

Dear Editor,

Thank you for allowing us to submit a revised version of our manuscript. We greatly appreciate the detailed and constructive comments of you and two reviewers which helped us to improve the manuscript. We addressed all comments as attached below (highlighted in red for reviewer 1's comments and in blue for reviewer 2's comments) and hope that we adequately solved the requests.

With kind regards,

Xiaojuan Feng

(We attached a marked version of the manuscript and the supplement with changes highlighted at the end of this pdf.)

Response to Editor

Comment:

(1) Your manuscript has now been seen by two reviewers, who both provided a very thorough assessment and gave several excellent suggestions to improve your work. From your reply, it appears that you will be able to address all their concerns. However, the required changes will alter the manuscript substantially. As such, I invite you to re-submit your manuscript, after which it will be sent out for a second review round.

(2) p.s. please note that your answer to comment 6 by reviewer 2 still needs a little work. At the start of the paragraph, you state that “Total respired CO₂ was higher in the litter-amended than non-amended soils before and after EPEs (Fig. S3), likely caused by the degradation of labile components in the fresh litter and/or increased degradation of SOC primed by litter additions (Fröberg et al., 2005; Ahmad et al., 2013).” However, towards the end of the paragraph you show that litter addition actually decreased the decomposition of native SOC. The first sentence clearly suggests the opposite, which makes the text a bit confusing.

Response:

(1) We appreciate reviewers’ critical and detailed assessment of our manuscript and we are grateful for his/her constructive comments which helped us to greatly refine our paper. Here we provide a point-to-point response to all the issues raised by the referee. We hope our replies and revisions will satisfy all the requests.

(2) Thank you very much for your kind suggestions. In this paragraph, we firstly would like to provide two possible reasons responsible for the higher total respired CO₂ in litter-amended than non-amended soils. However, whether and how these two reasons worked was not sure until our below isotope analysis. To avoid any possible confusion, we have revised this paragraph as follows:

“Total respired CO₂ was higher in the litter-amended than non-amended soils before and after EPEs (Fig. S3), likely caused by one or two following reasons: (1) the degradation of labile components in the fresh litter; (2) increased degradation of SOC primed by litter additions (Fröberg et al., 2005; Ahmad et al., 2013). These two reasons might affect CO₂ release in litter-amended soils in two ways: (1) both of them had positive effects; (2) one of them had positive effects and the other one had negative ones, but the total net effects was positive. To distinguish the influences of above two reasons on total respired CO₂ and further differentiate the contribution of litter (C4) and SOC (C3) to the respired CO₂, we examined the $\delta^{13}\text{C}$ values of CO₂ evolved from the GC soils after the first EPE. On the first day after EPE, CO₂ from the non-amended and litter-amended GC soils had a $\delta^{13}\text{C}$ value of -23.1‰ and -18.7‰ , respectively. The latter was close to the $\delta^{13}\text{C}$ signature of the added litter (-16.25‰). Using the two-endmember mixing model of Eq. (1) and (2), we calculated that litter contributed $\sim 64\%$ of the respired CO₂ in the litter-amended GC soils. However, along with the consumption of labile carbon in litter, the $\delta^{13}\text{C}$ signature of CO₂ decreased from -18.7‰ on Day 1 to -21.8‰ on Day 25 after EPE in the litter-amended soils (Fig. 2). Accordingly, the proportion of litter-derived CO₂ decreased from 64% to 20%. The litter-derived CO₂ flux in litter-amended GC soils was estimated to range from 7.0 to 17.5 mg C m⁻² h⁻¹, while the SOC-derived CO₂ flux increased from 6.2 to 15.7 mg C m⁻² h⁻¹ after the first EPE (Fig. S3). Compared with the SOC-derived CO₂ flux in non-amended GC soils (ranging from 17.2 to 27.1 mg C m⁻² h⁻¹), litter addition had a negative priming effect on the degradation of native SOC while increasing total respiration through labile litter degradation.”

Response to Referee #1

General Comments:

Comment 1:

The manuscript of Liu et al presents very interesting information regarding the triggering of soil carbon losses via respiration and leaching by extreme precipitation events. The results of the soil column experiments illustrate that leaching losses of carbon from soils as consequence of extreme precipitation events may well exceed carbon losses due to enhanced respiration. However, the overall relevance of dissolved organic and dissolved inorganic carbon leaching losses will ultimately depend on the fate of the leached carbon on its way to groundwater and further through rivers into the ocean. If the dissolved organic carbon and inorganic carbon are retained in subsoils, then the leached C might well be finally emitted from the soil to the atmosphere in the form of CO₂, if the dissolved organic carbon (DOC) is mineralized or if the soil water is lost via evapotranspiration, thus releasing the dissolved inorganic carbon (DIC)...This aspect of the importance of the downstream fate of leached carbon for the overall relevance of the leaching pathway for the carbon balance is missing in the manuscript.

Response:

This is a very good point! The downstream fate of the leached carbon is indeed very important as it will ultimately determine the relevance of leaching processes to the overall carbon budget or balance along the “soil-river-ocean” continuum. If part of the leached carbon is retained in the deeper soils or transformed and carried along from the river to the ocean in the form of DIC, it will not constitute a source of atmospheric CO₂ on a relatively short term (over years or decades). However, compared to DOC and DIC in the soil solution, the leached carbon is more likely to be subject to more intensified mineralization and outgassing during the land-ocean transfer, given more intensified mixing processes, oxygen exposure and photo-oxidation of terrestrial carbon upon releasing into the river (Hedges et al., 1997; Battin et al., 2009). Hence, we postulate that carbon leached from soils is more vulnerable to decomposition and/or release compared to that retained in the soil. That being said, it will be necessary to confirm our results and hypothesis using field-based leaching experiments to better understand the ultimate fate of leached soil carbon: whether it will be retained in the deeper soil or show a higher degradability upon leaving the soil matrix. Such information will be complementary to our study and further elucidate the importance of leaching processes in terms of ecosystem carbon budget.

The above considerations and discussions are now added as a separate paragraph in Section 3.4 in the revised paper:

“An uncertainty related to the importance of leaching processes in the overall carbon budget along the “soil-river-ocean” continuum lies in the ultimate downstream fate of the leached carbon. If part of this carbon is retained in the surrounding soils or carried along from the river to the ocean in the form of DIC without outgassing into the air, it will not constitute a source of atmospheric CO₂ on a relatively short term (over years or decades). However, soil columns used in our study has a depth (60 cm) typical of or even deeper than the average soil depth in the alpine grasslands of Qinghai-Tibetan Plateau (Wang et al., 2001). Hence, we assume that carbon leached in our experiments will have minimum retention in the soil. Furthermore, compared to DOC and DIC in the soil solution, the leached carbon is more likely to be subject to more intensified mineralization and outgassing during the land-ocean transfer, given more intensified mixing processes, oxygen exposure and photo-oxidation of terrestrial carbon upon releasing into the river (Hedges et al., 1997; Battin et al., 2009). Hence, we postulate that

carbon leached from soils is more vulnerable to decomposition and/or release compared to that retained in the soil. That being said, it will be necessary to confirm our results and hypothesis using field-based leaching experiments to better understand the ultimate fate of leached soil carbon: whether it will be retained in the deeper soil or show a higher degradability upon leaving the soil matrix. Such information will be complementary to our study and further elucidate the importance of leaching processes in terms of ecosystem carbon budget.”

References:

- Battin, T. J., Luysaert, S., Kaplan, L. A., Aufdenkampe, A. K., Richter, A., and Tranvik, L. J.: The boundless carbon cycle, *Nature Geoscience*, 2, 598-600, 2009.
- Hedges, J. I., Keil, R. G., and Benner, R.: What happens to terrestrial organic matter in the ocean?, *Org. Geochem.*, 27, 195-212, 1997.
- Wang, S.Q., Zhu, S.L., and Zhou, C.H.: Characteristics of spatial variability of soil thickness in China, *Geographical research*, 20, 161-169, 2001.

Comment 2:

When judging the relevance of dissolved inorganic carbon leaching for the carbon balance, it is also crucial to differentiate between the biogenic fraction of DIC and its lithogenic (carbonate-rock derived) fraction. In my opinion, it is much more straight forward to compare the biogenic leaching losses of DIC with the NEP, than total DIC leaching losses. The authors might want to consider this in their discussion of their results in lines 25ff on page 9. In this context the application of the isotopic mass balance model is important. The results of this model depend strongly on the delta 13C values of the end-members carbonate and CO₂ from soil respiration. The authors decided to use the delta 13C of the soil organic matter of -24 per-mille to calculate the biogenic fraction of dissolved inorganic carbon. Because isotopic fractionation occurs during the mineralization of soil organic matter, the authors might additionally use their delta 13C value of -23.1 per-mille as end member in order to assess the uncertainty that is associated with potential isotopic fractionation during mineralization and diffusive CO₂ transport in soil (Cerling et al., 1991. On the isotopic composition of carbon in soil carbon dioxide. *Geochim. Cosmochim. Acta* 55, 3403-3405). Quantitatively more important than the isotopic fractionation during mineralization and diffusion of CO₂ for the delta 13C value used as end member for the soil organic carbon derived fraction of DIC is the isotopic fractionation between CO₂ in the gas phase and bicarbonate (Zhang et al., 1995. Carbon isotope fractionation during gas-water exchange and dissolution of CO₂. *Geochim. Cosmochim. Acta* 59, 107-114). In the pH range of the investigated soils, the vast majority of the DIC will be present as bicarbonate (HCO₃⁻). According to Zhang et al. (1995), isotope fractionation between the gas phase and the aqueous phase will shift the delta 13C of bicarbonate in equilibrium with gaseous CO₂ by some 10-11 per-mille. Hence, the end member delta 13C of DIC in equilibrium with CO₂, which has a delta 13C value of -24 per-mille, can be around -14 to -13 per-mille. Considering this isotopic fractionation between gaseous CO₂ and bicarbonate will greatly increase the calculated fractions of biogenic (soil organic carbon-derived) DIC.

Response:

We must thank the reviewer for pointing out an excellent point that we overlooked. The dissolution of CO₂ produced by SOC degradation does cause large isotope fractionation on the biogenic carbonate/bicarbonate. Taking this into account, we have revised our endmember values ($\delta^{13}\text{C}_{\text{biogenic-DIC}}$ is estimated to be -16‰), and revised the following parts:

Section 2.4:

“Similarly, the relative contribution of lithogenic carbonate and biogenic DIC derived from SOC degradation to leached DIC was assessed according to the following isotopic mass balance model:

$$f_{\text{carbonate}} + f_{\text{biogenic-DIC}} = 1 \quad (3)$$

$$f_{\text{carbonate}} \times \delta^{13}\text{C}_{\text{carbonate}} + f_{\text{biogenic-DIC}} \times \delta^{13}\text{C}_{\text{biogenic-DIC}} = \delta^{13}\text{C}_{\text{DIC}} \quad (4)$$

where $f_{\text{carbonate}}$ and $f_{\text{biogenic-DIC}}$ are proportion of carbonate- and biogenic DIC in total DIC; $\delta^{13}\text{C}_{\text{carbonate}}$ is the $\delta^{13}\text{C}$ value of soil carbonate, equivalent to 0‰ (Edwards and Saltzman, 2016); and $\delta^{13}\text{C}_{\text{biogenic-DIC}}$ is the $\delta^{13}\text{C}$ value of biogenic carbonate/bicarbonate derived from the dissolution of CO_2 produced by SOC degradation, which is estimated to shift by approximately 8‰ compared with the $\delta^{13}\text{C}$ value of soil-respired CO_2 (–24‰ here) due to isotope fractionation during CO_2 dissolution (Zhang et al., 1995). Hence, $\delta^{13}\text{C}_{\text{biogenic-DIC}}$ is estimated to be –16‰. $\delta^{13}\text{C}_{\text{DIC}}$ is the measured $\delta^{13}\text{C}$ signature of leached DIC. According to Hendy (1971) and Doctor et al. (2008), isotopic fractionation of leached DIC due to CO_2 loss in an open system is insignificant when the partial pressure of CO_2 ($p\text{CO}_2$) in the solution is lower than twice that of the surrounding atmosphere. Therefore, due to the much lower $p\text{CO}_2$ in the XLHT leachates ($\sim 200 \mu\text{atm}$; Table S2) compared to that in the ambient atmosphere ($> 400 \mu\text{atm}$), the influence of CO_2 outgassing on the $\delta^{13}\text{C}$ of leached DIC was not considered in the present study.”

Section 3.3:

“Based on the isotopic mass balance Eq. (3) and (4), lithogenic carbonate (with a $\delta^{13}\text{C}$ value of 0‰) contributed 51.2% to the leached DIC while biogenic DIC produced by SOC degradation contributed 48.4% (Fig. 5). The $\delta^{13}\text{C}$ value of leached DIC decreased to –12.3‰ and –13.5‰ during the second and third EPEs, corresponding to a contribution of 77.0% and 84.4% by biogenic sources in the total DIC, respectively (Fig. 5). ... Combined with the total flux rate, we calculated that both lithogenic and biogenic DIC fluxes were $\sim 2.1 \text{ g C m}^{-2}$ in the first EPE. Subsequently, lithogenic DIC flux decreased to $\sim 1.3 \text{ g C m}^{-2}$ while biogenic DIC flux increased to 7.6 g C m^{-2} in the third EPE. This demonstrates that increased SOC degradation mainly contributed to the increased DIC fluxes with repeated EPEs.”

Section 3.4:

“It is worth mentioning that biogenic DIC loss ($16.0 \pm 3.4 \text{ g C m}^{-2}$) caused by SOC degradation accounted for 184% of NEP at XLHT, indicating the importance of biogenic DIC to SIC loss during EPEs.”

Reference:

Zhang, J., Quay, P. D., and Wilbur, D. O.: Carbon isotope fractionation during gas-water exchange and dissolution of CO_2 , *Geochim. Cosmochim. Acta*, 59, 107-114, [http://dx.doi.org/10.1016/0016-7037\(95\)91550-D](http://dx.doi.org/10.1016/0016-7037(95)91550-D), 1995.

Specific Comments:

Comment 3:

On page 3, line 19, the soil is classified as “chestnut soil”. This classification is not in line with the international soil classification system of the World Reference Base for Soil Resources (WRB, 2015). Please classify your soils also according to the WRB system (<http://www.fao.org/3/a-i3794e.pdf>).

Response:

Revised. Soils in Xilinhot and Keqi are classified as Calcic Chernozems according to the World Reference Base for Soil Resources (Steffens et al., 2008; IUSS working group WRB, 2015) while soils in Gangcha are

mainly Gelic Cambisol (IUSS working group WRB, 2015).

Comment 4:

On page 6, lines 20ff, the authors argue that the variation in SIC contents between sites are caused by the variation of pH values, suggesting a causality between pH (independent variable) and SIC content (dependent variable). The question is, whether the pH is really controlling SIC contents or vice versa. . .

Response:

SIC content is related to parent materials as well as soil pH. However, in the present study regions, soil pH is the key factor controlling SIC variation across the Mongolian and Tibetan grasslands according to the results of Shi et al. (2012). We have added one sentence to clarify this:

“This dependence of SIC content on soil pH is consistent with the results of Shi et al. (2012), showing that pH is the most important factor controlling SIC variation across the Mongolian and Tibetan grasslands.”

Comment 5:

On page 9, lines 29ff, the authors cite Kindler et al. (2011) for numbers of DIC leaching losses equaling 12% of NEP and DOC leaching losses equaling 2% of NEP. I do not understand how the authors extracted these numbers from the Kindler et al. (2011) publication.

Response:

This is clarified now in the revised paper:

According to the report of Kirschbaum et al. (2001), both Net Ecosystem Exchange (NEE) and NEP refer to net primary production minus carbon loss from heterotrophic respiration, R_h :

$$NEE = NEP = NPP - R_h = GPP - R_{\text{autotrophic respiration}} - R_h$$

These terms are used somewhat interchangeably, with NEE used more often when they are addressed based on measurements of gas exchange rates using atmospheric measurements over the time scales of hours, whereas NEP is more often used if measurements are based on ecosystem-carbon stock changes, typically measured over a minimal period of one year.

The NEE in Kindler et al. (2011) equals to gross primary productivity minus ecosystem respiration, excluding C deprivation with harvest, fires, etc. Therefore, we extracted NEE in Table 5 of Kindler et al. (2011) as NEP for calculating the proportion of leaching DOC and DIC in NEP. Relevant description has been added in Section 3.4 as follows:

“Nonetheless, the EPE-induced soil carbon loss relative to NEP was higher in this study than that estimated for grassland topsoil across Europe (12% for DIC loss, 2% for DOC loss; Kindler et al., 2011) where Net Ecosystem Exchange (NEE) reported by Kindler et al. was used as NEP according to the report of Kirschbaum et al. (2001).”

