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NEGATIVE EFFECTS OF CLIMATE CHANGE ON UPLAND GRASSLAND PRODUCTIVITY AND CARBON FLUXES ARE NOT ATTENUATED BY NITROGEN STATUS

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Abstract

Effects of climate change on managed grassland carbon (C) fluxes and biomass production are not well understood. In this study, we investigated the individual and interactive effects of experimental warming (+3 °C above ambient summer daily range of 9-12 °C), supplemental precipitation (333 mm +15%) and drought (333 mm -23%) on plant biomass, microbial biomass C (MBC), net ecosystem exchange (NEE) and dissolved organic C (DOC) flux in soil cores from two upland grasslands of different soil nitrogen (N) status (0.54% and 0.37%) in the UK. After one month of acclimation to ambient summer temperature and precipitation, five replicate cores of each treatment were subjected to three months of experimental warming, drought and supplemental precipitation, based on the projected regional summer climate by the end of the 21st Century, in a fully factorial design. NEE and DOC flux were measured throughout the experimental duration, alongside other environmental variables including soil temperature and moisture. Plant biomass and MBC were determined at the end of the experiment. Results showed that warming plus drought resulted in a significant decline in belowground plant biomass (-29 to -37%), aboveground plant biomass (-35 to -77%) and NEE (-13 to -29%), regardless of the N status of the soil. Supplemental precipitation could not reverse the negative effects of warming on the net ecosystem C uptake and plant biomass production. This was attributed to physiological stress imposed by warming which suggests that future summer climate will reduce the C sink capacity of the grasslands. Due to the low moisture retention observed in this study, and to verify our findings, it is recommended that future experiments aimed at measuring soil C dynamics under climate change should be carried out under field conditions. Longer term experiments are recommended to account for seasonal and annual variability, and adaptive changes in biota.

Keywords: climate change; warming; drought; carbon flux; grassland management; biomass productivity.

1. Introduction

Ecosystem sequestration of atmospheric carbon (C) in terrestrial ecosystems is a function of the balance between C uptake by plants (gross primary productivity – GPP) and C loss via processes such as ecosystem respiration (ER) and leaching, and these processes are sensitive to climate, particularly precipitation and temperature (Albaladejo et al., 2013; Bellamy et al., 2005; Rees et al., 2005). The global climate is warming and precipitation patterns are also changing, with regional differences reported (IPCC, 2013; Jenkins et al., 2009). Rising temperature and changing precipitation is also expected throughout the 21st Century, with global mean surface temperature projected to increase by 0.3 – 0.7 °C by 2035 and 1.7 – 4.8 °C by 2100, relative to 1986 – 2005 baseline (IPCC, 2014).

Climate change is expected to exert significant effects on terrestrial ecosystem C pools and their fluxes including plant biomass (Hartmann and Niklaus, 2012), microbial biomass (Rui et al., 2011), net ecosystem exchange (NEE, the difference between GPP and ER; De Boeck et al., 2007), and the leaching of dissolved organic C (DOC) (Hagedorn and Joos, 2014), with possible feedbacks to climate change. A very useful method for investigating the response of these ecosystem processes to climate change is experimental manipulation of climate variables such as temperature and precipitation (Bloor et al., 2010). Climate manipulation experiments have been set up to explore the responses of plant productivity and ecosystem C fluxes to climate change in different ecosystems (e.g. Allison and Treseder, 2008; Baldwin et al., 2014; Beierkuhnlein et al., 2011). A synthesis of data from 85 of these experimental studies (Wu et al., 2011) in different ecosystems across the globe including forests, shrublands and grasslands shows that: 1) warming without manipulated precipitation enhanced both ecosystem photosynthesis and respiration with no effect on net C uptake, 2) increased precipitation without warming enhanced both ecosystem photosynthesis and respiration with an overall increase in net C uptake, and 3) decreased precipitation without warming suppressed both ecosystem photosynthesis and respiration with an overall decrease in net C uptake. Another recent synthesis of results of about l60 climate manipulation experiments in different ecosystems also revealed that warming increased both soil C input and loss with no significant effect on net C pool, whereas increased precipitation stimulated soil C input, and drought suppressed it (Ni et al., 2017). Thus, the balance of evidence indicates that altered precipitation patterns have greater control on net soil C store than warming.

