

Article

Effects of Nitrogen and Water Addition on Ecosystem Carbon Fluxes in a Grazing Desert Steppe

Chao Wen ¹, Jianhui Huang ^{2,3}, Yumei Shan ^{1,*}, Ding Yang ¹, Lan Mu ¹, Pujin Zhang ¹, Xinchao Liu ¹, Hong Chang ¹ and Ruhan Ye ¹

¹ Inner Mongolia Academy of Agricultural and Animal Husbandry Sciences, Hohhot 010031, China; wenchao2000@163.com (C.W.); yangding81@126.com (D.Y.); mulan6202@163.com (L.M.); zhangpujin0819@163.com (P.Z.); liuxinchao3211@126.com (X.L.); chang-h11@163.com (H.C.); yiruhan1981@163.com (R.Y.)

² State Key Laboratory of Vegetation and Environmental Change, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China; jhhuang@ibcas.ac.cn

³ University of Chinese Academy of Sciences, Beijing 100049, China

* Correspondence: shanym2008@163.com

Abstract

Desert steppe ecosystems, characterized by water limitation and high sensitivity to global climate change and anthropogenic disturbance drivers, experience profound alterations in carbon (C) cycling processes driven by the multiplicative interactions among grassland grazing, altered precipitation regimes, and elevated atmospheric nitrogen deposition. However, how historical grazing legacies modulate ecosystem responses to concurrent changes in nitrogen deposition and precipitation regimes remains poorly resolved. To address this, we conducted a field experiment manipulating water and nitrogen addition across grazing intensities (no grazing, light grazing, moderate grazing, heavy grazing) in a *Stipa breviflora* desert steppe. Over three consecutive growing seasons (2015–2017), we continuously monitored net ecosystem CO₂ exchange (NEE), ecosystem respiration (ER), and gross ecosystem production (GEP) to quantify ecosystem CO₂ fluxes under these interacting global change drivers. Results revealed that water and nitrogen addition did not alter seasonal CO₂ flux dynamics across grazing intensities. Light grazing enhanced ecosystem C sink capacity, whereas heavy grazing reduced NEE and GEP, diminishing C sink strength. Water addition significantly increased CO₂ fluxes, strongly correlated with soil moisture. Nitrogen addition exerted a weak C source effect in a water-deficient year but enhanced the C sink in a water-rich year. Nitrogen plus water addition significantly boosted C sink potential, though this effect diminished along the grazing pressure gradient. Our findings demonstrate that the impacts of climate change on soil C fluxes in desert steppes are mediated by historical grazing intensity. Future manipulative experiments should explicitly incorporate grazing legacy effects, and integrate this factor into C models to generate reliable predictions of grassland C dynamics under global change scenarios.

Keywords: desert steppe; grazing intensity; precipitation increase; nitrogen addition; net ecosystem CO₂ exchange



Academic Editor: Małgorzata Szczepanek

Received: 15 July 2025

Revised: 15 August 2025

Accepted: 18 August 2025

Published: 21 August 2025

Citation: Wen, C.; Huang, J.; Shan, Y.; Yang, D.; Mu, L.; Zhang, P.; Liu, X.; Chang, H.; Ye, R. Effects of Nitrogen and Water Addition on Ecosystem Carbon Fluxes in a Grazing Desert Steppe. *Agronomy* **2025**, *15*, 2016. <https://doi.org/10.3390/agronomy15082016>

Copyright: © 2025 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

China possesses globally significant grassland resources, covering approximately 41% of its terrestrial area [1]. These grassland ecosystems deliver critical ecological services, including carbon sequestration, wind erosion control, water conservation, soil retention,

and biodiversity conservation. Consequently, they represent substantial carbon (C) sinks with considerable potential to mitigate global climate change [1,2]. As an integrative metric of ecosystem C source–sink dynamics, net ecosystem CO₂ exchange (NEE) reflects the balance between gross ecosystem productivity (GEP) and ecosystem respiration (ER) [3,4]. These component fluxes exhibit differential sensitivity to environmental drivers: GEP is primarily constrained by photosynthetically active radiation, nutrient availability, and plant phenological status, whereas ER responds strongly to soil temperature, moisture thresholds, and substrate supply. Consequently, NEE magnitude and directionality emerge from hierarchical interactions among biotic controls and abiotic forcings rendering it a highly responsive indicator of global change perturbations. Grasslands worldwide, particularly semi-arid grasslands in China, are subject to dual pressures from climate change (e.g., nitrogen deposition, altered precipitation) and land-use change (e.g., overgrazing) [5–7]. These drivers alter plant and microbial biodiversity, thereby modifying C cycling and generating divergent feedbacks to climate change [8,9]. Water and nitrogen represent key limiting resources in grassland ecosystems, while grazing serves as the dominant land-use practice in desert steppes [10]. These factors act in concert to modify multiple biotic and abiotic components [5,6], fundamentally altering the ecosystem structure, function, and stability [8,11], thereby impacting the ecosystem C cycle. Previous research on desert steppes remains limited, with most grazing experiments being short-term [10,12]. Consequently, the mechanisms by which long-term grazing influences ecosystem C processes through alterations in biotic and abiotic parameters remain unclear. While the individual effects of nitrogen deposition and precipitation changes on C fluxes are relatively well-established, how long-term grazing intensities modulate these effects has rarely been assessed.

Research on ecosystem C exchange in China spans the Qinghai–Tibet Plateau [13,14], meadow steppes [15,16], typical steppes [17,18], and desert steppes [19,20]. Grazing livestock influence ecosystem C fluxes' components by altering community biomass, vegetation structure, soil physicochemical properties, and microclimate through foraging, trampling, and excretion [21]. Intensive grazing exacerbates grassland degradation, triggering shifts in plant community composition and structure, reducing species diversity and productivity, and significantly diminishing belowground net primary production (BNPP). Consequently, soil nutrient content and retention capacity decline, accompanied by alterations in enzyme activity and organic carbon [22,23]. These changes accelerate CO₂ release, impacting grassland C cycling. Conversely, studies indicate that moderate grazing can enhance ecosystem CO₂ uptake potential [24,25], while others report negligible effects of grazing on CO₂ fluxes [26,27]. However, previous investigations into C fluxes' responses to grazing have typically assessed few intensity levels or short grazing histories [10]. A comprehensive assessment across multiple grazing intensities over extended periods (≥ 10 years) is therefore essential to elucidate grazing impacts on CO₂ fluxes.

Extensive evidence demonstrates that increased precipitation significantly enhances NEE in arid and semi-arid ecosystems [28,29]. This is attributed to the fact that the increase in precipitation leads to a greater rise in GEP compared to ER, with the extent of this increase depending on the degree of precipitation increase [28] and the level of environmental nutrient supply [30]. However, other studies have found that after precipitation increase, the rise in ER was equal to or greater than GEP, ultimately maintaining the ecosystem's C sink capacity or shifting it from a C sink to a source [31,32]. Consequently, there are many controversies surrounding the impact of precipitation change on NEE, and the mechanism underlying the positive or negative correlation effect is highly complex [33,34]. This complexity may be due to differences in climate, vegetation, and soil types. Moreover, variations in soil water retention capacity and the distribution of natural precipitation also lead to inconsistent responses of ecosystem C exchange to precipitation increase [35,36]. There-

fore, further research on the response of desert steppe ecosystem C fluxes to precipitation increase under future climate change holds great significance.

It is widely recognized that nitrogen input exerts an influence on the two key processes of C uptake and emission in grassland ecosystems. Nitrogen deposition elevates soil nitrogen availability, stimulating plant photosynthesis and growth. This enhances leaf area index, aboveground productivity, and GEP, while improving ecosystem water use efficiency [37,38]. However, long-term nitrogen addition experiments reveal that initial positive effects on ecosystem C exchange often progressively weaken or reverse over time. For example, in a temperate Inner Mongolian grassland, nitrogen-induced stimulation of net ecosystem C exchange consistently diminished with extended treatment duration [39]. This might be attributed to the increasing nitrogen deposition, which could acidify the soil or produce ammonia toxicity, inhibit microbial growth and activity, reduce species richness, or intensify the restriction of soil water on grasslands, thereby altering the ecosystem structure and function [40], and consequently impacting the ecosystem C cycle. Moreover, grassland productivity is co-limited by nitrogen and water availability. Consequently, nitrogen addition effects on ecosystem carbon exchange are inherently water dependent, indicating that both factors jointly regulate C fluxes [41,42]. For example, research findings in a grassland ecosystem in North America showed that the response of ecosystem C exchange to nitrogen addition was contingent upon the rainfall pattern, being promoted in wet years and suppressed in dry years [30]. Consequently, the direction and magnitude of interactive effects between climatic factors (e.g., nitrogen deposition, precipitation increase) and the anthropogenic driver (grazing) on water and nitrogen inputs vary across grassland types. This variation triggers divergent responses in ecosystem C fluxes' components. Therefore, how grasslands under chronic grazing pressure modify water and nitrogen inputs remains poorly constrained.

This study leverages an 11-year grazing platform (with four grazing intensities) established in a desert steppe. We investigated the effects of resource additions (water and nitrogen) on ecosystem CO₂ fluxes' components under different grazing scenarios. The grazing experiment was initiated in 2002, with water and nitrogen additions implemented from 2013 to 2017. By 2013 (baseline for this study), grazing had already altered vegetation and soil properties [19]. Our research addresses the following questions: (1) What were the effects of water, nitrogen, and their interactions on ecosystem CO₂ fluxes following nitrogen and water supplementation in a desert steppe under different grazing intensities, and how did the source and sink capacities change? Based on the Intermediate Disturbance Hypothesis, we propose that resource enhancement will most effectively promote carbon sequestration in grassland ecosystems under moderate grazing intensity. (2) How does resource enrichment modulate the relationship between ecosystem CO₂ fluxes and key abiotic drivers such as soil temperature and moisture? We hypothesize that soil temperature will assume a dominant role in regulating CO₂ fluxes' variations once resource limitations are alleviated.

