Long-term grazing effects on soil-atmosphere exchanges of CO_2, CH_4 and N_2O at different grasslands in Inner Mongolia: a soil core study

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Abstract

Regional greenhouse gas (GHG) budgets in vast grasslands may be changing due to overgrazing and grassland types. However, the comprehensive effects of grazing patterns, environmental factors and grassland types on soil carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O) exchanges have been poorly studied. This study investigates the effects of long-term grazing on the soil-atmosphere exchanges of CO_2, CH_4 and N_2O in important processes within different grasslands in Inner Mongolia, China. Using manual static chamber and gas chromatography, we measured the fluxes of CO_2, CH_4 and N_2O from intact soil cores of paired grazed/ungrazed sites collected from two typical steppes (S*tipa grandis* and *Leymus chinensis)*: one wetland in a flood plain and one desert steppe in the region of the Xilin River catchment, Inner Mongolia. Soil gas flux and concentration measurements were conducted in four simulated conditions (*i.e.*, drought, dry-wet, intense rainfall and freeze-thaw), which represent important processes in annual GHG exchanges. Extreme drought significantly inhibited CO_2 and N_2O emissions in all plots but did not change the CH_4 uptake by typical steppes. Dry-wet transition and intense rainfall could remarkably promote soil CO_2 emission pulses at different types, significantly decrease CH_4 uptake by typical steppes, and arouse N_2O emission pulses at all plots with different times of occurrence. During the freeze-thaw simulation, temperature-induced soil CO_2 emission and CH_4 uptake/emission presented a clear alternative variation, while soil thaw only slightly increased (<15 μg N m^{-2} h^{-1}) in the steppes and sand dunes and was significantly higher in the wetland (11-96 μg N m^{-2} h^{-1}). Long-term grazing significantly inhibited soil respiration rates at all grassland types, significantly decreased CH_4 uptake by the *Leymus chinensis* steppes, and did not show significant influence on N_2O emission due to large spatial variations for all types. Compared to the ungrazed *Leymus* steppes, *Stipa* steppes, sand dune and wetland, continuously grazed sites were significantly reduced by 22%, 38%, 48% and 47% in total GHG emissions, respectively. Our results indicate that the potential of the steppe soil CH_4 sink function can be offset by N_2O emission, especially in over-grazed plots. Furthermore, N_2O emissions should be considered in wetland rangelands with significantly higher N_2O emission potential (range: 0-343 μg N m^{-2} h^{-1}) more than steppes (range: 0-132 μg N m^{-2} h^{-1}) and sand dunes (range: 0-49 μg N m^{-2} h^{-1}). Nevertheless, comprehensive evaluation of the grazing effect on ecosystem GHG emissions merits consider in both field observation and incubation experiments because soil properties and environmental factors could be changed by vegetation growth in different grazing practices.

Keywords: **greenhouse gas, semiarid grassland, grazing, freeze-thaw, dry-wet, intense rainfall, Inner Mongolia**

Introduction

Temperate steppes, covering approximately one-tenth of the earth's land surface, play important roles in greenhouse gas (GHG) emissions or uptake (Sutie *et al.*, 2005). In general, steppe soils serve as a reservoir for large amounts of organic carbon and a sink or source of methane (CH_4) and nitrous oxide (N_2O) (Soussana et al., 2007). The size of sinks or sources in steppe soils is strongly controlled by the physical factors of soil (*e.g.*, texture, organic matter and soil temperature moisture) but also by land uses (*e.g.*, grazing and reclaiming) and climate change (*e.g.*, rainfall and air temperature) (Galbally *et al.*, 2008). Due to the large area and the mitigation potential, clarification of soil-atmosphere exchanges of GHG in temperate steppes is crucial to our understanding of the global C/N cycle and mitigation options.

The GHGs (*e.g*., CO_2, CH_4 and N_2O) are produced or consumed as a result of microbial processes in the soil (Galbally *et al*., 2008). The soil-atmosphere fluxes of GHGs heavily depends on the physical factors of soil, which affects the microbial processes of production, consumption and gas transport in the soil (Smith *et al*., 2003). Soil temperature and water content have been reported to be the two most important factors linked to soil GHG fluxes (*e.g*., Wang *et al.*, 2005; Wang and Fang, 2010). They directly affect production and consumption of GHGs through their effects on microorganisms and root activity. In addition, gas diffusivity, which depends on air-filled porosity (and thus varies inversely with water content), controls the movement of the gases to and from the atmosphere and indirectly controls the capacity of microbial processes by affecting soil aeration. Studies have shown that a large amount of water input during periods of freeze-thaw, dry-wet and intense rainfall could contribute greatly to annual GHG budgets (*e.g.*, Hao *et al*., 2010; Wolf *et al*., 2010; Yao *et al*., 2010). Wolf et al. (2010) found that the N_2O pulses during spring thaw dominate the annual N_2O emission in a typical steppe. The function of a carbon sink in the grassland ecosystem may change to the source during extreme dry years (Hao *et al*., 2010). Soil CH_4 uptake in grasslands have been reported to be sensitive to soil temperature and water content at the seasonal and annual scales (Chen *et al*., 2011). These studies have suggested these temperature- or water-related periods are important in affecting the greenhouse gas exchange mechanisms and regional GHG budgets.

At present, most of the grasslands suffer from degradation due to overgrazing and poor management, especially arid and semiarid grassland, where 73% of rangeland is degraded (Steinfeld *et al.*, 2006). Inner Mongolian steppes (approximately 87 million hectare), covering more than 20% of the total grassland area in China, are characterized by a typical temperate semiarid climate (Wang *et al.*, 2005). As a major base of animal husbandry, livestock numbers have increased significantly in the past few decades and most natural steppes have been facing the accelerating threats from overgrazing (Tong *et al.* 2004). Overgrazing has resulted in widespread rangeland degradation or desertification of steppes, associated with changes in soil properties (Kang *et al.*, 2008), plant diversity and productivity (Gao *et al.*, 2011) or activities of soil microorganisms (Su *et al.*, 2005). Recently, it was shown that the potential of soils to acts as a sink or source of atmospheric CH_4 or N_2O in steppes may have been reduced significantly if heavily grazed continuously for three years (Wolf *et al.*, 2010; Chen *et al.*, 2011). This indicates that greenhouses gas exchange in steppe ecosystems might have changed significantly with the expansion of degraded grasslands and global warming. Furthermore, these steppe types are characterized by a typical temperate semiarid climate with a significant inter-annual variability (Liu *et al.*, 2008). However, the comprehensive estimate of grazing practices and environmental factor effects on total GHG exchanges during freeze-thaw, dry-wet and intense rainfall periods have been rare until now. In addition to typical steppes, wetland rangelands (*e.g.*, flood plain and river valley meadows) and sand dune rangelands are very valuable grazing places due to plentiful water supply, rich vegetation, special water storage performance or leeward side for winter grazing. However, the grazing effect on GHG emissions from these two types of rangeland has been poorly studied. Thus, both aspects indicate the large uncertainties of these important parameters for regional GHG budgets.

The objective of this study is to understand the effects of steppe types, grazing practices and environmental factors on the potential of soil GHG exchanges. Using soil core incubation and gas chromatography methods, we determined the fluxes of CO_2, CH_4 and N_2O from intact soil cores of paired grazed/ungrazed sites collected from two typical steppes (S*tipa grandis* and *Leymus chinensis)*, one wetland in a flood plain and one desert steppe in the region of the Xilin River catchment of Inner Mongolia. The flux measurements were conducted in four simulated conditions (*i.e.*, dry, dry-wet, intense rainfall and freeze-thaw), which represent important processes in annual GHG exchanges. In addition, soil GHG concentrations at four depths (5, 10, 20 and 30 cm) were selectively measured to further understand soil-atmospheric GHG exchanges. The differences of

GHG exchange rates under various combined conditions and possible mechanisms are estimated and discussed.

Materials and methods

Study region

The study was conducted in different grassland ecosystems located in the Xilin River catchment near the Inner Mongolia Grassland Ecosystem Research Station (IMGERS, 43°38′ N, 116°42′ E; 1100 m a.s.l.) of the Chinese Ecosystem Research Network. The local climate is characterized as a temperate continental climate. In this region, the growing season usually starts in early May and ends in late September, and the remaining seven months are the non-growing season, when grass vegetation and roots are dormant. The 25-year mean annual air temperature (1982–2007) recorded by IMGERS was 0.7°C, with a mean January temperature of -21°C and a July mean of 19°C. The annual mean precipitation is 330 mm (range: 166–507 mm yr-1 in 1982–2007), of which 85% falls between May and September. The average air temperature and precipitation values (mostly in snowfall) during the non-growing season were -8.7°C and 52 mm.

The typical steppe covers approximately 85% of the area of the Xilin River catchment. The most common typical steppe types are communities either dominated by *Stipa grandis* or *Leymus chinensis*. The soil types of the *Stipa grandis* and *Leymus chinensis* steppes are *kastanozem* and *chernozem*, respectively. Sand dunes, occupying 6.5% of the study area, are characterized by undulating topography, where sand forms a sequence of depressions and hills. Dense vegetation, particularly *Ulmus pumila* and other tree genera, *i.e.*, *Betulus spp., Malus spp., Prunus spp.* and *Populus spp.* dominate north- to northwest-facing slopes, as well as in depressions. South- to southwest-facing slopes generally have sparser shrubs and grassland vegetation. Wetland covers only a tiny portion of the study area (approximately 0.4%). It borders most of the Xilin River, its tributaries and some flat areas of episodic streams. These areas are divided into two parts: dry and wet marshland. Dry marshland is frequently wet, but it becomes dry during one to three months of the summer. In contrast, wet marshland is wet all year round. However, wet marshland has also been noted to dry out during years with low summer rainfall (approximately 100 mm). The vegetation found in these areas consists mainly of marshland species such as *Phragmites australis, Carex appendiculata, Iris lactea var. chinensis* and *Hippuris vulgaris.*

Soil core sampling

Grazing practices (or stocking rate) and grassland type are indicators of soil GHG emissions or uptake in grasslands. It is crucial to consider these two indicators to find the best balance point in grassland quality and GHG emissions, and then select suitable grazing patterns for local grassland management. Paired experimental rangelands or plots under ungrazed and grazing practices in the Xilin River catchment were established by IMGERS. To determine the long-term grazing effect on the potential of soil GHG exchanges, four paired plots were chosen, including one plot that has remained ungrazed since 1979 (SUG79), and an adjacent, continuously grazed rangeland (SCG) at *Stipa grandis* steppe; one plot that has remained ungrazed since 1979 (LUG79) and an adjacent, continuously grazed rangeland (LCG) at the *Leymus chinensis* steppe; one plot that has remained ungrazed since 1989 (WUG89) and an adjacent, continuously grazed rangeland (WCG) in the Xilin river flood wetland; and one plot that has remained ungrazed since 2002 (SDUG02) and an adjacent, continuously grazed rangeland (SDCG) in a sand dune place near IMGERS. The paired plots have similar backgrounds regarding soil and vegetation characteristics. Animals (sheep and goats) could walk freely during the grazing seasons in all grazed plots. All long-term fenced plots had significantly higher values in vegetation height, coverage and plant density, compared to adjacent grazed rangelands. Detailed location, grassland managements and soil properties at paired ungrazed and grazed grasslands are shown in Table 1.

