Effect of Fertilizer Management on Soil Carbon Dioxide Fluxes in Grassland and Cornfield during Winter

P. A. Nugroho1*, U. Sudadi1, and S. Suwardi1

ABSTRACT

This study was conducted to investigate the effect of mineral (F) and Mineral-organic Fertilizers (MF) on soil CO2 flux during winter in grassland and cornfield in Southern Hokkaido, Japan, from May 2013 to April 2014. CO2 flux was measured by the static chamber method. Soil CO2 concentration was determined using silicone tubes pipe. The environmental variables, i.e. climate and soil, were also analyzed in this study. Results showed that, in freezing period, CO2 flux in MF was lower than F plots in grassland (0.1 and 0.4 Mg C ha\(^{-1}\) period\(^{-1}\), respectively). However, in melting period, CO2 flux in F was lower than MF plots (0.01 and 0.1 Mg C ha\(^{-1}\) period\(^{-1}\), respectively). CO2 flux in F and MF plot was similar in cornfield in freezing (0.5 Mg C ha\(^{-1}\) period\(^{-1}\)) and melting (0.01 Mg C ha\(^{-1}\) period\(^{-1}\)). These results were opposite to the annual CO2 flux where MF was higher than F plot in both grassland (10.9 and 8.5 Mg C ha\(^{-1}\) period\(^{-1}\)), respectively) and cornfield (8.7 and 6.2 Mg C ha\(^{-1}\) period\(^{-1}\)). Soil CO2 concentration during winter was relatively higher in grassland than cornfield. Soil NH4-N and Water Extractable Organic Carbon (WEOC) showed a positive correlation with soil CO2 concentration (\(r^2= 0.39\) and \(r^2= 0.19\), respectively). On the other hand, a negative correlation was observed between soil CO2 concentration and soil NO3-N content (\(r^2=-0.39\)).

Keywords: Carbon dioxide emission, Green-house gasses, Land use, Manure, Plant nutrient.

INTRODUCTION

Livestock is one of the important sectors for food security in Japan. It is highly intensive and still dependent on imported fodder (Hirata et al., 2013). To support this sector, the biomass from grassland and cornfield are harvested as a source of fodder. The grassland area in Japan is wide enough, the second largest after paddy field. It covers around 8,475 km\(^2\) area or about 18% of the total farmland area (Matsuura et al., 2012), hence, it is very important in the food sector.

To stimulate plant growth and improve land productivity, fertilizer and manure are applied in grassland and cornfield. Fertilizer and manure application will affect Net Primary Production (NPP), Soil Organic Matter (SOM), as well as microbial activities, which in turn enhance CO2 and N2O emissions (Shimizu et al., 2009; Gong et al., 2009; Nhu et al., 2012; Shimizu et al., 2013). Soil CO2 flux is often referred to as soil respiration and it is comprised of autotrophic respiration from plant roots and heterotrophic respiration from soil organisms. It may also include respiration from the litter layer on the top of the mineral soil (Kirschbaum et al., 2001). These processes are controlled by soil climate (temperature and water content), Carbon (C) source, nutrients, oxygen availability, and biological factors (Smith et al., 1997). Manure application increases the soil organic carbon and readily-available source of carbon for microbial activity that can increase CO2 emissions (Rochette et al., 2004; Alemu et al., 2015). Previous studies in Iran and USA indicated that incorporation of manure and

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nitrogen fertilizer elevated the CO₂ emission 9 and 30%, respectively, other than solitary N fertilizer application (Lee et al., 2007; Salehi et al., 2017).

As described above, the air and soil temperature play important role in CO₂ flux. In winter, the CO₂ flux has rarely been measured due to the difficulty of conducting measurements in snow-covered soil; furthermore, winter CO₂ emissions through the snow pack were are assumed to be negligible or are underestimated (Raich and Schlesinger, 1992; Raich and Potter, 1995).