Past climate manipulation experiments in different terrestrial ecosystems (see reviews by Liu et al., 2016; Ni et al., 2017; Wu et al., 2011) have usually explored the individual effects of warming, supplemental precipitation and drought on net C uptake. The interactive effects of changing temperature and precipitation on C cycling have rarely been examined and remain poorly understood (Lei et al., 2016), although global meta-analyses indicate that the interactive effects of warming and altered precipitation differ from their simple additive effects (Ni et al., 2017; Wu et al., 2011). This means that adding up the reported individual effects of manipulated climate variables will not give the true response of the ecosystem. The effects of experimental climate change on the terrestrial C cycle are also confounded by other site-specific characteristics and management practices such as vegetation type (Beierkuhnlein et al., 2011; Bloor and Bardgett, 2012; Miranda et al., 2009) and fertilizer application (Jonasson et al., 1999), especially nitrogen (N) fertilizers (Dukes et al., 2005). For example, Graham et al. (2014) found that the addition of 50 kg N/ha increased the positive effect of warming on soil C efflux by 12% in a New Zealand grassland. Thus, for an improved understanding of the response of managed ecosystems to climate change, there is need for multifactorial experiments where the interactive effects of management, warming, supplemental precipitation and drought will be investigated.

Grasslands store a significant amount (34%) of the global terrestrial C and provide important ecosystem services such as climate change mitigation and forage for livestock production (White et al., 2000). In European grasslands, which are already net C sinks (Chang et al., 2015), the majority of the climate manipulation experiments (see Tables A1 and A2) have focused on the effects of climate change on aboveground biomass (AGB) and soil respiration (SR), and less attention has been given to other important C cycling processes such as NEE and DOC leaching. In published studies (Table A1) the main effects were: 1) warming alone stimulated both an increase and a decrease in AGB; 2) increased precipitation alone resulted in both an increase and a decrease in AGB; 3) drought alone decreased AGB and SR; and 4) both positive and negative interactive effects were observed when warming was combined with either increased precipitation or drought. The lack of definitive pattern of response to climate change in these studies may be partly due to pre-existing differences in grassland productivity. This is possible because less productive grasslands tend to be more resistant to climate perturbations (Grime et al., 2000). Fertilizer is widely used to improve grassland productivity (Yue et al., 2016), hence it is likely to be a major confounding factor in interpreting climate change effects. Whereas some grasslands are fertilised to increase vegetation biomass for livestock production, fertilizer application is discouraged in other grasslands due to environmental concerns such as protecting and enhancing biodiversity, or protecting water courses from pollution (Reed et al., 2009). The effects of climate change on fertilized and unfertilized grasslands need to be investigated. This will help inform future management decisions for targeted outcomes in the face of climate change.

The need to investigate climate change effects on managed grasslands is a particular concern for UK uplands. These areas mainly occur at 250 – 300 m above sea level and have witnessed changes in climate that are much greater than in the lowlands (House et al., 2010). For example, between 1961 and 2000, minimum temperatures increased more than maximum temperatures in the uplands, whereas there was no difference in the changes between minimum and maximum temperatures in the lowlands (Burt and Holden, 2010). Morecroft et al. (2009) also found that temperature trends between 1993 and 2007 differed between upland and lowland sites in the UK, with an average temperature increase of 1.2 °C in the uplands and 0.7 °C in the lowlands. The UK upland grasslands are considered sensitive environments and have important conservation values because they contain species of plants that are scarce in Europe, and are breeding grounds for nationally rare birds and amphibians (English Nature, 2001). These upland grasslands are predominantly managed for livestock production (Stevens et al., 2008) under both extensive management regimes with no fertilizer application, and more intensive management regimes with fertilizer application to improve forage productivity for silage and grazing.

