



Original article

Carbon mineralization kinetics and soil biological characteristics as influenced by manure addition in soil incubated at a range of temperatures

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ABSTRACT

This study was conducted to investigate the effects of incubation temperature on mineralization of native pools of C from a soil with a history of manure application, compared to a non-manured soil. Net C mineralization, microbial community structure, biomass size, and metabolic quotient (qCO_2) were measured. Mineralization at cooler temperatures followed zero-order kinetics, indicating a non-limiting supply of substrate. First-order kinetics dominated at warmer temperatures as substrate supply increasingly limited microbial respiration. The soil with a history of manure application had a larger microbial biomass than the non-manured soil, and higher rates of C mineralization. There was a trend toward decreased biomass sizes with increasing incubation temperature. Bacterial DNA T-RFLP profiles were affected by incubation temperature and time with a significant difference in community structure detected after soils had been incubated for 120 days, as well as after incubation at 35 °C. Fungal DNA T-RFLP profiles indicated a distinct community in soils incubated at 35 °C, regardless of the length of the incubation. The key findings from the study were that C mineralization from native pools of organic matter does not follow Arrhenius kinetics at high temperatures, and that incubation of soils outside of their normal temperature range can alter soil biological characteristics which may impact estimates of mineralization parameters.

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1. Introduction

Arrhenius kinetic theory is commonly used to describe temperature effects on decomposition in soils. This theory is derived from fundamental chemistry and applies in situations where substrate is non-limiting and the size of the pool of substrate is not affected by temperature. However, many scientists have acknowledged that the observed temperature effects on soil C mineralization in incubation studies cannot be explained purely by Arrhenius kinetics [11,23]. In most cases in soils, substrate limitation inhibits microbial activity [11,37]; therefore, relying on an Arrhenius approach when modeling temperature effects on decomposition might result in unreliable predictions. Furthermore, the assumption that the size of the pool of

potentially mineralizable substrate in the soil remains constant at all temperatures has been challenged in a range of studies [9,10,14,27].

In spite of the recognized limitations of first-order, Arrhenius approaches to modeling soil C and N dynamics, a more appropriate alternative method has not yet been agreed upon. The incorporation of more biology into decomposition models has been suggested to improve the accuracy of predictions [31,35]. There are a number of ways in which soil biological processes could be affected by temperature, and thereby result in observed temperature effects on net C and N mineralization. For example, microbial biomass size, which can affect the capacity of the community to metabolize substrate, may be affected by temperature. Fang et al. [15] have argued that “the association between respiration rate and microbial biomass C (C_{mic}) during incubation suggests that the variation in microbial biomass may be a major cause of temporal changes in soil respiration”.

In Arrhenius approaches to modeling decomposition, the differences in soil biological characteristics which may impact on the process, including microbial biomass size and structure, are assumed to be incorporated within the first-order (k) or zero-order (z) rate constants: a larger capacity to metabolize substrate will be

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reflected in larger values for the rate constant. It is also assumed that the temperature response of the rate constant will be the same, regardless of the size or structure of the microbial community. But different management practices, including a history of manure use, or the incorporation of a long-term ley crop in the rotation, could result in microbial communities with significantly different sizes and structures, on the same soil type within the same climatic zone [16], and it has not yet been proven that temperature responses of the first-order or zero-order rate constants are the same where microbial community characteristics differ. This uncertainty has implications for modeling of decomposition, as it will determine whether it is important to explicitly include measured soil biological characteristics, as input parameters in decomposition models.

As well as changes in microbial biomass size, changes in microbial community composition could result in changes in the capability to metabolize specific substrates. Within the soil microbial community there is a huge diversity of taxa. Gans et al. [18] estimated that a typical non-contaminated soil contained 8.3×10^6 species of bacteria. Within this diverse microbial community there are a range of functional groups adapted to consume specific substrates. These communities may have differing temperature optima and hence the dominance of a particular functional group of decomposers may change as temperatures alter. Larkin et al. [24] reported differences in substrate specificities, estimated using the community level substrate utilization method, and differences in microbial community structure indicated by changes in fatty acid methyl ester profiles, for soils incubated at 18 °C compared with 25 °C. Andrews et al. [2] reported the use of lower quality substrates in soils incubated at higher temperatures. These changes in substrate specificities with temperature could lead to temperature-driven changes in the size of the mineralizable pool of substrate predicted by the first-order model.