Soil core sampling at the paired plots in four types of grasslands was conducted on September 20, 2008. The grass above ground were cut off to near the ground surface before sampling. At each plot or rangeland, undisturbed soil cores at a depth of 0–40 cm were taken in quintuplicates using specialized semi-column steel pipe and were put in manual chambers. Manual chambers were made from nontransparent polyvinyl chloride (PVC) pipe (diameter, 16 cm; length, 60 cm; volume, 4.0 L; basal area, 0.02 m^2) with a twist lid and a bottom (PVC) with a butyl rubber septum and a rubber O ring to form a gas-tight seal. After drying in the shade, all soil core samples were brought to the laboratory at the Institute of Meteorology and Climate Research - Atmospheric Environmental Research (IMK-IFU), Karlsruhe Institute of Technology (KIT) and stored in dry conditions and well aerated at 4°C for further analysis.

Experimental design of soil core incubation

Gas fluxes and soil gas concentrations were measured using the sampled soil core in manual chambers and gas chromatography techniques under different stimulated dry-wet, intense rainfall, freeze and thaw cycles. We conducted undisturbed soil core incubation and these measurements for each grassland type in sequence and the detailed steps as follows:

1) **Soil activation.** More than ten months in storage has resulted in completely dry soil cores, thus we activated soil cores by adding a simulated 15 mm rainfall event before starting the measurement. Standard rain mixture included 11 mg $CaCl_2$, 24.4 mg KCl, and 18.6 mg Na_2SO_4 in 1000 mL of distilled water.

2) **Drought simulation.** After activation, all soil cores were placed at room temperature for approximately 40 days until topsoil of soil cores was completely dried. Then, we put all soil cores in electronic controlled incubators, which can regulate air temperature, and kept the air temperature at 20°C. Fluxes of CO_2, CH_4 and N_2O were measured for three days during the simulated drought condition. Hereafter, the rainfall simulation and flux measurement were conducted in the incubator at room temperature.

3) **Dry-wet cycle simulation.** After the flux measurement under the simulated drought condition, we added 20 mm of rainfall water solution to the simulated dry-wet process. Then, GHG fluxes were continuously measured for seven to eight days until the flux variations tended to be stable. The topsoil moisture at the end time was approximately 10%-20%, which was similar to the normal values during the growing season in the grassland (mean: 15%).

4) **Intense rainfall simulation.** The 20 mm simulated rainfall solution was added to the soil core to simulate an intense rainfall event, aiming to evaluate the difference between grazed and ungrazed soil affected by intense rainfall in different grassland types. Similar GHG flux measurements were continuously conducted for seven to eight days until the flux was nearly stable.

5) **Freeze-thaw simulation.** In the steppes and sand dune, precipitation was generally small before the winter, so the topsoil moisture during the whole freeze period was low. In the following year, during snow melt and soil thawing, melted water from snow permeated into soil with air temperature increases. Soils were frozen at low temperatures, and the freeze-thaw cycle transition

occurred each day. Although soil water content mainly depends on the river flood situation, we used the same simulated condition to keep pace with other types. After completion of the second rainfall simulation and flux measurement, we removed all soil cores from incubators and kept them at room temperature until soil moisture was approximately 5%. This was to create a similar soil condition with that of the real field environment. Then, soil cores were put into freezers for 20 days to simulate the freeze period, and soil flux measurements were conducted in the end of this period. In the next step, soil cores were set into an incubator and regulated to the simulated temperature of 5°C, which was close to the actual temperature in the beginning of the spring-thaw period. We added 20 mm of rainfall water solution to simulate the penetration into soil from snow melted water. Four hours later, we measured the GHG fluxes from soil cores during the simulated thaw period. In addition, they were then put into freezers with a temperature of -10°C for 23-24 hours, and GHG fluxes were measured GHG fluxes, which represented the freeze process. In the following 10-12 days, the simulation of freeze (-10°C) and thaw (5°C) were transformed for the GHG flux measurement every day.

Measurements of GHG fluxes and soil gas GHG concentrations

The fluxes of CO_2, CH_4 and N_2O were measured using a static chamber method. The PVC tubes (soil cores) were sealed with stainless steel lids, enclosing a headspace of ~2 L. The lids were equipped with a circulating fan to ensure complete gas mixing. In addition, they had two ports, one with a septum for gas sampling and the other holding a Teflon tube (with an outer diameter of 1/8 inches and a length of 20 cm) to ensure the equilibration of air pressure between the inside and outside. A 20 mL gas sample was taken with a gas-tight syringe from the headspace immediately after sealing and again after 15, 30, 45, and 60 min. The air samples stored in the syringes were analyzed by gas chromatography (SRI 8610C, Texas Instruments, Torrance, CA, USA), which was equipped with an electron capture detector (ECD) for N_2O analysis and a flame ionization detector for CH_4 and CO_2 analysis. The CO_2 was converted to CH_4 in a methanizer prior to analysis. The analytical conditions for analyzing the concentrations of CO_2, CH_4 and N_2O and the methods for calculating the fluxes of each gas have been described by Holst *et al.* (2007) and Zheng *et al*. (2008). We used a purge gas (CO_2: N_2 = 10:90) for the ECD at a rate of 1–3 mL min-1 to avoid cross-interference between CO_2 and N_2O in the air samples. The gas fluxes were calculated from changes in the initial rate of concentration change within the chambers' headspace; this was calculated from the initial slope of a non-linear regression of concentration against time (Kroon *et al.*, 2008). Negative flux values indicate gas uptake from the atmosphere, and positive flux values indicate gas emissions to the atmosphere. The detection limits for our approach were 1.5μg N m^{-2} h^{-1} for N_2O, 4 μg C m^{-2} h^{-1} for CH_4 and 1 mg C m^{-2} h^{-1} for CO_2 based on the chamber dimensions, the sampling time, and the reproducibility of repeated measurements for standard gases (0.7% for CH_4, 0.8% for CO_2 and 0.6% for N_2O). To assess the grazing effect on the total GHG balance of steppes, we converted CH_4 and N_2O emissions into CO_2 equivalents (CO_2-*eq.*) using the global warming potential factors of 25 (for CH_4) and 298 (for N_2O) (IPCC, 2007).

Soil gas samples were taken from each soil core with a stainless-steel tube (inner diameter: 1 mm) near the surface of the soil cores and at 5, 10, 20, and 30 cm depth during the period of gas flux measurements. Ten milliliters of soil gas were collected every two days, and the CO_2, CH_4 and N_2O concentrations were analyzed using gas chromatography (SRI 8610C, Texas Instruments, Torrance, CA, USA). The detection method was the same as that described above for the gas flux measurement. In addition, among five soil cores, four soil cores were used for GHG flux measurements, and the other one was synchronously incubated to measure topsoil volumetric content (0–6 cm) by a portable FDR probe (ThetaKit, Delta-T Devices, Cambridge, UK).

Statistical analysis

The significance of differences in mean GHG flux and CO_2-*eq*. emissions among the treatments was investigated using a one-way ANOVA with Tukey's HSD test (SPSS 11.5, SPSS Inc., Chicago, USA). All plots (line, bar and contour) were made using SigmaPlot 10.0 (SPSS Inc., Chicago, USA).

Results

Soil properties

At the *Leymus* and *Stipa* steppes, long-term grazing significantly reduced topsoil (0-5 cm) bulk density, the contents of total carbon, nitrogen and silt (Table 1). Although grazing duration was much less than the steppes, there were also obvious decreasing trends in bulk density, pH and silt contents at the sand dune rangelands under heavy grazing practices. The wetland plots with long-term and moderate grazing did not change soil bulk density and pH but significantly reduced total carbon and nitrogen content and increased the silt content of the topsoil.

Carbon dioxide fluxes and concentrations

The CO_2 fluxes ranged from 0 to 92 mg C m^{-2} h^{-1} in the steppes, from 0 to 110 mg C m^{-2} h^{-1} at the sand dune rangeland and from 1 to 79 mg C m^{-2} h^{-1} at the wetland during the whole incubation period (Fig. 1). All investigated plots had similar varying trends of CO_2 emissions under simulated dry, dry-wet, intense rainfall and freeze-thaw periods (Fig. 1). The weak CO_2 emissions (<10 mg C m^{-2} h^{-1}) were determined at investigated plots, except WUG89 under the extreme dry period. The following two simulated rainfall events substantially promoted the CO_2 fluxes in the first day after watering and gradually decreased to stable levels; the CO_2 fluxes showed the alternative changes of “low-high-low” with simulated freeze and thaw practices, but the fluxes in freeze-thaw were significantly lower than those in dry-wet and intense rainfall processes.

In the steppe and sand dune rangelands, soil CO_2 concentrations greatly varied during dry, dry-wet, intense rainfall, and freeze-thaw periods (Fig. 4a-4c). Slightly higher soil CO_2 concentrations (400-500 ppmv) than ambient CO_2 concentrations during the dry period indicates low CO_2 emissions, whereas two simulated rainfall events (*i.e.*, dry-wet and intense rainfall practices) rapidly promoted soil CO_2 production with high concentrations (steppes: 1000-1500 ppmv; sand dune: 5000-7000 ppmv). Although soil CO_2 concentrations (< 1000 ppmv) were obviously lower than simulated dry-wet and intense rainfall events, the significant differences of CO_2 concentrations were determined between simulated frozen and thaw periods. In contrast to other rangeland types, we observed the continuously higher soil CO_2 concentrations (2000-6000 ppmv) at the depth of 20-30 cm in the wetland throughout all periods except the freeze-thaw period. Simulated rainfall increased the topsoil (5-15 cm) CO_2 concentrations, but the increased ranges were far lower than other types.

Compared with the lowest soil moisture condition (*i.e.*, 5%), the CO_2 fluxes in the steppe and sand dune rangelands substantially increased under the conditions of 12.5% and 20% with no significant differences between them (Fig. 7a). At the wetland, CO_2 fluxes gradually increased with the increase in soil moisture from 5% to 20% and had significant differences between 5% and 20%. In addition, the mean CO_2 flux under the 10°C condition (*i.e.*, 20.5 mg C m^{-2} h^{-1}) was approximately 3-13 times of that with -10°C (Fig. 7d). Our results clearly exhibited that grazing practices significantly reduced soil CO_2 fluxes by 25%-47% in all rangeland types (Fig. 8a). Using the data under all simulated conditions in all rangeland types, soil CO_2 fluxes followed the order: wetland (28 mg C m^{-2} h^{-1}) > sand dune (25 mg C m^{-2} h^{-1}) > steppe (22 mg C m^{-2} h^{-1}).

Methane fluxes and concentrations

All steppe and sand dune rangelands were CH_4 sinks with the fluxes varying from -3 to -59 μg C m^{-2} h^{-1}, while wetland rangelands were shifted between weak sinks or sources (-13~13 μg C m^{-2} h^{-1}) of atmospheric CH_4 under various conditions (Fig. 2). The CH_4 exchange responses to simulated dry, dry-wet and intense rainfall were quite different among the steppe, sand dune and wetland rangelands. At *Leymus* and *Stipa* steppes, soil had the strongest CH_4 uptake (-40~-60 μg C m^{-2} h^{-1}) under the extreme dry condition, a fast decrease in the first day after simulated rainfall and gradually increased to a similar level as the dry period in the following days. Compared with steppes, the variation range of CH_4 fluxes (-2~-25 μg C m^{-2} h^{-1}) in the sand dune were smaller and the maximum of CH_4 uptake occurred at the end of simulated intense rainfall. At the wetland rangelands, soils were very weak CH_4 sinks (-1~-4 μg C m^{-2} h^{-1}) under dry conditions, and then were shifted to weak sources (< 13 μg C m^{-2} h^{-1}) at WUG89 after simulated rainfall. During the freeze-thaw period, although the CH_4 fluxes also switched to "low-high-low" pattern (Fig. 2), the flux levels were comparable with other periods.