Comment 6:

Starting on page 9, line 31, the authors argue that the carbon loss due to extreme precipitation events was much greater than carbon losses through warming-enhances respiration. This comparison is perhaps

misleading, because it implies that extreme precipitation events occur only as consequence of climate change. More correct would be the comparison of carbon losses due to warming-enhanced respiration with carbon losses due to “climate change-enhanced” extreme precipitation events.

Response:

The reviewer raised a good point. It is not fair to compare carbon leaching through all annual EPEs with warming-induced respiration increase. To be more consistent and robust, we decide to delete the discussion on the comparison of carbon losses due to warming-induced respiration with carbon losses due to annual EPEs.

Response to Referee #2

General Comments:

Comment 1:

The manuscript presents a study that attempt to evaluate the effect of extreme precipitation events on soil carbon losses in arid and semi-arid grasslands. The objective was to distinguish between C losses due to respiration and leaching. Additionally, leaching losses were separated into DIC and DOC losses. Therefore, a soil column experiment was conducted, where respiration and leaching losses were measured after an artificial precipitation events. Soil inorganic carbon losses due to leaching was higher than due to an enhanced respiration.

(1) As already mentioned by the first referee, the relevance of C losses depends on the fate of DIC and this should be more pronounced in the discussion.

(2) In addition, soil carbon losses due to DIC leaching has to be more discussed in detail, especially the fact that about 50% (or even more with the already recalculated values) of the DIC originates from SOC degradation (dissolved CO₂). In consequence, the conclusion that most soil carbon during EPE is lost due to DIC (in particular SIC), might be not true. On the contrary, most of the DIC originates from dissolution of CO₂, which originates from SOC mineralization and not SIC leaching. This should be discussed much more in detail.

Response:

(1) We have added one paragraph to discuss the fate of DIC in Section 3.4 as follows:

“An uncertainty related to the importance of leaching processes in the overall carbon budget along the “soil-river-ocean” continuum lies in the ultimate downstream fate of the leached carbon. If part of this carbon is retained in the surrounding soils or carried along from the river to the ocean in the form of DIC without outgassing into the air, it will not constitute a source of atmospheric CO₂ on a relatively short term (over years or decades). However, soil columns used in our study has a depth (60 cm) typical of or even deeper than the average soil depth in the alpine grasslands of Qinghai-Tibetan Plateau (Wang et al., 2001). Hence, we assume that carbon leached in our experiments will have minimum retention in the soil. Furthermore, compared to DOC and DIC in the soil solution, the leached carbon is more likely to be subject to more intensified mineralization and outgassing during the land-ocean transfer, given more intensified mixing processes, oxygen exposure and photo-oxidation of terrestrial carbon upon releasing into the river (Hedges et al., 1997; Battin et al., 2009). Hence, we postulate that carbon leached from soils is more vulnerable to decomposition and/or release compared to that retained in the soil. That being said, it will be necessary to confirm our results and hypothesis using field-based leaching experiments to better understand the ultimate fate of leached soil carbon: whether it will be retained in the deeper soil or show a higher degradability upon leaving the soil matrix. Such information will be complementary to our study and further elucidate the importance of leaching processes in terms of ecosystem carbon budget.”

(2) The reviewer raised an excellent point and we agree that the source of DIC needs to be discussed in order to determine the contribution of SIC dissolution to soil carbon loss. We have therefore calculated the flux of biogenic and lithogenic DIC from soils and added discussions in Sections 3.3 and 3.4 as follows:

“... Based on the isotopic mass balance of Eq. (3) and (4), lithogenic carbonate (with a $\delta^{13}\text{C}$ value of

0‰) contributed 51.2% to the leached DIC while biogenic DIC produced by SOC degradation contributed 48.4% (Fig. 5). The $\delta^{13}\text{C}$ value of leached DIC decreased to -12.3‰ and -13.5‰ during the second and third EPEs, corresponding to a contribution of 77.0% and 84.4% by biogenic sources in the total DIC, respectively (Fig. 5). These results confirm our previous hypothesis that SOC decomposition contributed significantly to soil DIC fluxes. Combined with the total flux rate, we calculated that both lithogenic and biogenic DIC fluxes were $\sim 2.1 \text{ g C m}^{-2}$ in the first EPE. Subsequently, lithogenic DIC flux decreased to $\sim 1.3 \text{ g C m}^{-2}$ while biogenic DIC flux increased to 7.6 g C m^{-2} in the third EPE. This demonstrates that the increased DIC flux with repeated EPEs was mainly derived from increased contribution of SOC mineralization. Interestingly, increasing DIC fluxes with repeated EPEs were not observed in the KQ and GC soils (Fig. 4) despite their higher SOC contents and CO_2 release rates (Fig. S5). ...”

“Regardless of its source, the EPE-induced leaching loss of inorganic carbon was 31.5 and $10.6 \mu\text{g DIC g}^{-1}$ soil from the alkaline XLHT and GC soils, respectively, approximately three and five times higher than the corresponding DOC leaching loss (5.9 and $3.9 \mu\text{g DOC g}^{-1}$ soil, respectively). However, the KQ soil had a relatively lower EPE-induced DIC loss ($4.4 \mu\text{g DIC g}^{-1}$ soil) than the DOC leaching loss ($11.6 \mu\text{g DOC g}^{-1}$ soil) mainly due to its lower initial SIC content and relatively neutral soil pH value. Hence, DIC was the main form of soil carbon loss in alkaline soils during EPEs regardless of its source. When the source of the leached DIC is taken into account, dissolution of CO_2 produced by SOC mineralization (biogenic DIC) constituted more than half of the leached DIC (at least from the XLHT soils; Fig. 5), whose contribution increased with re-occurring EPEs (Fig. 5). This implies that SOC mineralization measured by CO_2 fluxes was under-estimated by approximately 8 times in the XLHT soils during the three EPEs (Fig. 5). In addition, DIC loss exclusively resulting from SIC dissolution or weathering was also a significant fraction of soil carbon loss, equivalent to 219% SOC loss in the form of CO_2 during EPEs (Fig. 5). These results collectively corroborate that inorganic carbon loss is the main form of soil carbon loss in alkaline soils during EPEs.”

“... In view of DIC sources, biogenic DIC loss derived from SOC mineralization contributed to more than half of the total leached DIC fluxes and accounted for 184% of the NEP at XLHT. ... These results also imply that SOC mineralization measured by CO_2 fluxes might be drastically under-estimated in alkaline grassland soils during EPEs. ...”

Comment 2:

The experimental setup seems appropriate for the objectives presented in the manuscript, however the presented results need some reconsideration and recalculation, especially respiration data should be presented as specific respiration to account for different SOC contents in the investigated soils (for more detail see specific comments).

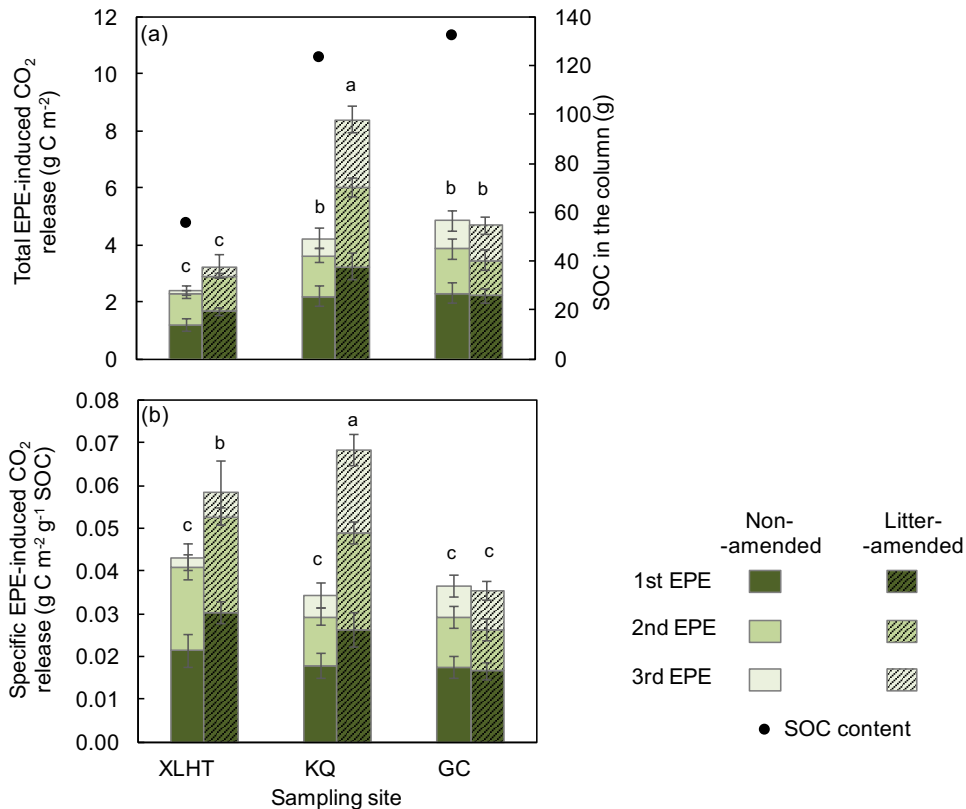
Response:

Following the reviewer’s suggestion, we calculate the specific respiration data normalization to SOC and added in Sections 3.2 and Figure 3b as follows:

“... The specific soil respiration rates normalized to SOC were 2.2 , 2.6 , and $7.9 \mu\text{g C g}^{-1} \text{SOC h}^{-1}$ in the GC, KQ, and XLHT soils, respectively. This indicated that SOC in the XLHT soils was easier to degrade despite

its low content.”

Figure:



“Figure 3: Total (a) and specific (b) extreme precipitation event (EPE)-induced CO₂ release in the litter-amended and non-amended grassland soils during three EPEs. Mean values are shown with standard deviation ($n = 3$). Lower-case letters (a, b, c) indicate significantly different levels among the litter-amended and non-amended soils determined by Duncan’s multiple range test (one-way ANOVA, $p < 0.05$).”

Section 3.2:

“Using data shown in Fig. S3-4, we calculated that total EPE-induced CO₂ release during three EPEs was higher in the KQ and GC soils than in the XLHT soil ($p < 0.05$; Fig. 3a) with a lower SOC content and a lower SOC:N ratio (Table 1). However, the specific EPE-induced CO₂ release normalized to SOC content showed no significant difference in the non-amended soils among three sites (Fig. 3b), indicating that a similar proportion of SOC (~4%) was subject to EPE-induced CO₂ release in the alpine and temperate grassland soils. The total EPE-induced CO₂ release (including CO₂ from both litter and SOC mineralization) was significantly higher in the litter-amended KQ soils than the non-amended ones, similar to the specific EPE-induced CO₂ in the KQ and XLHT soils. The specific EPE-induced CO₂ was significantly different for the litter-amended soils among sites ($p < 0.05$), showing a pattern of KQ > XLHT > GC. This pattern was consistent with the mean sand content in the order of KQ (46.9%) > XLHT (39.1%) > GC (27.2%). In addition, the higher total and specific EPE-induced CO₂ release in the litter-amended KQ soils may be related to its relatively lower soil pH (~7.7), which facilitates the release rather than the dissolution of respired CO₂ in soil solution. We therefore conclude that the KQ soil, with a coarser texture and a lower pH (Table 1), may have provided less sorptive protection for the labile DOC components after EPEs (Kell et al., 1994; Nelson et al., 1994) and allowed less dissolution of the respired CO₂, and hence showed a more responsive respiration to the precipitation events. These results suggest that SOC contents and SOC:N ratios are important factors influencing the total EPE-induced CO₂ release, while the availability of labile

organic carbon, soil texture and pH are key factors affecting the specific EPE-induced CO₂ release in these grassland soils.”

Specific Comments:

Comment 3:

Page 3, line 25: The dimension of the soil pits seems quite small 10 cm x 10 cm. Even by using a shovel I would expect that you need a bigger area to go down to 70 cm.

Response:

Thank you for pointing out this inaccurate description. We have confirmed that the pit was 25 cm × 25 cm × 70 cm. This is corrected in the text.

Comment 4:

Page 5, line 1: How was the water added to the soil columns? Did you have needles in the top lid of the soil column? Did you used a constant rate, like 0.5 mm per minute? How much time was in between the EPE events? How long did you wait until you start a new EPE? How where the soil columns treated in between the CO₂ measurements? Were they closed or flushed with constant air flow? Please provide more information about the experimental setup for the reader.

Response:

More information about the experimental setup is added at Section 2.2 as follows:

“...approximately 1 L of rainwater (rainfall of ~127 mm), comparable to 30% of the MAP of the investigated sites, was added to the surface of each soil column over 3–4 h at rates of one drop per second using syringes and allowed to leach through the column to be collected with a clean beaker within 12–14 h. ... To monitor soil respiration every 1–2 days following each EPE, soil columns were first aerated for 1 h using CO₂-depleted air that had been passed through saturated sodium hydroxide (NaOH) solutions (twice; Fig. 1) and then incubated for 4 h with lids closed. CO₂ gas in the column headspace was collected by gas-tight syringes for the subsequent measurement. After collection of CO₂ gas, the lids were open to allow the exchange with the ambient air. Soil respiration was monitored for 30 days after the first EPE and observed to stabilize approximately on the 20th day (Fig. S3). Hence, the first, second, and third EPEs were conducted on the 1st, 31st, and 51st day of incubation, and the CO₂ measurement was conducted for approximately 30, 20, and 20 days after the first, second, and third EPEs, respectively.”

Comment 5:

Page 7, line 8: Additionally, to the respiration rate I'd suggest to calculate a specific respiration rate, which is the respiration rate divided by the amount of SOC (mg CO₂-C g⁻¹ SOC h⁻¹). This would allow an easier comparison of the different soils with different SOC content.

Response:

The specific respiration rate has been added in Section 3.2 as follows:

“...The specific soil respiration rates normalized to SOC were 2.2, 2.6, and 7.9 μg C g⁻¹ SOC h⁻¹ in the GC, KQ, and XLHT soils, respectively. This indicated that SOC in the XLHT soils was easier to degrade despite

its low content. “

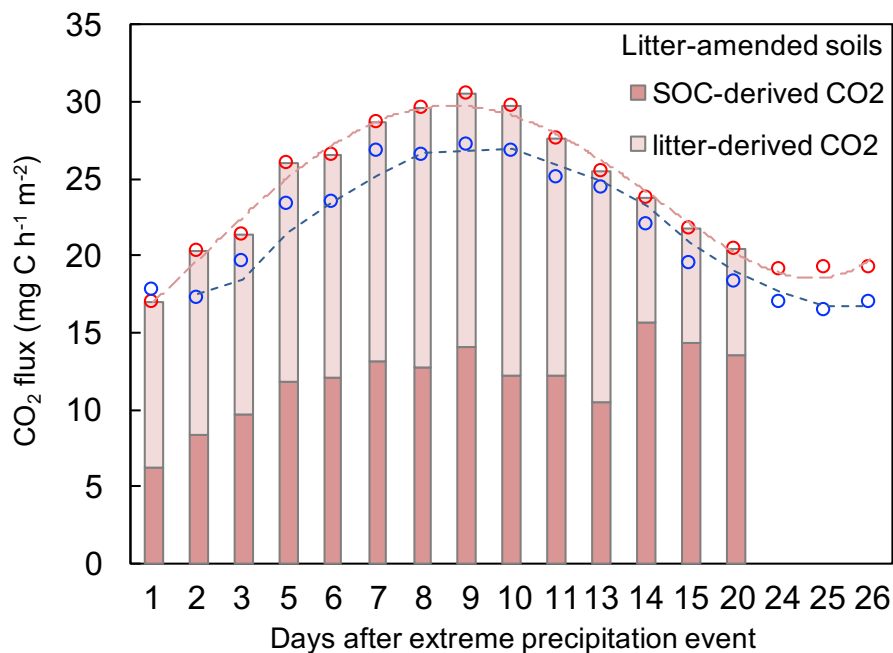
Comment 6:

Page 7 line 13: The authors argue that litter addition increase respiration due to mineralization of labile litter compounds and priming effects. However, here were no values presented which would underline this statement. Since the authors measured $\delta^{13}\text{C}\text{O}_2$, they should be able to separate litter mineralization from SOC mineralization.

Response:

Good point! We take the GC soils as an example to quantify the relative contribution of litter and SOC mineralization to the respired CO_2 in the following parts and added one inside-figure in Fig. S3:

Figure:



Inside figure of Fig. S3: Variations of litter-derived and SOC-derived CO_2 release with time in the litter-amended GC soils during the first EPE.

