was significantly higher for the infected than for the uninfected populations sieved with the 1,200- μm mesh size net (Fig. 3b; t -test $t = 4.141$, $df = 14$, $P = 0.001$).

Parasite prevalence increased as the net mesh size increased when all replicates were considered (Fig. 3c; Spearman's $\rho = 0.88$, $n = 39$, $P < 0.001$) and when only the replicates containing visibly infected *Daphnia* were included in the analysis (Spearman's $\rho = 0.70$, $n = 35$, $P < 0.001$). The mean number of parasite clusters per population increased with the increase in the net mesh size (Fig. 3c; Spearman's $\rho = 0.46$, $n = 21$, $P = 0.037$).

The mean size of the *Daphnia* after sieving (those which passed the nets) was positively correlated with the net mesh size both in week 3 (Fig. 4a; Spearman's $\rho = 0.46$, $n = 50$, $P = 0.001$) and in week 11 (Fig. 4a; Spearman's $\rho = 0.58$, $n = 38$, $P < 0.001$). The mean size of *Daphnia* retained in nets did not differ between the mesh size treatments in week 3 (Fig. 4a, Spearman's $\rho = 0.01$, $n = 41$, $P = 0.948$), but increased with the increase in net mesh size in week 11 (Fig. 4a; Spearman's $\rho = 0.45$, $n = 24$, $P = 0.028$). Only hosts larger than 1.2 mm were visibly infected (Fig. 4b–d). The number of parasite clusters per infected individual increased with an increase in *Daphnia* body length in week 11 in the populations treated with 1,000- μm nets (Fig. 4b; Spearman's $\rho = 0.62$, $n = 13$, $P = 0.024$) and 1,200 μm nets (Fig. 4b; Spearman's $\rho = 0.75$, $n = 18$, $P < 0.001$). For the 850- μm net treatment the correlation was not significant (Fig. 4b; Spearman's $\rho = 0.28$, $n = 10$, $P = 0.43$). Thus the larger individuals contained more