2. Materials and Methods

2.1. Study Site

The study site is located in the desert steppe of Siziwang Banner, Inner Mongolia, China (41°46'43.6" N, 111°53'41.7" E), at an average altitude of 1456 m. This region has a persistent mid-temperate continental climate, characterized by arid springs with persistent winds and hot, low-precipitation summers. The mean average precipitation is 280 mm, contrasting with evaporation rates 7–10 times higher. Prevailing northwesterly winds sustain an annual wind speed of 4–5 m·s⁻¹. Soils are classified as Kastanozem (FAO) with a sandy loam texture. Vegetation comprises sparse, low-stature communities (mean

height: 8 cm; coverage: 17–20%) dominated by the foundation species *Stipa breviflora*, with codominants including *Artemisia frigida* and *Cleistogenes songorica*. Over 20 species collectively characterize this ecosystem [43].

2.2. Experimental Design

A long-term grazing experiment (initiated 2002) in a desert steppe comprises four grazing intensities with three replicate plots per treatment: no grazing (NG), light grazing (LG), moderate grazing (MG), and heavy grazing (HG). Grazing intensities were defined as 0, 0.91, 1.82, and 2.71 sheep·hm^{−2}·half year^{−1}, respectively. Consequently, 12 experimental plots were established for research. The grazing period was six months from 1 June to 30 November [19]. In 2013, nested 8 m × 8 m subplots were established within each grazing plot. Four isolated 2 m × 2 m treatment quadrats (galvanized iron barriers: 20 cm buried + 5 cm aboveground) with 1 m buffers were implemented per subplot: control (CK), nitrogen addition (N), water addition (W), and nitrogen plus water addition (NW). This created 48 quadrats (4 grazing intensities × 3 replicates × 4 treatments). Water addition treatments received 108 mm annually (equivalent to 40% of mean annual precipitation), applied as 36 mm per month in four equal weekly increments of 9 mm during June–August. Nitrogen was applied as calcium ammonium nitrate at a rate of 10.00 g N·m^{−2}·yr^{−1}, evenly distributed across three application dates (1 June, 1 July, and 1 August) to minimize localized nutrient effects [44].

2.3. Measurements of Ecosystem Carbon Fluxes, Soil Moisture, and Temperature

Data collection was conducted during three consecutive growing seasons (June–September 2015–2017). Ecosystem CO₂ fluxes were measured using a static chamber by an infrared gas analyzer (LI-COR, Lincoln, NE, USA) [45]. The transparent assimilation chamber (50 cm × 50 cm × 50 cm plexiglass) featured two internal mixing fans and an integrated temperature probe for continuous air monitoring. A stainless steel base collar (50 cm × 50 cm; height: 8 cm) was permanently installed to 6 cm soil depth, creating a sealed measurement volume when coupled with the chamber. NEE measurements were performed monthly during 2015–2016 and biweekly from July to September 2017 (post two-year water–nitrogen amendment), consistently between 8:00 and 12:00 h under cloud-free conditions. A minimum of 60 s of continuous CO₂ concentration data was captured during each measurement to ensure valid flux calculations. Following NEE measurement, the assimilation chamber was removed and ventilated manually until internal CO₂ concentrations equilibrated with ambient air. It was then resealed on the base collar and immediately covered with a shading cloth to measure ER [46]. Soil temperature at 10 cm depth was measured using a soil temperature probe, while soil moisture (0–10 cm layer) was determined with a TDR-300 soil moisture meter (Spectrum Technologies, Aurora, IL, USA). Negative NEE values represented net CO₂ uptake by the ecosystem, whereas positive values denoted net CO₂ release. GEP was subsequently calculated from NEE and ER.

2.4. Data Analysis

To avoid operational disturbances, data from the first and last 10 s of each recording period were excluded prior to calculation. The rate of CO₂ concentration change (dc/dt) was calculated by a linear fitting method, that is, the slope. The CO₂ exchange rate (R_s , μmol·m^{−2}·s^{−1}) was calculated by the following formula:

$$R_s = \frac{VP_{av}(1000 - W_{av})}{RS(T_{av} + 273)} \times \frac{dc}{dt}$$

The key parameters are defined as follows: V denotes the volume of the assimilation chamber (m^3), calculated as the product of length, width, and total height (chamber height + base elevation above ground). P_{av} represents the mean atmospheric pressure (kPa) inside the chamber during measurement. W_{av} indicates the mean water vapor partial pressure ($\text{mmol} \cdot \text{mol}^{-1}$) in the chamber during the measurement. R is the universal gas constant $8.314 \text{ J} \cdot \text{mol}^{-1} \cdot \text{K}^{-1}$. S corresponds to the surface area covered by the assimilation chamber (m^2). T_{av} signifies the mean air temperature inside the chamber during measurement.

We employed linear mixed models (LMMs) to assess the effects of year (Y), historical grazing intensity (GI), water addition (W), nitrogen addition (N), and their interactions on soil temperature (ST), soil moisture (SM), NEE, ER, and GEP. GI, N, W, Y, and their interactions were treated as fixed effects, with subplot as a random effect. Differences in ST and SM among the control (CK), W, N, and NW treatments across three years were tested using one-way ANOVA followed by least square method (LSD) post hoc tests. Similarly, one-way ANOVA compared: (i) NEE, ER, and GEP across CK, W, N, and NW under the same GI; and (ii) NEE, ER, and GEP across GI levels within each treatment (CK/W/N/NW). Significance was set at $p < 0.05$. Linear regression analyzed the relationships between ecosystem CO_2 fluxes (NEE/ER/GEP) and abiotic drivers (ST/SM). Data in figures represent means $\pm$ standard error (SE). All analyses used R 4.2.3 (R Core Team 2023).

3. Results

3.1. Air Temperature and Precipitation

The annual mean temperatures during 2015–2017 exhibited a unimodal pattern, measuring 5.0°C , 4.5°C , and 5.7°C , respectively, with 2017 being the warmest year. Monthly temperatures during the growing season (June–September) showed a progressive increase across the three years, averaging 17.0°C , 17.5°C , and 18.9°C , respectively. July consistently recorded the highest monthly temperatures (20.6°C , 20.5°C , and 23.0°C), marginally exceeding the 50-year baseline mean (20.0°C , 1965–2014), while September exhibited the lowest temperatures. Annual precipitation totals were 276.7 mm, 422.4 mm, and 272.3 mm for 2015–2017, respectively, with corresponding growing season precipitation amounts of 204.7 mm, 322.8 mm, and 157.9 mm. The 2016 growing season precipitation surpassed the regional long-term average (230.7 mm, 1965–2014) by 39.9%, representing 1.58 times and 2.04 times the 2015 and 2017 values, respectively (Figure 1), qualifying as a high-rainfall year. Conversely, 2015 and 2017 growing season precipitation fell below the baseline by 11.27% and 31.6%, characteristic of low-rainfall years. Distinct seasonal distribution patterns emerged: 63.2% of 2016–2017 precipitation was concentrated in June–July, whereas 2015 showed peak accumulation in June and September. August consistently recorded minimal precipitation across all years, with particularly low values of 14.0 mm (2015) and 16.2 mm (2017).

3.2. Soil Temperature and Soil Moisture

The change rule of ST was basically consistent with that of atmospheric temperature. The peak value of ST occurred in July in 2015 and 2016, and the higher temperature occurred in late June and July in 2017. The peak value of SM occurred in late June in 2015, and both occurred in late July in 2016 and 2017. SM increased significantly due to W (Figure 2). According to the linear mixed model analysis results for Y, GI, W, and N (Table 1), GI and W had highly significant effects on ST ($p < 0.001$), while N showed no significant effect ($p > 0.05$). N, W, and their interaction significantly affected SM ($p < 0.05$). The interactions between Y and W, as well as Y and N, also significantly influenced SM ($p < 0.01$). GI had no significant effect on SM ($p > 0.05$).

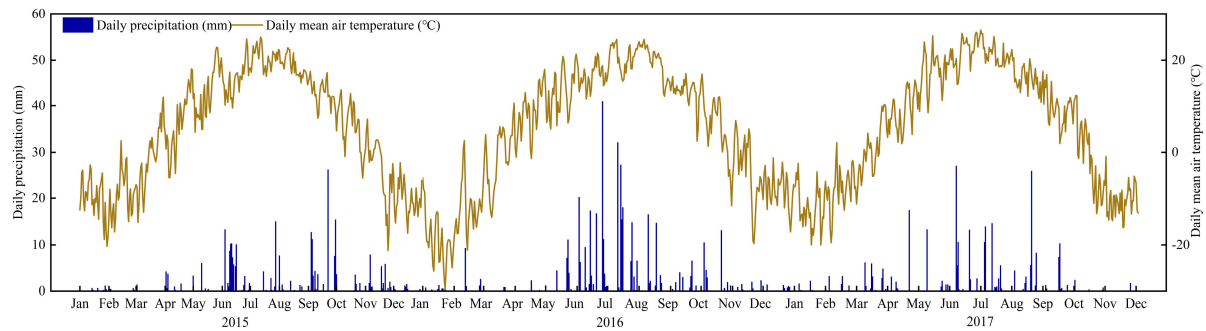


Figure 1. Daily mean air temperature and precipitation during growing seasons across the study period in Siziwang banner.

