# Elevational gradients do not significantly alter soil microbial respiration and temperature sensitivity in a subtropical forest

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# Abstract

Soil carbon (C) cycling plays critical role in regulating global C budget and atmosphere CO2 concentration. The ongoing global warming potentially accelerates soil C loss induced by microbial respiration (MR) and makes soil a large C source to atmosphere. Quantifying the drivers of MR and its response to rising temperature (also called temperature sensitivity, Q10) is a high priority in order to improve the modelling and prediction of terrestrial C cycle under global warming. In this study, we applied a standardized soil sampling along 9 gradients from 400 m to 1100 m in a subtropical forest in South China, and conducted the incubation experiment at the same temperature ranges (from 10 °C to 25 °C) to measure MR and Q10, then the measured MR was adjusted by the field temperature of sampling site. Our objectives were to examine the response of MR and Q10 to the environmental change induced by elevational gradients in the subtropical forest, and then quantify their main drivers. We totally collected 54 abiotic and biotic factors relative to the MR and Q10. Our results showed that the incubated MR increased from low to high elevation. However, significantly elevational trend of the adjusted MR was not examined after adjusted by the field temperature of sampling sites, due to the tradeoff between increasing soil C concentration and declining temperature as elevation increased. We further found that the 9 elevational gradients did not cause significant change of Q10. The variation of Q10 was negatively dominated by soil C quality. Since climate warming is predicted faster at high elevation than that at low elevation, C loss from high elevation might be accelerated in the future and need more attentions in the further studies

## Introduction

Soil stores twice carbon (C) more than the atmosphere (Jobbágy and Jackson, 2000; Lal, 2004; Scharlemann et al., 2014), and the decomposition of soil C (also called as soil microbial respiration, MR) releases 6-10 times more CO<sub>2</sub> into atmosphere than the current levels of fossil fuel consumption every year (Boden et al., 2009; Friedlingstein et al., 2021). Thus, soil C cycling plays critical role in regulating global C budget and atmosphere CO<sub>2</sub> concentration (Bond-Lamberty et al., 2018; Friedlingstein et al., 2021). Moreover, large amount of studies has confirmed that MR is critically sensitive to current climate change, especial rising temperature (Davidson and Janssens, 2006; Bond-Lamberty and Thomson, 2010; Bradford et al., 2021) and thus makes soil a large C source to atmosphere in the future (Davidson and Janssens, 2006; Bond-Lamberty and Thomson, 2010; Bond-Lamberty et al., 2018). Quantifying the variations and drivers of MR and its response to rising temperature (also called temperature sensitivity,  $Q_{10}$ ) is a high priority in order to better model and predict terrestrial C cycle under global warming (Zhou et al., 2009).

Elevational gradients are ideal platform to study the response of soil C cycling under climate warming (Kong *et al.*, 2022). Different elevational gradients cause various climate levels, such as temperature and precipitation (He *et al.*, 2021; Kong *et al.*, 2022), but with similar soil parent material and plant species pool. Therefore, elevational gradients could provide more realistic insight in the underlying mechanism driving MR and  $Q_{10}$  (Conant*et al.*, 2011; Longbottom *et al.*, 2014). Various studies found that MR declined along the elevational gradients (Garten and Hanson, 2006; Gutiérrez-Girón *et al.*, 2015), since warmer soils in low elevation contribute to more active soil microorganisms and high decomposition rates (Gutiérrez-Girón *et al.*, 2015). In contrast, there is also study suggesting that elevation positively affects MR (Kong *et al.*, 2022), due to high soil C concentration at high elevation that offset the negative effect of low temperature. Therefore, the net elevational effect on MR depends on the tradeoff between climate and respirated substrate along the elevation. More studies are still needed to figure out this question and its regional characteristic.

Lab incubation is a commonly used method to determine the microbial respiration, which usually incubates multiple soil samples at the same time under the same temperature or temperature range (Ding *et al.*, 2016; Liu *et al.*, 2017; Li *et al.*, 2020; Zhang*et al.*, 2022). The unified incubation temperature might be too high to samples from cold sites, while too low to samples from warm sites (Li *et al.*, 2020). However, it's critical difficulty to set a specific incubation temperature for each soil samples in the lab incubation. Instead, recent studies used an adjusted MR by the field temperature of each site based on the unify incubation temperature, which easily solve the difference between field temperature and incubated temperature (Li *et al.*, 2020).

Temperature sensitivity ( $Q_{10}$ ) of MR also serve as a reference for how regional C pools may respond to future warming (Davidson and Janssens, 2006). To date, there is still no consistent elevational trend of temperature sensitivity along the elevational gradients. Several studies suggest that high elevation increases  $Q_{10}$  (Gutiérrez-Girón *et al.*, 2015; Kong*et al.*, 2022; Okello *et al.*, 2022; Zeng *et al.*, 2022), while others reported higher  $Q_{10}$  at lower elevations (Lipson, 2007), or no significantly elevational trend (Schindlbacher *et al.*, 2010; Xu *et al.*, 2014; Wang*et al.*, 2016; Zuo *et al.*, 2021). This indicates that the current understanding of the elevational effect on  $Q_{10}$  is not comprehensive enough.

