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Relationship between Water Content and Mineralization of Carbon and Nitrogen in Soils Varying in Physical and Chemical Characteristics

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ABSTRACT

An understanding on relationship between water content and mineralization of carbon (C) and nitrogen (N) across soils varying in physical and chemical characteristics is required to assess the influence of soil physico-chemical properties on soil organic matter decomposition. However, such information is rarely available. Relationship between C and N mineralization of three soils varying in physico-chemical properties with different measurements of water content (water-filled pore space, gravimetric water content, volumetric water content, and water holding capacity) was studied through an incubation experiment for 8 weeks. Results of the experiment showed that C and N mineralization increased with increasing water content, reached a maximum, and then decreased with subsequent increasing water content levels. Maximum C and N mineralizations were observed at 70-80% and 50% water-filled pore space (WFPS), respectively. The ranges of WFPS for C and N mineralization were the narrowest among other measurements of water content. Therefore, it was likely that a single WFPS could be used in subsequent incubations to examine either C or N mineralization of soils with different characteristics. Result of this study suggests that the preliminary experiment on the relationship between mineralization of C and N and water content is necessary to do where mineralization is needed to be assessed in soils that have different physico-chemical characteristics.

Keywords: Carbon and nitrogen mineralization, percent of water-filled pore space, water content

INTRODUCTION

Mineralization of carbon (C) and nitrogen (N) are generally used to evaluate the effect of organic matter on changes in soil characteristics (Hooker and Stark 2008; Cayuela *et al*. 2009). Mineralization of C and N are also used to indicate the decomposition rate of organic waste in soils (Parnaudeau *et al*. 2006; Rivera-Espinoza and Dendooven 2004) and effect of land-use changes on soil quality (Kirschbaum *et al*. 2008; Mahaney *et al*. 2008). Mineralization of C and N are generally determined through incubation experiments in the laboratory at constant temperature and water content. Water content used in such incubation experiments is generally cited from the literature without concerning the soils used in the present experiments may have different characteristics to those of previous experiments.

It has been well established that water content is one of the environmental factors controlling the decomposition rates of soil organic matter. Relationships between carbon and nitrogen mineralization and water content have been the subject of several studies (Tietema *et al*. 1992; Sleutel *et al*. 2008). Howard and Howard (1993); Gulledge and Schimel (1998) examined the effect of water content on C mineralization of soils varying in physical and chemical properties. They reported that the response of C mineralization to water content expressed on a gravimetric basis varied widely among soils, depending mainly upon soil texture and organic matter. A possible consequence of this result is that the gravimetric water content is generally not useful for a direct comparison of the effect of water content on C mineralization across soils varying in texture and organic matter contents.

Most studies examining the response of C mineralization to soils had assumed that the relationship between water content and C mineralization would be similar among physically and chemically different soils. Many studies had been completed by incubating soils at a specific water content and determining C mineralization at that water content (Cayuela *et al*. 2009). Under such incubation conditions, it was assumed that variations in C mineralization must result from variations in soil characteristics. It was also assumed that relationship between C mineralization and water content would be similar to relation between N mineralization and water contents for a similar soil; thus, many studies determined C and N mineralization in a similar soil through an incubation experiment using a single water content. However, the relationship between soil water contents and C mineralization for different soils and the relationship between mineralization of C and N and water content for similar soil are unclear.

The measurements of water content for incubation experiments are volumetric water content, water holding capacity, water filled-pore space, water potential and gravimetric water content (Linn and Doran 1984; Ilstedt *et al*. 2000; Sleutel *et al*. 2008). However, a little information is available on which measurements of water content can allow a direct comparison of C and N mineralization of soils differing in physical and chemical characteristics. In this study, the influence of variations in water contents on the mineralization of carbon and nitrogen from three soils with different physico-chemical characteristics was evaluated.

The objectives were to quantify the changes in carbon and nitrogen mineralization with increasing water contents, to assess whether the relationship between C mineralization and water contents varied in different soils, and to determine whether relationship between C mineralization-water content and N mineralization-water content would be similar for the same soil.

