N and P Addition Reduce Semiarid Grassland Stability via Increasing the Dominance of Less-Stable Functional Groups

Zhifei Chen

State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau

Peifeng Xiong

Anhui Agricultural University

Junjie Zhou

State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau

Shuaibin Lai

State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau

Chunxia Jian

State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau

Weizhou Xu

Yulin University

Bingcheng Xu (✉ bcxu@ms.iswc.ac.cn)

State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau <https://orcid.org/0000-0002-1702-491X>

Research Article

Keywords: grassland stability, N and P fertilization, compensatory effect, asynchrony, functional group stability

Posted Date: February 17th, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-213567/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License. [Read Full License](#)

Abstract

Aims Extraneous fertilization could efficiently improve grassland productivity and promote grassland restoration. Increasing fertilization may profoundly affect community stability, whereas the underlying compensatory dynamics among species and functional groups in regulating grassland stability remain unclear.

Methods Three different grasslands, annuals and biennials (AB) community, perennial grass (PG) community and perennial forb (PF) community, on semiarid Loess Plateau were selected. We designed a 3-year split-plot experiment (main-plot: 0, 25, 50, and 100 kgN ha⁻¹ yr⁻¹; subplot: 0, 20, 40 and 80 kgP₂O₅ ha⁻¹ yr⁻¹) to explore how N and P addition affects community stability and its relationship with species richness, species asynchrony (SA) and functional group stability.

Results Temporal stability differed largely between functional groups under N and P addition, perennial forbs or grasses had higher stability than perennial legumes or annuals and biennials. Decreased stability of PG and PF communities was primarily due to increased dominance of perennial grasses or forbs and reduced SA under N addition alone, while it attributed to increased dominance of perennial legumes after P addition alone. 50 and 100 kgN ha⁻¹ yr⁻¹ combined with P addition notably increased dominance of annuals and biennials at the expense of perennial species, but decreased stability of annuals and biennials, which caused declines in stability of the three communities.

Conclusions Nutrient-induced shifts of functional groups, particularly increased dominance of less-stable functional groups, is the main driver reducing perennial grassland stability independent of diversity. Whereas N and P addition decreased annual grassland stability via additive effect of increased dominance of annuals and biennials and diversity decline. Targeted N and P combined addition was essential to maintain stability for different grasslands by regulating functional groups on semiarid Loess Plateau.

Introduction

Primary production and ecological process of natural grasslands are mainly co-limited by nitrogen (N) and phosphorus (P) (Fay et al. 2015; Harpole et al. 2011). Exogenous nutrient application has become an effective strategy to improve grassland production and promote vegetation restoration, as indicated by grassland studies in Europe (Fay et al. 2015; Lepš 2014), Australia (Rawnsley et al. 2019), North America (Avolio et al. 2014; Suding et al. 2005) and China (Bai et al. 2010; Yang et al. 2011). However, continued and increased fertilization often caused biodiversity reduction, species composition changes and soil acidification, threatening sustainability of ecosystem function (Hautier et al. 2014; Zhou et al. 2020). Temporal stability of productivity provides information about an ecosystem's ability to sustain sustainable functioning and services under environmental fluctuations (Craven et al. 2018; Isbell et al. 2009). Many studies have demonstrated the stabilizing effects of biodiversity on ecosystem stability, and suggested an integrative underlying mechanism for sustaining stability, including species asynchrony and compensatory dynamics, overyielding effects and portfolio effects (Grman et al. 2010; Melts et al. 2018). The underlying mechanisms in determining stability vary considerably among different ecosystems (Grman et al. 2010; Liu et al. 2018; Zhou et al. 2020).

The stabilizing effects of diversity on community productivity is mainly benefited from temporal asynchrony, occurring at both species and functional group levels (Craven et al. 2018; Huang et al. 2020). Species belong to different functional groups are more asynchronous in response to environment changes, whereas species within a specific functional group are more likely to perform synchrony (Collins et al. 2012; Mackie et al. 2013; Marquard et al. 2009). Meanwhile, different functional groups in a community potentially exhibit different intrinsic stability under environmental fluctuations (Tredennick et al. 2017). For example, perennial, tall and clonal species generally perform more stable than annuals, short and non-clonal species (Bai et al. 2004; Wang et al. 2017). High community-level stability is associated with dominance of species or functional groups with high stability. Dominant species or functional group usually had high stability than

subdominant and rare species because of long-term environmental adaptation (Sasaki and Lauenroth, 2011; Smith and Knapp, 2003). Community stability is primarily regulated by stability of dominant species or functional groups in short-term, whereas subdominant and rare species regulate stability in longterm by influencing biodiversity and species asynchrony (Sasaki and Lauenroth, 2011; Song et al. 2019).

N and P fertilization may influence community stability via multiple facets (Isbell et al. 2009; Liu et al. 2019a). Firstly, N and P addition could cause biodiversity decline and thus reduce community stability by decreasing species asynchrony, portfolio effect and overyielding effect (Hautier et al. 2014; Loreau and De Mazancourt, 2013; Marquard et al. 2009). Secondly, the reduction of community stability after N and P addition was also associated with large shifts in species composition even if biodiversity decline is not occurred (Huang et al. 2020; Liu et al. 2019a). This may be due to that dominant species shift from species with high stability to low stability after N and P addition (Mackie et al. 2019; Yang et al. 2011). Thirdly, increased plant height and community productivity may translate into changes in competition strength among species after N and P addition, which impacts species synchrony (Niu et al. 2018; Xu et al. 2015). Strong competition may benefit some opportunistic species with similar traits and allows them perform more synchronously (Niu et al. 2018; Song et al. 2019). Short species are susceptible to be suppressed by tall species because of light limitation after N and P addition (Collins et al. 2012; DeMalach et al. 2017). Clonal species perform high competitiveness for light and water than non-clonal species because of resource sharing via vegetative growth (Chen et al. 2020; Dickson, Mittelbach, Reynolds, and Gross, 2014).