To further quantify the underlying mechanism in driving MR and  $Q_{10}$ , we applied a standardized sampling along 9 elevational gradients from 400 m to 1100 m in a subtropical forest in South China. These soil samples were incubated in the lab to determine MR and  $Q_{10}$ . Our objectives were to examine the response of MR and  $Q_{10}$  to the environmental change induced by elevational gradients in the subtropical forest, and then quantify their main drivers. We hypothesized that: 1) high elevation reduces MR but increase  $Q_{10}$ , and 2) the varied MR and  $Q_{10}$  along elevation would be largely explained by the soil and plant community structure and environmental change induced by the elevational difference.

# Method and materials

## Site description

This study was conducted in Chebaling National Nature Reserve in the Guangdong Province of southern China (114°09'–114deg16'E, 24deg40'–24deg46'N), with elevation ranging from 330 m to 1256 m above sea level. The climate is a typical subtropical monsoon, with mean annual temperature and mean annual precipitation with 19.6 degC and 1,468 mm (He *et al.*, 2021). The vegetation is well-preserved subtropical evergreen broad-leaved forests, dominated by *Schima superba*, *Machilus chinensis*, and *Eurya nitida*. Soils are classified in the ultisol order and the adult suborder according to the USDA soil classification system (Zhou et al., 2013).

## Field sampling

The field samplings were conducted in an elevational gradients, including nine permanent plots  $(40 \times 40 \text{ m})$  and ranging from 300 to 1100 m asl. To reduce the influence of aspect, all plots were located on the south side. All trees with a diameter at breast height above 1 cm in each plot were surveyed.

The field soil sampling was conducted in October 2018. In each plot, five subplots  $(10 \times 10 \text{ m})$  were randomly selected. In each subplot, five litter samples were randomly collected in five 1 m x 1 m squares. After that,

five soil cores (3.5 cm in diameter and 20 cm in depth) were collected and mixed as one sample. In total, we have 45 soil samples from the elevational gradients (9 gradients \* 5 soil samples each gradient). All soil samples were seized by a 2-mm mesh to remove the stone, visible roots were collected as the root samples and the rest soil was collected as soil samples. The living roots were separated into coarse root and fine root (with diameter < 2 mm) by the root diameter. The litter and fine root samples were oven-dry at  $65^{\circ}$ C for 48 h, to determine the litter biomass and fine root biomass in each subplot. Meanwhile, soil water holding capacity (WHC) was measured using the ring knife method.

## Soil and plant physicochemical property

Then the litter and root samples were used to determine C and N concentration, by using CHNOS Elementar Analyzer (Vario EL III, Elementar Inc., Hanau, Germany), and P concentration by using ultraviolet spectrophotometer (UV-2550, Shimadzu, Kyoto, Japan).

Soil samples were separated into 4 parts. One was air-dried and used to determine soil physicochemical property. Soil C and Soil N were determined by CHNOS Elementar Analyzer, soil available N was determined by using continuous flow analyzer (San++, Skalar, Breda, the Netherlands), Soil P and available P were determined by using ultraviolet spectrophotometer, pH was determined by a pH meter (FE20–FiveEasy), soil texture as reflected by the weight percentages of sand, silt and clay was determined by the hydrometer method (Ashworth*et al.*, 2001). The second part was freeze-dried and used to measure the phospholipid fatty acids (PLFAs), to represent the microbial community structure, including community total PLFAs and the components of bacteria, gram-positive bacteria, gram-negative bacteria, actinomycetes, fungi (Frostegard and Baath, 1996). The ratios between these components were used to represent the relative composition of microbial community. The third part was stored at 4 °C and used to measure soil microbial biomass C, N and P, using chloroform funigation extraction technique. The forth part was incubated in the lab to determine microbial respiration (MR) and its temperature sensitivity ( $Q_{10}$ ).

## Soil incubation and measurements

We performed the lab incubation experiment under varying temperature to quantify the MR and  $Q_{10}$  of the 45 soil samples. The incubation and measurement were conducted using a 32-channel microbial respiration automatic measurement system in lab. For detail about this measurement system, please refer to (Zhang *et al.*, 2022).

Before measurement, all soil samples were adjusted to 60% WHC to maximum soil microbial activity (Zhou et al. , 2014). 40 gram dry-weight soil for each soil sample was placed in a bottle (250 ml) for incubation. The measurement last 4-weeks after one-week pre-incubation. During the pre-incubation and measurement, the artificial weather box experienced the same temperature cycle every day, i.e., 10 °C-15°C-20 °C-25 °C-20°C-15 °C-10 °C. All soil samples were maintained with 60% water holding capacity by weekly weighing the soil containers and adding distilled water to compensate for water loss. Each temperature last 240-minute, in which the former 80-minute was equilibration time and the latter 160-minute was for measurement. Each measurement lasts 150 s, with the  $60^{\rm th}$ -140<sup>th</sup> s data for determining the soil C decomposition rate. The rate of soil C decomposition at different temperature was used to calculate the daily accumulated MR and  $Q_{10}$  following Eq-1 (Lloyd and Taylor, 1994).

# $MR = ae^{bT}$ (1)

where MR was the rate of soil C decomposition at specific incubated temperature, T was the incubated temperature, *a* represent the base respiration at T = 0 for each soil sample (MR\_0). *b* was parameter of the exponential equation and used to calculate  $Q_{10}$  value, following Eq. (2)

$$Q_{10} = e^{10b}$$
 (2)

MR at each specific field temperature was calculated following:

 $MR_{-}MAT = ae^{bT_{-}MAT}$  (3)

Where MR\_MAT is MR at the specific field mean annual temperature (MAT) at each plot, and T\_MAT is the field MAT at each plot.

