Drought and Carbon Cycling of Grassland Ecosystems under Global Change: A Review

Tianjie Lei [1,2], Zhiguo Pang [1,2,*], Xingyong Wang [1,2], Lin Li [1,2], June Fu [1,2,†], Guangyuan Kan [1,2,†], Xiaolei Zhang [1,2,†], Liuqian Ding [1,2], Jiren Li [1,2], Shifeng Huang [1,2] and Changliang Shao [3,*]

[1]   State Key Laboratory of Simulation and Regulation of Water Cycle in River Basin, Beijing 100038, China; leitj@iwhr.com (T.L.); wangxy@iwhr.com (X.W.); lilin@iwhr.com (L.L.); fuje@iwhr.com (J.F.); kangy@iwhr.com (G.K.); ahuzhxl@163.com (X.Z.); dinglq@iwhr.com (L.D.); lijiren@iwhr.com (J.L.); huangsf@iwhr.com (S.H.)

[2]   China Institute of Water Resources and Hydropower Research (IWHR), Beijing 100038, China

[3]   Department of Geography/CGCEO, Michigan State University, East Lansing, MI 48823, USA

*   Correspondence: pangzg@iwhr.com (Z.P.); clshao@msu.edu (C.S.); Tel.: +86-10-6878-5406 (Z.P.)

†   These authors contributed equally to this work.

Abstract: In recent years, the increased intensity and duration of droughts have dramatically altered the structure and function of grassland ecosystems, which have been forced to adapt to this change in climate. Combinations of global change drivers such as elevated atmospheric CO_2 concentration, warming, nitrogen (*N*) deposition, grazing, and land-use change have influenced the impact that droughts have on grassland *C* cycling. This influence, to some extent, can modify the relationship between droughts and grassland carbon (*C*) cycling in the multi-factor world. Unfortunately, prior reviews have been primarily anecdotal from the 1930s to the 2010s. We investigated the current state of the study on the interactive impacts of multiple factors under drought scenarios in grassland *C* cycling and provided scientific advice for dealing with droughts and managing grassland *C* cycling in a multi-factor world. Currently, adequate information is not available on the interaction between droughts and global change drivers, which would advance our understanding of grassland *C* cycling responses. It was determined that future experiments and models should specifically test how droughts regulate grassland *C* cycling under global changes. Previous multi-factor experiments of current and future global change conditions have studied various drought scenarios poorly, including changes in precipitation frequency and amplitude, timing, and interactions with other global change drivers. Multi-factor experiments have contributed to quantifying these potential changes and have provided important information on how water affects ecosystem processes under global change. There is an urgent need to establish a systematic framework that can assess ecosystem dynamic responses to droughts under current and future global change and human activity, with a focus on the combined effects of droughts, global change drivers, and the corresponding hierarchical responses of an ecosystem.

Keywords: water stress; carbon sequestration; grassland; global change drivers; ecosystem function

1. Introduction

Grasslands cover around 40% of the global land surface and a large fraction of their biomass is below ground [1]. Therefore, grassland soils hold relatively large quantities of organic *C* and store around 28%–37% of the global soil organic *C* pool [2]. Grasslands are net sinks for the atmosphere, collecting nearly 0.5 PgC per year [3,4]. Grasslands have an irreplaceable role, as their contribution to climate change mitigation and adaptation improves land and ecosystem health, resilience, biological

diversity regimes, global productivity, and water cycles under future climate scenarios [5]. However, they are also projected to be among the most sensitive ecosystems to drought [6]. Therefore, research on grassland response to drought is of great significance. A comprehensive understanding of these responses would provide significant information to be used in drought mitigation policies and carbon management programs.

There is evidence that droughts have continued escalating on a global scale [7], influencing key ecosystem processes and functions [8]. Simultaneously, extreme climatic events such as droughts are predicted to become more intense, more frequent, and longer lasting in arid and semi-arid regions [7,9]. Droughts could fundamentally alter the composition, structure, and function of grassland ecosystems, posing a far stronger threat to ecosystem functionality than global trends and shifts in average regimes [10,11]. Moreover, while global change drivers such as elevated atmospheric CO_2 concentration, warming, *N* deposition, grazing, and other land-use changes are outside of the grassland ecosystems, they impose chronic, cumulative, and moderate stresses. Extreme events such as droughts, however, often lead to periodic, pulsing, and severe alterations [12]. Long-term declines in grassland productivity had been driven by increased dryness over four decades; to some extent, the water use efficiency that increased through CO_2 enrichment in grasslands may have slightly moderated the decline in production of native C3 grasslands, while variations in *N* had no effects [13]. So it seems that extreme droughts may change the structure, composition, and functionality of terrestrial ecosystems, thereby influencing *C* cycling and its feedback to the climate system [14–16].

