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The Influence of Intensity of Infection by a Trematode Parasite on the Reproductive Biology of *Gammarus insensibilis* (Amphipoda)

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Abstract—Thomas F., Verneau O., Santalla F., Cézilly F. & Renaud F. 1996. The influence of intensity of infection by a trematode parasite on the reproductive biology of *Gammarus insensibilis* (Amphipoda). *International Journal for Parasitology* 26: 1205–1209. Following the behavioural alterations induced by the trematode *Microphallus papillorobustus* Rankin 1940 (Trematoda, Microphallidae) on its second intermediate host, the amphipod *Gammarus insensibilis*, infected individuals are likely to mate among themselves. We investigated the influence of parasite intensity on the reproductive biology of infected hosts. In the mating system of amphipods, males compete severely for access to females and large males have greater ability to obtain large and more fecund females. We showed that the null hypothesis of random pair formation according to parasite intensity could not be rejected. In addition, infected males obtained females of the expected size according to their own sizes, whatever their parasite intensities. However, in both males and females, the parasite intensity increased the intermoult duration. Because size and reproductive success are strongly correlated in amphipods, we discuss the influence of this process on host fitness. Copyright © 1996 Australian Society for Parasitology. Published by Elsevier Science Ltd.

Key words: Sexual selection; parasitism; amphipod; trematode; *Gammarus insensibilis*; *Microphallus papillorobustus*.

INTRODUCTION

In recent years, there has been a growing interest in understanding how parasites affect the mating success of their hosts. Parasites can induce non-random mating in their hosts through various mechanisms, mainly by reducing mate attractiveness (Hamilton & Zuk, 1982; Milinski & Bakker, 1990; Rosenqvist & Johansson, 1995). In addition, because of depleted energy reserves or reduced survival, infected individuals may also invest less time and

energy in discriminating among mates (Poulin, 1994), and/or have a reduced competitiveness (Howard & Minchella, 1990). Therefore, we expect (i) parasitized individuals to obtain more frequently mates of low value compared with uninfected ones (Poulin, 1994) and (ii) mated and unmated individuals to differ in parasite loads (Møller, 1991).

The breeding biology of the brackish water crustacean *Gammarus insensibilis* (Amphipoda) is similar to that of the majority of *Gammarus* species (Hynes, 1954, 1955; Hartnoll & Smith, 1978; Birkhead & Clarkson, 1980). Fecundity increases with female size and each moult corresponds to 1 episode of reproduction, since a new batch of eggs is fertilized by

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males at this occasion (Hartnoll & Smith, 1978; Birkhead & Clarkson, 1980). Males compete severely for access to large females close to their moult and, in order to secure a mating, they guard females until the fertilization of eggs is possible (i.e. precopulatory mate guarding). This period of guarding is costly for the male since it carries the female under its body for several days (Ward, 1983) without feeding (Robinson & Doyle, 1985; Sutcliffe, 1993), while performing most of the swimming movements (Greenwood & Adams, 1984; Elwood, Gibson & Neil, 1987). Several studies have demonstrated that males and females within pairs are frequently matched for size (Ward, 1988; Thomas *et al.*, 1995a). Large males have a greater ability to access large and more fecund females, and are able to take over females held by smaller males (Ward, 1983). In this respect, male-male competition for access to larger, more fecund females has been proposed to be important in promoting size assortative mating in amphipods (Ward, 1983, 1984a, 1988).

The trematode *Microphallus papillorobustus* Rankin 1940 (Microphallidae) is a debilitating parasite that, in order to favour its transmission to charadriiform birds (definitive hosts), induces aberrant photophilic behaviour in *G. insensibilis* (Helluy, 1983a, b, 1984). Recently, Thomas *et al.* (1995a) found that this trematode strongly influenced the reproductive biology and the pattern of mating of *G. insensibilis*. In infected populations, fecundity of females was significantly reduced, and unpaired males and females were more parasitized than paired ones. Furthermore, there was an assortative mating by parasitic prevalence. The vertical segregation between photophilic infected and photophobic uninfected individuals (Thomas, Renaud & Cézilly, in press) appeared likely to explain the overrepresentation of pairs UU (Uninfected male-Uninfected female) and II (Infected male-Infected female). One obvious consequence of this vertical segregation in the field is that infected males mainly compete among themselves for access to infected females.