For the four weeks incubation, we calculated the accumulated microbial respiration, such as AccMR for the incubated respiration, AccMR\_0 for the base respiration at T = 0, AccMR\_MAT for the adjusted respiration. AccMR\_0 was further normalized by soil C concentration (AccMR\_0\_perSC) to represent the quality of soil C substrate (Creamer *et al.*, 2014; Ding *et al.*, 2016).

### Statistical analysis

MAT and mean annual precipitation (MAP) of each plot were extracted from the relevant latitude and longitude of the global climate layers of WorldClim (1 km<sup>2</sup> spatial resolution; http://www.worldclim.org/) using the extract function in the "raster" R package (v. 2.6.7), and the aspect of each plot was obtained by analyzing the digital elevation model data using the "raster" R package (Hijmans, 2020). We calculated the above-ground biomass (AGB) using a pantropical model (Chave *et al.*, 2014) in the "BIOMASS" R package (v. 2.1.1) (Rejou-Mechain *et al.*, 2017).

In this study, we totally collected 54 abiotic and biotic factors relative to the MR and  $Q_{10}$ . Regression analyses were used to examine the elevational trends of all response variables. Then, the 54 factors were clarified into seven groups: topography (elevation, aspect) and climate (MAT and MAP), soil environment (WHC, pH), soil texture (bulk density, sand content, silt content and clay content), plant community structure (AGB, species richness, Shannon diversity index (H'), Simpson index), plant carbon input (litter C, N, P concentration and C:N ratio (litter CNR), C:P ratio (litter CPR), N:P ratio (litter NPR), fine root biomass, fine root C, N, P concentration and C:N ratio (root CNR), C:P ratio (root CPR), N:P ratio (root NPR)), soil organic matter (soil C, N, P, C:N ratio (soil CNR), C:P ratio (soil CPR) and N:P ratio (soil NPR), soil available N and P), soil microbial biomass (soil microbial biomass C, N, P, C:N ratio (microbial CNR), C:P ratio (microbial CPR) and N:P ratio (microbial NPR)), soil microbial community structure (total phospholipid fatty acids, and its components of bacteria, gram-positive bacteria, gram-negative bacteria, actinomycetes, fungi, and gram-positive: negative bacteria ratio (GNR), actinomycetes: bacteria ratio (ABR), actinomycetes: fungi ratio (AFR), fungi: bacteria ratio (FBR)).

For each group of factors, we performed all subsets regression analysis to select the best model that had the lowest Bayesian information criterion (BIC) in predicting AccMR\_MAT and  $Q_{-10}$ , respectively (Table S1 and S2). If the difference of BIC was < 2 units (Burnham and Anderson, 2002), we obtained the model with the highest adjust  $R^{-2}$ . Using this approach, we selected 14 and 12 variables from the best models for AccMR\_MAT and  $Q_{-10}$ , respectively. The unselected variables either had no significant influence on AccMR\_-MAT or  $Q_{-10}$ , or were highly collinear with the selected variables. Then, the selected variables were used in structural equation modelling (SEM) to explain the variation of AccMR\_MAT and  $Q_{-10}$  along the elevation. We dropped the non-significant path and variables in the SEM to simplify the initial model and improve the model fit. The indirect effect of each predictor was calculated by multiplying the standardized direct effects of a given predictor on AccMR\_MAT or  $Q_{-10}$  via mediator in one route, and then we summed the multiple indirect effects and direct effect of the given predictor to quantify its total effect (Lefcheck, 2016). All the analyses were conducted in R 3.3.4. We used packages corrplot (Wei and Simko, 2013), leaps, piecewiseSEM (Lefcheck, 2016).

### Results

During the four weeks incubation, daily microbial respiration rates at the incubation temperature (MR) and adjusted at the field MAT (MR\_MAT), base respiration at 0 (MR\_0), and their normalizations by soil C concentration were all were all declining over time in all elevational gradients. While, there was no significant temporal trend of  $Q_{10}$  over the four weeks (Fig. S1-S2).

Four-week accumulated MR at incubated temperature (AccMR) and base respiration at 0 (AccMR\_0) increased as elevation increased, while the adjust accumulated MR at the field MAT (AccMR\_MAT) and  $Q_{10}$  did not vary significant along with the elevation. The normalized AccMR\_MAT and AccMR\_0 by soil C

concentration (AccMR\_MAT\_perSC and AccMR\_0\_perSC) both decreased nonlinearly as elevation increased (Fig. 1, S3-S6).

In the selected model for each group of factors (Fig. S7-S12), AccMR\_MAT was positively affected by pH and WHC in the soil environment factors, Root P concentration and Root NPR in the carbon input factors, soil available N and NPR in soil organic matter factors, negatively affected by Actinomyces in soil microbial factors. In total, soil organic matter explained the largest variation of AccMR\_MAT (0.24), followed by soil environment (0.13) and soil microbial community (0.10, Figure 2a).

 $Q_{10}$  was positively affected by fine root biomass in carbon input factors, negatively affected by litter N concentration in carbon input factors. In total, carbon input factors explained the largest variation of  $Q_{10}$  (0.13, Figure 2b).

SEM analysis showed that 71% variation of AccMR\_MAT was explained by the investigated abiotic and biotic factors (Figure 3a). The majority of this explanation was directly contributed by the positive effects of soil pH, AccMR\_0\_perSC and WHC, with standardized effects of 0.63, 0.54 and 0.51, respectively (Figure 4a). We further found that soil microbial C caused key indirect effect on AccMR\_MAT. Moreover, MAT, aspect and MAP also caused significant positive effect on the variation of AccMR\_MAT, with standardized total effects of 0.27, 0.10 and 0.09, respectively; while elevational gradients caused significant negative effect on AccMR\_MAT, with standardized total effect of -0.16 (Figure 4a).