Under all simulated conditions, soil CH_4 concentrations decreased with the increase in soil depth in the investigated plots except for WUG89 after intense rainfall (Fig. 5), which was consistent with the shift of the soil CH_4 sink and source. Soil CH_4 concentrations in the steppes and sand dune had clear gradual gradients in different depths during the dry period, and quickly decreased after simulated rainfall, especially in deep soil (< 1 ppmv). Compared with the steppes and sand dune, the ranges of decreasing gradient were much smaller in the wetland rangelands, and soil CH_4 concentrations were close to ambient concentrations after simulated intense rainfall. During the freeze-thaw period, the small differences of CH_4 concentrations in soil depths occurred with simulated freeze conditions, while the large ranges occurred with the thaw simulation.

The CH_4 uptake in the *Leymus* steppe did not greatly vary (42–46 μg C m^{-2} h^{-1}) among moistures of 5%, 12.5% and 20%, but they gradually decreased in *Stipa* steppes with increasing moisture (Fig. 7b). Opposite of the steppes, the CH_4 uptake in the sand dune showed the positive trend with the increase in moisture. In addition, the average CH_4 fluxes in wetland plots fluctuated around zero in three moisture conditions with no significant differences. With the change of air temperature from -10°C to 10°C, the CH_4 uptake increased from 27 to 38 μg C m^{-2} h^{-1} in the *Leymus* steppe, from -11 to 30 μg C m^{-2} h^{-1} in the *Stipa* steppe, and from 4 to 20 μg C m^{-2} h^{-1} in the sand dune, showing the substantial effect by temperature. In contrast to the CO_2 fluxes, we determined that only long-term grazing significantly reduced CH_4 uptake by 37% in the *Leymus* steppe. Among all rangelands, soil CH_4 uptake was highest in the *Leymus* and *Stipa* steppes (> 30 μg C m^{-2} h^{-1}), followed by the sand dune (~ 13 μg C m^{-2} h^{-1}) and lowest in the wetlands (Fig. 8b).

Nitrous oxide fluxes and concentrations

The range of N_2O fluxes from steppes, sand dunes and wetlands were 0-132, 0-49 and 0-343 μg N m^{-2} h^{-1} (Fig. 3). Under extreme dry conditions, wetlands were weak N_2O sources with an average flux of 14.5 μg N m^{-2} h^{-1}, while the N_2O fluxes were nearly 0 in other types. The first simulated rainfall aroused N_2O emissions at all rangelands with the peak occurring in the first or second day and gradually decreasing in the following days. Similarly, the second simulated rainfall (*i.e.*, intense rainfall) substantially promoted N_2O emissions with higher fluxes at all rangelands except the sand dunes (Fig. 3). The *Leymus* steppes and sand dunes had only a quick N_2O peak on the first day, but the N_2O peaks in the *Stipa* steppes and wetlands occurred on the third and fifth days with higher N_2O emissions. During freeze-thaw cycles, N_2O fluxes were nearly 0 under freeze conditions and slightly increased (< 15 μg N m^{-2} h^{-1}) with the thaw practices in the steppes and sand dunes. Although the wetlands had similar variation patterns of “low-high-low” with the simulated freeze and thaw practices, the N_2O emissions (11-96 μg N m^{-2} h^{-1}) were significantly higher than other types.

Soil N_2O concentrations in the steppe and sand dune rangelands were close to ambient N_2O

concentrations (~330 ppbv) at the extreme dry condition and quickly increased to 500-1036 ppbv following the first rainfall in the dry-wet cycle, indicating the production of N_2O. The second rainfall also increased the soil N_2O concentrations in the steppe rangelands, and had the maximum (2623 ppbv). Soil N_2O concentrations in the wetlands gradually increased with the first and second rainfall events and reached the maximum of 20401 ppbv. Simulated soil freeze and thaw cycles presented the pattern "low-high-low", with significant promotion in the steppe rangelands. Compared to the steppe and sand dune, the wetland kept a continuously high soil N_2O concentration, showing a stronger N_2O production.

In the steppe and wetland rangelands, soil N_2O emissions gradually increased with the moisture increase, from 5% to 20%, while N_2O emissions remained at continuously low levels at the sand dune rangeland under all moisture conditions (Fig. 7c). The increased ranges of soil N_2O fluxes were substantial at the *Stipa* steppe (2-83 μg N m^{-2} h^{-1}) and the wetland (10-220 μg N m^{-2} h^{-1}) rangelands. Compared with the frozen temperature (-10°C), the N_2O emissions had increasing trends at all rangelands, but we only determined significant differences of N_2O emissions at the *Stipa* steppes. Furthermore, the increasing ranges of soil N_2O emissions were significantly less under the two temperature conditions than that which occurred in the different moisture conditions.

Total soil GHG emissions

The average emissions of soil GHG (*i.e.*, CO_2, CH_4 and N_2O) were 83.1, 97.2, 95.5 and 14.4 tons CO_2 *eq*. ha^{-1} in the investigated *Leymus* steppes, *Stipa* steppes, sand dune and wetland rangelands, respectively. Of the total GWPs of soil GHG emissions, CO_2 emission, CH_4 uptake/emission and N_2O emission had average contributions of 97.7%, -1.6% and 3.9% in the *Leymus* steppes and 84.7%, -1.1% and 16.4% in the *Stipa* steppes. Sand dune rangelands had similar contributions (CO_2: 97.2%; CH_4: -0.5%; N_2O: 3.3%) to the *Leymus* steppes. Significantly different from other types, the wetland rangelands had higher N_2O shares (28.4%) in total GHG emissions, where CO_2 emissions accounted for 71.5% and the CH_4 contribution could be ignored. These results indicate that the total soil GHG emissions were dominated by CO_2 emissions in the *Leymus* steppe and sand dune rangelands and that the grazing effect on the soil GHG emissions was dependent on CO_2 changes. Nevertheless, the net CO_2 exchanges should also consider CO_2 uptake by plant photosynthesis, thus the roles of N_2O and CH_4 will be increased. Compared to the ungrazed *Leymus* steppes (*i.e.*, LUG79), the *Stipa* steppes (*i.e.*, SUG79) and sand dune (*i.e.*, SDUG02), both continuously grazed sites, were significantly reduced by 22%, 38% and 48% in total GHG emissions, respectively (Table 1). Furthermore, these results also indicate that the potential of steppe soil as a CH_4 sink function can be offset by the N_2O emission, especially in over-grazed plots, due to the significant reduction of the soil CH_4 sink and enhancement of the potential of soil N_2O sources. In contrast, N_2O emissions should be considered in the wetland rangelands, as grazing has reduced GHG emissions by 47%.

Discussion

Carbon dioxide

Soil CO_2 emission rates (0–110 mg C m^{-2} h^{-1}) under different conditions at the investigated rangelands were comparable with the reported fluxes (1-198 mg C m^{-2} h^{-1}) in the Eurasia steppes (*e.g.*, Cui *et al.*, 2000; Jia *et al.*, 2007; Wang *et al.*, 2007; Chen *et al.*, 2013; Gong *et al.*, 2014) and North America prairies (*e.g.*, Bremer *et al.*, 1998; Zhou *et al.*, 2006; Casasovas *et al.*, 2012). A large number of studies have shown that soil respiration rates are closely linked to surface soil temperature and moisture (Howard and Howard, 1993). Generally, high temperature and appropriate water conditions can greatly promote soil respiration, and conversely, soil respiration can be inhibited in low temperature and low water supply conditions. In addition, the high water supply causes soil surface water saturation, which can also weaken the soil respiration rate. In our study, under simulated drought conditions, soil respiration rates from all types of grassland were weak, mainly due to a severe lack of water to reduce the microbial activity, thereby reducing the microbial regulation of isotonic respiration (Nakano *et al.*, 2008.). There was some soil respiration in the wetland under the dry condition, which was mainly caused by CO_2 generation from the underlying soil by the wetting and CO_2 diffusion of the surface soil. Rainfall can affect soil respiration by influencing the biological activity of the soil and the amount of water required for root growth (Reference). In arid ecosystems or dry-wet transition seasons, rainfall events can strongly arouse soil respiration (Jia *et al.*, 2006; Hao *et al.*, 2010). Similarly, this phenomenon also appears in our study, with the appearance of a quick CO_2 emission pulse after each rainfall event. In arid or semiarid ecosystems, soil respiration has a positive correlation with water content that is below the saturated water content of the soil. Our study shows that, except for sand dunes, the surface soil moisture content after the second rainfall at the other three types of grassland were higher than the first after the rainfall, and the soil respiration after second rainfall is higher than the first time. Under frozen conditions, soil respiration rates were very weak, since very low temperature greatly inhibited the activity of soil microbes. When the soil was thawed, the surface soil temperature was recovered and therefore promoted the recovery of microbial activity. However, the soil respiration intensity was still weak compared to that under the 20°C condition because the soil temperature and microbial activity had a significant positive correlation.

Many studies have shown that overgrazing will reduce soil respiration in semiarid grassland soils, mainly because excessive grazing greatly reduces the substrate organic carbon of soil heterotrophic respiration and attenuates the autotrophic respiration of the underground part of a plant (Wang and Fang, 2010). For management of low grazing intensity, it is generally believed that the growth of plant roots underground will be promoted to increase soil respiration (Chen *et al.*, 2011). Thus, in the grazing tests with low grazing intensities, there was no significant difference or significant increase in soil respiration compared with the ungrazed site. In this study, all four selected types of grassland including a long-forbidden grazing plot, a long-term continuous grazing rangeland and long-term grazing have significantly changed the grassland vegetation and soil characteristics. Our incubation experiments showed that grazing had little effect on the soil respiration rate under the conditions of drought and freeze- thaw in all grass types because the activity of soil microbes in the ungrazed and grazing areas was strongly inhibited. However, long-term continuous grazing significantly reduced the soil respiration rate under appropriate temperature and rainfall conditions, which indicates that soil carbon, nitrogen and microbial activity reduction caused by grazing were only manifested under appropriate hydrothermal conditions.