Section 3.2

“Total respired CO_2 was higher in the litter-amended than non-amended soils before and after EPEs (Fig. S3), likely caused by one or two following reasons: (1) the degradation of labile components in the fresh litter; (2) increased degradation of SOC primed by litter additions (Fröberg et al., 2005; Ahmad et al., 2013). These two reasons might affect CO_2 release in litter-amended soils in two ways: (1) both of them had positive effects; (2) one of them had positive effects and the other one had negative ones, but the total net effects was positive. To distinguish the influences of above two reasons on total respired CO_2 and further differentiate the contribution of litter (C4) and SOC (C3) to the respired CO_2 , we examined the $\delta^{13}\text{C}$ values of CO_2 evolved from the GC soils after the first EPE. On the first day after EPE, CO_2 from the non-amended and litter-amended GC soils had a $\delta^{13}\text{C}$ value of -23.1‰ and -18.7‰ , respectively. The latter was close to the $\delta^{13}\text{C}$ signature of the added litter (-16.25‰). Using the two-endmember mixing model of Eq. (1) and (2), we calculated that litter contributed $\sim 64\%$ of the respired CO_2 in the litter-amended GC soils. However, along with the consumption of labile carbon in litter, the $\delta^{13}\text{C}$ signature of CO_2 decreased from -18.7‰ on

Day 1 to -21.8% on Day 25 after EPE in the litter-amended soils (Fig. 2). Accordingly, the proportion of litter-derived CO_2 decreased from 64% to 20%. The litter-derived CO_2 flux in litter-amended GC soils was estimated to range from 7.0 to $17.5 \text{ mg C m}^{-2} \text{ h}^{-1}$, while the SOC-derived CO_2 flux increased from 6.2 to $15.7 \text{ mg C m}^{-2} \text{ h}^{-1}$ after the first EPE (Fig. S3). Compared with the SOC-derived CO_2 flux in non-amended GC soils (ranging from 17.2 to $27.1 \text{ mg C m}^{-2} \text{ h}^{-1}$), litter addition had a negative priming effect on the degradation of native SOC while increasing total respiration through labile litter degradation.”

Comment 7:

Page 7, line 17: (1) The used mixing model should be mentioned in the method section. (2) Further with the given isotopic values I cannot understand how the authors calculated contribution of litter mineralization to total respiration. Using a mixing model of: $1 - ((c_{\text{mix}} - c_{\text{litter}}) / (c_{\text{control}} - c_{\text{litter}}))$, where c_{mix} is the isotopic value of CO_2 from the litter amended sample (-18.7% , c_{litter} the isotopic value of the added litter (-16.2% and c_{control} the isotopic value of CO_2 from the non-amended sample (-23.1% , the contribution of litter mineralization to total respiration was around 64% at day 1 and only 19% at day 25. Which values for $\delta^{13}\text{CO}_2$ values did you used for the control (non- amended) samples. Did you measured $\delta^{13}\text{CO}_2$ for the control only at the beginning or at the same resolution as $\delta^{13}\text{CO}_2$ for the litter-amended samples? (3) Further, are there any isotopic measurements of the other two sites. If so, why they are not shown here?

Response:

(1) The description on the mixing model has been added in Section 2.4 as follows:

“The relative contribution of litter- and SOC-derived CO_2 to total respired CO_2 in the litter-amended soils was estimated using the following mass balance model:

$$f_{\text{litter-derived}} + f_{\text{SOC-derived}} = 1 \quad (1)$$

$$f_{\text{litter-derived}} \times \delta^{13}\text{C}_{\text{litter-derived}} + f_{\text{SOC-derived}} \times \delta^{13}\text{C}_{\text{SOC-derived}} = \delta^{13}\text{C}_{\text{respired-CO}_2} \quad (2)$$

where $f_{\text{litter-derived}}$ and $f_{\text{SOC-derived}}$ are the proportion of litter- and SOC-derived CO_2 in the total respired CO_2 ; $\delta^{13}\text{C}_{\text{litter-derived}}$ is the $\delta^{13}\text{C}$ value of litter-derived CO_2 , equivalent to -16.25% ; $\delta^{13}\text{C}_{\text{SOC-derived}}$ is the $\delta^{13}\text{C}$ value of SOC-derived CO_2 , which assumes the same value as that in the non-amended soils at the beginning of incubation (-23.1%) according to Cerling et al. (1991); $\delta^{13}\text{C}_{\text{respired-CO}_2}$ is the measured $\delta^{13}\text{C}$ of respired CO_2 .”

(2) Regarding the calculation results on the proportion of litter-derived CO_2 , I must admit that I had made a very stupid mistake. I previously wrongly calculated the value of $(c_{\text{control}} - c_{\text{litter}})$ as -8.95% , which actually was -6.85% (i.e., $(-23.1\%) - (-16.25\%)$), and this mistake resulted in the incorrect proportions (72% to 39%). I have revised these values and re-checked all the data in this manuscript. The revised parts are as follows:

“Using the two-endmember mixing model of Eq. (1) and (2), we calculated that litter contributed $\sim 64\%$ of the respired CO_2 in the litter-amended GC soils. However, along with the consumption of labile carbon in litter, the $\delta^{13}\text{C}$ signature of CO_2 decreased from -18.7% on Day 1 to -21.8% on Day 25 after EPE in the litter-amended soils (Fig. 2). Accordingly, the proportion of litter-derived CO_2 decreased from 64% to 20%.”

(3) Due to budget constraints and logistic reasons, we measured the isotopes of respired CO_2 only in the GC soil during the first extreme precipitation event. The other two soils were not measured. As the present study mainly aimed to compare the relative importance of respiration and leaching in EPE-

induced soil carbon loss from grassland soils, the isotopic analysis was conducted for only one soil to provide some information regarding the source(s) of the leached and respired carbon.

Comment 8:

Page 7 line21.: (1) Despite the fact that the calculation described here might be simple, it should be part of the method section and not of the result/discussion section. (2) “EPE-induced CO₂ release was higher in the KQ and GC soils than in the XLHT soil ($p < 0.05$; Fig. 3) that had a lower SOC content and a lower SOC:N ratio (Table 1)”, as mentioned above, I suggest to calculate a specific respiration normalized to the absolute amount of SOC in the soil column. The specific respiration will provide more information about the stability and the loss of C from the different sites. A rough calculation based on figure 3 revealed that respiration of the 3 sites in the non-amended treatment might not differ. However, this has to be checked with the measurement values. It is also not clear to which EPE is shown in Fig 3, is it the first, second or third one?

Response:

(1) Calculation on EPE-induced CO₂ release has been moved to the method Section 2.4 as follows:

“EPE-induced CO₂ release via respiration was assessed following two steps. First, cumulative respiration during the first 20 days after each EPE (until respiration rate stabilized) was calculated. Second, difference between the measured cumulative respiration and that estimated using the stabilized basal respiration rate after each EPE was calculated as the EPE-induced CO₂ release.”

(2) Results on the specific EPE-induced CO₂ release normalized to the SOC content has been added as Fig. 3b, and discussed in Section 3.2 (please see our response to Comment 2).

Comment 9:

Page 7, line 25: “Litter amendment significantly increased the EPE-induced CO₂ release from the KQ soil ($p < 0.05$) but did not have any effect on the XLHT and GC soils.” - I disagree with this statement, because figure 3 only show total CO₂ release which is the sum of CO₂ from litter mineralization and SOC mineralization from the mineral soil. Therefore, it is only possible to say that EPE increased total CO₂ release. However, as mentioned above, I would recommend to use the isotopic data of CO₂ to separate respiration into the two sources litter and soil. Which would allow a comparison of SOC mineralization between non-amended and amended soils.

Further the authors explained the higher CO₂ release in the KQ soil with a coarser soil texture and less sorptive protection of DOC, however I can't follow this explanation since Table 1 show a lower sand content in the KQ than in the XLHT soils. “These results suggest that soil texture, SOC content and quality are important factors influencing EPE-enhanced soil respiration.” This is a quite strong statement, because there is no information given about the SOC quality in the different soils used in this study, also the soil texture of the used soils is more or less similar and the presented results does not justify such a strong statement. In fact, the higher CO₂ release in the KQ can be explained by several other factors, such as the higher pH of the GC and XLHT soils. The higher the pH the more CO₂ is dissolved, which can be seen in Fig 4 in the higher DIC contents of these sites. Also negative priming effects may occur in the GC and XLHT sites which lead to a reduced SOC mineralization, because microorganisms utilize the added litter first. This should be discussed in more detail.

Response:

We agree with the first part of the comment and have quantified the litter- and SOC-derived CO₂ as details below. As mentioned in our replies to previous comments, the isotopes of respired CO₂ were only measured in the GC soils during the first extreme precipitation event due to logistic reasons. Nonetheless, we have added detailed discussion on the total EPE-induced CO₂ release as well as its influencing factors in the following parts:

Section 3.2

“Total respired CO₂ was higher in the litter-amended than non-amended soils before and after EPEs (Fig. S3), likely caused by one or two following reasons: (1) the degradation of labile components in the fresh litter; (2) increased degradation of SOC primed by litter additions (Fröberg et al., 2005; Ahmad et al., 2013). These two reasons might affect CO₂ release in litter-amended soils in two ways: (1) both of them had positive effects; (2) one of them had positive effects and the other one had negative ones, but the total net effects was positive. To distinguish the influences of above two reasons on total respired CO₂ and further differentiate the contribution of litter (C4) and SOC (C3) to the respired CO₂, we examined the δ¹³C values of CO₂ evolved from the GC soils after the first EPE. On the first day after EPE, CO₂ from the non-amended and litter-amended GC soils had a δ¹³C value of −23.1‰ and −18.7‰, respectively. The latter was close to the δ¹³C signature of the added litter (−16.25‰). Using the two-endmember mixing model of Eq. (1) and (2), we calculated that litter contributed ~64% of the respired CO₂ in the litter-amended GC soils. However, along with the consumption of labile carbon in litter, the δ¹³C signature of CO₂ decreased from −18.7‰ on Day 1 to −21.8‰ on Day 25 after EPE in the litter-amended soils (Fig. 2). Accordingly, the proportion of litter-derived CO₂ decreased from 64% to 20%. The litter-derived CO₂ flux in litter-amended GC soils was estimated to range from 7.0 to 17.5 mg C m^{−2} h^{−1}, while the SOC-derived CO₂ flux increased from 6.2 to 15.7 mg C m^{−2} h^{−1} after the first EPE (Fig. S3). Compared with the SOC-derived CO₂ flux in non-amended GC soils (ranging from 17.2 to 27.1 mg C m^{−2} h^{−1}), litter addition had a negative priming effect on the degradation of native SOC while increasing total respiration through labile litter degradation.”

“Using data shown in Fig. S3-4, we calculated that total EPE-induced CO₂ release during three EPEs was higher in the KQ and GC soils than in the XLHT soil ($p < 0.05$; Fig. 3a) with a lower SOC content and a lower SOC:N ratio (Table 1). However, the specific EPE-induced CO₂ release normalized to SOC content showed no significant difference in the non-amended soils among three sites (Fig. 3b), indicating that a similar proportion of SOC (~4%) was subject to EPE-induced CO₂ release in the alpine and temperate grassland soils. The total EPE-induced CO₂ release (including CO₂ from both litter and SOC mineralization) was significantly higher in the litter-amended KQ soils than the non-amended ones, similar to the specific EPE-induced CO₂ in the KQ and XLHT soils. The specific EPE-induced CO₂ was significantly different for the litter-amended soils among sites ($p < 0.05$), showing a pattern of KQ > XLHT > GC. This pattern was consistent with the mean sand content in the order of KQ (46.9%) > XLHT (39.1%) > GC (27.2%). In addition, the higher total and specific EPE-induced CO₂ release in the litter-amended KQ soils may be related to its relatively lower soil pH (~7.7), which facilitates the release rather than the dissolution of respired CO₂ in soil solution. We therefore conclude that the KQ soil, with a coarser texture and a lower pH (Table 1), may have provided less sorptive protection for the labile DOC components after EPEs (Kell et al., 1994; Nelson et al., 1994) and allowed less dissolution of the respired CO₂, and hence showed a more responsive respiration to the precipitation events. These results suggest that SOC contents and SOC:N ratios are important factors influencing the total EPE-induced CO₂ release, while the availability of labile organic carbon, soil texture and pH are key factors affecting the specific EPE-induced CO₂ release in these grassland soils.”

Comment 10:

Page 8 line 8: The authors mentioned that CO₂ from litter decomposition contributed to DIC, however in the following lines they argue that this effect is more pronounced in the KQ soils than in the XLHT and GC, due to the higher SIC content of the latter. This is true speaking in relative amounts, but considering absolute amounts I would expect that much more CO₂ is dissolved in the GC and XLHT soils due to the higher pH. Therefore CO₂ might also be a source of DIC in these soils with high SIC content. Which is also shown in line 23-24 on the same page.

Response:

The relative effect of litter amendment on DIC fluxes is now discussed in this paragraph. Because the added litter-OC was only 0.7 g per column calculated using added litter biomass of 1.59 g and litter-OC content of 43%, and the maximum DIC content derived from dissolution of CO₂ produced by litter degradation was 0.8 g assuming that all the litter-OC was mineralized and that all the produced CO₂ was dissolved in soil solution. Compared with the SIC content (38.15 g C per column) in XLHT soils, litter-mineralization-derived DIC was far smaller, but the dissolved CO₂ derived from litter-OC mineralization could be more important in the KQ soils having only 0.7 g SIC per column. To clarify this, we have revised this paragraph as follows:

“... Due to the high SIC content in the XLHT soils (38.15 g per column) and the low litter-OC amendment (0.7 g per column), there was no significant difference of DIC fluxes between the non-amended and litter-amended XLHT soils (Fig. 4a). However, for the KQ soil having a relatively low SIC content which was similar to the added litter-OC (0.7 g per column; Table 1), the influence of litter addition on the DIC flux was quite obvious. Therefore, although the contribution of dissolved CO₂ to DIC fluxes should be more important in high-pH soils, the relative effect of litter amendment on DIC fluxes under EPEs seemed more significant in soils with a low SIC content.”

Comment 11:

Page 8, line 19f.: Why was this measurement only done for the XLHT soils? Or why the results from the other sites not shown?

Response:

Due to budget constraints and logistic reasons, we only measured the isotopes of leached DIC in the XLHT soils and took it as an example to trace the source of the leached DIC. This is now mentioned in the Methods.

Comment 12:

Page 8, line 27f.: “Interestingly, increasing DIC fluxes were not observed in the KQ and GC soils (Fig. 4), although they had higher SOC content and degradation (i.e., respiration) rates (Fig. S5)” - What do you mean with higher degradation?

Response:

Given the above comments and discussions, it may be inaccurate to treat the degradation rate as the same as the CO₂ release rate. To clarify, we have revised this sentence as follows:

“Interestingly, increasing DIC fluxes with repeated EPEs were not observed in the KQ and GC soils (Fig. 4) despite their higher SOC contents and CO₂ release rates (Fig. S5).”

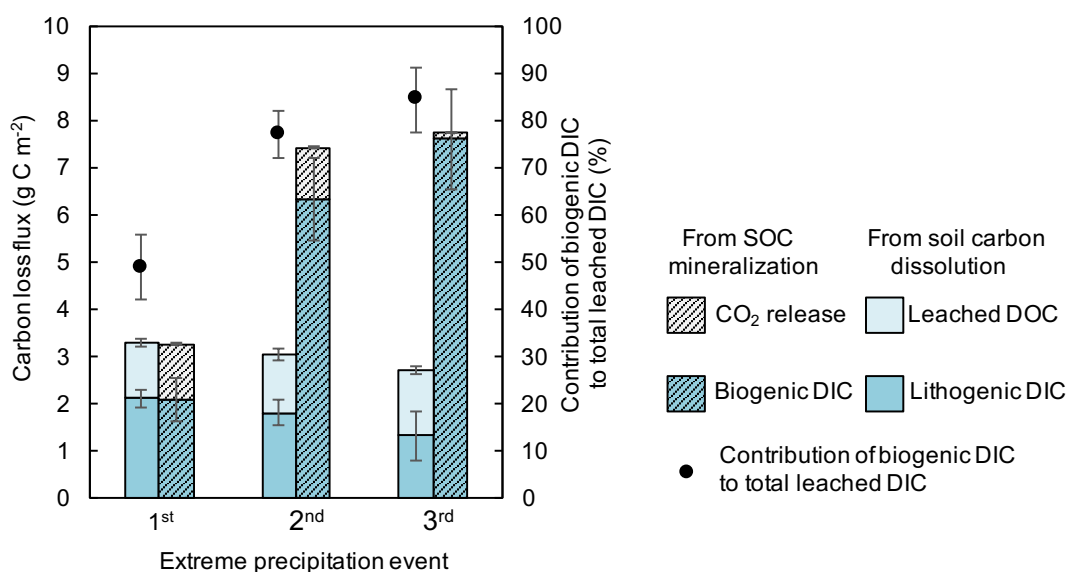
Comment 13:

Page 8, line 31: Here the authors calculated a specific leaching loss normalized to SIC content and compared it to DOC losses. However, in line 9 same page, it is mentioned that especially for the soils with low SIC content, the dissolution CO₂ from SOC and litter mineralization is a major source for DIC. Therefore, the high value of 32.5 mg C g⁻¹ SIC has to attributed to SOC mineralization and not to SIC loss. In consequence, the drawn conclusion that SIC loss is the main form of soil carbon loss in neutral to alkaline soils might be questioned, at least for the KQ soils.

