# *SUPPLEMENTARY INFORMATION: CONFIRMATION OF THE EFFECT OF FEEDING TREATMENTS ON PARASITE PERFORMANCE*

## *Statistical analysis*

To verify that our feeding treatments had an effect on parasite performance in our study, we used general linear models within the stats package in R (R Development Core Team, 2010). We modelled the presence or absence of a cercomer on 9 dpi, the parasite size 10 dpi (day-11 copepods) or 16 dpi (day 17 copepods) and the infection success in fish on day 11 (day-11 copepods) or day 17 (day-17 copepods) as response variables and the feeding treatment as fixed factor. For the presence or absence of a cercomer and the infection success we used a binomial error structure, for parasite size we used a gaussian error structure. For an overview of treatment groups and variables measured, see Figure 1 in the main text.

## *Effect of feeding treatment on day 11 copepods*

Parasites in day-11 copepods in the high food treatment developed significantly faster than those in the low food treatment. On day 9, 60 % of parasites in the high vs. 18 % in the low food treatment possessed a cercomer (Z1,119=-4.520, p<0.0001). Parasites in the high food treatment were also larger 10 dpi (mean +/- 95 % CI: 20425 +/- 862 um2 vs. 17199 +/- 671 um2, t1,120=-5.79, p<0.0001) and more likely to infect fish on day 11 (15 % vs. 3 %, Z1,119=-2.005, p=0.045).

## *Effect of feeding treatment on day 17 copepods*

In the day 17 copepods differences between feeding treatments were less pronounced. There were no significant differences in whether parasites possessed a cercomer 9 dpi (High vs. low food treatment: 47 vs. 52 %, Z1,120=0.496, p=0.620) and how likely they were to infect fish (High vs. low food treatment: 88 vs. 76 %, Z1,120=-1.583, p=0.114). However, parasites in a high food treatment again grew larger than those in a low food treatment (mean +/- 95 % CI: 25201+/- 851 um vs. 23514 +/- 890, t1,100=-2.685, p=0.0085). These results are consistent with previous claims that *S. solidus* growth is more responsive than ontogeny to resource availability (Benesh, 2010).

In summary, the feeding treatment was sufficiently aggressive to create variation in parasite traits considered fitness relevant.

## *Effect of feeding treatment on host lipids*

We additionally recorded lipid droplets in the hemocoel of infected copepods. As would be expected, under high food conditions copepods had more lipid droplets than under low food conditions (general linear model with poisson error family, z1,100 = -3.484, p=0.0005). Infection does not alter the number of lipid droplets (Franz and Kurtz, 2002). Hence our feeding treatments also affected the resource stores within hosts.

# *References*

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**Franz, K. and Kurtz, J.** (2002). Altered host behaviour: manipulation or energy depletion in tapeworm-infected copepods? *Parasitology* **125**, 187–196. doi:10.1017?S0031182002001932.

**Hothorn, T., Bretz, F. and Westfall, P.** (2008). Simultaneous inference in general parametric models. *Biometrical Journal* **50**, 346–363. doi:10.1002/bimj.200810425.

**R Development Core Team** (2010). R: a language and environment for statistical computing. *R Foundation for Statistical Computing*.

Table S1: Post hoc tests for the effect of the interaction between feeding treatment and infection for the distance copepods moved. Test statistics and p values were obtained using general linear hypotheses within the multcomp package in R (Hothorn *et al.*, 2008). Feeding treatment and infection were combined into a single factor with four different levels comprising all possible combinations between these two factors (FEED\_INF): Uninfected control, high food (c\_H); uninfected control, low food (c\_L); infected, high food (inf\_H); infected, low food (inf\_L). The comparisons were based on the following models (see Table 1 for more details): Day-11 copepods: FEED\_INF + INTERVAL + (INTERVAL | RE) + (1 | ID), Day 17: FEED\_INF + INTERVAL + AGE + INTERVAL : AGE + (INTERVAL | RE) + (AGE | ID). Significant p-values have been marked in bold.

|  |  |  |
| --- | --- | --- |
|  | Day-11 copepods | Day-17 copepods |
| Comparison | z | p | z | p |
| c\_H – c\_L | -4.771  | **<0.001** | 2.896 | **0.0194** |
| inf\_H – inf\_L | -2.140 | 0.140 | -0.556 | 0.9444 |
| c\_H – inf\_H | -8.518 | **<0.001** | -6.445 | **< 0.001** |
| c\_L – inf\_L | -5.383 | **<0.001** | -3.413 | **0.0038** |
| c\_H – inf\_L | -10.571  | **<0.001** | -6.918 | **< 0.001** |
| c\_L – inf\_H | 3. 383 | **0.004** | 2.953 | **0.0164** |

