

² Supplementary Information for

- ³ Changes in temperature alter the relationship between biodiversity and ecosystem
- 4 functioning

1

5 Francisca C. García, Elvire Bestion, Ruth Warfield, Gabriel Yvon-Durocher

- ⁶ Francisca C. Garcia.
- 7 E-mail: f.garcia-garcia@exeter.ac.uk

8 This PDF file includes:

- ⁹ Supplementary text
- ¹⁰ Figs. S1 to S6
- 11 Tables S1 to S12

Supporting Information Text 12

1. SI Materials and Methods 13

1.1. Identification of bacteria isolates. We identified the different isolates using 16S Polymerase Chain Reaction (PCR). A 15 master-mix solution was prepared using 7.2 μ l of DNA free water, 0.4 μ l 27 forward primer, 0.4 μ l 1492 reverse primer and 10 μ l of Taq polymerase per sample. To create a template solution 2 μ l of sample 100 x diluted in DNA free water was added to 16 $18 \ \mu$ l of master-mix solution. Samples were then placed in a thermal cycler (Applied Biosystems Veriti Thermal Cycler). This 17 procedure included 1 cvcle at 94 °C for 4 minutes, 35 cvcles at 94, 48 and 72 °C for 1 minute, 30s and 2 minutes, respectively, 18 and finally, 1 cycle at 72 °C for 8 minutes. The PCR product was cleaned up using Exonuclease I and Antartic Phosphatase 19 and high quality samples were Sanger sequenced using the 27F, 1492R primers (Core Genomic Facility, University of Sheffield). 20 Sequences were trimmed in Genious (version 6.1.8) removing the bp from the 5' end and trimming the 3' end to a maximum 21

length of 1000bp. 22

14

1.2. Species-levels thermal tolerance curves. Prior to characterising the growth curve, each species was acclimated for 24 h 23 at each temperature. The acclimated strains were then inocculated at a common density into a 96-well plate with 200 μ L 24 of protozoan media. Plates were stored inside of plastic boxes with water soaked sponges to maintain humidity during the 25 experiment and avoid changes in the volume due to evaporation. The optical density of each replicate was measured every 26 2 hours using a Biotek reader synergy 2 at 600 nm wavelength and measurements were corrected with a blank (to account 27 for particulate organic matter in the protozoa medium). The optical density (OD) was monitored until each species reached 28 carrying capacity. 29

Population growth rates $(r (h^{-1}))$ and carrying capacities (OD_{600}) were quantified by fitting the logistic growth equation to 30 the time-series of biomass measurements using non-linear least squares regression: 31

$$N_{t} = \frac{K}{1 + Ae^{-rt}}; A = \frac{K - N_{0}}{N_{0}}$$
[1]

where N_t is the biomass (OD₆₀₀) at time, t, K is the carrying capacity (OD₆₀₀), N_0 is the biomass at the start of the 33 experiment and r is the rate of exponential population growth (h^{-1}) . The thermal tolerance curve was quantified by fitting 34 the four parameter Sharpe-Schoolfield equation to the mean population growth rate (across the 6 replicates at each assay 35 temperature) measured along the thermal gradient for each taxa: 36

$$\ln(r(T)) = Ea(\frac{1}{kT_{\rm c}} - \frac{1}{kT}) + \ln(r(T_{\rm c})) - \ln(1 + e^{E_{\rm h}(\frac{1}{kT_{\rm h}} - \frac{1}{kT})})$$
[2]

where r(T), is population growth rate (h⁻¹), k is Boltzman's constant (8.62*10⁻⁵ eV K⁻¹), Ea is the activation energy 38 (in eV) that characterises the steepness of the slope leading up to a thermal optimum, T is temperature in Kelvin (K), $E_{\rm h}$ 39 characterizes temperature-induced inactivation of growth above $T_{\rm h}$, the temperature where half the enzymes are rendered non 40 functional and $r(T_c)$ is the rate of growth normalized to an arbitrary reference temperature, $T_c = 18$ °C, where no low or high 41 temperature inactivation is experienced. Equation (2) yields a maximum growth rate at an optimum temperature: 42

32

37

$$T_{\rm opt} = \frac{E_{\rm h} T_{\rm h}}{E_{\rm h} + k T_{\rm h} \ln(\frac{E_{\rm h}}{E_{\rm a}} - 1)}$$
[3]