Response:

Good point! It is inaccurate to say that SIC loss is the main form of soil carbon loss in neutral to alkaline soils. We have now used DIC flux rates in the units of mg C g⁻¹ soil to compare with SOC loss. In addition, regardless of the DIC sources, DIC (not SIC) was a major form of carbon loss from these grassland soils. In view of its sources, biogenic DIC should not be neglected. Therefore, we have revised Figure 5 and added one paragraph in Section 3.3 to discuss the role of biogenic DIC in leached DIC.

Figure :



“Figure 5: The flux of carbon loss from soil organic carbon (SOC) mineralization including CO₂ release and biogenic dissolved inorganic carbon (DIC), and that from soil carbon dissolution including leached dissolved organic carbon (DOC) and lithogenic DIC in the XLHT soils. Mean values are shown with standard error (n = 3).”

Section 3.3

“Regardless of its source, the EPE-induced leaching loss of inorganic carbon was 31.5 and 10.6 μg DIC g⁻¹ soil from the alkaline XLHT and GC soils, respectively, approximately three and five times higher than the corresponding DOC leaching loss (5.9 and 3.9 μg DOC g⁻¹ soil, respectively). However, the KQ soil had a relatively lower EPE-induced DIC loss (4.4 μg DIC g⁻¹ soil) than the DOC leaching loss (11.6 μg DOC g⁻¹ soil) mainly due to its lower initial SIC content and relatively neutral soil pH value. Hence, DIC was the main form of soil carbon loss in alkaline soils during EPEs regardless of its source. When the source of the

leached DIC is taken into account, dissolution of CO₂ produced by SOC mineralization (biogenic DIC) constituted more than half of the leached DIC (at least from the XLHT soils; Fig. 5), whose contribution increased with re-occurring EPEs (Fig. 5). This implies that SOC mineralization measured by CO₂ fluxes was under-estimated by approximately 8 times in the XLHT soils during the three EPEs (Fig. 5). In addition, DIC loss exclusively resulting from SIC dissolution or weathering was also a significant fraction of soil carbon loss, equivalent to 219% SOC loss in the form of CO₂ during EPEs (Fig. 5). These results collectively corroborate that inorganic carbon loss is the main form of soil carbon loss in alkaline soils during EPEs.”

Comment 14:

Page 9, line 33: “It is also worth mentioning that soil carbon leaching fluxes in this study (10.1–25.3 g C m⁻² yr⁻¹) far exceed annual SOC loss through warming-enhanced respiration at these sites (0.2–0.6 g C m⁻² yr⁻¹) given an assumed temperature sensitivity of 2 in climate...” - I think the authors can not make such a comparison, since most of the DIC in this study originates from SOC degradation, therefore DIC losses has to be attributed to respiration losses and not DIC losses. Meaning that dissolution of CO₂ needs first mineralization of SOC by microorganism.

Response:

After considering both Reviewer #1 and #2's comments, we have removed this part to make this manuscript more rigorous and accurate.

Comment 15:

Technical corrections Fig. S4 and S5 are swapped, the caption of Fig. S4 belongs to Fig. S5 and vice versa.

Response:

Revised. Thank you!

Comparing soil carbon loss through respiration and leaching under extreme precipitation events in arid and semi-arid grasslands

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Abstract.

Respiration and leaching are two main processes responsible for soil carbon loss. While the former has received considerable research attention, studies examining leaching processes are limited especially in semiarid grasslands due to low precipitation. Climate change may increase the extreme precipitation event (EPE) frequency in arid and semiarid regions, potentially enhancing soil carbon loss through leaching and respiration. Here we incubated soil columns of three typical grassland soils from Inner Mongolia and Qinghai-Tibetan Plateau and examined the effect of simulated EPEs on soil carbon loss through respiration and leaching. EPEs induced transient increase of **CO₂ release through** soil respiration, equivalent to 32% and 72% of the net ecosystem productivity (NEP) in the temperate grasslands (Xilinhot and Keqi) and 7% in the alpine grasslands (Gangcha). By comparison, leaching loss of soil carbon accounted for 290%, 120% and 15% of NEP at the corresponding sites, respectively, with dissolved inorganic carbon (DIC) as the main form of carbon loss in the alkaline soils **regardless of DIC sources**. Moreover, **DIC loss increased with re-occurring EPEs in the soil with the highest pH due to elevated contribution of dissolved CO₂ from organic carbon degradation (indicated by DIC- $\delta^{13}\text{C}$)**. These results highlight that leaching loss of soil carbon (**particularly in the form of DIC**) is important in the regional carbon budget of arid and semiarid grasslands, **and also imply that SOC mineralization measured by CO₂ fluxes might be drastically under-estimated in alkaline soils**. With a projected increase of EPEs under climate change, soil carbon leaching processes and its influencing factors warrant better understanding and should be incorporated into soil carbon models when estimating carbon balance in grassland ecosystems.

1 Introduction

Soils store approximately 2500 Pg of carbon (including organic and inorganic carbon) globally, equivalent to 3.3 and 4.5 times the carbon in the atmosphere (760 Pg) and terrestrial plants (560 Pg), respectively (Lal, 2004). Slight variations of the soil carbon pool will hence severely influence atmospheric CO₂ concentrations and have important implications for climate change (Davidson and Janssens, 2006; Trumbore and Czimczik, 2008). Respiration and leaching are two main processes responsible for soil carbon loss. While respiration has received considerable research attention (Raich and Schlesinger, 1992; Raich and Potter, 1995; Hoover et al., 2016; Burri et al., 2015; Escolar et al., 2015), leaching is relatively poorly constrained despite its importance in certain ecosystems (Cole et al., 2007; Battin et al., 2008). For instance, soil carbon leached from forests, grasslands, and croplands is estimated to be 15.1, 32.4, and 20.5 g C m⁻² yr⁻¹ across Europe, representing 4%, 14%, and 8% of net ecosystem exchange (NEE), respectively (Kindler et al., 2011). Additionally, leaching of carbon previously preserved in surface litter and soil layers is believed to be a main source of dissolved organic and inorganic matter in inland waters (Spencer et al., 2008). In particular, soil inorganic carbon (SIC) that occurs widely in the arid and semiarid regions is more prone to leaching during sporadic high precipitation events (Lal and Kimble, 2000). Despite the importance of leaching loss in regional soil carbon budget, very few detailed data exist to investigate and compare the relative contribution of respiration and leaching processes to soil carbon loss.

Climate change is reported to increase the frequency as well as intensity of extreme precipitation events (EPEs; Knapp et al., 2002; Goswami et al., 2006; Parry et al., 2007; Min et al., 2011; Reichstein et al., 2013), especially in arid regions (Donat et al., 2017). In northwestern China, the frequency and intensity of EPEs have showed an increasing trend in the recent 50 years, constituting a much higher proportion of total precipitation than light precipitation events (Liu et al., 2005; Chen et al., 2012; Wang et al., 2012; Fu et al., 2013; Wang et al., 2014). Increasing EPEs will not only enhance soil carbon leaching but also affect soil respiration processes through increasing soluble substrates for microbial decomposition and potentially inducing hypoxic conditions. (Knapp et al., 2002; Harper et al., 2005; Morel et al., 2009; Unger et al., 2010). Hence, it is critical to evaluate the effects of EPEs on soil respiration and leaching processes in order to better understand the impact of climate change on terrestrial carbon cycling, especially in the arid and semiarid regions.

Grasslands, containing 20% of global soil carbon pool, are the most widespread ecosystems in arid and semiarid regions globally (Jobbagy and Jackson, 2000). The deposition rate of carbonate is relatively high in the grassland soils with a high alkalinity and aridity (Lal, 2008; Yang et al., 2012), and hence SIC is the major form of soil carbon in many grasslands (Mi et al., 2008). SIC storage in China is approximately 53.3–77.9 Pg (Li et al., 2007; Mi et al., 2008), 54% of which is mainly distributed in the temperate and alpine grasslands located in Inner Mongolia and Qinghai-Tibetan Plateau (Mi et al., 2008). From 1980s to 2000s, SIC in the topsoil of Chinese grasslands was estimated to decrease by 26.8 g C m⁻² yr⁻¹, mainly attributed to soil acidification (Yang et al., 2012). Alternatively, precipitation is one of the main factors influencing the distribution and storage of SIC in arid and semiarid regions (Batjes, 1998; Lal and Kimble, 2000). Mi et al. (2008) found that

84% of SIC in China was distributed in areas with a mean annual precipitation (MAP) of < 500 mm and that SIC content decreased significantly with the increase of MAP. Given the high leaching potential of SIC in grassland soils under altered precipitation patterns in the future, we hypothesize that EPEs may significantly enhance SIC loss through leaching processes and further reduce SIC storage in grasslands.

5 In this study, soils were collected from varied depths of three typical temperate and alpine grasslands in Inner Mongolia and Qinghai-Tibetan Plateau to construct soil columns for a laboratory incubation study. Using simulated EPEs, we examined soil carbon loss through respiration and leaching processes and compared their fluxes after EPEs. In addition, leaf litter of a C4 grass was added to the surface of one set of soil columns to compare soil carbon loss from bare versus litter-covered soils and to estimate the contribution of litter-derived carbon to soil respiration after EPEs. Our research objectives were: (1) to
10 investigate the influence of EPEs on soil respiration; (2) to quantify the loss of SIC and soil organic carbon (SOC) through leaching during EPEs; and (3) to compare the relative importance of respiration and leaching in EPE-induced soil carbon loss from grassland soils.

2 Materials and Methods

2.1 Study area

15 For the incubation experiment, soils were collected from three different sites of temperate and alpine grasslands of China with varied environmental characteristics. Temperate grasslands were sampled near Xilinhot (XLHT, 116°22' E, 44°8' N, mean elevation of 1170 m) and Keqi (KQ, 117°15' E, 43°18' N, mean elevation of 1250 m) within the arid and semiarid regions of Inner Mongolia (Fig. S1) with MAP of 299 and 402 mm and mean annual temperature (MAT) of 1.2 and 0.4°C, respectively. **Soil in this region is mainly chestnut soil, classified as Calcic Chernozems according to the World Reference
20 Base for Soil Resources (Steffens et al., 2008; IUSS working group WRB, 2015), with *Stipa klemenzii*, *Stipa Goboca*, *Stipa breviflora*, and *Stipa glareosa* as the dominating species (Sui and Zhou, 2013).** The alpine grassland was sampled in Gangcha (GC, 100°7' E, 37°19' N, mean elevation of 3500 m) located north of the Qinghai Lake on the northeastern edge of the Qinghai-Tibetan Plateau. The GC site has an MAT of 0.4°C, an MAP of 370 mm and a mean annual evaporation (MAE) of 607 mm. **Soils at this site are mainly Gelic Cambisol (IUSS working group WRB, 2015), with *Potentilla ansrina*
25 *Rosaceae*, *Elymus nutans* Griseb, and *Deyeuxia arundinacea* as the dominant species.**

Soils were collected by digging soil pits of 25 cm × 25 cm × 70 cm from the temperate (XLHT and KQ) and alpine (GC) sites in October, 2014 and August, 2015, respectively. At each site, three plots (200 m × 200 m) were selected (> 200 m in between) with three random soil pits (distance of ~ 5 m in between) sampled within each plot. Soils from the same depth (0–20, 20–40, and 40–60 cm) of the three soil pits were mixed *in situ* for each plot, shipped back to the laboratory immediately,
30 and stored at 4°C before the experiment started within one month. As a result, each sampling site had three “true” replicates from the field for the soil column experiment.

2.2 Soil column experiment and simulated EPEs

For the laboratory experiment, we reconstructed soil columns of similar structures and texture under controlled conditions and used gravity to collect soil leachates. This approach is commonly used in process-related research (Hendry et al., 2001; Thaysen et al., 2014; Ahmad and Walworth, 2009; Aslam et al., 2015) as it minimizes experimental errors and bias caused by unknown factors including soil heterogeneity and microbial community variations. It is also more favourable in terms of quantifying soil carbon leaching loss as it circumvents pore-water contamination by vacuum suction in the field. In particular, leachate sampling by gravity from soil columns prevents alterations to DIC concentrations, which may be caused by CO₂ outgassing using vacuum suction in field studies. Artificial soil columns were constructed in the laboratory with polymethyl methacrylate frames (diameter: 10 cm; height: 70 cm; Fig. 1). The bottom of each column had an aperture (inner diameter: 0.6 cm; height: 3 cm) for the collection of soil leachates, and the column top was fitted with an airtight lid connected to two tubes for gas exchange and collection. Empty columns were soaked in 0.1 mM hydrochloric acid (HCl) solutions for 12 h and rinsed with distilled water before use. Column bottoms were packed with pre-cleaned quartz sand (5-cm thick; soaked in 0.1 mM HCl and combusted at 450°C for 6 h before use) with a layer of nylon net (pore size: 150 μm; diameter: 10 cm) on both sides to prevent the movement of soil particles. Subsequently, soils were passed through 2-mm sieves with roots removed and packed into each column at the corresponding depths (in the sequence of 40–60, 20–40, and 0–20 cm). Soils were compacted gently to maintain a similar bulk density as in the field (Table 1). Water content of each soil layer was separately adjusted to 60% of the maximum water holding capacity (Table 1) to provide an ideal moisture condition for microbial growth (Howard and Howard, 1993; Rey et al., 2005). There was a 10-cm headspace unfilled with soil for each column.

Six soil columns (one litter-amended and one non-amended column for each of the three sampling plots) were set up for each site as described above, and pre-incubated for two weeks in the laboratory to allow the recovery of microbial communities after disturbance. Subsequently, leaf litter of a C4 grass, *Cleistogenes squarrosa*, a dominant species in the grasslands of northern China (Tian et al., 2015), was added to the surface of three columns in an amount equivalent to the aboveground biomass in the field (1.26 g for the XLHT and KQ sites and 1.59 g for the GC site; Bai et al., 2008). The isotopic signal of the leaf litter ($\delta^{13}\text{C}$ of -16.2‰) would allow us to estimate the contribution of litter-derived CO₂ to total soil respiration. The columns were pre-incubated again for seven days. Basal respiration rate was measured by collecting CO₂ gas in the column headspace after 4 h of incubation. Temperature was recorded every day during the whole incubation period ($23 \pm 1^\circ\text{C}$).

According to historical precipitation records (Fig. S2), more than 70% of the annual precipitation occurs from June to August in the study area, mainly in the form of 2-4 heavy precipitation events. Therefore, a total of three EPEs were simulated over a period of 2 months for each soil using artificial rainwater prepared according to rainwater's composition at the corresponding sites (pH of 7.3; Table S1; Tang et al., 2014; Zhang et al., 2013). A maximum rainfall intensity of ~100 mm per precipitation event has been recorded in the past two decades in the study area (Fig. S2) and is predicted to increase by 18.1% in the late 21st century in north China (Chen et al., 2012). Hence, approximately 1 L of rainwater (rainfall of ~127

mm), comparable to 30% of the MAP of the investigated sites, was added to the surface of each soil column over 3–4 h at rates of one drop per second using syringes and allowed to leach through the column to be collected with a clean beaker within 12–14 h. The leachates were weighed, filtered through a 0.45- μ m PTFE syringe filter and analyzed for dissolved organic carbon (DOC) and dissolved inorganic carbon (DIC) concentrations immediately. To monitor soil respiration every 1–2 days following each EPE, soil columns were first aerated for 1 h using CO₂-depleted air that had been passed through saturated sodium hydroxide (NaOH) solutions (twice; Fig. 1) and then incubated for 4 h with lids closed. CO₂ gas in the column headspace was collected by gas-tight syringes for the subsequent measurement. After collection of CO₂ gas, the lids were open to allow the exchange with the ambient air. Soil respiration was monitored for 30 days after the first EPE and observed to stabilize approximately on the 20th day (Fig. S3). Hence, the first, second, and third EPEs were conducted on the 1st, 31st, and 51st day of incubation, and the CO₂ measurement was conducted for approximately 30, 20, and 20 days after the first, second, and third EPEs, respectively. Basal respiration was considered to be represented by the stabilized respiration rate at the end of each EPE cycle.