Methane

Negative CH_4 fluxes from soil core incubation indicate the uptake is by steppes, which are consistent with field measurements in this region (Wang *et al.*, 2005; Liu *et al.*, 2007; Chen *et al.*, 2011). The CH_4 uptake rates (3–59 μg C m^{-2}) in the investigated steppes fall into a range of different ecosystems in arid and semiarid regions (2–105 μg C m^{-2} h^{-1}), including deserts, prairies, croplands and steppes (*e.g.*, Mosier *et al.*, 1996; Galbally *et al.*, 2008; Chen *et al.*, 2011). The results of our experiments showed that typical steppes and sand dune grasslands still obviously absorbed the CH_4 in the atmosphere under the very dry condition of the surface soil. Before the start of incubation experiments, we added 15 mm of water to activate the soil microbes, which would be more likely to penetrate deeper into the soil in the arid soil cores. Thus, after more than a month of evaporation, although the surface soil moisture content has been very low (< 5% v/v) and the activity of CH_4 oxidizing bacteria was very low at the surface, the atmospheric CH_4 can also be diffused to the more humid soil and then oxidized. In the first day after the simulated rainfall, soil surface gas diffusion reduced CH_4 diffusion from atmosphere to the soil due to a large amount of water input, so there appeared to be short and weak CH_4 absorption, and CH_4 absorption gradually increased followed by the surface soil water evaporation. These change characteristics were more obvious in typical steppes than other types. Because the soil texture is loose and the water permeability is better, the input of water was transferred to the deep soil over time, and the surface soil did not accumulate a large amount of water to affect the soil diffusion. Thus the response of the CH_4 absorption to the rainfall in the sand dune rangeland may not be sensitive. For soil cores collected from wetlands, these soils have been in place for nearly a year and the surface soil had been very dry when we started measurements. However, the soils in the lower part (20-40 cm) remain moist. At this time, the wetland soil can be considered a natural grassland soil after the river water retreated. The CH_4 flux was almost zero, meaning there was a balance between soil CH_4 emission and absorption. Our simulated 20 mm rainfall looks as though it is not enough to change this situation. The alteration of CH_4 uptake in typical steppes and sand dune rangeland was also obvious under freezing and thawing conditions due to the large alteration of temperature (-10^{o} C and 10^{o} C). This mainly accounted for the change of microbial activity in the surface soil, which influenced CH_4 absorption (Chen *et al.*, 2011). Generally, when the soil temperature is lower than -5^{o} C, the activity of soil CH_4 oxidizing bacterial will be greatly reduced (Smith *et al.*, 2000).

The order of CH_4 uptake rates in the different types of grassland was *Leymus chinensis* steppe ≥ *Stipa Grandis* steppe > Sand dune > Wetland. For the typical steppe, the results showed that the grazing significantly inhibited the CH_4 uptake by the *Leymus chinensis* steppe, which was consistent with the field observation (Liu *et al.*, 2007; Chen *et al.*, 2011). Severe grazing significantly reduced the annual CH_4 uptake, mainly due to the remarkable decrease in surface soil gas permeability. Although we did not detect the gas permeability of the soil cores during the incubation periods, the soil bulk density (0.99 g cm^{-3}) of the long-term grazing in the *Leymus chinensis* steppe was higher than that of the forbidden grazing area (0.87 g cm^{-3}), which may also suggest that the gas permeability of the grazing rangeland was low. However, the soil bulk density (1.24 g cm^{-3}) was also significantly higher than that of the forbidden grazing plot (1.06 g cm^{-3}), but we did not detect the significant effect of grazing on the *Stipa grandis* steppe. This suggests that in addition to the gas permeability reduction, the reducing soil microbial activity by vegetation changes and induced environmental factors (*e.g.*, soil temperature and moisture) was also important for the grazing reduction effect on CH_4 uptake by the *Stipa grandis* steppe.

The soil CH_4 sink function of the two types of typical steppes was significantly higher than that in the sand dune and wetland rangelands ($p < 0.05$). Most of sand dune areas used for grazing are formed by overgrazing or reclamation in a typical grassland. The grass coverage on the sand dune rangeland is very sparse, and the microbial activity in the soil was significantly lower than that of the typical steppes. The sand dune rangelands can be regarded as typical steppes under extreme overgrazing conditions; therefore, CH_4 uptake capacity of sand dune rangelands were significantly lower than that of the typical steppes (Chen *et al*., 2013). For the seven years of forbidden sand dune rangelands, although vegetation has been restored, our incubation experiment indicates that the soil CH_4 sink capacity has not been improved. This result may be different from field observations in view of the effects of vegetation alone on the activity of soil microorganisms or the changing environmental factors (*e.g*., soil temperature and moisture). However, for degraded grasslands or sand dunes under excessive grazing practices, there is still large uncertainty as to whether the CH_4 sink capacity could be restored to natural grassland or how long it will take to recover. Mosier *et al*. (1997) found that the soil CH_4 oxidation rate was only one-third that of the natural grassland when the arable land was converted into grassland for 50 years. Smith *et al*. (2000) considered the CH_4 oxidation rate when the original grass was reclaimed or fertilized and found that the recovery was slow and takes more than 100 years to recover to the CH_4 absorption rate before interference.

In addition, our incubation experiment suggests that the seasonal river floodplain wetland did not function as a CH_4 sink after river water retreat, and grazing had no significant effect on the weak absorption or discharge of CH_4. This means that with the depletion or reduction of grassland water resources, the wetland soil was the only weak source or sink and can be ignored in the regional CH_4 budget. For the flood plain, when the river flows over the surface of the wetlands, the wetlands are a strong source of atmospheric CH_4. In addition, Hirota *et al*. (2005) have reported that grazing can reduce CH_4 emissions from alpine wetlands in the short term by affecting the ventilation of aquatic plants. Although some CH_4 emission observations were carried out in the Inner Mongolia floodplain wetlands, most of them were on grazing wetlands. For grazed and ungrazed comparisons, there was little research to understand the impact of grazing on CH_4 emissions from floodplains, which would also help to estimate regional CH_4 budget.

Nitrous oxide

The N_2O emission rates from the soil core incubation of steppes (0-132 μg N m^{-2} h^{-1}) and sand dunes (0-49 μg N m^{-2} h^{-1}) fall in the range of field measurements (Mosier *et al.*, 1996; Wang *et al.*, 2005; Wolf *et al.*, 2010). Many studies have shown that soil N_2O emissions are positively correlated with soil moisture (Mosier *et al.*, 1996; Steffens *et al.*, 2008). In our study, the N_2O emissions of the three types of grassland under drought conditions were weak. Insufficient water supply would reduce the activity of microorganisms during nitrification and denitrification, resulting in a decrease in soil N_2O production. In general, heavy rainfall during the wet and dry transition process will stimulate the N_2O pulses, which are reflected in many field observations and indoor experiments (*e.g.*, Teepe *et al.*, 2004; Holst *et al.*, 2008; Yao *et al.*, 2010). We found that, in addition to the *Leymus chinensis* steppe, there was a N_2O emission pulse in the other three types of grassland in the short term after rainfall, especially after continuous rainfall. The surface soil moisture content will be greatly increased, which greatly improved the nitrification and denitrification process by providing hydrothermal conditions to increase the amount of soil N_2O production. The N_2O emissions were greatly reduced during the freeze-thaw conditions compared to the field snowmelt period. This may due to the large consumption of soil substrate in N_2O production during two rainfall simulations and to relatively low temperatures that inhibit microbial activity. Because the texture of the surface soil was relatively loose at the typical steppes and sand dunes, the rainfall water was easy to penetrate the deep soil; therefore, the surface soil moisture was easily evaporated to reduce the water conditions of soil N_2O production. The surface soil density of the *Stipa grandis* steppe was 0.2 g cm^{-3} higher than that of the *Leymus chinensis* steppe. Therefore, the soil moisture in the topsoil was easy to accumulate after rainfall and formed a high humidity condition, which was beneficial to the production of soil N_2O. The wetland soils in the flood plains were wetter at the deeper depth. After wetting, the wetland soil had a strong water retention capacity and provided a high humidity condition. Moreover, wetland soil can provide more organic substrate, so the wetland soil N_2O emissions should be much higher than the other three types.

The order of soil N_2O emission rate of different types of grassland was Wetland > *Stipa Grandis* steppe > Sand dune > *Leymus chinensis* steppe. In general, long-term grazing affects the bulk density and porosity of the surface soil, which could easily form an anaerobic zone to increase the N_2O

emission potentials under rainfall conditions (Steffens *et al.*, 2008). In addition, sheep feces in heavily grazed plots were obviously greater than in other soil cores. Thus, through leaching with rainfall, animal feces nutrients may provide more reaction substrates for soil denitrification bacteria. However, in the four types of grasslands surveyed, we did not find that grazing had a significant effect on the soil N_2O emissions. Soil N_2O emission has high spatial heterogeneity, and the variation coefficient of emissions between different soil cores were as high as 80%-290% (Fig. 3), especially in the wetland. In the undisturbed soil we collected, the surface area of the soil column was only 0.02 m^2, and the height of gas collection in soil cores was 10 cm. This may result in large differences in hydrothermal conditions or soil organic carbon and nitrogen content in the microenvironment and thus affect the soil N_2O production. Therefore, it is important to improve the spatial distribution of N_2O emission in different grassland types and to increase the number of spatial samples, which plays an important role in the accurate detection of the impact of grazing management on the N_2O emission in grasslands.

Conclusions

All steppe and sand dune rangelands were CH_4 sinks (-3~-59 μg C m^{-2} h^{-1}), while wetland rangelands were shifted between weak sinks or sources (-13~13 μg C m^{-2} h^{-1}) under various conditions. The order of CH_4 uptake rate of different types of grassland is *Leymus chinensis* Steppe ≥ *Stipa grandis* steppe > Sand dune > Wetland. Wetland showed significantly higher N_2O emission potential (range: 0-343 μg N m^{-2} h^{-1}) than the steppes (range: 0-132 μg N m^{-2} h^{-1}) and sand dune (range: 0-49 μg N m^{-2} h^{-1}). The order of soil CO_2 emission in the four types of forbidden grazing sites was Wetland > Sand dune > steppes, but there was no statistically significant difference considering both grazed and ungrazed sites. Grazing significantly reduced the soil respiration rate of different types of soils and only significantly inhibited the absorption of CH_4 in *Leymus chinensis* steppe; we did not find a significant effect on soil N_2O emissions. Compared to the ungrazed *Leymus* steppes, *Stipa* steppes, sand dune and wetland, continuously grazed sites were significantly reduced by 22%, 38%, 48% and 47% in total GHG emissions, respectively. Our study found the potential of the steppe soil CH_4 sink function can be offset by N_2O emission, especially during overgrazing practices, which indicates a net GHG budget might mainly depend on the net CO_2 exchange. In addition, N_2O emissions in wetland rangelands during dry periods should be considered in the regional N_2O budget due to the higher emission potential. However, it is important to comprehensively consider results from both field observation and incubation experiments to evaluate the grazing effect on ecosystem GHG emissions because soil properties and environmental factors could be changed by vegetation growth in the field.

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References

Bremer, D.J., Ham, J.M., Owensby, C.E., Knapp, A.K., 1998. Responses of soil respiration to clipping and grazing in a tallgrass prairie. Journal of Environmental Quality. 27, 1539–1548.

Casasovas, N.G., Matamalar, A.R., Cook, D., Gonzalez-Meler, M.A., 2012. Net ecosystem exchange modifies the relationship between the autotrophic and heterotrophic components of soil respiration with abiotic factors in prairie grasslands. Global Change Biology. 18, 2532–2545.

Chen W., Wolf B., Zheng X., Yao Z., Brüggemann N., Butterbach-Bahl K., Liu C., Han S., Han X., 2011. Annual methane uptake by temperate continental steppes under a semiarid climate as regulated by stocking rates, plant growth and soil-air permeability. Global Change Biol. 17, 2803–2816.

Chen, W., Wolf, B., Zheng, X., Yao, Z., Brüggemann, N., Butterbach-Bahl, K., Liu, C., Han, S., Han, X., 2011. Annual methane uptake by temperate continental steppes under a semiarid climate as regulated by stocking rates, plant growth and soil-air permeability. Global Change Biology. 17, 2803–2816.

Chen, W., Zheng, X., Chen, Q., Wolf, B., Butterbach-Bahl, K., Brüggemann, N., Lin, S., 2013. Effects of increasing precipitation and nitrogen deposition on CH_4 and N_2O fluxes and ecosystem respiration in a degraded steppe in Inner Mongolia, China. Geoderma. 192, 335-340.