Grassland accounts for nearly 40% of the national land in China. However, about 61% of grassland is experiencing serious degradation due to human activity in the northwest China (Liu et al. 2019b). The semiarid Loess Plateau is typical vulnerable ecoregion because of soil erosion, over-grazing and frequent drought (Deng, Shangguan, and Sweeney, 2014; Zhang, 2005). The “Grain-for-Green” eco-restoration program was proposed since the 1990s to promote revegetation across the Loess Plateau (Deng et al. 2014; Gang et al. 2018). Up to now, grassland has accounted for about 42.86% of area, which has various grassland types dominated by different species due to different abandoned years and terrain conditions (Gang et al. 2018; Zhang and Dong, 2010). During the early succession, the dominant species are annual and biennial species, and community composition and function are simple and unstable. Perennial species progressively become dominant species in 6-15 years after abandonment, leading to a high diversity and stability. As succession extends to 15-40 years, the stable communities mainly comprise perennial Poaceae, Asteraceae and Fabaceae (Chen et al. 2020; Zhang, 2005; Zhang and Dong, 2010).

Grasslands production and ecosystem services are widely limited by low soil nutrient availability in this region (Zhang and Dong, 2010). Extraneous fertilization could efficiently improve grassland productivity and functions (Chen et al. 2020). However, reconciliations of productivity improvement and stability conservation among various grasslands are community-specific and rate-dependent when using N and P fertilization. The relative importance of biodiversity decline, species and functional group asynchrony, functional group stability for determining community-level stability in this region under N and P addition remains unclear. Thus, a three-year N and P addition experiment in three representative grassland communities were conducted on semiarid Loess Plateau, to examined the effects of N and P addition on community stability and its relationship with species richness, species and functional groups asynchrony and functional group stability. The specific aims were to: (1) clarify the effect of different N and P addition on species richness, species asynchrony, functional group asynchrony and stability. (2) assessed the relative importance of diversity and compensatory dynamics of functional groups in maintaining stability of grassland communities after N and P addition. (3) explore the specific underlying mechanism in stabilizing productivity of different grasslands under different N and P addition.

Materials And Methods

Study site

The study site is located in the Zhifanggou watershed at Ansai district (36°42'42"–36°46'28"N, 109°13'46"–109°16'03"E, 1010–1431 m altitude), Yan'an City, Shaanxi Province, northwest China. The region is characterized by semiarid temperate climate with a mean annual temperature of 8.8 °C (min. -6.9°C in January and max. 22.6°C in July) and a mean annual precipitation of 528.8 mm (1951-2016), of which about 82.2% falling during growing season (May–September). The zonal soil is mainly Huangmian soil (Calcaric Cambisol, FAO classification), developed on wind deposited loess parent material. The landform and vegetation are representative loess hilly gullied landscape. The common vegetation in this region are mainly xeric herbaceous species including *Bothriochloa ischaemum*, *Artemisia gmelinii*, *Artemisia giraldii*, *Lespedeza davurica*, and *Stipa bungeana*.

Experimental design

Based on succession process, one representative grassland in early successional stage (annual and biennial (AB) community), two representative grasslands in late successional stage (perennial grass (PG) community and perennial forb (PF) community) were selected in 2017. Geographical and vegetation information of the three grassland communities before N and P additions as shown in Table 1. At each community, a 20 × 30 m area was fenced for grazing exclusion in early May 2017. A split-plot field experiment with N addition in main-plot and P addition in subplot was conducted from 2017 to 2019. Each community consisted of three randomly blocks. Each block was split into four 4.0×4.0 m main-plots and each main-plot split into four 2.0×2.0 m subplots. Four N addition rates (0, 25, 50, and 100 kg N ha⁻¹ yr⁻¹; coded as N0, N25, N50, and N100) were applied to main-plots as calcium ammonium nitrate [5Ca(NO₃)₂·NH₄NO₃·10H₂O]. Four P addition rates (0, 20, 40 and 80 kg P₂O₅ ha⁻¹ yr⁻¹; coded as P0, P20, P40, and P80) were applied to four subplots randomly as triple superphosphate [Ca(H₂PO₄)₂·H₂O]. The N0P0 treatment was regarded as control. Each community included 12 main-plots and 48 subplots. The N and P fertilization were applied manually to each subplot once a year during natural rain event in early growing season on 4 June 2017, 21 May 2018 and 13 June 2019, respectively. Precipitation data was obtained from meteorological station in the Zhifanggou watershed (Fig. 1).

Plant sampling

Vegetation sampling were carried out in late August from 2017 to 2019. A 1.0×1.0 m quadrat was setup to measure abundance, coverage and height of each species in each subplot. The 1.0×1.0 m quadrat was quartered into four 0.5×0.5 m parts to measure the coverage of each species and whole community. Then two 0.5×0.5 m parts were clipped to ground level. To describe compensatory dynamics of functional groups, we classified species into perennial grasses (PG), perennial legumes (PL), perennial forbs (PF) and annuals and biennials (AB). Biomass of different functional groups were oven-dried for 48 h (80°C). Relative biomass contribution of different functional groups was calculated by the percentage of functional group biomass to community biomass.

Soil sampling and analysis

In each subplot, three 0–20 cm soil samples were collected and mixed into one composite sample per subplot after vegetation investigation. Soil samples were air-dried and sieved through 0.25-mm mesh for soil total N and P measurement. Soil total N was determined following Kjeldahl digestion (Kjektec System 2300 Distilling Unit, Foss, Sweden). Soil total P was determined by molybdenum-antimony colorimetric method after wet digestion with sulphuric acid and perchloric acid.

Statistical analysis

Species richness (*S*) and Simpson's dominance index (*D*) were selected to estimate diversity, *D* was calculated as (Niu et al. 2018):

$$= 1 - \sum_{i=1}^S P_i^2 \quad (1)$$

P_i represent the relative important values (IV) of species i , which was calculated as: IV = (relative abundance + relative height + relative coverage)/3.