Droughts may affect *C* cycling under the influence of global change drivers. However, a full understanding of drought impact on carbon dynamics has not yet been achieved in climate impact research on grasslands. It is not clear whether a combination of drought and global change drivers affect grassland *C* cycling. In this review, however, we found that the relation between drought-derived water stresses imposed on grassland plants is indeed affected by multiple factors and, consequently, changes grassland *C* cycling [17,18].

2. Drought and Grassland *C* Cycling under Global Change

2.1. Overview

In this paper, drought is defined as water stress for plants and includes natural and human-manipulated droughts. The predominant vegetation of grasslands, including the Eurasian steppes, prairies, rangelands, or savannas, is grass [19]. The *C* sequestration and cycling in grasslands include *C* sequestration, allocation, turnover, emissions, and storage, such as GPP (gross primary productivity), NPP (net primary productivity), soil respiration, and SOC (soil organic carbon). The biomass of grasslands is allocated largely below ground with a large root to shoot ratio, which slows decomposition and weathering rates where significant accumulations of SOM (soil organic matter) and highly fertile soils were present [20]. Currently, grasslands possess about 12% of global SOM [21]. The belowground system can play an important role in controlling terrestrial *C* sequestration and cycling.

Grassland may be exceedingly vulnerable to droughts [10], which may result in shifts in the magnitude and patterns of *C* cycling [22]. In addition, effects from other changing parameters may interact with drought and grassland *C* cycling, such as elevated CO_2 concentrations, global warming, *N* deposition, grazing, other land-use change, and grassland ecosystem components (Figure 1) [23]. *C* cycling may mitigate or exacerbate climate change, depending upon the relative responses of grassland *C* sequestration and emissions to global change factors and droughts.
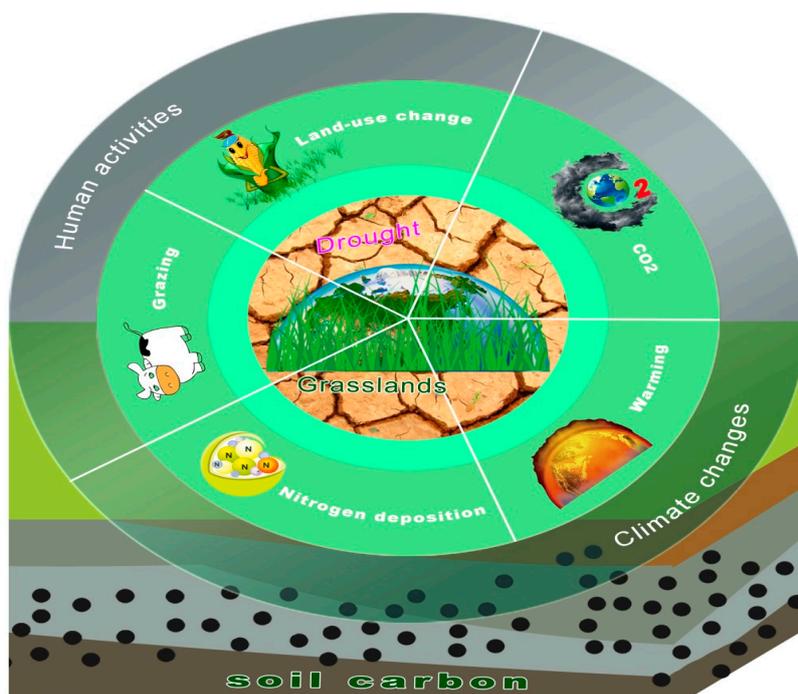
Figure 1. The schematic diagram describes the complex interaction between drought and global change drivers on grassland *C* cycling. In the paper, global change drivers include elevated CO_2 concentrations, global warming, *N* deposition, grazing, and land-use changes. To some degree, the chronic and gradient factors can counteract the negative effects of drought on grassland *C* cycling via their positive effects driven by climate change, such as elevated CO_2 concentrations, global warming, and *N* deposition; on the other hand, the periodic and pulsing factors can amplify the negative effects of drought on grassland *C* cycling via human activities such as grazing and land-use change. In grasslands, the ecosystem has the ability to buffer impacts from drought and large amounts of soil *C* in its surface layers. As climate change and human activities continue, droughts will have a stronger and more complicated effect on *C* cycling in grassland ecosystems in the future.