Temperature-driven differences in microbial community assimilation (e_{ass} ; assimilated C per unit C consumed) and production (e_{prod} ; biomass C per unit assimilated C) efficiencies could alter the proportion of C respired for every unit of C metabolized. Andren et al. [1] established assimilation and production efficiencies for a number of functional groups in soils, that are commonly used in soil food web models. For example, in de Ruiter et al.'s [12] food web model of N mineralization, both bacteria and fungi are assigned production efficiency (e_{prod}) values of 0.30, which means that 30% of carbon consumed is apportioned to biomass production while the remaining 70% is respired. In the same model protozoa have e_{prod} values of 0.40, indicating that communities with high populations of protozoa partition more C for biomass and respire a smaller proportion of utilized substrate as CO_2 .

Overall efficiency of substrate use (the combined effects of production and assimilation efficiencies) is also affected by the quality of substrate consumed [20]. Readily available substrates like glucose can have substrate use efficiency as high as 70%, while more recalcitrant substrates may have an efficiency of use of only 15–20%. So if temperature alters the type of substrate metabolized, it may also alter the proportion that is released as CO_2 .

We can postulate about the effects of temperature-driven changes in substrate use efficiency of the soil microbial population; however, it is difficult to measure the effects on each functional group separately in a natural soil system. In soil incubation studies respiration of the whole soil microbial community is generally measured. Efficiency is commonly reported as metabolic efficiency or the metabolic quotient ($q\text{CO}_2$), which is calculated as the rate of basal respiration per unit biomass, and expressed as $\text{mg CO}_2\text{--C mg}^{-1} \text{C}_{\text{mic}}$ per unit time [16]. This measure is not directly related to e_{ass} or e_{prod} ; however $q\text{CO}_2$ has been related to the complexity of the microbial food web with lower values indicating

higher efficiency and food web complexity (Fliessbach et al., 2007). Higher levels of $q\text{CO}_2$ have been linked to nutritional stresses (i.e. substrate limitations), increases in the bacterial to fungal ratio [22], and communities in the initial stages of development with a high ratio of active to dormant biomass [5].

In this study we selected soil from an experiment that included two different manure management treatments: zero manure additions (M0) and 300 kg manure-N $\text{ha}^{-1} \text{y}^{-1}$ (M300). We expected the soil from the treatment receiving annual applications of semi-solid cattle manure to have a relatively large, labile pool of soil organic matter (SOM), while in contrast we expected there to be less labile SOM in the soils that had received no amendments for the previous ten years. We also expected the soil microbial communities to differ between the two manure treatments, and for these substrate quality and microbial community differences to impact the kinetics of C mineralization (i.e. shape of the mineralization curves and response to increasing temperature).

The objectives of this research were to: (1) determine the effects of temperature on the potentially mineralizable pool of substrate (C_0) and the first-order (k) or zero-order (z) rate constants of C mineralization, (2) investigate temperature effects on soil biological characteristics (microbial biomass size, metabolic quotient and community structure), and (3) determine if temperature effects on decomposition parameters and soil biological processes were the same in a soil with two contrasting manure management histories.

2. Materials and methods

2.1. Site and soil

The study site was a long-term forage fertility trial located at the Agriculture and Agri-Food Canada Research Station at Nappan, Nova Scotia, Canada (45° 45' N, –64° 14') on a Gleyed Eluviated Sombric Brunisol (Typic Eutrochrept in US classification). The soil is an imperfectly drained, coarse loamy till, with <5% gravel by volume [43]. The site was seeded to timothy (*Phleum pratense* L., cv Champ), a forage species common to the area, in the spring of 1994. The control and manure treatment plots were first established in the spring of 1995, and continued annually until 2004, as part of a field experiment with a randomized complete block design (four replicate blocks). Forage was harvested for silage up to three times per year when growing conditions allowed. The manure treatment (M300) received an annual early fall application of semi-solid beef cattle manure at a rate of 300 kg total N ha^{-1} . The control treatment (M0) received no amendments throughout the experiment.

In July 2004, a sample (20 × 5 cm diam. cores; ~2 kg fresh weight) was collected from the top 15 cm of each treatment in replicates 1, 2 and 3 of the field experiment. The sample from each plot was passed through a 4 mm sieve and divided into four equal portions which were pre-conditioned at the actual field water content for each plot (see Table 1) for four weeks at each of the following temperatures: 5, 15, 25 and 35 °C. A subsample (~200 g fresh weight) was air dried at 22 °C, passed through a 2 mm sieve,

Table 1

Selected properties of soil collected under a grass/legume forage crop from a long-term fertility trial with treatments of without (M0) or with (M300) a history of annual applications of 300 kg total N ha^{-1} of semi-solid beef manure.

Fertility management	pH	Organic C ^a (mg g ⁻¹)	Total N ^a (mg g ⁻¹)	C:N	Field water content (g g ⁻¹)
M0	6.1	24 ± 2	1.5 ± 0.2	16	0.24–0.27
M300	6.3	30 ± 4	2.0 ± 0.3	15	0.26–0.31

^a Determined by dry combustion, means ± SD reported; n = 3.

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