Cui, X.Y., Chen, S.Q., Chen, Z.Z., 2000. CO_2 release from typical Stipa grandis grassland soil. Chinese Journal of Applied Ecology. 11, 390–394 (in Chinese).

Galbally, L.E., Kirstine, W.V., Meyer, C.P., Wang, Y.P., 2008. Soil–atmosphere trace gas exchange in semiarid and arid zones. Journal of Environmental Quality. 37, 599–607.

Gao, Y., Chen, Q., Shan, L., Giese, M., Brueck, H., 2011. Resource manipulation effects on net primary production, biomass allocation and rain-use efficiency of two semiarid grassland sites in Inner Mongolia, China. Physiological Ecology. 165, 855–864.

Gong, J., Wang Y., Liu, M., Huang, Y., Yan, X., Zhang, Z., 2014. Effects of land use on soil respiration in the temperate steppe of Inner Mongolia, China. Soil and Tillage Research. 144, 20–31.

Hao, Y., Wang, Y., Mei, X., Cui, X., Zhou, X., Huang, X., 2010. The sensitivity of temperate steppe CO_2 exchange to the quantity and timing of natural interannual rainfall. Ecological Informatics. 5, 222–228.

Hirota M., Tang Y.H., Hu Q.W., Kato T., Hirata S., Mo W.H., Cao G.M., Mariko S., 2005. The potential importance of grazing to the fluxes of carbon dioxide and methane in an alpine wetland on the Qinghai-Tibetan Plateau. Atmospheric Environment. 39, 5255-5259.

Holst J., Liu C., Yao Z., Brueggemann N, Zheng X, Giese M, Butterbach-Bahl K., 2008. Fluxes of nitrous oxide, methane and carbon dioxide during freezing–thawing cycles in an Inner Mongolian steppe. Plant and Soil. 308, 105-117.

Howard, D.M., Howard, P.J.A., 1993. Relationships between CO_2 evolution, moisture content and temperature for a range of soil types. Soil Biology and Biochemistry. 25, 1537-1546.

IPCC, 2007. Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, UK and New York, NY, USA, 539–542.

Jia B., Zhou G., Wang Y., Wang F., Wang X., 2006. Effect of temperature and soil water-content on soil respiration of grazed and ungrazed Leymus Chinensis steppes, Inner Mongolia. Journal of Arid Environment. 67, 60-76.

Jia, B.R., Zhou, G.S., Yuan, W.P., 2007. Modeling and coupling of soil respiration and soil water content in fenced Leymus chinensis steppe, Inner Mongolia. Ecological Modelling. 201, 157–162.

Kang L., Han X.G., Zhang Z.B.., Sun O.J., 2007. Grassland ecosystems in China: review of current knowledge and research advancement. Philosophical Transactions of the Royal Society B:Biological Sciences. 362, 997-1008.

Kroon, P.S., Hensen, A., Bulk, W.C.M., Jongejan, P.A.C., Vermeulen, A.T., 2008. The importance of reducing the systematic error due to non-linearity in N_2O flux measurements by static chambers. Nutrient Cycling in Agroecosystems. 82, 175–186.

Liu C., Holst J., Brüggemann N., Butterbach-Bahl K., Yao Z., Han S., Han X., Zheng X., 2008. Effects of water input on nitrous oxide, methane and carbon dioxide fluxes in an Inner Mongolian steppe. Advances in Atmospheric Sciences. 25, 748-756.

Mosier A.R., Delgado J.A., 1997. Methane and nitrous oxide fluxes in grasslands in western Puerto Rico. Chemosphere. 35, 2059–2082.

Mosier, A.R., Parton, W.J., Valentine, D.W., Ojima, D.S, Schimel, D.S., Delgado, J.A., 1996. CH_4 and N_2O fluxes in the Colorado shortgrass steppe: 1. Impact of landscape and nitrogen addition. Global Biogeochemical Cycles. 10, 387–399.

Nakano T., Nemoto M., Shinoda M., 2008. Environmental controls on photosynthetic production and ecosystem respiration in semi-arid grasslands of Mongolia. Agricultural and Forest Meteorology. 148, 1456-1466.

Smith K.A., Ball T., Conen F., Dobbie K.E., Massheder J., Rey A., 2003. Exchange of greenhouse gases between soil and atmosphere: Interactions of soil physical factors and biological processes. Eur J Soil Sci. 54, 779–791.

Smith K.A., Dobbie K.E., Ball B.C., Bakken L.R., Sitaula B.K., Hansen S., Brumme R., Borken W., Christensen S., Priemé A., Fowler D., Macdonald J.A., Skiba U., Klemedtsson L., Kasimir-Klemedtsson A., Degórska A., Orlanski P., 2000. Oxidation of atmospheric methane in Northern European soils, comparison with other ecosystems, and uncertainties in the global terrestrial sink. Global Change Biology. 6, 791–803.

Soussana, J.F., Allard, V., Pilegaard, K., et al., 2007. Full accounting of the greenhouse gas (CO_2, N_2O, CH_4) budget of nine European grassland sites. Agriculture, Ecosystems and Environment. 124, 121-134.

Steffens, M., Kölbl, A., Totsche, K.U., Kögel-Knabner, I., 2008. Grazing effects on soil chemical and physical properties in a semiarid steppe of Inner Mongolia (P.R. China). Geoderma. 143, 63–72.

Steinfeld, H., P. Gerber, T. Wassenaar, V. Castel, M. Rosales, and C. de Haan., 2006. Livestock's long shadow, Environmental Issue and Options, Food and Agriculture Organization of the United Nations, Rome. (http://www.virtualcentre.org/n/library/key_pub/longshad/A0701E00.pdf)

Su Y., Li Y., Cui J., Zhao W., 2005. Influences of continuous grazing and livestock exclusion on soil properties in a degraded sandy grassland, Inner Mongolia, northern China. CATENA. 59, 267-278.

Sutie, J.M., Reynolds, S.G., Batello, C. (Eds.), 2005. Grassland of the world. : Plant Production and Protection Series, No. 34. Food and Agriculture Organization of the United Nations, Rome. 1 pp.

Teepe, R., Vor A., Beese F., Ludwig B., 2004. Emissions of N_2O from soils during cycles of freezing and thawing and the effects of soil water, texture and duration of freezing. Eur. J. Soil Sci. 55, 357–365.

Tong, C., Wu, J., Yong, S., Yang, J., Yong, W., 2004. A landscape-scale assessment of steppe degradation in the Xilin River Basin, Inner Mongolia, China. Journal of Arid Environment. 59, 133-149.

Wang, W., Fang, J., 2010. Soil respiration and human effects on global grasslands. Global and Planetary Change. 67, 20–28.

Wang, Y., Xue, M., Zheng, X., Ji, B., Du, R., Wang, Y., 2005. Effects of environmental factors on N_2O emission from and CH_4 uptake by the typical grasslands in the Inner Mongolia. Chemosphere. 58, 205–215.

Wolf B., Zheng X., Brüggemann N., Chen W., Dannenmann M., Han X., Sutton M.A., Wu H., Yao Z., Butterbach-Bahl K., 2010. Grazing-induced reduction of natural nitrous oxide release from continental steppe. Nature. 464, 881–884.

Zheng, X., Mei, B., Wang, Y., Xie, B., Wang, Y., Dong, H., Xu, H., Chen, X., Cai, Z., Yue, J., Su, F., Zou, J., 2008. Quantification of N_2O fluxes from soil-plant systems may be biased by the applied gas chromatograph methodology. Plant and Soil. 311, 211–234.

Zhou, X.H., Sherry, R.A., An, Y.,Wallace, L.L., Luo, Y.Q., 2006. Main and interactive effects of warming, clipping, and doubled precipitation on soil CO_2 efflux in a grassland ecosystem. Global Biogeochemical Cycles. 20, GB1003. doi:10.1029/2005GB002526.

List of Tables

Table 1 Grassland managements and soil properties at paired ungrazed and grazed grasslands in Inner Mongolia

Grassland type	Site code	Location	Management	Stocking rate (sheep ha^{-1} yr^{-1})	Bulk density (g cm^{-3})	pH	Total C (mg g^{-1})	Total N (mg g^{-1})	CaCO$_3$ (mg g^{-1})	Sand (%)	Silt (%)	Clay (%)
Stipa grandis steppe	SUG79	N 43°32′32″ E 116°40′15″	Ungrazed since 1979	0	1.06^{cd}	7.5^{a}	1.54^{d}	0.22^{b}	0.1	60.3	31.3	8.4
	SCG	N 43°32′22″ E 116°40′01″	Continuously grazed	2.34	1.24^{b}	6.9^{b}	1.22^{e}	0.17^{c}	0.1	62.4	28.1	9.5
Leymus chinensis steppe	LUG79	N 43°33′10″ E 116°40′27″	Ungrazed since 1979	0	0.87^{e}	6.9^{b}	1.63^{d}	0.19^{bc}	0	73.5	17.3	9.2
	LCG	N 43°33′05″ E 116°40′54″	Continuously grazed	2.05	1.00^{cd}	6.7^{b}	0.51^{f}	0.07^{d}	0	86.7	9.5	3.8
Sand dune	SDUG02	N 43°37′57″ E 116°42′04″	Ungrazed since 2002	0	0.98^{d}	7.4^{a}	2.20^{b}	0.33^{a}	0.1	80.5	14.2	5.3
	SDCG	N 43°38′01″ E 116°41′47″	Continuously grazed	1.56	1.45^{a}	6.7^{b}	2.14^{bc}	0.25^{ab}	0.2	92.4	4.6	3.0
Wetland	WUG89	N 43°37′39″ E 116°41′13″	Ungrazed since 1989	0	1.11^{c}	6.7^{b}	2.57^{a}	0.24^{b}	0.9	45.7	49.4	4.9
	WCG	N 43°37′09″ E 116°41′12″	Continuously grazed	1.32	1.14^{c}	6.7^{b}	1.90^{cd}	0.16^{c}	0.1	51.6	39.0	9.4

The lower cases on the upper-right corner of each data indicate significant ($p < 0.05$) differences among each column.

Table 2 Soil CO_2 (mg C m^{-2} h^{-1}), CH_4 (μg C m^{-2} h^{-1}) and N_2O (μg N m^{-2} h^{-1}) fluxes investigated grasslands during simulated dry, dry-wet, intense rainfall and freeze-thaw periods.