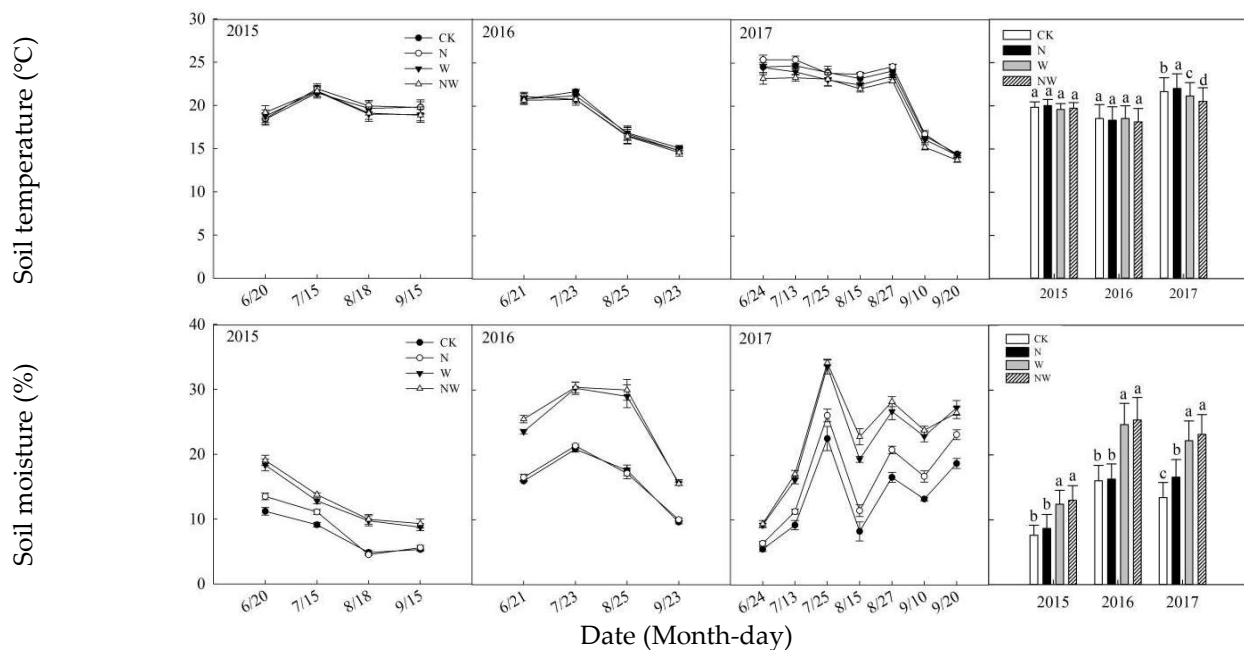


Figure 2. Changes in soil temperature and soil moisture in 2015–2017 growing season. CK, N, W, and NW represent no water and nitrogen addition, nitrogen addition, water addition, and water plus nitrogen addition, respectively. Each data point is the average of all repeated observations in that day under the treatment. Different lowercase letters indicate significant differences among treatments. The same notes are applicable as used in the following figures.

3.3. Characteristics of Ecosystem CO_2 Fluxes Under Water and Nitrogen Addition Treatments

LMM analysis revealed highly significant effects of GI on NEE and GEP ($p < 0.01$), but no significant effect on ER ($p > 0.05$). Both N and W significantly affected NEE, ER, and GEP ($p < 0.05$). The interaction between Y and W demonstrated highly significant effects on NEE, ER, and GEP ($p < 0.001$). Conversely, the Y and N interaction showed no significant effects ($p > 0.05$). Significant interactions were observed between GI and W for ER and GEP, and between N and W for NEE and GEP ($p < 0.05$; Table 1).

The seasonal dynamics of NEE, ER, and GEP from 2015 to 2017 are shown in Figure 3. Overall, the peak value of CO_2 fluxes mostly occurred in July in the desert steppe ecosystem. In 2015, without water supplement (CK and N) showed a weak C source, and only the CK treatment in the NG plot showed a weak C sink in June and July. The peak value of NEE appeared in July under water supplement (W and NW), and appeared as a weak C sink mostly. The peak value of ER appeared mostly in July, and the highest ER values occurred in W and NW in moderate grazing, which were $4.18 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ and $4.20 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, respectively. The lowest ER appeared in heavy grazing in September ($0.51 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$).

The GEP peaked in late July. Net C uptake occurred in the whole growing season in 2016, and was enhanced with plant growth. The peak value of NEE appeared in July, and gradually decreased in September. The peaks value of ER and GEP also occurred in July. Net C uptake occurred mostly in the 2017 growing season, with NEE peaking at the end of July, while ER and GEP also reached their highest levels.

Table 1. The effects of year (Y), grazing intensity (GI), water addition (W), and nitrogen addition (N) on soil temperature (ST), soil moisture (SM), net ecosystem CO₂ exchange (NEE), ecosystem respiration (ER), and gross ecosystem productivity (GEP) were analyzed by linear mixed model.

	ST	SM	NEE	ER	GEP
Y	0.17	3.80	0.01	0.08	0.03
GI	29.73 ***	1.64	24.37 ***	0.62	12.88 **
W	21.39 ***	1367.44 ***	358.07 ***	312.52 ***	455.71 ***
N	1.26	45.73 ***	14.57 ***	5.20 *	13.24 ***
Y × GI	4.10 *	1.63	0.05	5.72 *	1.92
Y × W	7.09 **	33.42 ***	14.04 ***	30.20 ***	27.66 ***
Y × N	1.54	8.66 **	1.95	1.17	2.13
GI × W	1.29	2.13	1.78	5.67 *	4.36 *
GI × N	3.15	0.58	0.09	1.33	0.63
W × N	3.78	6.57 *	4.07*	3.70	5.28 *
Y × GI × W	0.01	8.77 **	0.01	0.52	0.09
Y × GI × N	0.03	1.23	0.26	0.29	0.003
Y × W × N	2.12	4.54 *	1.77	2.28	2.69
GI × W × N	0.08	1.02	0.27	1.65	1.00
Y × GI × W × N	0.06	0.32	0.01	0.41	0.17

*, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.001$. Using grazing intensity (GI), water addition (W), nitrogen addition (N), and year (Y) as fixed factors, and subplot as random factor.

Analysis of three-year mean values demonstrated significant grazing legacy effects on CO₂ fluxes: NEE was significantly lower in NG and LG than in MG and HG ($p < 0.05$; Figure 4a). ER reached minimal levels under HG, showing a significant reduction compared to LG and MG (Figure 4b), while GEP peaked in LG but was most suppressed under HG ($p < 0.05$; Figure 4c). Annual patterns further demonstrated treatment interactions: In 2015, NG exhibited the lowest NEE (significantly below all grazed treatments; $p < 0.05$), while LG was lower than MG; ER peaked in MG ($p < 0.05$ vs. NG/HG), and HG consistently suppressed GEP ($p < 0.05$). During the water-rich 2016 (growing-season precipitation: 158% of 2015), enhanced CO₂ fluxes activity coincided with HG reducing NEE relative to NG/LG ($p < 0.05$) while altering ER and GEP versus all other treatments ($p < 0.05$). By 2017, HG significantly depressed NEE while elevating ER, whereas LG stimulated GEP ($p < 0.05$; Figure 4).

Analysis of the three-year mean ecosystem CO₂ fluxes revealed that water supplement (W and NW) significantly enhanced fluxes compared to without water supplement (CK and N) ($p < 0.05$; Figure 5). In 2015, NEE represented a net C sink under CK in the NG, but a net source in all grazed CK. All N resulted in weak C sources, with source strength increasing with grazing intensity (NG < LG < MG < HG). Conversely, water supplement (W and NW) shifted NEE to a weak sink, with sink capacity decreasing as grazing intensity increased (NG > LG > MG > HG). Both annual ER and GEP were significantly higher under water supplement (W, NW) than without water supplement (CK, N) ($p < 0.05$). In 2016, N and W significantly promoted NEE in the NG. Within LG and HG, the annual mean NEE under NW was significantly lower than under other grazing treatments. In the HG plot, both N and W showed significantly lower NEE than CK. ER was significantly higher under N compared to CK in NG and LG, while W significantly increased ER versus CK in HG. Across all grazing intensities, NW significantly elevated ER relative to CK. Annual GEP

was significantly higher under N than CK in NG and HG; W also significantly increased GEP versus CK in HG. Furthermore, NW significantly enhanced GEP compared to both CK and N across all grazing intensities ($p < 0.05$). During 2017, water supplement (W, NW) significantly increased the annual means of all CO₂ fluxes (NEE, ER, GEP) compared to without water supplement (CK, N). N reduced annual NEE, transforming the HG into a weak C source; N under MG was significantly lower than CK. Within HG, the annual mean ER under NW was significantly higher than all other grazing treatments ($p < 0.05$). N significantly decreased GEP in MG and HG, while NW in HG resulted in significantly higher GEP than other treatments ($p < 0.05$; Figure 4).

