Supporting Information

Appendix S1

In this Appendix, we provide methods used to determine the nutrient content of the two resource species (first experiment), as well as methods for the preparation of 'control' and 'extract' food treatments (second experiment). Then we describe statistical methods used to estimate components of transmission potential. We also present data on host survivorship in both experiments. In Tables S1 and S2 we provide *P*-values for pairwise contrasts of those parameters between food treatments. Ratios of carbon (C), nitrogen (N), and phosphorus (P) in both resource species are presented in Table S3.

Additional methods

Nutrient content of food

We assessed whether elemental nutrient stoichiometry (i.e., C:N:P) drove differences in food quality in the first experiment. To determine C:N:P of the two food species, we collected stationary phase samples of the high quality (*Ankistrodesmus falcatus*) and low quality (*Microcystis aeruginosa*) phytoplankton onto precombusted filters (GF/F, 0·7 µm pore size, Whatman, Piscataway, NJ, USA). C and N content (five replicates each) were measured on a 2400 series CHN analyser (Perkin Elmer, Waltham, MA, USA). P content (three replicates) was measured on a UV-1800 spectrophotometer (Shimadzu Scientific Instruments, Columbia, MD, USA) using the ascorbic acid method following persulfate digestion (APHA 1995). We compared nutrient ratios between the two food species (Table S3) using one-tailed two sample *t*-

tests with equal variances. In both experiments, food levels were standardized to C content based on absorbance (750 nm)–C regressions for each species.

Microcystis extraction, fractionation, and coating of food

In the second experiment, we tested whether protease inhibitors (or other chemical compounds) in the low quality food (*M. aeruginosa* strain NIVA-Cya 43) could have driven its effects on transmission potential in the earlier experiment. To do this, we extracted material from *M. aeruginosa* cells (using methods adapted from von Elert, Zitt & Schwarzenberger [2012]), and verified that it contained nostopeptin BN920 (Ploutno & Carmeli 2002) and cyanopeptolin CP954 (von Elert *et al.* 2005). These two compounds inhibit digestive proteases (chymotrypsins) of a related zooplankter, *Daphnia magna* (von Elert *et al.* 2012). We created food treatments by coating the extracted material onto high quality *A. falcatus*, as described below. By using *A. falcatus* as a substrate, we could isolate effects of the focal compounds while controlling for other factors, such as cell morphology or nutritional content.

Lyophilised *M. aeruginosa* (0·87 g dry mass) was exhaustively extracted in methanol (MeOH). We separated this crude extract on a reversed-phase C_{18} silica gel column (10 g Supelclean ENVI-18 SPE, Supelco, Bellefonte, PA, USA) using a stepwise MeOH/H_2O mobile phase (20%, 40%, 60%, 80%, and 100% aqueous MeOH, ending with a 100% ethyl acetate wash). This yielded six fractions that differed in polarity. To determine which fraction(s) contained the compounds of interest, we used liquid chromatography–mass spectrometry. We analysed each fraction at 1 mg mL^{-1} with a a Waters 2695 high performance liquid chromatograph coupled to a Waters 2996 photodiode array UV detector and a Waters Micromass ZQ 2000 mass spectrometer (Waters Corporation, Milford, MA, USA). A Grace Alltima C_{18}

silica gel column (Grace Alltech, Deerfield, IL, USA) was employed with a gradient of 30% to 100% aqueous acetonitrile (with 0·1% acetic acid), and the focal compounds were detected via positive and negative electrospray ionisation modes (mass-to-charge ratios: BN920: m/z = 921·5; CP954: m/z = 955·4). We estimated the relative abundance of the focal compounds across the six fractions by comparing integrated mass peak areas for each molecular ion. Approximately 90% of both compounds was contained in the fractions eluted with 60% and 80% MeOH, and the remaining 10% was in the fractions eluted with 20% and 40% MeOH.