The four-parameter Sharpe-Schoofield model assumes that there is a single rate limiting enzymatic reaction that is reversibly 44 inhibited by temperature. The parameters of the equation $(r(T_c), E_a, E_h, T_h, and T_{opt})$ characterize the thermal tolerance 45 curve. We consider these parameters to be thermal "traits" that play a key role in shaping the impacts of temperature change 46 on community dynamics and ecosystem functioning (Fig. S6, Table S1). 47

1.3. Biodiversity ecosystem functioning experiment. The optical density of each community at each temperature level was 48 49 measured every 2 hours using a Biotek reader synergy 2 at 600 nm wavelength and was monitored until each microcosm reached carrying capacity. In all microcosms, biomass increased exponentially and then reached a stationary phase. Ecosystem 50 functioning was quantified as the asymptotic biomass (i.e. the yield) of the community in the stationary phase, determined by 51 fitting the logistic growth equation to the biomass time-series. For richness = 1, the average yield (across the 6 replicates) of 52 each of the 24 taxa grown in monoculture at each assay temperature and characterised in the "species-level thermal tolerance 53 54 curves", were used.

The experiment structure was well defined by a power function: 55

56

$$\log_{10} Y(S) = b(\log_{10} S - \log_{10} S_{\rm c}) + \log_{10} Y(S_{\rm c})$$
^[4]

where $\log_{10} Y$ is the natural logarithm of ecosystem function (here yield) at a given level of species richness, S, b is the 57 exponent, that captures the shape of the diversity functioning relationship, $(\log_{10} S - \log_{10} S_c)$, is the natural log transformed 58 centred species richness. Centring the species richness variable means that the intercept, $\log_{10} Y(S_c)$, gives the yield at an 59 average level of species richness, and decreases the correlation between the exponent and the intercept. The exponent can 60 indicate that ecosystem function is either a decellerating function of species richness when b < 1 (i.e. there is some degree of 61 functional redundancy) or that functioning increases linearly with richness when b = 1 (i.e. there is no functional redundancy 62

and each species has a positive contribution to ecosystem functioning). We quantified the effects of warming on the relationship 63 between biodiversity and ecosystem functioning by fitting Eq. (4) to the experimental data using a linear mixed effects model 64 via the "lmer" function in the "lme4" package of R software. We included "temperature" as a fixed categorical factor to 65 assess how b and $\log_{10} Y(S_c)$ varied among the various temperature treatments. Because our experimental design entailed 66 exposing each unique randomly assembled community at each level of species richness to the 8 temperature treatments, we 67 included "community ID" nested within "richness" as a random effect on the intercept to account for non-independence among 68 measurements made on the same communities across temperature treatments. To quantify the significance of differences in the 69 parameters among all pairwise combinations of the 8 temperature treatments, we used post-doc Tukey tests via the "glht" 70

⁷¹ function in the "multcomp" package for R statistical software (see Table S9).

1.4. Re-isolation of species. At the end of the biodiversity ecosystem functioning experiment we re-isolated the bacterial taxa from maximum richness treatments (S = 24) exposed to the ambient (20 °C) and the two extreme temperatures (10 & 40 °C). Samples from each community were prepared by spreading 10 μ L serial dilutions onto R2 agar plates and incubated at ambient temperature (20 °C). A total of 2512 colonies were identified across all replicates and counted by visual inspection. Of these, 121 colonies were identified using 16S rRNA Sanger sequencing (see section 1.1) to confirm the taxonomic identity based on colony morphology.

1.5. Linking thermal traits to the impacts of warming and species loss on ecosystem functioning. In this analysis, community 78 yield was treated as the response variable, $\langle T_{opt} \rangle_c$ was continuous predictor and temperature (10, 15, 20, 25, 27.5, 30, 35, 30, 35, 30, 35). 79 40° C) and species richness (S = 2, 4, 8, 16) were treated as categorical factor variables. To account for the hierarchical nature 80 of our experimental design we included "community ID" nested within "richness" as a random effect on the intercept. Note, 81 we did not include the maximum level of species richness, S = 24, because there was no variation in $\langle T_{opt} \rangle_c$ as each of the 82 10 replicates received an identical set of taxa. We started with the most complex model which included all variables and 83 their interactions and carried out model simplification by sequentially removing terms and testing for their significance using 84 likelihood ratio tests. Model simplification was stopped when all terms were significant at p < 0.05 (Table S8). 85

1.6. Partitioning the net biodiversity effect into selection and complementarity. Using information on the relative abundance of species isolated from the high diversity treatments at 10, 20 and 40 °C, the net effect of biodiversity was calculated as the difference between the observed yield and the expected (weighted average yield of the monocultures). The complementarity effect was estimated as $\overline{N\Delta RYM}$, where N is the number of species, $\overline{\Delta RY}$ is the average change in the relative yield for all species in the mixture and \overline{M} is the average monoculture yield. The selection effect was calculated as the covariance between the monoculture yield of species and their change in relative yield in the mixture (Ncov(ΔRY , M)).