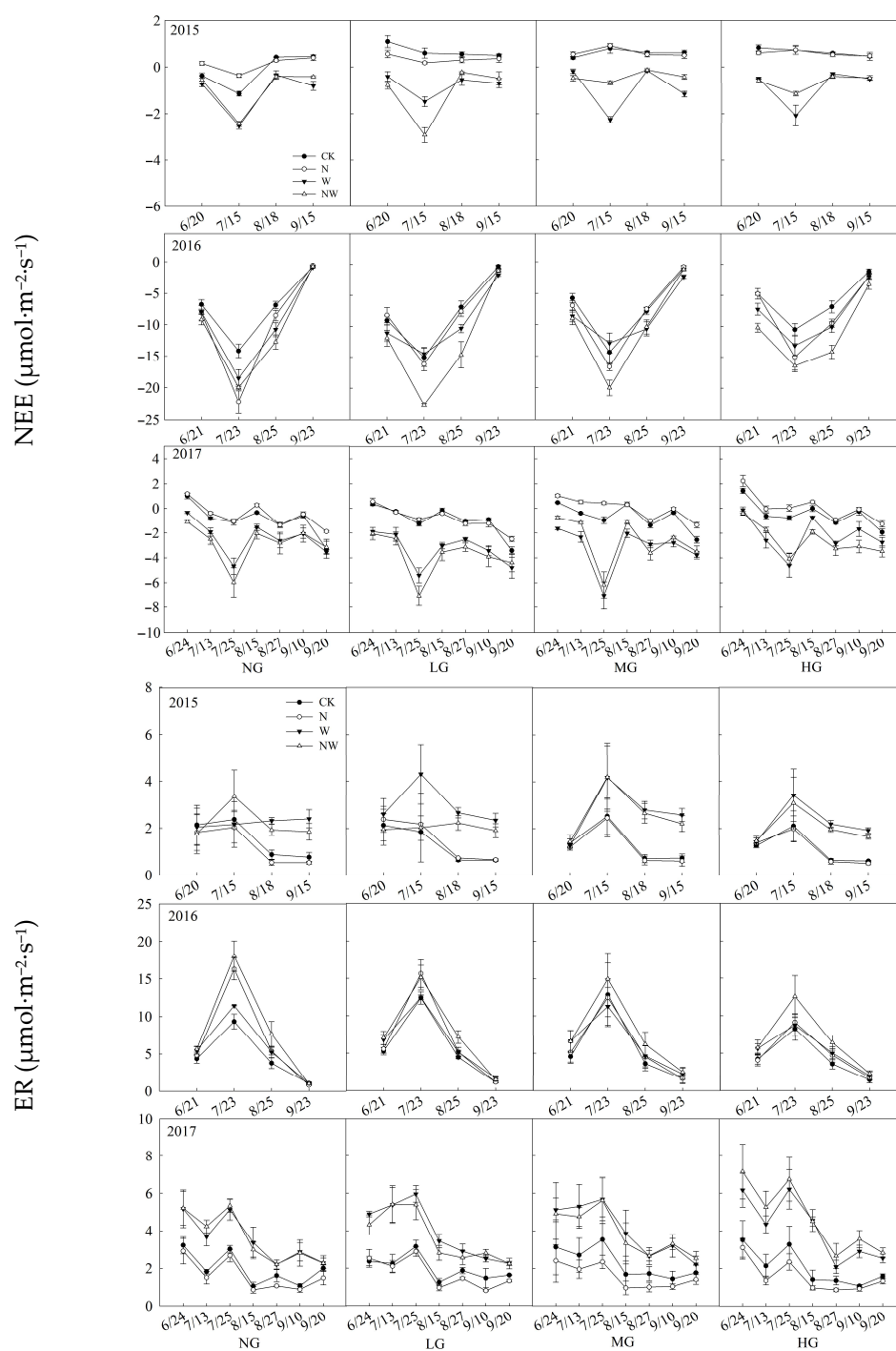


Figure 3. Cont.

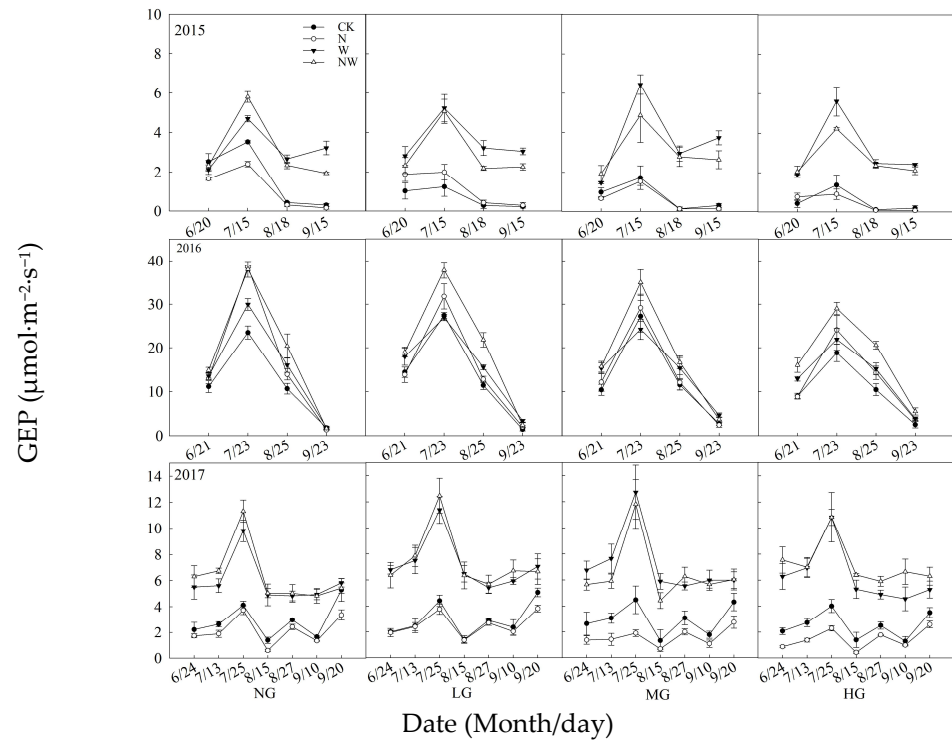


Figure 3. Seasonal dynamics of NEE, GEP, and ER in 2015, 2016, and 2017. NG, LG, MG, and HG represent no grazing, light grazing, moderate grazing, and heavy grazing, respectively.

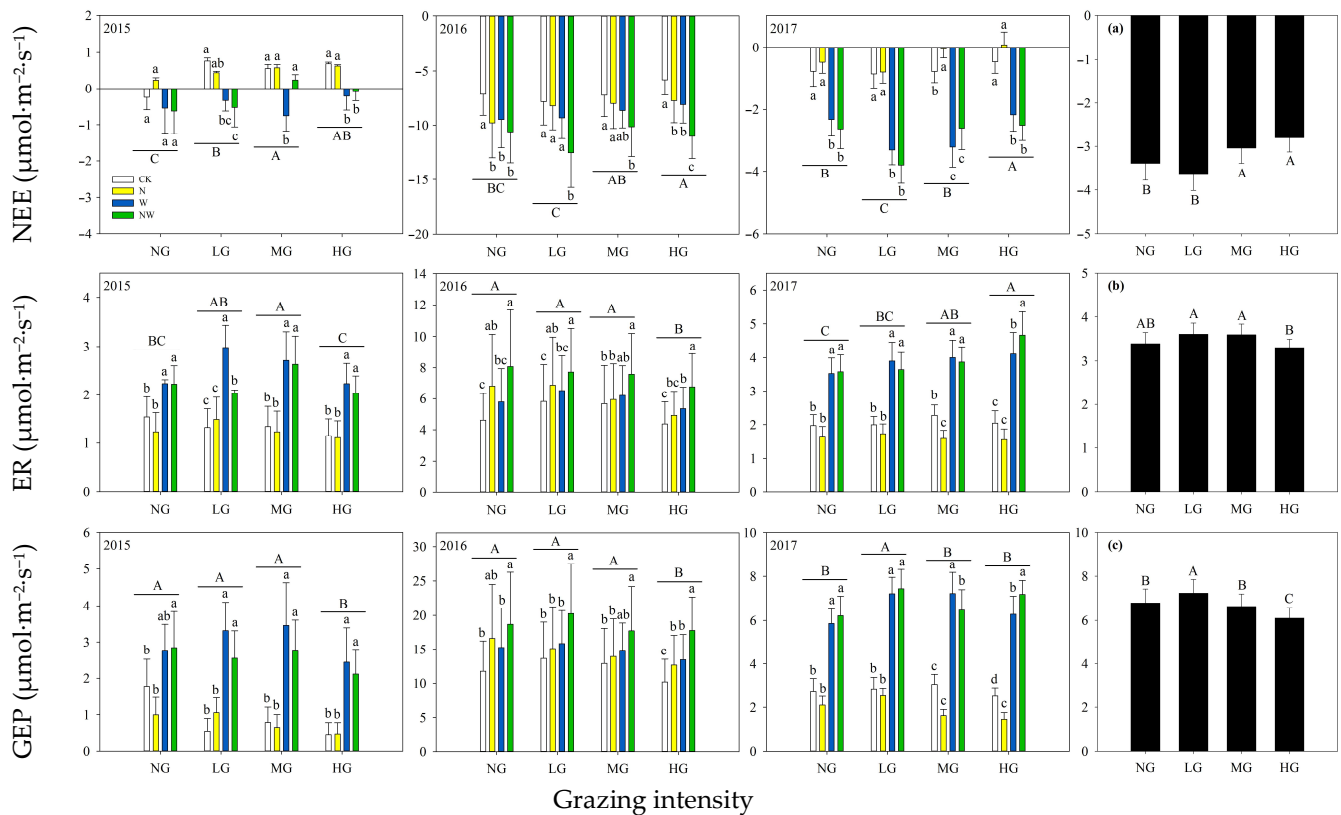


Figure 4. Annual mean values of NEE, GEP, and ER in 2015, 2016, and 2017. The 3-year average of NEE (a), ER (b), and GEP (c) under different grazing intensities. Different lowercase letters indicate significant difference between treatments under the same grazing intensity; different capital letters indicate significant difference between different grazing intensities. The means with different letters are significantly different based on LSD's multiple-range tests ($p < 0.05$). Bars indicate mean SE.

