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How does temperature affect forest "fungus breath"? Diurnal non-exponential temperature-respiration relationship, and possible longer-term acclimation in fungal sporocarps

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ABSTRACT

Fungal respiration contributes substantially to ecosystem respiration, yet its field temperature response is poorly characterized. I hypothesized that at diurnal time scales, temperature-respiration relationships would be better described by unimodal than exponential models, and at longer time scales both Q_{10} and mass-specific respiration at 10 °C (R_{ms10}) would show signs of acclimation. I measured respiration on intact sporocarps over the course of several days, and modeled temperature-respiration relationships using exponential and unimodal Gaussian functions. Unimodal models provided a better fit than exponential models. R_{ms10} and Q_{10} also declined with increasing temperature, consistent with longer-term temperature acclimation. There was some evidence of diurnal hysteresis. When exponential models were appropriate, Q_{10} values averaged ~3.5, and R_{ms10} averaged 0.02 μmol CO_2 g^{-1} sec^{-1}. The observed high mass-specific respiration rates, peaked temperature responses, decline in R_{ms10} and Q_{10} with increasing temperature, and hysteresis could contribute to observed non-exponential and hysteretic patterns in soil and ecosystem respiration.

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1. Introduction

Fungal contributions to ecosystem respiration could be large, yet are poorly constrained. Assuming (1) 10–25% of microbial biomass is fungal (Fierer et al., 2009), (2) microbial biomass is about 4% of root biomass (Fierer et al., 2009), (3) fungal mass-specific respiration is 10× higher than fine roots (e.g., Andrew et al., 2014), (4) fine roots mass-specific respiration is about 3–10× coarse roots (Desrochers et al., 2002; Chen et al., 2010), and (5) fine roots are about 14% of total root mass (Jackson et al., 1997), then soil fungal respiration would equal from 11 to 86% of total root respiration on a per area basis. This back-of-the-envelope calculation makes it clear that fungal respiration could be a major contributor to ecosystem respiration, and hence could play a very important role in the terrestrial C cycle and global climate. It is, therefore, surprising how poorly we understand rates and environmental regulation of fungal respiration, especially given the need to accurately model potential ecosystem feedbacks to climate change.

The effect of temperature on ecosystem respiration is still not fully understood, with efforts focused on quantifying the effects on whole-ecosystem respiration (e.g., Janssens et al., 2001) and two major components— plant respiration (e.g., Ryan, 1991; Atkin and Tjoelker, 2003) and soil respiration (e.g., Lloyd and Taylor, 1994; Davidson and Janssens, 2006), both of which typically include poorly constrained fungal components. This lack of clear partitioning arises from a variety of factors, not the least of which is that partitioning of respiration between autotrophs and heterotrophs can be quite difficult (Hanson et al., 2000). Soil respiration is produced by a combination of broadly defined autotrophic and heterotrophic components. These are often operationally distinguished as autotrophic organisms and their symbiome, i.e., roots plus endorhizal and rhizoplane mycorrhizal, commensal and parasitic fungi and bacteria, as well as mycorrhizosphere organisms that consume exudates; and free-living heterotrophs that consume tissues (mostly saprotrophic and predatory biotrophic organisms). Although less commonly discussed, 'plant' respiration also contains fungal respiration, especially in roots because of mycorrhizal

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associations, but also other plant tissues because of the pervasive presence of fungal endophytes. Given the large differences noted between plant and fungal respiration rates (e.g., Andrew et al., 2014) even a small fungal component could contribute a significant fraction of empirical measurements of plant respiration. In addition it is likely that patterns of plant and fungal respiration respond differently to environmental cues such as temperature and moisture. Therefore, understanding controls on fungal respiration will enhance our ability to model plant symbiotic, soil and ecosystem respiration.

Temperature effects on respiration are sometimes simplistically modeled using Q_{10} values that specify an exponential rate of change in respiration as a function of temperature. For a variety of reasons we expect Q_{10} to be insufficient to adequately describe *in situ* field temperature responses of fungal respiration, as has previously been observed for soil respiration (Lloyd and Taylor, 1994; Davidson et al., 2006; Tuomi et al., 2008). First, we know that exponential rates of change in respiration are only valid over a narrow temperature range, beyond which organelle membranes, enzymes and other cellular constituents function poorly or break down entirely (Tansey and Brock, 1972; Robinson, 2001). Second, the conceptual basis for Q_{10} is that enzyme kinetics regulate cellular metabolism and should respond exponentially to temperature, yet we know that metabolism is more complex, because it is sensitive to environmental cues that can rapidly up- or down-regulate gene expression, enzyme production, and cellular metabolism (Davidson et al., 2006). Thus, specific biochemical and physiological responses to environmental cues could cause significant deviation from fixed Q_{10} responses to environmental stimuli such as temperature. Over the long term these could contribute to acclimation responses (Atkin and Tjoelker, 2003; Malcolm et al., 2008), but over the short-term these could result in non-exponential responses to temperature, or variation in Q_{10} values. Given that fungal tissues are often exposed to widely fluctuating temperatures over diurnal cycles, cellular physiology will be continually responding to these dynamic conditions, potentially leading to more complex response functions. Third, temperature often cycles strongly diurnally, and circadian and other endogenous rhythms can be superimposed on these diurnal cycles (Smith, 1973), potentially altering apparent temperature responses. Fourth, carbon supply can vary as a function of C fixation, transport, and competition with plant sinks, so respiration of mycorrhizal fungi could be linked to cyclic variation or trends in substrate supply (Heinemeyer et al., 2006, 2012; Kuzyakov and Gavrichkova, 2010). Fifth, water availability can also vary as a function of evaporative losses, hydraulic redistribution and plant sinks (Lilleskov et al., 2009), so respiration responses to temperature could be dampened as a function of diurnal cycles or trends in water availability.

Studies of respiration in the field are either destructive (e.g., Andrew et al., 2014) or non-destructive (e.g., Heinemeyer et al., 2006, 2007, 2012). The former provide the opportunity to determine time-point estimates of mass-specific rates of respiration of soil hyphae, which can be useful for scaling based on biomass estimates. They also permit a rapid snapshot of respiration temperature relationships for a large number of individual samples with little time investment. By contrast, the latter provide greater insights into temporal variation in respiration within an *in situ* organism or community—e.g., as a function of temperature, moisture, and substrate supply—and can be combined with terminal destructive harvests to provide information on mass-specific respiration.

Targets of respiration research include vegetative mycelia (e.g., Heinemeyer et al., 2006, 2007) and fungal sporocarps (Andrew et al., 2014). Although the former are targets of many recent studies on soil respiration, and represent the majority of fungal biomass in soils (e.g., Wallander et al., 2001), the advantages of the latter are several. First, they provide an opportunity to examine isolated fungal tissues with high biomass per unit area, enabling better estimates of fungal respiratory parameters. Second, the respiration rate can be linked to a species, enabling better estimates of taxon-specific fungal C costs for different functional classes of organisms (e.g., saprotrophs, ectomycorrhizal fungi). Third, understanding the respiratory costs of reproduction is essential to quantifying parameters of fungal carbon balance and fitness. Fourth, our previous work suggests that mass-specific respiration rates are fairly similar between vegetative mycelium and sporocarps (Andrew et al., 2014).

Remarkably, to my knowledge no studies have non-destructively examined sporocarp temperature-respiration relationships *in situ*. To fill this gap, I designed a custom chamber for examining *in situ* sporocarp respiration. My goals were to determine whether this system would provide robust estimates of diurnal sporocarp respiration-temperature relationships, to explore whether exponential or unimodal relationships are better for characterizing these relationships, and to begin to accumulate species-specific and functional group estimates of respiration of mycorrhizal and saprotrophic fungi. I hypothesized that (H1) under higher temperatures sporocarp respiration-temperature relationships would deviate downward from a simple exponential Q_{10}, and Gaussian regressions would be better than exponential regressions at representing these relationships; (H2) when approaching freezing temperatures, respiration would exhibit upward deviations from exponential relationships consistent with metabolic responses to cold stress; and (H3) when comparing sporocarps growing at different temperatures respiration would decline with increasing temperature, consistent with acclimation responses. Additionally I compare respiration estimates with published estimates for tree fine roots in order to begin to provide some additional insight into relative root and fungal contributions to soil respiration.

