The temperature sensitivity of the decomposition of physical fractions of organic matter of soils with different vegetation coverage 2019-2024

Background and objectives of the study

Soil organic matter (SOM) plays a key role in the global carbon cycle, and it acts as an important carbon reservoir. Therefore, understanding the relationship between environmental changes and rates of the decomposition of soil organic matter is critical for projecting changes in soil carbon fluxes under changing climate conditions (Houghton, 2001). The global warming could thus lead to a net increase of C release to the atmosphere by the terrestrial biosphere, because the extra CO_2 efflux from SOM may cause further warming by forming a positive feedback (Jenkinson et al., 1991). Hence, the investigation of the temperature sensitivity of SOM decomposition, commonly referred to as Q_{10} , is very important for modelling changes in soil C stock (Davidson and Janssens, 2006). In spite of the intensive research, the temperature sensitivity of soil organic carbon (SOC) decomposition remains controversial (Davidson and Janssens, 2006; Hakkenberg et al., 2003; Kirschbaum, 2006). Some studies have shown that a warmer climate may accelerate rates of SOC decomposition and, consequently, the CO_2 emissions (e.g. Sanderman et al., 2003; Trumbore et al., 1996), while others have provided evidence that the response of SOC decomposition to warming is either transient or insensitive (e.g. Domeignoz-Horta et al., 2023; Giardina and Ryan, 2000).

So the sensitivity of the decomposition of soil carbon to warming is a major uncertainty in projections of carbon dioxide concentration and climate. However, no comprehensive study to date that trying to evaluate the effect of confounding factors on the temperature sensitivity of SOM decomposition. Our objectives were (i) to investigate and evaluate the effect of different vegetation cover/land use on the temperature sensitivity of organic matter decomposition, (ii) to investigate the temperature sensitivity of organic matter pools with different stabilisation mechanism, and (iii) to study the effect of organic matter quality on Q_{10} .

Materials and Methods

Site description and sampling

Composite soil samples were collected from the top 20 centimetres of soils with different land use, from a native grassland, a spruce forest, an oak forest and arable area. The sampling sites were on areas very close to each other (within 20 km) in order to prevent climatic factors affecting the transformation of organic matter. The mean annual temperature (MAT) of this area is 9.2 °C, while the mean annual precipitation (MAP) is 725 mm year⁻¹. The soil samples were transported to the laboratory, air dried and passed through a 2 mm sieve. The basic data of the sites are presented in Table 1.

Soil fractionation and incubation experiment

The SOM fractionation was carried out following the Zimmermann fractionation method (Poeplau and Don, 2013). Briefly, each soil sample was subjected to ultrasonic dispersion (22 J ml-1), after which the samples were wet-sieved over a 63 μ m screen in order to separate the silt and clay fraction (s+c) from the sand and stabile aggregates (S+A) and particulate organic

matter (POM). The sieved s+c fraction was centrifuged for 30 minutes at 2000g and dried at 40 °C. The sieving residue (S+A, POM) was isolated by density, using sodium iodide (NaI, $\rho = 1.6 \text{ g cm}^{-3}$). The sodium iodide solution was added to the S+A + POM fraction (> 63 µm) and centrifuged for 15 minutes at a speed of 1000g. The floating POM fraction was decanted and the step was repeated to ensure the complete extraction of POM. Both fractions were carefully washed with deionised water to remove the sodium iodide and then dried at 40 °C.

30 g samples of the s+c and S+A fractions and 1.5 g of the POM fraction were incubated for 357 days at 15, 25 and 35 °C in three replicates. The samples were preincubated at 20 °C for two weeks, because rewetting and sieving affect the availability of SOM for microorganisms and may cause an increase in the rate of respiration (Franzluebbers, 1999). The moisture content of the fractions was determined using the column method (Govindasamy et al., 2023) and the samples were adjusted to 60 % WHC. The evaporated water was replenished at two-week intervals during the incubation.