Climate manipulation studies in the UK upland grasslands are few (e.g. Briones et al., 2009; Grime et al., 2008), and how grasslands under different management regimes might respond to warming and altered precipitation has not been considered. Briones et al. (2009) investigated the response of an unimproved acid grassland in Scotland to a 2-year soil warming (+3.5 °C) and found an increase in both ER and root biomass, and a decrease in AGB. Similarly, Grime et al. (2008) studied the response of an unfertilized grassland in Buxton (northern England) to 13 years of winter warming (+3.0 °C), supplemental summer precipitation (+26%) and summer drought (-77%). Warming, increased precipitation and drought both separately and in combination, had little effect on the ecosystem, however, there was a reduction in AGB under all the treatments (Grime et al., 2008). It remains to be known how fertilised upland grasslands respond to climate change. Specifically, there is a dearth of information on the effects of warming and altered precipitation on the net C uptake by plants and soil microbes as well as DOC flux.

The main aim of this study was to assess the individual and interactive effects of experimental warming, supplemental precipitation and drought on plant biomass, microbial biomass C (MBC), NEE, and DOC flux in two upland grassland fields of different soil N status in northern England. Earlier work (Eze et al., 2018a) showed that these grasslands store significant amount of soil organic C (SOC, 59 – 101 Mg ha^{-1}) and that about 70% of these C stock is occluded within the soil mineral mass. Whereas Eze et al. (2018a) identified the relative size of current SOC fractions, in this study we are investigating how the main fluxes and labile ecosystem C pools respond to short term climate manipulation. Changes in the bulk soil C stock or the relatively stable mineral-occluded C can only be detected in the long term (e.g. 10 – 100 years) (Smith, 2004). In contrast, changes in the less stable C pools such as the DOC, MBC and biomass accumulation, and C fluxes such as NEE, which are important indicators of changing environmental conditions, can be detected in the short term. The study was therefore based on the following hypotheses: 1) experimental warming and drought will separately and in combination reduce plant biomass, NEE, MBC, and DOC flux, 2) supplemental precipitation alone and experimental warming plus supplemental precipitation will stimulate higher plant biomass, NEE, MBC, and DOC flux, 3) the effects of experimental warming and altered precipitation will be greater in the high N field.

2. Methodology

2.1 Study area

Soil mesocosms from upland grasslands in Nidderdale (54^{o}09’N, 01^{o}53’W; Figure 1), northern England, were used for this study. Detailed site characteristics and management information are as described by Eze et al. (2018a). Briefly, the site has cool and wet climate with mean annual temperature (MAT) of 7.4 °C and mean annual precipitation (MAP) of 1550 mm (1981 – 2010). The soil is a sandy loam stagnohumic gley (Humic Gleysol in the World Reference Base), formed from clay drift with siliceous stone content. We selected two fields for this experiment. One field (high N field, HNF) receives inorganic N addition and has a significantly higher soil N (0.54%) and organic C stock (101 Mg/ha) than the low N field (LNF, total soil N = 0.37%, soil organic C stock = 59 Mg/ha). The herbaceous species common to the two fields are Ranunculus repens L. and Trifolium repens L. Other grasses in the HNF are Holcus lanatus L., Agrostis gigantea Roth, and Anthoxanthum odoratum L., whereas the LNF has Lolium perenne L.