2. Methods

2.1. Study site

Ectomycorrhizal sporocarp respiration was measured in the field at the Houghton Rhizotron Facility at the US Forest Service Forestry Sciences Laboratory in Houghton, Michigan (47.115N, −88.548W). The Rhizotron has two forest types: on one side is a young white pine (*Pinus strobus*) stand planted in 2006 on imported Kalkaska sand, and on the other is a mature second growth deciduous forest stand dominated by red oak (*Quercus rubra*), sugar maple (*Acer saccharum*), basswood (*Tilia americana*), and white ash (*Fraxinus americana*), growing on native fine sandy loam.

2.2. Respiration measurements

To measure sporocarp respiration *in situ* I used an automated soil CO_2 flux system (LI-8100, LICOR, Lincoln, Nebraska, USA) with the 10 cm survey chamber (8100-102, LICOR, Lincoln, Nebraska, USA) fitted to a custom PVC collar that seals around the sporocarp stalk with easily conformable closed cell foam strips (Fig. S1). A mini-hypodermic thermocouple probe (model HYP0; Omega Engineering, Inc., Stamford, Connecticut, USA) was inserted ~1 cm into the center of the cap and wired into the LI-8100 for continuous temperature logging during respiration measurements. The system was attached to an AC to DC power supply (LI-8150-770, LICOR, Lincoln, Nebraska, USA) provided with AC power via the Rhizotron. Respiration measurements were programmed on the LI-8100 at

repeated intervals, typically every 30 min, for at least 24 h. Each measurement was run for 3 min with a 30 s dead band. At the end of the repeat measurement period the sporocarp was cut off at the point of entry to the chamber, put in a sealed plastic bag, weighed fresh, dried at 55 °C and reweighed. In the deciduous forest site sporocarps were more exposed to wind, increasing the chance of leakage and wind-stripping artifacts (Lai et al., 2012). To avoid any possibility of wind artifacts on measurements, I placed an open-topped 85 cm tall × 39 cm diameter opaque PVC pipe around the respiration system as a windbreak.

Field sporocarp measurements were performed opportunistically. I used sporocarps that were large and robust enough to tolerate being sealed into the chamber without damage to the stem or connection to soil mycelium. To be able to determine biomass-specific rates of respiration I minimized sporocarp growth over the *in situ* measurement period by choosing individuals with fully expanded caps. Although they were healthy and intact throughout the measurement period, they were not sterile, so colonization (and contributions to respiration) by fungivores is possible, but their activity was low enough that they did not damage the integrity of the sporocarp, which always remained intact and firm at the end of measurements. Sporocarps with visible decay or significant insect damage were not included in the analysis. In late October–early November 2011 mushrooms of *Laccaria laccata* were fruiting under the young white pine during an exceptionally mild fall. Sporocarp temperatures during this period ranged between −2 and 8 °C. I successfully measured respiration on three of these sporocarps over 1–3 d each. In late September-early October 2013 I measured respiration under the deciduous forest adjacent to the Rhizotron. One sporocarp each of *Hebeloma* sp. and *Cortinarius* sp. were monitored for several days. Temperatures ranged between 5 and 18 °C.

To measure saprotroph respiration and test for endogenous rhythms I used a commercial culture of *Lentinula edodes* (catalog # LKLE, Fungi Perfecti, Olympia, WA, USA), supplied growing on sawdust. I followed supplier's instructions for initiating fruiting. After fruiting I selected a sporocarp fruiting on the top surface of the culture block to facilitate sealing into the custom chamber, and used the same system as for the mycorrhizal sporocarps to examine respiration under a range of conditions. I placed the *L. edodes* culture in a Conviron PGR15 growth chamber (Controlled Environments Ltd., Winnipeg, Canada), and set initial conditions to 15 °C and 12 h day:12 h night for 2 d. Chamber humidity was regulated to ~65% by use of a humidifier. The sporocarp was enclosed in the *in situ* collar and monitored for respiration. To check for endogenous rhythms in respiration, after the first 2 d I switched off the lights, maintained a constant 15 °C air temperature, and logged respiration for 4 d more. Next, to test respiration-temperature relationships in a range of conditions, under 12 h day:night cycles I ramped temperature from 14° down to 4 °C over 12 h, next cycled up to 14 °C and back down to 4 °C over 24 h, next cycled up to 25 °C over 24 h and down to 7 °C over 4 h, and last cycled back up to 30 °C over the course of 4.5 h.

2.3. Data analysis

LI-8100 output was examined for quality control. Highly predictive ($r^2 > 0.99$) linear fits to chamber CO_2 accumulation rates over the course of individual measurements were used to confirm that a good seal had been achieved between the stem and the chamber. In some cases I observed strongly asymptotic and/or noisier CO_2 accumulation curves, in which cases the seal between the stem and the chamber or the sporocarp connection to vegetative mycelium was observed to be bad at the end of measurement. Only results from sporocarps that were undamaged, with no evidence of leakage, and which had a strong connection to vegetative mycelium at the end of the sampling period were used in the following analyses.

I fitted a variety of models to the sporocarp temperature-respiration data using SigmaPlot 13 (Systat Corporation version 13). All of the respiration rates were converted to dry mass-specific rates (i.e., μmol CO_2 per g dry weight per second) prior to fitting. I fitted these regressions for T-R relationships for increasing and decreasing temperature periods (approximately day and night) during individual 24 h cycle for as many days as the sporocarp was examined. To characterize temperature respiration relationships I started with two parameter exponential fits of van 't Hoff (Davidson et al., 2006):

$$R_{ms} = ae^{bT} \tag{1}$$

in which R_{ms} is mass specific sporocarp respiration, e is the base of the natural logarithm, T is the temperature (°C) and a and b are fitted coefficients. I evaluated exponential fits for homogeneity of variance, normality, and bias in residuals. When exponential fits were adequate I used the Q_{10} derived from the van 't Hoff equation (Davidson et al., 2006):

$$Q_{10} = e^{b*10} \tag{2}$$

To compare mass specific respiration rates among sporocarps, and between sporocarps and published rates for root mass specific respiration, I also solved for Equation (1) at 10 °C (R_{ms10}).

To include estimates of Q_{10} for data that did not follow an exponential curve, I also calculated Q_{10} for all sample intervals with ≥2 °C temperature range using the following modified van 't Hoff equation (Davidson et al., 2006):

$$Q_{10} = \left(\frac{r_2}{r_1}\right)^{\frac{10}{T_2 - T_1}} \tag{3}$$

where r_1 and r_2 are the measured mass-specific respiration rates at minimum and maximum temperatures T_1 and T_2, respectively. This allowed me to test the effect of change in temperature on Q_{10} with a larger set of sample intervals and a broader range of temperatures.

I also fitted Gaussian 3 or 4 parameter models, as appropriate. Perhaps best known for describing the normal distribution, Gaussian models are one of a class of unimodal models that can capture any decrease in Q_{10} with increasing temperature, identify a maximum value for respiration, and permit negative slopes at higher temperatures, which is more biologically realistic than assuming monotonic positive slopes, especially over broader temperature ranges (Hunt, 1977; O'Connell, 1990; Tuomi et al., 2008). In a comparison of models describing soil heterotrophic respiration—exponential, Arrhenius, modified Arrhenius (Lloyd and Taylor, 1994), asymptotic (Del Grosso et al., 2005), and Gaussian—the Gaussian models performed best (Tuomi et al., 2008).

I first attempted to fit the data with Gaussian 3 parameter models:

$$R_{ms} = ae^{\left[-0.5\left(\frac{T-T_0}{b}\right)^2\right]} \tag{4}$$

where a, b, and T_0 are fitted parameters: T_0 is a fitted term for the temperature at peak respiration, and a is the fitted parameter for peak respiration rate.

When 3 parameter models could not be fitted (arrays ill conditioned at final iteration or not meeting test assumptions), I fitted the data with Gaussian 4 parameter models:

$$R_{ms} = R_{ms0} + ae^{\left[-0.5\left(\frac{T-T_0}{b}\right)^2\right]} \quad (5)$$

where R_{ms0} is the y intercept for mass specific respiration. Note that $R_{ms0} + a$ is the fitted peak respiration rate (when $T = T_0$).