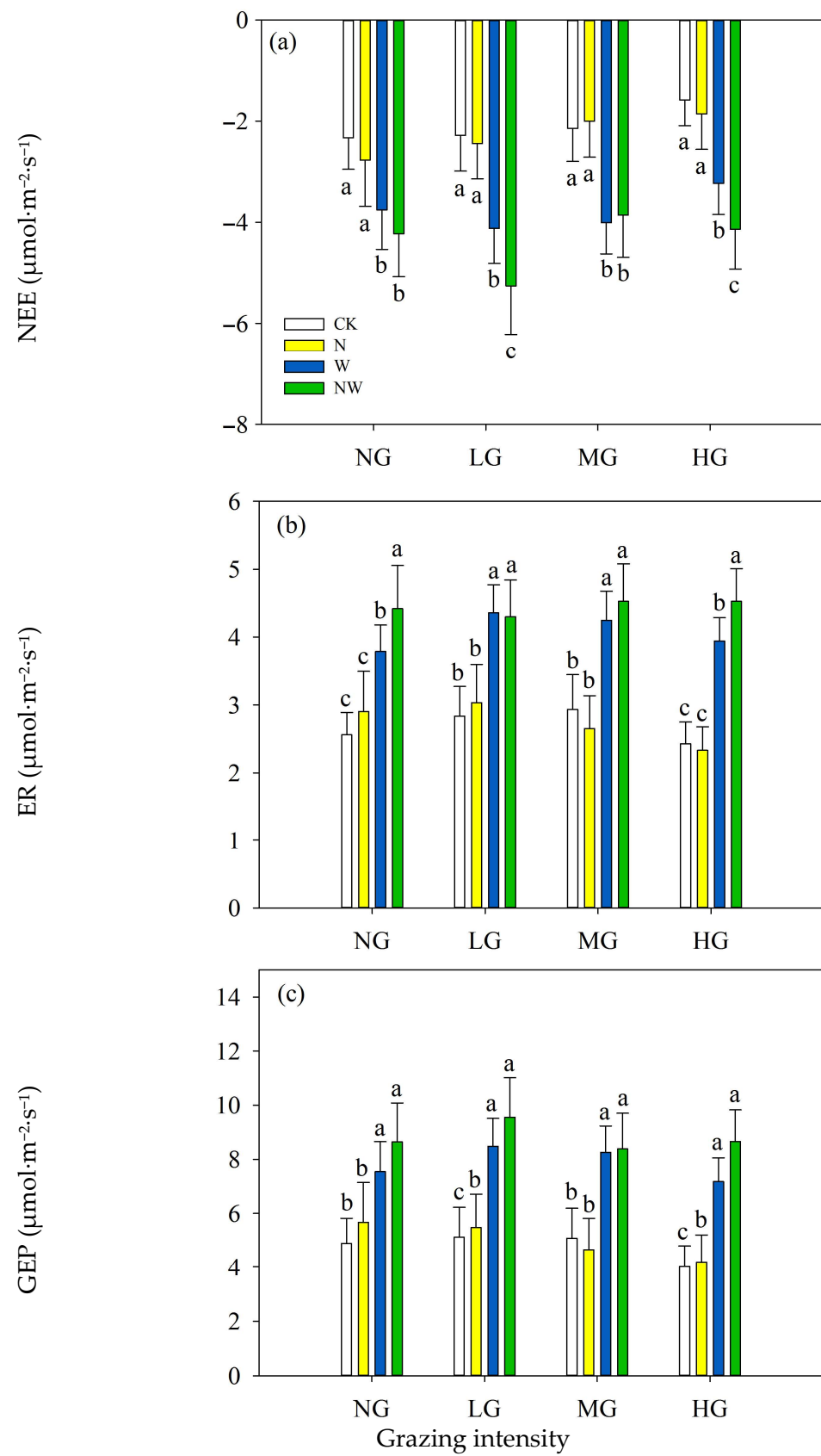


Figure 5. The 3-year average of NEE (a), ER (b), and GEP (c) in different treatments under different grazing intensities. Different lowercase letters indicate significant difference between treatments under the same grazing intensity.

3.4. Relationships of Ecosystem CO₂ Fluxes with Soil Temperature and Soil Moisture

Regression analyses examining the relationships between ecosystem CO₂ fluxes and soil temperature revealed distinct treatment effects. Neither NEE nor GEP exhibited significant correlations with soil temperature under any nutrient addition treatment ($p > 0.05$). Similarly, ER showed no significant correlation with soil temperature under CK or N ($p > 0.05$). In contrast, significant positive correlations ($p < 0.05$) emerged between ER and soil temperature under both water supplements (W and NW). The strongest correlation was observed for W alone, explaining 25% of the variation in ER (Figure 6). In contrast to temperature relationships, NEE, ER, and GEP all demonstrated highly significant linear relationships ($p < 0.01$) with soil moisture across treatments. However, the explanatory power of soil moisture varied considerably. Under nitrogen addition, soil moisture accounted for a consistently lower proportion of the variation in CO₂ fluxes ($R^2 = 0.13$ – 0.19). Conversely, under water supplement (W and NW), soil moisture explained over 40% of the variation in NEE (Figure 7).

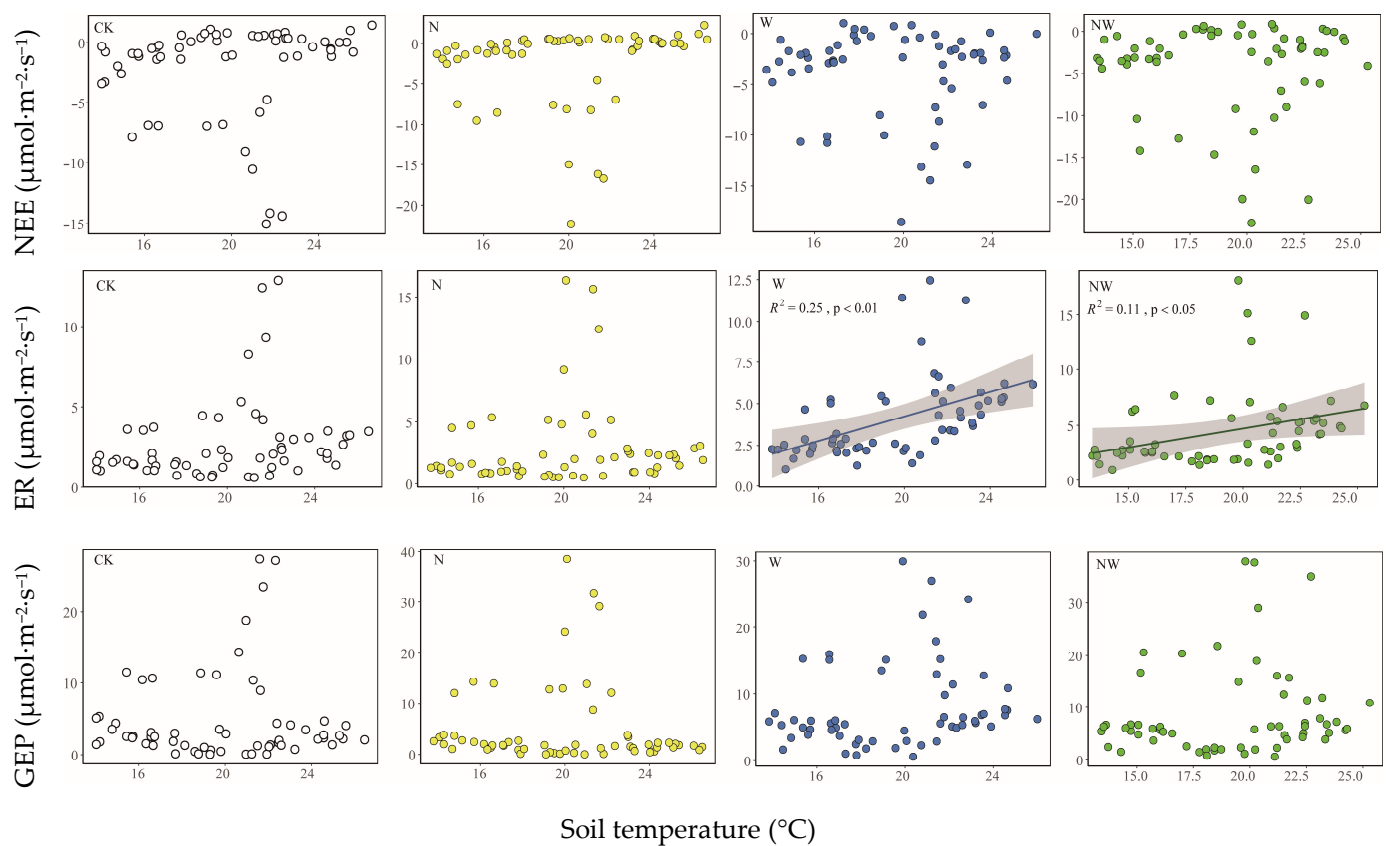


Figure 6. Relationship of the CO₂ fluxes of the ecosystem with soil temperature.

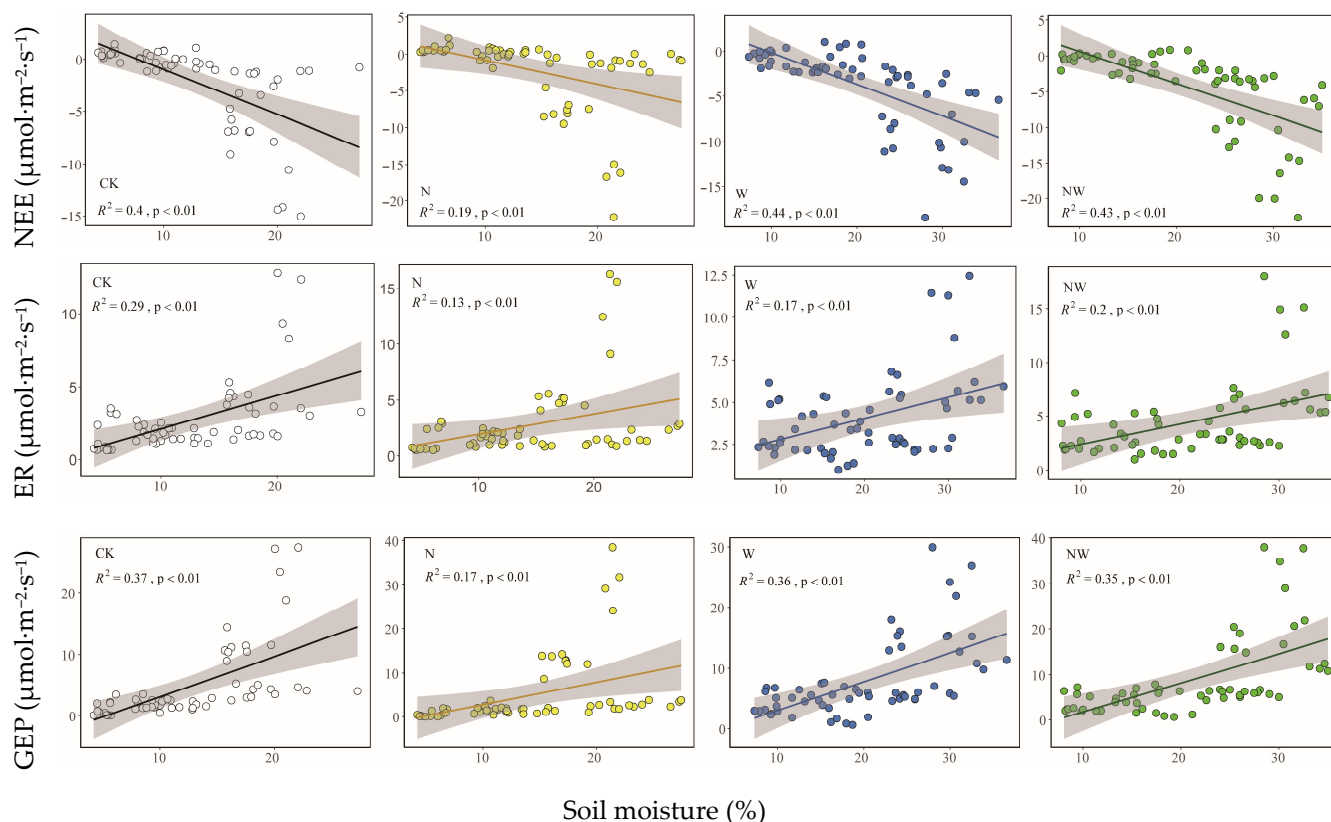


Figure 7. Relationship of the CO₂ fluxes of the ecosystem with soil moisture.