Hence, NE, SE and CE were estimated at a local scale for each replicate (10 replicates per temperature) and the total effects were estimated for each temperature treatment (10, 20 and 40 °C) (Table S5). An analysis of variance (Anova) was performed to test for significant differences among temperature treatments for each mechanism (i.e. NE, SE, CE) (Table S10). To determine whether differences among all pairwise temperature levels were significant, we used Tukey-Kramer post hoc tests (Table S11). We additionally test whether NE, SE and CE estimated for each temperature treatment were significantly different

⁹⁷ from zero using a *t*-tests (Table S12).



Fig. S1. Temperature-driven selection of community composition linked to thermal tolerance traits. At the end of the diversity experiment we re-isolated the bacterial taxa from maximum richness treatments (S = 24) that had been exposed to the ambient ($20 \degree C$) and the two extreme temperatures ($10 \& 40 \degree C$). The red box represents the range of optimal temperatures of the 24 taxa and the blue box represents the optimal temperature of the taxa recovered at the end of the experiment. We found that the presence of taxa in the different temperature treatments was strongly associated with their thermal tolerance curves, with those present in the 40 °C treatments being species with the highest T_{opt} . Notably no taxa with $T_{opt} < 27 \degree C$ were present at the end of experiments. These results demonstrate that "selection effects" played an important role in determining the effects of temperature community composition that likely influenced the diversity-functioning relationship. Point size corresponds to the number of colonies of a determined taxa that appeared at each incubation temperature.



Fig. S2. Linking thermal traits to the impacts of warming and species loss on ecosystem functioning. Coupling between ecosystem functioning (community yield) and the community-mean optimum temperature $\langle T_{opt} \rangle_c$ at different richness levels from 2 to 16. Analyses reveal that $\langle T_{opt} \rangle_c$ becomes an increasingly important predictor of ecosystem function as species richness increases, highlighting a strong positive interaction between biodiversity and mean optimum temperature. The red lines represent the fitted curves derived from the linear mixed effect model. The different point shapes represents the level of temperature (T).



Fig. S3. Covariation between monoculture yields and mixture relative biomass. For species we assess the correlation between monoculture yield and the relative abundance of each species in the 24 species mixtures at the ambient (20 °C) and the two extreme temperatures (10 & 40 °C). Grey lines represent the fit of a linear model. Dashed grey lines represent cases where we obtained a positive slope (7 of the 24 species) and indicates that there is dominance by species in the mixture for those temperatures at which the species growth in monoculture was optimal.



Fig. S4. Temperature changes the nature of species interactions. For 10 randomly assembled pairwise combinations of species we compare the yield of the mixture (red circles) to the yield of each species in monoculture (black and grey circles) across the thermal gradient. We see substantial variation in the outcome (i.e. which species dominates) and the nature (i.e. whether mixture yield exceeds monoculture yield) across the thermal gradient. These results imply that temperature change altered species interactions in the biodiversity-ecosystem functioning experiment. Error bars represent the standard deviation of the yield across 6 replicates of each species in monoculture.



Fig. S5. Overview of the study site and isolation process. a: Map of Iceland showing geographic location of the field site, Hvergerdi Valley, 45 km east of Reykjavik. b: Map of the drainage basin in the Hvergerdi Valley, identifying the streams from which taxa were isolated. c: Phylogenetic tree with the 24 different strains isolated from the streams (see table S1 for taxonomic and associated information of each taxa). The colour code represents the temperature for the streams.



Fig. S6. Thermal tolerance curves for each isolate fitted using the Sharpe-Schoofield model. Y-axis represents the average natural log-tansformed population growth rate (lnr) quantified from the logistic growth curves of 6 replicates at each assay temperature. X-axis represents the growth temperature.