The aim of this paper is to evaluate, among infected individuals, the influence of parasite intensity (i.e. the number of parasites per infected individual, Margolis *et al.*, 1982) of *M. papillorobustus* on its host's reproductive fitness. Given the existence of competition between males in order to gain access to larger, more fecund females, we first analysed within homogamic pairs II, the potential influence of the male parasite intensity on its sexual performance (i.e. the size of its partner). Then, because size and reproductive fitness are

strongly correlated for both males and females, we examined indirectly the influence of parasite intensity on host growth, through its effect on the moult cycle.

MATERIALS AND METHODS

Parasite load and pattern of mating. Two samples (S1 and S2) of pairs of *G. insensibilis* in precopula were collected following the methodology described in Thomas *et al.* (1995a), in the brackish lagoon of Thau (southern France, 43°25'N, 3°35'E) during December 1995 and February 1996, respectively. Pairs involving infected males were identified in the field through the aberrant behaviour induced by the parasite. Assuming that assortative mating for parasitic prevalence was predominating in the field (Thomas *et al.*, 1995a, 1996), we obtained *a priori* 74 (S1) and 75 (S2) II pairs. Males and females of each pair were killed in alcohol (70 v/v), measured in length (from head to tip of telson) and dissected in order to count cerebral metacercariae of *M. papillorobustus*. Metacercariae of this trematode are permanent ovoid cysts (270 × 350 µm; Rebecq, unpublished data) located within the amphipod brain (Helluy, 1983b). For both males and females, we estimated the parasite intensity and the prevalence (proportion of infected individuals, Margolis *et al.*, 1982). The exact determination of the parasite load in S1 and S2 provides for both samples an estimation of the parasite distribution within the population of paired individuals. To test the null hypothesis of random pair formation according to parasite intensity within II pairs, we used sampled randomization tests (Sokal & Rohlf, 1981) to compare the pairings observed with a large enough sample of possible (random) pairings. Thus, for each sample S1 and S2, we considered the observed sets of pairs as 1 of many but equally possible outcomes that could have arisen by chance. We formed 2500 possible outcomes by randomly rearranging males and females in pairs and obtained the distribution of the difference between male and female parasite intensity (DMF). Using the Kolmogorov-Smirnov 1-sample test, the significance of assortative pairing according to parasite intensity was estimated by comparing the observed distribution of DMF with that obtained by randomly rearranging males and females. The potential parasite-mediated alteration of male competitiveness was examined through the relationship between the residuals of the regression of female size on male size (i.e. male performance corrected for size) and parasite intensity in males. To analyse the significance of the relationship, we formed 20,000 possible outcomes by randomly rearranging male performance corrected for size and male parasite intensity, and computed for each combination the value of the Spearman rank-order correlation coefficient between the 2 variables. When the observed value of the Spearman rank-order correlation coefficient was positive, the significance of the relationship was then calculated as the probability of obtaining a value superior or equal to that observed.

Host growth. Because growth in gammarids is dependent on the moulting process, we investigated the influence of