Wang et al. (2010) reported that CO₂ emission during winter in a forest-steppe ecotone in north China was 3.48-7.30% of its annual CO₂ emission. Another study by Hubbard et al. (2005) indicated that about 8% of the annual CO₂ emission was emitted from the subalpine forests in winter. In an Austrian mountain forest, CO₂ in winter contributed 12% of the annual CO₂ flux (Schindlbacher et al., 2007), and in winter, a mixed conifer forest in Washington State released around 17% of the annual CO₂ budget (Mc Dowell et al., 2000).

The combination of mineral and organic fertilizer application in grassland and cornfield is widely used in Japan; therefore, it is very important to enhance the information about the CO₂ flux during winter in managed grassland and cornfield. The differences in management of fertilizer probably will affect the CO₂ gas that is emitted in winter.

This study aimed to find out the effect of fertilizer management practice on soil CO₂ flux in grassland and cornfield during winter.

**MATERIALS AND METHODS**

The study was conducted in a grassland in the livestock experimental farm of Hokkaido University and a cornfield in Niikapu station, the National Livestock Breeding Centre. Both sites were located in Southern Hokkaido with geographical position of 42° 26’ N; 142° 29’ E and 42° 24’ N; 142° 28’ E, respectively, with elevation of about 50 m above sea level and a slope of 1-2%.

The climatic zone is categorized as humid continental climate with cold winters and cool summers without apparent wet or dry seasons. The mean annual precipitation and annual temperature (1981–2010) are 1,290 mm and 8.1°C, respectively. Soil was developed from volcanic ash and classified as Andisols (Soil Survey Staff, 2006) or Andosol (FAO, 1998). Grassland soil is categorized as poorly drained; on the other hand, cornfield is classified as well drained soil (Shimizu et al., 2013). Grassland soil had 0.77 g g⁻¹ sand and 0.19 g g⁻¹ silt and 0.037 g g⁻¹ clay. In cornfield, soil fractions contents were dominated by sand i.e. 0.71 g g⁻¹ followed by silt 0.21 g g⁻¹ and clay 0.08 g g⁻¹.

**TREATMENTS**

Two treatments [mineral (F) and Mineral-organic Fertilizers (MF)] were applied consisting of four replications in 10×10 m plots. Twenty one kg ha⁻¹ seed of Timothy grass (Phleum pretense L.) and 4 kg ha⁻¹ white clover (Trioplium repens L.) were sowed in grassland on May 25, 2013. Since the precipitation in May was higher than usual, the seeding of grass was delayed. The grass was harvested on July 19, 2013. The higher precipitation also happened in June and July. This situation enhanced high weed growth in the grassland site. It was decided to renovate the grassland by herbicide application. Four days after the application of herbicide, the whole grass was cut. The residue which remained on the ground was then incorporated with rotary ploughed on September 20, 22 and 24. Eighty one kg ha⁻¹ seed of Timothy grass (Phleum pretense L.) was sown on 27 September along with the application of 40 kg N ha⁻¹ (which was supplied by Urea) to F and MF plots by topdressing. A total of 37 t ha⁻¹ (containing 448 kg N ha⁻¹ and 7.3 t C ha⁻¹) of mixed woody bark and cattle manure was applied on October 17, 2012 to MF plot by broadcasting machine. Due to the renovation of grassland, a 17 t fresh manure ha⁻¹ (194 kg N ha⁻¹ and 3.5 t C ha⁻¹) was reapplied on September 12, 2013.
On April 26, 2013, the cornfield was ploughed and 24.6 kg ha$^{-1}$ of corn (Zea mays L.) seed was sown on May 10, 2013. Four months after (September 20, 2013), corn was harvested. In harvesting, top parts of the plant were removed from the field by harvesting machine and 30 cm of stubble and roots were left in the field.