MATERIALS AND METHODS

Soil Sampling and Analysis

Soil samples were collected from sites that exhibiting different land-uses (secondary forest, maize farm, and grasslands) in the Kabupaten Tanah Laut, South Kalimantan Province, Indonesia. A composite sample was collected from the 0-10 cm layer at each site using a cylindrical core (10 cm diameter). Living plant material was selectively removed from the collected samples by hand, and the samples were thoroughly homogenized and stored in polyethylene containers at 5 °C. Sub-samples were air-dried and ground to pass a 2-mm sieve for physical and chemical analyses.

Bulk density was determined immediately after sampling in each sampling point using the core method described by Blake and Hartge (1986a). Soil samples were analysed for soil texture (Gee and Bauder 1986), particle density (Blake and Hartge 1986b), soil pH (McLean 1982) and cation exchange capacity (Rhoades 1982). Organic C content was determined using Walkley and Black method(Nelson and Sommers 1996), and total nitrogen was determined using Kjehdahl method (Bremer and Mulvaney 1982).

Laboratory Incubation

A portion of each composite soil sample was oven-dried at 40° C to constant mass and gently crushed to pass a 2.00 mm screen. For each incubated sample, an appropriate mass of soil was placed into 130 ml containers (diameter = 5 cm) to give a depth of 20 mm after compacting the sample to the bulk density measured in the field. Distilled water was added drop-wise using a fine jet pipette to obtain 30%, 40%, 50%, 60%, 70%, 80%, and 90% water-filled pore space (WFPS). WFPS was calculated from the equation (Linn and Doran 1984):

$$\text{WFPS} = (\text{GWC x BD}) / (1 – (\text{BD/PD})$$

where GWC is the gravimetric water content (g water g^{-1} soil), BD is the bulk density (Mg m^{-3}), and PD is the particle density (Mg m^{-3}). Each sub-sample was placed into a 1 litre jar along with a vial containing 10 mL 0.5 *M* NaOH to trap evolved CO_2. A jar with a vial containing NaOH solution and a container without soil was used as a blank. The jars were sealed and incubated in the dark at 25 ± 1° C for 8 weeks. For each combination of soil and WFPS, three replicated samples were prepared and incubated. During the incubation period, distilled water was added periodically to compensate for evaporative losses and ensure constant WFPS. The alkali traps were replaced every week and total CO_2-C was determined soon after sampling using the method described by Schinner *et al.* (1996).

Inorganic nitrogen (NH_4^+ and NO_3^-) was determined after 8 weeks. At the end of incubation, containers were removed from the incubation, and sub-samples (5 g) were extracted with 2 *M* KCl at an extractant: sample ratio of 10 : 1 (v/w). The extracts were filtered through a Whatman 42 ash-free filter paper. Concentrations of NH_4^+ and NO_3^- in the extracts were measured colorimetrically using a hydrazine reduction method for NO_3-N and an indophenol blue method for NH_4-N (Bundy and Meisinger 1994). At the end of incubation, gravimetric and volumetric water contents and water holding capacity were determined for each sample using the methods as described by Ilstedt *et al*. (2000).

Statistical Analysis

Statistical analysis of experimental data was accomplished by analysis of variance (ANOVA) using a completely randomised design for cumulative C mineralization and N mineralization using GenStat 12th Edition (Payne 2008). The data were checked for normal distribution with the Shapiro–Wilk test. In the case of significance in ANOVAs, means were compared by the least significant difference (LSD) multiple comparison procedure at $P < 0.05$.

RESULTS AND DISCUSSION

Soil Characteristics

Soil samples used in this experiment varied in soil textures: clay loam (maize farm), sandy loam (grassland), and clay (secondary forest). The three soils were acidic, with pH values ranged from 3.3 to 5.4 (Table 1). Bulk density ranged from 0.96 Mg m^{-3} in grassland to 1.32 Mg m^{-3} in secondary forest.

Table 1 also shows that organic carbon contents of three soils varied considerably from low carbon content (3.9 g C kg^{-1} soil) in grassland to high carbon content (48.8 g C kg^{-1} soil) in secondary forest. Total nitrogen of three soils varied within a relatively small range of 1.3 to 4.3 g N kg^{-1} soil. However, combination of C and N content did not result in significant differences in C/N ratio (3.1 – 11.4). Cation exchange capacity (CEC) of secondary forest soil was 1.9-fold and 2.7-fold higher than of grassland and maize farm soils, respectively. Based on the differences in soil characteristics, it could be concluded that three soils used in this experiment varied in physical and chemical characteristics.