We pooled the 60% and 80% MeOH fractions and coated them onto lyophilised *A. falcatus* cells using dimethyl sulfoxide (DMSO) as a carrier solvent; this became our 'extract' food treatment. The amount of material extracted from 1 mg C of *M. aeruginosa* was coated onto each 1 mg C of *A. falcatus*. The 'control' diet consisted of *A. falcatus* coated only with DMSO. We also coated *A. falcatus* with the pooled 20% and 40% MeOH or pooled 100% MeOH and 100% ethyl acetate fractions; these two treatments did not affect any of the components of transmission potential (R.M. Penczykowski, unpublished data). To minimize degradation of the food treatments over the course of the experiment, we prepared aliquots for each day. The aliquots were lyophilised to remove the DMSO, stored at -20 °C, and rehydrated in Artificial *Daphnia* Medium (ADaM; Klüttgen *et al.* 1994) immediately before being fed to hosts.

Statistical methods for estimating parameters

In the first experiment, we estimated components of infection risk (transmission rate, β) by simultaneously fitting models to our infection data and feeding rate data (Bertram *et al.* 2013). The two model fits had separate likelihood values, which we added together to estimate parameters, as described below. To the infection data, we fitted the dynamical transmission

model (equ. 1), where β was broken down into its constituent parts (per spore infectivity, *u*, and feeding rate, *f*, both of which increase with length squared, L_β^2; equ. 2):

$$dS/dt = -\hat{u}\hat{f}L_\beta^4 SZ \quad \text{(S1.a)}$$

$$dI/dt = \hat{u}\hat{f}L_\beta^4 SZ \quad \text{(S1.b)}$$

$$dZ/dt = -\hat{f}L_\beta^2 (S+I)Z\,. \quad \text{(S1.c)}$$

This model (equ. S1) can be solved analytically to give the predicted proportion of hosts infected, *p*, at the end of the spore exposure period of duration t_β:

$$p = \frac{I(t_\beta)}{N} = 1 - \exp\left(\hat{u}L_\beta^2\left(\frac{Z(0)}{N}\right)\left(\exp\left(-\hat{f}L_\beta^2 N\, t_\beta\right) - 1\right)\right), \quad \text{(S2)}$$

where N and $Z(0)$ are initial densities of hosts and spores, respectively, in the infection assays, and $N = S + I$ is fixed. The binomial-based likelihood function for the infection process is:

$$\ell_\beta = \binom{N}{I} p^I (1-p)^{N-I}, \quad \text{(S3)}$$

where I is the density of infected hosts at the end of the experiment.

We simultaneously estimated size-corrected feeding rate, $\hat{f}$, by fitting a natural log-transformed version of a standard formula for calculating foraging-based "clearance rate" (Sarnelle & Wilson 2008):

$$\log(C_t) = \log(C_0) - ft/V + \varepsilon, \quad \text{(S4)}$$

where C_t is the concentration of algae remaining at the end of the grazing period of length t, C_0 is the concentration of algae in ungrazed reference tubes at the end of the grazing period, $f = \hat{f}L_\beta^2$ (with size-corrected feeding rate $\hat{f}$ and body length L_β during the feeding assay), V is the volume

of medium in the tube, and errors (ε) were assumed to be normally distributed. Fitting this model (equ. S4) produced a likelihood value for feeding rate, ℓ_f. Then we estimated the parameters $\hat{u}$ and $\hat{f}$ by minimizing the sum of the two negative log-transformed likelihoods, ℓ_β and ℓ_f.

To summarize, we simultaneously fit two datasets (infection and 'clearance rate') to obtain point estimates (with bootstrapped 95% confidence intervals) for size-corrected per spore susceptibility, $\hat{u}$, and size-corrected feeding rate, $\hat{f}$. Using these estimates, we could then calculate (with bootstrapped 95% confidence intervals) the effective per spore susceptibility, *u*, as the product of $\hat{u}$ and mean length at infection squared (L_β^2); the rate of spore exposure, *f*, as the product of $\hat{f}$ and L_β^2; and total infection risk, β, as the product of *u* and *f*. In Figure 3, we show how these constituent parameters combine to form infection risk (β). Finally, we calculated transmission potential as the product of β and spore yield (σ), and bootstrapped over the infection and spore yield data to generate 95% confidence intervals (Fig. 4).