Later, 103 kg N ha$^{-1}$ in the form of urea was applied by topdressing to F and MF plots on June 2, 2013. A total of 29 t fresh manure ha$^{-1}$ (191 kg N ha$^{-1}$ and 4.3 t C ha$^{-1}$) was broadcasted to MF plots on September 9-17, 2013. The N application rates of mineral fertilizer in grassland and cornfield were at the recommended level on the basis of soil tests done by the livestock farm staff, whereas the manure application rates were the optimum rates used by farmers in the region and were based on adequacy of potassium application to the fields (Jin et al., 2010).

**Environmental Variables**

The environmental variables like precipitation and snow depth was obtained from AMeDAS (Automated Meteorological Data Acquisition System) by Japan Meteorological Agency. Soil temperature was measured continuously at 5 cm depth using thermocouple thermometer (CT220, Custom, Tokyo-Japan). The Soil freezing depth was measured using methylene blue dye method (Richard et al., 1976).

**Soil Sampling and Analysis**

Soil composite sampling was carried out concurrently with gas sampling from 0-10 cm depth with 4 replications. In the beginning of freezing and ice melting, soil was taken from 0-10; 10-20; 20-30; 30-40 and 50-60 cm depths. Samples were then extracted using deionized water (ratio 1: 5) and 2M KCl (1:10), then filtered through 0.2 µm membranes. Soil pH was measured in soil-water extract (by F-52, Horiba, Japan) just before filtering.

The extraction of soil and deionized water was then used to measure NO$_3$-N content by ion chromatograph (Dionex, Sunnyvale, CA, USA) and Water Extractable Organic Carbon (WEOC) by TOC analyzer (TOC-5000A, Shimadzu, Kyoto-Japan). NH$_4$-N by the indophenol-blue method was measured using soil-KCl extract by UV-Vis Mini Spectrophotometer 1240, Shimadzu, Kyoto-Japan.

**CO$_2$ Flux Measurement and Calculation**

Soil CO$_2$ flux was measured from May 2013 to April 2014 by closed chamber with 40 cm x 30 cm in size (Toma and Hatano, 2007). The measurement was conducted in 2 to 28-day intervals during the growing season and 10 to 30-day intervals during the non-growing season. Gas sampling was conducted between 8:00 and 11:00 in each measuring day to minimize the effect of diurnal temperature variation.

Gas sample was taken from the chambers using a 50 mL syringe and it was collected at 0 and 6 minutes. For 0 minute sampling, gas was taken before the chamber was closed and after chamber was closed for 6 minutes sampling. Afterwards, gas samples were placed in a Tedlar bag. CO$_2$ was analyzed using an infrared CO$_2$ analyzer (ZFP9FC11, Fuji Electric System, Tokyo, Japan) just after sampling.

Gas flux is the gradient of gas concentration in chamber over time. Positive flux indicates gas emission from soil surface into the atmosphere, while negative flux indicates gas uptake from the atmosphere. It was calculated by the equation below.

\[ F = \rho \times V/\Delta t \times A \times \Delta c/\Delta t \times 273/T \times \alpha \]  

Where, $F$ is CO$_2$ flux (mg C m$^{-2}$ h$^{-1}$); \( \rho \) is the density of CO$_2$ at the standard conditions (1.98 \times 10^6 mg m$^{-3}$); \( V \) (m$^3$) and \( A \) (m$^2$) are the effective volume and bottom area of the chamber; \( \Delta c/\Delta t \) (10$^6$ m$^{-3}$ m$^{-3}$ h$^{-1}$) is the gradient of gas concentration in the chamber during the close of chamber; \( T \) is the absolute temperature (K); and \( \alpha \) is the ratio of C atom to CO$_2$ molecule (CO$_2$= 12/44).
The cumulative gas flux was calculated by Equation (2):

\[
\sum_{i=1}^{n} R_i x 24 x D_i
\]

(2)

Where, \( R_i \) is the mean gas flux (mg m\(^{-2}\) h\(^{-1}\)) of the two successive sampling dates, \( D_i \) is the number of days in the sampling interval and \( n \) is the number of sampling times.