Variation of  $Q_{10}$  in the SEM was explained 38%, which was contributed by the directly negative effect from AccMR\_0\_perSC, elevation and litter N concentration, with standardized effects of -0.62, -0.43 and -0.28, respectively (Figure 3b). In total, we further found that WHC, Aspect, fine root biomass, MAP and microbial NPR caused indirectly positive effect on  $Q_{10}$ , all via MR\_0\_perSC (Figure 4b).

## Discussion

## Elevational variation and drivers of soil microbial respiration

In this study, the incubated AccMR increased along with the elevation. This phenomenon is similar to other incubation studies (Kong *et al.*, 2022), but opposite to the field measurements that soil respiration declines from low to high elevation (Garten and Hanson, 2006; Gutierrez-Giron *et al.*, 2015). In the lab measurement, since all soil samples were incubated at the same temperature, high AccMR at high elevation was largely contributed by its high soil C concentration and microbial biomass C (Figs. S10-S11). However, field temperature is declining from low to high elevation (Fig. S7). After adjust by the field MAT, no significant elevational trend of AccMR\_MAT was examined, because the low MAT of sampled high elevation limits microbial activity, thus, offsetting the positive effect from high SOC concentration. However, most previous incubation studies used a unify temperature (or temperature range) no matter where soil was sampled, the unified incubation temperature is relatively high to cold site samples and potentially overestimates their soil C release (Li *et al.*, 2020; Zhang *et al.*, 2022). Future incubation experiment should pay more attention on this uncertainty, by using different incubating temperature or at least adjusting the C release by the natural temperature gradients of each sampling site (Li*et al.*, 2020).

In this study, the declining AccMR\_MAT from low to high elevation was dominantly contributed by soil environments (pH and WHC) and soil C quality (AccMR\_0\_perSC), followed by soil microbial biomass C, MAT, aspect and MAP. The dominant factors were similar to previous studies that soil micro-environments (Ding *et al.*, 2016; Li *et al.*, 2020; Zhang *et al.*, 2022) and initial quality of soil C (Guo *et al.*, 2022) played critical role in regulating microbial respiration. While our results were differ to other studies that suggests the critical role of soil microbe in determining soil C decomposition (Colman and Schimel, 2013). Possible explanation is that soil microbe is highly collinear with soil pH and WHC, the influence of soil microbe was implied in the effect of soil pH and WHC. These results suggest that soil environment and soil C quality are much important in determining soil C decomposition.

Elevational variation and drivers of temperature sensitivity of microbial respiration

Moreover, we also found that  $Q_{10}$  did not significantly vary along with the elevation. Similar result was also observed in a recent global meta-analysis (Li *et al.*, 2022) and several observation studies (Schindlbacher *et al.*, 2010; Xu*et al.*, 2014; Zuo *et al.*, 2021), suggesting that microbial response to temperature was similar among different elevational gradients. Such results did not support the microbial thermal adaption that  $Q_{10}$  declines from low to high temperature site (or from high to low elevation) (Bradford *et al.*, 2019). Moreover, our SEM analysis revealed no significant direct and indirect effect from MAT on  $Q_{10}$ , suggesting that no significant influence of elevation and temperature on  $Q_{10}$  in the elevational gradients. Since many studies predicted climate warming is faster at high than that at low elevation (Pepin *et al.*, 2015), the unified  $Q_{10}$  means future climate warming will cause more C loss at the high elevation. Meanwhile, other studies reported different elevational trend of  $Q_{10}$  that  $Q_{10}$  increased (Gutierrez-Giron *et al.*, 2015; Kong *et al.*, 2022; Okello *et al.*, 2022; Zeng*et al.*, 2022) or decreased (Lipson, 2007) along with the elevation, suggesting that the elevational trend of  $Q_{10}$  still need more studies to confirm.

In this study, the unchanged  $Q_{10}$  along with elevation might be due to tradeoff between the directly negative effect from elevation and its indirectly positive effect. All these indirect influences were via the pathway of AccMR\_0\_perSC, showing that high elevation indirectly enhanced  $Q_{10}$  via reducing AccMR\_0\_perSC. Low AccMR\_0\_perSC means low soil C quality and contributes to high  $Q_{10}$ , similar result was also observed in previous studies (Ding *et al.*, 2016). These findings support previous carbon-quality-temperature hypothesis that  $Q_{10}$  of low quality soil C is greater than that of the high quality soil C (Fierer *et al.*, 2006; Liu *et al.*, 2017; Wang *et al.*, 2018; Bradford *et al.*, 2021), and confirm that soil C quality is a good predictor of  $Q_{10}$  value (Davidson and Janssens, 2006).

In summary, elevational variation of soil microbial respiration and its temperature sensitivity in a subtropical forest in South China were assessed by using lab incubation experiment. We found that incubated AccMR increased from low to high elevation. However, significantly elevational trend of AccMR\_MAT was not examined after adjusted by the field temperature, due to the tradeoff between increasing Soil C concentration and declining field temperature. We further found that  $Q_{10}$  did not vary significantly along the subtropical forest elevational gradients, the variation of  $Q_{10}$  was negatively dominated by soil C quality (AccMR\_0\_perSC). Since climate warming is predicted faster at high elevation than that at low elevation, C loss from high elevation might be accelerated in the future and need more attentions.