2.2. Drought and Grassland C Cycling

Water is a limiting factor in grasslands, many of which experience periodic droughts [10,24]. On a global scale, the frequency, duration, and intensity of droughts have increased strongly in recent decades [25], especially in arid and semi-arid regions [7]. Droughts are the main source of inter-annual variation in terrestrial *C* sequestration, as they cause large reductions in GPP as well as in the net ecosystem exchange (NEE) of terrestrial ecosystems [26,27]. Droughts also have negative effects on soil biodiversity, the content of SOM, and water retention [28]; they may have implications on ecosystem functions that last longer than the drought itself [29,30]. Most notably, the ecosystem *C* sequestration accumulated over a number of years could be undone by a single severe drought [26,31]. When severe droughts occur, soil microbes struggling for 'food' resources eat into the 'old' soil *C* accumulated over a number of years in grasslands [32]. Additionally, under future climate scenarios, productivity is predicted to increase in North American grasslands despite rising aridity [33]. Therefore, droughts are key determinants of *C* cycling in grassland ecosystems, as they alter water provision and use [34,35].

C cycling has different responses to various drought intensities, as is supported by many studies [36–38]. Depending on their intensity, frequency, duration, and timing, not all droughts have an equal impact on grassland *C* cycling [39]. Drought intensity partly determines the fate of *C* allocation for photosynthate in grassland plants [40]. However, extreme drought limited *C* translocation from aboveground to belowground storage, while a moderate drought provoked allocation, assigning *C* translocation to the area of the plant where it was most urgently needed at high water stress [41].

In another study, soil CO_2 flux decreased by 8% under reduced rainfall amounts, by 13% under altered rainfall timing, and by 20% when both were combined [42]. Thus, it was suggested that drought timing was a factor more critical than intensity in affecting *C* dynamics in semi-arid regions [43–45]. Similarly, the developmental stage of a plant could be determined by a response to drought [46]. Seedlings are known to be highly susceptible to drought stress [47]. A shift in spring drought impacts the structure and function of grasslands more than a drought in summer/fall for the North American Great Plains [48].

Furthermore, drought has mixed effects on grassland ecosystem processes. In the short term, drought adversely affects root biomass, litter decomposition rates, and short-term CO_2 fluxes, but increases soil nutrient retention, soil fertility, and longer term *C* fixation rates [49]. In addition, droughts may have different impacts on aboveground and belowground productivity or communities [50]. Aboveground organisms grow fast, are more susceptible to drought, and have a quicker recovery rate post-drought, while belowground organisms grow slowly, do not easily suffer from drought, and have slower recovery rates post-drought [51]. Also, plant belowground inputs can affect the recovery of belowground communities after drought [52]. Drought may change plants and soil microorganisms by altering the *C* transfer process at the plant–microbial interface [53,54]. Drought can also change contemporary rates of biogeochemical processes by inducing a shift of abiotic drivers and microbial community structure [40,55].

In hostile environments, droughts can cause the functional thresholds of an ecosystem to shift rapidly, changing a *C* sink to a source [56–58] or reducing resiliency to pests, fires, and disease [59]. In the future, increases in the frequency and intensity of droughts could turn temperate grasslands from *C* sinks into sources, with positive *C*-climate feedback [60]. In fact, ecosystems suffer from the effects of a single event by switching into alternative ecological regimes and cannot withstand the combination of multiple extreme events [61]. Grassland ecosystems are able to withstand moderate drought and maintain ecosystem functions [38,62], but severe, extended droughts may induce catastrophic consequences, such as the 1930s Dust Bowl in North America [12,63]. Droughts can significantly and divergently alter the resilience to new disturbances, such as insects, disease, or the next drought [59]. Droughts can trigger other disturbances such as wildfires [60] and pest invasion [64]. While fire and drought can increase short-term SOC accumulation, their long-term impacts on *C* cycling are still unclear [65]. Droughts interacting with high temperatures can cause plant mortality and accelerate seed germination and ecological invasion due to favorable weather conditions [66]. According to the 'fluctuating resources hypothesis', the rainfall after drought may even enhance the chance of invasion [67]. These effects of droughts on diversity, productivity, reproduction, phenology, nutrient cycling, and community resistance to invasion indirectly affect grassland *C* cycling [68].

Usually, the impacts of droughts are regulated by many environmental factors [69], as well as ecosystem traits, such as species and functional diversity [70,71], and succession timing or growth stage [8]. Therefore, alongside global change and human activity, droughts with novel magnitudes, timing, and durations are out of synchrony with the resistance of ecosystems and will have stronger and more complicated effects on *C* cycling in the future [17,72].