Temporal stability in both community and functional groups levels was quantified as μ/σ , where μ is the interannual mean aboveground biomass and σ is the interannual standard deviation during 2017-2019 (Huang et al. 2020).

Species and functional group asynchrony ($1 - \phi_b$) were quantified as:

$$1 - \phi_b = 1 - \frac{\sigma_{bT}^2}{(\sum_{i=1}^S \sigma_{bi}^2)} \quad (2)$$

where ϕ_b is species and functional group synchrony, σ_{bT}^2 is temporal variance of community aboveground biomass over 2017-2019 and σ_{bi} is temporal standard deviation of species and functional group i aboveground biomass over 2017-2019 (Huang et al. 2020)

Structural equation modelling (SEM) was used to analyze hypothetical paths how N and P addition impacted community stability in the three communities. In the SEM analyses, we assumed that N and P addition potentially altered community stability indirectly through functional groups stability, species richness and species asynchrony. SEM analyses were conducted using AMOS 24.0 (IBM SPSS Inc, Chicago, IL, USA). The goodness of models fit were based on both chi-square (χ^2) test ($0 \leq \chi^2/df \leq 2$ and $0.05 < p \leq 1.00$) and root mean square error of approximation ($0 \leq RMSEA \leq 0.1$ and $0.10 < p \leq 1.00$) (Xu et al. 2015).

The relationship heat map between the relative biomass contribution of different functional groups, Simpson's dominance, species richness, species asynchrony, functional group asynchrony and community stability were run in R 3.5.2 (R Development Core Team) using packages corplot and car. SPSS 20.0 (IBM SPSS Inc, Chicago, IL, USA) was used to test the least significant difference at $p < 0.05$.

Results

Precipitation

Annual precipitations were 485.6 mm and 464.4 mm in 2017 and 2018, respectively, which were obviously lower than mean precipitation of 1951-2016 (528.8 mm). From May to September, growing season precipitations of 2017 and 2018 (2017: 328.8 mm, 2018: 399.8 mm), which account for 67.7% and 86.1% of annual precipitation, were lower than mean of 1951-2016 (434.8 mm). In 2019, the annual precipitation (538.6 mm) and growing season precipitations (424.2 mm) was similar to mean of 1951-2016.

Community and functional group stability

For AB community, community stability decreased significantly after N50 addition alone as compared to no N and P addition (N0P0) ($p < 0.05$). Community stability had no significant response to P addition alone or combined with N25 compared to N0P0, whereas it decreased significantly after N50 and N100 combined with P40 and P80 addition ($p < 0.05$; Fig. 2). Perennial forb stability was significantly higher than perennial grass and perennial legume stability across all N and P treatments (Fig. S3). Only annual and biennials stability decreased significantly under N50 and N100 addition

alone compared to N0P0. Annual and biennials stability decreased significantly after N50 and N100 combined with P addition compared to N0P0, while significant reductions of perennial forb stability occurred at N100 combined with P addition ($p < 0.05$; Fig. 3).

For PG community, community stability decreased significantly after N50 and N100 addition alone or P addition alone compared to N0P0 ($p < 0.05$). Community stability showed no significant reduction after N25 combined with P addition compared to N0P0, while the significant decreases were found under N50 and N100 combined with P addition ($p < 0.05$; Fig. 2). Perennial grass and perennial forb performed significantly higher stability than annual and biennials and perennial legume across all N and P treatments (Fig. S3). Only annual and biennials stability decreased significantly after N50 and N100 addition alone. Perennial legume stability showed significant decreases under N25 combined with P addition compared with N0P0 ($p < 0.05$). Annual and biennials stability after N50 and N100 combined with P addition showed significant reductions compared to N0P0 ($p < 0.05$; Fig. 3).

For PF community, community stability decreased significantly after N50 and N100 addition alone or P40 and P80 addition alone compared with N0P0. Community stability showed no significant response to N25 combined with P addition compared with N0P0, while it showed significant declines under N50 and N100 combined with P addition ($p < 0.05$; Fig. 2). Perennial forb performed significantly higher stability than the other three functional groups across all N and P treatments (Fig. S3). Perennial legume stability showed significant declines under N25 combined with P addition compared to N0P0. Perennial forb stability increased significantly after N25P20 addition, but decreased significantly under N100P20 and N100P80 addition compared to N0P0 ($p < 0.05$). Annual and biennials stability showed significant decreases after N50 and N100 combined with P addition compared with N0P0 ($p < 0.05$; Fig. 3).

Compensatory dynamics of functional groups

For AB community, relative biomass of annuals and biennials performed apparent compensatory dynamics with perennial forb and grass under N50 and N100 addition in 2017. Relative biomass of annual and biennials and perennial forb under P addition alone notably increased at the expense of perennial grass and legume in 2018 and 2019. The relative biomass of annual and biennial had a notable increase under N50 and N100 combined with P addition, and thus caused declines in perennial grass and legume in both 2018 and 2019 (Fig. 4).

For PG community, different functional groups had no obvious changes in 2017. Relative biomass contribution of perennial grass increased notably under N addition alone at the expense of perennial legume in both 2018 and 2019. The reduced relative biomass of perennial grass under P addition alone or combined with N25 and N50 addition was compensated by the increased perennial legume in 2018 and 2019. Under N100 combined with P addition, the decreased relative biomass of perennial grass was compensated by increased perennial forb in 2018, but compensated by increased annual and biennial in 2019 (Fig. 4).

For PF community, Only relative biomass of perennial forb and perennial grass showed compensatory dynamics in 2017. Reduced relative biomass of perennial forb and grass under P addition alone was compensated by increased perennial legume and annual and biennials in 2018 and 2019. Relative biomass of perennial forb increased under N25 and N50 combined with P addition which caused notable reductions of perennial grass and legume in 2018 and 2019. After N100 combined with P addition, relative biomass contribution of annual and biennials increased substantially at the expense of perennial forb in 2019 (Fig. 4).