Site	Description	pH _{H2O}	рНксі	Sand	Silt	Clay	TOC	C/N
					%			
Faber meadow,	Native Grass (Festuca	5.48	4.21	21.7	70.9	7.4	3.1	10.9
Sopron, Hungary	rubra) cca 140 years old							
(N 47°39'56,508";								
E 16°32'57,480")								
Sopron Mountains,	Medium-aged spruce	3.87	2.92	26.4	69.4	4.2	9.8	21.8
Hungary	stands (30-40 years, Picea							
(N 47°39'20,082";	abies)							
E 16°32'28,595")								
Sopron Mountains,	Medium-aged oak stands	4.28	3.40	16.8	78.7	4.5	4.6	13.6
Hungary	(30-40 years, Quercus							
(N 47°39'21,101";	petraea)							
E 16°32'26,645")								
Harka, Hungary	Arable, cultivated,	5.58	5.06	25.8	69.0	5.2	1.9	10.8
(N 47°37'08,208";	fertilised (for 30 years)							
E 16°34'58,404")								

Table 1 Basic properties of the sites

Measurement of CO₂ evolution and elemental analysis

The respired CO_2 of the soil fractions was trapped in 15 ml of 2 M NaOH. The NaOH traps were replaced on days 7, 14, 21, 28, 35, 49, 63, 77, 105, 133, 161, 189, 217, 245, 273, 302, 330 and 357. The amount of CO_2 released from the fractions was measured by titrating the NaOH solutions with 1M HCl in the presence of BaCl₂ (Anderson, 1982).

Calculation of k and Q_{10}

The decomposition reactions were assumed to be first-order single pool kinetics for each fraction. To calculate the decomposition rate constant (k) of SOC in individual fractions the measured points of CO_2 evolution with time were fitted by means of non-linear curve fitting. The response of decomposition the rate (k) to temperature (T) was described by an exponential equation (Hamdi et al., 2013):

 $\mathbf{k} = \mathbf{a} \times \mathbf{e}^{\mathbf{b} \times \mathbf{T}}$,

where a and b are fitted parameters, the unit of T is degrees Celsius. Q_{10} was calculated using the van't Hoff empirical equation (Hamdi et al., 2013): $Q_{10} = e^{10 \times b}$

The chemical composition of SOM

To determine the chemical composition of SOM of fractions Fourier transform infrared (FT-IR) spectroscopy and 13C nuclear magnetic resonance (NMR) spectroscopy was used. All the samples taken on days 0, 35, 77, 105, 133, 166, 189, 217, 273 and 357 were dried at 60 °C before FT-IR measurements. The FT-IR DRIFT (diffuse reflectance) measurements were carried out. Temporal changes in the FT-IR spectra of the samples were evaluated using 2D correlation spectroscopy (2DCOS). The NMR analysis was done only for the initial (day 0) stage of the SOM fractions.

Results

Mass distribution and C and N storage among the soil organic fractions

The soil in the spruce forestland contained a very high mass fraction of POM, more than 13% of the total soil mass, while in oak, grassland and arable soils with mineral organic matter fractions (S+A and s+c) accounting for 95-99% of the mass (Figure 1a).



Figure 1 Mass distribution of soil organic matter fractions (a) and carbon and nitrogen accumulation in the organic matter fractions (b) for sites with different vegetation

Very different proportions of carbon and nitrogen pools were observed for the four soils with different vegetation types. For example, in the case of forest soils high amounts of carbon and nitrogen accumulated in the POM fractions (30-48% for C and 22-43% for N; Figure 1b), despite the fact that the POM fraction was neither physically nor chemically protected. Both inhibiting soil environmental parameters (e.g. pH, texture) and high C input into a given vegetation type may lead to the accumulation of the POM fraction (Hayashi et al., 2023; Soucémarianadin et al., 2019). An unfavourably high C to N ratio (lignin/N ratio) has been found for coniferous leaves (Vesterdal et al., 2008), which was supported by the present data:

the POM fraction of spruce was characterized by a very high C/N ratio, which may be one of the reasons for inhibited decomposition. However, not only the C/N ratio of POM, but the C/N ratio of the whole soil may be limiting factor for decomposition. Consistent with this, the present study revealed a significant correlation between soil C/N ratio and POM-C (Figure 2a). Another environmental factor that should be taken into account in the present case is soil acidity: the unfavourable pH of pine soil reduces microbial activity and thus litter degradation, which can lead to POM accumulation (Figure 2b), as reported by John et al. (2005).



Figure 2 Relationships between the carbon accumulated in POM fractions (POM-C) and the pH (a) and soil C/N ratio (b)

Changes in chemical composition of SOM during the incubation

2D correlation spectroscopy was performed to extract compositional changes in soil organic fractions with time as perturbation (Figure 3). All major organic matter groups were found to be significantly variable during the incubation; even the amount of lipid and lignin-like substances, which are thought to be difficult to degrade, was markedly altered. No clear pattern was seen in this respect between the fractions and soils, but perhaps it is worth noting that the amount of protein-like substances in the mineral fractions was not as susceptible as in POM.