R^2, tests of normality, and examination of residuals of both exponential and Gaussian models were used to determine adequacy of the fit. When both exponential and Gaussian models could be fitted, the corrected Akaike Information Criterion (AIC_c) was calculated for all regressions, and the weighted likelihood (w_i) of exponential and Gaussian fits were calculated (Burnham and Anderson, 2002). In weighted likelihoods, the values of the alternate models sum to 1, with higher values indicating higher likelihoods.

To characterize hysteresis in the temperature-respiration relationship, I fitted regressions and calculated Q_{10} for sporocarp respiration separately for the increasing temperature (typically day) and decreasing temperature (typically night) intervals of the diel temperature-respiration cycle. I did this separately for each 24 h period to account for any shifts in basal respiration. To test for evidence of longer-term acclimation, I also used non-linear regression, or rank correlations when regression assumptions were not met, to examine the relationship between the maximum temperature of sporocarps and both the average and individual R_{ms10} and Q_{10} for sporocarps. This was first done only for the intervals that met the criteria for inclusion in the Q_{10} estimates. However, this excluded many intervals best fit by Gaussian regression, yet these intervals are the ones most likely to capture changes in Q_{10} as a function of temperature. To incorporate these Q_{10} estimates for periods better fitted by unimodal relationships, I next relaxed these criteria to include all intervals that had a temperature change of at least 2 °C. I then fitted the Q_{10} using Equation (3). When testing for relationships with maximum temperature, these data did not meet the test criteria for linear or non-linear regressions and so were fitted with a negative exponential smoothing function for display purposes, and the relationship was tested with Spearman rank correlation.

To determine the potential influence of fungal respiration on field root respiration (in roots which contain a percentage of mycorrhizal or endophytic fungal tissue) I also compared the estimates of sporocarp respiration with published estimates of root respiration at a common temperature, and assuming that this represented the respiration rate for hyphae in roots, I estimated the fungal fraction of root respiration using different published estimates of ectomycorrhizal fraction of fine root biomass.

3. Results

3.1. What models are best for describing fungal respiration-temperature relationships?

For all sporocarps exponential models were highly predictive and provided an adequate fit under some conditions, especially at intermediate temperatures and during decreasing temperature intervals (Table 1, Figs. 1–4). However, there were clear deviations from these exponential models, especially in increasing temperature intervals and when the diurnal cycle attained higher (>17 °C) and lower (<0 °C) temperatures. In seven intervals Gaussian models could not be fitted. When both exponential and Gaussian models could be fitted, the w_i for exponential models were never significantly better than Gaussian, whereas Gaussian model w_i were significantly better in nine of 25 cases (Table 1). In all cases even when residuals were unbiased, variance tended to increase at higher temperatures, especially for increasing temperature intervals for *Cortinarius*, *L. edodes*, and *Hebeloma* (Figs. 2–4), but not for *L. laccata* (Fig. 1).

In *L. laccata*, when temperatures fell below zero, the decreasing interval of the diurnal temperature cycle was under-predicted by an exponential model, and the early part of the increasing interval was over-predicted, although respiration typically recovered to previous rates by the end of the warming interval (Fig. 1). During warming, this led to positive residuals at the lowest temperatures, and negative residuals at intermediate temperatures for exponential (Fig. 1C) but not Gaussian (Fig. 1D) models. Deviations from exponential fits were also evident during the increasing temperature intervals of the warmest diurnal temperature cycles for *Cortinarius* and *L. edodes,* and perhaps for *Hebeloma* (Figs. 2A and B, 3A and B, 4B). For *Cortinarius*, this was evident as an upward deviation above an exponential fit above intermediate temperatures, leading to poorly-distributed residuals in exponential fits (Fig. 2C). These were followed by a rapid decline as peak temperatures were approached near the end of the day. For *L. edodes* there was no strong evidence of an upward deviation from exponential (Fig. 3A and C), only a decline at higher temperatures (Fig. 3B and D). Under these warmer conditions Gaussian 3 or 4 parameter regressions provided better fits for both *L. edodes* and *Cortinarius* (Table 1; Figs. 2B and D, 3B and D). For *Hebeloma*, exponential and Gaussian fits were both adequate (Table 1; Fig. 4A-D), except that at the highest temperature there was a rapid shift from an exponential increase in temperature to an erratic spike in respiration with no change in temperature at 17 °C that was not adequately fit by exponential or Gaussian models (Fig. 4B).

3.2. Patterns of diel hysteresis in respiration

As evident from the differences between the fits for day and night respiration, hysteresis was clearly evident in the respiration of *L. laccata* and *Cortinarius* sp., but the pattern of hysteresis differed (Figs. 1B and 2B). For *L. laccata* sporocarps, which were examined under colder temperatures, the observed responses were more linear on the nocturnal decreasing interval, whereas on the diurnal increasing interval rates had a strong concave curvature, leading to a counterclockwise hysteresis. In *Cortinarius*, which was examined under warmer temperatures, curves were similar for the lower half of the increasing and decreasing intervals, but deviated steeply above the exponential curve then fell rapidly back to the exponential curve at the warmest part of the increasing interval, but not the decreasing interval (Fig. 2), leading to a clockwise hysteresis. In general decreasing temperature intervals were less likely than increasing intervals to exhibit a strong improvement in fits using Gaussian models as indicated by their lower average Gaussian w_i and by the fact that of the seven intervals that could not be fitted by Gaussian models, five were decreasing temperature intervals, and only two were increasing temperature intervals (Table 1).

In examining the multi-day patterns of respiration under constant air temperature for *L. edodes*, I found no evidence of circadian or other endogenous rhythms (data not shown).

3.3. Q_{10}, mass-specific respiration rates, and acclimation

All sporocarps had some increasing or decreasing temperature intervals that were adequately fit by exponential regressions ($r^2 > 0.90$, normal distribution). Where exponential fits were adequate, Q_{10} values had a mean of 3.74 ± 0.41 (SE) and R_{ms10} had

Table 1
Comparison of exponential and Gaussian regression models (r^2, normality, constant variance, AICc metrics for different models), of sporocarp temperature-respiration relationships. AICc are only calculated where both exponential and Gaussian models were fitted.