Site	Dry period			Dry-wet period			Intense rainfall period			Freeze-thaw period		
code	CO_2	CH_4	N_2O	CO_2	CH_4	N_2O	CO_2	CH_4	N_2O	CO_2	CH_4	N_2O
SUG79	7 ± 2^{bc}	-48 ± 2^{b}	2 ± 1^{b}	33 ± 15^{ab}	-37 ± 7^{bc}	33 ± 11^{c}	52 ± 20^{a}	-33 ± 6^{bc}	102 ± 31^{b}	13 ± 13^{a}	-23 ± 12^{b}	8 ± 5^{c}
SCG	2 ± 1^{d}	-46 ± 5^{ab}	1±1	24 ± 8^{c}	-40 ± 10^{ab}	21 ± 8^{c}	33 ± 5^{b}	-33 ± 7^{bc}	72 ± 18^{c}	7 ± 8^{b}	-17 ± 12^{b}	8 ± 7^{c}
LUG79	7 ± 1^{c}	-52 ± 1^{a}	0 ± 0^{b}	35 ± 12^{ab}	-53 ± 4^{a}	5 ± 2^{d}	45 ± 23^{a}	-49 ± 8^{a}	5 ± 1^{e}	12 ± 11^{a}	-44 ± 9^{a}	2 ± 1^{c}
LCG	8 ± 1^{bc}	-41 ± 3^{c}	1 ± 1^{b}	25 ± 11^{c}	-40 ± 3^{c}	12 ± 11^{cd}	35 ± 20^{b}	-34 ± 8^{c}	21 ± 31^{d}	8 ± 7^{b}	-21 ± 7^{b}	3 ± 2^{c}
SDUG02	1 ± 1^{d}	-9 ± 2^{d}	2 ± 0^{b}	59 ± 29^{a}	-10 ± 7^{d}	21 ± 15^{c}	54±19a	-15 ± 5^{d}	16 ± 2^{d}	12 ± 12^{a}	-15 ± 11^{bc}	2 ± 2^{c}
SDCG	0 ± 0^{e}	-12 ± 3^{d}	1 ± 1^{b}	31 ± 18^{b}	-14 ± 6^{d}	9 ± 4^{d}	25 ± 9^{b}	-17 ± 4^{d}	5 ± 1^{e}	9 ± 8^{b}	-9 ± 8^{c}	2 ± 2^{c}
WUG89	34 ± 1^{a}	-1 ± 1^{e}	16 ± 1^{a}	44 ± 13^{ab}	4 ± 5^{e}	97 ± 20^{a}	58±11a	4 ± 7^{f}	288 ± 64^{a}	18 ± 8^{a}	3 ± 3^{d}	51 ± 22^{a}
WCG	10 ± 1^{b}	-2 ± 1^{e}	14 ± 1^{a}	22 ± 7^{d}	-2 ± 3^{e}	70 ± 13^{b}	31 ± 8^{b}	-5 ± 5^{e}	130 ± 37^{b}	14 ± 7^{ab}	1 ± 2^{d}	18 ± 8^{b}

The lower cases on the upper-right corner of each data indicate significant ($p < 0.05$) differences among each column.

List of Figures

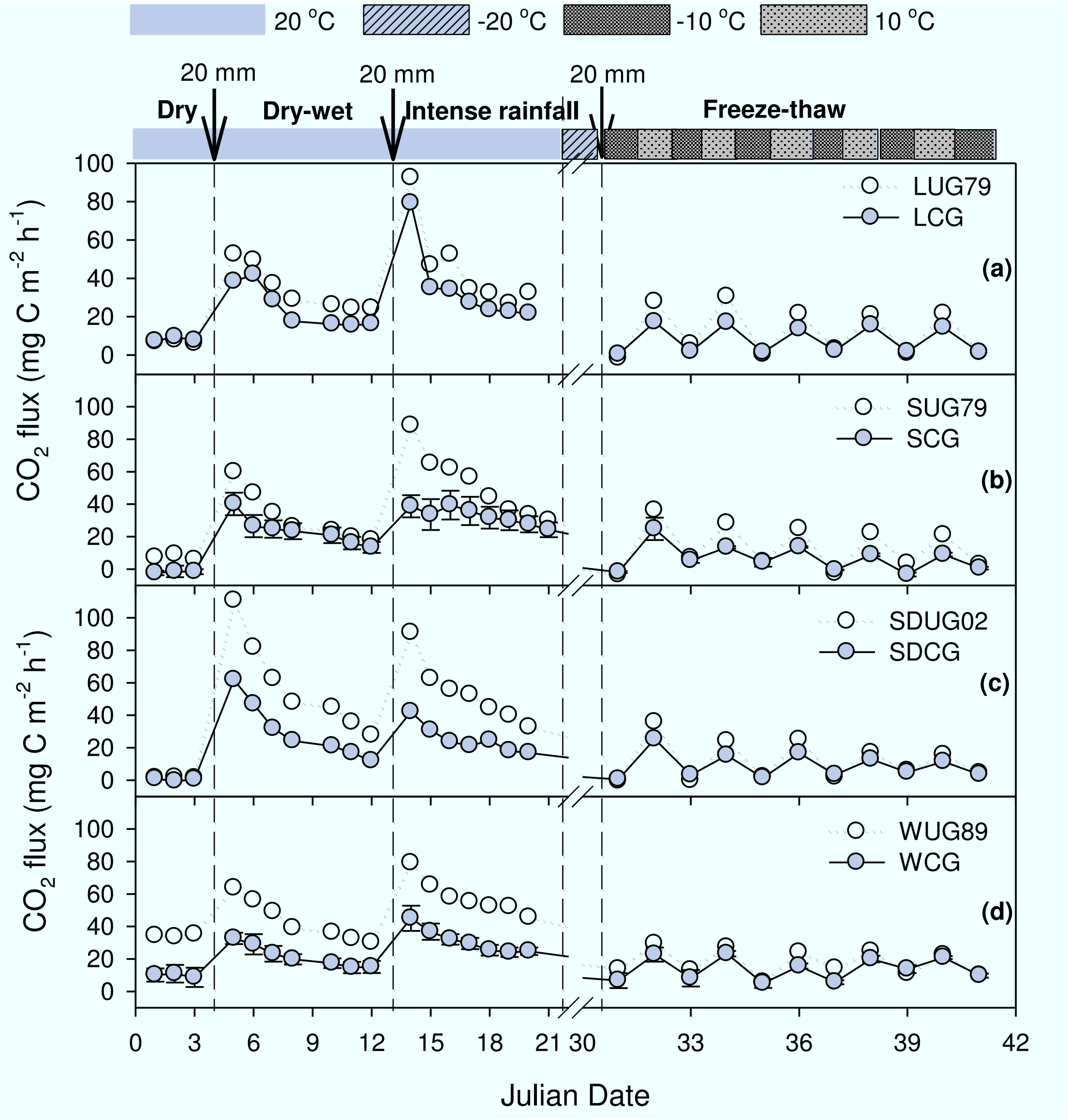

Fig.1 Variations of soil CO_2 fluxes from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) during simulated dry, dry-wet, intense rainfall and freeze-thaw contitions. Black down arrows indicate time to add water as simualted rainfall. Different corlors and patterns of rectangles represent different incubation temperatures. The given data were the means of four replicates and standard deviations.

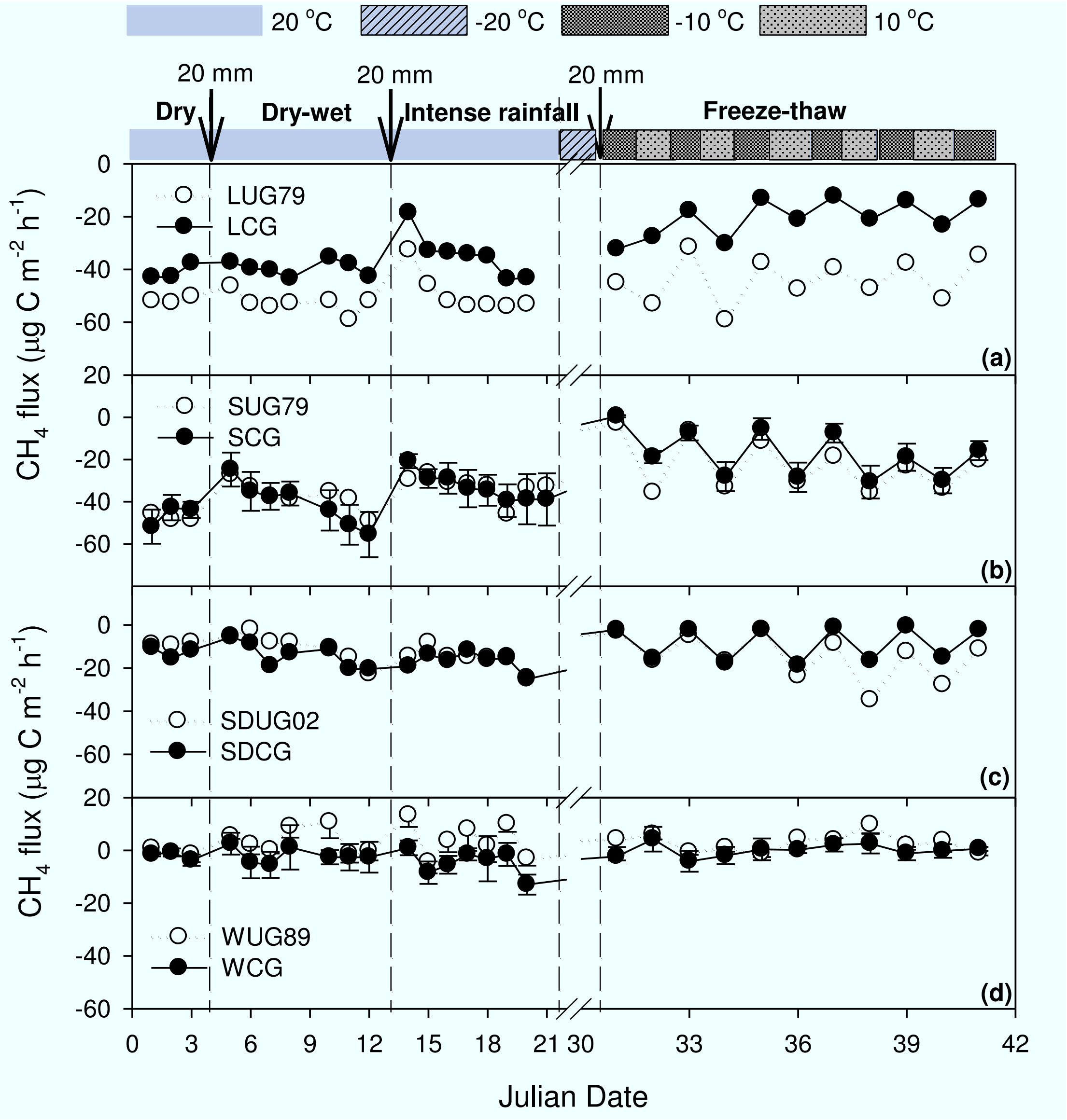

Fig.2 Variations of soil CH_4 fluxes from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) during simulated dry, dry-wet, intense rainfall and freeze-thaw contitions. Black down arrows indicate time to add water as simualted rainfall. Different corlors and patterns of rectangles represent different incubation temperatures. The given data were the means of four replicates and standard deviations.