During incubation, the most common carbohydrate-protein and carbohydrate-lignin correlations were most frequently observed. The carbohydrate-protein correlations were negative in all cases, meaning that they changed in opposite directions, one decreasing when the other increased. A similar result was obtained for the carbohydrate-lignin relationship, where there was a negative correlation for all but the arable soil S+A fraction. A carbohydrate-carboxyl correlation was also found in the mineral fractions.

The temporal order of the spectral changes was very varied both for the vegetation types and even more so for the SOM fractions. Typically, changes in compounds considered to be easily mineralizable (carbohydrate, protein) preceded those in compounds that are more difficult to break down (lipid, lignin). However, particularly in the s+c fractions, surprising sequences were found, such as Lipid/Lignin \rightarrow Carbohydrate or Lipid \rightarrow Lignin/Carboxyl/Protein possibly because of the readily available abundant N compounds present in MAOM.



Figure 3 Synchronous and asynchronous spectra for the SOM fractions (POM, S+A and s+c) for grassland, spruce, oak and arable sites. The dark regions show positive, while the light ones show negative correlations

Decomposition rate constants and Q_{10}

Temperature had a significant effect on the rate of organic matter decomposition (k); an increase of magnitude was measured for all fractions as the temperature increased from 15 °C to 35 °C (Table 2). At all three temperatures, the soil of spruceland showed the lowest degradation values for all SOM fractions. In contrast, the organic matter of the grassland soil showed outstanding degradation at all three temperatures. However, the highest k values were found in the S+A fraction of arable soil with values of 0.12, 0.25 and 1.50 year⁻¹ at 15, 25 and 35 °C. No significant difference in degradation rate was found between MAOM and aggregates, with the exception of the arable soil, where the S+A fraction had a very high rate constant at all temperatures. In general, the degradation rate of the labile POM fraction was significantly higher than that of the protected fractions (aggregates and MAOM).

When considering the individual fractions, great variability in Q_{10} values were found in the s+c fraction, with the highest value for grass (3.7) and the lowest for oak (3.0). Variability was less pronounced in the other two fractions, although the high value recorded for aggregates in the arable soil (S+A: 5.4) stood out not only among the S+A fractions, but for all the organic matter fractions. No significant differences were found between the labile (POM) and stabilised C pools (aggregates and MAOM) for either grassland or spruce soil. In contrast, the s+c fraction of oakland had significantly lower temperature sensitivity than the S+A and POM fractions of the same soil.

Although no significant differences were generally found in the Q_{10} values of the SOM fractions of different land uses, the aggregate fraction of the cultivated soil exhibited significantly higher temperature sensitivity (Table 2). Previous studies demonstrated that frequent tillage, combined with chemical fertilisers, reduced the formation and stability of macro-aggregates, and exposed the formerly occluded C to microbial attack (Poeplau et al., 2020; Tamura and Suseela, 2021). Furthermore, the disintegration of macro-aggregates could cause higher Q_{10} for micro-aggregate carbon in tilled, fertilised soil, because of the greater substrate supply within micro-aggregates and the poor protection of carbon (Wankhede et al., 2020). The extremely high Q_{10} value obtained for cultivated land in the present study, compared to the other land uses, highlighted the fact that the temperature sensitivity of cultivated soils could not be fitted to current organic C degradation models, since land use changes that reduce soil C may accelerate decomposition-derived CO₂ emission during warming to a greater extent than is predicted by currently used SOM models (Wagai et al., 2013).