4. Discussion

The impact of grazing on ecosystem CO₂ fluxes remains subject to considerable uncertainty. Beyond grassland type, this uncertainty largely depends on grazing intensity. In this study, over the three-year experimental period, sites with a history of light grazing exhibited increases in NEE, ER, and GEP compared to no grazing (by 7.94%, 10.28%, and 6.55%, respectively), although only the increase in GEP was statistically significant ($p < 0.05$; Figure 4). These findings indicate that the legacy effects of light grazing history exert a positive effect on ecosystem CO₂ fixation. This result deviates slightly from our initial hypothesis, which posited that moderate grazing intensity would exert the strongest influence on C fluxes. This outcome aligns with previous studies suggesting that NEE may improve under light or moderate grazing due to plant compensatory growth, a concept known as the Intermediate Disturbance Hypothesis [47,48]. However, water scarcity in the desert steppe likely constrains the potential for post-grazing plant compensatory growth [48], which could further impact C fluxes within the grassland ecosystem. Our results also demonstrate that heavy grazing significantly reduced both NEE and GEP ($p < 0.05$), resulting in decreases of 17.58% and 10.19%, respectively. Notably, the inhibitory effect on GEP was stronger than that on ER (Figure 4). The underlying cause of this phenomenon may be attributed to the long-term cumulative grazing pressure, which significantly reduced both aboveground and belowground biomass and vegetation cover in heavily grazed areas [19]. Concurrently, livestock foraging preferences likely inflict greater damage on plant photosynthetic organs (e.g., leaves) [49,50]. Our findings align with previous studies reporting the potential negative impacts of grazing or mowing on C exchange in arid grassland ecosystems [51,52]. Notably, even in more humid meadow steppes, studies have found that moderate grazing can still lead to a decline in net C sequestration [49]. This inconsistency with the Intermediate Disturbance Hypothesis

may stem from variations in vegetation composition, grazing regimes, and local climate across different study sites [53]. In conclusion, the effects of grazing on grassland C flux mechanisms are complex. Furthermore, a deeper understanding of the underlying mechanisms driving these C flux changes requires integrated analyses of both plant and microbial dynamics.

Numerous studies have documented that increased precipitation enhances C uptake in grassland ecosystems [29,54]. Given that our study area is arid, grassland C budgets are strongly constrained by precipitation variability [47]. Our three-year measurements revealed a pronounced influence of precipitation on the interannual dynamics of ecosystem CO₂ fluxes. Notably, the CO₂ fluxes in 2016 were significantly higher than in 2015 and 2017. During water deficit years, low soil moisture may induce stomatal closure in plants, leading to carbon starvation due to reduced photosynthesis. It may also suppress soil microbial activity, thereby diminishing soil nutrient supply capacity [55,56]. These mechanisms collectively disrupt the C sequestration process in grassland ecosystems. Furthermore, severe drought stress can cause leaf rolling in *S. breviflora*, the dominant species in the desert steppe we studied. This reduces the Leaf Area Index, further exacerbating the inhibitory effect of soil moisture on NEE, driving it towards less negative values [57]. Therefore, desert steppes, characterized by low water availability, exhibit high sensitivity to water addition. Increased precipitation elevates soil moisture, thereby enhancing community photosynthetic capacity [58] and increasing GEP. Elevated GEP provides more substrate for root growth, microbial activity, and soil respiration, subsequently stimulating ER. Because GEP, governed by photosynthesis, is solely plant-related, while ER involves additional factors like soil microbes and fauna, GEP typically demonstrates greater sensitivity to precipitation than ER and exerts a dominant influence on NEE changes. These results demonstrate that precipitation serves as a key regulatory factor governing CO₂ flux dynamics in the *S. breviflora* desert steppe. Crucially, in years with abundant precipitation, increased rainfall can significantly enhance the C sink strength of this ecosystem.

Nitrogen addition can modulate the impacts of climate change on ecosystem C cycling and significantly affect the global C balance [39]. Our results indicate that the effect of nitrogen addition on CO₂ fluxes varied across grazing intensities. While generally stimulatory in most grazed areas, it exhibited a slight inhibitory effect under moderate grazing. However, during the relatively water-rich year (2016), nitrogen addition significantly increased NEE and ER in the no grazing condition, and NEE and GEP in the heavy grazing condition. This suggests that the efficacy of nitrogen addition is influenced by interannual precipitation variability. Our findings differ slightly from other studies in desert steppes. Research applying similar nitrogen addition levels reported significant increases in NEE, ER, and GEP [59] or in GEP and ER [60]. A key factor explaining this discrepancy may be land-use history. Previous studies were often conducted on grasslands without grazing history, whereas our sites had varying long-term grazing intensities. Prolonged grazing has likely induced different degrees of degradation and severe soil nutrient depletion, potentially leading to co-limitation by multiple nutrients. Thus, adding only nitrogen may not consistently stimulate productivity and C fixation significantly. Supporting this, a 30-year nitrogen addition experiment in northern Europe revealed negative long-term effects on NEE, ER, and GEP [22], possibly due to reduced plant diversity and constrained growth diminishing the initial positive NEE response. The significant enhancement of CO₂ fluxes under combined water and nitrogen addition (NW) compared to water addition (W) indicates a synergistic interaction. The effect of nitrogen addition appears modulated by precipitation; increased rainfall enhances soil moisture and nutrient availability, potentially unlocking a greater fertilizer effect, thereby boosting vegetation productivity, photosynthetic C fixation, and overall ecosystem C sequestration capacity.

Extensive research identifies soil temperature and moisture content as primary drivers of ecosystem CO₂ fluxes [61,62]. Comparing the relationships over three years, soil moisture consistently exhibited highly significant linear relationships with all CO₂ fluxes (NEE, ER, GEP) and generally explained a larger proportion of their variation than soil temperature. Soil temperature only showed a significant effect on ER under water supplement (W and NW). While many studies identify soil temperature as the dominant factor constraining NEE [61,63] where warming increases root and microbial respiration, when promoting CO₂ release, our results differ. We posit that in the *Stipa breviflora* desert steppe, CO₂ fluxes are primarily governed by soil moisture. The inherent aridity and low soil moisture in this system subject plants to water stress, triggering stomatal closure [64], reduced transpiration, and increased water-use efficiency. A water deficit directly impairs photosynthesis, thereby controlling net C uptake. The dominant limiting factor varies across grassland types, leading to divergent responses of CO₂ fluxes to climate change. Our findings in the desert steppe further substantiate this pattern, revealing that variations in soil moisture serve as the primary driver of C flux reduction in grazed ecosystems. Additionally, soil temperature plays a critical role in regulating ER, with increased precipitation acting primarily to strengthen the correlation between ER and soil temperature.

It is important to note that this study employed only a single application rate for both water and nitrogen addition. Grassland communities comprise multiple species with varying growth, development, and photosynthetic responses to water and nitrogen availability, which in turn influence C cycling dynamics. Future research should therefore incorporate gradients of water and nitrogen application levels to better elucidate their interactive effects on grassland ecosystem functioning.

5. Conclusions

Historical grazing intensity exerts significant effects on ecosystem scale C fluxes within the desert steppe. Light grazing enhanced GEP by 6.55% ($p < 0.05$), while heavy grazing suppressed NEE and GEP by 17.58% and 10.19%, respectively ($p < 0.05$). Water availability emerged as the primary limiting factor for CO₂ fluxes in this desert steppe. Water addition significantly elevated NEE. Soil moisture content exhibited significant linear correlations with all ecosystem CO₂ fluxes, explaining over 40% of the variation in NEE. Furthermore, water addition enhances the soil temperature coupling of ER. The effect of nitrogen addition on ecosystem CO₂ fluxes was contingent upon precipitation. During water-deficient years (2015, 2017), nitrogen addition primarily exerted a slight inhibitory effect. In contrast, during the water-rich year (2016), it exerted a stimulatory effect, significantly increasing the annual mean NEE. This demonstrates that the nitrogen fertilizer effect is constrained by water availability. Combined water and nitrogen addition significantly boosted CO₂ fluxes compared to water addition. This indicates that in arid regions, the efficacy of nutrient addition is regulated by water availability; increased precipitation can unlock the potential fertilizer effect of nitrogen. Currently, most grasslands are experiencing intensified climate change and excessive grazing pressure. Our findings demonstrate that historical grazing intensity modulates both the direction and magnitude of soil C flux responses to climate change. Consequently, future climate change experiments in grassland ecosystems must crucially incorporate grazing intensity and its interactions with climatic factors (e.g., nitrogen deposition, precipitation regimes) to enable robust predictions of soil C dynamics under future scenarios.

Author Contributions: Methodology, J.H.; formal analysis, Y.S.; investigation, D.Y., L.M., P.Z., R.Y., H.C. and X.L.; data curation, C.W.; writing—original draft preparation, C.W.; writing—review and editing, J.H. and Y.S.; visualization, C.W. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the National Natural Science Foundation of China (Grant No. 31860138), Inner Mongolia Autonomous Region Science and Technology Planning (Grant No. 2025YFDZ0082), Innovation Fund of Inner Mongolia Academy of Agricultural and Animal Husbandry Science (Grant No. 2021CXJJM06, 2023CYZX03, 2024CYZX04), and Zhengxiangbai Banner Xilingol League Inner Mongolia Grassland Ecological Restoration Experimental Study.