In the second experiment, we tested whether the cyanobacterial extract affected overall transmission potential (βσ), but we did not tease apart the two components of infection risk (*u* and *f*). Thus, we estimated β by fitting only the transmission model to the infection data, and bootstrapped over the infection and spore yield data to generate 95% confidence intervals for transmission potential (βσ; Fig. 5).

In Tables S1 and S2, we present *P*-values for pairwise contrasts of these parameters between food treatments in the first (Figs 2, 3, and 4) and second (Fig. 5) experiments, respectively. We performed 9999 randomizations of the datasets in each contrast (Gotelli & Ellison 2004).

Additional results

Nutrient content of food

In both phytoplankton species, amounts of N and P (relative to C; Table S3) were well above limiting levels for *Daphnia* (Sterner & Hessen 1994; Urabe, Clasen & Sterner 1997). Furthermore, high quality *A. falcatus* had significantly higher C:N, C:P, and N:P (i.e., significantly lower N:C, P:C, and P:N) compared to low quality *M. aeruginosa* (two sample *t*-test for each ratio, all $P < 0{\cdot}0001$). Thus, stoichiometric composition of the two resources did not explain differences in their quality as food for hosts.

Host survivorship

Survival of hosts to the end of the experiment (10 days after spore exposure) was similar across food treatments. In the first experiment, there was 59%, 59%, and 56% survival in the 'high', 'high-to-low', and 'low' quality manipulations, respectively, and no significant effect of food treatment on survivorship (GLM with quasibinomial error distribution for overdispersed binomial data: $F_{1,190} = 0{\cdot}13$, $P = 0{\cdot}72$). In the second experiment, there was 59%, 57%, and 74% survival in the 'control', 'control-to-extract', and 'extract' treatments, respectively, and also no significant effect of food treatment on survivorship ($F_{1,202} = 3{\cdot}65$, $P = 0{\cdot}06$).

Table S1. *P*-values for comparisons of parameter estimates between food quality manipulations in the first experiment (Figs 2, 3, and 4). Bolding denotes significant pairwise differences after Holm–Bonferroni correction.

		Food quality manipulation		
Parameter	**Symbol**	**High vs. High-to-low**	**High-to-low vs. Low**	**High vs. Low**
Infection risk (Fig. 2B)	β	**0·0005**	0·30	< **0·0001**
Size at exposure (Fig. 2C)	L_β	1·0	< **0·0001**	< **0·0001**
Size-corrected exposure rate (Fig. 3A)	$\hat{f}$	**0·0001**	< **0·0001**	< **0·0001**
Exposure rate (Fig. 3B)	f	< **0·0001**	**0·003**	< **0·0001**
Size-corrected per spore susceptibility (Fig. 3C)	$\hat{u}$	1·0	0·44	0·50
Per spore susceptibility (Fig. 3D)	u	1·0	0·099	**0·015**
Transmission potential (Fig. 4D)	$\beta\sigma$	< **0·0001**	0·061	**0·0001**

Table S2. *P*-values for comparisons of parameter estimates between food quality manipulations in the second experiment (Fig. 5). There were no significant pairwise differences after Holm–Bonferroni correction.

		Food quality manipulation		
Parameter	**Symbol**	**Control vs. Control-to-extract**	**Extract vs. Control-to-extract**	**Control vs. Extract**
Infection risk (Fig. 5B)	β	0·087	0·058	0·97
Size at exposure (Fig. 5C)	L_β	N/A[†]	N/A	0·18
Transmission potential (Fig. 5F)	$\beta\sigma$	0·020	0·040	0·65

[†]N/A indicates that there is no associated pairwise comparison

Table S3. Molar ratios (mean ± 1 SD) of carbon (C), nitrogen (N), and phosphorus (P) in the two food species.

Quality: Food species	**C:N**	**C:P**	**N:P**
High quality: *Ankistrodesmus falcatus*	7·85 ± 0·13	46·08 ± 0.83	5·87 ± 0·13
Low quality: *Microcystis aeruginosa*	6·22 ± 0·05	23·21 ± 0·45	3·73 ± 0·07

References in supporting information

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