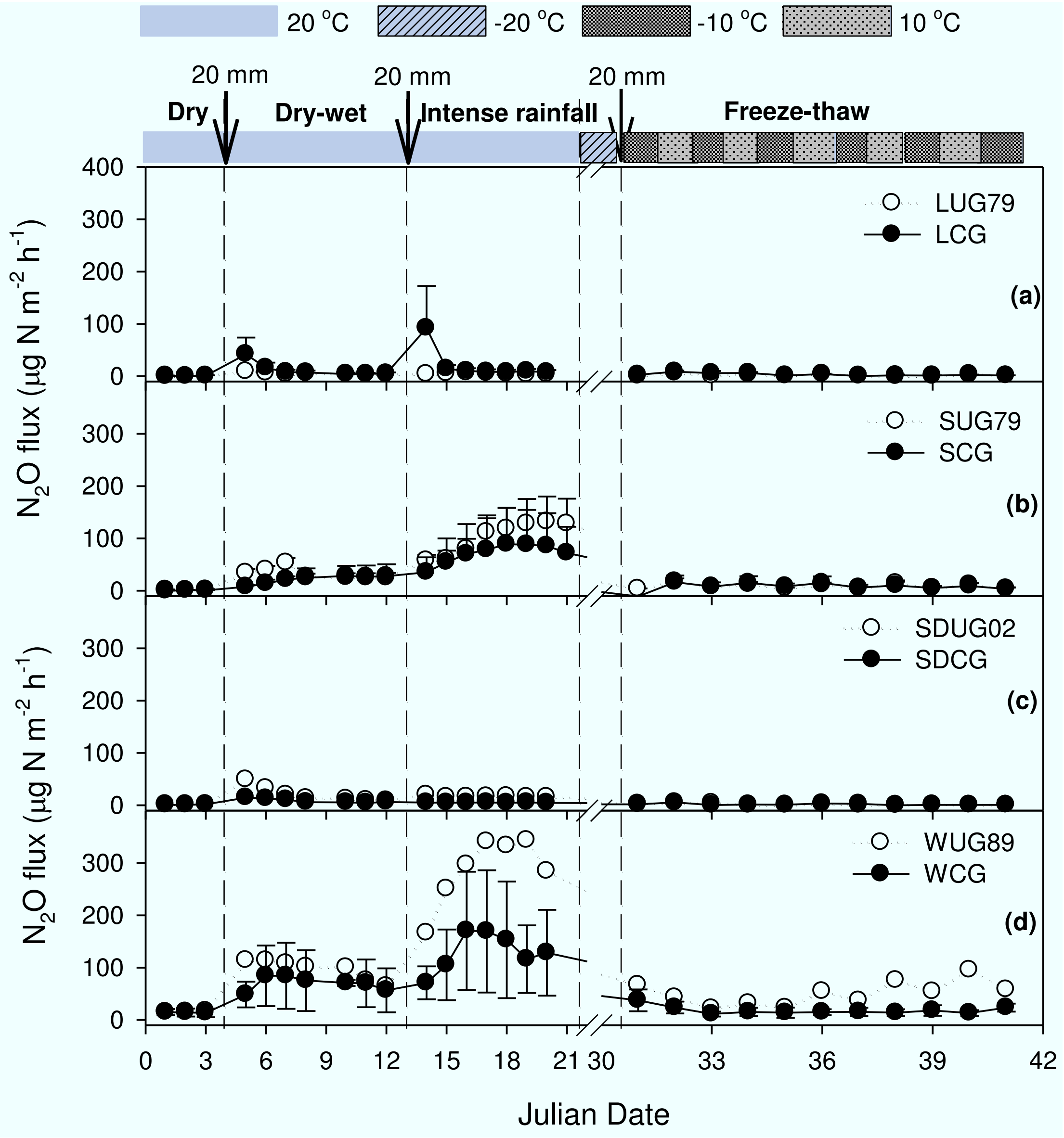

Fig.3 Variations of soil N_2O fluxes from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) during simulated dry, dry-wet, intense rainfall and freeze-thaw contitions. Black down arrows indicate time to add water as simualted rainfall. Different corlors and patterns of rectangles represent different incubation temperatures. The given data were the means of four replicates and standard deviations.

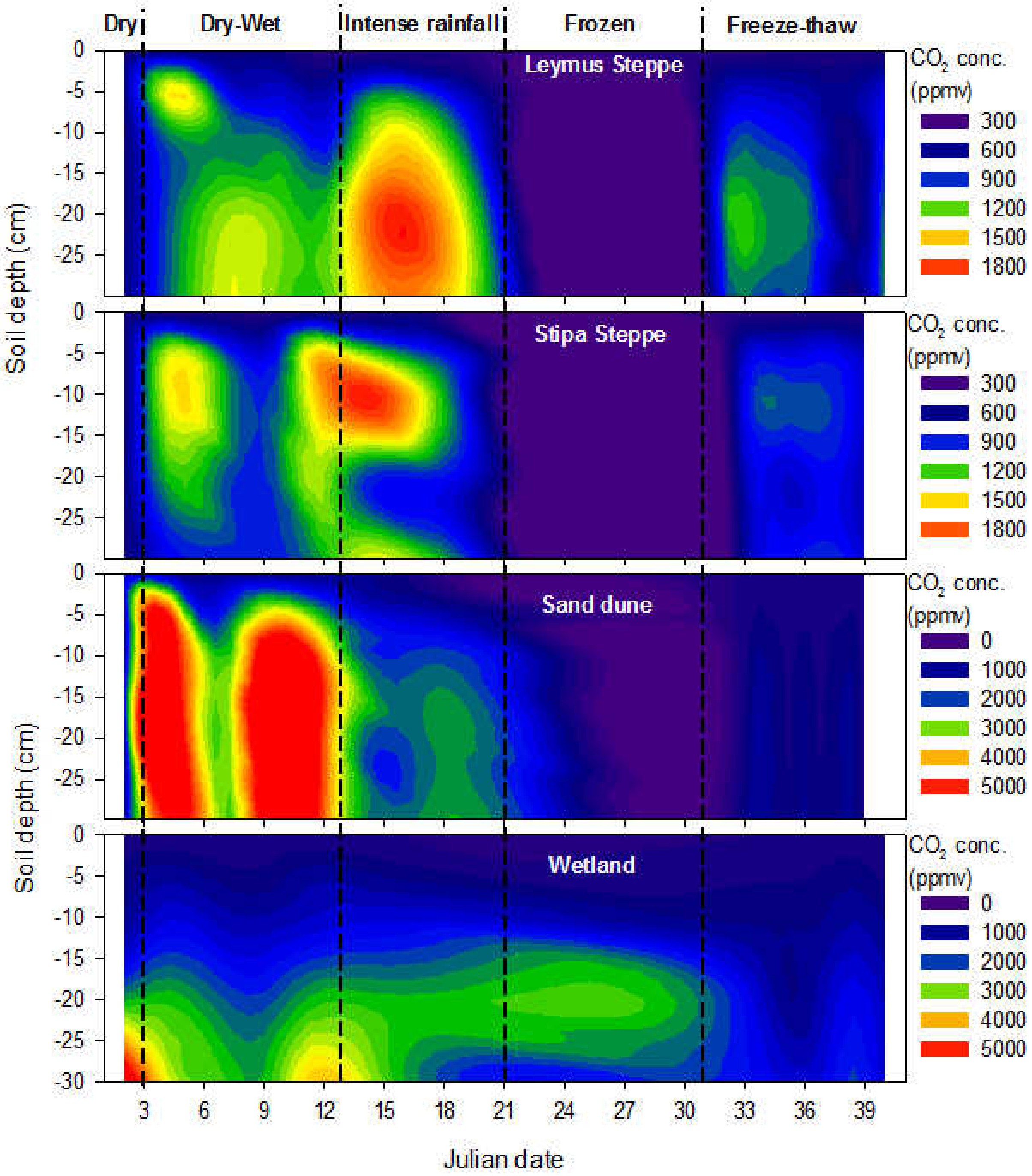

Fig.4 Variations of soil CO_2 concentrations at different depths from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) during simulated dry (1-3 days), dry-wet (4-13days), intense rainfall (13-21 days) and freeze-thaw periods (31-40 days). Frozen period reprsents soil cores were frozen in -20 °C for a month.

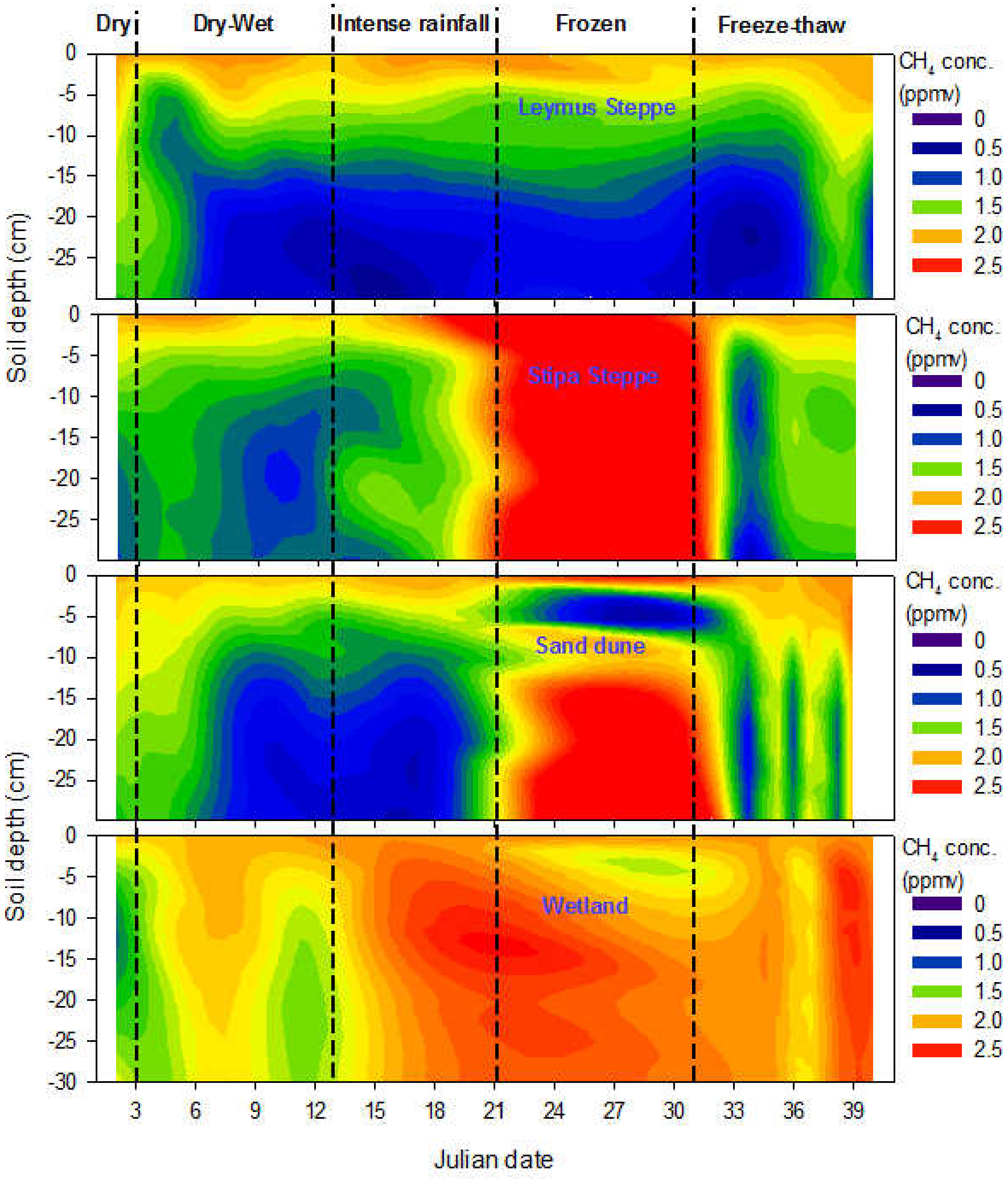

Fig.5 Variations of soil CH_4 concentrations at different depths from investigated grasslands (Leymus steppe, Stipa steppe, Sand dune and Wetland) during simulated dry (1-3 days), dry-wet (4-13days), intense rainfall (13-21 days) and freeze-thaw periods (31-40 days). Frozen period reprsents soil cores were frozen in -20 °C for a month.