parasite load on intermolt duration. A sample of unpaired *G. insensibilis* ($n=211$) was collected in November 1995 at the same station. We collected mainly infected individuals with an aberrant behaviour. In the laboratory, individuals were maintained individually in small cups (diameter, 2 cm; height, 5 cm) filled with constantly aerated sea water (20°C , 38°‰), at the natural photoperiod (10 h light:14 h dark) and with an excess of fish food (Tetra Ani Min). The cups were examined daily and the delay between 2 consecutive moults was recorded for each gammarid. The amphipod's moult was detected by the presence of a whole or fragmented exuvium (Ward, 1984b). After their second moult, gammarids were sexed, measured and dissected to determine their parasite load of *M. papillorobustus*. Following Margolis *et al.* (1982), we estimated the mean abundance defined as the mean parasite load of all individuals. The influence of the parasite load of *M. papillorobustus* on its host intermolt duration was examined through the relationship between the residuals of the regression of host intermolt duration on host size (i.e. intermolt duration corrected for size) with the parasite abundance. To analyse the significance of the relationship, we formed 20,000 possible outcomes by randomly rearranging intermolt duration corrected for size and parasite abundance, and analysing the significance of the Spearman rank-order correlation coefficient. Other statistical tests (ANOVA, linear regression, ANCOVA, Mann-Whitney U-test, Wilcoxon signed rank test) were performed following Sokal & Rohlf (1981) and Siegel & Castellan (1988). All tests were 2-tailed. Throughout the paper values given are mean \pm S.D. The significance level chosen was 5%. All analyses were performed using *Logithec* (V. Boy, Station biologique Tour du Valat).

RESULTS

Parasite intensity in pairs

The exact determination of parasite loads revealed that for S1 and S2 the parasite prevalence was 100%. Mean parasite loads between the 2 samples were not significantly different (Mann-Whitney test, males $U=2425.5$, $P=0.17$, females $U=2608$, $P=0.52$). The frequency distribution of *M. papillorobustus* within the 2 sexes conforms to a negative binomial distribution (Kolmogorov-Smirnov test, $P>0.05$ in all cases). The values of k (the aggregation coefficient) were for males and females 1.65, 1.3 (S1) and 1.11, 1.89 (S2). Within pairs, the mean intensity of males was slightly, but not significantly, higher than that of females (mean \pm S.D., S1: males 5.21 ± 5.2 , females 5 ± 6.3 , Wilcoxon signed rank test $z=0.44$, $P=0.66$; S2: males 5.1 ± 6.1 , females 4.8 ± 4.6 , Wilcoxon signed rank test $z=0.15$, $P=0.88$). For both S1 and S2, the null hypothesis of random pair formation with respect to parasite intensity could not be rejected (Kolmogorov-Smirnov 1-sample test, S1: $D=0.06$; S2: $D=0.05$; $P>0.05$ in both cases).

Male competitiveness and parasite intensity

Males in S1 were significantly smaller than those of S2 (mean \pm S.D., S1: 14.7 ± 1.4 mm, S2: 15.2 ± 1.7 mm; ANOVA, $F_{1,147}=4.09$, $P=0.03$), whereas females were significantly larger (mean \pm S.D., S1: 12.07 ± 1.43 mm, S2: 10.7 ± 1.8 mm, Mann-Whitney test, $U=1605.5$, $P<0.001$). Within the 2 samples there was a positive relationship, significant only for S2, between male size and female size within the pair (S1, $n=74$, $r=0.12$, $P=0.32$; S2, $n=75$, $r=0.57$, $P<0.001$). Differences between slopes were significant (analysis of covariance, $F_{1,146}=9.67$, $P=0.003$). For both S1 and S2, residuals of the regressions of female size on male size were not correlated with parasite intensity in males (Spearman's rank-order correlation coefficient: S1, $r_s=0.16$, S2, $r_s=-0.06$; $P>0.05$ in each case). Thus, at equal size, variance in male performance was not explained by their parasite intensities.