## Data accessibility

All data will be available in the Supplementary materials after acceptance.

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## References

Ashworth, J., Keyes, D., Kirk, R., Lessard, R., 2001. Standard procedure in the hydrometer method for particle size analysis. Commun. Soil. Sci. Plan. 32, 633-642.

Boden, T.A., Marland, G., Andres, R.J., 2009. Global, regional, and national fossil-fuel CO2 emissions. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, US Department of Energy, Oak Ridge, Tenn., USA doi 10.

Bond-Lamberty, B., Bailey, V.L., Chen, M., Gough, C.M., Vargas, R., 2018. Globally rising soil heterotrophic respiration over recent decades. Nature 560, 80-83.

Bond-Lamberty, B., Thomson, A., 2010. Temperature-associated increases in the global soil respiration record. Nature 464, 579-582.

Bradford, M., Wood, S., Addicott, E., Fields, N., Gonzalez-Rivero, J., Jevon, F., Maynard, D., Oldfield, E., Polussa, A., Ward, E., Wieder, W., 2021. Quantifying microbial control of soil organic matter dynamics at macrosystem scales. Biogeochemistry, In press.

Bradford, M.A., McCulley, R.L., Crowther, T.W., Oldfield, E.E., Wood, S.A., Fierer, N., 2019. Cross-biome patterns in soil microbial respiration predictable from evolutionary theory on thermal adaptation. Nature Ecology & Evolution.

Burnham, K.P., Anderson, D.R., 2002. Model selection multimodel inference: a practical information-theoretic approach, 2nd ed. Springer, New York.

Chave, J., Rejou-Mechain, M., Burquez, A., Chidumayo, E., Colgan, M.S., Delitti, W.B.C., Duque, A., Eid, T., Fearnside, P.M., Goodman, R.C., Henry, M., Martinez-Yrizar, A., Mugasha, W.A., Muller-Landau, H.C., Mencuccini, M., Nelson, B.W., Ngomanda, A., Nogueira, E.M., Ortiz-Malavassi, E., Pelissier, R., Ploton, P., Ryan, C.M., Saldarriaga, J.G., Vieilledent, G., 2014. Improved allometric models to estimate the aboveground biomass of tropical trees. Global. Change. Biol. 20, 3177-3190.

Colman, B.P., Schimel, J.P., 2013. Drivers of microbial respiration and net N mineralization at the continental scale. Soil Biology and Biochemistry 60, 65-76.

Conant, R.T., Ryan, M.G., Agren, G.I., Birge, H.E., Davidson, E.A., Eliasson, P.E., Evans, S.E., Frey, S.D., Giardina, C.P., Hopkins, F.M., Hyvonen, R., Kirschbaum, M.U.F., Lavallee, J.M., Leifeld, J., Parton, W.J., Steinweg, J.M., Wallenstein, M.D., Wetterstedt, J.A.M., Bradford, M.A., 2011. Temperature and soil organic matter decomposition rates - synthesis of current knowledge and a way forward. Global. Change. Biol. 17, 3392-3404.

Creamer, R.E., Schulte, R.P.O., Stone, D., Gal, A., Krogh, P.H., Lo Papa, G., Murray, P.J., Peres, G., Foerster, B., Rutgers, M., Sousa, J.P., Winding, A., 2014. Measuring basal soil respiration across Europe: Do incubation temperature and incubation period matter? Ecological Indicators 36, 409-418.

Davidson, E.A., Janssens, I.A., 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. Nature 440, 165-173.

Ding, J., Chen, L., Zhang, B., Liu, L., Yang, G., Fang, K., Chen, Y., Li, F., Kou, D., Ji, C., 2016. Linking temperature sensitivity of soil CO2 release to substrate, environmental, and microbial properties across alpine ecosystems. Global. Biogeochem. Cy. 30, 1310-1323.

Fierer, N., Colman, B.P., Schimel, J.P., Jackson, R.B., 2006. Predicting the temperature dependence of microbial respiration in soil: A continental-scale analysis. Global. Biogeochem. Cy. 20, GB3026.

Friedlingstein, P., Jones, M.W., O'Sullivan, M., Andrew, R.M., Bakker, D.C.E., Hauck, J., Le Quere, C., Peters, G.P., Peters, W., Pongratz, J., Sitch, S., Canadell, J.G., Ciais, P., Jackson, R.B., Alin, S.R., Anthoni, P., Bates, N.R., Becker, M., Bellouin, N., Bopp, L., Chau, T.T.T., Chevallier, F., Chini, L.P., Cronin, M., Currie, K.I., Decharme, B., Djeutchouang, L., Dou, X., Evans, W., Feely, R.A., Feng, L., Gasser, T., Gilfillan, D., Gkritzalis, T., Grassi, G., Gregor, L., Gruber, N., Gurses, O., Harris, I., Houghton, R.A., Hurtt, G.C., Iida, Y., Ilyina, T., Luijkx, I.T., Jain, A.K., Jones, S.D., Kato, E., Kennedy, D., Klein Goldewijk, K., Knauer, J., Korsbakken, J.I., Kortzinger, A., Landschutzer, P., Lauvset, S.K., Lefevre, N., Lienert, S., Liu, J., Marland, G., McGuire, P.C., Melton, J.R., Munro, D.R., Nabel, J.E.M.S., Nakaoka, S.I., Niwa, Y., Ono, T., Pierrot, D., Poulter, B., Rehder, G., Resplandy, L., Robertson, E., Rodenbeck, C., Rosan, T.M., Schwinger, J., Schwingshackl, C., Seferian, R., Sutton, A.J., Sweeney, C., Tanhua, T., Tans, P.P., Tian, H., Tilbrook, B., Tubiello, F., van der Werf, G., Vuichard, N., Wada, C., Wanninkhof, R., Watson, A., Willis, D., Wiltshire, A.J., Yuan, W., Yue, C., Yue, X., Zaehle, S., Zeng, J., 2021. Global Carbon Budget 2021. Earth Syst. Sci. Data Discuss. 2021, 1-191.