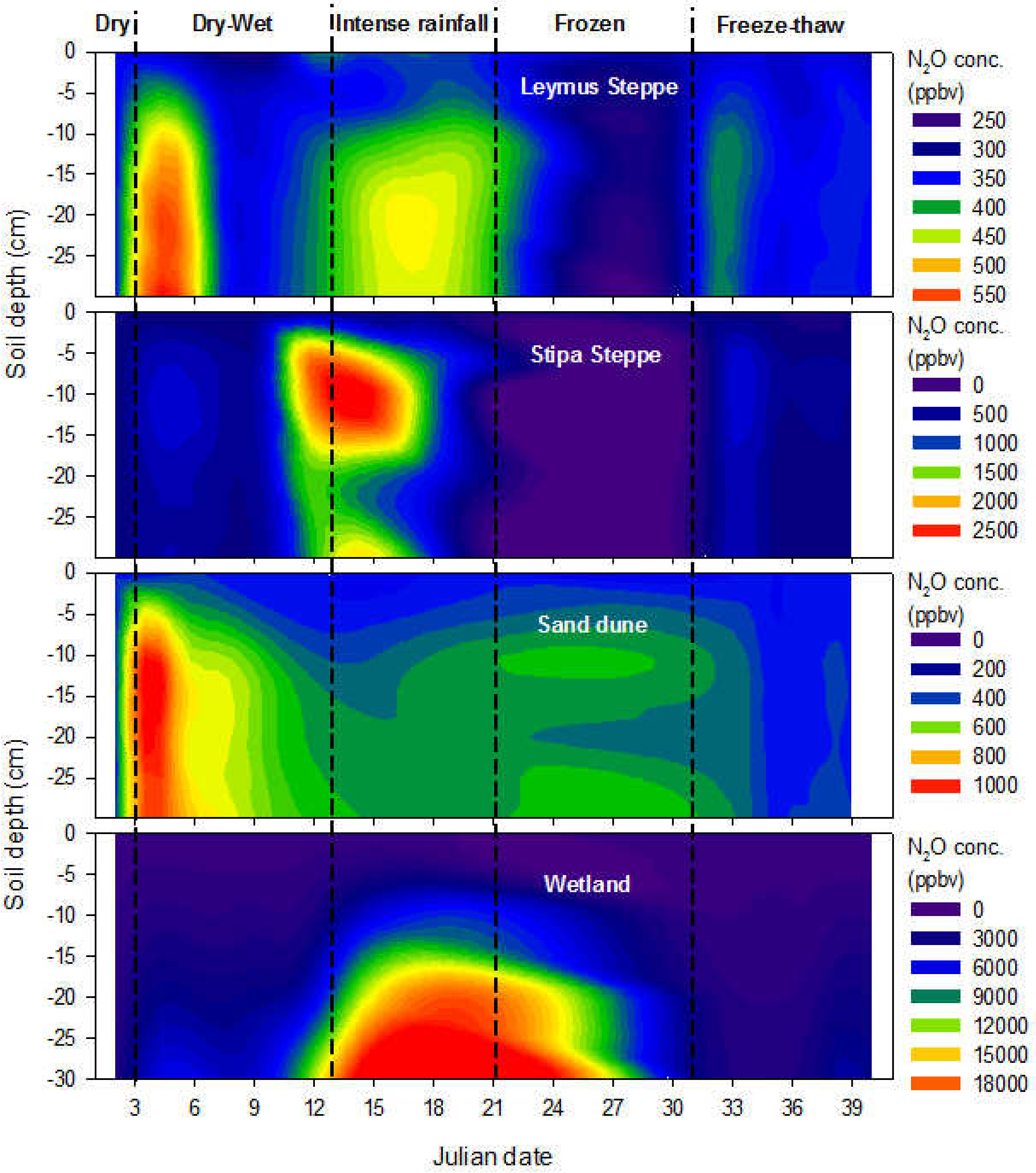

Fig.6 Variations of soil N_2O concentrations at different depths from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) during simulated dry (1-3 days), dry-wet (4-13days), intense rainfall (13-21 days) and freeze-thaw periods (31-40 days). Frozen period reprsents soil cores were frozen in -20 °C for a month.

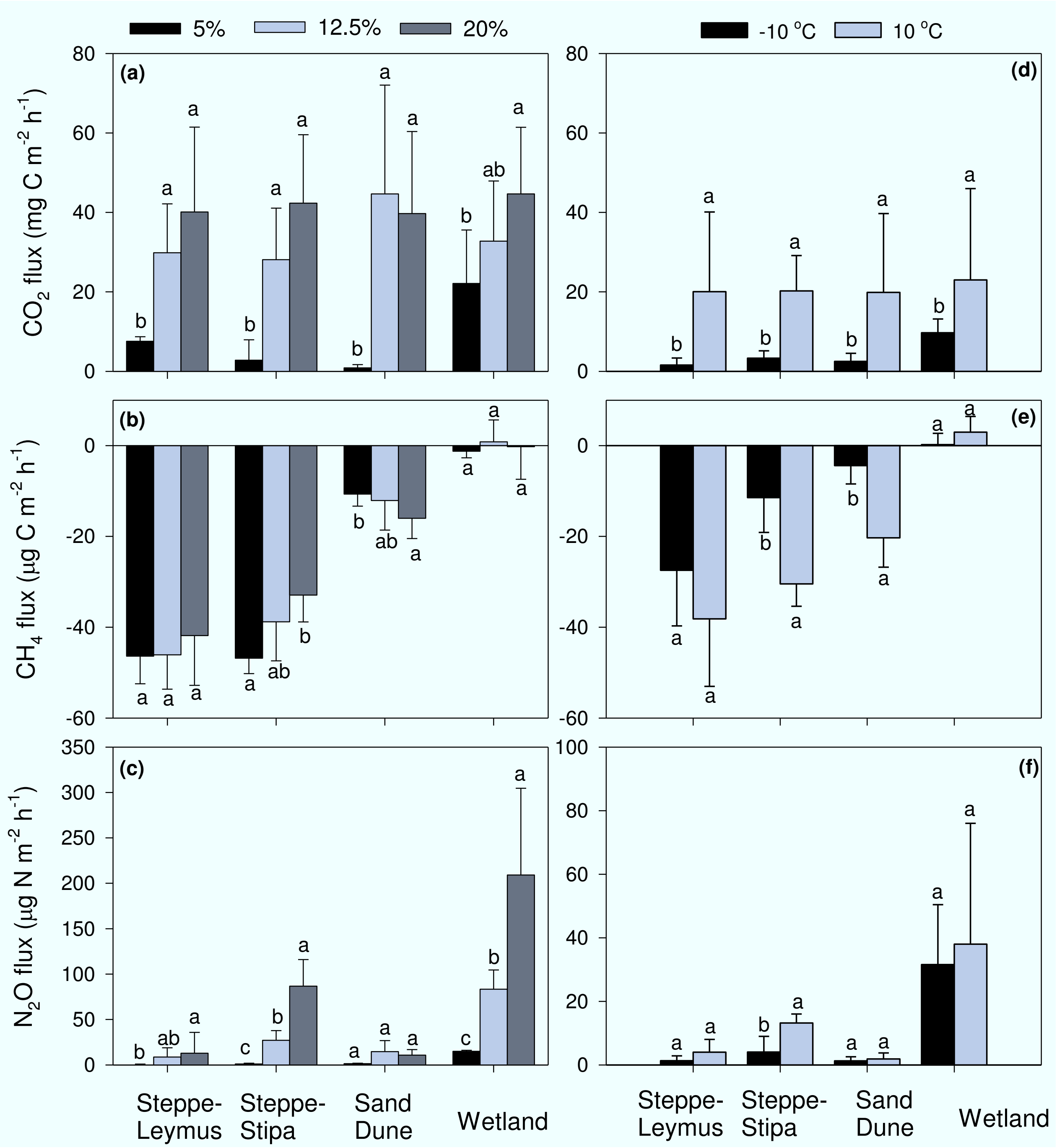

Fig.7 Soil CO_2, CH_4 and N_2O fluxes from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) in different air tempreature and soil moisture conditions. Two temperatures represent conditions of freeze period and thaw period, respectively. Three values of soil moisture (5%, 12.5% and 20%) averaged from dry period, dry-wet simulation period, and intense rainfall and freeze-thaw periods, respectively. These values of soil moistures were rounded to nearest value. Temporal-spatial standard deviations of four soil core replicates were calcuated for each data. The lowercases above or below the bars indicate the signficant differences between two air temperatures

and among three soil moistures.

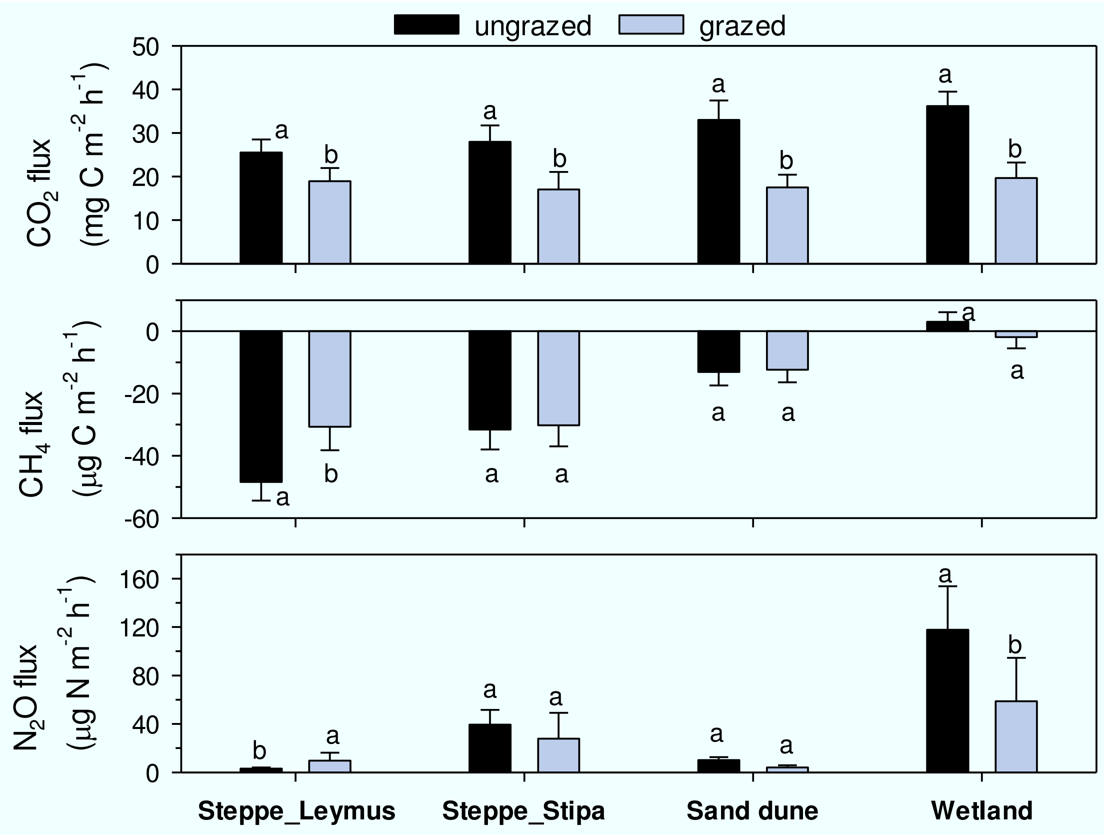

Fig.8 The average of soil CO_2, CH_4 and N_2O fluxes from investigated grasslands (Leymus steppe, Stipa steppe, Sand due and Wetland) during whole incubation peirods. Spatial standard deviations of four soil core replicateswere calcuated for each data. The lowercases above or below the bars indicate the signficant differences between ungrazed and grazed plots.