

# DIVISION S-3—SOIL BIOLOGY & BIOCHEMISTRY

## Soil Carbon and Nitrogen Mineralization: Influence of Drying Temperature

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### ABSTRACT

Carbon and N mineralization in dried soils that are rewetted has been proposed as a rapid index of C and N mineralization potential and to reflect soil management, but further research is needed on effects of soil type and drying temperature for this approach. The objective of this study was to determine the effect of maintaining soil field moisture or drying soil at 40, 60, or 100°C followed by rewetting and a 3-d incubation on C and N mineralization across diverse soil types. Strong correlations between C mineralized in 24 d from field moist soils vs. C mineralized in 24 h from soils dried at 40 or 60°C were observed. Carbon mineralization values for 24 vs. 3 d resulted in nearly linear relationships for all drying treatments. Nitrogen mineralization in 24 d from moist vs. dried at 40 or 60°C and rewetted soils were also highly correlated with field moist N mineralization. The drying and rewetting pre-incubation of soil followed by a 3-d incubation was shown to be a useful indicator of longer-term (24 d) C mineralization potential. Nitrogen mineralization potential may also be obtained after drying/rewetting at 40 or 60°C without the need for keeping soil in a continuously field-moist state.

CARBON AND N MINERALIZATION can be a useful tool for quantifying the impact of various organic and inorganic amendments on soil functions. Carbon mineralization is generally determined by monitoring CO<sub>2</sub> fluxes from field-moist samples that are wetted to roughly 50% of field capacity and subsequently incubated in the laboratory for various periods of time. The incubation period is generally several weeks long, depending on the objective of the study. Short incubations and air-drying soil facilitate routine soil testing procedures and for certain tests air-drying avoids biochemical artifacts that could occur if soils are kept moist before analysis.

Adopting a technique that uses dried soil may significantly reduce variability within the same soil sample and reduce the amount of refrigerated space necessary for storage of moist soils. Soil samples from the same site can vary greatly in moisture content, depending on season or short-term weather patterns. Drying soil holds potential to minimize this variability. Drying and rewetting soil may also permit researchers to determine C and N mineralization potentials on dry, archived soil

samples. When a soil analysis requires field-moist soil, a pre-incubation time of 7 to 10 d after rewetting dried soil may be used to equilibrate the samples before analysis (Franzluebbers et al., 1996).

Rewetting dried soil is thought to alter the soil physiochemical environment and make it an unrealistic treatment (Martens, 1995). On the other hand, laboratory drying and rewetting tends to produce a uniform release of C and N and is a natural process that occurs under field conditions (Birch, 1958, 1959, 1960). Furthermore, short-term C mineralization (1–3 d) of soil after drying (40 or 60°C) followed by rewetting correlates strongly with longer-term (100-d) CO<sub>2</sub> evolution and soil microbial biomass C (Franzluebbers et al., 2000; Haney et al., 1999).

Marumoto et al. (1982) and Sparling et al. (1995) have shown that it may be possible to estimate soil C and N mineralization potential by monitoring the fluxes of CO<sub>2</sub> following the rewetting of dried soil. Other authors have stated that the amount and quality of substrates available for mineralization may be quantified using CO<sub>2</sub> evolution (Sorensen, 1974; Sparling and Ross, 1988). Anderson and Domsch (1978) suggested that the size of the soil microbial biomass is reflected by the short-term flush of CO<sub>2</sub> after amending labile substrates. This is the basis for substrate induced respiration (SIR) method for determination of soil microbial biomass. If the evolution of CO<sub>2</sub> following rewetting of dried soils can be related to soil microbial biomass and potential mineralizable C and N for different soils under different environments then this method might serve as a rapid indicator of potential C and N mineralization.

Chemical and physical disturbances of soil organic matter have been proposed as mechanisms for increasing the flush of CO<sub>2</sub> associated with soil drying and rewetting (van Gestel et al., 1991). For example, Franzluebbers and Arshad (1999) ground dry soils to a powder then rewetted the soils and trapped evolved CO<sub>2</sub>. This treatment resulted in a greater flush of CO<sub>2</sub> than from undisturbed soil. However, C mineralized in 3 d from the disturbed soils was strongly correlated with 24-d C mineralization in undisturbed samples. Similar results were observed when comparing N mineralization as affected by drying temperature.