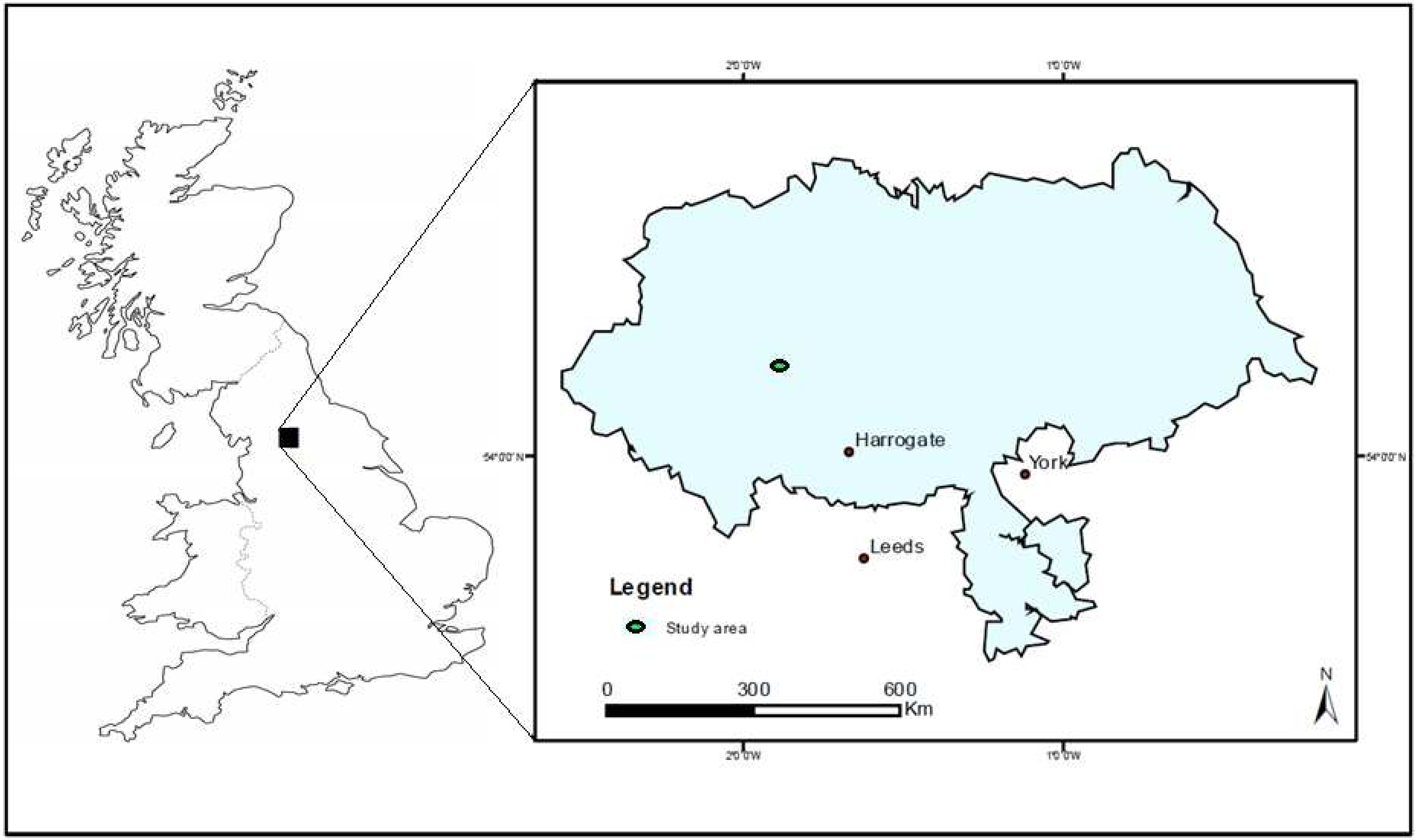

Figure 1: Map of North Yorkshire in the UK showing the area where soil cores were extracted.

2.2 Soil core sampling, experimental design and setup

We extracted 30 soil cores of 15 cm depth with intact vegetation (approx. 5 cm in height) from each of the two fields using un-plasticized polyvinyl chloride pipes (PVCu pipes, 11 cm diameter and 20 cm length). One end of each core was bevelled for ease of driving into the soil and the soil cores were extracted on the 31st of May 2017. After extraction, the cores were immediately taken to an environmental room in the School of Geography, University of Leeds.

The experiment consisted of six climate treatments in a fully factorial design of two temperature (ambient air temperature, T_0; ambient air temperature plus 3 °C, T_3) and three precipitation (ambient precipitation, P_0; ambient precipitation plus 15%, P_{+15}; ambient precipitation minus 23%, P_{-23}) conditions (Table 1). The six treatment combinations were applied to five replicated cores of each field type (LNF and HNF), resulting in a total of 60 experimental cores. The 30-year (1981 – 2010) mean summer (June – August) precipitation and temperature (recorded at Malham Tarn station located 18 km from the site) were used as the P_0 and T_0. The warming (T_3) and altered precipitation (P_{+15} and P_{-23}) treatments were based on the most recent UK climate projection (UKCP09) for the latter part of the 21st Century (2070 – 2100) (http://ukclimateprojections.metoffice.gov.uk/). The changes projected for our site under medium emission scenarios and 50% probability are: 3.0 °C increase in mean winter temperature, 3.3 °C increase in mean summer temperature, 15% increase in mean winter precipitation and 23% decrease in mean summer precipitation. We chose the medium emission and 50% probability scenario because it represents the change in climate that is likely not to be exceeded (Jenkins et al., 2009). Although summer condition was the focus of our study, we included supplemental precipitation (P_{+15}) in order to account for possible uncertainties in the prediction of seasonal precipitation, which is known to be highly variable (Jenkins et al., 2009).