Day	T trend[a]	T min	T max	Gaussian model[b]	r^2		Normal (P)[c]		Const. var. (P)[d]		AIC_c[e]		Δ_i[f]		w_i[g]	
					Exp.	Gauss.	Exp.	Gauss.	Exp.	Gauss.	Exp.	Gauss.	Exp.	Gauss.	Exp.	Gauss.
Laccaria laccata 1																
1	dec	1.8	7.4	NA	0.97	NA	0.62	NA	0.01	NA	NA	NA	NA	NA	NA	NA
2	inc	2.1	5.8	3	0.98	0.98	0.69	0.46	0.18	0.48	−285.3	−288.7	3.45	0.000	0.151	0.849
2	dec	−0.5	4.6	4	0.89	0.92	0.07	0.64	0.12	0.01	−450.2	−454.2	4.04	0.000	0.117	0.883
3	inc	−0.5	7.2	4	0.97	0.97	0.78	0.80	0.15	0.11	−234.1	−233.0	0.00	1.087	0.633	0.367
Laccaria laccata 2																
1	dec	−0.4	6.1	3	0.97	0.99	0.00	0.82	0.18	0.63	−453.6	−472.3	18.77	0.000	0.000	**1.000**
2	inc	−0.5	6.3	4	0.95	0.98	0.36	0.88	0.63	0.04	−210.9	−223.1	12.25	0.000	0.002	**0.998**
2	dec	−0.4	5.3	3	0.98	0.99	0.52	0.79	0.05	0.88	−490.9	−501.2	10.23	0.000	0.006	**0.994**
3	inc	−0.1	6.1	NA	0.97	NA	0.04	NA	0.75	NA	NA	NA	NA	NA	NA	NA
4	inc	5.6	8	3	0.95	0.97	0.62	0.22	0.13	0.20	−216.9	−239.8	22.92	0.000	0.000	**1.000**
Laccaria laccata 3																
1	dec	−1.9	3.8	4	0.94	0.94	0.74	0.74	0.83	0.97	−469.0	−465.5	0.00	3.485	0.851	0.149
2	inc	−1.6	6.9	4	0.90	0.95	0.25	0.21	0.08	0.53	−210.2	−214.4	4.16	0.000	0.111	0.889
Lentinula edodes																
1	dec	10.1	20.59	4	0.98	0.98	0.95	0.86	0.75	0.17	−265.1	−264.4	0.00	0.678	0.584	0.416
8	dec	3.97	13.79	3	0.99	1.00	0.95	0.29	0.71	0.81	−348.6	−365.0	16.40	0.000	0.000	**1.000**
9	inc	3.97	13.77	3	0.96	0.96	1.00	0.88	0.00	<0.0001	−364.9	−362.2	2.68	0.000	0.208	0.792
9	dec	4.14	13.58	3	0.98	0.98	0.12	0.07	0.36	0.06	−411.1	−410.6	0.46	0.000	0.443	0.557
10	inc	4.25	25.65	3	0.96	0.98	0.01	0.27	0.80	<0.0001	−750.5	−787.5	36.95	0.000	0.000	**1.000**
10	dec	5.57	25.31	3	0.89	0.97	0.91	0.83	0.46	0.26	−88.6	−90.2	1.59	0.000	0.311	0.689
11	inc	8.71	29.88	3	0.84	0.97	1.00	0.47	0.61	0.41	−111.1	−125.3	14.21	0.000	0.001	**0.999**
11	dec	7.69	25.31	3	0.90	0.98	0.91	0.83	0.46	0.26	−55.0	−57.3	2.29	0.000	0.242	0.758
Cortinarius sp.																
1	dec	4.96	9.71	3	0.92	0.94	0.90	0.99	0.26	0.34	−371.0	−371.7	0.68	0.000	0.415	0.585
2	inc	5.66	11.71	3	0.96	0.96	0.06	0.09	0.10	0.24	−366.9	−365.1	0.00	1.772	0.708	0.292
2	dec	5.73	10.83	NA	0.96	NA	0.07	NA	0.37	NA	NA	NA	NA	NA	NA	NA
3	inc	5.87	15.42	4	0.93	0.97	0.08	0.01	0.00	0.12	−291.9	−296.5	4.58	0.000	0.092	0.908
3	dec	8.49	15.25	3	0.93	0.93	0.21	0.11	0.17	0.35	−347.5	−344.7	0.00	2.788	0.801	0.199
4	inc	7.62	16.73	4	0.75	0.94	0.03	0.19	<0.0001	<0.0001	−259.7	−279.8	20.06	0.000	0.000	**1.000**
4	dec	10.1	16.6	NA	0.90	NA	0.61	NA	0.27	NA	NA	NA	NA	NA	NA	NA
5	inc	9.76	17.75	4	0.81	0.91	0.01	0.39	0.02	<0.0001	−264.5	−276.7	12.13	0.000	0.002	**0.998**
5	dec	10.4	17.39	NA	0.97	NA	0.96	NA	0.06	NA	NA	NA	NA	NA	NA	NA
Hebeloma sp.																
5	dec	9.38	13.23	3	0.92	0.94	0.21	0.42	0.42	0.76	−370.2	−371.8	1.66	0.000	0.304	0.696
6	inc	9.25	12.34	3	0.90	0.91	0.54	0.77	0.85	0.96	−383.3	−382.7	0.00	0.642	0.580	0.420
6	dec	7.07	12.11	NA	0.98	NA	0.16	NA	0.02	NA	NA	NA	NA	NA	NA	NA
7	inc	6.59	17.22	NA	0.91	NA	0.00	NA	<0.0001	NA	NA	NA	NA	NA	NA	NA
Average decreasing					0.95	0.96	0.52	0.62	0.32	0.46	−343.39	−347.41	4.68	0.58	0.34	0.66
Average increasing					0.92	0.96	0.36	0.43	0.33	0.34	−303.86	−313.44	10.26	0.27	0.19	0.81
Average all					0.93	0.96	0.45	0.52	0.33	0.41	−322.84	−329.75	7.58	0.42	0.26	0.74

[a] Temperature trend over measurement interval: dec = decreasing temperature trend; inc = increasing temperature trend.
[b] '3' indicates 3 parameter Gaussian model; '4' indicates 4 parameter Gaussian model; 'NA' indicates not available because of lack of convergence of 3 or 4 parameter Gaussian models.
[c] Normal = P value of Shapiro-Wilk Normality test. Lower p values indicate deviation from normality.
[d] Const. variance = P value of Constant Variance Test. Lower p values indicate deviation from constant variance.
[e] AICc = corrected Akaike Information Criterion.
[f] Δ_i = AICc differences from the best model.
[g] w_i = Akaike weights: weights for alternative models sum to 1, with higher values indicative of higher probability.

a mean of 0.021 ± 0.005 μmol g^{-1} s^{-1} (Table 2).

For all sporocarps combined, when I regressed maximum temperature during the measurement period against predicted R_{ms10} I found a strong negative exponential decay relationship when I looked at both individual measurement periods (Fig. 5A) and sporocarp averages (Fig. 5B), consistent with acclimation to warmer temperatures. Similar results were found with average temperature as the predictor (data not shown). When I regressed maximum temperature during the measurement period against Q_{10} for samples with good exponential fits I found a non-significant negative relationship both for individual measurement periods (Fig. 5C) and sporocarp averages (Fig. 5D). However, when I included all sample intervals with a temperature difference >2 °C, including those better fit by Gaussian equations, and fit those with Equation (3), I found a significant negative rank correlation (Fig. 6) consistent with an acclimation response.

4. Discussion

4.1. H1 and 2. Deviations from exponential in warmer and colder sporocarps

Although both exponential and unimodal models were highly predictive of fungal respiration, the significantly better fits for the unimodal models for three of four species is consistent with hypothesized deviations from exponential growth. This was most evident during the warmer portions of increasing temperature intervals. These results contrast with the finding of a positive temperature response for respiration in soil saprotroph communities, but not in mycorrhizal extraradical hyphae (Heinemeyer et al., 2006, 2007), which they attribute to carbon limitation of fungal respiration. Although I found sporocarp respiration to be strongly predicted by temperature, I was examining sporocarps, which are typically produced later in the season when belowground carbon

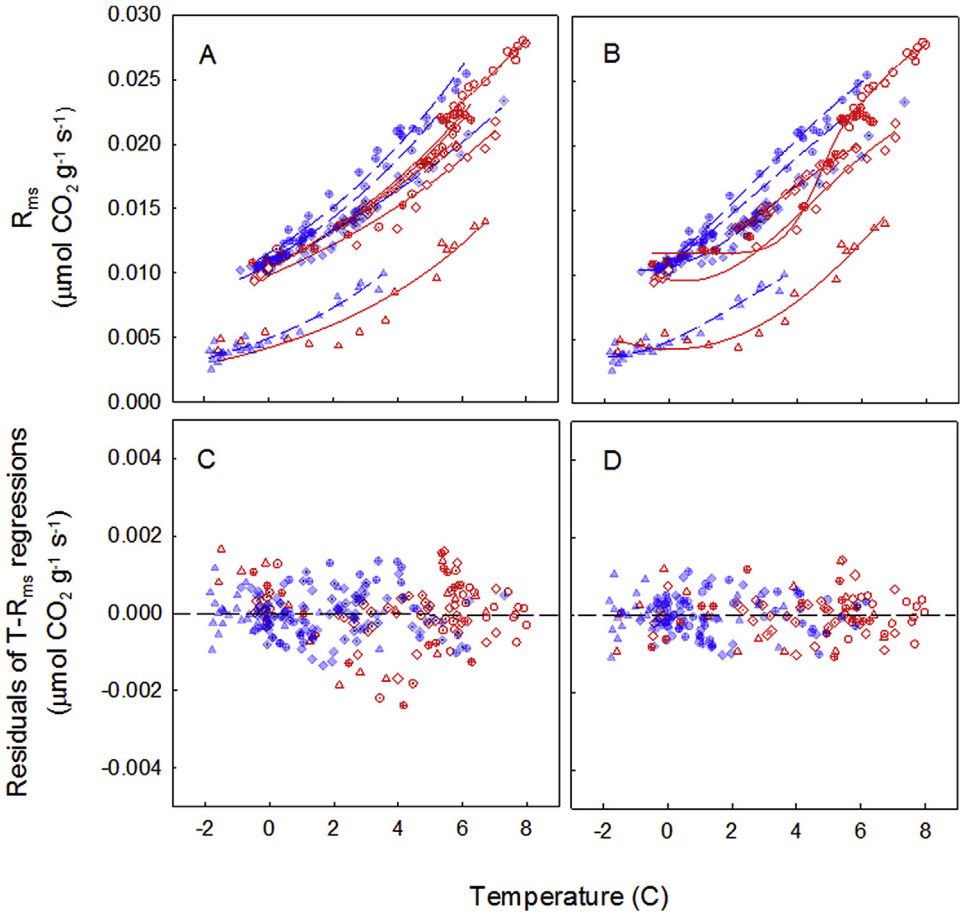

Fig. 1. *Laccaria laccata* 1–3 individual day and night mass-specific respiration-temperature fits: (A) exponential 2 parameter and (B) Gaussian 3 or 4 parameter fits; (C) and (D) are residuals of (A) and (B), respectively. In (A) and (B) fitted lines are for individual day (solid red) or night (dashed blue). In (C) note the daytime residuals are biased positive as temperatures begin to warm early morning, and biased negative as sporocarps cool at night, whereas in (D) residuals are unbiased for both day and night. Each symbol represents an individual decreasing or increasing temperature interval. Red open symbols are for increasing temperatures, blue filled symbols are for decreasing temperature intervals.

allocation is high, and so are less likely to be strongly carbon limited. Thus rates during the fruiting season are likely to be higher than those at equivalent temperatures during seasons when aboveground sinks are competing for carbohydrates (e.g., Högberg et al., 2010).