2.3 Sample analyses

Soil pH was measured at a soil:water ratio of 1:2.5 (w:v) using a pH meter (Sartorius PB-10). Soil texture was examined by laser diffraction using Malvern Mastersizer 2000 (Malvern Instruments Ltd., UK) after removal of organic matter and calcium carbonates. Soil field water content was determined by difference between moist and dried soils (dried at 105°C for 8 h). Maximum water holding capacity was estimated by weighing soils before and after removal of redundant water from fully soaked soils (in water for 8 h). For SOC analysis, dried soils were decarbonated by exposure to concentrated HCl vapor for 72 h, followed by saturated NaOH solutions for 48 h to neutralize extra HCl, and then dried at 45°C. Total soil carbon, SOC (after decarbonation) and nitrogen (N) contents were measured by combustion using an elemental analyser (Vario EL III, Elementar, Hanau, Germany). SIC was calculated as the difference between total carbon and SOC contents. Small aliquots of the soil leachates were analyzed immediately on a Multi N/C 3100-TOC/TN Analyzer (Analytik Jena, Germany) for DIC and DOC concentrations (with the latter acidified to pH < 2 with concentrated HCl before analysis). It should be mentioned that the DIC concentration may be underestimated due to CO₂ outgassing during leachate collection. However, the potential underestimation is lower than 7% owing to the low proportion of outgassed CO₂ in total DIC (Table S2) as calculated using formulas in Ran et al. (2015). CO₂ concentration in the soil column headspace was determined by gas chromatograph (Agilent 7890A, USA) coupled with a flame ionization detector (FID).

To examine the contribution of SOC- and litter-derived carbon to soil respiration, the $\delta^{13}\text{C}$ values of SOC and CO₂ gas were determined on an isotope ratio mass spectrometer (Delta plus xp, Thermo, Germany) with a precision of $\pm 0.2\%$. To estimate the contribution of SOC degradation to leached DIC, the $\delta^{13}\text{C}$ values of DIC were determined on a Picarro isotopic CO₂ analyzer equipped with an automated DIC sample preparation system (AutoMate) based on wavelength scanned cavity ring down spectroscopy technique (Picarro AM-CRDS, USA). The precision for the DIC- $\delta^{13}\text{C}$ measurement was $\pm 0.3\%$. Due to

budget constraints and logistic reasons, we only measured the $\delta^{13}\text{C}$ of the respired CO_2 in the GC soils during the first EPE and the leached DIC in the XLHT soils.

2.4 Data analysis and statistics

5 The relative contribution of litter- and SOC-derived CO_2 to total respired CO_2 in the litter-amended soils was estimated using the following mass balance model:

$$f_{\text{litter-derived}} + f_{\text{SOC-derived}} = 1 \quad (1)$$

$$f_{\text{litter-derived}} \times \delta^{13}\text{C}_{\text{litter-derived}} + f_{\text{SOC-derived}} \times \delta^{13}\text{C}_{\text{SOC-derived}} = \delta^{13}\text{C}_{\text{respired-CO}_2} \quad (2)$$

10 where $f_{\text{litter-derived}}$ and $f_{\text{SOC-derived}}$ are the proportion of litter- and SOC-derived CO_2 in the total respired CO_2 ; $\delta^{13}\text{C}_{\text{litter-derived}}$ is the $\delta^{13}\text{C}$ value of litter-derived CO_2 , equivalent to -16.25% ; $\delta^{13}\text{C}_{\text{SOC-derived}}$ is the $\delta^{13}\text{C}$ value of SOC-derived CO_2 , which assumes the same value as that in the non-amended soils at the beginning of incubation (-23.1%) according to Cerling et al. (1991); $\delta^{13}\text{C}_{\text{respired-CO}_2}$ is the measured $\delta^{13}\text{C}$ of respired CO_2 .

Similarly, the relative contribution of lithogenic carbonate and biogenic DIC derived from SOC degradation to leached DIC was assessed according to the following isotopic mass balance model:

$$f_{\text{carbonate}} + f_{\text{biogenic-DIC}} = 1 \quad (3)$$

$$15 \quad f_{\text{carbonate}} \times \delta^{13}\text{C}_{\text{carbonate}} + f_{\text{biogenic-DIC}} \times \delta^{13}\text{C}_{\text{biogenic-DIC}} = \delta^{13}\text{C}_{\text{DIC}} \quad (4)$$

where $f_{\text{carbonate}}$ and $f_{\text{biogenic-DIC}}$ are proportion of carbonate- and biogenic DIC in total DIC; $\delta^{13}\text{C}_{\text{carbonate}}$ is the $\delta^{13}\text{C}$ value of soil carbonate, equivalent to 0% (Edwards and Saltzman, 2016); and $\delta^{13}\text{C}_{\text{biogenic-DIC}}$ is the $\delta^{13}\text{C}$ value of biogenic carbonate/bicarbonate derived from the dissolution of CO_2 produced by SOC degradation, which is estimated to shift by approximately 8% compared with the $\delta^{13}\text{C}$ value of soil-respired CO_2 (-24% here) due to isotope fractionation during CO_2 dissolution (Zhang et al., 1995). Hence, $\delta^{13}\text{C}_{\text{biogenic-DIC}}$ is estimated to be -16% . $\delta^{13}\text{C}_{\text{DIC}}$ is the measured $\delta^{13}\text{C}$ signature of leached DIC. According to Hendy (1971) and Doctor et al. (2008), isotopic fractionation of leached DIC due to CO_2 loss in an open system is insignificant when the partial pressure of CO_2 ($p\text{CO}_2$) in the solution is lower than twice that of the surrounding atmosphere. Therefore, due to the much lower $p\text{CO}_2$ in the XLHT leachates ($\sim 200 \mu\text{atm}$; Table S2) compared to that in the ambient atmosphere ($> 400 \mu\text{atm}$), the influence of CO_2 outgassing on the $\delta^{13}\text{C}$ of leached DIC was not considered in the present study.

EPE-induced CO_2 release via respiration was assessed following two steps. First, cumulative respiration during the first 20 days after each EPE (until respiration rate stabilized) was calculated. Second, difference between the measured cumulative respiration and that estimated using the stabilized basal respiration rate after each EPE was calculated as the EPE-induced CO_2 release.

30 Independent samples T test (group size = 2) and One-way ANOVA analysis (group size > 2) was used to compare the dissolved carbon concentrations and fluxes among different columns. Linear regression analysis was used to assess correlations between leachate carbon flux and influencing factors (carbon content, soil pH, soil texture, etc.). All these

analyses were performed using IBM SPSS Statistics 22. Differences and correlations are considered to be significant at a level of $p < 0.05$.

3 Results and Discussion

3.1 Bulk properties of grassland soil samples

5 In the investigated grassland soils, SOC represented 59–99% of soil carbon and exhibited $\delta^{13}\text{C}$ values typical of C3 plant inputs (ranging from -24.1‰ to -26.3‰ ; Table 1). The XLHT soil had a much lower SOC and nitrogen (N) contents than the KQ and GC soils despite a similar soil texture ($p < 0.05$; Table 1). The SOC:N ratio was also lowest in XLHT (7.09–8.03), indicating a more decomposed state of soil organic matter (Weiss et al., 2016). Conversely, the SIC content was highest in XLHT and lowest in KQ, mainly due to soil pH variations at these sites, i.e., lowest pH in KQ and highest in
10 XLHT. **This dependence of SIC content on soil pH is consistent with the results of Shi et al. (2012), showing that pH is the most important factor controlling SIC variation across the Mongolian and Tibetan grasslands.** In terms of depth variations, soils became coarser with depth in XLHT and GC but became finer in KQ. The SOC and N contents decreased with depth in all soils due to declining plant inputs ($p < 0.05$; Table 1), while the SOC:N ratio remained relatively similar (except a small decrease with depth in XLHT). By contrast, XLHT and GC soils showed an increasing SIC content with depth ($p < 0.05$;
15 Table 1), because SIC, with a good solubility, is prone to leaching from the topsoil and subsequently gets deposited in the deeper soil via salt formation (Mi et al., 2008; Tan et al., 2014). The KQ soil, showing an almost neutral pH, had an invariant SIC content and pH with depths. Overall, the varied properties (including SOC, SIC, pH, etc.) of these soils allowed us to compare the effects of EPEs on soil respiration and leaching processes in different grassland soils.

3.2 EPE-induced changes to soil respiration

20 Shortly after each simulated EPE, soil respiration was similar to or lower than basal respiration (Fig. S3). The latter case may be attributed to hypoxic conditions induced by water saturation during EPEs (Hartnett and Devol, 2003; Jessen et al., 2017). Subsequently, soil respiration increased and peaked after approximately one week due to the recovery of microbial activity with improved soil aeration (Borken and Matzner, 2009). It then decreased to a constant level approximately 20 days after each EPE (Fig. S3). The transient increase of respiration was consistent with the “Birch Effect” proposed by (Birch, 1964),
25 i.e., a pulse of soil respiration after rewetting events due to resuscitation of microorganisms and improved diffusive transport of substrate and extracellular enzymes (Borken and Matzner, 2009; Navarro-García et al., 2012; Placella et al., 2012). The maximum soil respiration rates were 40.6 and 37.3 $\text{mg C m}^{-2} \text{h}^{-1}$ after EPEs in the non-amended KQ and GC soils, respectively. These rates were significantly higher than that in the XLHT soil (13.7 $\text{mg C m}^{-2} \text{h}^{-1}$), likely related to the higher SOC content in the former soils. **The specific soil respiration rates normalized to SOC were 2.2, 2.6, and 7.9 $\mu\text{g C g}^{-1} \text{SOC}$**

h^{-1} in the GC, KQ, and XLHT soils, respectively. This indicated that SOC in the XLHT soils was easier to degrade despite its low content.

Total respired CO_2 was higher in the litter-amended than non-amended soils before and after EPEs (Fig. S3), likely caused by one or two following reasons: (1) the degradation of labile components in the fresh litter; (2) increased degradation of SOC primed by litter additions (Fröberg et al., 2005; Ahmad et al., 2013). These two reasons might affect CO_2 release in litter-amended soils in two ways: (1) both of them had positive effects; (2) one of them had positive effects and the other one had negative ones, but the total net effects was positive. To distinguish the influences of above two reasons on total respired CO_2 and further differentiate the contribution of litter (C4) and SOC (C3) to the respired CO_2 , we examined the $\delta^{13}\text{C}$ values of CO_2 evolved from the GC soils after the first EPE. On the first day after EPE, CO_2 from the non-amended and litter-amended GC soils had a $\delta^{13}\text{C}$ value of -23.1‰ and -18.7‰ , respectively. The latter was close to the $\delta^{13}\text{C}$ signature of the added litter (-16.25‰). Using the two-endmember mixing model of Eq. (1) and (2), we calculated that litter contributed $\sim 64\%$ of the respired CO_2 in the litter-amended GC soils. However, along with the consumption of labile carbon in litter, the $\delta^{13}\text{C}$ signature of CO_2 decreased from -18.7‰ on Day 1 to -21.8‰ on Day 25 after EPE in the litter-amended soils (Fig. 2). Accordingly, the proportion of litter-derived CO_2 decreased from 64% to 20%. The litter-derived CO_2 flux in litter-amended GC soils was estimated to range from 7.0 to 17.5 $\text{mg C m}^{-2} \text{h}^{-1}$, while the SOC-derived CO_2 flux increased from 6.2 to 15.7 $\text{mg C m}^{-2} \text{h}^{-1}$ after the first EPE (Fig. S3). Compared with the SOC-derived CO_2 flux in non-amended GC soils (ranging from 17.2 to 27.1 $\text{mg C m}^{-2} \text{h}^{-1}$), litter addition had a negative priming effect on the degradation of native SOC while increasing total respiration through labile litter degradation.

Using data shown in Fig. S3-4, we calculated that total EPE-induced CO_2 release during three EPEs was higher in the KQ and GC soils than in the XLHT soil ($p < 0.05$; Fig. 3a) with a lower SOC content and a lower SOC:N ratio (Table 1). However, the specific EPE-induced CO_2 release normalized to SOC content showed no significant difference in the non-amended soils among three sites (Fig. 3b), indicating that a similar proportion of SOC ($\sim 4\%$) was subject to EPE-induced CO_2 release in the alpine and temperate grassland soils. The total EPE-induced CO_2 release (including CO_2 from both litter and SOC mineralization) was significantly higher in the litter-amended KQ soils than the non-amended ones, similar to the specific EPE-induced CO_2 in the KQ and XLHT soils. The specific EPE-induced CO_2 was significantly different for the litter-amended soils among sites ($p < 0.05$), showing a pattern of $\text{KQ} > \text{XLHT} > \text{GC}$. This pattern was consistent with the mean sand content in the order of $\text{KQ} (46.9\%) > \text{XLHT} (39.1\%) > \text{GC} (27.2\%)$. In addition, the higher total and specific EPE-induced CO_2 release in the litter-amended KQ soils may be related to its relatively lower soil pH (~ 7.7), which facilitates the release rather than the dissolution of respired CO_2 in soil solution. We therefore conclude that the KQ soil, with a coarser texture and a lower pH (Table 1), may have provided less sorptive protection for the labile DOC components after EPEs (Kell et al., 1994; Nelson et al., 1994) and allowed less dissolution of the respired CO_2 , and hence showed a more responsive respiration to the precipitation events. These results suggest that SOC contents and SOC:N ratios are important factors influencing the total EPE-induced CO_2 release, while the availability of labile organic carbon, soil texture and pH are key factors affecting the specific EPE-induced CO_2 release in these grassland soils.

3.3 EPE-induced leaching of soil carbon

During three EPEs, a total of 0.57–0.71, 0.56–0.94, and 0.73–0.89 L of leachates were collected for the XLHT, KQ, and GC soils, respectively. DIC was the main form of carbon in the leachates from the alkaline soils with a high SIC content (XLHT and GC) but low from the KQ soil with a neutral pH and low SIC content (Fig. 4). The resulting DIC flux was much higher for the XLHT soils ($\sim 21.3 \text{ g C m}^{-2}$) than the other two (2.9 g C m^{-2} for KQ and 7.4 g C m^{-2} for GC soils) during three EPEs, equivalent to five times of its DOC flux ($3.8\text{--}4.2 \text{ g C m}^{-2}$, Fig. 4). In contrast, DIC flux in the KQ soils was only one third of its DOC flux during EPEs. The form of leached carbon was mainly linked to the amount of SOC and SIC in the columns (shown in Fig. S5).

Litter amendment did not increase DOC fluxes in any of the investigated soils but increased DIC fluxes leached from the KQ soil during the second and third EPEs and from the GC soil during the second EPE ($p < 0.05$, Fig. 4b-c). We postulate that, while litter contribution to DOC was minor, CO_2 derived from litter degradation contributed to dissolved CO_2 in soils and hence increased DIC in the leachates (Monger et al., 2015). This effect was not evident during the first EPE when litter decomposition just started and was not significant for the third EPE in the GC soil due to a high sample variability associated with the litter-amended soil (Fig. 4c). Due to the high SIC content in the XLHT soils (38.15 g per column) and the low litter-OC amendment (0.7 g per column), there was no significant difference of DIC fluxes between the non-amended and litter-amended XLHT soils (Fig. 4a). However, for the KQ soil having a relatively low SIC content which was similar to the added litter-OC (0.7 g per column; Table 1), the influence of litter addition on the DIC flux was quite obvious. Therefore, although the contribution of dissolved CO_2 to DIC fluxes should be more important in high-pH soils, the relative effect of litter amendment on DIC fluxes under EPEs seemed more significant in soils with a low SIC content.

Between different EPEs, leachate DOC fluxes did not vary in any of the investigated soils. By comparison, DIC fluxes increased in the XLHT soil from 4.5 g C m^{-2} after the first EPE to 9.0 g C m^{-2} after the third EPE ($p < 0.01$, Fig. 4). This increase may be caused by (i) an increased contribution of SOC degradation to soil DIC and/or (ii) an elevated dissolution of soil carbonates induced by higher soil CO_2 concentrations with repeated EPEs (Gulley et al., 2014; Ren et al., 2015). To evaluate these contributions, the $\delta^{13}\text{C}$ values of DIC were measured for the non-amended XLHT soil. The $\delta^{13}\text{C}$ of leached DIC ranged from -10.0‰ to -6.6‰ during the first EPE. Based on the isotopic mass balance of Eq. (3) and (4), lithogenic carbonate (with a $\delta^{13}\text{C}$ value of 0‰) contributed 51.2% to the leached DIC while biogenic DIC produced by SOC degradation contributed 48.4% (Fig. 5). The $\delta^{13}\text{C}$ value of leached DIC decreased to -12.3‰ and -13.5‰ during the second and third EPEs, corresponding to a contribution of 77.0% and 84.4% by biogenic sources in the total DIC, respectively (Fig. 5). These results confirm our previous hypothesis that SOC decomposition contributed significantly to soil DIC fluxes. Combined with the total flux rate, we calculated that both lithogenic and biogenic DIC fluxes were $\sim 2.1 \text{ g C m}^{-2}$ in the first EPE. Subsequently, lithogenic DIC flux decreased to $\sim 1.3 \text{ g C m}^{-2}$ while biogenic DIC flux increased to 7.6 g C m^{-2} in the third EPE. This demonstrates that the increased DIC flux with repeated EPEs was mainly derived from increased contribution of SOC mineralization. Interestingly, increasing DIC fluxes with repeated EPEs were not observed in the KQ

and GC soils (Fig. 4) despite their higher SOC contents and CO₂ release rates (Fig. S5). Given that the XLHT soil had the highest soil pH, the high alkalinity may have favored the retention of respired CO₂ in the soil solution compared with the other soils (Parsons et al., 2004; Yates et al., 2013; Liu et al., 2015), leading to its high contribution to DIC fluxes.