Isolate ID	$\ln(r(T_{\rm c}))$	$E_{\mathbf{a}}$	$E_{\rm h}$	$T_{\rm h}$	quasi r 2	$T_{\rm opt}$
lc1B(b)	-0.34	0.50	1.87	309.89	0.92	32.32
lc4(c)	-0.22	0.29	3.00	313.64	0.91	34.31
lcV(d)	0.07	0.75	1.29	297.66	0.25	26.42
lc1(e)	-0.22	0.40	1.61	304.64	0.74	26.15
lc10(f)	-0.23	0.29	3.68	311.20	0.77	32.58
lc5(g)	-0.42	0.21	2.45	307.87	0.89	27.1
lc1A(h)	0.09	0.88	2.94	298.74	0.91	23.4
lc4(i)	-0.37	0.25	2.60	308.14	0.86	28.07
lc5(j)	-0.46	0.32	3.43	310.78	0.76	32.17
lc11A(k)	-0.57	0.49	2.04	303.39	0.79	25.86
lc1B(l)	-0.63	0.56	2.28	308.53	0.59	31.37
lc4(m)	-0.30	0.47	2.18	306.30	0.63	28.47
lc7(n)	-0.29	0.32	2.37	309.94	0.39	30.46
lc5(o)	0.41	0.43	3.17	300.69	0.74	23.08
lc5(p)	-0.12	0.38	3.80	310.58	0.76	32.67
lc5(q)	-0.64	0.42	3.66	310.98	0.77	33.26
lc7(r)	-0.58	0.24	2.30	311.02	0.30	30.23
lc11B(s)	-0.26	0.60	2.91	301.98	0.83	25.21
lc11B(t)	-1.23	0.40	2.27	308.88	0.84	30.27
lcV(u)	-0.44	0.38	3.18	315.00	0.97	36.59
lcV(v)	-0.43	0.42	2.94	313.55	0.92	35.32
lc1A(w)	-0.20	0.64	2.00	297.73	0.89	21.71
lc1(x)	-0.01	0.54	1.94	301.12	0.32	24.15
lc1A(y)	-0.04	0.36	3.45	309.47	0.82	31.26
Fit all sps	-0.01	0.60	1.00	298.84	0.92	28.76

Table S1. Thermal tolerance traits for the 24 isolates

Data shown are the parameters obtained from the Schoofield model and characterize the thermal tolerance curve for each taxa and an overall thermal tolerance curve derived by aggregating the growth rate data across species for each temperature (Fig. 1c). Isolate ID is the identity of each isolated taxa (see Table S1). $\ln(r(T_c))$ is the rate of growth normalized to an arbitrary reference temperature, $T_c = 18$ °C, where no low or high temperature inactivation is experienced. E_a is the activation energy (in eV) that characterizes the steepness of the slope leading to a thermal optimum. E_h characterizes temperature-induced inactivation of growth above T_h , the temperature where half the enzymes are rendered non functional. The quasi r^2 represents

the quality of the model fitting and T_{opt} is the optimal temperature at which the maximum growth rate is reached.

Table S2. Details of the linear mixed effects model analysis for the biodiversity - ecosystem functioning relationship.

Model	d.f.	AIC	Log Lik	Chisq	р
Random effects structure					
random = $\sim 1/Replicate$					
Fixed effects structure					
1.log $_{10}Y \sim$ 1 + log $_{10}S$ * T	19	-1055.15	546.57	108.11	< 0.001 * **
$2.\log_{10}Y \sim 1 + \log_{10}S + T$	12	-961.04	492.52		

Random effects on intercept were determined at the level of replicates nested within richness level. The results of the model selection procedure on the fixed effect terms are given and the most parsimonious model is model 1 (p < 0.01). Analyses reveal that ecosystem functioning (log_{10} Y) changed significantly through the different richness levels ($log_{10}S$) and that the exponent and intercept were significantly different between selection temperatures. The degrees of freedom (d.f.), AIC, logLik, Chi square (Chisq) and p-value are given.

Table S3. Results from the pairwise comparison of the Permutational Multivariate Analysis of Variance (PERMANOVA) to test the effect of temperature on community composition.

Temperature	F	R^2	p-value	p.adjusted
10-20	4.56	0.21	0.010	0.030*
10-40	9.22	0.34	0.001	0.003 * **
20-40	19.75	0.54	0.001	0.003 * **

The table shows the parameters obtained from the pairwise Permutational Multivariate Analysis of Variance (PERMANOVA) comparison to compare whether there is significant differences between all pairwise combinations of the temperature treatments (10, 20 and 40 °C). p is the p-value and p.adjusted is the adjusted p value using Bonferroni correction.

Table S4. Results from the Permutational Multivariate Analysis of Variance (PERMANOVA) analysis to test the effect of temperature on community composition

Parameter	d.f.	Sum of Sqs	Mean Sqs	F-value	R^2	p-value
Temperature	1	2.76	2.76	10.00	0.27	0.001 * **
Residuals	27	7.44	0.28		0.73	
Total	28	10.20			1.00	

The table shows the parameters obtained from Permutational Multivariate Analysis of Variance (PERMANOVA) test to analyse whether there is significant differences between the species composition at each temperature treatments (10, 20 and 40 °C).