**Soil CO\(_2\) Concentration Measurement**

Soil CO\(_2\) concentration was measured using a method proposed by Katayanagi and Hatano (2012). Silicon tubes with PVC pipes (inner diameter of 15 mm and outer diameter of 20 mm) were installed in the soil at 0, 10, 20, 30, 40, 50, and 60 cm depths with four replications at each depth in each plot. Pipes were left in the field throughout the winter period (December to April) and a composite sample of soil gas was collected from each pipe at the same time of CO\(_2\) flux measurement. Sampling was collected slowly to prevent contamination by gas from non-targeted depths and inserted to a Tedlar® bag and N\(_2\) gas was inserted to replace the removed gases. Ten mL gas from Tedlar® bag was then transferred into a vial bottle for CO\(_2\) analysis.

**Statistical Analysis**

Statistical analysis (ANOVA and Pearson correlation) was performed using SPSS 16.0 (SPSS Inc., Chicago, US).

**RESULTS**

**Environmental Variables**

The annual precipitation during the study period was 1,239 mm. Daily precipitation (Figure 1-A) showed that the highest precipitation was 97.5 mm d\(^{-1}\) and occurred on September 16, 2013. During winter season, the precipitation was meager and the medium precipitation (40.5 mm day\(^{-1}\)) occurred in the end of the melting period. Snowfall started in the middle of November 2013. The maximum snow height (38 cm) was reached on February 16, 2014, and it coincided almost with the minimum air and soil temperature (~11 and ~2°C, respectively).

Frozen depth in grassland was deeper than in cornfield i.e. around 20 and 30 cm, respectively, and it disappeared on April 02, 2014 (Figure 1-B). In the end of March, soil temperature raised above 0°C and there was a rain that accelerated melting process in both sites in the middle of March. Snow completely disappeared on April 02, 2014. Air and soil temperature (Figures 1-C and -D) indicated the similar trend. The maximum temperature occurred in August 2013 (summer season) and started to decrease in September, then reached 0°C in December 2013.

Winter season lasted 4 months, during which we determined the snow height (Figure 1-A), soil freezing depth (Figure 1-B), soil temperature (Figure 1-C) and field observation as well. Winter started on December 12, 2013, when the soil temperature decreased to near 0 degree and soil in the surface began to freeze. On April 3, 2014, the temperature increased to above 0 degree and there was no frozen soil in the surface, this situation showed that the winter was over (Table 1).

**Soil NH\(_4\)-N, NO\(_3\)-N, WEOC Concentration in Winter Period**

Soil chemical properties in 0-10 cm depth during winter in grassland and cornfield are shown in Figure 3. In grassland, NH\(_4\)-N concentration in F plots in the freezing period was significantly higher (P< 0.05) than in the melting period. Similar situation also occurred in MF plots in cornfield. Whereas, in MF plots of grassland and F plots of cornfield, NH\(_4\)-N concentration in freezing period was lower than in melting period (Figure 3-A).
Table 1. Cumulative CO$_2$ flux in each period of grassland and cornfield.

<table>
<thead>
<tr>
<th>Period</th>
<th>Cumulative CO$_2$ emissions (Mg C ha$^{-1}$ period$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Grassland</td>
</tr>
<tr>
<td></td>
<td>F</td>
</tr>
<tr>
<td>Before freezing</td>
<td>(1/5/2013 - 22/12/2013)</td>
</tr>
<tr>
<td>Freezing</td>
<td>(22/12/2013 - 13/3/2014)</td>
</tr>
<tr>
<td>Melting</td>
<td>(13/3/2014 - 2/4/2014)</td>
</tr>
<tr>
<td>After melting</td>
<td>(3/4/2014 - 30/4/2014)</td>
</tr>
<tr>
<td>Annual</td>
<td>(1/5/2013 - 30/4/2014)</td>
</tr>
</tbody>
</table>

Figure 1. Seasonal pattern of daily precipitation and snow depth (A), soil freezing depth (B), daily mean soil temperature ($^\circ$C) in grassland and cornfield from May 2013 to 30 April, 2014.