2.3. Elevated CO₂ Concentrations

CO_2 fertilization effects may be affected when droughts occur [73]. An elevated CO_2 environment may also increase the water use efficiency of plants and thus soil moisture—due to the reduction in stomata conductance—and alleviate the impacts of drought [74]. Elevated CO_2 could mitigate the effects of drought on grassland net carbon uptake by increasing root growth and plant *N* uptake [75]. In fact, elevated CO_2 directly or indirectly affects plant water loss and may be crucial to understanding the combined effects of drought on the *C* cycling processes of ecosystems. The long-term effects of CO_2 on ecosystem functions are more likely to be indirect responses, such as changes in the biochemical cycle, soil moisture, and species structure, than direct responses through exchanges in production [76]. Elevated CO_2 not only increases long-term mean soil moisture [77], but also enhances microbial

biomass and density [11]. To a certain extent, rising atmospheric CO_2 concentrations might decrease the vulnerability of grassland *C* to droughts [78].

Atmospheric CO_2 enrichment can enhance NPP and mitigate the negative effects of droughts on GPP and NEE [31,79]. Likewise, elevated CO_2 concentrations alleviate the negative effects of droughts on soil respiration, principally due to the promotion of carbon assimilation, which increases the substrate supply for respiration in both roots and soil microorganisms [80]. Given a number of adverse environmental and edaphic conditions, the increased resource use efficiency of plant growth under high atmospheric CO_2 concentrations cannot prevent a decline in productivity and quality [78]. In one study, elevated atmospheric CO_2 reduced the sensitivity of grassland ecosystems to drought and increased grassland productivity by ~5%–15%, depending on water and nutrient availability [81,82]. However, CO_2 fertilization decreases when water and *N* are limited. In fact, these positive effects are unlikely to offset the negative impacts of high temperature changes and decreased summer rainfall, which would lead to more frequent and intense droughts [83]. Consequently, beneficial CO_2 fertilization effects are suppressed, and water use efficiency is not enough to compensate for the negative effects of droughts [84]. As the intensity and duration of droughts increasingly result in stomatal closure, the ecosystem can become CO_2 limited, which may stimulate *C* starvation and hydraulic failure [85,86]. Concurrently, droughts can also lead to increasing GHG (Green House Gas) emissions, limiting the vegetation *N* supply, and change the amount of fixed carbon [87].

The effects of concurrent elevated CO_2 levels and droughts on a plant's water use efficiency are intertwined. As global climate models predict rising temperatures, it is important to acknowledge that CO_2 concentrations will interact with the change in precipitation patterns, thereby affecting *C* cycling. In dry soils, elevated CO_2 concentrations coincide with or even contribute to drier conditions, resulting in negative soil respiration responses to temperatures [88]. Elevated CO_2 concentrations and extended droughts show a positive interaction, and elevated CO_2 can reduce the effects of drought on soil respiration [80]. In contrast, elevated CO_2 concentrations restrain diversity, but have little effect on the relative abundance or the production of a community [89]. Thus, those effects indicate the importance of a multi-factor experimental approach to understanding an ecosystem's response to droughts [18]. Indeed, the interactive effects of multiple factors on grassland *C* cycling under different drought scenarios are lacking in the literature, especially studies discussing underlying mechanisms. Therefore, further work is needed to evaluate the integrated impacts of multiple factors on grassland *C* cycling under different drought scenarios.

2.4. Global Warming

Global warming may accelerate the turnover of water [68]. Warming is often accompanied by drought and could reduce the primary production in many temperate grasslands, which is not necessarily mitigated by efforts to maintain or increase species richness [90]. In grassland communities, mild droughts and warming do not lead to enhanced resistance or recovery from an extreme recurrent drought. Also, grasslands experiencing recurrent drought demonstrate a larger decrease in green vegetation cover [91]. For some drought-affected areas, however, annual NPP and NEE are not reduced, likely because higher temperatures enhance photosynthesis, counteracting the drought effects on NPP in those grasslands [91,92]. Meanwhile, it is expected that climate warming may directly accelerate soil respiration by stimulating the activities of soil fauna, microbes, and plant roots, and may indirectly accelerate soil respiration by stimulating *N* mineralization, litter production, and substrate [93,94]. Warming and prolonged droughts may strongly alter SOM decomposition, but also the quantity and quality of litter input [95,96].

In general, under drought scenarios, warming can cause further aridity of the ecosystem and hence act to further reduce soil respiration. In contrast, many experiments have shown that rising temperatures increase the rate of soil respiration [97,98]. In addition, warming can cause droughts that increase the carbon in roots and weaken the overall acclimation of plants to drought by regulating *C* allocation between source and sink organs [41]. It is expected that rising temperatures will affect

decomposition more than primary productivity [99], the consequence being a net loss of soil *C* and a positive feedback to the climate system in the long term. However, there is still no agreement on how temperature sensitivity varies with the labiality of SOC [100]. Instead, warming and drought have a direct impact on soil *C* storage, mainly by altering the mineralization rates of SOM [101]. Also, warming likely affects ANPP in grasslands, and warming effect is moderated by shifts in the C3/C4 ratios of plant communities [102]. Forecasting *C* cycling between droughts will interact with elevated temperatures in the future has been a significant subject of many models. However, to date, a clear mechanism to cope with this interaction has not been developed.