Host intermolt duration

For both males and females, intermolt duration was positively correlated with size (males: $r=0.60$, $n=107$, $P<0.001$; females: $r=0.49$, $n=104$, $P<0.001$). Residuals from the previous regressions were positively correlated with the number of parasites per host (Spearman rank correlation, males: $r_s=0.37$, $n=107$, $P<0.001$; females: $r_s=0.47$, $n=104$, $P<0.001$; Fig. 1). These relationships were still significant when uninfected hosts were excluded (Spearman rank correlation, males: $r_s=0.21$, $n=81$, $P=0.03$; females: $r_s=0.37$, $n=63$, $P=0.001$). Thus, heavily infected individuals, compared with uninfected ones of the same size, moulted less frequently. We then compared intermolt duration corrected for size between uninfected and singly infected individuals. Although it is significant only for males (ANCOVA, $F_{1,52}=0.703$, n.s.; comparison of intercepts, $F_{1,52}=7.083$, $P=0.01$), the same tendency was observed for both males and females (intermolt duration, mean \pm S.D., uninfected males: 8.07 ± 2.6 , $n=26$; singly infected males: 9.96 ± 2.7 , $n=28$; uninfected females: 8.6 ± 2.1 , $n=41$; singly infected females: 9 ± 2.2 , $n=21$).

DISCUSSION

Size assortative pairing is a common phenomenon in the mating system of gammarids (Birkhead & Clarkson, 1980; Ward, 1983, 1984a; Elwood *et al.*, 1987; Crespi, 1989; Thomas *et al.*, 1995a, b). Previous studies (Thomas *et al.*, 1995a, 1996) that compared UU with II pairs found no significant influence of the prevalence of *M. papillorobustus* on this pattern of mating. Here, variation in the size

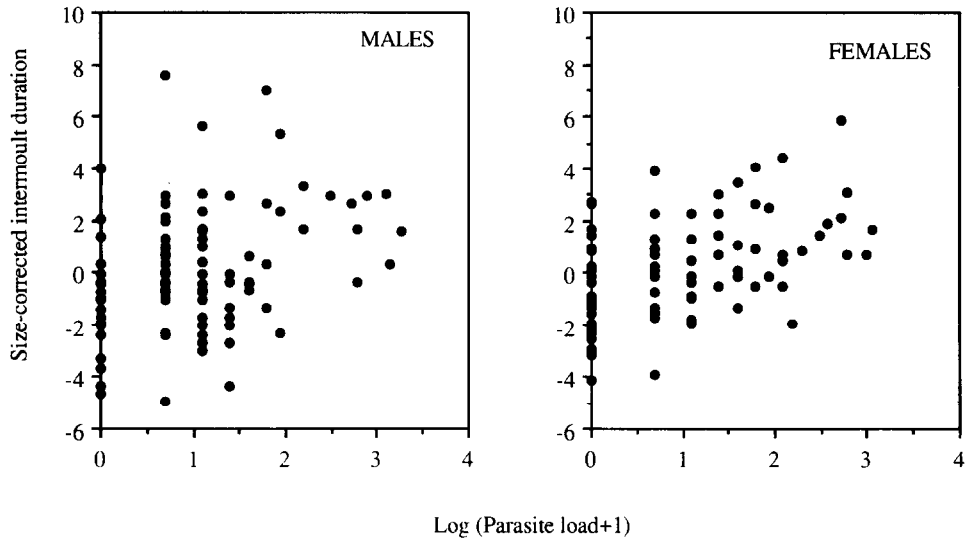


Fig. 1. Relationships between intermoult duration (corrected for size) and number of parasites per host in males and females.

assortative pairing occurred over time, whereas the mean parasite intensity remained stable between the 2 sampling dates. The exact role of parasitism in this pattern of mating remains unclear and would require monitoring of pairing patterns in an experimental setting. The absence of a relationship between (i) the parasite intensities of pair members and (ii) between male performance (corrected for size) and male parasite intensity suggests that, among infected individuals, *M. papillorobustus* has no direct influence on sexual selection through its intensity. Infected males obtained partners of the expected sizes according to their own size, irrespective of their parasite intensity. However, since size and mating success are positively correlated in males (Ward, 1988), the detrimental effect of the parasite load of *M. papillorobustus* on growth has to be considered in the context of sexual selection. By increasing the intermoult duration, the parasite load in *M. papillorobustus* indirectly reduces, in heavily infected males of small or medium size, the probability to access later in their life the larger females of the population. This phenomenon could explain the higher proportion of heavily infected males of smaller size in unpaired males compared with paired ones (Thomas *et al.*, 1995a).