Soil NO$_3$-N concentration in melting period in all plots (except in MF plot of grassland) were higher than freezing period (Figure 3-B). The significant difference ($P<0.05$) between NO$_3$-N concentrations in freezing and melting periods occurred in cornfield (Figure 3-B).

In F plots of both grassland and cornfield, soil WEOC concentration was higher in freezing than melting period. On the other hand, soil WEOC in MF plots of grassland
and cornfield was almost similar in freezing and melting period.

**Annual CO₂ Flux**

CO₂ flux in both F and MF plots of grassland generally showed similar pattern (Figure 2): it increased after melting period in May 2013 then reached the peak in July and August 2013 (summer). Flux decreased gradually until winter started (December 2014). CO₂ flux in MF plots was significantly higher than in F plots, particularly in June, July, and August. Flux decreased when herbicide was applied on August 13. Herbicide killed the grass including root; therefore, there was no contribution of CO₂ flux from root respiration. CO₂ flux increased again after soil plowing in renovation grassland (August 28). Soil plowing replaced sub-soil with top soil and made the dead root abundant on the top soil. It stimulated decomposition, hence, increased CO₂ flux.

CO₂ flux during winter in both grassland and cornfield showed a similar pattern. CO₂ flux in F plots of grassland was relatively higher than MF plots; meanwhile, in MF plots of cornfield, CO₂ flux was relatively higher than in F plots. However, we observed no significant interaction between period of winter (freezing and melting), fertilizer treatments (F and MF), and field (grassland and cornfield) in CO₂ fluxes.

**Soil CO₂ Concentration**

Soil CO₂ concentration near the soil surface (0-5 cm) of grassland during winter was lower than CO₂ concentration in the deeper soil layer. Soil CO₂ concentration increased gradually from the beginning of freezing period and reached the maximum concentration in the beginning of the melting period (the end of March). From this time on, the concentration decreased and reached the minimum concentration in the end of melting period (Figure 4-A). In cornfield, soil CO₂ concentration from 0-30 cm depth during winter in F and MF plots.
Figure 3. Concentration of NH$_4^-$N, NO$_3^-$N, Water Extractable Organic Carbon (WEOC) from soil at 0-60 cm depth in grassland and cornfield in 12/03/2013 and 04/02/2014.

Figure 4. Soil CO$_2$ concentration in F plots (A) and MF plots of grassland (B), and F plot (C) and MF plots of cornfield (D).
was relatively similar (Figure 4-C). The maximum concentration in MF plot occurred in the deeper soil depth in freezing period (December to February). On the other hand; the maximum CO$_2$ concentration in MF plots was found in melting period (March to April), almost in the entire depth (Figure 4-D).

**DISCUSSION**

**Effect of Fertilizer Management on CO$_2$ Flux**

In this study, about 2-10% of the cumulative annual CO$_2$ flux occurred during winter season and the flux was relatively higher in cornfield than in grassland. Less than 1% of annual CO$_2$ flux in all plots of grassland and cornfield was released in melting period, whereas in freezing period, CO$_2$ flux was relatively higher than melting period in both grassland and cornfield (Table 1). Meanwhile, the CO$_2$ flux in F and MF plots in cornfield is apparently similar in both freezing and melting periods. On the other hand, during the freezing time in grassland, CO$_2$ flux was lower in MF than in F plots. However, a contrary circumstance occurred in melting period of grassland site where the CO$_2$ flux in F was lower than MF plots.