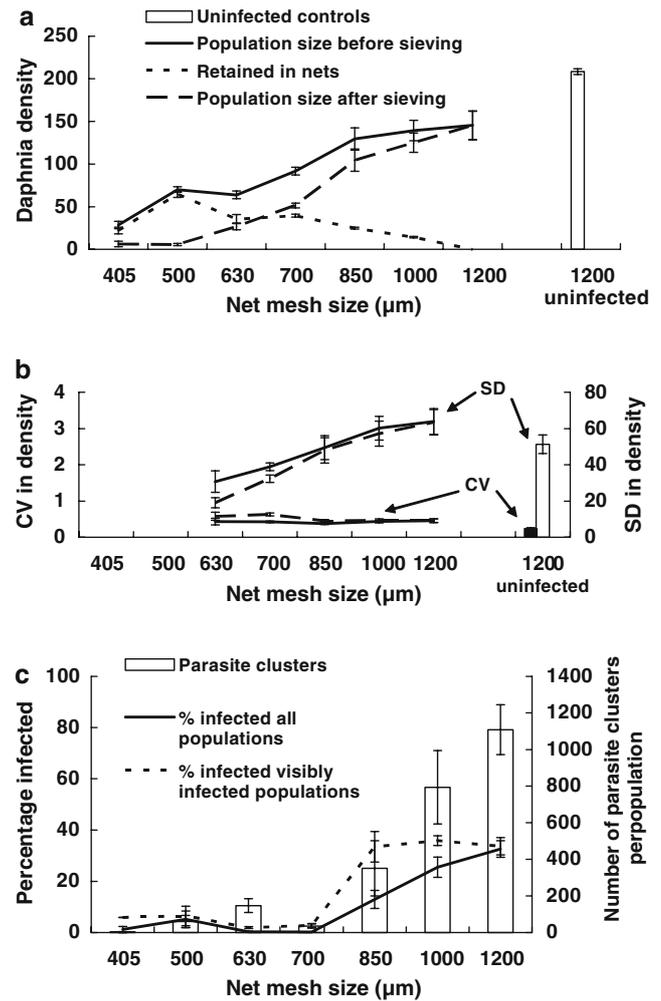


Fig. 3 **a** Mean densities (\pm SE) and **b** means of the coefficient of variation in density (\pm SE; two lower lines and filled bar) and means of the standard deviation in density (\pm SE; two upper lines and open bar) of the *Daphnia* populations for each net mesh size. Animals retained in the nets were removed from the populations at each weekly census. **c** Percentage of *Daphnia* infected with *Caullerya mesnili* (mean \pm SE) for each net mesh size among all replicates or only among replicates containing visibly infected *Daphnia* and number of *C. mesnili* clusters per population (mean \pm SE) for each net mesh size. Since the density values for the treatments with 405- and 500- μm nets are only for 1 week for most replicates due to very early extinction, they were not included in the statistical analyses but are shown for comparison

parasite clusters than the smaller ones. The populations treated with 630- and 700- μm nets contained only a few visibly infected individuals in week 3, and infected individuals were not found from these populations after week 4. The proportion of infected individuals left in the population after sieving increased with the increase in net mesh size both in week 3 (Fig. 4c; Spearman's $\rho = 0.99$, $n = 6$, $P < 0.001$) and in week 11 (Fig. 4d; Spearman's $\rho = 0.98$, $n = 5$, $P = 0.005$). Thus the proportion of large individuals with high loads of parasite clusters remaining in the populations after the sieving treatment was increased when the size-selective removal intensity decreased.