		Temperature treatments				
		10	20	40		
Biodiversity effects						
NBE	$\sum_{k}^{P} \sum_{j}^{T} \sum_{i}^{N} \Delta R_{ijk} Y_{ijk} [5]$	0.50	0.16	0.38		
ТС	$PTN\overline{\overline{\Delta RY}}$ [6]	0.43	0.12	0.24		
TS	$PTNcov(\Delta RY_{ijk}, M_{ijk})$ [7]	0.07	0.06	0.13		
local <nbe></nbe>	$\sum_{i}^{N} \Delta R_{i} Y_{i} [8]$	0.050	0.018	0.038		
local <ce></ce>	$N\overline{\Delta}\overline{RYM}$ [9]	0.043	0.012	0.024		
local <se></se>	$Ncov(\Delta RY_i, M_i)$ [10]	0.007	0.006	0.014		

Table S5. Local scale and total biodiversity partitions.

NBE is the net biodiversity effect, TC is the total complementarity, TS is the total selection effect, local $\langle NBE \rangle$ is the average local net biodiversity effect, local $\langle CE \rangle$ is the average local complementarity effect and local $\langle SE \rangle$ is the average local selection effect. $\overline{\Delta RY}$ is the average change in the relative yield for all species in the mixture and M is the monoculture yield, i is the indexes of one of the number of species (N = 24), j is the indexes of one of T times (T = 1), k is the indexes of one of P places (P = 10).

Isolate ID	Genus	Family	Phylum	Stream	Temp stream	Accession number
lc1B(b)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	1B	8.2	MH801968
lc4(c)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	4	7.3	MH801970
lcV(d)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	vent	38	MH801982
lc1(e)	Chryseobacterium sp	Flavobacteriaceae	Bacteroidetes	1	16.5	MH801963
lc10(f)	Chryseobacterium sp	Flavobacteriaceae	Bacteroidetes	10	16.5	MH801979
lc5(g)	Chryseobacterium sp	Flavobacteriaceae	Bacteroidetes	5	16.5	MH801973
lc1A(h)	Janthinobacterium sp	Janthinobacteriaceae	Betaproteobacteria	1A	8	MH801965
lc4(i)	Erwinia sp	Enterobacteriaceae	Gammaproteobacteria	4	7.3	MH801971
lc5(j)	Hebaspirillum sp	Betaproteobacteria	Oxalobacteriaceae	5	26.9	MH801976
lc11A(k)	Flavobacterium sp	Flavobacteriaceae	Bacteroidetes	11A	14.4	MH801981
lc1B(l)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	1B	8.2	MH801969
lc4(m)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	4	7.3	MH801972
lc7(n)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	7	11.4	MH801974
lc5(o)	Chryseobacterium sp	Flavobacteriaceae	Bacteroidetes	5	26.9	MH801975
lc5(p)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	5	26.9	MH801977
lc5(q)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	5	26.9	MH801978
lc7(r)	Aeromonas sp	Aeromonadaceae	Gammaproteobacteria	7	11.4	MH801980
lc11B(s)	Buttiauxella sp	Gammaproteobacteria	Enterobacteriaceae	11B	17.2	MH801983
lc11B(t)	Bacillus sp	Bacillaceae	Bacilli	11B	17.2	MH801984
lcV(u)	Acinetobacter sp	Moraxellaceae	Gammaproteobacteria	vent	38	MH801985
lcV(v)	Acinetobacter sp	Moraxellaceae	Gammaproteobacteria	vent	38	MH801986
lc1A(w)	Pseudomonas sp	Gammaproteobacteria	Pseudomonaceae	1A	8	MH801964
lc1(x)	Pseudomonas sp	Gammaproteobacteria	Pseudomonaceae	1	16.5	MH801967
lc1A(y)	Pseudomonas sp	Gammaproteobacteria	Pseudomonaceae	1A	8	MH801966

Table S6. List of bacterial isolates used in the experiment.

Isolate identification codes, taxonomic information, stream ID, average daily water temperature at the time of sampling and GenBank accession numbers are given.