Currently, most soil incubations use field-moist soil. The flush of C and N after drying and rewetting could possibly become a rapid assessment tool for monitoring changes in C and N mineralization potential due to organic or inorganic inputs as well as inputs from different management strategies. The objective of this study was to compare soil C and N mineralization after drying and rewetting with those maintained field-moist to ex-

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**Table 1. Soil location, classification, and land management.**

Location	Soil Classification	Land Management	pH	Clay %	Organic C g C kg <sup>-1</sup> soil
Weslaco, TX	Hidalgo sandy clay loam (fine-loamy, mixed, active, hyperthermic Typic Calcistoll)	Irrigated maize ( <i>Zea mays</i> L.) under no-tillage	8.0	28	9.3
Corpus Christi, TX	Victoria clay (Fine, smectitic, hyperthermic Udic Haplustert)	Sorghum [ <i>Sorghum bicolor</i> (L.) Moench], conventional tillage with 60 kg N ha <sup>-1</sup>	6.5	41	11.5
Overton, TX	Bowie fine sandy loam (Fine-loamy, siliceous, semiactive, thermic Plinthic Paleudult)	Bermuda grass hay [ <i>Cynodon dactylon</i> (L.) Pers.] receiving 100 kg N ha <sup>-1</sup> as poultry litter	5.9	6	6.5
Stephenville, TX	Windthorst fine sandy loam (Fine, mixed, active, thermic Udic Paleustalf)	Bermuda grass hay receiving 400 kg N ha <sup>-1</sup> as dairy manure	6.3	13	13.0
Lubbock, TX	Acuff loam (Fine-loamy, mixed, superactive, thermic Aridic Paleustoll)	Sorghum receiving 200 kg N ha <sup>-1</sup>	7.4	22	9.3
Clinton, LA	Providence silt loam (Fine-silty, mixed, active, thermic Oxyaquic Fragiudalf)	Alamo switch grass	6.6	18	22.5
Kenai, AK	Kenai silt loam (Medial over loamy, mixed, superactive Typic Haplocryod)	Bluegrass pasture	7.2	19	48.3
Oakwood, OK	Lincoln loamy fine sand (Sandy, mixed, thermic Typic Ustifluvent)	Wheat ( <i>Triticum aestivum</i> L.), continuous tillage with 20 kg N ha <sup>-1</sup>	6.5	10	10.8

plore the possibility of using of dried soil in routine incubations as opposed to field-moist soil.

## MATERIALS AND METHODS

Soil samples were collected from four states. Five of the soils were from Texas (Windthorst, Acuff, Hildalgo, Bowie, Victoria series), and one each was from Alaska (Kenai), Louisiana (Providence), and Oklahoma (Lincoln) (see Table 1 for soil descriptions). Sampling depth in each case was 0 to 7.5 cm. (Table 1). Soil pH was measured with 2:1 water/soil ratio. Soil texture was determined by the hydrometer method and soil organic C from the modified Mebius method (Thomas, 1996).

Each sample was homogenized and passed through a 5-mm sieve, and then split into four treatment groups with each soil sample having 160 g on an oven-dried basis. All soil samples were incubated for 7 d at about 50% of field capacity (range of water addition was 6–14 mL per 40 g of soil). After that, four treatments (three replicates each) were imposed: 24 h drying at 40, 60, 100°C, or continuously moist at 50% field capacity.

Dried soil treatments were subsequently rewetted with water to approximately 50% field capacity. After rewetting, soil samples were incubated at 25°C in 1-L glass jars with an alkali trap containing 10 mL of 1 M KOH to adsorb CO<sub>2</sub> and a container with 10 mL water to maintain humidity. Traps were changed at 1, 2, 3, 4, 5, 7, 14, and 24 d after incubation began and titrated with 1 M HCl (Anderson, 1982).