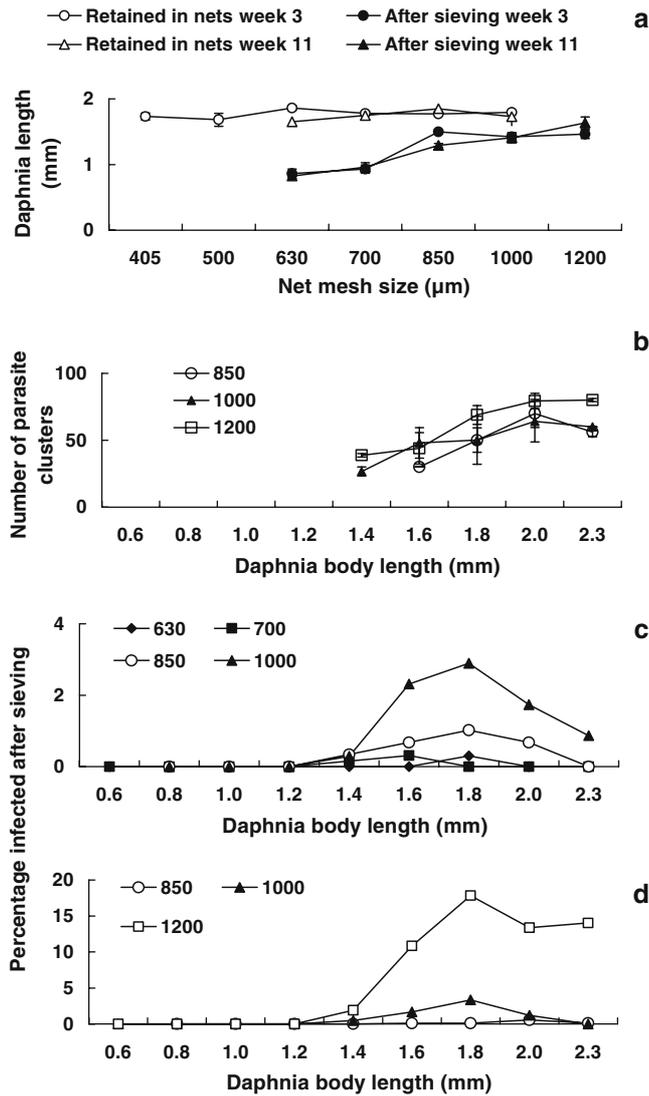


Fig. 4 **a** The mean size of the *Daphnia* retained in nets or left in populations after sieving for each net mesh size in weeks 3 and 11. **b** The number of parasite clusters (mean±SE) per *Daphnia* infected with *Caullelya mesnili* in each size class for each net mesh size in week 11. **c** The percentage of individuals with visible infections of *C. mesnili* in the infected populations in each *Daphnia* size class for net mesh sizes 630–1,200 µm that were left in the population after sieving in week 3 and **d** in week 11. The percentages were calculated from the sum of all replicates within a treatment for each week from the population density before sieving. Note that in week 3 the data for 1,200-µm net is missing and in week 11 the populations sieved with 630- and 700-µm nets did not contain anymore infected individuals

Discussion

Our results showed that size-selective removal of the largest host individuals can indeed eliminate a parasite from a host population. Under highly selective regimes (<850-µm mesh size), the parasite went extinct in all populations. When the removal rate was at its highest, the host populations went extinct as well (Fig. 2a). With no culling (1,200-µm mesh size net) the parasite drove

most of the host populations, and consequently itself, to extinction. At intermediate levels of removal (850- and 1,000-µm mesh size nets), however, the proportion of simultaneous extinctions decreased, and the outcome in which the parasite went extinct while the host populations persisted became more common (Fig. 2a).

Host populations sieved with mesh size nets smaller than 630 µm were most likely driven to extinction because of the sieving treatment. Under favourable food conditions at 20°C *D. galeata* produces the first eggs at an age of 6 days (Stich and Lampert 1984), but the first offspring are released 3 days later. Thus, only large adolescents would be able to produce eggs and release a clutch of offspring in the time period between cullings. On the last census before extinction, the size of all the animals in the populations exceeded the retention limit of the nets and were thus removed from the populations. Even in the 630-µm treatment the parasites were unlikely to have contributed to host extinction, since we recorded parasite extinction several weeks before host extinction in all except one replicate for this treatment.

In the populations sieved with the 700-µm mesh size net and in most of the populations sieved with the 630-µm net, the parasite went extinct within 3–5 weeks while *Daphnia* populations persisted several weeks more or until the end of the experiment. The mean sizes of the populations sieved with these nets were lower than the mean sizes of the populations sieved with the larger mesh sizes. Since transmission probability depends on host population size (Anderson and May 1981), low population sizes may have contributed to parasite extinction, but the removal of the large and most intensely infected individuals from the populations may also explain this finding. The size distributions from weeks 3 and 11 show that the mean size of the *Daphnia* in populations after sieving was significantly lower for the 630 and 700 µm net treatments than in the treatments with larger mesh sizes (Fig. 4a). *C. mesnili* first produce visible clusters 6 days post infection and the number of clusters increases with time since infection (Bittner et al. 2002). Visible (and thus likely infectious) infections of *C. mesnili* occurred in the size classes larger than 1.2 mm (Fig. 4b–d), but young infected females have typically only few spore clusters in their gut and thus do not produce many secondary infections. Thus we think that the likely explanation for the parasite extinctions is that the selection regime of the 630- and 700-µm mesh size nets removed many of the large infectious animals from the populations and that under conditions of reduced host density the remaining infectious animals did not produce enough secondary infections for continued, successful transmission. Epidemiologically speaking, the parasite R_0 was <1 (Anderson and May 1981).

In the populations sieved with the net mesh sizes 850 µm or larger, the host extinctions cannot be explained by the selective removal of reproducing *Daphnia*. In these treatments, the proportion of infected *Daphnia* and the number of parasite clusters per population increased relative to smaller net mesh size treatments

(Fig. 3c). Our data suggest that the proportion of extinct populations increased as an indirect consequence of a less stringent removal of *Daphnia*. The parasite suffered lower mortality and influenced the host populations more strongly, which led to host extinctions. In the 1,200- μm net treatment, parasite-induced host extinction was directly observed in the comparison of the infected and uninfected populations. Parasite-induced host extinction has been shown for non-selected populations of the same system before (Bittner et al. 2002), and the same study also showed that *C. mesnili* strongly reduces host fecundity and survival. Castrating parasites are known to be powerful regulators of host populations (Anderson and May 1981) and may also drive their hosts to extinction (Ebert et al. 2000a). *Daphnia* populations infected with *Caullerya* consistently have lower mean densities than uninfected populations treated identically (Fig. 3a; Bittner et al. 2002).