Table S7. Experimental design

Richness	Replicate	Isolates ID	Vol (μ l)
S2	1	c,t	12
S2	2	x,h	12
S2	3	f,b	12
S2	4	f,p	12
S2	5	b,y	12
S2	6	k,t	12
S2	7	k,j	12
S2	8	h,l	12
S2	9	l,n	12
S2	10	q,d	12
<i>S</i> 4	1	d,p,o,w	6
S4	2	v,p,b,f	6
S4	3	q,m,y,x	6
S4	4	h,w,y,s	6
<i>S</i> 4	5	h,y,f,w	6
S4	6	i,h,e,b	6
<i>S4</i>	7	g,t,m,u	6
<i>S</i> 4	8	u,c,l,g	6
<i>S</i> 4	9	i,m,e,q	6
<i>S4</i>	10	f,g,w,r	6
<i>S8</i>	1	v,h,y,t,b,k,i,r	3
<i>S8</i>	2	u,d,f,v,w,l,q,p	3
<i>S8</i>	3	v,r,w,m,g,u,i,h	3
<i>S8</i>	4	k,y,l,q,h,o,r,c	3
<i>S8</i>	5	j,o,w,g,n,s,u,f	3
<i>S8</i>	6	l,o,b,f,j,r,v,y	3
<i>S8</i>	7	b,s,p,q,o,v,t,g	3
<i>S8</i>	8	i,t,j,b,g,d,c,w	3
<i>S8</i>	9	n,l,u,e,x,y,k,j	3
<i>S8</i>	10	j,d,c,t,k,m,g,o	3
S16	1	h,j,n,u,f,s,t,m,l,b,d,o,x,v,i,q	1.5
S16	2	s,x,j,r,t,f,m,d,u,g,b,q,l,e,n,p	1.5
S16	3	m,f,t,p,q,d,o,h,s,k,l,i,r,j,b,x	1.5
S16	4	s,q,l,t,j,f,c,y,g,i,k,x,w,e,v,d	1.5
S16	5	q,g,l,r,c,v,h,p,x,y,s,m,w,f,i,	1.5
S16	6	l,r,j,h,q,e,n,d,t,x,u,b,i,k,m,y	1.5
S16	7	l,k,s,n,o,h,f,r,y,e,c,t,m,v,x,q	1.5
S16	8	j,k,e,b,p,c,y,l,q,i,h,d,x,f,g,t	1.5
S16	9	g,o,n,c,b,w,r,l,j,i,x,h,q,t,d,k	1.5
S16	10	s,l,e,q,d,r,m,k,g,h,i,u,p,b,n,	1.5
S24	1	all 24 sps	1
S24	2	all 24 sps	1
S24	3	all 24 sps	1
S24	4	all 24 sps	1
S24	5	all 24 sps	1
S24	6	all 24 sps	1
S24	7	all 24 sps	1
S24	8	all 24 sps	1
S24	9	all 24 sps	1
S24	10	all 24 sps	1

The table shows the experimental design of the diversity experiment. Richness represents the number of species assembled. Replicate identifies each of the 10 independent communities at each diversity level. Isolate IDs, give the identity of each isolate assembled into each community (we have only used the letter in brackets of the isolates ID in order to simplify the experimental design, see table S1 for further information about the bacterial isolates) and, finally, the volume describes the volume of media for each isolate added to each of the communities.

Table S8. Model selection results linking the species thermal traits to the observed ecosystem functioning in the biodiversity experiment

Random effects structure						
random = \sim 1S/Replicate	Parameters	d.f.	AIC	logLik	Chisq	p-value
Fixed effects structure	Model 3	24	-2043.1	1045.5		
Model 1 Y \sim $<\!T_{ m opt}\!>_{ m c}$ * T * S	Model 2	45	-2099.1	1094.6	98.11	< 0.001 * **
Model 2 Y $\sim < T_{opt} >_c + T + S + < T_{opt} >_c:T + < T_{opt} >_c:S + T:S$	Model 1	66	-2087.7	1109.8	30.59	0.08
Model 3 Y $\sim < T_{opt} >_c + T + S + < T_{opt} >_c :T + < T_{opt} >_c :S$						

We used a linear mixed effects model to assess whether the community-mean optimum temperature, $\langle T_{opt} \rangle_c$, was a significant predictor of ecosystem functioning across the various temperature (T) and diversity levels (S). To test for the significance of all terms in the model we sequentially removed terms from the most complex model that included all variables and their interactions using likelihood ratio tests. We found that the most parsimonious model included all variables and all two-way interactions, but did not include the 3-way interaction between $\langle T_{opt} \rangle_c$: T : S.