Although respiration deviated from exponential, our hypothesis that respiration under warmer temperatures would deviate downward from exponential slopes was supported in the case of *L. edodes* but not *Cortinarius*. Although *Cortinarius* was also best fit by a Gaussian regression and peaked below the highest temperatures, the deviations above exponential curves during the middle part of the warming intervals (as evident from the residual analysis and the comparison with respiration under decreasing temperatures) were not consistent with hypothesis 1, suggesting this fungus experienced accelerated metabolism likely in response to thermal stress, although some influence of circadian rhythms or other drivers of hysteresis (see below) cannot be ruled out. Gaussian models were only superior to exponential models for one of the three *L. laccata*, perhaps because under colder conditions rates are well below the peak, making Gaussian models less appropriate, or perhaps because of idiosyncrasies of the species. For *Hebeloma* there was no significant difference between Gaussian and exponential models for growth under 17 °C, although neither captured the sudden spike in respiration observed above 17 °C. It is possible that in this species there is a threshold response of respiration to temperature, but as this spike was only observed once, artifacts (machine error, non-fungal sources of respiration, etc.) cannot be ruled out.

The generally equivalent or improved fits when using unimodal models suggests that they should be used in place of exponential models to provide accurate estimates of short-term responses for fungal respiration. This conclusion is consistent with our previous finding for fungal respiration rates at the Aspen FACE site using destructive sampling, where Gaussian were a much better fit than exponential regressions once temperatures rose above 18 °C (Andrew et al., 2014). Furthermore, this conclusion is also consistent with results for soil respiration, which show that a declining Q_{10} with increasing temperatures (Lloyd and Taylor, 1994) is best fit with Gaussian models (Tuomi et al., 2008). Although these Gaussian functions appear able to capture diurnal peaks, it should be kept in mind that as temperatures rise much above peak respiration, the relationship would likely deviate downward from the symmetrical Gaussian function, because of rapid loss of metabolic activity as temperature thresholds are crossed and cells lose integrity.

Physiological and biochemical responses to changing thermal conditions likely affect respiration and fungal respiration-temperature relationships, e.g., cold stress tolerance responses such as the production of sugars, sugar alcohols, antifreeze proteins, unsaturated lipids and cold-adapted enzymes (Duman and Olsen, 1993; Robinson, 2001); and heat and moisture stress responses such as the production of sugar alcohols, heat shock

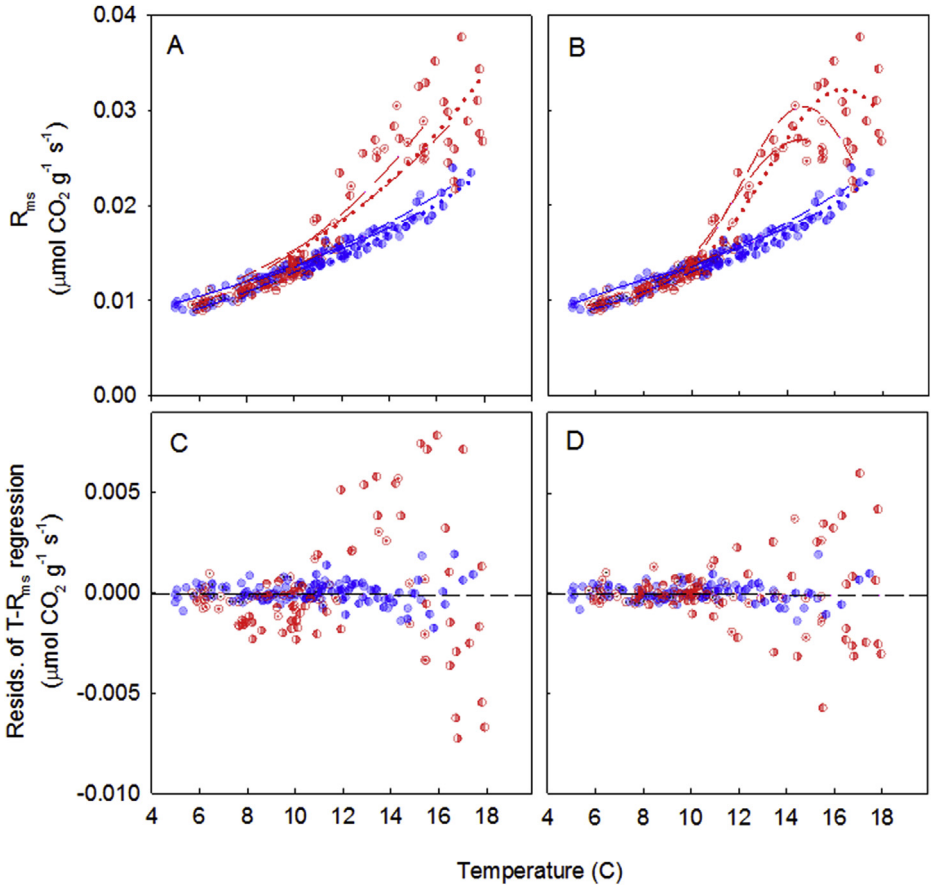

Fig. 2. *Cortinarius* sp. separate day and night temperature *vs.* mass-specific respiration regression fits over the course of 5 days. (A) Exponential 2 parameter regression fits, (B) Gaussian 3 or 4 parameter regression fits; (C) residuals of (A); (D) residuals of (B). In the exponential fits, note the larger bias in residuals for the upper half of the increasing intervals compared with the decreasing intervals. Each symbol represents an individual decreasing or increasing temperature interval. Red partially open symbols are for increasing temperatures, blue filled symbols are for decreasing temperature intervals.

proteins, and oxidative repair mechanisms (Abrashev et al., 2008). These responses could explain the clearly non-exponential upward deviations in Q_{10} during warming followed by exponential declines during subsequent cooling, as observed in the *Cortinarius* sp. They would also explain the slightly higher than exponentially declining respiration rates observed in some intervals during the cooling of *L. laccata* when approaching freezing temperatures. Respiration in plants and fungi has been observed to increase during heating but become depressed afterward (e.g., Sundberg et al., 1999; Kurets et al., 2003). Similarly, cold hardening and frost damage can both alter temperature-respiration relationships in plants, leading to increases in Q_{10} during cold hardening (Kurets et al., 2003). Under diurnally varying conditions of sufficient magnitude these costs would be incurred daily, increasing the overall respiratory rate of fungi. In contrast, the effect of damage to cells could be to reduce respiration as respiratory pathways are interrupted, and subsequently increase respiration during the repair process. This pattern is consistent with what was observed in warming *L. laccata* after sub-freezing temperatures, although it is also possible that the initial lag in respiratory response to warming was due to effects of cold acclimation on respiratory machinery.