2.5. N Deposition

CO_2 fertilization effects can depend on the amount of available nitrogen and water [73]. Therefore, nitrogen becomes a limiting factor when droughts occur due to strong interactions between water and *N* [17]. Although variances in droughts can have different effects on ecosystem productivity and *C* cycling [10], how these might interact with *N* deposition is not clear.

There are more rapid ecosystem alterations caused by the interactive effects of *N* deposition and droughts than when *N* deposition or droughts occur alone [12]. Drought and *N* additions increase enzymatic efficiency and induce faster decomposition of litter [103]. In the Mongolian steppe, elevated *N* deposition can enhance the recovery of grassland productivity after drought [104]. Drought can cause nutrient deficiencies, even in fertilized soil. The increased duration and intensity of drought are usually linked to decreasing *N* mineralization [105], mobility, and absorbance of inorganic nutrients [106,107]. Moreover, plant biomass, *N* concentration levels, and the amount of *N* in the whole plant increases by adding water after a short-term or medium-term drought, while they decrease after a long-term drought [108]. *N* addition increases grassland productivity after a drought [104]. In contrast, *N* fertilization has been shown to reduce the ability of grassland ecosystems to sustain net CO_2 assimilation. *N* addition enhances within-plot variability in plant size structure at the species level, but did not change total aboveground biomass [109]. Indeed, droughts affect soil respiration by influencing the feedback from soil *N* pools, because the microbial processes that regulate soil *N* availability are sensitive to short-term variations in soil moisture [110]. Meanwhile, *N* and water supplies limit potential NEE. Conversely, soil *N* availability has little effect on the short-term stability of ecosystem processes to extreme drought [49]. Some results suggest that increases in ANPP associated with *N* deposition may be offset by precipitation-mediated shifts, with negative consequences for the strength of the ecosystem *C* sink [111,112]. For grasslands, the frequency and intensity of the disturbances have a key role in *C* balance [113]. Thus, annual grasslands experiencing higher rates of *N* may provide lower-than-expected *C* storage. In short, how *C* storage responds to *N* depends on the longer term variations of droughts.

2.6. Grazing

Grazing has been a way of life and a common land use for people in arid and semiarid regions for centuries [114]. Grazing may have uncertain impacts on grassland *C* cycling [115], as it can have high intensity and relate to drought [116]. Low-intensity grazing coupled with droughts can impact aboveground productivity (AGP) and belowground productivity (BGP) [117]. Moderate grazing was less important than droughts in reducing plant cover, but heavy grazing was at least as influential [118]. Sensitivity to droughts can override grassland productivity, significantly altering *C* dynamics in grazing management systems [119]. The interaction between grazing and droughts can change diversity, alter dominance, and affect productivity [120]; it also restrains grass stem density and delays recovery in mesic grasslands in North America and South Africa [121]. In grazed and ungrazed North American tallgrass prairie sites, ANPP was relatively resilient under drought. Moreover, grasslands with different evolutionary histories respond similarly to grazing and drought. At frequently burned sites, droughts increase grassland sensitivity to grazing. The effects of droughts were not the primary driver of grassland productivity; instead, the magnitude of the effects is similar to those of grazing and fire [122].

Grazing also interacts with droughts to affect BGP, and grazed grassland can be more or less sensitive to changes in precipitation than ungrazed grassland [38]. Frequently grazed grassland has a higher probability of experiencing a drought during a regrowth stage. Drought controls major trends in plant species composition and production, with grazing playing a secondary role [123]. The role of rangelands as sinks or sources of GHG, however, is determined by complex interactions between drought, vegetation, and grazing.

Collectively, grazing and drought have uncertain effects on grassland *C* dynamics, contingent on the intensity of grazing and the drought as well as grassland type. In the event of extreme drought gives way to a wet year, both enhanced GPP and ER cause greater *C* uptake on the grazed (uptake 6 g·C·m^{-2}) than on the ungrazed desert steppe (release 43 g·C·m^{-2}). In an extreme year-long drought, a grazed desert steppe (release 70 g·C·m^{-2}) has greater *C* release due to reduced GPP than ungrazed (release 48 g·C·m^{-2}), but the grazed and ungrazed steppes have comparable ecosystem respiration [117]. The primary challenges in maximizing the potential of rangeland GHG mitigation are to create a complete account of GHG balances across many rangeland ecosystems and to quantify the magnitude and direction of GHG changes, due to interactions between management and environment [119]. Long-term monitoring is needed to thoroughly document mechanisms contributing to *C* dynamics in semi-arid rangelands in order to fully understand their role as CO_2 sinks, given the anticipated trajectory of future climate change [57,124].

