Main question:

Does the temperature history of rotifers affect competition with protists?

Sub-questions:

1. Did the evolution period lead to changes to rotifers’ response to temperature?
2. Do rotifers have a competitive response to the protist?
3. Do rotifers have a competitive effect on the protist?

Following are answers to the above questions, observed in two growth parameters: maximum population growth rate (hereafter: growth rate, *r*) and carrying capacity (*K*).

1. Population growth rate (*r*)

Table 1. Effects of evolved temp, current temp, and competition on rotifer growth rate. Three-factor type III ANOVA. Data are log-transformed to meet test assumptions.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Predictor | SS | df | *F* | *p* |
| Evolved temperature | 0.0285 | 1 | 0.1747 | 0.6772 |
| Current temperature | 1.5953 | 1 | 9.7838 | 0.0025\*\* |
| Competition | 0.0765 | 1 | 0.4694 | 0.4955 |
| Evolved T x Current T | 0.2332 | 1 | 1.4302 | 0.2356 |
| Evolved T x Competition | 0.2020 | 1 | 1.239 | 0.2694 |
| Current T x Competition | 0.8302 | 1 | 5.0913 | 0.0271\*\* |
| Evolved T x Current T x Competition | 0.1287 | 1 | 0.7895 | 0.3772 |
| Residuals | 11.74 | 72 |  |  |

Table 2a. Effects of current temp and competition on protist growth rate. Two-factor type III ANOVA. Data are log-transformed to meet test assumptions.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Predictor | SS | df | *F* | *p* |
| Current temperature | 6.2274 | 1 | 30.9951 | <0.001\*\*\* |
| Competition | 0.6339 | 2 | 1.5774 | 0.2159 |
| Current T x Competition | 2.1513 | 2 | 5.3536 | 0.0076\*\* |
| Residuals | 10.8495 | 54 |  |  |

Table 2b. Comparisons among competition treatment effects on log-transformed protist growth rate. All p-values are Tukey’s HSD-adjusted.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Current T | Comparison | *estimate* | SE | *df* | *t* | *p* |
|  | control - rotif25 | -0.3453 | 0.2005 | 54 | -1.7224 | 0.2062 |
| 25 °C | control - rotif30 | -0.248 | 0.2005 | 54 | -1.2369 | 0.4369 |
|  | rotif25 - rotif30 | 0.0973 | 0.2005 | 54 | 0.4854 | 0.8785 |
|  | control - rotif25 | 0.406 | 0.2005 | 54 | 2.0253 | 0.1159 |
| 30 °C | control - rotif30 | 0.5989 | 0.2005 | 54 | 2.9879 | 0.0116\* |
|  | rotif25 - rotif30 | 0.1929 | 0.2005 | 54 | 0.9625 | 0.6035 |

i. Evolution of temperature response in rotifers (*r*)

The growth rate of rotifers did not differ between the two evolutionary histories of temperature (F1,72 = 0.469, p = 0.677; Table 1).

ii. Rotifer competitive response (*r*)

Competition did affect rotifer growth rate, but was dependent on current temperature (*F*1,72 = 5.091, *p* = 0.0271; Table 1). Growth rate was nearly doubled (98% higher, back-transformed ES) in the absence of a competitor at 30 °C (*estimate* = 0.682 ± 0.13 SE, *t*72 = 5.34, *p* < 0.001) but competition had only a weak effect on growth rate at 25C (*estimate* = 0.27 ± 0.13 SE, *t*72 = 2.08, *p* = 0.041). In the absence of competition, protists grew faster (51%, back-transformed ES) at 30 °C than at 25 °C (*estimate* = 0.41 ± 0.13 SE, *t*72 = 3.23, *p* = 0.002), whereas temperature had no detectable effect when a competitor was present (*p* = 0.98).

iii. Rotifer competitive effect (*r* of protists)

Competition affected the growth rate of protists, but the effect was dependent on current temperature (*F*1,54 = 5.354, *p* = 0.008; Table 2a). Post-hoc comparisons using Tukey’s HSD-adjusted p values of the levels of competition (competition with rotifers from each evolutionary history, and a control without rotifers) show protist growth rate was further dependent on the evolutionary history of their rotifer competitors. At a current temperature of 30 °C, protist growth rate was about 46% lower (back-transformed data) in the presence of rotifers, but only if the rotifers evolved at 30 °C (*estimate* = 0.599 ± 0.200 SE, *t*54 = 2.988, *p* = 0.0116). At the same current temperature of 30 °C, rotifers with an evolutionary history of 25 °C did not have an effect on protist growth rate (*estimate* = 0.406 ± 0.200 SE, *t*54 = 2.025, *p* = 0.1159). Protist growth rate was unaffected by the presence of competitors when the current temperature was 25 °C (all *p* > 0.05, Table 2b).