4.2. H3. Q10 and evidence for acclimation?

Although exponential models of temperature-invariant Q_{10} have their limitations in describing fungal respiration, exponential fits were valid for intermediate data. In these cases I observed a mean Q_{10} of 3.74 ± 0.41 (SE), with a range of 1.96–7.09, which is higher than Q_{10} of 1.67–2.56 observed by Malcolm et al. (2008) in pure cultures. The present study differed from theirs in numerous ways (e.g., rapid *vs.* slow warming, field *vs.* lab, connected to host *vs.* pure culture, sporocarp *vs.* vegetative mycelium, wider *vs.* narrower temperature ranges, uncontrolled *vs.* controlled time-course of warming) all of which could have affected observed Q_{10} values. The present results should be more predictive of unacclimated diurnal responses, and so it is worth noting that higher Q_{10} values for these short-term fungal responses to elevated temperatures could have significant impacts on models partitioning diurnal respiration between fungi and other organisms, with the caveat that these would only be valid over a narrow intermediate temperature range.

The among-sporocarp and among-diurnal-interval results showing lower R_{ms10} and Q_{10} (under relaxed estimation criteria) with increasing temperatures are consistent with acclimation responses to longer-term changes in temperature. Although the present results could also have arisen from species-specific or non-thermal environmental differences between the measurement periods, they are consistent with other studies. Acclimation reduces respiratory costs under warmer conditions in plant (e.g., Atkin et al., 2005; Jarvi and Burton, 2013) and fungal respiration (e.g., Lange and Green, 2005; Heinemeyer et al., 2006; Malcolm et al., 2008), and colonization of roots by mycorrhizal fungi can reduce the magnitude of acclimation responses (Atkin et al., 2009). The latter

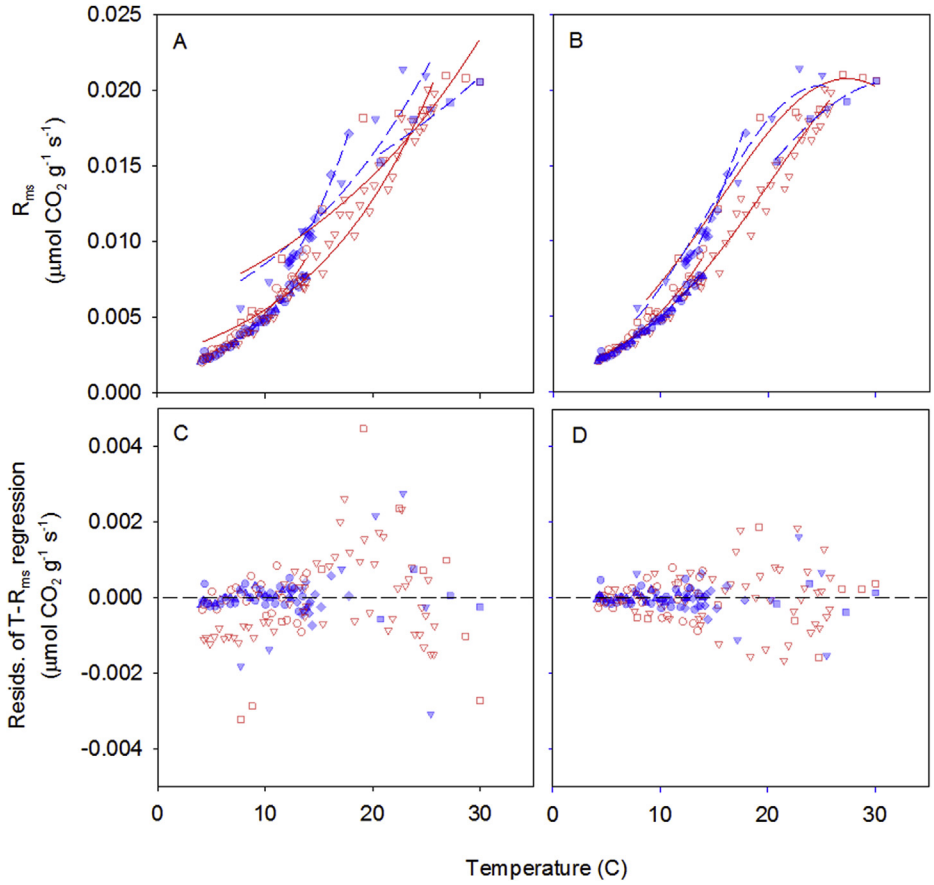

Fig. 3. *Lentinula edodes* sporocarp temperature-respiration regressions with (A) exponential fits; (B) Gaussian fits; (C) residuals of exponential fits; (D) residuals of Gaussian fits. All fits were significant except day 11 decreasing (filled blue squares), with only four data points contributing to the regression. Each symbol represents an individual decreasing or increasing temperature interval. Red open symbols are for increasing temperatures, blue filled symbols are for decreasing temperature intervals.

might arise directly from the interspecific interaction, or alternatively from the differential temperature acclimation profiles, of the fungal and plant partners, reinforcing the need for characterization of the long-term acclimation patterns of both symbiotic partners. Understanding the nature of acclimation responses in fungi is of course critical to improved modeling of ecosystem carbon cycling.

These data suggest that, given the large diurnal temperature cycles that can be experienced by sporocarps and other fungal tissues, the temperature responses of fungal respiration will represent a longer-term trend of acclimation modified by unimodal responses to short-term temperature dynamics that are especially evident at higher temperatures and high belowground carbon flux. The importance of shorter and longer-term temperature responses will depend on the relative magnitude of diurnal *vs.* seasonal variation in temperature. This will be driven by the degree of seasonality and thermal buffering. Although mycelium deeper in the soil would experience a more thermally stable environment, many hyphae experience unstable thermal environments similar to those experienced by sporocarps, e.g., in leaves, surface organic or exposed mineral horizons, fine woody debris, and surfaces of live or dead wood. For example, in a northern hardwood forest in northern Michigan, soils at 1 and 5 cm depth commonly experienced diurnal temperature amplitude of ~5 and 1.5 °C respectively, and subweekly variation of approximately 10 and 5 °C, respectively (Lilleskov unpublished). Given the concentration of fungal mycelia from litter-feeding and mycorrhizal fungi in these organic-rich substrata it is clear that a large fraction of fungal biomass experiences a continuously shifting thermal environment that could drive short-term dynamic responses in respiration similar to those seen in the present study. In some cases this might involve bursts of respiration to avoid or tolerate stress, leading to respiration that deviates upward from an exponential curve as observed for the increasing interval of *Cortinarius* under warm conditions, and the latter half of the increasing interval of *L. laccata* as it warmed out of freezing conditions.

4.3. Implications of fungal respiration rates for root versus fungal respiration partitioning

As noted earlier, fungi are intimately involved in root processes as symbionts, so it is worth comparing respiration rates of fungi and fine roots. Estimates of mean R_{ms10} (0.021 ± 0.005 μmol g^{-1} s^{-1}) in the present study are remarkably similar to those for sporocarp and mycelial fungal respiration at similar temperatures in another study using destructive sampling methods (Andrew et al., 2014). Fungal respiration rates in the present study were ~14× higher than published R_{ms10} for fine roots of *Quercus* and *Pinus* in similar ecosystems (0.0015 ± 0.00009 μmol g^{-1} s^{-1}; calculated from data in Burton et al., 2002). This difference between fungal and root respiration is consistent with previous findings of approximately an order of magnitude higher R_{ms} for both fungal mycelium and sporocarps in aspen stands compared with published values for aspen fine roots (Andrew et al., 2014).