Regardless of its source, the EPE-induced leaching loss of inorganic carbon was 31.5 and 10.6 μg DIC g⁻¹ soil from the alkaline XLHT and GC soils, respectively, approximately three and five times higher than the corresponding DOC leaching loss (5.9 and 3.9 μg DOC g⁻¹ soil, respectively). However, the KQ soil had a relatively lower EPE-induced DIC loss (4.4 μg DIC g⁻¹ soil) than the DOC leaching loss (11.6 μg DOC g⁻¹ soil) mainly due to its lower initial SIC content and relatively neutral soil pH value. Hence, DIC was the main form of soil carbon loss in alkaline soils during EPEs regardless of its source. When the source of the leached DIC is taken into account, dissolution of CO₂ produced by SOC mineralization (biogenic DIC) constituted more than half of the leached DIC (at least from the XLHT soils; Fig. 5), whose contribution increased with re-occurring EPEs (Fig. 5). This implies that SOC mineralization measured by CO₂ fluxes was underestimated by approximately 8 times in the XLHT soils during the three EPEs (Fig. 5). In addition, DIC loss exclusively resulting from SIC dissolution or weathering was also a significant fraction of soil carbon loss, equivalent to 219% SOC loss in the form of CO₂ during EPEs (Fig. 5). These results collectively corroborate that inorganic carbon loss is the main form of soil carbon loss in alkaline soils during EPEs.

As for the influencing factors on soil carbon leaching loss, the DIC flux was positively correlated to the amount of SIC in the soil columns and soil pH ($p < 0.05$; Fig. 6a-b). These two relationships may be self-correlated due to a positive relationship between soil pH and SIC (Liu et al., 2016). By comparison, DOC flux was linked with the amount of SOC in the soil columns, but decreased with an increasing content of silt and clay ($p < 0.05$; Fig. 6c). This may be explained by the stronger retention of SOC on small-sized particles with more sorption sites (Barré et al., 2014; Mayer, 1994). Overall, total soil carbon loss through leaching under EPEs was positively related to soil pH values ($p < 0.05$; Fig. 6d), suggesting that soil pH is a critical factor determining the magnitude of soil carbon loss under EPEs.

3.4 Main pathways of grassland soil carbon loss under EPEs

In this study, EPE-induced soil carbon loss was composed of three parts: leachate DIC including lithogenic and biogenic DIC, leached DOC and EPE-induced CO₂ release through respiration. Regardless of the carbon sources, DIC and DOC fluxes accounted for 90%, 62%, and 68% of EPE-induced total loss at XLHT, KQ, and GC, respectively, representing the major pathway of soil carbon loss in these grassland soils under EPEs. Soil carbon leaching fluxes were 25.3, 10.4, and 10.1 g C m⁻² yr⁻¹ in XLHT, KQ, and GC soils during three EPEs, respectively, with DIC as the dominant form in XLHT and GC soils. While DIC fluxes in this study generally fell within the range reported for grassland soils (1.3–47.8 g C m⁻² yr⁻¹; Parfitt et al., 1997; Brye et al., 2001; Kindler et al., 2011), the XLHT soil had a DIC flux higher than the majority (> 50%) of the reported values (Fig. 7). This may be attributed to the higher SIC content and stronger dissolution of respired CO₂ in the XLHT soils due to its higher soil pH (9.1 ± 0.1) relative to other grassland soils (pH: 5.4–7.5; Kindler et al., 2011), and the high intensity of our simulated EPEs (precipitation: 40 mm h⁻¹). Nonetheless, DIC fluxes in grassland soils reported in this

study and elsewhere (Brye et al., 2001; Kindler et al., 2011) were significantly higher than in forest and cropland ecosystems ($p < 0.05$; Rieckh et al., 2014; Lentz and Lehrs, 2014; Gerke et al., 2016; Herbrich et al., 2017; Siemens et al., 2012; Walmsley et al., 2011; Wang and Alva, 1999; Kindler et al., 2011), highlighting the importance of leaching as a major pathway of soil carbon loss in grasslands. By contrast, DOC fluxes in this study ($4.8 \pm 2.5 \text{ g C m}^{-2}$) were lower than most of the reported values in forest and grassland ecosystems due to the low SOC contents in our soils (Fig. 7).

Net ecosystem production (NEP) in the temperate steppe of Inner Mongolia (XLHT and KQ) is $8.7 \text{ g C m}^{-2} \text{ yr}^{-1}$ (Sui and Zhou, 2013). While the EPE-induced CO_2 release (2.8 ± 0.6 and $6.3 \pm 3.0 \text{ g C m}^{-2}$) accounted for 32% and 72% of the NEP at XLHT and KQ, respectively; soil carbon leached during three EPEs was equivalent to 290% and 120% of NEP, with total DIC loss accounting for 244% and 33%, respectively. It is worth mentioning that biogenic DIC loss ($16.0 \pm 3.4 \text{ g C m}^{-2}$) caused by SOC degradation accounted for 184% of NEP at XLHT, indicating the importance of biogenic DIC to leached inorganic carbon loss during EPEs. By comparison, NEP in the studied alpine grassland ($68.5 \text{ g C m}^{-2} \text{ yr}^{-1}$; Fu et al., 2009) is much higher than in typical temperate steppe. Hence, soil carbon loss through leaching and respired CO_2 release accounted for 15% (DIC: 11%, DOC: 4%) and 7% of the NEP at GC, respectively. Nonetheless, the EPE-induced soil carbon loss relative to NEP was higher in this study than that estimated for grassland topsoil across Europe (12% for DIC loss, 2% for DOC loss; Kindler et al., 2011) where Net Ecosystem Exchange (NEE) reported by Kindler et al. was used as NEP according to the report of Kirschbaum et al. (2001). This was partially attributed to the lower NEP and higher SIC content in XLHT and KQ soils, underscoring that soil carbon leaching is more important in fragile ecosystems with low productivity.

An uncertainty related to the importance of leaching processes in the overall carbon budget along the “soil-river-ocean” continuum lies in the ultimate downstream fate of the leached carbon. If part of this carbon is retained in the surrounding soils or carried along from the river to the ocean in the form of DIC without outgassing into the air, it will not constitute a source of atmospheric CO_2 on a relatively short term (over years or decades). However, soil columns used in our study has a depth (60 cm) typical of or even deeper than the average soil depth in the alpine grasslands of Qinghai-Tibetan Plateau (Wang et al., 2001). Hence, we assume that carbon leached in our experiments will have minimum retention in the soil. Furthermore, compared to DOC and DIC in the soil solution, the leached carbon is more likely to be subject to more intensified mineralization and outgassing during the land-ocean transfer, given more intensified mixing processes, oxygen exposure and photo-oxidation of terrestrial carbon upon releasing into the river (Hedges et al., 1997; Battin et al., 2009). Hence, we postulate that carbon leached from soils is more vulnerable to decomposition and/or release compared to that retained in the soil. That being said, it will be necessary to confirm our results and hypothesis using field-based leaching experiments to better understand the ultimate fate of leached soil carbon: whether it will be retained in the deeper soil or show a higher degradability upon leaving the soil matrix. Such information will be complementary to our study and further elucidate the importance of leaching processes in terms of ecosystem carbon budget.

In summary, this study quantified and compared soil carbon loss through respired CO_2 release and leaching in three typical grassland soils of northern China under simulated EPEs. Soil CO_2 release was stimulated shortly after each EPE, leading to an EPE-induced CO_2 release equivalent to 32% and 72% of the NEP at XLHT and KQ (temperate grasslands) and 7% at GC

(alpine grassland). By comparison, [regardless of the carbon sources](#), soil carbon leaching fluxes accounted for 290%, 120% and 15% of the NEP at XLHT, KQ, and GC, respectively, with DIC as the main form of carbon loss in the SIC-enriched XLHT and GC soils. [In view of DIC sources, biogenic DIC loss derived from SOC mineralization contributed to more than half of the total leached DIC fluxes and accounted for 184% of the NEP at XLHT](#). Moreover, DIC loss increased with re-occurring EPEs in the XLHT soil with the highest pH due to increased dissolution of soil carbonates as well as elevated contribution of dissolved CO₂ from SOC degradation. [These results also imply that SOC mineralization measured by CO₂ fluxes might be drastically under-estimated in alkaline grassland soils during EPEs](#). Admittedly, our results are based on artificial soil columns which destroyed natural soil structures, hence potentially increasing the contact between pore water and soil particles through eliminating macropore structures and preferential flow (Seyfried et al., 1987; Singh et al., 1991). Hence, our estimate may represent an upper limit of soil carbon leaching potential under EPEs. Nonetheless, these results highlight that leaching loss of soil carbon, [especially in the form of DIC originated from biogenic and lithogenic carbonates](#), plays an important role in the regional carbon budget of grasslands located in arid and semiarid regions. Further research effort is needed to combine short-term laboratory experiments with long-term field measurements to fully assess the impacts of EPEs on soil carbon budget in these areas. In addition, with a projected increase of EPEs under climate change, soil carbon leaching processes and its influencing factors warrant better understanding and should be incorporated into soil carbon models when estimating carbon balance in grassland ecosystems.

Data availability. All data is available within this paper (Table 1) and in the Supplement (Dataset S1).

Competing interests. The authors declare that they have no conflict of interest.

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Table 1: Bulk properties of soil samples collected from the temperate and alpine grasslands for the soil column experiment (mean \pm standard error; n = 3).

Station	Depth (cm)	SOC (%)	SIC (%)	N (%)	SOC:N ratio	pH	$\delta^{13}\text{C}$ (‰)	FWC (%)	Max WHC (%)	BD (g cm^{-3})	Clay (%)	Silt (%)	Sand (%)
Xilinhot (XLHT)	0–20	1.48 \pm 0.02	0.41 \pm 0.01	0.18 \pm 0.00	8.03 \pm 0.18	8.98 \pm 0.03	–24.1	10.65 \pm 0.11	47.12 \pm 0.37	1.06 \pm 0.02	0.4	64.6	35.0
	20–40	1.00 \pm 0.05	0.64 \pm 0.00	0.13 \pm 0.00	7.69 \pm 0.22	9.09 \pm 0.01	–24.1	6.48 \pm 0.24	44.92 \pm 0.25	1.24 \pm 0.05	0.5	58.2	41.3
	40–60	0.67 \pm 0.03	1.05 \pm 0.01	0.09 \pm 0.00	7.09 \pm 0.22	9.09 \pm 0.04	–23.7	5.56 \pm 0.11	39.78 \pm 0.39	1.31 \pm 0.03	0.6	58.5	41.0
Keqi (KQ)	0–20	3.36 \pm 0.05	0.02 \pm 0.00	0.29 \pm 0.00	11.48 \pm 0.24	7.79 \pm 0.10	–26.0	19.59 \pm 0.22	65.57 \pm 0.82	1.14 \pm 0.03	0.4	41.0	58.6
	20–40	2.52 \pm 0.04	0.01 \pm 0.00	0.22 \pm 0.00	11.59 \pm 0.27	7.63 \pm 0.04	–25.9	8.56 \pm 0.05	53.59 \pm 1.98	1.22 \pm 0.01	0.2	55.7	44.1
	40–60	1.65 \pm 0.03	0.02 \pm 0.00	0.14 \pm 0.00	11.49 \pm 0.42	7.57 \pm 0.12	–25.5	8.00 \pm 0.27	42.92 \pm 0.57	1.19 \pm 0.01	0.2	61.6	38.1
Gangcha (GC)	0–20	3.32 \pm 0.23	0.34 \pm 0.04	0.31 \pm 0.03	10.70 \pm 1.28	8.53 \pm 0.07	–26.3	33.24 \pm 0.68	60.79 \pm 0.21	n.d.	1.3	75.9	22.8
	20–40	2.90 \pm 0.18	0.44 \pm 0.10	0.29 \pm 0.01	9.93 \pm 0.69	8.60 \pm 0.03	–24.0	36.15 \pm 0.52	62.03 \pm 0.30	n.d.	0.9	75.8	23.3
	40–60	2.12 \pm 0.22	0.52 \pm 0.06	0.20 \pm 0.02	10.55 \pm 1.50	8.76 \pm 0.10	–25.3	35.79 \pm 0.91	62.85 \pm 0.61	n.d.	0.6	64.0	35.4

SOC: soil organic carbon; SIC: soil inorganic carbon; N: nitrogen; FWC: field water content; Max WHC: maximum water holding capacity; BD: bulk density; Clay: soil particle size $<$ 0.2 μm ; Silt: 0.2 μm $<$ soil particle size $<$ 20 μm ; Sand: soil particle size $>$ 20 μm ; n.d.: not determined.

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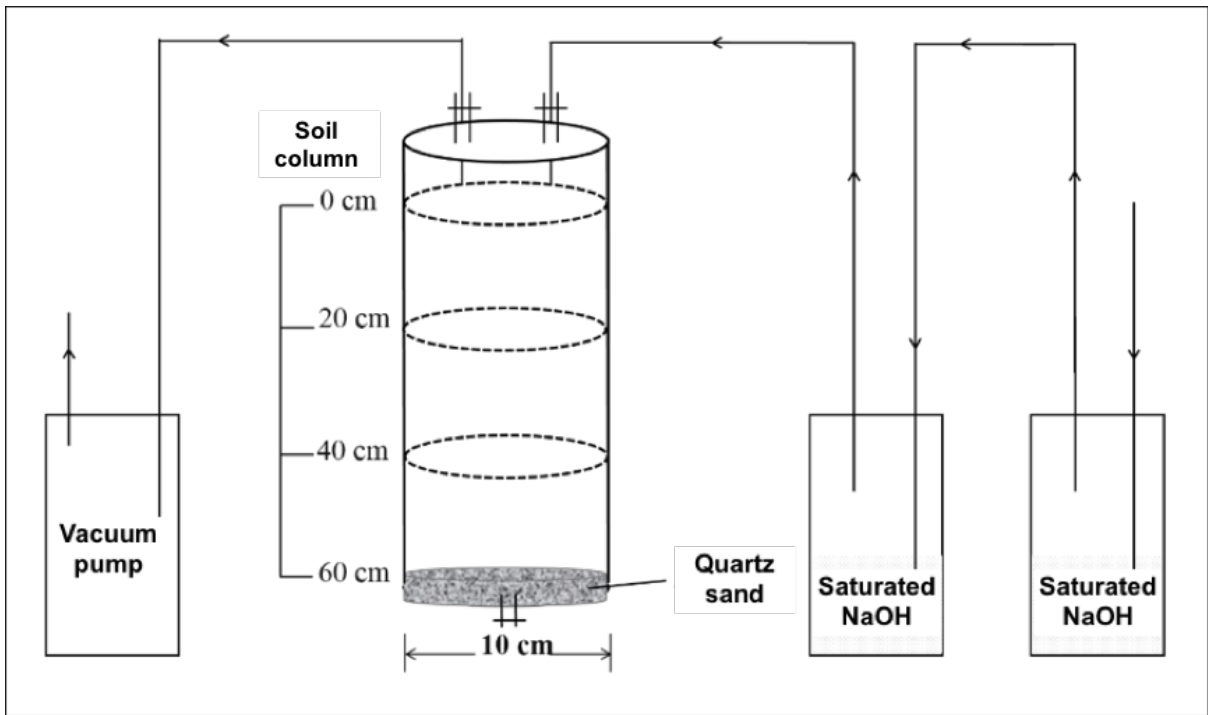


Figure 1: Design of the soil column experiment for monitoring soil respiration and leaching after simulated extreme precipitation events (EPEs).

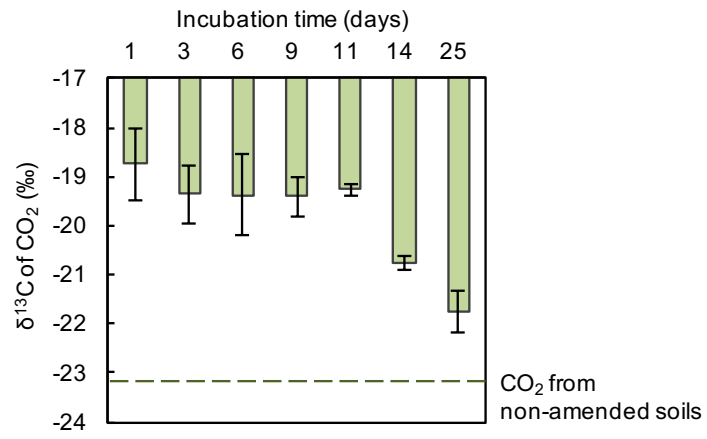


Figure 2: The $\delta^{13}\text{C}$ values of respired CO_2 in the litter-amended Gangcha (GC) soils after the first extreme precipitation event (EPE). Mean values are shown with standard error ($n = 3$).