Species and functional group asynchrony

Species asynchrony showed significant decreases under N50 and N100 addition alone compared to N0P0 in AB community ($p < 0.05$). Under N50P40 and N50P80 addition, species asynchrony increased significantly compared with N50P0 in AB community ($p < 0.05$; Fig. 5). Species and functional group asynchrony of PG community after N100 addition

alone showed significant declines compared to N0P0. Under N50 and N100 combined with P addition, functional group asynchrony of PG community decreased significantly compared to N0P0 ($p < 0.05$; Fig. 5). Species and functional group asynchrony of PF community showed significant decreases under N100 addition alone or P40 and P80 addition alone compared to N0P0 ($p < 0.05$). Species and functional group asynchrony increased significantly under N50P20 and N50P40 addition compared with N50P0, and increased significantly under N100P20 addition compared to N100P0 ($p < 0.05$; Fig. 5).

Factors influencing community stability

For AB community, relative biomass of annual and biennials was negatively related to perennial grass ($r = -0.62$, $p < 0.001$), perennial legume ($r = -0.36$, $p < 0.05$) and perennial forb ($r = -0.61$, $p < 0.001$). Functional group asynchrony showed positive relationship with species richness ($r = 0.42$, $p < 0.01$) and relative biomass of perennial grass ($r = 0.37$, $p < 0.01$), but negatively related with relative biomass of annual and biennials ($r = -0.35$, $p < 0.05$). Community stability had positive relationship with species asynchrony ($r = 0.71$, $p < 0.001$), functional group asynchrony ($r = 0.68$, $p < 0.001$) and species richness ($r = 0.35$, $p < 0.05$), while negatively related with community biomass ($r = -0.44$, $p < 0.01$; Fig. 6; Fig. S2).

For PG community, relative biomass contribution of perennial grass showed negative relationship with perennial legume ($r = -0.63$, $p < 0.001$) and perennial forb ($r = -0.51$, $p < 0.001$). Functional group asynchrony was negatively related with relative biomass of perennial grass ($r = -0.45$, $p < 0.001$), but positively related to that of perennial legume ($r = 0.46$, $p < 0.001$). Community stability showed positive relationship with species asynchrony ($r = 0.63$, $p < 0.001$) and functional group asynchrony ($r = 0.54$, $p < 0.001$), while showed negative relationship with community biomass ($r = -0.30$, $p < 0.05$; Fig. 6; Fig. S2).

For PF community, relative biomass contribution of perennial forb and annual and biennials were negatively related with perennial grass ($r = -0.42$, $p < 0.01$; $r = -0.46$, $p < 0.001$) and perennial legume ($r = -0.51$, $p < 0.001$; $r = -0.36$, $p < 0.05$). Species asynchrony showed negative relationship with relative biomass of perennial legume ($r = -0.32$, $p < 0.05$). Community stability showed positive relationship with species asynchrony ($r = 0.69$, $p < 0.001$) and functional group asynchrony ($r = 0.66$, $p < 0.001$) and relative biomass of perennial forb ($r = 0.33$, $p < 0.05$), but negatively related with relative biomass of annual and biennials ($r = -0.40$, $p < 0.01$; Fig. 6; Fig. S2).

Structural equation model revealed that total variations of AB community stability was mainly explained (65%) by species richness, species asynchrony, annual and biennials and perennial forb stability. Reduced species richness, annual and biennials and perennial forb stability induced by N and P addition directly decreased AB community stability. In addition, decreased annual and biennial stability indirectly reduced AB community stability through species asynchrony (Fig. 7). Species richness, functional groups stability and species asynchrony explained 57% of total variance in PG community stability. Decreased annual and biennial stability and perennial legume stability directly reduced PG community stability. Perennial forb stability showed indirectly positive effects while perennial grass stability showed indirectly negative effects on PG community stability through species asynchrony (Fig. 7). Total variations in PF community stability were mainly explained (67%) by species asynchrony, perennial grass and annual and biennial stability. Decreased annual and biennial stability induced by N addition directly caused decreases in PF community stability. In addition, decreased annual and biennial stability reduced PF community stability indirectly through species asynchrony (Fig. 7).

Discussion

Our results demonstrated that the underlying mechanisms contributing to semiarid grassland temporal stability after N and P addition largely depend on interannual rainfall variability. Aboveground biomass of the three grassland communities showed interannual variation in response to N and P addition from 2017 to 2019, which was ascribed to the strong year-to-year precipitation fluctuations (Wang et al. 2017). The low annual precipitations in 2017, especially low

growing season precipitation, limited the response size of community productivity to N and P addition. The response of community productivity to N and P addition increased notably with increased precipitation in 2018 and 2019. High responsiveness of productivity in relative wet years after N and P addition driven a reduction in temporal stability in all three communities. N and P addition resulted in large shifts in functional groups, and their temporal stability differed largely. The community-specific shifts of functional group had large but variable effects on community stability in different grassland types. We found that compensatory dynamics of functional group and its stability changes were more important than diversity in regulating semiarid grassland stability under short-term N and P addition (Craven et al. 2018; Liu et al. 2018).

Various diversity–stability relationships have been detected in natural grassland communities (positive relationships: Craven et al. 2018; Huang et al. 2020; negative relationships: Sasaki and Lauenroth 2011; Yang et al. 2011; no relationships: Liu et al. 2019a; Xu et al. 2015). The different effects of diversity on community stability were associated with the magnitude of diversity decline and species composition (Isbell et al. 2009; Melts et al. 2018). Species richness decreased significantly and resulted in negative effects on AB community stability after N and P addition. This was because increasing tall and dense annuals and biennials under N50 and N100 combined with P addition produced strong and uniformly light limitation and thus caused short species loss (Avolio et al. 2014; DeMalach et al. 2017). In contrast to AB community, N and P addition had minimal effects on species richness in PG and PF communities, suggesting that diversity changes were not the main driver affecting community stability in perennials grassland (Liu et al. 2019a; Wang et al. 2017). Meanwhile, N and P addition induced overyielding effect may overwhelmed the role of diversity changes in regulating perennial grassland stability (Liu et al. 2018; Xu et al. 2015). Whereas this result did not deny the positive effect of diversity in stabilizing natural communities (Hautier et al. 2014). Likewise, some studies in semiarid grasslands have demonstrated that species diversity is not the primary factor influencing community stability, they identified compensatory dynamics and stability among functional groups are frequently more important drivers for community stability under N and P addition (Bai et al. 2004; Wang et al. 2017; Yang et al. 2011).