It is also important to remember that fine roots used for the

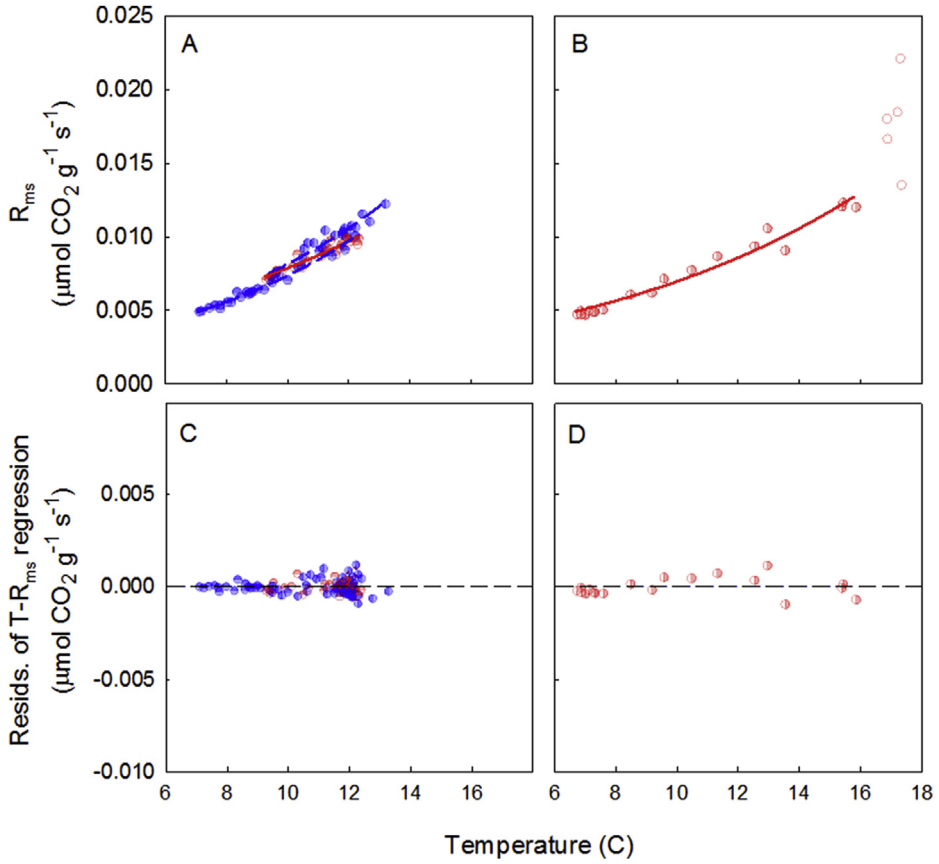

Fig. 4. *Hebeloma* sp. separate day and night temperature *vs.* mass-specific respiration exponential regression fits for data successfully fit from (A) day 1–6 and (B) the increasing interval on day 7 and (C) and (D) residuals of A and B, respectively. Gaussian fits not shown because they did not improve the fit significantly. Note the open symbols in (C) indicating the spike in respiration that was not included in the fit. Each symbol represents an individual decreasing or increasing temperature interval. Red open symbols are for increasing temperatures, blue filled symbols are for decreasing temperature intervals.

Table 2
Averages of Q_{10}, r^2 for two parameter exponential fits, minimum and maximum temperature, and fitted mass-specific respiration at 10 °C separately for all increasing and decreasing temperature intervals of diurnal cycles that had an $r^2 \geq 0.90$ and passed the normality test. n = number of separate days on which the preceding criteria were met for that sporocarp. Overall averages were calculated from the individual sporocarp averages. SE for individual sporocarps was calculated from replicate measures on multiple intervals for that sporocarp. SE for overall among-sporocarp average was calculated from the SE of the mean values from individual sporocarps.

ID	Temp. trend	n	Q_{10}		r^2	T min	T max	R_{ms10}
			Mean	SE	(adj)	°C	°C	µmol CO_2 g^{-1} s^{-1}
Laccaria laccata 1	increasing	2	2.93	0.02	0.97	0.80	6.50	0.030
	decreasing	2	2.91	0.14	0.93	0.65	6.00	0.031
Laccaria laccata 2	increasing	2	3.08	0.48	0.95	2.55	7.15	0.036
	decreasing	1	3.89	–	0.98	−0.40	5.30	0.042
Laccaria laccata 3	increasing	1	5.90	–	0.90	−1.60	6.90	0.025
	decreasing	1	7.09	–	0.94	−1.90	3.80	0.035
Lentinula edodes	increasing	1	3.94	–	0.96	3.97	13.77	0.005
	decreasing	3	3.65	0.03	0.98	6.07	15.99	0.005
Cortinarius sp.	increasing	2	2.70	0.43	0.94	5.77	13.57	0.015
	decreasing	5	2.13	0.08	0.94	7.94	13.96	0.013
Hebeloma sp.	increasing	1	2.79	–	0.90	9.25	12.34	0.008
	decreasing	2	3.85	0.13	0.95	8.23	12.67	0.008
Average	increasing	6	3.56	0.51	0.94	3.46	10.04	0.020
Average	decreasing	6	3.92	0.69	0.95	3.43	9.62	0.023
Average	all	12	3.74	0.41	0.94	3.44	9.83	0.021

above respiration estimates contain some mycorrhizal and other endophytic fungal biomass, so the pure root respiration term might be even lower if fungal respiration were excluded. For example, assuming (1) sporocarp and extraradical vegetative fungal mycelium R_{ms10} equals fungal R_{ms10} in fine roots, (2) the R_{ms10} for fine roots reported above is representative, and (3) epi-and intraradical fungal biomass from mycorrhizas and endophytes is 2% of fine root biomass (e.g., Satomura et al., 2003), then the equation

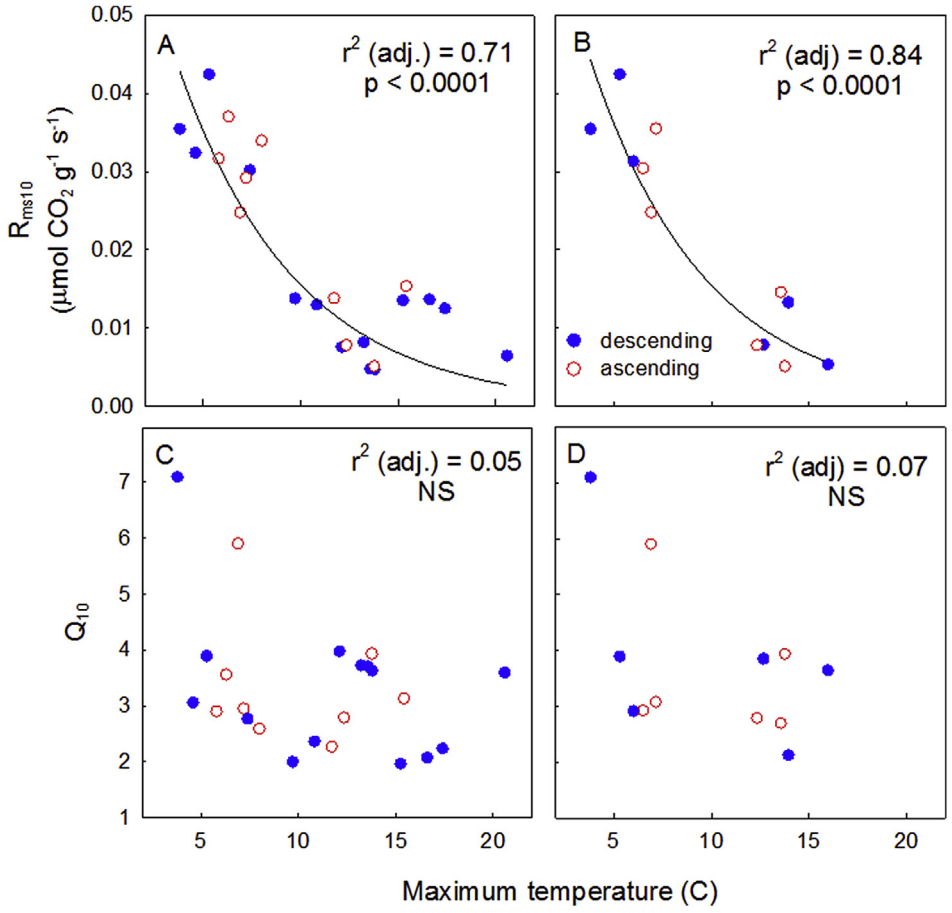

Fig. 5. Maximum temperature *vs.* fitted values of R_{ms10} (A and B) and Q_{10} (C and D). In (A) and (C) each point represents values for a single increasing (solid blue) or decreasing (open red) interval of a diel temperature cycle that was adequately fit by an exponential relationship between temperature and respiration. In (B) and (D) each point represents average values for each sporocarp for all increasing or decreasing intervals of diel temperature cycles that were adequately fit by an exponential relationship between temperature and respiration. Lines represent exponential decay functions for combined increasing and decreasing data.