2.7. Land-Use Change

Land-use change enhanced the vulnerability to drought and posed a threat to grasslands ecosystem. Land-use and management change can strongly influence variations in grassland SOC [125]. The impacts of land-use change on drought mostly focus on land-use change as a driver of climate change [126]. Grasslands could be influenced by potential variations in droughts and by alterations in land-use changes [127]. Land-use change and droughts may induce multidirectional impacts on an ecosystem [128]. In addition, land-use change can cause a decline in grassland biodiversity and plant cover [129]. Land use can also strongly affect the resistance and resilience of soil food webs to drought [130]. Drought has significant effects on vegetation and may enhance the risk of wildfires [131]. Also, large-scale and destructive shifts in grasslands can change climate conditions and affect vegetation regimes and the surface energy budget, deteriorating water depletion [132,133]. However, drought and land-use change can deteriorate soil erosion, causing soil *C* to be lost at a faster rate and in larger amounts [134]. It is difficult to distinguish which one is the driver. Land-use change can influence the frequency and duration of drought [135]. Changes in land-use and plant species components can affect the decomposition of litter via a number of mechanisms, such as alterations of litter quality, but also reshape the pattern of temperature and moisture at the soil surface [136,137]. Furthermore, grasslands serve as a significant *C* sink via improved management, and are highly regulated by biome type and climate conditions [115]. To mitigate drought effects on SOC, land-use change is important to avoid in management practice within a dryland ecosystem [2,138]. Drought and land-use change have a direct impact on the *C* source/sink function of a grassland ecosystem, and may be major drivers affecting the *C* budget of grasslands.

2.8. Grassland Ecosystem Properties

To a certain extent, the effects of drought on an ecosystem are related to the traits of the dominant species, as well as plant functional diversity [139]. The ecosystem functions and species composition of grasslands are likely to be impacted by droughts [140]. Drought may have pronounced effects on functional performance, such as C-fixation as well as fluxes and pools [36,56]. However, a grassland's plant diversity provides a buffer against environmental fluctuations because different species respond differently to these fluctuations, triggering functional compensations among species [11,141]. Plant diversity has a stronger effect on soil microbial functions and enzymes [142]. Plant diversity is a major determinant of soil microbial biomass in a changing environment [143]. Specifically the ability for a

community to maintain ecosystem functions during a stressful event (resistance) or to recover rapidly from it (resilience) is a component of stability [144]. Likewise, a grassland may withstand moderate drought in areas with rich biodiversity [104,145]. In particular, greater ecological diversity means higher resistance to drought because of the complementary use of available water and other resources when compared to communities with low ecological diversity [146]. Accordingly, at the species level, the individual may not respond to drought, but the ecosystem significantly responds [147]. Because of high diversity, the ecosystem exhibits greater stability and ensures species persistence and ecosystem functions [71,148]. Post-drought recovery of an ecosystem is much more rapid where greater levels of biodiversity are conserved than in less diverse areas [149]. On the other hand, droughts are not buffered by increased biodiversity richness. Intriguingly, frequent mild droughts did not change the productivity patterns and point to a higher resistance to severe droughts, with increasing richness levels not necessarily enhancing resistance [91]. Similarly, higher drought stress with increased biodiversity richness led to greater mortality, yet all communities are able to recover similar green cover post-drought [150]. In fact, another threat to *C* sequestration in grassland stems from the rapid loss of plant diversity, which is projected under climate change [151].

In a grassland, soil plays an important role in buffering the impacts from drought. Plant–soil interaction plays an important role in regulating *C* cycling [152]. The adaptive strategies of plant–soil interactions play a key role in the short-term stability of *C* cycling to extreme drought events [49]. To better withstand times of drought, increased amounts of *C* sequestered as SOM into soil enhance rainfall effectiveness through increased infiltration, water-holding capacity, and water source replenishment. Moreover, grasslands may be most likely missing *C*-sinks [3,113], owing to grasslands potential capacity to sequester and store *C* in soils [5,153]. Higher soil *C* levels can reduce the impacts of droughts [154,155]. The relative extent of drought impacts on soil respiration depends on the level of belowground biomass and soil *C* [156]. Sequestering *C* in grassland soils brings about a number of positive environmental outcomes, or co-benefits, beyond offsetting GHG emissions [157]. SOM plays an important role in determining soil chemical properties, including pH, nutrient availability and cycling, and buffer capacity [158]. Thus, increasing SOM is an effective method for increasing drought resistance in arid and semiarid areas.