Data Availability Statement: Data are contained within the article.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

NEE	Net ecosystem CO ₂ exchange
GEP	Gross ecosystem productivity
ER	Ecosystem respiration
C	Carbon
NG	No grazing
LG	Light grazing
MG	Moderate grazing
HG	Heavy grazing
CK	No water and nitrogen addition
N	Nitrogen addition
W	Water addition
NW	Combined water and nitrogen addition
GI	Historical grazing intensity
Y	Year
ST	Soil temperature
SM	Soil moisture

References

- Shen, H.H.; Zhu, Y.K.; Zhao, X.; Geng, X.Q.; Gao, S.Q.; Fang, J.Y. Analysis of current grassland resources in China. *Chin. Sci. Bull.* **2016**, *61*, 139–154.
- Bai, Y.F.; Huang, J.H.; Zheng, S.X.; Pan, Q.M.; Zhang, L.X.; Zhou, H.K.; Xu, H.L.; Li, Y.L.; Ma, J. Drivers and regulating mechanisms of grassland and desert ecosystem services. *Chin. J. Plant Ecol.* **2014**, *38*, 93–102.
- Niu, S.; Wu, M.; Han, Y.; Xia, J.; Li, L.; Wan, S. Water-mediated responses of ecosystem carbon fluxes to climatic change in a temperate steppe. *New Phytol.* **2007**, *177*, 209–219. [[CrossRef](#)] [[PubMed](#)]
- Sloat, L.L.; Henderson, A.N.; Lamanna, C.; Enquist, B.J. The effect of the foresummer drought on carbon exchange in Subalpine meadows. *Ecosystems* **2015**, *18*, 533–545. [[CrossRef](#)]
- Bobbink, R.; Hicks, K.; Galloway, J.; Spranger, T.; Alkemade, R.; Ashmore, M.; Bustamante, M.; Cinderby, S.; Davidson, E.; Dentener, F.; et al. Global assessment of nitrogen deposition effects on terrestrial plant diversity: A synthesis. *Ecol. Appl.* **2010**, *20*, 30–59. [[CrossRef](#)]
- Hilker, T.; Natsagdorj, E.; Waring, R.H.; Lyapustin, A.; Wang, Y. Satellite observed widespread decline in Mongolian grasslands largely due to overgrazing. *Glob. Change Biol.* **2013**, *20*, 418–428. [[CrossRef](#)]
- Lemoine, N.P.; Sheffield, J.; Dukes, J.S.; Knapp, A.K.; Smith, M.D. Terrestrial Precipitation Analysis (TPA): A resource for characterizing long-term precipitation regimes and extremes. *Methods Ecol. Evol.* **2016**, *7*, 1396–1401. [[CrossRef](#)]
- Zhang, H.; Wang, B.; Wu, Y.; Wu, L.; Yue, L.; Bai, Y.; Chen, D. Plants and soil biota co-regulate stability of ecosystem multifunctionality under multiple environmental changes. *Ecology* **2025**, *106*, e4534. [[CrossRef](#)] [[PubMed](#)]
- Xu, F.; Li, J.; Su, J.; Sasaki, T.; Lu, X.; Wang, Y.; Chen, D.; Bai, Y. Understanding the drivers of ecosystem multifunctionality in the Mongolian steppe: The role of grazing history and resource input. *Agric. Ecosyst. Environ.* **2023**, *359*, 108748. [[CrossRef](#)]
- Zhou, G.; Zhou, X.; He, Y.; Shao, J.; Hu, Z.; Liu, R.; Zhou, H.; Hosseinibai, S. Grazing intensity significantly affects belowground carbon and nitrogen cycling in grassland ecosystems: A meta-analysis. *Glob. Change Biol.* **2016**, *23*, 1167–1179. [[CrossRef](#)]
- Post, A.K.; Knapp, A.K. The importance of extreme rainfall events and their timing in a semi-arid grassland. *J. Ecol.* **2020**, *108*, 2431–2443. [[CrossRef](#)]

12. Zhao, F.; Ren, C.; Shelton, S.; Wang, Z.; Pang, G.; Chen, J.; Wang, J. Grazing intensity influence soil microbial communities and their implications for soil respiration. *Agric. Ecosyst. Environ.* **2017**, *249*, 50–56. [\[CrossRef\]](#)
13. Wu, J.; Wang, H.; Li, G.; Lu, Y.; Wei, X. Effects of rainfall amount and frequency on carbon exchange in a wet meadow ecosystem on the Qinghai-Tibet Plateau. *Catena* **2022**, *219*, 106629. [\[CrossRef\]](#)
14. Liu, Y.; Chen, R.; Han, C.; Liu, Z.; Yang, Z.; Zhao, Y. Diurnal pattern and characteristic of soil respiration and net ecosystem carbon exchange in alpine meadow ecosystem on the northeastern Qinghai-Tibet Plateau. *Ecol. Indic.* **2024**, *165*, 112180. [\[CrossRef\]](#)
15. Hasi, M.; Zhang, X.; Niu, G.; Wang, Y.; Geng, Q.; Quan, Q.; Chen, S.; Han, X.; Huang, J. Soil moisture, temperature and nitrogen availability interactively regulate carbon exchange in a meadow steppe ecosystem. *Agric. For. Meteorol.* **2021**, *304–305*, 108389. [\[CrossRef\]](#)
16. Wang, Y.; Meng, B.; Zhong, S.; Wang, D.; Ma, J.; Sun, W. Aboveground biomass and root/shoot ratio regulated drought susceptibility of ecosystem carbon exchange in a meadow steppe. *Plant Soil* **2018**, *432*, 259–272. [\[CrossRef\]](#)
17. Li, J.; Huang, Y.; Xu, F.; Wu, L.; Chen, D.; Bai, Y. Responses of growing-season soil respiration to water and nitrogen addition as affected by grazing intensity. *Funct. Ecol.* **2018**, *32*, 1890–1901. [\[CrossRef\]](#)
18. Xu, F.; Li, J.; Wu, L.; Zhu, B.; Chen, D.; Bai, Y. Grazing legacy mediates the diverse responses of grassland multidimensional stability to resource enrichment. *Agric. Ecosyst. Environ.* **2024**, *378*, 109313. [\[CrossRef\]](#)
19. Wang, Y.; Zhao, Q.; Wang, Z.; Zhao, M.; Han, G. Overgrazing leads to decoupling of precipitation patterns and ecosystem carbon exchange in the desert steppe through changing community composition. *Plant Soil* **2023**, *486*, 607–620. [\[CrossRef\]](#)
20. Jin, Y.X.; Liu, F.; Zhang, J.; Han, M.Q.; Wang, Z.W.; Qu, Z.Q.; Han, G.D. Net ecosystem carbon exchange characteristics in *Stipa breviflora* desert steppe with different stocking rates. *Chin. J. Plant Ecol.* **2018**, *42*, 361–371.
21. Huian, G. Effects of grazing on carbon and nitrogen in plants and soils in a semiarid desert grassland, China. *J. Arid Land* **2014**, *7*, 341–349.
22. Shen, C.; Zhang, J.; Yang, X.; Liu, J.; Han, G. Effects of grazing on temperate grassland ecosystems: A meta-analysis. *Agric. Ecosyst. Environ.* **2024**, *381*, 109452. [\[CrossRef\]](#)
23. Bai, Y.; Cotrufo, M.F. Grassland soil carbon sequestration: Current understanding, challenges, and solutions. *Science* **2022**, *377*, 603–608. [\[CrossRef\]](#)
24. Wang, H.; Li, Y.; Zhang, J.; Zhang, T.; Wang, Y.; Li, F.Y. Moderate grazing reduces while mowing increases greenhouse gas emissions from a steppe grassland: Key modulating function played by plant standing biomass. *J. Environ. Manag.* **2025**, *374*, 124142. [\[CrossRef\]](#)
25. Chen, J.; Shi, W.; Cao, J. Effects of grazing on ecosystem CO₂ exchange in a meadow grassland on the Tibetan Plateau during the growing season. *Environ. Manag.* **2014**, *55*, 347–359. [\[CrossRef\]](#)
26. Hou, L.; Liu, Y.; Du, J.; Wang, M.; Wang, H.; Mao, P. Grazing effects on ecosystem CO₂ fluxes differ among temperate steppe types in Eurasia. *Sci. Rep.* **2016**, *6*, 29028. [\[CrossRef\]](#)
27. Zhou, W.C.; Suolang, D.E.J.; Cui, L.J.; Wang, Y.F.; Li, W. Effects of fencing and grazing on the emissions of CO₂ and CH₄ in Zoige Peatland, East Qinghai-Tibetan Plateau. *Ecol. Environ. Sci.* **2015**, *24*, 183–189.
28. Chen, S.; Lin, G.; Huang, J.; Jenerette, G.D. Dependence of carbon sequestration on the differential responses of ecosystem photosynthesis and respiration to rain pulses in a semiarid steppe. *Glob. Change Biol.* **2009**, *15*, 2450–2461. [\[CrossRef\]](#)
29. Yu, J.; Zhang, Y.; Wang, Y.; Luo, X.; Liang, X.; Huang, X.; Zhao, Y.; Zhou, X.; Li, J. Ecosystem photosynthesis depends on increased water availability to enhance carbon assimilation in semiarid desert steppe in northern China. *Glob. Ecol. Conserv.* **2022**, *38*, e02202. [\[CrossRef\]](#)
30. Harpole, W.S.; Potts, D.L.; Suding, K.N. Ecosystem responses to water and nitrogen amendment in a California grassland. *Glob. Change Biol.* **2007**, *13*, 2341–2348. [\[CrossRef\]](#)
31. Risch, A.; Frank, D. Effects of increased soil water availability on grassland ecosystem carbon dioxide fluxes. *Biogeochemistry* **2007**, *86*, 91–103. [\[CrossRef\]](#)
32. Koerner, S.E.; Collins, S.L.; Blair, J.M.; Knapp, A.K.; Smith, M.D. Rainfall variability has minimal effects on grassland recovery from repeated grazing. *J. Veg. Sci.* **2013**, *25*, 36–44. [\[CrossRef\]](#)
33. Guan, K.Y.; Good, S.P.; Caylor, K.K.; Medvigy, D.; Pan, M.; Wood, E.F.; Sato, H.; Biasutti, M.; Chen, M.; Ahlström, A.; et al. Simulated sensitivity of African terrestrial ecosystem photosynthesis to rainfall frequency, intensity, and rainy season length. *Environ. Res. Lett.* **2017**, *13*, 025013. [\[CrossRef\]](#)
34. Liu, J.; Ma, X.; Duan, Z.; Jiang, J.; Reichstein, M.; Jung, M. Impact of temporal precipitation variability on ecosystem productivity. *Wires Water* **2020**, *7*, e1481. [\[CrossRef\]](#)
35. Huang, G.; Li, Y.; Padilla, F.M. Ephemeral plants mediate responses of ecosystem carbon exchange to increased precipitation in a temperate desert. *Agric. For. Meteorol.* **2014**, *201*, 141–152. [\[CrossRef\]](#)
36. Hao, G.; Hu, Z.; Di, K.; Li, S. Rainfall pulse response of carbon exchange to the timing of natural intra-annual rainfall in a temperate grass ecosystem. *Ecol. Indic.* **2020**, *118*, 106730. [\[CrossRef\]](#)