Frostegard, A., Baath, E., 1996. The use of phospholipid fatty acid analysis to estimate bacterial and fungal biomass in soil. Biol. Fert. Soils. 22, 59-65.

Garten, C.T., Hanson, P.J., 2006. Measured forest soil C stocks and estimated turnover times along an elevation gradient. Geoderma 136, 342-352.

Guo, M., Zhao, B., Wen, Y., Hu, J., Dou, A., Zhang, Z., Rui, J., Li, W., Wang, Q., Zhu, J., 2022. Elevational pattern of soil organic carbon release in a Tibetan alpine grassland: Consequence of quality but not quantity of initial soil organic carbon. Geoderma 428, 116148.

Gutierrez-Giron, A., Diaz-Pines, E., Rubio, A., Gavilan, R.G., 2015. Both altitude and vegetation affect temperature sensitivity of soil organic matter decomposition in Mediterranean high mountain soils. Geoderma 237-238, 1-8.

He, X., Chu, C., Yang, Y., Shu, Z., Li, B., Hou, E., 2021. Bedrock and climate jointly control the phosphorus status of subtropical forests along two elevational gradients. Catena 206, 105525.

Hijmans, R.J., 2020. raster: Geographic Data Analysis and Modeling. R package version 3.0-12.

Jobbagy, E.G., Jackson, R.B., 2000. The vertical distribution of soil organic carbon and its relation to climate and vegetation. Ecol. Appl. 10, 423-436.

Kong, J., He, Z., Chen, L., Zhang, S., Yang, R., Du, J., 2022. Elevational variability in and controls on the temperature sensitivity of soil organic matter decomposition in alpine forests. Ecosphere 13, e4010.

Lal, R., 2004. Soil carbon sequestration to mitigate climate change. Geoderma 123, 1-22.

Lefcheck, J.S., 2016. piecewiseSEM: Piecewise structural equation modelling in r for ecology, evolution, and systematics. Methods in Ecology and Evolution 7, 573-579.

Li, C., Li, M., Xu, L., He, N., 2022. A global synthesis of patterns in soil organic matter and temperature sensitivity along the altitudinal gradient. Frontiers in Environmental Science 10, 959292.

Li, J., Nie, M., Pendall, E., Reich, P., Pei, J., Noh, N., Zhu, T., Li, B., Fang, C., 2020. Biogeographic variation in temperature sensitivity of decomposition in forest soils. Global. Change. Biol. 26, 1873-1885.

Lipson, D.A., 2007. Relationships between temperature responses and bacterial community structure along seasonal and altitudinal gradients. Fems. Microbiol. Ecol. 59, 418-427.

Liu, Y., He, N., Zhu, J., Xu, L., Yu, G., Niu, S., Sun, X., Wen, X., 2017. Regional variation in the temperature sensitivity of soil organic matter decomposition in China's forests and grasslands. Global. Change. Biol. 23, 3393-3402.

Lloyd, J., Taylor, J.A., 1994. On the Temperature-Dependence of Soil Respiration. Funct. Ecol. 8, 315-323.

Longbottom, T.L., Townsend-Small, A., Owen, L.A., Murari, M.K., 2014. Climatic and topographic controls on soil organic matter storage and dynamics in the Indian Himalaya: Potential carbon cycle–climate change feedbacks. Catena 119, 125-135.

Okello, J., Bauters, M., Verbeeck, H., Bode, S., Kasenene, J., Francoys, A., Engelhardt, T., Butterbach-Bahl, K., Kiese, R., Boeckx, P., 2022. Temperature sensitivity of soil organic carbon respiration along the Rwenzori montane forests elevational transect in Uganda. Biogeosciences Discuss. 2022, 1-33.

Pepin, N., Bradley, R.S., Diaz, H.F., Baraer, M., Caceres, E.B., Forsythe, N., Fowler, H., Greenwood, G., Hashmi, M.Z., Liu, X.D., Miller, J.R., Ning, L., Ohmura, A., Palazzi, E., Rangwala, I., Schoner, W., Severskiy, I., Shahgedanova, M., Wang, M.B., Williamson, S.N., Yang, D.Q., Mountain Research Initiative, E.D.W.W.G., 2015. Elevation-dependent warming in mountain regions of the world. Nat. Clim. Change. 5, 424-430.

Rejou-Mechain, M., Tanguy, A., Piponiot, C., Chave, J., Herault, B., 2017. biomass: an r package for estimating above-ground biomass and its uncertainty in tropical forests. Methods in Ecology and Evolution 8, 1163-1167.

Scharlemann, J.P., Tanner, E.V., Hiederer, R., Kapos, V., 2014. Global soil carbon: understanding and managing the largest terrestrial carbon pool. Carbon Management 5, 81-91.