Nitrogen mineralization was determined by subtracting the initial inorganic N concentration (NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N) of nonincubated soil samples from soil N extracted after 24-d of incubation. Inorganic N was extracted from 7-g soil subsamples using 28 mL of 2 M KCl. Samples were shaken for 30 min on a reciprocal shaker, filtered, and the extracts analyzed for NH<sub>4</sub><sup>+</sup>-N and NO<sub>2</sub><sup>-</sup> plus NO<sub>3</sub><sup>-</sup>-N using an autoanalyzer (Technicon Industrial Systems, 1977a, 1977b). The sum of the above N forms was designated inorganic N.

The experiment was analyzed as a completely randomized factorial design with eight soil types and four incubation treatments. Linear regression was determined to show the strength of relationships. We used SigmaStat ver. 2.03 (SPSS Inc. Chicago, IL.) for all the statistical work.

## RESULTS AND DISCUSSION

Increasing the drying temperature of soils from 40 to 100°C caused a substantial change in the evolution of CO<sub>2</sub> during the first day after soil rewetting. Of the eight soils studied, seven reached their peak rate of CO<sub>2</sub> evolution on or before the second day of incubation (Fig. 1). The flush of CO<sub>2</sub> from drying and rewetting was essentially complete for all soils by the fourth day of incubation.

The quantity of CO<sub>2</sub> produced (C mineralized) in the first 4 d of the incubation was highly correlated to drying temperature ( $r = 0.98$  data not shown). This suggests that short-term CO<sub>2</sub> flux after soil drying and rewetting may provide a stable index for comparative analysis. Results for continuously moist soils vs. soils dried at 40°C (data not shown) were not significantly different from one another at  $P < 0.05$ , as analyzed by ANOVA.

We observed close correlations between 1-d CO<sub>2</sub> evolution following rewetting of soils dried at 40 or 60°C and 24-d cumulative CO<sub>2</sub> release from continuously moist soils, with  $r$  values of 0.99 and 0.98, respectively (Table 2). This CO<sub>2</sub> evolution may partially be due to reestablishment of the microbial population following microbial death due to drying (Sorensen, 1974) and osmotic shock following rewetting (Kieft et al., 1987).

Soils dried at 100°C exhibited both increased C mineralization and greater variability as compared with the 40 or 60°C treatments (Table 2). Birch (1959) used various soils that were air-dried (25°C) or dried at 100°C (with soil drying lasting for 24 to 48 h) and observed a flush of C and N after rewetting. He stated that desiccation of microbial biomass likely occurred during drying. It is likely that the difference in the flush of CO<sub>2</sub> from soils following drying and rewetting originates predominantly from killed soil microbial biomass that is quickly mineralized by the remaining heat-resistant or protected microorganisms. The poor correlation of 1-d CO<sub>2</sub> to