37. Brueck, H.; Erdle, K.; Gao, Y.; Giese, M.; Zhao, Y.; Peth, S.; Lin, S. Effects of N and water supply on water use-efficiency of a semiarid grassland in Inner Mongolia. *Plant Soil* **2009**, *328*, 495–505. [\[CrossRef\]](#)
38. Jiang, L.; Guo, R.; Zhu, T.; Niu, X.; Guo, J.; Sun, W. Water- and plant-mediated responses of ecosystem carbon fluxes to warming and nitrogen addition on the Songnen grassland in northeast China. *PLoS ONE* **2012**, *7*, e45205. [\[CrossRef\]](#) [\[PubMed\]](#)
39. Niu, S.; Wu, M.; Han, Y.; Xia, J.; Zhang, Z.; Yang, H.; Wan, S. Nitrogen effects on net ecosystem carbon exchange in a temperate steppe. *Glob. Change Biol.* **2009**, *16*, 144–155. [\[CrossRef\]](#)
40. Chen, D.; Xing, W.; Lan, Z.; Saleem, M.; Wu, Y.; Hu, S.; Bai, Y. Direct and indirect effects of nitrogen enrichment on soil organisms and carbon and nitrogen mineralization in a semi-arid grassland. *Funct. Ecol.* **2018**, *33*, 175–187. [\[CrossRef\]](#)
41. Yan, L.; Chen, S.; Huang, J.; Lin, G. Increasing water and nitrogen availability enhanced net ecosystem CO₂ assimilation of a temperate semiarid steppe. *Plant Soil* **2011**, *349*, 227–240. [\[CrossRef\]](#)
42. Song, W.; Ochoa-Hueso, R.; Cui, H.; Yang, X.; Fan, M.; Sun, W. Effects of nitrogen addition on ecosystem-level carbon fluxes and water use efficiency are more dependent on early growing season precipitation than on annual precipitation in a semi-arid meadow steppe. *Agric. For. Meteorol.* **2023**, *330*, 109316. [\[CrossRef\]](#)
43. Wang, Z.; Jiao, S.; Han, G.; Zhao, M.; Ding, H.; Zhang, X.; Wang, X.; Ayers, E.L.; Willms, W.D.; Havsatad, K.; et al. Effects of stocking rate on the variability of peak standing crop in a desert steppe of Eurasia grassland. *Environ. Manag.* **2013**, *53*, 266–273. [\[CrossRef\]](#)
44. Zhang, P.J.; Huang, J.H.; Mu, L.; Shan, Y.M.; Ye, R.H.; Wen, C.; Chang, H.; Ren, T.T.; Chen, S.P.; Bai, Y.F. Influence of nitrogen and water addition on the primary productivity of *Stipa breviflora* in a desert steppe under different grazing intensities. *Acta Ecol. Sin.* **2022**, *42*, 5458–5470. [\[CrossRef\]](#)
45. Chen, S.; Lin, G.; Huang, J.; He, M. Responses of soil respiration to simulated precipitation pulses in semiarid steppe under different grazing regimes. *J. Plant Ecol.* **2008**, *1*, 237–246. [\[CrossRef\]](#)
46. Zhang, X.; Tan, Y.; Li, A.; Ren, T.; Chen, S.; Wang, L.; Huang, J. Water and nitrogen availability co-control ecosystem CO₂ exchange in a semiarid temperate steppe. *Sci. Rep.* **2015**, *5*, 15549. [\[CrossRef\]](#)
47. Kang, X.; Hao, Y.; Cui, X.; Chen, H.; Li, C.; Rui, Y.; Tian, J.; Kardol, P.; Zhong, L.; Wang, J.; et al. Effects of grazing on CO₂ balance in a semiarid steppe: Field observations and modeling. *J. Soils Sediments* **2013**, *13*, 1012–1023. [\[CrossRef\]](#)
48. Zhang, F.; Quan, Q.; Ma, F.; Zhou, Q.; Niu, S. Clipping increases ecosystem carbon sequestration and its sensitivity to precipitation change in an alpine meadow. *Plant Soil* **2019**, *458*, 165–174. [\[CrossRef\]](#)
49. Chang, Q.; Xu, T.; Ding, S.; Wang, L.; Liu, J.; Wang, D.; Wang, Y.; Li, Z.; Zhao, X.; Song, X.; et al. Herbivore Assemblage as an Important Factor Modulating Grazing Effects on Ecosystem Carbon Fluxes in a Meadow Steppe in Northeast China. *J. Geophys. Res. Biogeosci.* **2020**, *125*, e2020JG005652. [\[CrossRef\]](#)
50. Zhu, L.; Johnson, D.A.; Wang, W.; Ma, L.; Rong, Y. Grazing effects on carbon fluxes in a Northern China grassland. *J. Arid Environ.* **2014**, *114*, 41–48. [\[CrossRef\]](#)
51. Fan, F.; Liang, C.; Tang, Y.; Harker-schuch, I.; Porter, J.R. Effects and relationships of grazing intensity on multiple ecosystem services in the Inner Mongolian steppe. *Sci. Total Environ.* **2019**, *675*, 642–650. [\[CrossRef\]](#)
52. Koncz, P.; Pintér, K.; Balogh, J.; Papp, M.; Hidy, D.; Csintalan, Z.; Molnár, E.; Szaniszló, A.; Kampfl, G.; Horváth, L.; et al. Extensive grazing in contrast to mowing is climate-friendly based on the farm-scale greenhouse gas balance. *Agric. Ecosyst. Environ.* **2017**, *240*, 121–134. [\[CrossRef\]](#)
53. Giltrap, D.L.; Kirschbaum, M.U.; Laubach, J.; Hunt, J.E. The effects of irrigation on carbon balance in an irrigated grazed pasture system in New Zealand. *Agric. Syst.* **2020**, *182*, 102851. [\[CrossRef\]](#)
54. Xu, F.; Li, J.; Su, J.; Lu, X.; Wang, Y.; Wu, L.; Wang, C.; Chen, D.; Bai, Y. Seasonality regulates the effects of resource addition on plant diversity and ecosystem functioning in semi-arid grassland. *J. Plant Ecol.* **2021**, *14*, 1143–1157. [\[CrossRef\]](#)
55. Hawkes, C.V.; Waring, B.G.; Rocca, J.D.; Kivlin, S.N. Historical climate controls soil respiration responses to current soil moisture. *Proc. Natl. Acad. Sci. USA* **2017**, *114*, 6322–6327. [\[CrossRef\]](#) [\[PubMed\]](#)
56. Wang, Y.H.; Chen, J.Q.; Zhou, G.S.; Shao, C.L.; Chen, J.; Wang, Y.; Song, J.M. Predominance of precipitation event controls ecosystem CO₂ exchange in an Inner Mongolian desert grassland, China. *J. Clean. Prod.* **2018**, *197*, 781–793. [\[CrossRef\]](#)
57. Mu, L.; Zhang, P.J.; Shan, Y.M.; Wen, C.; Ye, R.H.; Chang, H.; Qiu, X.; Sun, H.L.; Ren, T.T.; Chen, S.P.; et al. Influence of short-term nitrogen and water addition on the leaf traits of *Stipa breviflora* in a desert steppe under different grazing intensities. *Acta Ecologica Sin.* **2021**, *41*, 522–534.
58. Ye, R.; Liu, G.; Chang, H.; Shan, Y.; Mu, L.; Wen, C.; Te, R.; Wu, N.; Shi, L.; Liu, Y.; et al. Response of plant traits of *Stipa breviflora* to grazing intensity and fluctuation in annual precipitation in a desert steppe, northern China. *Glob. Ecol. Conserv.* **2020**, *24*, e01237. [\[CrossRef\]](#)
59. Li, Y.L.; Hong, M.; Bai, W.M.; Han, G.D.; Wang, H.M. The response of gas exchange on water supplied and N addition in the *Stipa breviflora* steppe. *Ecol. Environ. Sci.* **2014**, *23*, 217–222.
60. Wu, Q.; Han, G.D.; Wang, Z.W.; Pan, Z.L.; Liu, F.; Wang, R.Z.; Zhang, R.Y.; Qin, J.; Li, J.W. Effects of warming and N addition on ecosystem carbon exchange in a desert steppe. *Chin. J. Ecol.* **2016**, *35*, 1427–1434.

61. Wang, J.; Liu, Y.; Wang, S.; Ma, P.; Li, Y.; Wang, R.; Liu, W.; Jia, Z.; Li, W.; Niu, Y.; et al. Enhanced ecosystem carbon sink in shrub-grassland ecotone under grazing exclusion on Tibetan plateau. *Ecol. Indic.* **2024**, *160*, 111854. [[CrossRef](#)]
62. Li, G.; Han, H.; Du, Y.; Hui, D.; Xia, J.; Niu, S.; Li, X.; Wan, S. Effects of warming and increased precipitation on net ecosystem productivity: A long-term manipulative experiment in a semiarid grassland. *Agric. For. Meteorol.* **2016**, *232*, 359–366. [[CrossRef](#)]
63. Du, L.; Luo, Y.; Zhang, J.; Shen, Y.; Zhang, J.; Tian, R.; Shao, W.; Xu, Z. Reduction in precipitation amount, precipitation events, and nitrogen addition change ecosystem carbon fluxes differently in a semi-arid grassland. *Sci. Total Environ.* **2024**, *927*, 172276. [[CrossRef](#)] [[PubMed](#)]
64. Agurla, S.; Gahir, S.; Munemasa, S.; Murata, Y.; Raghavendra, A.S. *Advances in Experimental Medicine and Biology*; Springer: Singapore, 2018; pp. 215–232.

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.