N and P addition induced large shifts in dominance of functional groups (Avolio et al. 2014; Wang et al. 2017). The negative relationships between biomass contribution of different functional groups, a potential complementary effect, could support community stability (Grman et al. 2010). Different N and P addition modulated compensatory dynamics of functional group through intensified competition between N-fixing and non-N-fixing species, tall and short species and annual and perennial species (Avolio et al. 2014; Song et al. 2019; Suding et al. 2005). Dominance of perennial grasses increased after N addition alone primary due to its superior N acquisition ability in PG and PF communities in relative wet year (Huang et al. 2020; Suding et al. 2005). The compensatory dynamics under P addition alone or combined with N25 often occurs between perennial legumes and perennial grasses or forbs in relative wet year. P addition could promote biological nitrogen fixation process by facilitating root development and the nitrogenase activity of nodule to increase abundance of legumes, thus suppressing perennial grasses or forbs (Avolio et al. 2014). Compensatory dynamics between annuals and biennials and perennial species frequently take place at high N and P combined addition (Avolio et al. 2014; Suding et al. 2005). Under N100 combined with P addition, the decreased stability in all three communities was driven by plastic responses of increased annuals and biennials in wet year (Wang et al. 2017). This is because annuals and biennials could best take advantage of higher nutrient availability and growth rapidly, which was facilitated by their functional traits, including abundant seed production, acquisitive resource use strategy and tall stature (Suding et al. 2005; Wang et al. 2017). As a consequence, strong light limitation induced by tall annuals and biennials suppressed other functional groups, which characterized with uncompetitive functional traits (conservative resource use strategy and short stature) (Chen et al. 2020; DeMalach et al. 2017).

The effect of functional group shifts on community stability was tightly associated with the intrinsic stability of functional groups (Huang et al. 2020). Different functional groups performed different stabilizing effects on community stability (Fig. 3; Fig. S3) (Bai et al. 2004). Our results indicated that perennial forbs and grasses had higher stability than

perennial legumes and annuals and biennials. The intrinsic stability of functional group was related to functional trait, relative biomass and species numbers within functional groups. Firstly, functional traits largely determine intrinsic stability of functional groups. Perennial grasses had relative higher stability in PG and PF communities, while performed relative lower stability in AB community (Fig. 3; Fig. S3). This was because most species within perennial grasses in PG and PF communities were clonal grasses (i.e. rhizome grasses and bunchgrasses). Clonal species that spread vegetative growth can effectively integrate and share limiting soil moisture and light resource through ramets, and thus performed high stability (Bai et al. 2004; Dickson et al. 2014). However, perennial grasses in AB community were primarily non-clonal species and showed low stability, which was susceptible to light and water limitation. These results are comparable to that rhizome grasses and bunchgrasses performed the lowest coefficients of variation (highest stability) among all functional groups in natural Inner Mongolia grassland over 24-year study (Bai et al. 2004). Perennial legumes had consistently lower stability in all three communities. The low stability of perennial legumes was associated with its uncompetitive for light because of short stature. Meanwhile, N addition could lead to the potential breakdown of symbiotic nitrogen fixation to reduce legumes stability (Regus et al. 2017). The low stability of annuals and biennials was attributed to its acquisitive resource use strategy and shallow root system (Bai et al. 2010; Wang et al. 2017). Annuals and biennials growth rapidly in relative wet years, while they disappeared and retreated to soil seed bank in subsequent dry years (Bai et al. 2010; Wang et al. 2017). Secondly, the stability of dominant functional group which had high aboveground biomass contribution was considered as a significant contributor to community stability (Sasaki and Lauenroth 2011; Smith and Knapp 2003). Many studies have demonstrated the crucial role of dominant species stability in regulating community stability (Sasaki and Lauenroth 2011; Song et al. 2019). For example, annuals and biennials were rare and had low biomass contribution under N0 or N25 addition in PG and PF communities. The negative effect of annuals and biennials on community stability was not significant. However, annuals and biennials became dominant species and resulted in significantly negative effect on community stability under N50 and N100 combined with P addition. Thirdly, functional groups with more species were generally highly stabilized (Fig. S3), which indicated that species within functional groups were not completely redundant with respect to community stability (Huang et al. 2020; Marquard et al. 2009). Annuals and biennials in AB community which contained more species performed relative higher stability than that in PG and PF communities. Perennial forbs in all three communities performed consistent relatively higher stability, which was associated with its highest species number.

Compensatory effect and stability of different functional groups also affect community stability via influencing species and functional group asynchrony (De Mazancourt et al. 2013; Liu et al. 2018). Species asynchrony promotes community stability when species sustains functional complementary interactions and response divergent under N and P addition (Loreau and De Mazancourt, 2013; Tredennick et al. 2017). N or P addition alone caused notable decreases in species and functional groups asynchrony, in turn, reduced stability in PG and PF communities. This was because N or P addition alone only relieved perennial grasses or perennial legumes from nutrient limitation and allow them to respond more strongly and synchronously (Fig. 4) (Collins et al. 2012; Wang et al. 2017). This supported the fact that the similar response of species belonging to a specific functional group caused reduction in species and functional group asynchrony (Tredennick et al. 2017). However, N and P combined addition promoted species asynchrony compared to N or P addition alone in all three communities. This pattern may be possibly attributed to perennial grasses, forbs, legumes and annuals and biennials free of N and P limitation simultaneously, allowing all functional groups response more asynchrony (Yang et al. 2011). In addition, the growing season of perennial species runs from early May to late September on Loess Plateau, whereas annuals and biennials usually germinate in early July following rainy season and had shorter lifespan which could promote species asynchrony (Bai et al. 2004; Wang et al. 2017). However, the positive effects of increased species asynchrony might be outweighed by the negative effects of decreased annuals and biennials stability under N50 and N100 combined with P addition, leading to reductions in community stability (Bai et al. 2010; Wang et al. 2017).