**Table S2: Associations between parasite performance and host activity and distance.** Mixed models used whether or not a copepod moved within a two second interval (Activity) or, if it moved, how far it moved (distance, log transformed) as response variables. Copepod identity (ID), the recording event (RE, i.e. a combination of copepod identity and the day of the recording), and the time interval in the recording (i.e. before vs. after the simulated predator attack, INTERVAL) were incorporated into the models’ random effect structure. INTERVAL, feeding treatment (FEED), and their interaction were additionally included as fixed effects. For day 17-copepods we included whether or not parasites were infective for fish as both fixed and random effect (AGE, together with ID) and its interaction with FEED and INTERVAL. Subsequently, we separately added measures of parasite performance (PERFORM (i.e. presence or absence on a cercomer on day 9 as an indicator of development, parasite size on day 10 (day-11 copepods) or day 16 (day-17 copepods) and infection success in fish) and all their pairwise interactions with INTERVAL, FEED and AGE (day-17 copepods). Test statistics and MCMC-estimated p-values are for the comparison with the preceding model. Null models: Day-11 copepods: INTERVAL + FEED + INTERVAL : FEED + (INTERVAL | RE) + (1 | ID), day-17 copepods: INTERVAL + FEED + AGE+ INTERVAL : FEED + INTERVAL : AGE + FEED : AGE (INTERVAL | RE) + (AGE | ID). Since we used multiple tests, according to bonferroni adjustment only p-values below 0.0042 should be considered significant at α=0.05. They have been marked in bold. P-values significant only prior to adjustment have been put in italics.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Day-11 copepods | Factors | Parasite size | Development (Cercomer present or absence 9 dpi) | Infection success in fish |
| Activity | Distance | Activity | Distance | Activity | Distance |
| DF | Chisq | p | DF | Chisq | p | DF | Chisq | p | DF | Chisq | p | DF | Chisq | p | DF | Chisq | p |
| +PERFORM | 1,9 | 0.447 | 0.5040 | 1,10 | 9.883 | **0.0017** | 1,9 | 6.591 | 0.0103 | 1,10 | 0.417 | 0.5186 | 1,9 | 3.217 | 0.0729 | 1,10 | 7.934 | 0.0049 |
| +PERFORM: INTERVAL | 1,10 | 3.490 | 0.0617 | 1,11 | 0.898 | 0.3433 | 1,10 | 0.950 | 0.3298 | 1,11 | 3.884 | 0.0488 | 1,10 | 0.332 | 0.5644 | 1,11 | 3.056 | 0.0805 |
| +PERFORM:FEED | 1,11 | 0.048 | 0.8272 | 1,12 | 1.564 | 0.2111 | 1,11 | 0.216 | 0.6418 | 1,12 | 0.089 | 0.7651 | 1,11 | 0.280 | 0.5964 | 1,12 | 0.902 | 0.3424 |
|  | 31240 observations on 355 recording events and 122 copepods | 10949 observations on 352 recording events and 122 copepods | 30712 observations on 349 recording events and 121 copepods | 10609 observations on 346 recording events and 121 copepods | 30976 observations on 352 recording events and 121 copepods | 10801 observations on 348 recording events and 121 copepods |
|  |
| Day-17 copepods | Factors | Parasite size | Development (Cercomer present or absence 9 dpi) | Infection success in fish |
| Activity | Distance | Activity | Distance | Activity | Distance |
| DF | Chisq | p | DF | Chisq | p | DF | Chisq | p | DF | Chisq | p | DF | Chisq | p | DF | Chisq | p |
| +PERFORM | 1,14 | 12.323 | **0.0004** | 1,15 | 9.099 | **0.0026** | 1,14 | 0.382 | 0.5368 | 1,15 | 0.012 | 0.9113 | 1,14 | 0.030 | 0.8637 | 1,15 | 1.242 | 0.2651 |
| +ERFORM: INTERVAL | 1,15 | 0.046 | 0.8306 | 1,16 | 3.617 | 0.0572 | 1,15 | 0.077 | 0.7813 | 1,16 | 5.435 | *0.0197* | 1,15 | 0.067 | 0.7951 | 1,16 | 1.954 | 0.1621 |
| +PERFORM:AGE | 1,16 | 0.001 | 0.9752 | 1,17 | 0.022 | 0.8830 | 1,16 | 1.574 | 0.2096 | 1,17 | 0.518 | 0.4717 | 1,16 | 5.598 | *0.0180* | 1,17 | 0.519 | 0.4713 |
| +PERFORM:FEED | 1,17 | 5.722 | *0.0168* | 1,18 | 0.696 | 0.4041 | 1,17 | 0.980 | 0.3221 | 1,18 | 1.458 | 0.2273 | 1,17 | 0.734 | 0.3917 | 1,18 | 1.421 | 0.2332 |
|  | 50512 observations on 574 recording events and 102 copepods | 18433 observations on 557 recording events and 102 copepods | 50072 observations on 569 recording events and 101 copepods | 18316 observations on 552 recording events and 101 copepods | 48928 observations on 556 recording events and 99 copepods | 17603 observations on 539 recording events and 99 copepods |