Changes in Water Content and Carbon Mineralization

Analysis of variance showed that C mineralization, expressed as mg CO_2-C kg^{-1} soil was significantly influenced by changes in water contents expressed in WFPS. Carbon mineralization of three soils were influenced by changes in the WFPS. Carbon mineralization ranged from 643 mg CO_2-C kg^{-1} soil to 2,706 mg CO_2-C kg^{-1} soil, depending on soils and WFPS levels. The highest C mineralization was observed at 70-80% WFPS for secondary-forest and grassland soils and 80% WFPS for maize-farm soils (Figure 1). Carbon mineralization of three soils with water contents expressed in water-filled pore space, gravimetric water content, volumetric water content and water-holding capacity is described in Figure 1. Carbon mineralization increased with increasing water content, reached a maximum and then decreased with subsequent increasing water content. Other experiments also demonstrated that C mineralization exhibited a parabolic response with the increasing water contents (Ilstedt *et al*. 2000; Ruser *et al*. 2006; Beare *et al*. 2009). Low carbon mineralization at low water contents was likely due to insufficient water for microbial decomposition and limited diffusion of soluble substrate supply (Taggard *et al*. 2012). Increasing in C mineralization at initial water contents were due to increasing water availability for microorganisms in soil pores, reached a maximum, and then decreased because of reduction in oxygen availability in soil pores for microorganisms with subsequent increasing in water contents (Skopp *et al*. 1990; Wen *at al*. 2006).

Maximum C mineralization of three soils was observed at different ranges of water content under different measurements. Maximum C mineralization for all soils were observed at 70–80% WFPS, 22-43% of gravimetric water content, 29-44% of volumetric water content, and 51-70% of water holding capacity (Figure 1). For WFPS, the optimum C mineralization in this study was occurred at a higher WFPS than the proposed optimum reported

Table 1. Selected soil physical and chemical characteristics.

Characteristics	Unit	Secondary forest	Grassland	Maize farm
Texture				
Sand	%	16.62	63.32	44.41
Silt	%	26.44	24.43	25.12
Clay	%	56.94	12.25	30.47
Bulk density	Mg m^{-3}	1.32	0.96	1.03
Particle density	Mg m^{-3}	2.33	2.28	2.29
pH (H_2O)	-	4.24	3.30	5.44
Organic C	g C kg^{-1}	48.82	3.93	10.60
Nitrogen	g N kg^{-1}	4.27	1.25	1.67
C/N ratio	-	11.43	3.14	6.35
CEC	cmol (+) kg^{-1}	52.17	27.69	19.26

in other studies. Beare *et al*. (2009) observed the optimum WFPS for C mineralization of uncompacted (bulk density of 1.01 Mg m^{-3}) and compacted (bulk density of 1.49 Mg m^{-3}) soils were occurred at 45% and 55%, respectively. In addition, Linn and Doran (1984) found 60% WFPS for maximum microbial activity in an experiment using soils with bulk density of 1.14 and 1.40 Mg m^{-3}. A relatively wide range of water content for C mineralization could lead a serious error in the interpretation of C mineralization data from an experiment that uses only single water content. For example, if 51% water holding capacity was used for the incubation experiment C mineralization of grassland soil would be maximized but C mineralization of secondary forest soil would only reach 63% from its maximum potential (Figure 1). Variation in C mineralization of three soils incubated at 51% water holding capacity was not only resulted from variation in physical and chemical properties of soils, but it was also resulted from differences in relation between water contents and C mineralization among three soils. Results of this experiment suggested that water-holding capacity was not good to be used for a direct comparison of soil C mineralization with different characteristics.