Table S0 Multiple comp	arieon contract analycic for the	nower function between biodi	versity and ecceveter function
Table 33. Multiple compa	anson contrast analysis for the	power function between blou	versity and ecosystem function

Estimate	SE	t value	p-value
-1.33	0.02	-80.74	< 0.01 * **
-1.36	0.02	-82.35	< 0.01 * **
-1.36	0.02	-82.60	< 0.01 * **
-1.36	0.02	-82.40	< 0.01 * **
-1.39	0.02	-84.34	< 0.01 * **
-1.38	0.02	-84.34	< 0.01 * **
-1.44	0.02	-86.70	< 0.01 * **
-0.03	0.01	-1.78	0.86
-0.03	0.01	-1.96	0.75
-0.03	0.01	-1.83	0.83
-0.06	0.01	-3.98	< 0.01 * *
-0.05	0.01	-3.06	0.09
-0.11	0.02	-7.39	< 0.01 * **
-0.003	0.01	-0.18	1.00
-0.001	0.01	-0.05	1.00
-0.03	0.01	-2.20	0.56
-0.02	0.01	-1 29	0.99
-0.08	0.02	-5.63	< 0.01 * **
0.001	0.01	0.12	1 00
-0.03	0.01	-2.03	0.70
-0.05	0.01	-2.00	1.00
-0.02	0.01	-5.46	۲.00 ح ∩ ∩1 × ××
-0.00	0.02	-5.40	0.61
-0.03	0.01	-2.10	0.01
-0.02	0.01	-1.23	0.99
-0.08	0.02	-0.01	< 0.01 * **
0.01	0.01	0.91	1.00
-0.05	0.02	-3.45	0.03*
-0.07	0.02	-4.34	< 0.01 * **
0.22	0.03	7.00	< 0.01 * **
0.13	0.03	4.21	< 0.01 * **
0.19	0.03	5.94	< 0.01 * **
0.19	0.03	5.82	< 0.01 * **
0.18	0.03	5.71	< 0.01 * **
0.30	0.03	9.53	< 0.01 * **
0.33	0.03	10.18	< 0.01 * **
-0.09	0.03	-3.27	0.05*
-0.03	0.03	-1.27	0.99
-0.04	0.03	-1.38	0.98
-0.04	0.03	-1.51	0.96
0.08	0.03	3.00	0.10
0.11	0.03	3.84	< 0.01 * *
0.05	0.03	2.01	0.71
0.05	0.03	1.89	0.80
0.05	0.03	1.75	0.87
0.17	0.03	6.23	< 0.01 * **
0.20	0.03	7.02	< 0.01 * **
-0.003	0.03	-0.11	1.00
-0.007	0.03	-0.25	1.00
0.11	0.03	4.28	< 0.01 * **
0.14	0.03	5.10	< 0.01 * **
-0.004	0.03	-0.13	1.00
0.12	0.03	4.37	< 0.01 * **
0.15	0.03	5.18	< 0.01 * **
0.12	0.03	4,50	< 0.01 * **
0.15	0.03	5.31	< 0.01 * **
0.03	0.03	0.88	1.00
	Estimate -1.33 -1.36 -1.36 -1.39 -1.38 -1.44 -0.03 -0.03 -0.03 -0.06 -0.05 -0.11 -0.03 -0.02 -0.08 0.001 -0.03 -0.02 -0.08 0.001 -0.03 -0.02 -0.08 0.001 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.03 -0.02 -0.08 0.01 -0.05 -0.07 0.22 0.13 0.19 0.03 -0.007 0.02 0.03 -0.04 -0.04 0.05 0.05 0.05 0.17 0.20 -0.003 -0.007 0.11 0.19 0.19 0.19 0.19 0.19 0.19 0.19 0.19 0.19 0.19 0.19 0.19 0.11 0.19 0.19 0.19 0.19 0.19 0.02 -0.03 -0.04 -0.04 0.05 0.05 0.17 0.20 -0.003 -0.007 0.11 0.12 0.15 0.12 0.15 0.03	Estimate SE -1.33 0.02 -1.36 0.02 -1.36 0.02 -1.36 0.02 -1.37 0.02 -1.38 0.02 -1.39 0.02 -1.38 0.02 -1.38 0.02 -1.38 0.02 -1.38 0.02 -0.03 0.01 -0.03 0.01 -0.03 0.01 -0.05 0.01 -0.05 0.01 -0.01 0.01 -0.02 0.01 -0.03 0.01 -0.03 0.01 -0.03 0.01 -0.02 0.01 -0.03 0.01 -0.04 0.02 -0.05 0.02 -0.07 0.02 -0.07 0.02 -0.07 0.02 -0.07 0.02 -0.07 0.02 -0.07 0.02 <	Estimate SE t value -1.33 0.02 -80.74 -1.36 0.02 -82.35 -1.36 0.02 -82.60 -1.37 0.02 -84.34 -1.38 0.02 -84.34 -1.38 0.02 -84.34 -1.38 0.02 -84.34 -1.38 0.02 -84.34 -1.38 0.02 -86.70 -0.03 0.01 -1.78 -0.03 0.01 -1.83 -0.05 0.01 -3.98 -0.05 0.01 -3.06 -0.11 0.02 -7.39 -0.003 0.01 -0.18 -0.01 0.01 -0.12 -0.02 0.01 -1.29 -0.03 0.01 -2.03 -0.02 0.01 -1.12 -0.08 0.02 -5.63 0.01 0.11 1.23 -0.02 0.01 -1.12 -0.03