		k 15	k 25	k 35	MRT ₁₅	MRT ₂₅	MRT ³⁵	Q10
Land								
use	Fraction	year ⁻¹			year			
Grass	s+c	0.07^{e^*}	0.15 ^f	0.61 ^f	14.2	6.6	1.6	3.71 ^b
	S+A	0.07 ^c	0.14 ^{ef}	0.57 ^{ef}	14.7	7.0	1.8	3.66 ^b
	POM	0.09^{f}	0.21 ^g	0.83 ^g	11.0	4.7	1.2	3.60 ^b
Spruce	s+c	0.03 ^a	0.06 ^a	0.20^{a}	36.2	17.3	4.9	3.24 ^{ab}
	S+A	0.04 ^b	0.08^{ab}	0.28 ^{ab}	27.4	12.7	3.6	3.27 ^{ab}
	POM	0.06 ^{de}	0.16 ^f	0.56 ^{ef}	15.5	6.2	1.8	3.3 ^{ab}
Oak	s+c	0.05 ^c	0.11 ^{cd}	0.33 ^{bc}	21.0	9.4	3.0	2.96 ^a
	S+A	0.05 ^c	0.10 ^{bc}	0.40 ^{cd}	20.2	10.0	2.5	3.58 ^b
	POM	0.07 ^e	0.16 ^f	0.65^{f}	14.2	6.1	1.5	3.66 ^b
Arable	s+c	0.06 ^d	0.13 ^{de}	0.49 ^{de}	17.0	8.0	2.0	3.58 ^b
	S+A	0.12 ^g	0.25 ^h	1.50 ^h	8.4	4.0	0.7	5.38 ^c
	POM	0.07 ^e	0.16 ^f	0.64 ^f	14.5	6.2	1.6	3.67 ^b

Table 2 Decomposition rate constant (k), mean residence time (MRT) and Q_{10} for soils with different land use

^{*} Different letters mean significant differences within column based on Tukey's post hoc test.

Relationship between the chemical composition of SOM and Q10

Among the factors that determine the degradation of organic matter, the amount of di-O-alkyl and O-aryl compounds showed a clear positive correlation with the k values calculated at the three temperatures, while the hydrophobic and hydrophilic ratio (HB/HI), alkyl and Alkyl/O-alkyl ratio exhibited a negative correlation (Figure 4a). The chemical composition of the organic matter, in terms of the relative proportions of aryl, methoxy and carbonyl-like compounds and the GP/GN ratio, was positively correlated with Q_{10} , the parameter expressing the temperature sensitivity of organic matter degradation. In addition, a strong negative correlation was found between the C/N ratio of the organic matter, the relative amount of O-alkyl compounds and the Q_{10} values.

The high abundance of aromatic compounds in OM is often considered to make it more stable, since these energy-rich compounds restrict microbial availability and decomposition (Weishaar et al., 2003). Gram-positive bacteria are known to prefer older, more complex organic carbon, whereas Gram-negative bacteria preferentially use fresh OM as SOC sources (Kramer and

Gleixner, 2006). The positive relationship between Q_{10} and the GP/GN ratio thus supports the kinetic theory. A strong negative correlation was found between the Q_{10} values and the O-alkyl content of SOM (Fig. 2a). Wagai et al. (2013) noted that the low temperature sensitivity of organic matter could be due to the abundance of labile, O-alkyl type compounds, which is consistent with the present findings. The C/N ratio also proved to have a negative influence on the temperature sensitivity of OM. There is controversy in the literature as to whether the C/N ratio controls the temperature sensitivity of organic matter decomposition, since some experiments indicate a positive correlation between Q10 and the C/N ratio (Meyer et al., 2018; Wang et al., 2018), and others a negative relationship (Liu et al., 2024; Wei et al., 2022). These differences are mainly due to methodological differences, since both the experiments performed by Meyer et al. and Wang et al. were very short incubations (12 h and 21 days), in which the temperature sensitivity is dominated by the degradation of labile organic matter, whereas protected pools also played a role in the temperature sensitivity of SOM in the one-year incubation in the present experiment.



Figure 4 RDA triplot for the relationship between the chemical composition of organic matter and Q_{10} (a), and variance partitioning analysis for Q_{10} (b)

The variation partitioning results showed that land use could explain 34.9% of the variance, while the carbon availability (SOM fractions) only accounted for a smaller part (15.4%) of the variance for Q_{10} values (Fig. 4b). Nevertheless, the present findings indicated that the quality of the organic matter, i.e. its chemical composition, also played a very important role in shaping the temperature sensitivity of organic matter degradation, as shown by the variance partitioning results (Fig. 2b), where land use explained a larger proportion of the total variance than the "C availability" factor representing the availability and stabilisation mechanisms of organic C. Thus, the significant differences in the organic matter composition (SOM quality) of the fractions were partly responsible for the results obtained. Furthermore, it should be noted here that these two factors, quality and availability, are difficult to separate in an experiment of this nature, since, the intrinsic properties of organic molecules also largely determine their availability, for instance, the hydrophobic domains of the organic matter could be protected from further degradation (Spaccini et al., 2002).

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