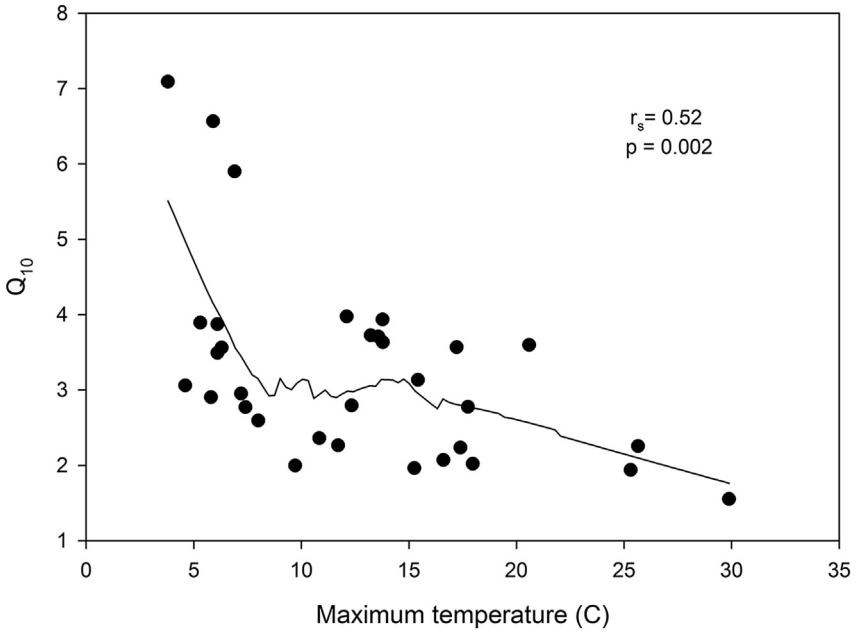

Fig. 6. Maximum temperature *vs.* Q_{10} calculated using Equation (3) for individual ascending and descending intervals that had a temperature range ≥2 °C. Line is a negative exponential smoothing fit. R_s is the Spearman rank correlation, used because data did not meet the normality criteria for linear or non-linear regression.

$$R_{ms10\ \ pure\ \ root} = R_{ms10\ \ mycorrhizal\ \ roots} - \frac{R_{ms10\ \ fungal}{*}fraction\ \ fungal\ \ biomass}{fraction\ \ pure\ \ root\ \ biomass} \quad (6)$$

can be used to estimate that:

$$R_{ms10\ \ pure\ \ root} = 0.0015\ - \frac{0.021{*}0.02}{0.98} = 0.0011\ \ \mu mol\ \ CO_2\ g^{-1}s^{-1} \quad (7)$$

in which case the fungal:pure root R_{ms10} ratio increases to ~19×, and fungal respiration is estimated at 28% of total fine root respiration. If fine roots contain 3% fungal biomass (e.g., Kårén and Nylund, 1996), then using Equation (6) pure root respiration declines to 0.0008 μmol CO_2 g^{-1} s^{-1}, fungal:pure root R_{ms10} ratio climbs to ~23×, and fungal respiration is estimated at 42% of total fine root respiration.

Hobbie (2006) speculated that hyphal respiration would scale based on the ¾ power of mass, which was proposed by Enquist et al. (2003) as a general scaling factor for respiration. Assuming a root diameter of 100 μm and hyphal diameter of 10 μm Hobbie (2006) predicted a 32× higher respiration rate in fungal hyphae than fine roots. This is higher than the R_{ms10} ratio I calculated here (~14×) and in Andrew et al. (2014), but closer to the rate estimated when fungal respiration is removed from the root respiration estimates. Of course scaling laws break down in the present case, because they would predict lower respiration for the larger-diameter sporocarps than fine roots, which is not supported by the present study. Based on previous field estimates for aspen-dominated ecosystems, rates of vegetative mycelium respiration were comparable to (~1.5×) sporocarp respiration (Andrew et al., 2014), suggesting that fungal respiration is relatively invariant to the scale of the tissue and might have more to do with tissue function.

These 'back-of-the-envelope' calculations clearly indicate that with the large observed differences in fungal and root mass-specific respiration, small changes in fungal biomass might have a very large impact on both root respiration rates and the fungal fraction of that respiration. To be clear, sporocarp and vegetative mycelial mass-specific respiration rates may not be identical. Andrew et al. (2014) found that sporocarp R_{ms} was 75 ± 8% of vegetative, but I am not aware of other direct comparisons of field rates from the same system. If this relationship holds, sporocarp respiration estimates should be considered as reasonable, and perhaps conservative, proxies for vegetative mycelium. A summary of studies exhibits high variation and overlapping ranges of field sporocarp respiration and culture-based hyphal respiration (Andrew et al., 2014). Future studies should test the mass-specific respiration rates of reproductive, extraradical vegetative, and intraradical fungal mycelium more broadly, and compare their temperature response functions during periods of high and low belowground carbon flux and soil moisture. As noted earlier, there is some evidence that extraradical mycorrhizal fungal respiration might be less responsive to diurnal temperature cycles than saprotrophs under certain circumstances (e.g., Heinemeyer et al., 2007).

4.4. Hysteresis and soil respiration

We found clear evidence of diurnal hysteresis in some ectomycorrhizal fungal sporocarps. Although not discussed as hysteresis, Heinemeyer et al. (2007) noted differences in diurnal respiration *vs.* temperature for mycorrhizal fungi in root-exclusion collars consistent with a counterclockwise hysteresis, i.e., nighttime respiration higher than daytime for a given temperature. The observed hysteresis in fungal respiration seen most clearly in *Cortinarius* sp. and *L. laccata* could be an additional biotic contributor to hysteresis in soil respiration, which has previously been attributed in part to abiotic processes such as gas transport through the soil, and in part to biotic process such as diurnal variation in photosynthetic rate and associated lags in phloem C transport from leaves to soil (Zhang et al., 2015 and references therein). The proposed physical mechanisms for hysteresis in soil respiration do not apply to aboveground fungal tissues, but it is possible that ectomycorrhizal fungal respiration could be constrained by temporal variation in host C supply driven by diurnal variation in photosynthetically active radiation (PAR), given their dependence on host carbohydrates. Although we were not able to test this hypothesis, with more information on the lags associated with transport from leaves to mycelium it might be possible to infer the importance of PAR-mediated variation in carbohydrate supply from the pattern of hysteresis. In the present study, the pattern in *L. laccata* (associated with young white pine) was counterclockwise, whereas the hysteresis in *Cortinarius* sp. (associated with large canopy oak trees) was clockwise. Zhang et al. (2015) modeled hysteresis in soil respiration and found that as the lag between photosynthesis and soil respiration increased from 0 to 7 h, the direction of the hysteresis reversed from clockwise to counterclockwise. Although the time lags for photosynthate transport in the present study are unknown, delays can be on the order of hours to weeks depending on plant size (Zhang et al., 2015), pointing to the possibility that the observed opposing patterns of hysteresis are extrinsically driven by differences in host size (the oaks were larger than pines) rather than driven by intrinsic responses to thermal cues. An alternate possibility is that hysteresis is an artifact of lags in temperature response of the whole sporocarp relative to the point of temperature measurement. While this is possible, it does not fit with the variation among sporocarps. For example, the *Laccaria* sporocarps had very thin stipes and caps, and yet showed strong hysteresis, whereas *Hebeloma*, which had thicker sporocarps, did not exhibit hysteresis.

Whatever the driver for these hysteresis patterns, it is clear that hysteretic fungal respiratory dynamics could influence complex ecosystem responses to varying temperature. However, it is important to consider that fungal community respiration will be the sum of the respiratory responses of many species of fungi, each of which might differ in patterns of hysteresis depending on the underlying causes. Therefore we must expand our understanding of the patterns and drivers of thermal responses to temperature in order to understand the diversity of these respiratory responses, and to accurately model fungal community respiration-temperature relationships. Understanding these relationships could also help us predict differential seasonal and interannual responses of species to climatic variation.

5. Conclusions

Our novel approach to *in situ* sporocarp respiration measurement provides a window into the ecophysiology of fungal respiration-temperature relationships, indicating that non-exponential unimodal models provided equal or better fits than exponential models under most conditions, especially for daytime/warming conditions. Incorporation of diurnal unimodal temperature response functions combined with seasonal acclimation and expected patterns of carbon limitation should improve our ability to understand and model terrestrial ecosystem CO_2 exchange. Furthermore, the observed high rates of mass-specific respiration in fungi under field conditions point to their large potential contributions to symbiotic, soil, and ecosystem respiration. Future

studies should develop better field estimates of mass-specific vegetative mycelial respiration in roots and soil.

Author contributions statement

EAL is responsible for the conception, design, data collection and writing of this manuscript.

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Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.funeco.2017.02.001.

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