Table 1: Experimental design based on observed 30-year mean summer (June – August) air temperature and precipitation and their 2070 – 2100 projection.

	Ambient summer temperature, T_0 (minimum = 9 °C, maximum = 16 °C)	Ambient summer temperature plus 3 °C, T_3, (minimum = 12 °C, maximum = 19 °C)
Low nitrogen field (LNF)	Ambient summer precipitation (P_0, 333 mm)	Ambient summer precipitation (P_0, 333 mm)
	Drought (P_{-23}, 256 mm)	Drought (P_{-23}, 256 mm)
	Supplemental precipitation (P_{+15}, 383 mm)	Supplemental precipitation (P_{+15}, 383 mm)
High nitrogen field (HNF)	Ambient precipitation (P_0, 333 mm)	Ambient precipitation (P_0, 333 mm)
	Drought (P_{-23}, 256 mm)	Drought (P_{-23}, 256 mm)
	Supplemental precipitation (P_{+15}, 383 mm)	Supplemental precipitation (P_{+15}, 383 mm)

The environmental room was set to T_0 with diurnal changes from a minimum of 9 °C at 0200 hr to a maximum of 16 °C at 1200 hr. In order to apply the two temperature treatments (T_0 and T_3) within the environmental room, we constructed two environmental chambers, ECs, (200 cm height × 116 cm width) with wooden frames (Rough sawn timber, 25 mm thickness × 38 mm width) and Celotex insulation board (Celotex TB4025 General Application Insulation Board, 25 mm thickness × 1200 mm width). Each of the ECs was fitted with an LED growth light (Heliospectra RX30), an Ecoheater (1500 mm Slimeline Greenhouse & Shed Heater HHT315 – 190W), two axial fans (Sunon Maglev DC 12V HA series) and a humidity/temperature data logger (EXTECH RH10). The Heliospectra lights were programmed to simulate 16 hours of light and 8 hours of darkness, which corresponds to the average summer photoperiod of the site. Light intensity was set to simulate diurnal variation, increasing from the first light-hour (40 µmol m^{-2} s^{-1}) to a maximum intensity at the ninth light-hour (850 µmol m^{-2} s^{-1}) and then decreasing until it becomes dark. The photosynthetically active radiation (PAR) recorded on-site from June 2016 to May 2017 was used to set the diurnal light intensity in the ECs. After setting up the lights, the environmental room temperature settings were adjusted to correct the increase in temperature resulting from heats generated by the lamps.

After the ECs were set up, wooden platforms (15 cm height) were placed on the floors of the chambers to support the soil cores. A total of 30 lysimeters, which served as the base for the soil cores, were arranged in five rows on the wooden platform. The lysimeters were made with 110 mm double socket PVCu couplers and plugs, fitted with high-density polyethylene funnels (Azlon FWC104, 300mL capacity). The top of the funnels were covered with 1 mm nylon mesh to keep soil particles in the cores intact and allow easy drainage of water. A transparent plastic container (125 ml) was placed directly underneath each lysimeter to collect leachates from the soil cores. To avoid any leachate loss, a silicone tubing (Food Grade Flexible Hose, 11 mm diameter) was connected to each funnel to direct leachates into the containers. Five replicate soil cores for each precipitation treatment and from each of the two Nidderdale fields (HNF and LNF) were placed on the lysimeters in a completely randomized design, resulting in 30 cores in each EC. One core from each replicated treatment combination was fitted with soil moisture and temperature sensors (5TM Decagon sensors) coupled to Arduino loggers (Kona 328 Arduino Uno compatible development board).