The proportion of populations in which the host and the parasite coexisted until the end of the experiment was highest among the non-selected populations. This is contrary to our expectation, since based on the previous knowledge of the parasite virulence we predicted most extinctions to occur in the non-selected populations. However, the extinctions in the non-selected populations were always simultaneous, thus supporting the expectation of parasite-induced host extinction. Possibly the rest of the populations might have gone extinct as well if the experiment had been extended. In the 850- or 1,000- μm mesh size treatments there were also extinctions of the parasite populations only. Simultaneous extinctions were always preceded by a high prevalence of the parasite (near 100%), while in cases of parasite extinctions only, parasite prevalence and/or host density were low (Fig. 2b). This suggests that some parasite extinctions in the 850- or 1,000- μm mesh size treatments were facilitated by low absolute host population sizes, but it is impossible to tell whether this was due to deterministic or stochastic effects.

Increased population variability has been suggested to increase the risk of extinction especially for small populations, since density is more likely to drop to zero during a decrease in population size (Bengtsson and Milbrink 1995; Ebert et al. 2000a; Pimm et al. 1988). Since parasites are able to decrease the host population density considerably (Anderson 1979; Ebert et al. 2000a; Hudson et al. 1998), it has been suggested that they even further increase the potential for host population extinction. In addition, parasites may also increase the variation in host population density (Ebert et al. 2000a). On the other hand, predation may decrease variation by levelling off the density peaks caused by simultaneous reproduction (Bengtsson and Milbrink 1995). In our experiment, there was no clear indication of increased variance with decreasing removal among the infected populations, as expected, since the two measures of variability gave opposite results as compared to each other. However, the uninfected non-selected populations had a slightly lower although not statistically different SD, but

statistically significantly lower CV than the infected non-selected populations. None of the uninfected populations went extinct, whereas five of eight infected non-selected populations did. Thus, it seems that the variation in density combined with the reduction in the mean population size may have contributed to raising the likelihood of host extinction in the infected non-selected populations.

Our experimental design does not allow separating between the possibilities that the effects we observed for the parasite (e.g., extinction events) were due to changes in the host populations further impinged on the parasite populations or due to interaction between host and parasite populations. In addition, we cannot exclude the possibility that the observed effects were due to changes in the populations other than size-selective removal caused by the sieving treatment. For example, it is possible that infected hosts had a different likelihood to pass through a net of a given size than uninfected hosts. However, we think that such an effect would not explain much of the variation in our experimental results, and so we think that our interpretation captures most of the ecologically relevant aspects in the system.

The intensity of the sieving treatments we used in our experiment encompassed a range from very severe culling to a non-removal treatment. The sieving treatments that mimicked the behaviour of visually hunting fish predator most closely were the 700- and 850- μm mesh size net treatments, which had their 50% removal rate at the *Daphnia* size classes between 1.3 and 1.8 mm. Fish have been shown to prefer *D. galeata* larger than 1.5 mm in Lake Constance (Stich 1989). The per capita mortalities in our experiment (range 0.02–0.31 day⁻¹ in the culled populations) fall in the range observed for example for natural populations of *D. dentifera* (Duffy et al. 2005). Thus, treatments mimicking fish predation with natural mortality rates led either to parasite extinctions only (700- μm net) or to variable dynamics ranging from simultaneous host and parasite extinctions to coexistence (850- μm net). The *Daphnia* densities in our experiment were, however, 5–50 times higher than those reported generally for most lakes (Stich 1989; Wagner et al. 2004; Wolinska et al. 2004). High densities may have facilitated the parasite spread in our experimental populations as compared to natural systems with lower host densities. Furthermore, the daily constant addition of food resources for the hosts may have further facilitated the parasite persistence. Increased feeding rate of the hosts facilitates parasite transmission and increased growth of the hosts also increases parasite propagule production (Bittner et al. 2002; Ebert et al. 2000b). In natural environments, the lower densities of the *Daphnia* populations decrease the potential for parasite spread. Thus, the culling regime needed to eradicate the parasite from a natural host population might be even less severe than the ones effective in this experiment.