Schindlbacher, A., de Gonzalo, C., Diaz-Pines, E., Gorria, P., Matthews, B., Inclan, R., Zechmeister-Boltenstern, S., Rubio, A., Jandl, R., 2010. Temperature sensitivity of forest soil organic matter decomposition along two elevation gradients. Journal of Geophysical Research: Biogeosciences 115.

Wang, D., He, N., Wang, Q., LU, Y., Wang, Q., Xu, Z., Zhu, J., 2016. Effects of Temperature and Moisture on Soil Organic Matter Decomposition Along Elevation Gradients on the Changbai Mountains, Northeast China. Pedosphere 26, 399-407.

Wang, Q., Liu, S., Tian, P., 2018. Carbon quality and soil microbial property control the latitudinal pattern in temperature sensitivity of soil microbial respiration across Chinese forest ecosystems. Global. Change. Biol. 24, 2841-2849.

Wei, T., Simko, V., 2013. corrplot: Visualization of a correlation matrix. R package version 0.73 230, 11.

Xu, W., Li, W., Jiang, P., Wang, H., Bai, E., 2014. Distinct temperature sensitivity of soil carbon decomposition in forest organic layer and mineral soil. Scientific Reports 4, 6512.

Zeng, X.-M., Feng, J., Chen, J., Delgado-Baquerizo, M., Zhang, Q., Zhou, X.-Q., Yuan, Y., Feng, S., Zhang, K., Liu, Y.-R., Huang, Q., 2022. Microbial assemblies associated with temperature sensitivity of soil respiration along an altitudinal gradient. Sci. Total. Environ. 820, 153257.

Zhang, B., Jiang, Y., Chen, D., Wu, Z., Li, B., Chu, C., 2022. Highly spatial variation of soil microbial respiration and temperature sensitivity in a subtropical forest. Sci. Total. Environ. 808, 152197.

Zhou, T., Shi, P., Hui, D., Luo, Y., 2009. Global pattern of temperature sensitivity of soil heterotrophic respiration (Q10) and its implications for carbon-climate feedback. Journal of Geophysical Research: Biogeosciences 114, G02016.

Zhou, W., Hui, D., Shen, W., 2014. Effects of Soil Moisture on the Temperature Sensitivity of Soil Heterotrophic Respiration: A Laboratory Incubation Study. PLOS ONE 9, e92531.

Zuo, Y., Zhang, H., Li, J., Yao, X., Chen, X., Zeng, H., Wang, W., 2021. The effect of soil depth on temperature sensitivity of extracellular enzyme activity decreased with elevation: Evidence from mountain grassland belts. Sci. Total. Environ. 777, 146136.

Figure 1 Elevational variation of accumulated MR (AccMR (a), AccMR\_MAT (b) and AccMR\_0 (c)), temperature sensitivity ( $Q_{10}$ , d) and the normalized AccMR\_0 and AccMR\_MAT by soil C concentration (AccMR\_MAT\_perSC (e) and AccMR\_0\_perSC (f)) from 400 m to 1100 m.



Figure 2 Standardized regression coefficients for the explanatory effects included in the selected (a) AccMR<sub>-</sub>MAT and (b)  $Q_{10}$  models. Model  $R^2$  is reported in each subpanel (factors of soil texture, soil environment, soil texture, plant community, carbon input, soil organic matter, soil microbial community in respective) for the selected model. Close circles and open circles indicate a significant (P < 0.05) and nonsignificant (P [?] 0.05) effect on AccMR<sub>-</sub>MAT or  $Q_{10}$  and lines indicate standard errors.



Figure 3 Structural equation modelling analyses to examine the overall effect on accumulated microbial respiration rate (AccMR\_MAT, a) and temperature sensitivity ( $Q_{10}$ , b). The model fit the data well (AccMR\_MAT: Fisher's C=172.98, P=0.00, Df=94, AIC=256.98, BIC=332.86, N=45;  $Q_{10}$ : Fisher's C=170.27, P=0.00, Df=86, AIC=236.27, BIC=295.89, N=45; AccMR\_0\_PerSC: Fisher's C=275.83, P=0.00, Df=126, AIC=367.83, BIC=450.94, N=45;). Solid arrows indicate significant (P < 0.05) positive (black) or negative (red) relationships. Values associated with the arrows represent standardized path coefficients. Widths of significant paths are scaled by standardized path coefficients.





Figure 4 Standardized total effects derived from the SEM for the influences of abiotic and biotic factors on accumulated microbial respiration rate (AccMR\_MAT, a) and temperature sensitivity ( $Q_{10}$ , b).



	Model	Bulk density	Sand	Silt	Clay		
Soil texture	1		*				
	2	*	*				
	3	*	*	*			
	4	*	*	*	*		
		WHC	$_{\rm pH}$				
Soil environment	1	*	-				
	2	*	*				
		AGB	H'	Simpson	Richness		
Plant community	1		*				
	2		*		*		
	3	*	*		*		
	4	*	*	*	*		
		Litter C	Litter N	Litter P	Litter CNR	Litter CPR	Litter NPR
Carbon input	1						
	2						
	3	*					
	4	*			*		
	5	*			*		
	6	*			*		
	7	*			*		
	8	*			*	*	
		Soil C	Soil N	Soil P	Soil CNR	Soil CPR	Soil NPR
Soil organic matter	1						*
	2						*
	3		*	*			
	4		*	*	*		
	5		*	*	*		
	6	*	*	*	*		
	7		*	*	*	*	*
	8	*	*	*	*	*	*
		Microbial C	Microbial N	Microbial P	Microbial CNR	Microbial CPR	Microbial N
Soil microbe	1	*					
	2	*					
	3	*			*		
	4	*				*	*
	5	*			*		
	6	*				*	*
	7	*			*		
	8	*				*	*

Table S1 Summary of all-subsets regressions for the influence of different factor combinations in the six groups on AccMR\_MAT. \* indicates the selected variables in each model. The selected model is marked in bold.