3. Syntheses and Perspectives

3.1. Combined Effects of Multiple Factors

Ecosystems exist in a multi-factor world. Global change drivers can mediate the rate and efficiencies of both photosynthesis and water use to affect ecosystem productivity and other processes. Realistic combinations of global change drivers show small diversity effects, but a remarkable effect on dominant species [159]. In addition, global change drivers can alter available resources in an ecosystem by directly affecting the biota and driving ecosystem responses, causing chronic changes in water balance and modifying the biogeochemical cycle of nutrients [160]. Drivers can also change ecosystem nutrient dynamics indirectly by affecting plant litter quality [161]. The natural ecosystem response to global change drivers may be constrained by different perturbations [162]. Collectively, current disturbance regimes and global changes impose a suite of impacts on available resources in an ecosystem [163]. Effects from global change drivers and droughts may interact within ecological diversity and in the composition of natural communities [164]. *C* cycling may mitigate or exacerbate climate change, depending upon the relative responses of grassland *C* sequestration and emissions to global change factors and droughts [165].

Climate, fire, and grazing are three important drivers affecting the composition, structure, and function of grasslands. There are also many interactions between these drivers, which affect ecological patterns and processes in grasslands differently than if they were single drivers [166]. Grassland composition and diversity are primarily governed by long-term regional climate. To some extent, grassland composition and diversity can alleviate the impacts of a drought on an ecosystem [121]. In a

warmer world, elevated CO_2 concentrations will increase both soil water content and productivity in semi-arid grasslands [167]. Adversely, elevated CO_2 alone, or in combination with warming, alters ER to a greater extent than GPP, resulting in net *C* loss by the stimulated decomposition of SOM. For instance, an elevated CO_2 concentration causes a greater increase in *C* cycling than in *C* storage in grasslands [168], which can alleviate or offset the impacts of a drought on *C* cycling. Also, anthropogenic pressures such as grazing and land-use change could be key drivers of biodiversity loss, with serious consequences for ecosystem functioning [12]. Simultaneously, the combined effects of environmental factors have great potential to interact and indirectly or directly mediate soil moisture, affecting the main process of *C* cycling [165].

Similarly, analyses of the roles of other factors in grassland *C* cycling or ecosystem functions should not overlook the influence of drought. The contribution of each factor to grassland *C* cycling should be diagnosed and quantified, especially with regard to influencing the relationship between droughts and grassland *C* cycling in a realistic multi-factor world. Clearly, there has been much progress in the sophistication of both models and experiments. It is no longer considered acceptable to make projections about drought response without incorporating the likely effects from other factors, such as elevated CO_2 concentrations and global warming [169]. Doing so is significant in discussing how and why ecosystems have different sensitivities to chronic global change and pulsing droughts [12].

The multi-factor effects differ greatly from simple combinations of single-factor responses because the impacts of abrupt changes and alternative multi-factors induced nonlinear changes in the ecosystem [18]. Concurrent changes in multiple factors potentially trigger complex interactive impacts on ecosystem structure and functioning. For example, CO_2 enrichment restrains the effects of increasing temperature, precipitation, and available *N* on NPP annually in Californian grasslands [170]. Moreover, the amplification or suppression of one factor's impact by another factor on soil *C* has not been observed in most studies [161,171]. These effects are the key to identifying the most sensitive factors affecting *C* cycling. Therefore, evaluating multi-factor interactions influencing ecosystem structure and functioning is critical to understanding their response to global change. Indeed, once interactive effects can regulate the main effects of single factors, single-factor experiments will become less useful in understanding ecosystem changes [171]. There are a number of single factors that help assess the impacts of global change, such as rainfall manipulation experiments [10], FACE experiments, and N-addition experiments [12]. A multi-factor experimental approach explains ecosystem responses to multiple factors, especially under different drought stress scenarios [172].

Usually, multi-factor experiments are quite expensive and provide imprecise results and undefined interactive mechanisms. They also cannot be properly managed in many ecosystems due to financial constraints. To make multi-factor experiments more effective, models can help imitate the formation of a scientific hypothesis in its initial stages and extrapolate experimental results [171]. Ecosystem models should incorporate the direct and indirect effects of climate change on soil moisture to accurately predict drought feedback and the long-term effects of *C* cycling. Models can be informed by single-factor experiments that provide ecosystem-level information for single-factor responses. Multi-factor experiments are important for testing concepts, thus indicating the reality of multi-factor influences. However, large uncertainties remain in most current models that evaluate the feedbacks between *C*-cycle and climate change over the past few decades.