2.3 Treatments and C flux measurement

The soil cores were kept under control conditions (i.e. P_0 and T_0) for 30 days (1st to 30th June 2017) for acclimation. Under the control condition, we applied 7.4 mm of artificial rain to the cores every other day. The frequency of rainfall was based on the average monthly summer rain-days for the site which was 14. In the absence of rainfall chemistry data for the site, the artificial rain was prepared in the laboratory using a published rainfall chemistry data from a long-term monitoring site (Moor House – Upper Teesdale; 54°41' N, 2°23' W) about 60 km away (Table A3). After the acclimation period, the temperature of one EC was raised by 3 °C using the Ecoheater that had been fitted in the chamber. One-third of the soil cores in each chamber were still maintained under P_0 treatment, whereas the remaining two-thirds of the soil cores were spilt between P_{+15} and P_{-23} treatments. For the P_{+15} and P_{-23} treatments, 8.5 mm of synthetic rain was applied every other day. However, the drought treatment (P_{-23}) received rain only in the first and the third months of the treatments. The one month of no rain in the P_{-23} treatment was used to simulate an extreme drought event of 30 days which is similar to the 100-year recurrent drought event in both the UK (Bloor and Bardgett, 2012) and Germany (Mirzaei et al., 2008). The treatments lasted for three months (1st July to 30th September 2017) after the acclimation period. Throughout the treatment period, we recorded air temperature and relative humidity (EXTECH RH10 data loggers) at 1 hr intervals, and soil moisture and temperature (5TM Decagon sensors) at 15 minutes intervals. We also collected leachates from the cores weekly, which were analyzed for DOC using thermal oxidation analysis of aqueous samples (Analytik Jena Multi N/C 2100).

Weekly measurements of CO_2 and CH_4 fluxes were made throughout the experiment using an ultra-portable greenhouse gas (GHG) analyzer (Los Gatos Research UPGHGA, model 915-0011, California). The gas measurements were separated into NEE, the CO_2 exchange measured under light, and ER, the CO_2 exchange measured when light was excluded. During each gas measurement, the soil cores were fitted with cylindrical covers 34 cm in height with internal diameter similar to those of the soil cores. The cover used for NEE measurement was constructed of transparent PVC pipe (110 mm diameter; > 90% light transmission), whereas opaque terracotta drainage pipe (110 mm diameter; 0% light transmission) was used to construct the cover used for ER measurement. The top of each cover was fitted with an inlet and outlet gas tubing connected to the gas analyser, an axial fan for headspace air mixing and pressure equilibration gas bags. A pipe coupling (110 mm diameter) was attached to the base of each cover to ensure that it fitted tightly on the soil cores and prevent exchange of gases with the surrounding environment.

CO_2 and CH_4 fluxes were measured from 48 (4 per treatment) out of the 60 soil cores as the remaining 12 cores had soil moisture and temperature sensors in them and could not be fitted with the covers. To measure NEE, the core was fitted with the transparent cover after being connected to the gas analyser. After 60 seconds of placing the transparent cover on the core, the CO_2 concentration in the cover was measured continuously for a period of 120 seconds. To measure ER, the transparent cover was removed from the core immediately after taking the last NEE reading, the core was vented for 60 seconds and fitted with the opaque cover connected to the gas analyser. After 60 seconds of placing the opaque cover on the core, the CO_2 and CH_4 concentrations in the cover were measured continuously for a period of 120 seconds. During each gas measurement, a respirator (3M 7501 Silicone half mask respirator) connected to the outside of the environmental room was worn to remove any exhaled CO_2. This was done to prevent an increase in the CO_2 concentration in the environmental room. We also recorded temperature and pressure of the chamber environment (Comet Thermo-hygro-barometer), and PAR (Skye Quantum sensor) during each gas flux measurement.

The fluxes of CO_2 and CH_4 were calculated based on the rates of increase or decrease in their concentrations (Denmead, 2008):

$$F = \frac{V}{A}\frac{d\rho}{dt} \qquad 1$$

where F = flux density at the soil core surface (mg m^{-2} s^{-1}), V = headspace volume (m^3), A = internal area of soil core (m^2), ρ = mass concentration of the gas in the cylindrical cover headspace (mg m^{-3}) and t = time (s). The CO_2 and CH_4 flux values in mg m^{-2} s^{-1} were converted to mg m^{-2} day^{-1} by multiplying with 86400 (i.e. the number of seconds in a day).