Culling of parasitized size classes of hosts may lead to complex changes in the host population, which may induce changes in the exploitation pattern of the hosts by parasites. Changes in the demographic structure of the

host population may lead to changes in the behaviour of the remaining individuals, such as exploitation of previously unavailable food resources which may expose them to parasites. On a longer time scale, upon which natural populations likely operate, size-selective culling may induce evolutionary changes in the life history parameters of the host (growth, age and size of maturation) (Edley and Law 1988) or the virulence of the parasite (Choo et al. 2003; Ebert and Mangin 1997). The increase in mean size observed for *Daphnia* populations in week 11 in this study is likely to be the result of the size-selective culling rather than an evolutionary change induced by culling. *C. mesnili* has also been shown to reduce the size at maturity of its host *D. galeata* (Lass and Bittner 2002). These life history changes might be visible at the population level of the host when certain parasitized size-classes are removed. In our system, the removal of large individuals from the *Daphnia* populations and the consequent increase in relative resource level for the individuals remaining in the populations might have increased their fecundity or decreased the size at first reproduction (Brett 1992), and thus facilitated the persistence of the host populations under those culling regimes which led to eradication of the parasite populations.

Our data support the suggestion that *Daphnia* parasites might suffer in nature in the presence of strong fish predation (Duffy et al. 2005; Ebert et al. 1997). A model based on realistic empirically derived parameters from a *Daphnia*–microparasite–fish predator system indicated that a selective predator may crucially change the parasite's ability to persist even after a successful invasion (Hall et al. 2005a). The model also shows, paradoxically, that a predator which is selectively neutral or even avoids infected hosts may cause host and consequently parasite extinction (Hall et al. 2005a). The model by Hall et al. (2005a, b) does not take into account the size-dependent infection probability and infection intensity which has been shown for infected *Daphnia* populations (Mangin et al. 1995; Stirnadel and Ebert 1997). We speculate that incorporating these effects into the model would strengthen their results. Our results show that removal of the largest individuals, irrespective of their infection status, can be sufficient to eradicate a parasite from *Daphnia* populations. Thus, predators might control the occurrence and epidemics of also such parasites which do not make their hosts more conspicuous. The effect is probably strongest for parasites with a long latent (non-infectious growth stage) period, because in this case the hosts responsible for spreading the disease are older and therefore less likely to still be alive. Some parasites of *Daphnia*, which usually occur in fishless ponds, are known to have latent periods of up to 20 days (Ebert et al. 2000a). In nature, *C. mesnili* has been found in *D. galeata* and *D. hyalina* in large lakes with intensive predation by size-selective fish (Bittner et al. 2002; Wolinska et al. 2004). However, prevalences of *C. mesnili* are usually low in spring and summer, when fish predation is high, and high in fall when predation strongly declines. Thus, we suggest that the occurrence of *C. mesnili* is controlled by fish predation.

Theoretical and empirical studies have shown that populations with virulent parasites are highly unstable (Anderson and May 1991; Ebert et al. 2000a). Predators can thus have a strong impact on the host-parasite dynamics in these systems (Packer et al. 2003). Removal of infected individuals or over-infected size/age classes by predation might be a mechanism allowing the coexistence of virulent parasites with their hosts. For our experimental *Caullerya*–*D. galeata* populations, the parameter space providing stable coexistence was rather limited. Although weak culling reduces the chance that the parasite drives its host to extinction, it increases stochastic effects for the parasite, leading to its extinction. Although low densities are likely to restrict the spread of parasites in natural populations, and increase the potential for predator control, the larger absolute population sizes and spatial heterogeneity in parasite distributions (Hall et al. 2005b) are likely to provide conditions for stable coexistence in natural populations.

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