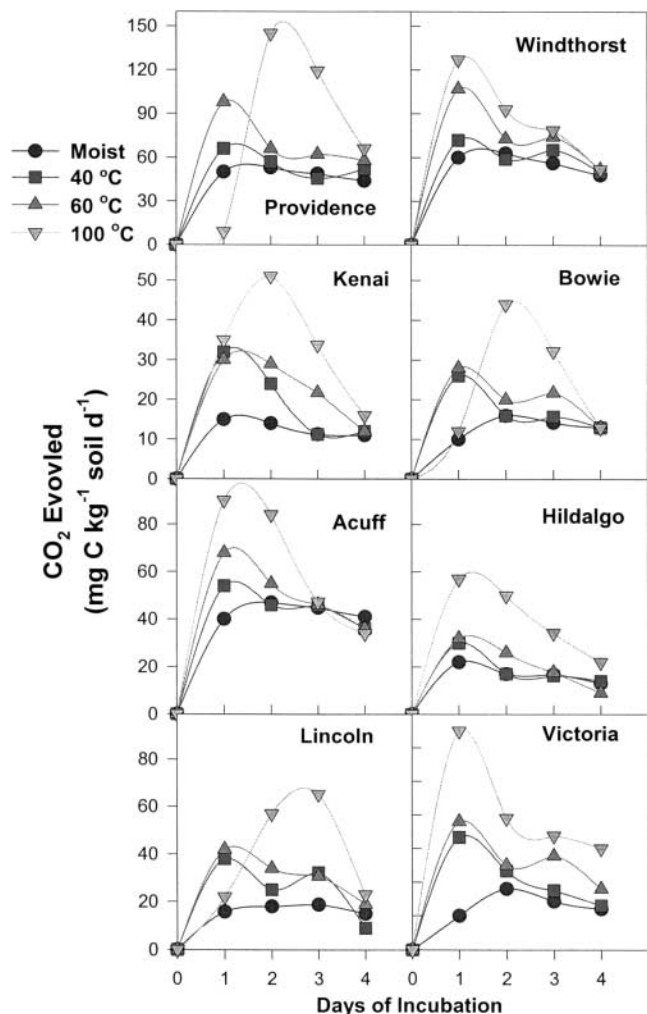


Fig. 1. Rate of CO<sub>2</sub> evolution during the first 4 d of incubation after drying at 40, 60, and 100°C and then rewetting.

24-d CO<sub>2</sub> shown in Table 2 for the 100°C drying treatment was most likely due to a greater portion of the indigenous microbial community affected by 100°C over the 40 or 60°C treatments. By Day 3 of the incubation, however, the evolution of CO<sub>2</sub> from all dried treatments was strongly correlated with field moist 24-d C mineralization, indicating the relatively rapid recovery of the more heat-sensitive microorganisms even when dried at 100°C (Table 2). Therefore, a 24-h recovery period after drying at 100°C and rewetting was insufficient to estimate the complete flush of CO<sub>2</sub>.

Nitrogen mineralized after 24 d from soils that were dried at different temperatures and then rewetted was strongly related to N mineralized after a 24-d incubation of continuously moist soils (Fig. 2). Essentially a 1:1 relationship in N mineralized was observed for soils dried at 40°C and rewetted compared with soils kept continuously moist. As the drying temperature increased, the relationships between N mineralized in dried/rewetted vs. moist soils became more variable, although they were still significantly correlated. Nitrogen immobilization due to microbial uptake or N volatilization may have occurred following the higher temper-

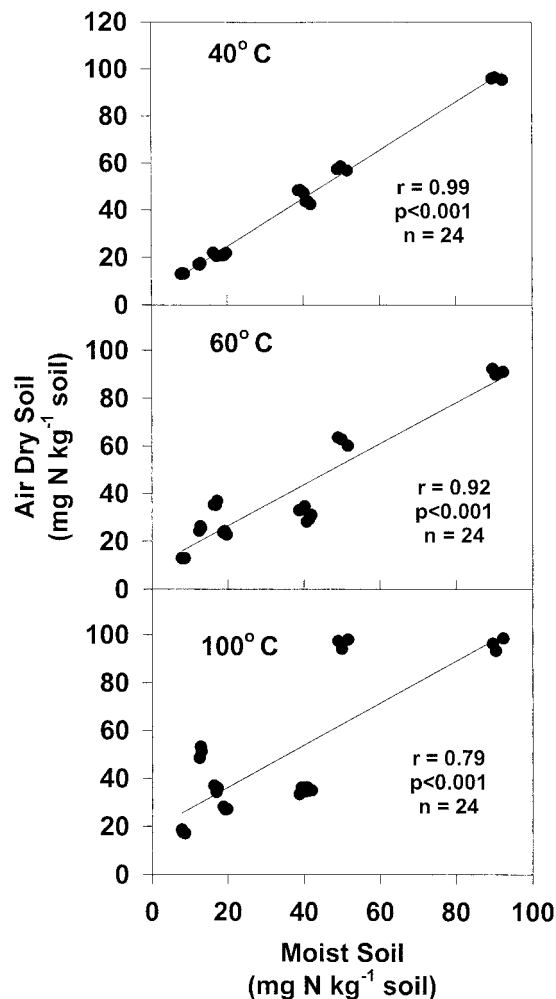


Fig. 2. Nitrogen mineralized after 24 d in soils dried at 40, 60, or 100°C following rewetting compared with 24-d N mineralization in soils that were kept continuously moist.

ature treatments during the 24-d incubation and may possibly explain the increasing variability with increasing drying temperature.

The intercepts of the regression lines increased as soil drying temperature increased, suggesting that the pool of mineralizable N was larger in soils that were dried and rewetted than in field-moist soils (data not shown). One possible explanation may be that protein denaturation begins around 60°C, which suggests that proteins may have been degraded to amino acids and NH<sub>4</sub><sup>+</sup> by

Table 2. Correlation coefficients of potential C mineralization from soils with pretreatment drying and rewetting followed by 3-d incubation with 24-d incubation of field-moist soil (n = 24).