Based on ranges of water content with achieved maximum C mineralization, water filled pore space exhibited the narrowest range (70-80% WFPS) compared to other measurement water contents. Variation in C mineralization resulted from variation in WFPS was relatively small, 2-15% lower than the maximum C mineralization at 70% WFPS or 80% WFPS (Figure 1). This implied that WFPS resulted in more accurate C mineralization compared to other water content measurements if only single water content was used in an incubation experiment of soils differing in physical and chemical characteristics. This result is in agreement with

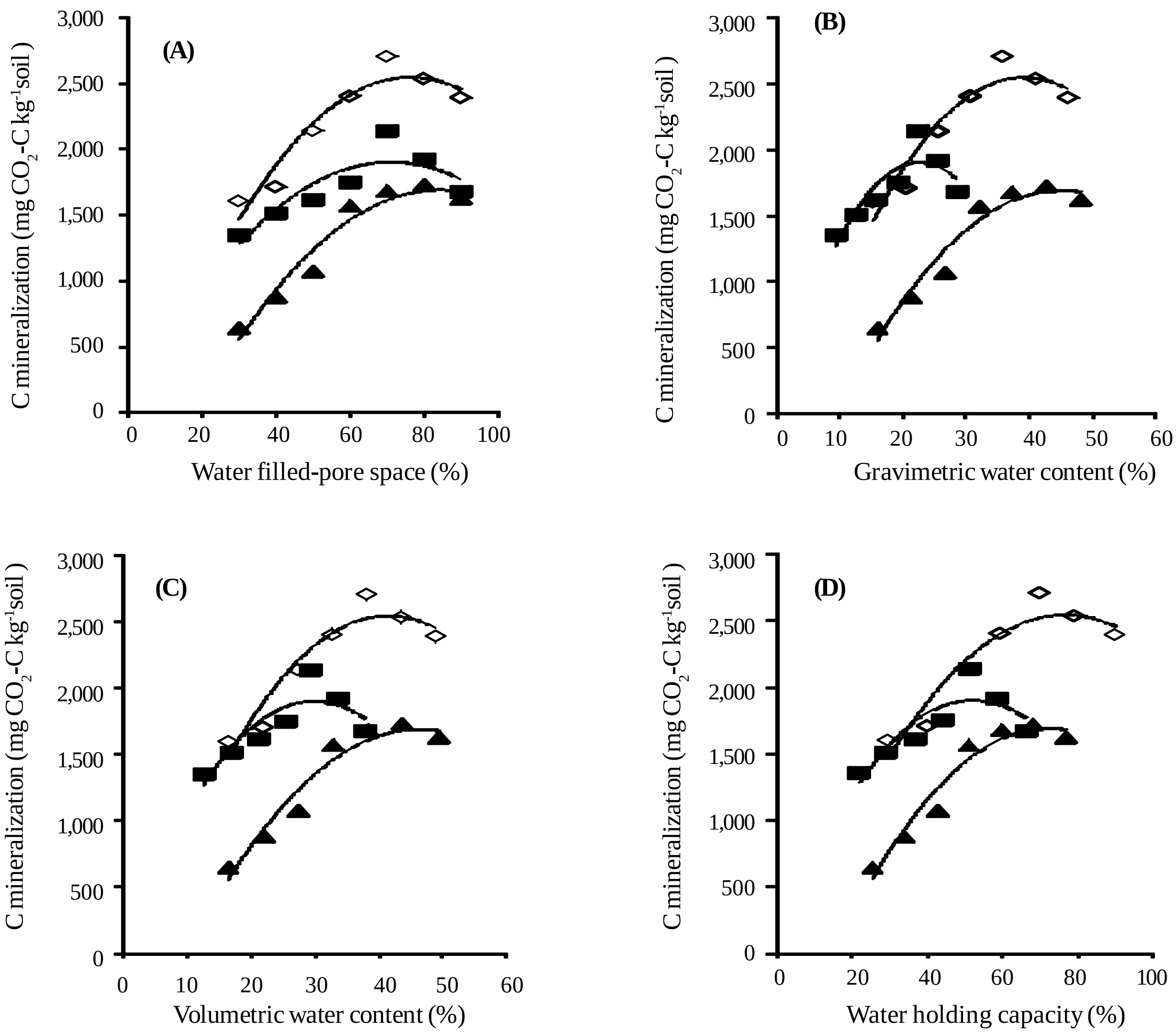

Figure 1. Relationship between C mineralization and changes in water content. Water filled-pore space (A), gravimetric water content (B), volumetric water content (C), and water holding capacity (D). ◇ = secondary forest, ▲ = maize farm, and ■ = grass land.