Data shown are the contrast analysis for the biodiversity-ecosystem functioning relationship. This analysis tests whether exponent (b) and the intercept $(\log_{10} Y(S_c))$ are significantly different between all pairwise combinations of temperature treatments. The standard error (SE), t value and p-value were estimated for all possible pairwise combinations are given.

Table S10. Results from the analysis of variance (ANOVA) on the local scale biodiversity partitions.

	Net Effect					Sel effect				Comp effect					
Resp	d.f.	SumSqs	MeanSqs	F	р	d.f.	SumSqs	MeanSqs	F	р	d.f.	SumSqs	MeanSqs	F	р
Temp	2	0.005	0.002	207.6	< 0.001	2	$3^{*}10^{-4}$	$1.64^{*}10^{-4}$	3.1	0.06	2	0.005	0.002	48.8	< 0.001
Res	26	$3*10^{-4}$	1*10 ⁻⁵			26	0.001	5.29*10 ⁻⁵			26	0.001	4.8*10 ⁻⁵		

ANOVAs were carried to determine whether the net biodiversity effects (NE), selection effects (SE) and complementarity effects (CE) were significantly different between temperature treatments (10, 20 and 40 °C). Analyses reveals that there is a significant effect of temperature for all biodiversity partitions (p < 0.05) except for selection effects (p = 0.06).

Table S11. Posthoc Tukey tests for the biodiversity partitions.

	Net Effect				Sel effect				Comp effect			
Temperature	diff	lwr	upr	р	diff	lwr	upr	р	diff	lwr	upr	р
20-10	-0.03	-0.04	-0.03	< 0.001 * **	-0.001	-0.01	0.01	0.94	-0.03	-0.04	-0.02	< 0.001 * **
40-10	-0.01	-0.02	-0.01	< 0.001 * **	0.005	-0.002	0.01	0.13	-0.02	-0.03	-0.02	< 0.001 * **
40-20	0.02	0.02	0.02	< 0.001 * **	0.005	-0.003	0.01	0.25	0.01	0.01	0.02	< 0.01 * **

Post hoc Tukey tests were carried to assess whether the net biodiversity effect (NE), the selection effect (SE), and complementarity effect (CE) differed among all pairwise combinations of the temperature treatments. Analyses reveal significant differences across temperature treatments for NE and CE, but not for SE.

Table S12. One sample *t*-test to assess whether the net biodiversity effect, the selection effect and the complementarity effects were significantly different from zero.

			Net Effect				Sel effect				Comp effect	
Temperature	t	df	CI[95%]	р	t	df	CI[95%]	р	t	df	CI[95%]	р
10	49.21	9	0.047 to 0.052	< 0.001 * **	3.45	9	0.003 to 0.012	< 0.01 * *	23.47	9	0.038 to 0.047	< 0.001 * **
20	13.6	8	0.015 to 0.021	< 0.001 * **	4.72	8	0.003 to 0.009	< 0.01 * *	5.71	8	0.007 to 0.016	< 0.001 * **
40	35.90	9	0.035 to 0.040	< 0.001 * **	4.58	9	0.007 to 0.02	< 0.01 * *	9.10	9	0.018 to 0.031	< 0.001 * **

Data shown are the parameters obtained from the t statistical test to analyse whether the net biodiversity effects (NE), selection effects (SE) and complementarity effects (CE) values obtained at the different temperature treatments levels (10, 20 and 40 $^{\circ}$ C) were or not significantly different from zero. t is the t-value, df the degrees of difference, CI the confidence intervale at 96 % and p the p-value. Analyses suggest that there are that there is a positive effects at all temperature treatments for the three mechanisms studied (NE, SE, CE).