Debilitating parasites may be expected to induce alterations of several traits in their hosts for at least 2 main reasons. First, manipulative changes may be accompanied by additional effects which have no particular adaptive significance for the parasite, but that substantially alter the host's population biology (Lefcort & Blaustein, 1995; Thomas *et al.*, 1995a).

Second, because debilitating parasites are generally harmful to the host, changes in life history traits can theoretically evolve in order to temper the negative effect of the parasite (Michalakis *et al.*, 1992; Poulin, 1994). Theoretically, female amphipods could increase the number of reproductive events by increasing moult frequency. Because infected females experience a reduced survival, there should be greater urgency for them to reproduce than for uninfected females. Contrary to this expectation, we did not find a reduced intermoult duration in infected females. The detrimental effect on growth may have several repercussions for female reproductive fitness. Indeed, fecundity in *G. insensibilis*, as in the majority of amphipods, is largely influenced by size. Large females produce more eggs than smaller ones (Ward, 1988; Thomas *et al.*, 1995a). Thus, for the same age, females infected by *M. papillorobustus* are less fecund because they are smaller and, in addition, given the constraint of exuviation for egg fertilization, they are less frequently available for mating. This may explain why in the field there is a larger proportion of females with more than 4 metacercariae in unpaired females when compared with paired ones (Thomas *et al.*, 1995a).

In conclusion these results, and those obtained by Thomas *et al.* (1995a, 1996), suggest that in populations of *G. insensibilis* infected by the trematode *M. papillorobustus* a strong perturbation is induced by the mere presence of the parasite, while its intensity reduces indirectly the host's reproductive success through its detrimental effects on the moult cycle.