2. Carrying capacity (*K*)

Table 2a. Rotifers: three-factor ANOVA of *K*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Predictor | df | SS | MS | F | p |  |
| evolved temp | 1 | 599.8 | 599.8 | 4.2537 | 0.04277 | \* |
| current temp | 1 | 522.1 | 522.1 | 3.7028 | 0.05827 | . |
| competition | 1 | 6211.0 | 6211.0 | 44.0512 | <0.001 | \*\*\* |
| evol:current | 1 | 20.4 | 20.4 | 0.1450 | 0.70449 |  |
| evol:comp | 1 | 144.1 | 144.1 | 1.0223 | 0.31537 |  |
| current:comp | 1 | 20.3 | 20.3 | 0.1442 | 0.70525 |  |
| evol:current:comp | 1 | 1.5 | 1.5 | 0.0106 | 0.91811 |  |
| residuals | 72 | 10151.6 | 141.0 |  |  |  |

Table 2b. Protists: two-factor ANOVA of *K*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Predictor | df | SS | MS | F | p |  |
| current temp | 1 | 11129 | 11129 | 0.4755 | 0.49341 |  |
| competition | 2 | 327742 | 163871 | 7.0021 | 0.00198 | \*\* |
| current:comp | 2 | 802975 | 401488 | 17.1554 | <0.001 | \*\*\* |
| residuals | 54 | 1263761 | 23403 |  |  |  |

i. Evolution of temperature response in rotifers (*K*)

Evolved temperature affected the carrying capacity of rotifers independent of current temperature and the presence of a competitor (F1,72 = 4.254, p = 0.043; Table 2a). The carrying capacity of rotifers that evolved at 25C was 9% higher than those that evolved at 30C (estimate = 5.48 ± 2.66, t72 = 2.062, p = 0.0428).

ii. Rotifer competitive response (*K*)

Competition affected the carrying capacity of rotifers independent of current temperature and evolved temperature (F1,72 = 44.051, p = <0.001; Table 2a). Rotifer carrying capacity was 26% higher in the absence of the protist competitor (estimate = 17.6 ± 2.66, t72 = 6.637, p = <0.001).

iii. Rotifer competitive effect (*K* of protists)

Competition affected the carrying capacity of protists, but the effect was dependent on current temperature (F1,54 = 17.1554, p = <0.001; Table 2b). Protist carrying capacity was 36% lower in the presence of rotifer competitors, but only at a current temperature of 25C (estimate = -395.9 ± 59.2, t54 = -6.681, p = <0.001). The carrying capacity of protists was unaffected by the evolutionary history of their rotifer competitors (p = >0.05).

Supplemental Table 1. EMMs from a two-factor ANOVA of ln-transformed protist *r*. No competition = “no comp", rotifers evolved at 25C = “rotif25”, rotifers evolved at 30C = “rotif30”. All *p-*values are Tukey’s HSD-adjusted.

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Current temp | | Contrast | Estimate | | SE | Df | t | p |  |
| 25C | noComp – rotif25 | | -0.326 | 0.207 | 54 | -1.575 | 0.2653 |  |
| noComp – rotif30 | | -0.215 | 0.207 | 54 | -1.039 | 0.5555 |  |
| rotif25 – rotif30 | | 0.111 | 0.207 | 54 | 0.535 | 0.8544 |  |
| 30C | noComp – rotif25 | | 0.404 | 0.207 | 54 | 1.953 | 0.1339 |  |
| noComp – rotif30 | | 0.577 | 0.207 | 54 | 2.789 | 0.0196 | \* |
| rotif25 – rotif30 | | 0.173 | 0.207 | 54 | 0.836 | 0.6823 |  |