Fertilizer treatments (F and MF) did not induce the CO$_2$ fluxes that were released in all plots during winter (Table 2). Generally, in cooler climate, soil has more organic matter than in warmer climate, because of slower mineralization/decomposition rates (Bot and Benites, 2005). The cumulative CO$_2$ flux in all plots of grassland and cornfield was generally low in melting period (Table 1). Although on 2 April, 2014, the WEOC in all plots (Figure 3-C) was relatively high and available for soil microorganism in 0-10 cm depth, the decomposition process was delayed due to the water saturated soil. When the soil temperature increases after winter and water is drained from the soil, the CO$_2$ flux in all plots inclines to increase (Figure 2). This condition is an affirmation of the previous study by Mukumbuta et al. (2017) who reported the WFPS (water-filled pore space) had a significant negative correlation with the CO$_2$ flux. Our statistical analysis also supported this phenomenon where there was no significant difference between CO$_2$ flux of grassland and cornfield in melting period (Table 2).

The concentration of NH$_4$-N and NO$_3$-N did not affect the CO$_2$ flux during winter season. This is proven by the results of the cumulative CO$_2$ flux in cornfield, which was lower in MF plots than in F plots, although the NH$_4$-N concentration in 0-10 cm depth was almost equal in both plots (Figure 3-A). This result clarified the study of Mukumbuta et al. (2017) that suggested NO$_3$-N concentration had a significant negative correlation with CO$_2$ flux. Our data also confirm the previous study by Liang et al. (2015) who reported that there was no significant effect of N addition on soil CO$_2$ flux during winter season.

The cumulative CO$_2$ flux in winter from whole plots in grassland and cornfield was 0.2 to 0.6 Mg C ha$^{-1}$ period$^{-1}$. These results

<table>
<thead>
<tr>
<th>Source</th>
<th>$df$</th>
<th>Mean square</th>
<th>$F$</th>
<th>Sig</th>
</tr>
</thead>
<tbody>
<tr>
<td>Field</td>
<td>1</td>
<td>5336.460</td>
<td>13.668</td>
<td>0.001</td>
</tr>
<tr>
<td>Fertilizer</td>
<td>1</td>
<td>1012.960</td>
<td>2.594</td>
<td>0.120</td>
</tr>
<tr>
<td>Period</td>
<td>1</td>
<td>1400.406</td>
<td>3.587</td>
<td>0.070</td>
</tr>
<tr>
<td>Field ×Fertilizer×Period</td>
<td>1</td>
<td>1291.564</td>
<td>3.308</td>
<td>0.081</td>
</tr>
<tr>
<td>Error</td>
<td>24</td>
<td>390.433</td>
<td></td>
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</tr>
</tbody>
</table>
were lower compared to CO₂ flux from *Querqus/Betulla* forest (0.9 Mg C ha⁻¹ period⁻¹) (Mariko *et al*., 2000) and higher than cropland in Northern China that released 0.28 to 0.45 Mg C ha⁻¹ period⁻¹ (Shi *et al*., 2012). These results also confirm the previous studies by Alm *et al*. (1999) and Van Bochove *et al*. (2000) that suggested the activity of microorganism did not stop in winter even in colder temperature. Mariko *et al*. (2000) also reported that fungi in tundra soil are able to respire at temperatures as low as -6.5 to -7.5°C. Soil CO₂ flux in winter was not influenced by snow height and frozen depth even though CO₂ flux tended to be low when the soil freezing depth was deepened. These results confirm the previous study by Ohkubo *et al*. (2012).

Another result showed that the cumulative CO₂ flux was higher in freezing than in melting period, in both grassland and cornfield. This phenomenon could be explained by the soil that was almost saturated in melting period and inhibited the microbial activity.

**Soil CO₂ Concentration and CO₂ Flux during Winter**

The CO₂ concentration during winter indicated the opposite pattern for each site (grassland and cornfield). Generally, the CO₂ concentration was higher in grassland than in cornfield for the entire soil depth. This circumstance was, perhaps, due to differences in CO₂ contribution by root respiration in grassland and cornfield. Root respiration contributed to around 38% of soil respiration in Osaka grassland (Yoneda and Okata, 1987) and 31-51% in Tsukuba grassland (Wang *et al*., 2005).