Ilstedt *et al*. (2000) who found that WFPS was better than volumetric and gravimetric water contents for measurement of C mineralization in an incubation experiment of forest soils with average bulk density of 0.64 Mg m^{-3}. Results of this study emphasized the importance of preliminary experiment to describe relationship between water content and C mineralization for incubation studies using soils with different characteristics.

Changes in Water Content and Nitrogen Mineralization

Analysis of variance showed that N mineralization was significantly affected by changes in water content. Nitrogen mineralization of three soils were varied in a range of 53-157 mg N kg^{-1} soil, depending on soils and water contents (Figure 2). The effect of changes in water contents on net N mineralization was similar to that on C mineralization. N mineralization increased with increasing water content, reached a maximum at a specific water content and then decreased with subsequent increase in water content. However, the pattern of C mineralization in response to changes in water contents was quite different to N mineralization. Turning point, point of either C or N mineralization began to decrease, of C mineralization was occurred at relatively high water contents (Figure 1) while N mineralization at low water contents (Figure 2). This suggested that the water requirement for microorganisms to result in optimum C mineralization was higher than that for N mineralization.

Different measurements of water content resulted variation in ranges at which N mineralization was maximized (Figure 2). WFPS had the narrowest range of water content for maximum N mineralization compared to others (Figure 2). Mineralization of N of three soils reached a maximum at 50% WFPS, 15-27% gravimetric water content, 17-29% volumetric water content, and 36-50% water holding capacity. This result suggested that the use of a single WFPS level (50% WFPS) for incubation experiment would result in better N mineralization compared to other water content measurements. The fact that WFPS gave a better N mineralization compared to other water content measurements were supported by the study of Sleutel *et al*. (2008), who found that the relationship between soil moisture and N mineralization was described very well when using WFPS to measure soil water status.

Nitrogen mineralization of three soils in response to changes in WFPS are described in Figure 2. It can be seen that N mineralization showed a parabolic response to the increasing of WFPS in which the maximum N mineralization in all samples were occurred at 50% WFPS. Above 50% WFPS, the N mineralization decreased with increasing WFPS levels. This value was slightly lower than other published estimations of WFPS for optimum N mineralization (60% WFPS, Linn and Doran 1984; 56% WFPS, De Neve and Hofman 2002; 57-78% WFPS, Sleutel *et al*. 2008).

Decrease in N mineralization at WPFS > 50% could have been related to an increase in N requirement for microorganisms. At 50-70% WFPS, C mineralization increased from 1065 to 2705 mg CO_2-C kg^{-1} soil, depending on the soil used in this study (Table 2). As C mineralization increased with increasing WFPS levels, the requirement of nitrogen by microorganisms would also increase, therefore a larger fraction of nitrogen released to the soils would be immobilised. This finding is in agreement with Franzluebbers (1999), who found that at WFPS in which N mineralization began to decrease, C mineralization continued to increase to a maximum with further increase in WFPS. Loss of mineral nitrogen could also have occurred through denitrification, which would give the impression of reducing N mineralization or immobilisation. Losses of nitrogen through denitrification were suggested by decreasing in nitrate concentration in all soils with increasing WFPS levels (data not shown). This finding is consistent with Pandey *et al.* (2009) who reported reduction in NO_3^- concentration of soils differing in land use systems with increasing WFPS. Previous studies by Torbert and Wood (1992) and Aulakh *et al*. (2000) have shown that nitrogen losses through denitrification occurred at 60% WFPS. In addition, nitrous oxide (N_2O) production in mineral and organic soils increased with increasing water contents (Menendez *et al*. 2012; Paul *et al.* 2012; Weerden *et al*. 2012).