Fortunately, close cooperation between experimentalists and modelers make it easier to understand water and *C* cycling via explicit programs of model–data fusion, such as data sharing, data assimilation, and clarity of model processes. A major improvement in current observations may be gained by the combination of long-term, multi-factor experiments at the ecosystem level, such as whole-ecosystem flux measurements. To some extent, the coupled *C*-cycle–climate models can demonstrate the importance of potential carbon-cycle–climate system feedbacks [17]. However, much work towards these experiments and models is needed to increase our understanding of the mechanisms between drought and grassland *C* cycling in the multi-factor world at the ecosystem level. In general, the emerging gaps are as follows:

(1)　Under particular scenarios such as droughts, recognizing the key environmental factors impacting grassland *C* cycling in the real, multi-factor world is important. It is necessary to carry out multi-factor experiments to determine the contribution of each factor and their interactive effects with other factors on grassland *C* cycling at the ecosystem scale.

(2)　The quantitative impacts between drought and other factors (e.g., elevated CO_2 concentrations, global warming, *N* deposition, grazing, and other land-use change) on grassland *C* cycling are not clearly proposed at the ecosystem level.

(3)　The next challenge is to establish the quantitative relationships between different *C* fluxes and different global change factors under different drought scenarios as soon as possible.

(4)　Multi-factor response models should be developed with better coupling mechanisms to examine the interactive effects of global change multi-factors on the carbon and water processes of ecosystems, especially under different levels of drought stress or other extreme scenarios. The data–model fusion has become essential to assess the interactive effects of multiple factors in global change research.

3.2. A Framework for Assessment and Application of Combined Effects

Practical fluctuations with the background of global changes would result in an intricate blend of internal processes and external forces on ecosystem, which can trigger a regime shift in grasslands [173]. According to the 'fluctuating resources hypothesis', ecosystems become more susceptible to biological encroachment whenever the amount of idled resources increases in two basic ways: declines in the supply and use of ecosystem or external resources. A disturbance such as drought, grazing, or other land-use change could damage or kill resident vegetation and reduce resource uptake (light, water, and nutrients) [174]. Drought coupled with global change drivers or with favorable weather conditions can induce plant mortality or promote seed germination and ecological invasion, respectively [175]. It is clear that several interacting global change drivers and droughts trigger shifts in ecological thresholds [12].

However, there is not enough knowledge about how drought affects biodiversity and ecosystem function. The framework describes a hierarchy of mechanisms that climate change uses to impact ecosystem *C* dynamics, generating three levels of response, and also provides a new approach to the studying climate change impacts on *C* cycling [176]. The expression of functional thresholds is affected by many factors including droughts, other global change drivers, and ecosystem characteristics. Combined multi-factor effects may make ecosystems surpass ecological thresholds, resulting in a reconfigured ecosystem structure and function and a profound influence on *C* cycling. Rapid ecosystem shifts are triggered by droughts occurring within the background of gradual global change, overgrazing, land-use change, and invasive species [177], and is projected to increase in the future because ecosystem resilience is corroded by chronic global changes [12]. The interactions between drought and global change drivers influence and accelerate the trajectories of ecosystem response [178]. An ecotone shift caused by drought has severe consequences on ecological and accelerates soil erosion, which induces large losses of soil *C* [173]. The shifts of grassland composition in response to seasonal or annual precipitation regimes represent drought-induced alterations of vegetation dynamics that may trigger threshold development [12]. Ecological thresholds must further evaluate and develop to effectively manage grassland *C* cycling and fully assess the ecological consequences of climate extremes for ecosystem structure and function [179]. Threshold recognition and prediction is significant to help managers prevent the emergence of undesirable states and promote the management of grassland *C* cycling [180]. We do not know the ecological mechanisms trigger the ecological threshold, or the speed and degree of response to the triggering threshold. Therefore, a systematic framework that assesses ecosystem dynamics in response to drought under current and future global change and human activity is urgently needed to focus on the combined effects that drought and global change drivers have on an ecosystem and to assess the hierarchical response of that ecosystem [181,182]. More research is needed to create a framework that meets the following needs:

(1) Defines the intensity and duration of drought at which ecological thresholds are triggered;

(2) Represents ecological mechanisms of response to drought under current and future global change scenarios at different spatial–temporal scales;

(3) Assesses the hierarchical responses of an ecosystem to drought and global change, including individual, species, and ecosystem responses;

(4) Quantifies the contribution of other global change drivers that prevent ecological thresholds triggered by droughts;

(5) Provides a baseline to assess the impact of drought under global change;

(6) Assesses the contribution of ecological thresholds to the fate of grassland *C* cycling;

(7) Gives suggestions to managers can use to enhance *C* sink of grasslands.

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