We assumed that there was no contribution of root activity/respiration to soil CO₂ concentration. It was due to the corn in the field was in residue (dead corn) form. Therefore, CO₂ concentration in cornfield was lower than in grassland. Another explanation was that corn residue consisted of stubble and roots that were somewhat difficult to decompose. Our visual observation showed that stubble and roots of corn did not significantly change in winter compared to the period before freezing. This is because the slow decomposition during winter resulted in low CO₂ concentration in the soil. Moreover, the slow decomposition was due to C/N ratio of cornfield in the study site, which was quite high viz. 49.90% (Nugroho *et al*., 2015).

Some variables corresponded to soil CO₂ concentration in grassland and cornfield during winter (Table 3). These relationships showed that mineralization was the major process of CO₂ production in winter. Mineralization released NH₄-N and carbon in to the soil. Therefore, the concentration of NH₄-N and carbon (WEOC) in the soil increased during freezing to melting (Figure 3). Mineralization also emitted CO₂ into the atmosphere. The previous observation at the same site by Nugroho *et al*. (2015) suggested that WFPS in grassland and cornfield during winter was 80 to 90%. It means that CO₂ was still probably emitted into the atmosphere even when soil was almost saturated.

Soil CO₂ concentration changes was not linear with CO₂ flux. In grassland, high concentration during freezing was contrary to CO₂ flux. However, our investigation suggested that after soil frost melting and the when temperature was around 0°C, CO₂ flux increased drastically. This condition was also reported by Elberling and Brandt (2003), Dörsch *et al*. (2004) and Maljanen *et al*.

**Table 3.** Relationship between soil CO₂ concentration and soil properties.

<table>
<thead>
<tr>
<th></th>
<th>NH₄-N</th>
<th>NO₃-N</th>
<th>WEOC</th>
</tr>
</thead>
<tbody>
<tr>
<td>CO₂ concentration</td>
<td>0.39**</td>
<td>-0.39**</td>
<td>0.19</td>
</tr>
<tr>
<td>n</td>
<td>48</td>
<td>48</td>
<td>48</td>
</tr>
</tbody>
</table>

** Correlation is significant at the 0.01 level.
al. (2007). High CO\textsubscript{2} flux might be originated from CO\textsubscript{2} that was trapped on frozen soil during freezing period and was released to the atmosphere when soil unfreezed. In addition, increasing soil temperature will also increase the microbial activity that influences CO\textsubscript{2} flux (Dörsch et al., 2004). Even though CO\textsubscript{2} flux seems to be correlated with CO\textsubscript{2} concentration in melting period, further studies that involve gas diffusion are required to comprehensively understand the correlation between CO\textsubscript{2} flux and soil CO\textsubscript{2} concentration.

CONCLUSIONS

During winter season, soil CO\textsubscript{2} was continuously released from F and MF plots in grassland and cornfield. This proves that low temperature does not stop the activity of soil microorganism. Moreover, the addition of fertilizer and manure-fertilizer to grassland and cornfield does not stimulate the winter CO\textsubscript{2} emission. CO\textsubscript{2} flux that was emitted in all plots of grassland and cornfield in freezing period was relatively higher than CO\textsubscript{2} flux in melting period, even though statistically the difference was not significant. Another result indicated that CO\textsubscript{2} flux was different between grassland and cornfield during winter season. The difference in soil properties (soil texture, drainage, WFPS, C/N ratio) in both locations was most probably the cause of these variations, although we did not conduct a detailed analysis in this study. Soil CO\textsubscript{2} concentration was also measured to observe the correlation between CO\textsubscript{2} flux in the soil and CO\textsubscript{2} flux in the atmosphere. It turned out that soil CO\textsubscript{2} concentration was not in linear correlation with CO\textsubscript{2} released in the atmosphere during winter. However, CO\textsubscript{2} concentration in the soil had a positive correlation with soil NH\textsubscript{4}-N and WEOC content and a negative correlation with soil NO\textsubscript{3}-N concentration. A comprehensive study, particularly in gas diffusion from sub soil into the soil surface, is still necessary to be conducted.

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