Use of Single Water Content for Mineralization of Carbon and Nitrogen

Another important experimental issue for this study is the difference in WFPS level for maximum C and N mineralization. Carbon and nitrogen mineralization of three soils at different level of WFPS are illustrated in Figure 3. It was observed that N mineralization reached a maximum at 50% WFPS while C mineralization at 70-80% WFPS. Differences in WFPS level for optimum C and N mineralization in this study are in agreement with Franzuelebbers (1999), who reported lower WFPS level for optimum N mineralization than C

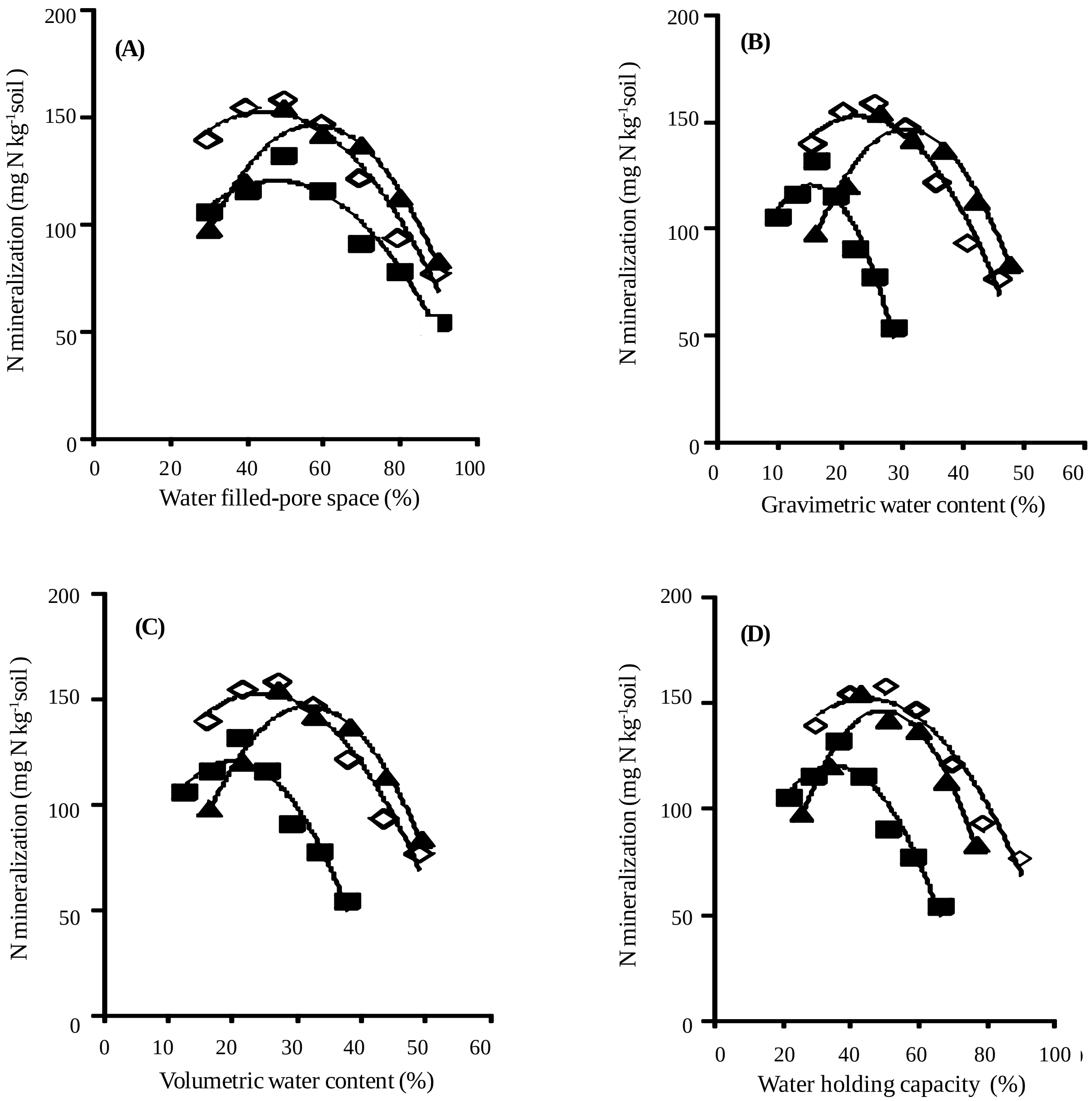

Figure 2. Relationship between N mineralization and changes in water content in different units: water filled-pore space (A), gravimetric water content (B), volumetric water content (C), and water holding capacity (D). ◇ = secondary forest, ▲ = maize farm, and ■ = grass land.

