

## Supporting Information

The following Supporting Information is available for this article online:

**Table S1.** Results from Experiment 1 by diluted (Dilution 1 and 2) and undiluted treatment and day during the experiment when colonies were sampled. Results are: the number of *Fredericella sultana* colonies that were uninfected, covertly infected, overtly infected, dead, and that contained statoblasts and the number of *Tetracapsuloides bryosalmonae* sacs.

**Table S2.** Results from Experiment 2 (transplant experiment) by source and final treatments and day during the experiment when colonies were sampled. Results are: the number of *Fredericella sultana* colonies that were uninfected, covertly infected, overtly infected, dead, and that contained statoblasts and the number of *Tetracapsuloides bryosalmonae* sacs.

**Figure S1.** Frequencies of infected maternal colonies according to the proportion of statoblasts infected in their brood.

**Appendix S1.** Methods used to extract chlorophyll-a from water samples taken from each treatment.

**Appendix S2.** Methods used to detect covert infections and to estimate infection intensity semi-quantitatively.

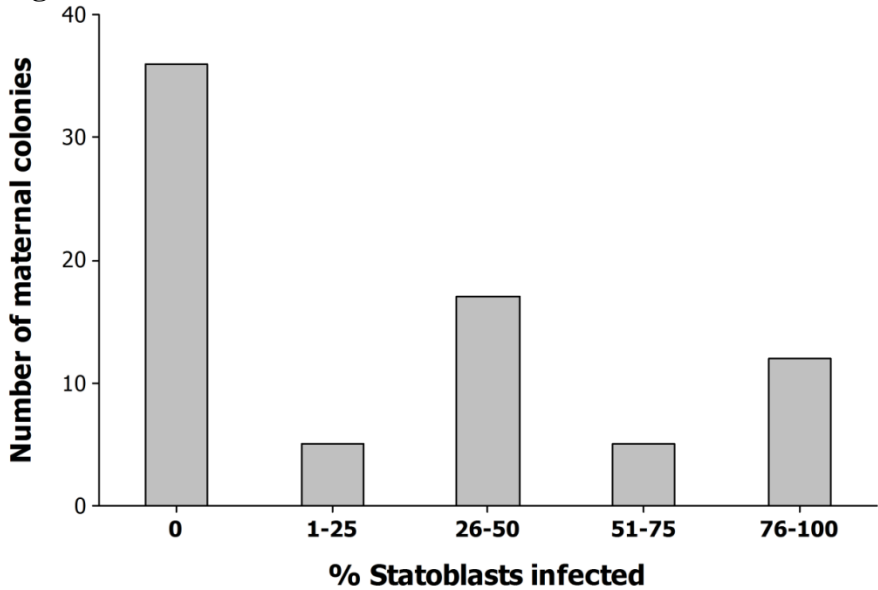
**Table S1**

Treatment	Day in study	Number of colonies					Number of sacs
		Uninfected	Covertly infected	Overtly infected	Dead	Containing Statoblasts	
Dilution 1	3	3 (27.3%)	6 (54.6%)	2 (18.2%)	0	0	2
	5	5 (50.0%)	2 (20.0%)	3 (30.0%)	0	0	7
	7	3 (30.0%)	7 (70.0%)	0	0	0	0
	9	3 (30.0%)	6 (60.0%)	1 (10.0%)	0	1 (10.0%)	2
	11	0	10 (100.0%)	0	1 (9.1%)	0	0
Dilution 2	3	1 (6.7%)	11 (73.3%)	3 (20.0%)	0	0	8
	5	5 (50.0%)	2 (20.0%)	3 (30.0%)	0	0	4
	7	1 (11.1%)	5 (55.6%)	3 (33.3%)	0	0	21
	9	2 (18.2%)	5 (45.5%)	4 (36.4%)	1 (8.3%)	1 (9.1%)	17
	11	0	8 (88.9%)	1 (11.1%)	1 (10.0%)	0	3
Undiluted	3	4 (36.4%)	7 (63.6%)	0	0	0	0
	5	2 (22.2%)	7 (77.8%)	0	0	1 (11.1%)	0
	7	2 (20.0%)	7 (70.0%)	1 (10.0%)	0	1 (10.0%)	2
	9	0	10 (90.9%)	1 (9.1%)	0	0	3
	11	0	10 (100.0%)	0	0	0	0