Table S2 Summary of all-subsets regressions for the influence of different factor combinations in the six groups on AccMR\_MAT. \* indicates the selected variables in each model. The selected model is marked in bold.

	Model	Bulk density	Sand	Silt	Clay		
Soil texture	1			*			
	2		*		*		
	3	*	*		*		
	4	*	*	*	*		
		WHC	PH				
Soil environment	1		*				
	2	*	*				
		AGB	$\mathbf{H}'$	Simpson	Richness		
Plant community	1	*					
	2	*		*			
	3	*		*	*		
	4	*	*	*	*		
		Litter C	Litter N	Litter P	Litter CNR	Litter CPR	Litter NPR
Carbon input	1						
	2		*				
	3		*			*	
	4		*	*		*	
	5	*	*			*	*
	6	*	*			*	*
	7	*	*		*	*	*
	8	*	*		*	*	*
		Soil C	Soil N	Soil P	Soil CNR	Soil CPR	Soil NPR
Soil organic matter	1						
	2			*			
	3		*	*			*
	4		*	*	*	*	
	5		*	*	*	*	
	6		*	*	*	*	
	7		*	*	*	*	*
	8	*	*	*	*	*	*
		Microbial C	Microbial N	Microbial P	Microbial CNR	Microbial CPR	Microbial N
Soil microbe	1						*
	2						
	3						*
	4						*
	5						*
	6				*	*	
	7	*	*			*	
	8	*	*			*	

Figure S1 Temporal variation of MR and  $Q_{10}$  during the incubation period (28 days) at different elevations.



Figure S2 Accumulated MR (AccMR, AccMR\_0 and AccMR\_MAT) and the normalized by soil C concentration (AccMR\_perSC, AccMR\_0\_perSC and AccMR\_MAT\_perSC) of different elevational gradients during the 4-week incubation from 400 m to 1100 m.



Figure S3 Elevational variation of the first week accumulated MR (AccMR (a), AccMR\_MAT (b) and AccMR\_0 (c)), temperature sensitivity ( $Q_{10}$ , d) and the normalized AccMR\_0 and AccMR\_MAT by soil C concentration (AccMR\_MAT\_perSC (e) and AccMR\_0\_perSC (f)) from 400 m to 1100 m.



Figure S4 Elevational variation of the second week accumulated MR (AccMR (a), AccMR\_MAT (b) and AccMR\_0 (c)), temperature sensitivity ( $Q_{10}$ , d) and the normalized AccMR\_0 and AccMR\_MAT by soil C concentration (AccMR\_MAT\_perSC (e) and AccMR\_0\_perSC (f)) from 400 m to 1100 m.



Figure S5 Elevational variation of the third week accumulated MR (AccMR (a), AccMR\_MAT (b) and AccMR\_0 (c)), temperature sensitivity ( $Q_{10}$ , d) and the normalized AccMR\_0 and AccMR\_MAT by soil C concentration (AccMR\_MAT\_perSC (e) and AccMR\_0\_perSC (f)) from 400 m to 1100 m.



Figure S6 Elevational variation of the forth week accumulated MR (AccMR (a), AccMR\_MAT (b) and AccMR\_0 (c)), temperature sensitivity ( $Q_{10}$ , d) and the normalized AccMR\_0 and AccMR\_MAT by soil C concentration (AccMR\_MAT\_perSC (e) and AccMR\_0\_perSC (f)) from 400 m to 1100 m.



Figure S7 Elevational variation of MAT (a), MAP (b), bulk density (c), WHC (d), soil sand content (e), soil silt content (f), soil clay content (g) and soil pH value (h) from 400 m to 1100 m.



Figure S8 Elevational variation of AGB (a), NDVI (b), Shannon index (c), Simpson index (d), species richness (e), fine root density (f) from 400 m to 1100 m.



Figure S9 Elevational variation of litter C concentration (a), litter N concentration (b), litter P concentration (c), litter C:N ratio (CNR, d), litter C:P ratio (CPR, e), litter N:P ratio (NPR, f), root C concentration (g), root N concentration (h), root P concentration (i), root C:N ratio (CNR, j), root C:P ratio (CPR, k), root N:P ratio (NPR, l) from 400 m to 1100 m.







Figure S11 Elevational variation of soil microbial C (a), soil microbial N (b), soil microbial P (c), soil microbial C:R ratio (CNR, d), soil microbial C:P ratio (CPR, e), soil microbial N:P ratio (NPR, f) from 400 m to 1100 m.



Figure S12 Elevational variation of total PLFA (a), actinomyces PLFA (b), AMF PLFA (c), fungi PLFA (d), gram\_neg\_bacteria PLFA (e), gram\_pos\_bacteria PLFA (f), bacteria PLFA (g), fungi: bacteria ratio (FBR, h), gram positive:negative ratio (GPNR, i), actinomyces:bacteria ratio (ABR, j), actinomyces:fungi ratio (AFR, k) from 400 m to 1100 m.