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REFERENCES

- Birkhead T. R. & Clarkson K. 1980. Mate selection and precopulatory guarding in *Gammarus pulex*. *Zeitschrift für Tierpsychologie* **52**: 365–380.
- Crespi B. J. 1989. Causes of assortative mating in arthropods. *Animal Behaviour* **38**: 980–1000.
- Elwood R., Gibson J. & Neil S. 1987. The amorous *Gammarus*: size assortative mating in *G. pulex*. *Animal Behaviour* **35**: 1–6.
- Greenwood P. J. & Adams J. 1984. Sexual dimorphism in *Gammarus pulex*: the effect of current flow on pre-copula pair formation. *Freshwater Biology* **14**: 203–209.
- Hamilton W. D. & Zuk M. 1982. Heritable true fitness and bright birds: a role for parasites. *Science* **218**: 384–387.
- Hartnoll R. G. & Smith S. M. 1978. Pair formation and the reproductive cycle in *Gammarus duebeni*. *Journal of Natural History* **12**: 501–511.
- Helluy S. 1983. Un mode de favorisation de la transmission parasitaire: la manipulation du comportement de l'hôte intermédiaire. *Revue d'Ecologie (Terre et Vie)* **38**: 211–223.
- Helluy S. 1983. Relations hôtes-parasites du trématode *Microphallus papillorobustus* (Rankin, 1940). II Modifications du comportement des *Gammarus* hôtes intermédiaires et localisation des métacercaires. *Annales de Parasitologie Humaine et Comparée* **58**: 1–17.
- Helluy S. 1984. Relations hôtes-parasites du trématode *Microphallus papillorobustus* (Rankin, 1940). III Facteurs impliqués dans les modifications du comportement des *Gammarus* hôtes intermédiaires et tests de prédation. *Annales de Parasitologie Humaine et Comparée* **59**: 41–56.
- Howard R. D. & Minchella D. J. 1990. Parasitism and mate competition. *Oikos* **58**: 120–122.
- Hynes H. B. N. 1954. The ecology of *Gammarus duebeni* Lilljeborg and its occurrence in freshwater in western Britain. *Journal of Animal Ecology* **23**: 38–84.
- Hynes H. B. N. 1955. The reproductive cycle of some British freshwater Gammaridae. *Journal of Animal Ecology* **24**: 352–387.
- Lefcort H. & Blaustein A. R. 1995. Disease, predator avoidance, and vulnerability to predation in tadpoles. *Oikos* **74**: 469–474.
- Margolis L., Esch G. W., Holmes J. C., Kuris A. M. & Schad G. A. 1982. The use of ecological terms in parasitology (report of an *ad hoc* committee of the American Society of Parasitologists). *Journal of Parasitology* **68**: 131–133.
- Michalakis Y., Olivieri I., Renaud F. & Raymond M. 1992. Pleiotropic action of parasites: how to be good for the host. *Trends in Ecology and Evolution* **7**: 59–62.
- Milinski M. & Bakker T. C. M. 1990. Female sticklebacks use male coloration in mate choice and hence avoid parasitized males. *Nature* **344**: 330–333.
- Møller A. P. 1991. Parasites, sexual ornaments, and mate choice in the barn swallow. In: *Bird-parasite interactions. Ecology, Evolution and Behaviour* (Edited by Loye J. E. & Zuk M.), pp. 328–343. Oxford University Press, Oxford.
- Poulin R. 1994. Mate choice decisions by parasitized female upland bullies, *Gobiomorphus breviceps*. *Proceedings of the Royal Society of London (Series B)* **256**: 183–187.
- Robinson B. W. & Doyle R. W. 1985. Trade-off between male reproduction (amplexus) and growth in the amphipod *Gammarus lawrencianus*. *Biological Bulletin* **168**: 482–488.
- Rosenqvist G. & Johansson K. 1995. Male avoidance of parasitized females explained by direct benefits in a pipefish. *Animal Behaviour* **49**: 1039–1045.
- Siegel S. & Castellan N. J. 1988. *Nonparametric Statistics for the Behavioural Sciences*, 2nd edition. McGraw-Hill, New York.
- Sokal R. R. & Rohlf F. J. 1981. *Biometry*, 2nd edition. Freeman, New York.
- Sutcliffe D. W. 1993. Reproduction in *Gammarus* (Crustacea: Amphipoda): male strategies. *Freshwater Forum* **3**: 97–109.
- Thomas F., Renaud F., Derothe J. M., Lambert A., De Meeüs T. & Cézilly F. 1995. Assortative pairing in *Gammarus insensibilis* (Amphipoda) infected by a trematode parasite. *Oecologia* **104**: 259–264.
- Thomas F., Lambert A., De Meeüs T., Cézilly F. & Renaud F. 1995. Influence of *Microphallus hoffmanni* (Trematoda, Microphallidae) on the survival, sexual selection, and fecundity of *Gammarus aequicauda* (Amphipoda). *Canadian Journal of Zoology* **73**: 1634–1639.
- Thomas F., Renaud F. & Cézilly F. 1996. Assortative pairing by parasitic prevalence in *Gammarus insensibilis* (Amphipoda): patterns and processes. *Animal Behaviour* **52**: 1–8.
- Ward P. I. 1983. Advantages and a disadvantage of large size for male *Gammarus pulex* (Crustacea Amphipoda). *Behavioural Ecology and Sociobiology* **14**: 69–76.
- Ward P. I. 1984. The effects of size on the mating decisions of *Gammarus pulex* (Crustacea Amphipoda). *Zeitschrift für Tierpsychologie* **64**: 174–184.
- Ward P. I. 1984. *Gammarus pulex* control their moult timing to secure mates. *Animal Behaviour* **32**: 927.
- Ward P. I. 1988. Sexual selection, natural selection and body size in *Gammarus pulex* (Amphipoda). *American Naturalist* **131**: 348–359.