Incubation time	Temperature	Correlation coefficient
d	°C	r
1	40	0.99***
1	60	0.99***
1	100	0.54 <sup>NS†</sup>
3	40	0.98***
3	60	0.99***
3	100	0.98***

\*\*\* Significant at P < 0.001.

† Not significant at P < 0.05.



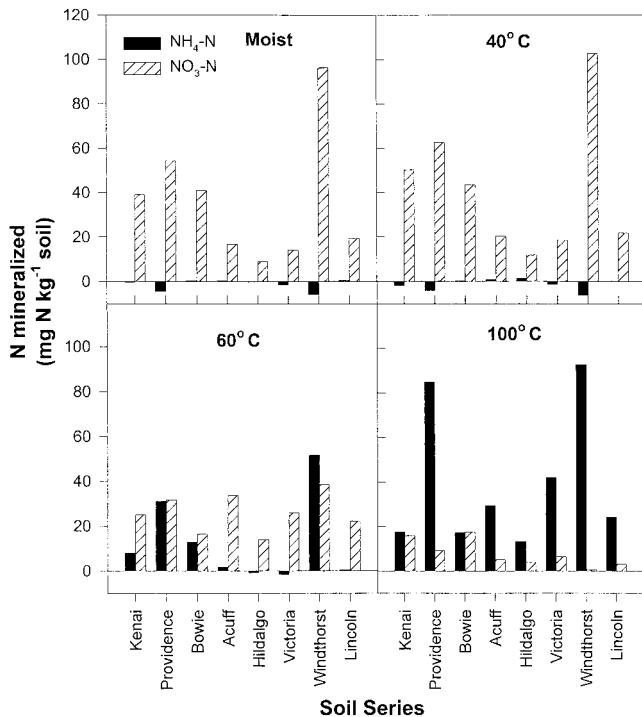


Fig. 3. Nitrogen mineralized ( $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_2^-\text{-N}$ ,  $\text{NO}_3^-\text{-N}$ ) after 24 d in soils kept continuously moist vs. soils that were dried and rewetted.

the drying process. As drying temperature was increased from 40 to 100°C,  $\text{NH}_4^+\text{-N}$  also became more prevalent than  $\text{NO}_3^-\text{-N}$  (Fig. 3). Total inorganic N concentrations from summing both  $\text{NH}_4^+\text{-N}$  and  $\text{NO}_3^-\text{-N}$  were similar in all drying treatments, but  $\text{NH}_4^+\text{-N}$  increased with increasing temperature, indicating that soil-nitrifying bacteria were more sensitive to desiccation at higher drying temperatures, but were not completely eliminated even at 100°C. It is also interesting to note that soils dried at 40°C and rewetted, total N mineralization and individual quantities of  $\text{NH}_4^+$  and  $\text{NO}_3^-$  were not significantly different compared with continuously moist samples. Therefore, as long as standardized laboratory techniques are followed, the use of soil dried at 40°C and rewetted should be useful to evaluate potential N mineralization across a wide range of soils without having to maintain soil in a field-moist state.

## CONCLUSIONS

The flush of  $\text{CO}_2$  after 3 d of incubation from soils that were dried at 40, 60, or even 100°C and rewetted were highly correlated to 24-d incubations with soil that was incubated in a field-moist state. Drying soil at 100°C, rewetting, and then determining  $\text{CO}_2$  evolution for 1 d was not reliable for estimating longer-term C mineralization (24 d). However, even when dried at 100°C, soil microbial respiration rebounded sufficiently within 3 d of rewetting to strongly correlate with longer-term C mineralization. Drying at increasing temperatures then rewetting soils produced a near linear increase in  $\text{CO}_2$  release across a variety of soil types. The flush of  $\text{CO}_2$  after drying and rewetting reaches its peak on the first

or second day of incubation, depending on drying temperature, however, we recommend a 3-d, as opposed to a 1-d incubation following drying and rewetting, to ensure a more complete recovery of the  $\text{CO}_2$  flush.