The underlying mechanisms in regulating temporal stability and ecological implications for different grassland types under short-term N and P addition were totally different on Loess Plateau. Species diversity is not the primary driver in regulating perennial grassland stability under N and P addition, which was instead driven by functional groups shifts, i.e. dominant species shifts from perennial species to annuals and biennials (Wang et al. 2017, Liu et al. 2018). However, the decreased community stability in annuals and biennials grassland was due to additive negative effect of diversity decline and increased dominance of annuals and biennials. Our findings shed new light on that the stability decline of farming-withdrawn grassland ecosystem function could be avoided by controlling dominance of less-stable functional groups during grassland restoration using fertilization on Loess Plateau (Bai et al. 2010; Liu et al. 2018). For restoring the vast areas of farming-withdraw grasslands in Loess Plateau, it is highly necessary to employ targeted N and P combination for various grassland communities based on dominant species and functional group composition to simultaneously sustain high productivity, high diversity and high stability.

Conclusion

N and P addition resulted in large shifts in functional groups and apparent compensatory dynamics in all three communities. Temporal stability differed largely between functional groups under N and P addition, and perennial forbs or grass had higher stability than perennial legumes or annuals and biennials. In perennial grassland, community stability decreased significantly under N addition alone because of decreased species and functional group asynchrony. The significant reduction in community stability under P addition alone primarily was due to increased dominance of less-stable perennial legumes. Increased dominance and reduced stability of annuals and biennials under N50 and N100 combined with P addition had significantly negative impacts on community stability in perennial grassland. The reductions of perennial grassland stability were primarily driven by increases in dominance of less-stable functional groups, such as perennial legumes and annuals and biennials. However, decreased community stability in annuals and biennials grassland was additive effect of species loss and increased dominance of annual and biennials. Our findings point to the need of targeted N and P combination for various grasslands to conserve the function and service stability when applying fertilization to promote grassland restoration in semiarid Loess Plateau.

Declarations

Acknowledgments

The study was financially supported by National Key Research and Development Program of China (2016YFC0501703), National Natural Science Foundation of China (41371509; 41771553) and the Foundation of State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau (A314021402-2015).

Funding

National Key Research and Development Program of China (2016YFC0501703), National Natural Science Foundation of China (41371509; 41771553) and the Foundation of State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau (A314021402-2015)

Conflicts of interest/Competing interests

The authors declare no conflicts of interest/competing interests.

Availability of data and material

The data generated during the current study are available from the corresponding author on reasonable request.

Author contributions

Bingcheng Xu and Zhifei Chen designed the entire experiment. Zhifei Chen, Peifeng Xiong, Junjie Zhou, Shuaibin Lai and Chunxia Jian conducted the experiments, and data collection. Xu B. C. supervised the research. Zhifei Chen, Bingcheng Xu, Peifeng Xiong, Weizhou Xu, Junjie Zhou and Chunxia Jian analyzed the data, wrote and edited the manuscript.

Code availability

The code used in this study is available from the corresponding author on reasonable request.

Ethics approval (include appropriate approvals or waivers)

Not applicable.

Consent to participate (include appropriate statements)

Not applicable.

Consent for publication (include appropriate statements)

Not applicable.

References

- Avolio ML, Koerner SE, La Pierre KJ, Wilcox KR, Wilson GWT, Smith MD (2014). Changes in plant community composition, not diversity, during a decade of nitrogen and phosphorus additions drive above-ground productivity in a tallgrass prairie. *J Ecol* 102:1649–1660. <https://doi.org/10.1111/1365-2745.12312>
- Bai YF, Han XG, Wu JG, Chen ZZ, Li LH (2004). Ecosystem stability and compensatory effects in the Inner Mongolia grassland. *Nature* 431:181–184. <https://doi.org/10.1038/nature02850>
- Bai YF, Wu JG, Clark CM, Naeem S, Pan QM, Huang JH, Zhang LX, Han XG (2010) Tradeoffs and thresholds in the effects of nitrogen addition on biodiversity and ecosystem functioning: evidence from Inner Mongolia grasslands. *Global Change Biol* 16:358–372. <https://doi.org/10.1111/j.1365-2486.2009.02142.x>
- Chen ZF, Xiong PX, Zhou JJ, Yang Q, Wang Z, Xu BC (2020). Grassland productivity and diversity changes in responses to N and P addition depend primarily on tall clonal and annual species in semiarid Loess Plateau. *Ecol Eng* 145:105727. <https://doi.org/10.1016/j.ecoleng.2020.105727>
- Collins SL, Koerner SE, Plaut JA, Okie JG, Brese D, Calabrese LB, Evansen RJ, Nonaka E (2012). Stability of tallgrass prairie during a 19-year increase in growing season precipitation. *Funct Ecol* 26:1450–1459 <https://doi.org/10.1111/j.1365-2435.2012.01995.x>
- Craven D, Eisenhauer N, Pearse WD, et al (2018). Multiple facets of biodiversity drive the diversity–stability relationship. *Nat Ecol Evol* 2:1579–1587. <https://doi.org/10.1038/s41559-018-0647-7>
- De Mazancourt C, Isbell F, Larocque A, et al (2013). Predicting ecosystem stability from community composition and biodiversity. *Ecol Lett* 16:617–625. <https://doi.org/10.1111/ele.12088>
- DeMalach N, Zaady E, Kadmon R (2017). Light asymmetry explains the effect of nutrient enrichment on grassland diversity. *Ecol Lett* 20:60–69. <https://doi.org/10.1111/ele.12706>