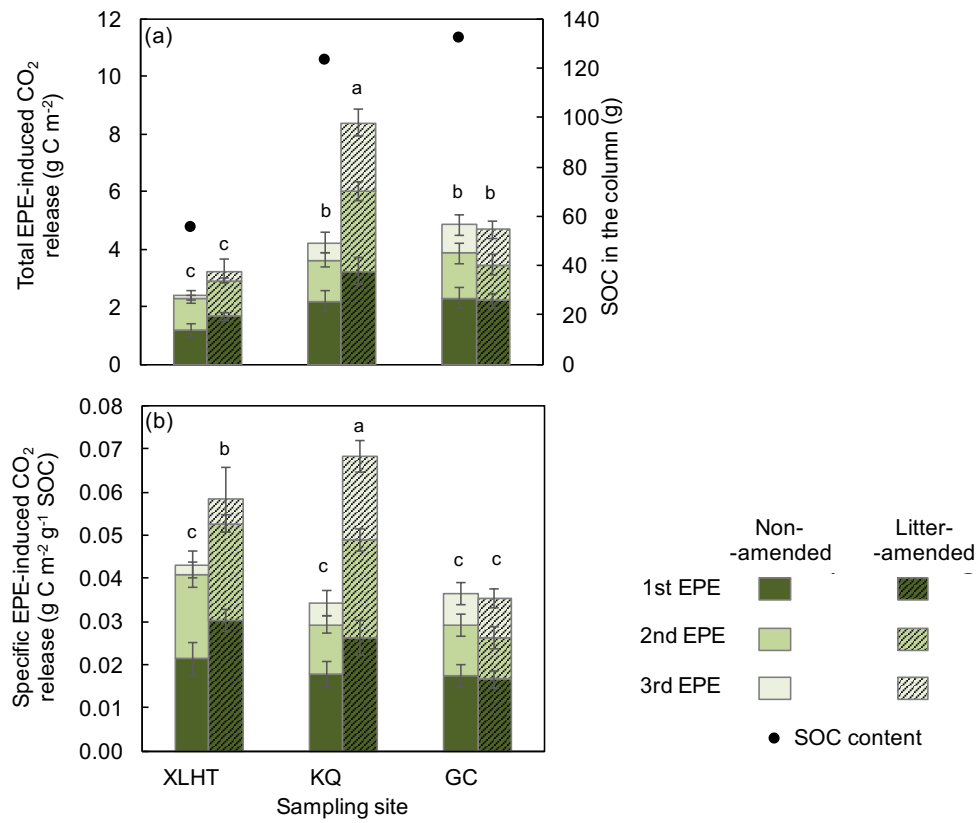


Figure 3: Total (a) and specific (b) extreme precipitation event (EPE)-induced CO₂ release in the litter-amended and non-amended grassland soils during three EPEs. Mean values are shown with standard deviation (n = 3). Lower-case letters (a, b, c) indicate significantly different levels among the litter-amended and non-amended soils determined by Duncan's multiple range test (one-way ANOVA, $p < 0.05$).

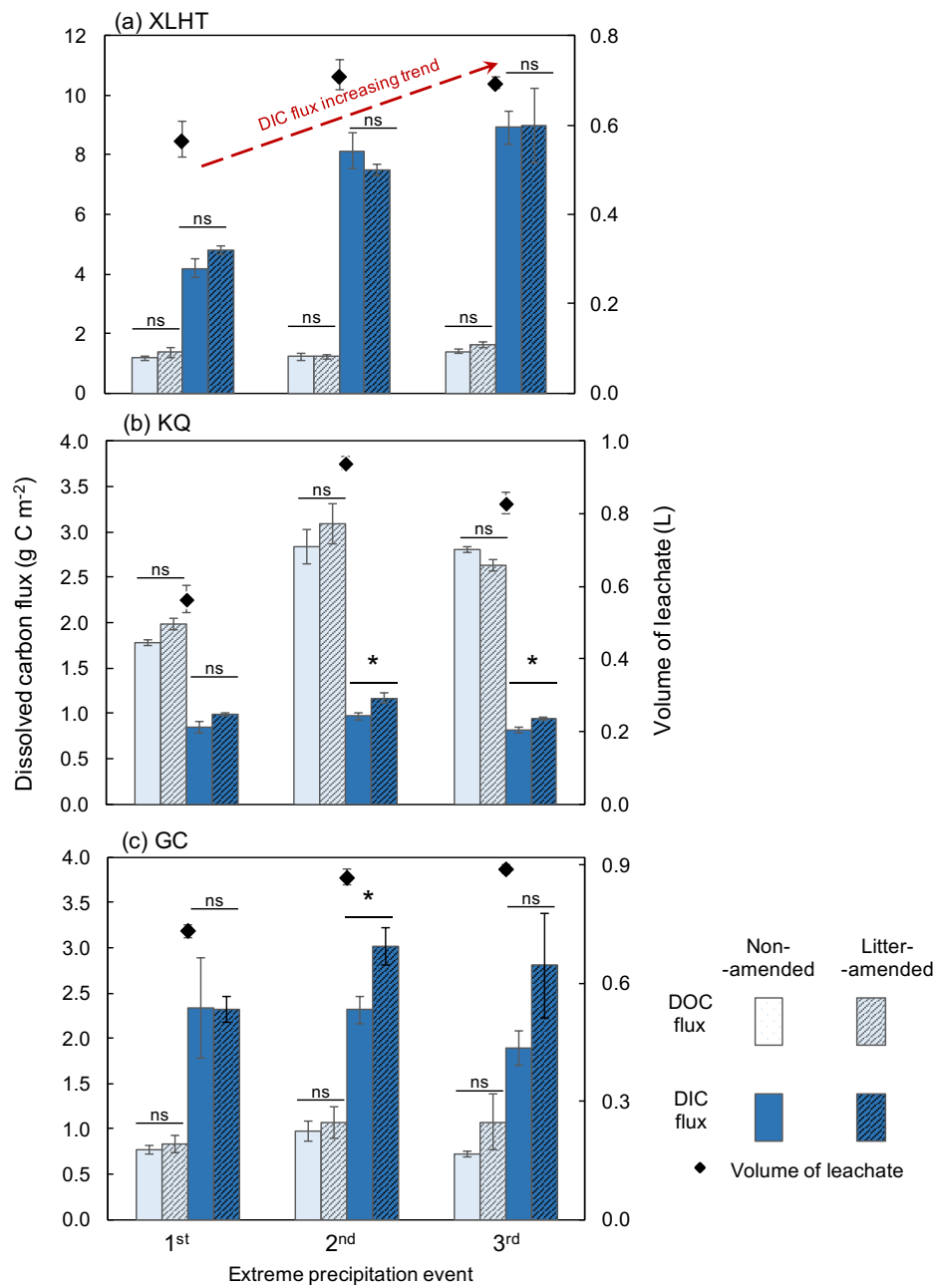


Figure 4: Fluxes of dissolved organic carbon (DOC) and dissolved inorganic carbon (DIC) and volume of leachates from soil columns after extreme precipitation events (EPEs). Mean values are shown with standard error ($n = 3$). * and ns denote significant and no difference between the litter-amended and non-amended soils determined by independent samples T test, respectively ($p < 0.05$).

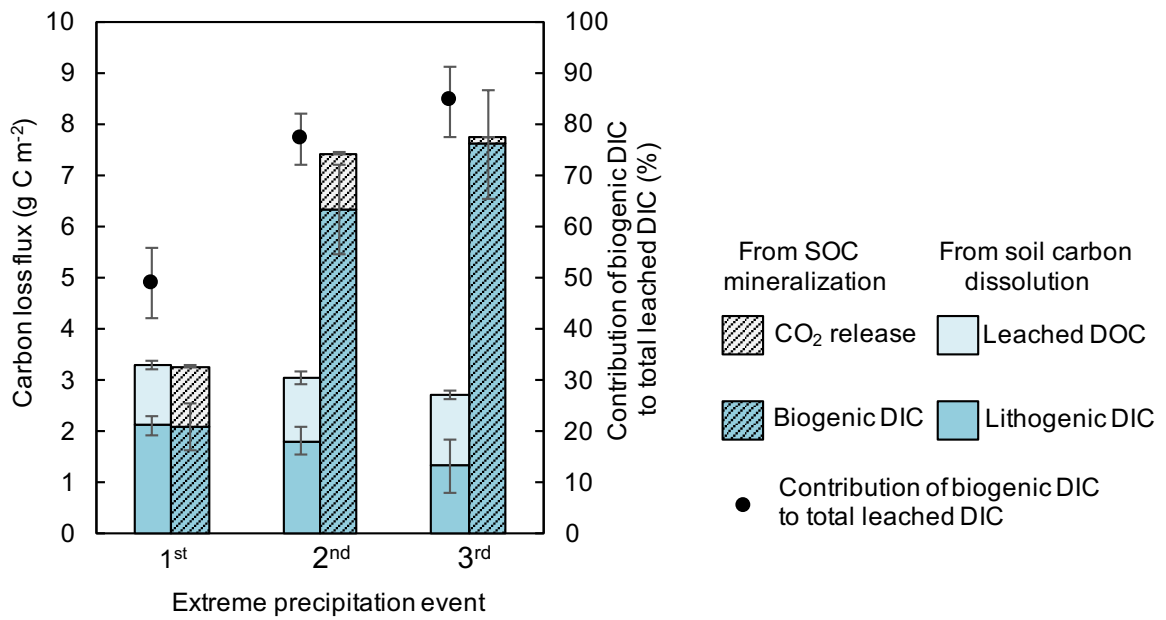
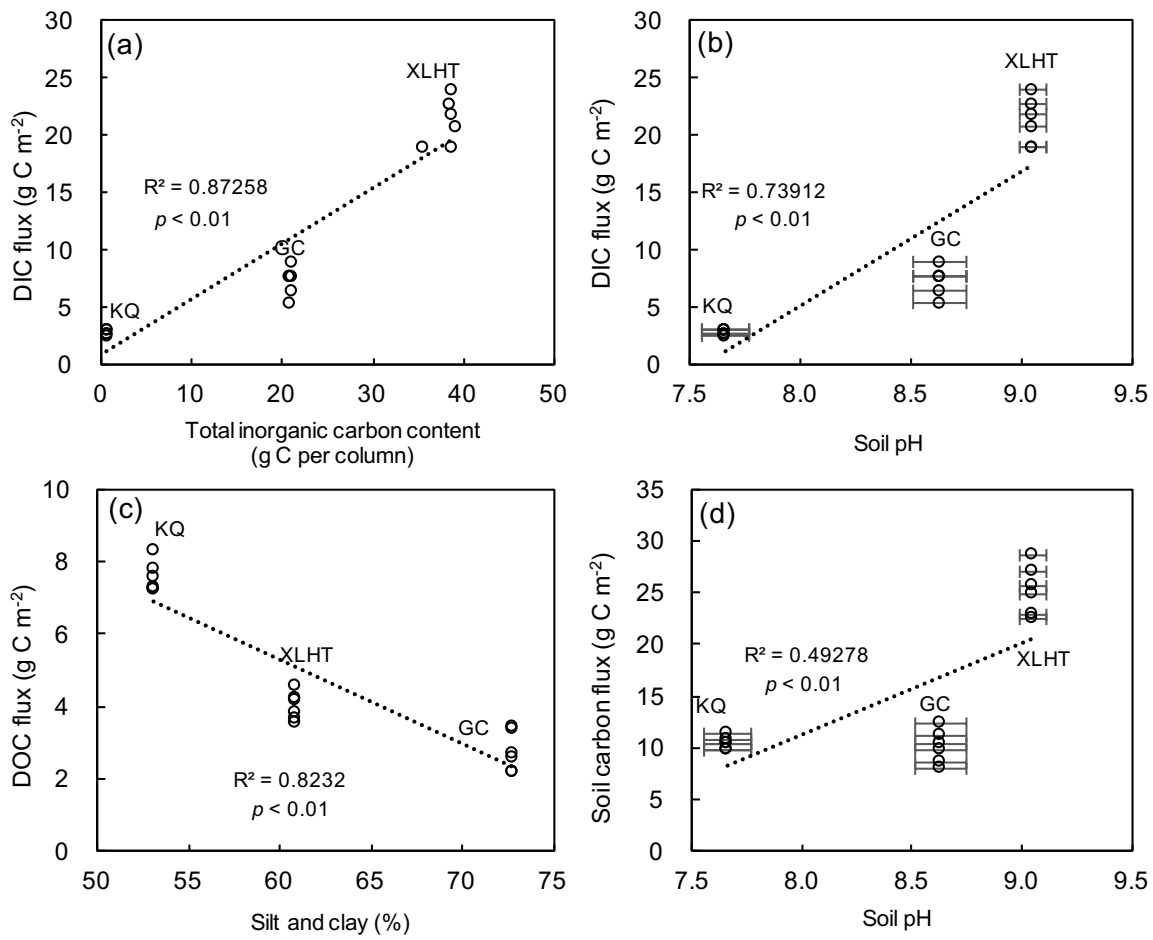


Figure 5: The flux of carbon loss from soil organic carbon (SOC) mineralization including CO₂ release and biogenic dissolved inorganic carbon (DIC), and that from soil carbon dissolution including leached dissolved organic carbon (DOC) and lithogenic DIC in the XLHT soils. Mean values are shown with standard error (n = 3).

5



5 **Figure 6: Relationship of dissolved inorganic carbon (DIC) and dissolved organic carbon (DOC) fluxes with soil properties: (a) DIC flux with total inorganic carbon in the soil columns; (b) DIC flux with soil pH, (c) DOC flux with silt and clay content of soils, (d) total soil carbon flux with soil pH. Mean pH values are shown with standard error ($n = 3$).**

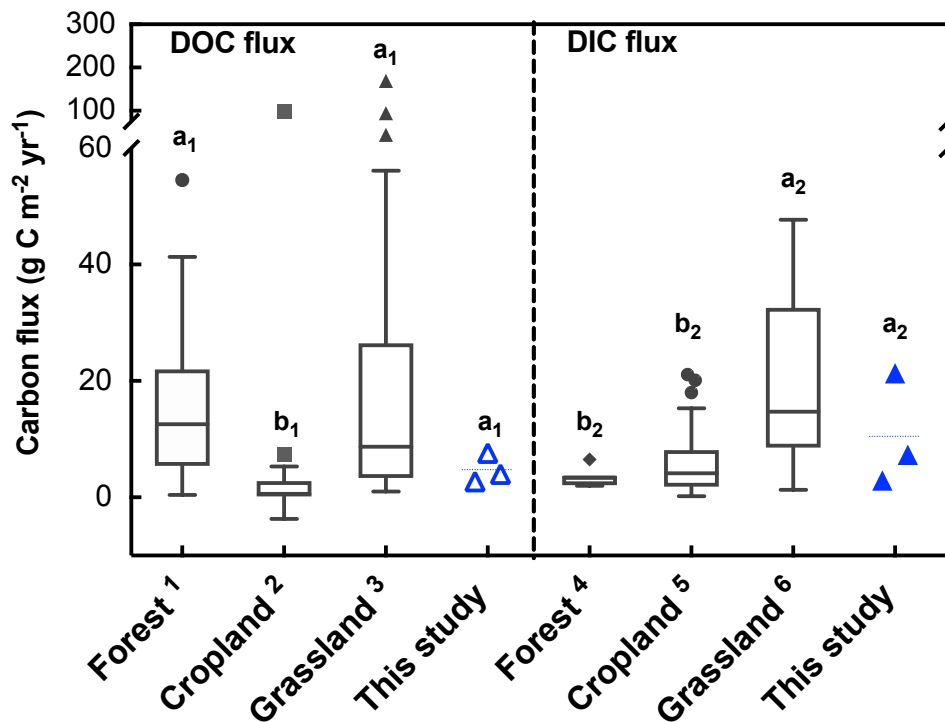


Figure 7: Leaching fluxes of dissolved organic carbon (DOC) and dissolved inorganic carbon (DIC) in this study compared with that reported in the literature. ¹n = 110, data from Brooks et al., 1999; Froberg et al. 2005, 2006, 2011; Gielen et al., 2011; Kindler et al., 2011; Lu et al., 2013; Michalzik et al., 2000; Sanderman et al., 2008; ²n = 33, data from Brye et al., 2001; Kindler et al., 2011; Siemens et al., 2012; Walmsley et al., 2011; Wang and Alva, 1999; Gerke et al., 2016; Herbrich et al., 2017; Rieckh et al., 2014; Lenz, 2014; ³n = 46, data from Brooks et al., 1999; Brye et al., 2001; Ghani et al., 2010; Kindler et al., 2011; Mctiernan et al., 2001; Parfitt et al., 2009; Sanderman et al., 2008; Tipping et al., 1999; ⁴n = 8, data from Kindler et al., 2011; ⁵n = 32, data from Kindler et al., 2011; Siemens et al., 2012; Walmsley et al., 2011; Wang and Alva, 1999; Gerke et al., 2016; Herbrich et al., 2017; Rieckh et al., 2014; Lenz, 2014; ⁶n = 9, data from Brye et al., 2001; Kindler et al., 2011. Lower-case letters (a₁, b₁) and (a₂, b₂) represent significant different levels of DOC and DIC fluxes in different ecosystems determined by Duncan's multiple range test, respectively, (one-way ANOVA, *p* < 0.05). Dash lines represent mean values for the investigated soils.