For potential N mineralization, soil dried at 40°C was highly related to soil kept at field-moist conditions and no significant differences were detected in the amount of  $\text{NH}_4\text{-N}$  or  $\text{NO}_3\text{-N}$  when comparing dried (40°C) vs. field-moist soil.

Drying and rewetting may serve as a useful alternative to maintaining soils in a field-moist state for estimating potential C and N mineralization.

## REFERENCES

- Anderson, J.P.E. 1982. Soil respiration. p. 831–871. *In* A.L. Page et al. (ed.) Methods of soil analysis. Part 2. 2nd ed. Agron Monogr 9, ASA and SSSA, Madison, WI.
- Anderson, J.P.E., and K.H. Domsch. 1978. A physiological method for the quantitative measurement of microbial biomass in soils. *Soil Biol. Biochem.* 10:215–221.
- Birch, H.F. 1958. The effect of soil drying on humus decomposition and nitrogen. *Plant Soil* 10:9–31.
- Birch, H.F. 1959. Further observations on humus decomposition and nitrification. *Plant Soil* 3:262–286.
- Birch, H.F. 1960. Nitrification in soils after different periods of dryness. *Plant Soil* 7:81–96.
- Franzluebbers, A.J., and M.A. Arshad. 1997. Particulate organic carbon content and potential mineralization as affected by tillage and texture. *Soil Sci. Soc. Am. J.* 61:1382–1386.
- Franzluebbers, A.J., R.L. Haney, F.M. Hons, and D.A. Zuberer. 1996. Active fractions of organic matter in soils with different texture. *Soil Biol. Biochem.* 28:1367–1372.
- Franzluebbers, A.J., R.L. Haney, C.W. Honeycutt, H.H. Schomberg, and F.M. Hons. 2000. Flush of carbon dioxide following rewetting of dried soil relates to active organic pools. *Soil Sci. Soc. Am. J.* 64:613–623.
- Haney, R.L., A.J. Franzluebbers, F.M. Hons, and D.A. Zuberer. 1999. Soil C extracted with water or  $\text{K}_2\text{SO}_4$ : pH effect on determination of microbial biomass. *Can. J. Soil Sci.* 79:529–533.
- Kieft, L.T., E. Soroker, and F.K. Firestone. 1987. Microbial biomass response to a rapid increase in water potential when dry soil is wetted. *Soil Biol. Biochem.* 19:119–126.
- Martens, R. 1995. Current methods for measuring microbial biomass C in soil: Potentials and limitations. *Biol. Fertil. Soils* 19:87–99.
- Marumoto, T., J.P.E. Anderson, and K.H. Domsch. 1982. Mineralization of nutrients from soil microbial biomass. *Soil Biol. Biochem.* 14:469–475.
- Sorensen, L.H. 1974. Rate of decomposition of organic matter in soil as influenced by repeated air-drying-rewetting and repeated additions of organic material. *Soil Biol. Biochem.* 6:287–292.
- Sparling, G.P., D.V. Murphy, R.B. Thompson, and I.R.P. Fillery. 1995. Short-term net N mineralization from plant residues and gross and net N mineralization from soil organic matter after rewetting of a seasonally dry soil. *Aust. J. Soil Res.* 33:961–973.
- Sparling, G.P., and D.J. Ross. 1988. Microbial contributions to the increased nitrogen mineralization after air-drying of soils. *Plant Soil* 105:163–167.
- Technicon Industrial Systems. 1977a. Determination of nitrogen in BS digests. Technicon Industrial Method 487–74W/B. Technicon Industrial Systems, Tarrytown, NY.
- Technicon Industrial Systems. 1977b. Nitrate and nitrite in soil extracts. Technicon Industrial Method 487–77A. Technicon Industrial Systems, Tarrytown, NY.
- Thomas, G.W. 1996. Soil pH and soil acidity. P.475–490. *In* D.L. Sparks (ed.) Methods of soil analysis. Part 3. SSSA Book Ser. 5. SSSA, Madison, WI.
- van Gestel, M., J.N. Ladd, and M. Amato. 1991. Carbon and nitrogen mineralization from two soils of contrasting texture and microaggregate stability: Influence of sequential fumigation, drying and storage. *Soil Biol. Biochem.* 23:313–322.