- Deng L, Shangguan ZP, Sweeney S (2014). "Grain for Green" driven land use change and carbon sequestration on the Loess Plateau, China. *Sci Rep* 4:1–8. <https://doi.org/10.1038/srep07039>
- Dickson TL, Mittelbach GG, Reynolds HL, Gross KL (2014). Height and clonality traits determine plant community responses to fertilization. *Ecology* 95:2443–2452. <https://doi.org/10.1890/13-1875.1>
- Fay PA, Prober SM, Harpole WS, Knops JMH, Bakker JD, Borer ET (2015). Grassland productivity limited by multiple nutrients. *Nat Plants* 1:15080. <https://doi.org/10.1038/nplants.2015.80>
- Gang CC, Zhao W, Zhao T, Zhang Y, Gao XR, Wen ZM (2018). The impacts of land conversion and management measures on the grassland net primary productivity over the Loess Plateau, Northern China. *Sci Total Environ* 645:827–836. <https://doi.org/10.1016/j.scitotenv.2018.07.161>
- Grman E, Lau JA, Schoolmaster DR, Gross KL (2010). Mechanisms contributing to stability in ecosystem function depend on the environmental context. *Ecol Lett* 13:1400–1410. <https://doi.org/10.1111/j.1461-0248.2010.01533.x>
- Harpole WS, Ngai JT, Cleland EE, Seabloom EW, Borer ET, Bracken ME (2011). Nutrient co-limitation of primary producer communities. *Ecol Lett* 14:852–862. <https://doi.org/10.1111/j.1461-0248.2011.01651.x>
- Hautier Y, Seabloom EW, Borer ET, et al (2014). Eutrophication weakens stabilizing effects of diversity in natural grasslands. *Nature* 508:521–525. <https://doi.org/10.1038/nature13014>
- Huang MJ, Liu X, Zhou SR (2020). Asynchrony among species and functional groups and temporal stability under perturbations: Patterns and consequences. *J Ecol* 108:2038–2046. <https://doi.org/10.1111/1365-2745.13418>
- Isbell F, Polley HW, Wilsey BJ (2009). Biodiversity, productivity and the temporal stability of productivity: patterns and processes. *Ecol Lett* 12:443–451. <https://doi.org/10.1111/j.1461-0248.2009.01299.x>
- Lepš J (2014). Scale- and time-dependent effects of fertilization, mowing and dominant removal on a grassland community during a 15-year experiment. *J Appl Ecol* 51:978–987. <https://doi.org/10.1111/1365-2664.12255>
- Liu HY, Mi ZR, Lin L, et al (2018). Shifting plant species composition in response to climate change stabilizes grassland primary production. *PNAS* 115:4051–4056. <https://doi.org/10.1073/pnas.1700299114>
- Liu JS, Li XF, Ma QH, Zhang X, Chen Y, Isbell F, Wang DL (2019a). Nitrogen addition reduced ecosystem stability regardless of its impacts on plant diversity. *J Ecol* 107:2427–2435. <https://doi.org/10.1111/1365-2745.13187>
- Liu YY, Wang Q, Zhang ZY, Tong LJ, Wang ZQ, Li JL (2019b). Grassland dynamics in responses to climate variation and human activities in China from 2000 to 2013. *Sci Total Environ* 690:27–39. <https://doi.org/10.1016/j.scitotenv.2019.06.503>
- Loreau M, De Mazancourt C (2013). Biodiversity and ecosystem stability: a synthesis of underlying mechanisms. *Ecol Lett* 16:106–115. <https://doi.org/10.1111/ele.12073>
- Mackie KA, Zeiter M, Bloor JMG, Stampfli A (2019). Plant functional groups mediate drought resistance and recovery in a multisite grassland experiment. *J Ecol* 107:937–949. <https://doi.org/10.1111/1365-2745.13102>
- Marquard E, Weigelt A, Temperton VM, Roscher C, Schumacher J, Buchmann N, Fischer M, Weisser WW, Schmid B (2009). Plant species richness and functional composition drive overyielding in a six-year grassland experiment. *Ecology* 90:3290–3302. <https://doi.org/10.1890/09-0069.1>