mineralization for 15 soil samples differing in texture and organic matter contents. Franzuelebbers (1999) observed C mineralization was optimum at 53-66 % WFPS while N mineralization at 34-40% WFPS. The difference in WFPS for optimizing C and N mineralization could lead a serious error in the interpretation of soil organic matter mineralization data from an experiment that used only a single water content for the incubation. Carbon and N mineralization of secondary forest soil was a good example to describe the error. If the level of WFPS selected for an incubation experiment was 70%, carbon mineralization of secondary forest soil would be maximised, but net nitrogen mineralization would only reach 81% of its maximum potential (Figure 3). If 50% WFPS was used in the incubation experiment, N mineralization of secondary forest soils would be maximized but C mineralization of the soil only reached 79% of its maximum potential. Therefore, it is important to note that in experiments where carbon and nitrogen mineralization are measured at a single WFPS, the relationship between carbon and nitrogen mineralization rates is specific for that WFPS and may not be applied to either wetter or drier conditions. Work completed in this experiment stresses the need for preliminary experimentation to define the influence of water content on mineralization of carbon and nitrogen to ensure that acquired data are interpreted correctly.

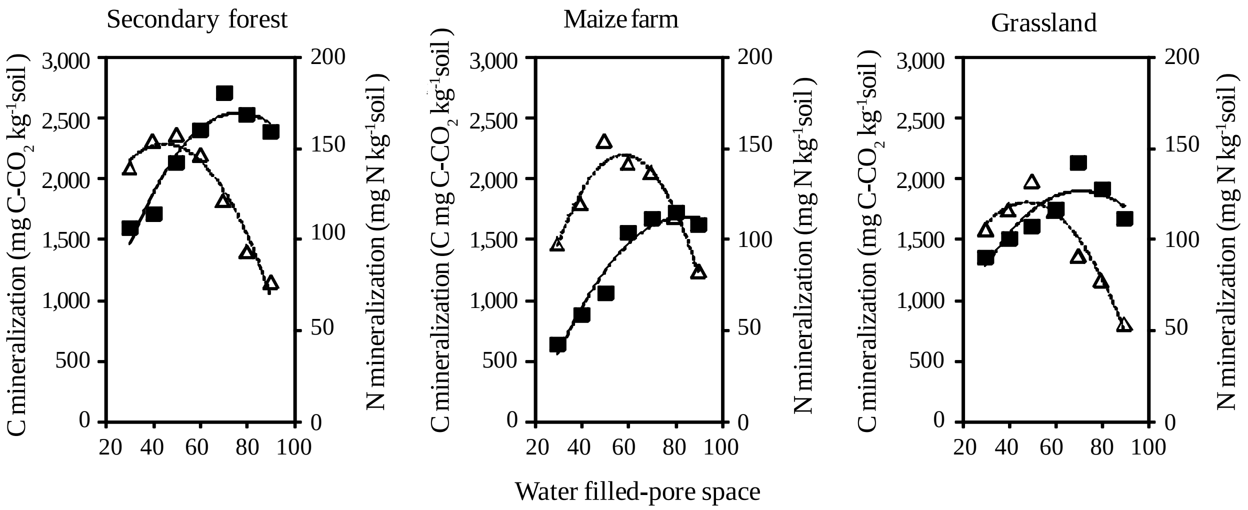

Figure 3. Relationship between C and N mineralization and changes in water content expressed in water filled-pore space. ■ = C mineralization and ∆ = N mineralization.

CONCLUSIONS

This study showed that C and N mineralizations increased with increasing water content, reached a maximum, and then decreased with further increasing water content. The measurement of water content in WFPS resulted in a narrower range of water for maximum C and N mineralization compared to other measurements (gravimetric water content, volumetric water content, and water holding capacity). Carbon and nitrogen mineralization of three soils was maximised at 70-80% WFPS and 50% WFPS, respectively. There is now a clear evidence to suggest that WFPS would result in more accurate C and N mineralization data compared to other water content measurements in an incubation study using single water content and soils varying in physical and chemical characteristics. Data of C and N mineralization also revealed that maximum C and N mineralization were observed at different levels of WFPS, indicating that the use of a single WFPS in an incubation study of a certain soil can not be useful to describe C and N mineralization.

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