**Table S2**

	Final Treatment	Day in study	Number of colonies					Number of sacs
			Uninfected	Covertly infected	Overtly infected	Dead	Containing Statoblasts	
Dilution 1	Dilution 2	7	7 (50.0%)	7 (50.0%)	0	0	1 (7.1%)	0
		14	13 (65.0%)	7 (35.0%)	0	4 (0.2%)	0	0
Dilution 2		7	3 (30.0%)	7 (70.0%)	0	1 (0.1%)	1 (10.0%)	0
		14	2 (22.2%)	7 (77.8%)	0	2 (0.2%)	0	0
Undiluted		7	2 (14.3%)	12 (85.7%)	0	1 (0.1%)	2 (14.3%)	0
		14	9 (81.8%)	2 (18.2%)	0	0	1 (9.1%)	0
Dilution 1	Undiluted	7	0	13 (100.0%)	0	0	0	0
		14	10 (62.5%)	6 (37.5%)	0	0	0	0
Dilution 2		7	0	13 (86.7%)	2 (13.3%)	0	0	5
		14	6 (85.7%)	1 (14.3%)	0	1 (0.1%)	0	0
Undiluted		7	0	6 (100.0%)	0	0	0	0
		14	4 (50.0%)	4 (50.0%)	0	0	0	0

**Figure S1**



## Appendix S1

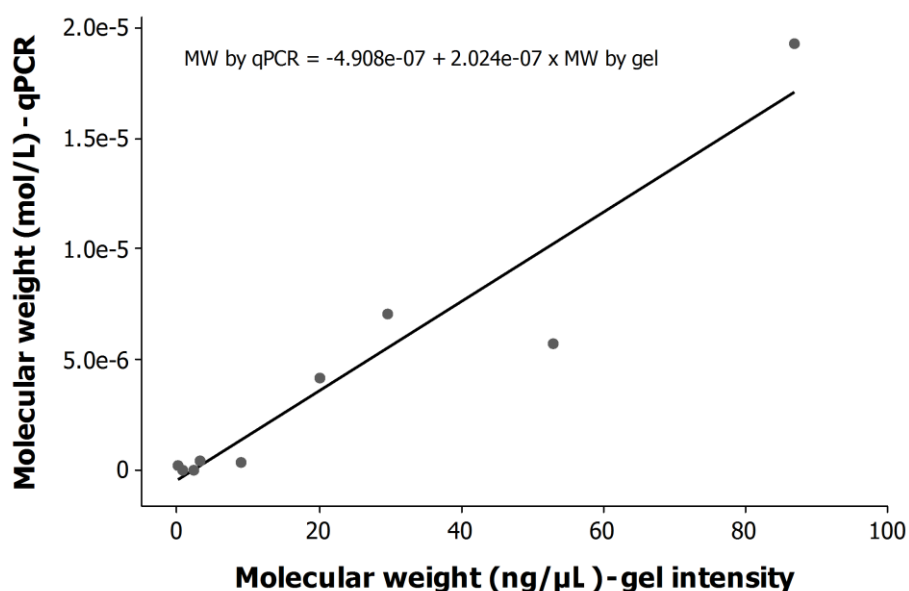
To estimate chlorophyll-a concentrations 3 x 2L ( $V_{\text{sample}}$ ) water samples from each treatment and a control (deionised water) were collected at the end of the experiment with the exception of the undiluted treatment where 1L replicates were collected. Each water sample was filtered through a Whatman<sup>®</sup> glass microfiber filter, Grade GF/D (diameter 4.25cm), which was then preserved at -80°C in a 50mL plastic tube wrapped in foil to exclude light. An ethanol extraction protocol was used on the frozen filters. Each filter was suspended in 20mL of 100% EtOH and boiled for 3sec in a water bath (at 78.5°C). The tubes were agitated and allowed to stand in the dark overnight at room temperature to extract the pigments. The next day 2.22mL of water was added to make a 90% EtOH solution, the filters were removed and the tubes were centrifuged for 7min at 3000rpm. The extract was decanted into 15mL plastic tubes and its volume measured ( $V_{\text{extract}}$ ). The pigment extract solution was pipetted into cuvettes with 10mm cell pathlength (d) and the absorbance was measured at wavelengths of 665nm (for chlorophyll-a;  $A_{665}$ ) and 750nm (to correct for background turbidity;  $A_{750}$ ) using a Shimadzu UV-1800 UV/Visible Scanning Spectrophotometer. The undegraded chlorophyll-a ( $\mu\text{g/L}$ ) was calculated using the formula:

$$[12.2*(A_{665}-A_{750})*V_{\text{extract}}]/(d*V_{\text{sample}}).$$

## Appendix S2

DNA was extracted from bryozoan samples (fragments, maternal colonies and statoblasts) using a hexadecyltrimethylammonium bromide (CTAB) protocol (Tops & Okamura 2003). To detect covert infections we undertook a PCR assay using primers diagnostic for *T. bryosalmonae* 18S rDNA [514F\_new (5'-ATTCAGGTCCATTCGTGAGTAACAAGC-3') and 776R (5'-GCTGATACACCCAATTAAGGGCAG-3')] to produce an amplicon of 244bp using PCR cycling conditions as described in Hartikainen, Fontes and Okamura (2013). Each sample was subjected to two separate PCR reactions, which were analysed in parallel as described below. PCR products were run in a standardised fashion on 1.5% agarose electrophoretic gels, stained with GelRed™ (250mL 1x TAE (Tris-acetate-EDTA), 3.3g agarose and 8.3µL GelRed™). 3µL of each PCR product ( $V_{\text{product}}$ ) and 2.5µL of Bioline HyperLadder™ I were loaded to gels, the latter being added to every gel row. Gels were run in fresh 1x TAE at 100v in a large Fisherbrand® Horizontal Electrophoresis Apparatus Model HU13W for 45min and were visualised on a UVP EpiChemi™ II Darkroom with a UVP UV transilluminator, always using the same settings. Infection intensity (ng of amplified parasite DNA per µL) was characterised semi-quantitatively (see below) from gel images inferred from two separately-run PCR replicates. The maximum pixel intensity per gel band was measured using the 1D-Multi analysis tool in AlphaEase®FC version 6.0.0 with a “rubber band” background subtraction method. The product’s molecular weight (MW) was then obtained by calibrating the average maximum pixel intensity of the band (I) against the molecular weight of the 200bp ladder ( $MW_{200\text{bp band}}$ ) based on 10ng (in 2.5µL). The following equation was then used to calculate the product’s molecular weight:  $[(I_{\text{product}} \times MW_{200\text{bp band}}) / I_{200\text{bp band}}] / V_{\text{product}}$ . Values for the average parasite DNA concentration were transformed to natural logarithms to linearise the data. This method is based on that used in Neuhauser, Huber and Kirchmair (2009). We tested its efficiency by comparing the gel band

intensity values of nine randomly selected covertly infected colonies with SYBRGreen qPCR quantification values (Fontes *et al.*, submitted). There was a significant correlation between the quantification methods: MW in mol/L (by qPCR) =  $-4.908e-07 + 2.024e-07 * \text{MW in ng}/\mu\text{L}$  (by gel intensity) ( $R^2 = 90.6\%$ ; correlation coefficient = 0.952; Analysis of Variance test: d.f. = 8, F = 67.66,  $P < 0.001$ ) (see Fig. S2). These results support the use of gel band intensity as an estimate of infection intensity.



**Figure S2.** Linear relationship between the mean values of two duplicate measures of infection intensity: molecular weight (MW) of the gel band intensity, following standard PCR, and MW in mol/L quantified by qPCR. The regression equation is presented at the top of the graph.

## References

Hartikainen, H., Fontes, I. and Okamura, B. (2013). Parasitism and phenotypic change in colonial hosts. *Parasitology* **140**, 1403-1412. doi: 10.1017/S0031182013000899.

**Neuhauser, S., Huber, L. and Kirchmair, M.** (2009). A DNA based method to detect the grapevine root-rotting fungus *Roesleria subterranea* in soil and root samples. *Phytopathologia Mediterranea* **48**, 59-72. doi: 10.14601/Phytopathol\_Mediterr-2875.

**Tops, S. and Okamura, B.** (2003). Infection of bryozoans by *Tetracapsuloides bryosalmonae* at sites endemic for salmonid proliferative kidney disease. *Diseases of Aquatic Organisms* **57**, 221-226. doi: 10.3354/dao057221.