- Melts I, Lanno K, Sammul M, Uchida K, Heinsoo K, Kull T, Laanisto L (2018). Fertilising semi-natural grasslands may cause long-term negative effects on both biodiversity and ecosystem stability. *J Appl Ecol* 55:1951-1955. <https://doi.org/10.1111/1365-2664.13129>
- Niu D, Yuan XB, Cease AJ, Wen HY, Zhang CP, Fu H, Elser JJ (2018). The impact of nitrogen enrichment on grassland ecosystem stability depends on nitrogen addition level. *Sci Total Environ* 618:1529–1538. <https://doi.org/10.1016/j.scitotenv.2017.09.318>
- Rawnsley RP, Smith AP, Christie KM, Harrison MT, Eckard RJ (2019). Current and future direction of nitrogen fertilizer use in Australian grazing systems. *Crop Pasture Sci* 70:1034-1043. <https://doi.org/10.1071/CP18566>
- Regus JU, Wendlandt CE, Bantay RM, Gano-Cohen KA, Gleason NJ, Hollowell AC, O'Neill MR, Shahin JL, Sachs JL (2017). Nitrogen deposition decreases the benefits of symbiosis in a native legume. *Plant Soil* 414:159–170. <https://doi.org/10.1007/s11104-016-3114-8>
- Sasaki T, Lauenroth WK (2011). Dominant species, rather than diversity, regulates temporal stability of plant communities. *Oecologia* 166:761–768. <https://doi.org/10.1007/s00442-011-1916-1>
- Smith MD, Knapp AK (2003). Dominant species maintain ecosystem function with non-random species loss. *Ecol Lett* 6:509–517. <https://doi.org/10.1046/j.1461-0248.2003.00454.x>
- Song MH, Zong N, Jiang J, Shi PL, Zhang XZ, Gao JQ, Zhou HK, Li YK, Loreau M (2019). Nutrient-induced shifts of dominant species reduced stability via increase in species synchrony and population variability. *Sci Total Environ* 692:441-449. <https://doi.org/10.1016/j.scitotenv.2019.07.266>
- Suding KN, Collins SL, Gough L, Clark CM, Cleland EE, Gross KL, Milchunas DG, Pennings S (2005). Functional- and abundance-based mechanisms explain diversity loss due to N fertilization. *P Natl Acad Sci USA* 102:4387–4392. <https://doi.org/10.1073/pnas.0408648102>
- Tredennick AT, De Mazancourt C, Loreau M, Adler PB (2017). Environmental responses, not species interactions, determine synchrony of dominant species in semiarid grasslands. *Ecology* 98:971–981. <https://doi.org/10.1002/ecy.1757>
- Wang JF, Knops JMH, Brassil CE, Mu CS (2017). Increased productivity in wet years drives a decline in ecosystem stability with nitrogen additions in arid grasslands. *Ecology* 98, 1779–1786. <https://doi.org/10.1002/ecy.1878>
- Xu ZW, Ren HY, Li MH, Van Ruijven J, Han XG, Wan SQ, Li H, Yu Q, Jiang Y, Jiang L (2015). Environmental changes drive the temporal stability of semi-arid natural grasslands through altering species asynchrony. *J Ecol* 103:1308–1316. <https://doi.org/10.1111/1365-2745.12441>
- Yang Z, Van Ruijven J, Du GZ (2011). The effects of long-term fertilization on the temporal stability of alpine meadow communities. *Plant Soil* 345:315–324. <https://doi.org/10.1007/s11104-011-0784-0>
- Zhang JT (2005). Succession analysis of plant communities in abandoned croplands in the eastern Loess Plateau of China. *J Arid Environ* 63:458–474. <https://doi.org/10.1016/j.jaridenv.2005.03.027>
- Zhang JT, Dong YR (2010). Factors affecting species diversity of plant communities and the restoration process in the loess area of China. *Ecol Eng* 36:345–350. <https://doi.org/10.1016/j.ecoleng.2009.04.001>
- Zhou M, Yang Q, Zhang HJ, Yao XD, Zeng WJ, Wang W (2020). Plant community temporal stability in response to nitrogen addition among different degraded grasslands. *Sci Total Environ* 729:138886. <https://doi.org/10.1016/j.scitotenv.2020.138886>

Tables

Table1 Geographical and vegetation information of the three grassland communities before N and P additions in 2017

Grassland type	Restoration years (a)	Latitude and longitude	Altitude (m)	Aspects	Slope	Soil total N (TN) and total P (TP) content	Dominant species	Minor species
Annual and biennial (AB) community	7	109°14'30.68"E 36°44'36.29"N	1272	E	12°	TN: 0.50 g kg ⁻¹ TP: 0.62 g kg ⁻¹	<i>Setaria viridis</i> (Linn.) Beauv. <i>Artemisia capillaris</i> Thunb.	<i>Bidens pilosa</i> Linn. <i>Geranium wilfordii</i> Maxim. <i>Artemisia scoparia</i> Waldst. & Kit.
Perennial grass (PG) community	20	109°14'53"E 36°44'38"N	1164	E46°S	9°	TN: 0.60 g kg ⁻¹ TP: 0.58 g kg ⁻¹	<i>Bothriochloa ischcemum</i> (Linn.) Keng.	<i>Stipa bungeana</i> Trin. <i>Lespedeza davurica</i> (Laxm.) Schindl. <i>Artemisia gmelinii</i> Web. ex Stechm.
Perennial forb (PF) community	20	109°14'56"E 36°44'41"N	1153	E46°S	13°	TN: 0.69 g kg ⁻¹ TP: 0.53 g kg ⁻¹	<i>Artemisia gmelinii</i> Web. ex Stechm., <i>Artemisia giraldii</i> Pamp.	<i>Artemisia capillaris</i> Thunb. <i>Artemisia scoparia</i> Waldst. & Kit. <i>Lespedeza davurica</i> (Laxm.) Schindl.

Figures

Figure 1

Daily and monthly precipitation during 2017-2019, and mean monthly precipitation over 66 years (1951-2016). Grey arrows are N and P addition dates.

Figure 2

Temporal stability of the three grassland communities under N and P additions during 2017-2019. AB community: annual and biennial community; PG community: perennial grass community; PF community: perennial forb community. Blue bars are l.s.d. values among all N and P addition treatments ($p < 0.05$). Same as following.

Figure 3

Temporal stability of four functional groups in the three grassland communities under N and P additions during 2017-2019. PG: perennial grass; PL: perennial legume; PF: perennial forb; AB: annuals and biennials.

Figure 4

Compensatory dynamics of functional group dominance indicated by relative aboveground biomass contribution in the three grassland communities under N and P addition during 2017-2019.

Figure 5

Species and functional groups asynchrony of the three grassland communities under N and P addition during 2017-2019.

Figure 6

The relationship heat map between mean relative aboveground biomass contribution of different functional groups, Simpson's dominance index (SD), species richness (SR), species asynchrony (SA), functional group asynchrony (FGA) and community stability (CS). *p ≤ 0.05, **p ≤ 0.01, ***p ≤ 0.001.

Figure 7

Structural equation modeling results of the effect of N and P addition, community stability relating functional group stability, species richness and species asynchrony in the three grassland communities. Numbers beside lines are standardized path coefficients. The solid and dotted lines indicate significant positive and negative paths ($^{\wedge}p \leq 0.1$, $^*p \leq 0.05$, $^{**}p \leq 0.01$, $^{***}p \leq 0.001$). Path width is scaled by the size of standardized path coefficient. The model fitting for AB community: $\chi^2 = 18.785$, $df = 15$, $p = 0.224$; RMSEA = 0.073, $p = 0.33$. The model fitting for PG community: $\chi^2 = 23.375$, $df = 15$, $p = 0.076$; RMSEA = 0.109, $p = 0.14$. The model fitting for PF community: $\chi^2 = 16.523$, $df = 15$, $p = 0.348$; RMSEA = 0.046, $p = 0.47$.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [SupplementaryMaterial.docx](#)