Supplementary Material for

Comparing soil carbon loss through respiration and leaching under extreme precipitation events in arid and semi-arid grasslands

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Contents of this file

Table S1 to S2

Figure S1 to S5

15 **Additional Supporting Information (Files upload separately)**

Dataset S1

Introduction

The supplementary material contains Table S1-S2, Figures S1-S5, and Dataset S1. Table S1 presents the major ion concentrations of the artificial rainwater used for the simulated extreme precipitation events (EPEs). Table S2 shows
20 proportion of outgassed CO₂ in total dissolved inorganic carbon under given pH, temperature and alkaline conditions. Fig. S1 provides the locations of sample sites and their mean annual precipitation (MAP) levels. Fig. S2 provides daily and monthly precipitation in Xilinhot. Fig. S3 shows soil respiration rate variation after EPEs. Fig. S4 provides information on soil carbon content in each column. Fig. S5 provides information on cumulative respiration in soils of different treatments. Dataset S1 (separate file) shows data displayed in the figures of main text and supporting information.

Table S1: Major ion concentrations ($\mu\text{mol L}^{-1}$) of the artificial rainwater used for the simulated extreme precipitation events (EPEs).

Ion	For XLHT and KQ soils	For GC soils
Ca^{2+}	113	180
Mg^{2+}	17	37
$\text{Na}^{+} + \text{K}^{+}$	41	87
HCO_3^{-}	180	12
Cl^{-}	45	75
SO_4^{2-}	161	67

5

Table S2: Proportion of outgassing CO₂ in total dissolved inorganic carbon under given pH, temperature and alkaline conditions according to formulas shown in Ran et al. (2015).

pH ^a	T ^b	Alkaline ^c ($\mu\text{mol/l}$)	Proportion of outgassing CO ₂ (%)	Partial Pressure of CO ₂ (μatm)
9.0	23	300	0.2	18
8.5	23	300	0.7	59
7.5	23	300	6.8	601
9.0	23	500	0.2	29
8.5	23	500	0.7	98
7.5	23	500	6.8	1002
9.0	23	1000	0.2	58
8.5	23	1000	0.7	195
7.5	23	1000	6.8	2004
9.0	23	2000	0.2	117
8.5	23	2000	0.7	391
7.5	23	2000	6.8	4008
9.0	23	3000	0.2	175
8.5	23	3000	0.7	586
7.5	23	3000	6.8	6013

^a: pH of the soil leachates in this study, pH_{XLHT} ~ 9, pH_{KQ} ~ 7.6, pH_{GC} ~ 8.6;

^b: incubation temperature;

^c: alkaline range of natural waters.

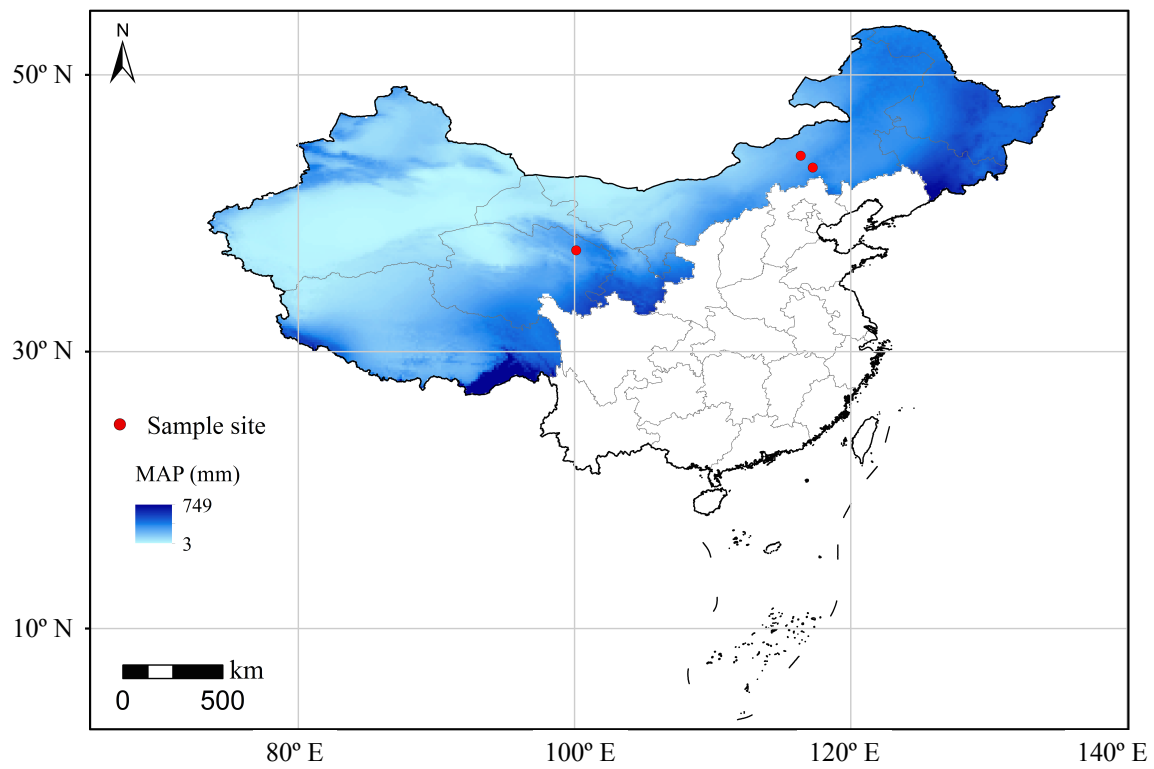


Figure S1: Map of sampling sites and their mean annual precipitation (MAP) levels.

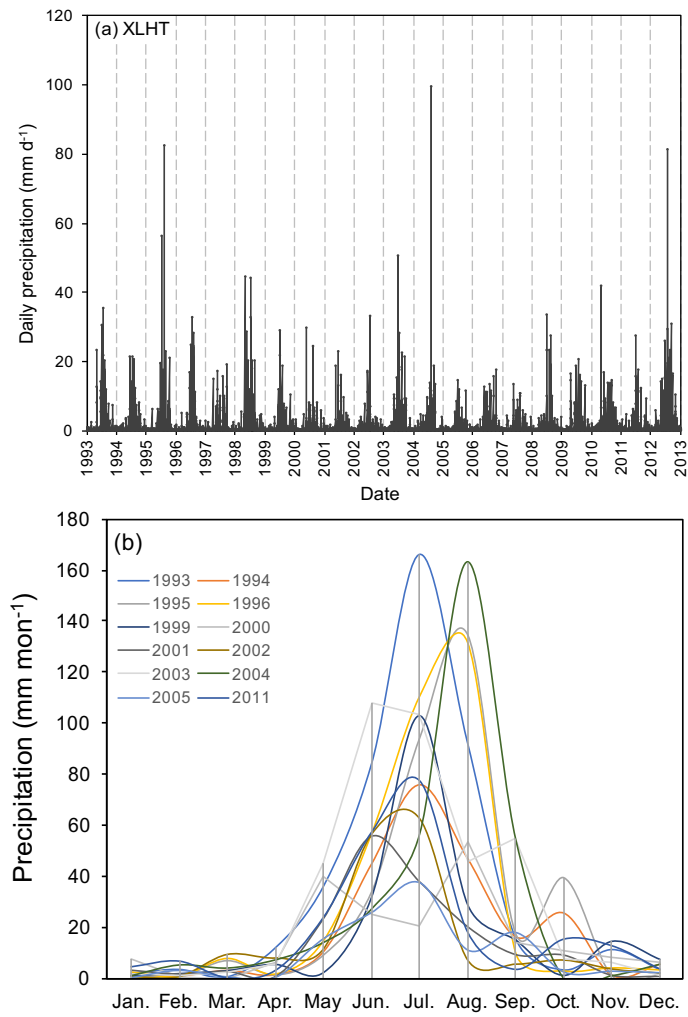
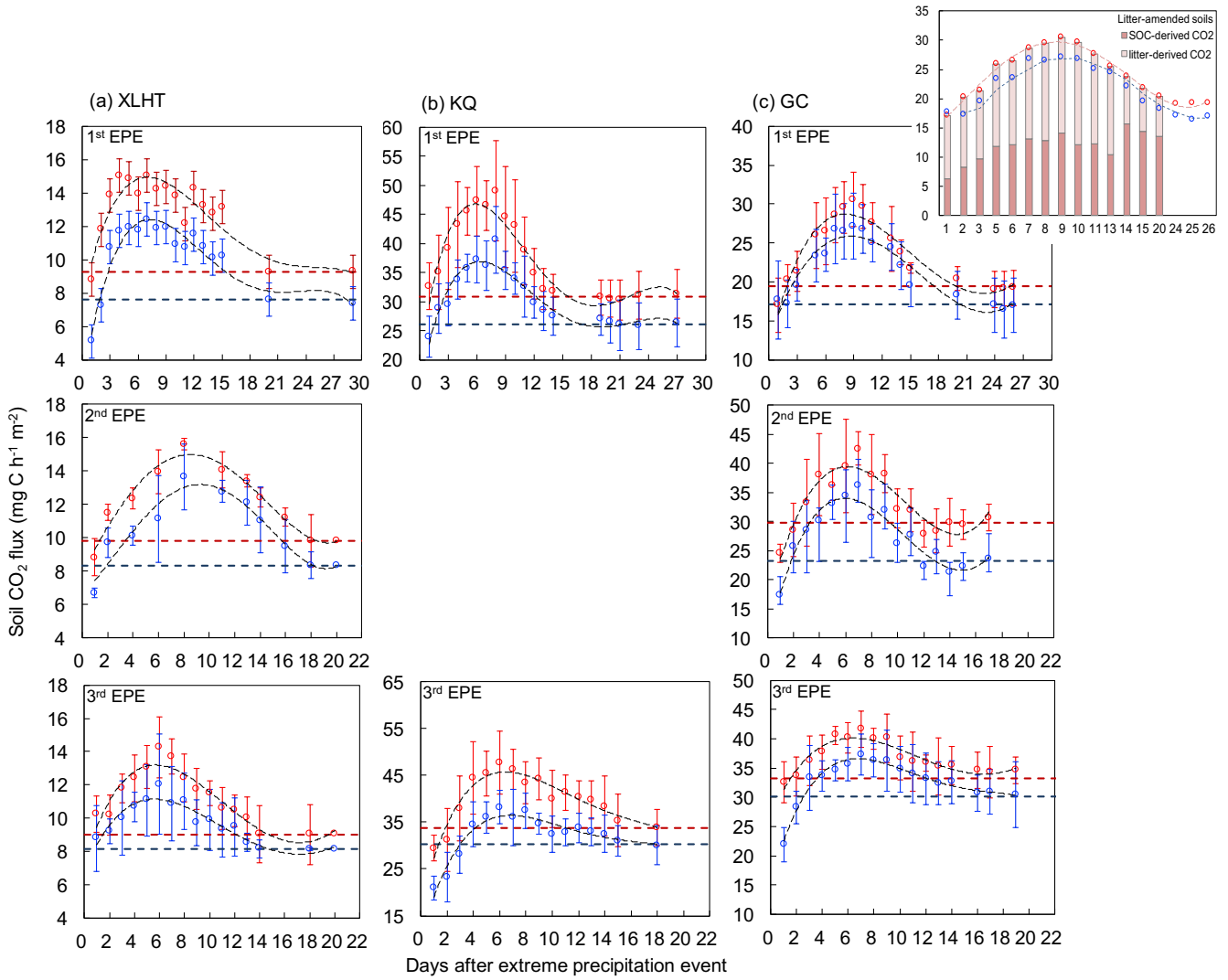


Figure S2: Variation of the daily precipitation (a) and monthly precipitation (b) during recent two decades at Xilinhot in Inner Mongolia (data modified from <http://data.cma.cn/data/index/6d1b5efbdcfb9a58.html>).



5 **Figure S3: Variations of soil CO₂ release rate after the simulated extreme precipitation events (EPEs). The inside-figure shows the variations of litter-derived and SOC-derived CO₂ release with time in litter-amended GC soils during the first EPE. Red and blue dots represent respiration in the litter-amended and non-amended soils, respectively. Mean values are shown with standard error (n = 3). Red and blue dash lines represent the stabilized respiration rate after EPEs in the litter-amended and non-amended soils, respectively. Black dash lines represent regressive lines using polynomial fitting method.**

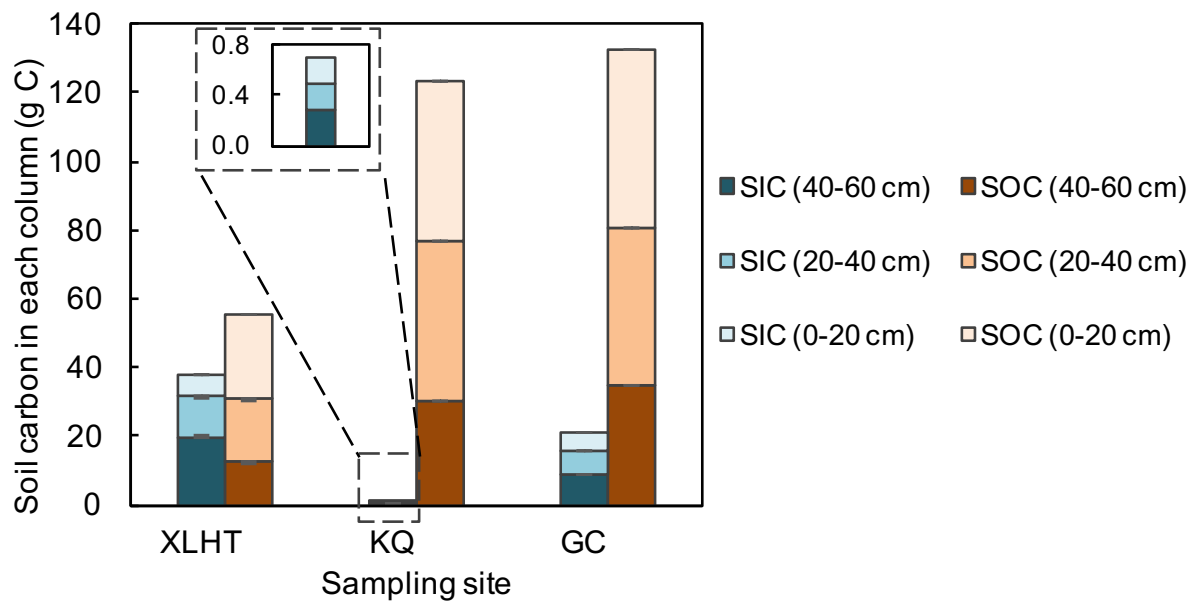
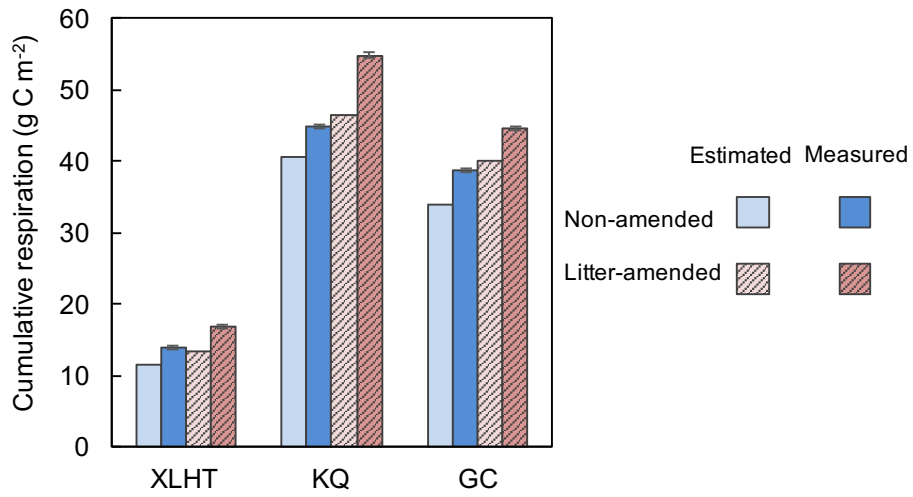


Figure S4: Total amount of soil organic carbon (SOC) and soil inorganic carbon (SIC) in different layers of the artificial soil columns.



5 **Figure S5: Cumulative respiration in the non-amended and litter-amended soils during the first 20 days after three extreme precipitation events (EPEs) relative to that estimated using basal respiration rates stabilized after EPEs. Mean values are shown with standard error (n = 3).**