The gas fluxes were estimated as the slope of the linear regression of CO_2 and CH_4 concentrations against time, after air temperature and pressure were corrected to standard values. Flux values were recorded if the slope of the linear regression was significant ($p < 0.05$) and the coefficient of determination (R^2) was equal to or greater than 0.75. The mean fluxes for each treatment combination were calculated by averaging the fluxes from four replicate soil cores. We adopted the atmospheric sign convention which defines a negative NEE as a net C uptake by the soil cores whereas positive NEE indicates C loss to the atmosphere (Imer et al., 2013).

2.4 Modelling CO_2 and CH_4 fluxes

After we calculated the fluxes of CO_2 and CH_4 using Equation 1, the CH_4 fluxes were not significantly different from zero, hence only CO_2 fluxes were modelled. To estimate CO_2 flux for the treatment period (i.e. 3 months), we filled gaps in the daily time series based on the relationships between CO_2 flux and meteorological variables. The GPP and ER for the treatment period were modelled for each of the 48 soil cores and NEE was calculated using Equation 2.

$$NEE = ER - GPP \qquad 2$$

2.4.1 Modelling GPP

A rectangular hyperbolic saturation curve (Equation 3) is widely used for modelling GPP in grasslands (e.g. Dyukarev, 2017; Elsgaard et al., 2012; Huth et al., 2017).

$$GPP = \frac{(\alpha \times PAR \times Gmax)}{(\alpha \times PAR + Gmax)} \qquad 3$$

where Gmax refers to the theoretical maximum rate of photosynthesis at infinite PAR (photosynthetic capacity), α is the initial slope of the hyperbolic equation (photosynthetic efficiency). To model GPP, we tested the performance of Equation 3 for predicting the GPP we measured (measured GPP was calculated with Equation 2 using measured NEE and ER). Equation 3 was fitted to PAR data recorded in the environmental chambers and the 'Solver' function in Microsoft Office Excel (2010 version) was used to estimate the best fit parameters for α and Gmax based on values that produced the smallest error term (sum of the squared difference between measured GPP and the GPP predicted by the curve). Using best fit parameters for the equation, GPP values were predicted for the days with measured GPP. The predicted GPP were plotted against their corresponding measured GPP and a regression line fitted through the plots (e.g. Figure A1). The regression line had a relatively high slope (0.77) and coefficient of determination ($R^2 = 0.97$), hence, Equation 3 was considered appropriate for modelling GPP. The parameters of Equation 3 derived for each of the 48 soil cores were then applied to a daily set of PAR data recorded in the environmental chambers. Daily GPP was generated for the period 1st July 2017 to 30th September 2017 and summed.

2.4.2 Modelling ER

To model ER, we tested the performance of two exponential models (Equations 4 and 5) that have been used to model ER in grassland studies (e.g. Du et al., 2014; Elsgaard et al., 2012; Huth et al., 2017).

Arrhenius model: $$ER = R_{10}\, e^{E_0\left(\frac{1}{T_{ref}-T_0} - \frac{1}{T-T_0}\right)} \qquad 4$$

Multiplicative model: $$ER = ae^{bT} \cdot SWC^{c} \qquad 5$$

where T is soil temperature; R_{10} is ER rate at a reference temperature (T_{ref} = 283.15 K); T_0 is temperature when ER is zero, usually constrained to 227.13 K to avoid over-parameterisation (Elsgaard et al., 2012; Huth et al., 2017); E_0 is temperature sensitivity coefficient; SWC is soil volumetric water content; a, b, and c, are fitting parameters. We parameterised the two models using the same procedure for parameterising the rectangular hyperbolic curve in Section 2.4.1. The equations were fitted to soil temperature and moisture data recorded for our soil cores and the ‘Solver’ function in Microsoft Office Excel (2010 version) was used to estimate the best fit parameters for R_{10}, E_0, a, b, and c based on values that produced the smallest error term. Using best fit parameters for the two equations, ER values were predicted for the days with measured ER. The predicted ER were plotted against their corresponding measured ER and a regression line fitted through the plots.

Compared to the Arrhenius model, the multiplicative model performed better in predicting measured ER as shown by its higher slope and coefficient of determination (Figures A2A and A2B). However, we found that the slope of the multiplicative model was always less than 0.5. We therefore tested the performance of a multiple linear regression based on soil temperature and soil moisture (Equation 6). The ER predicted by Equation 6 were plotted against their corresponding measured ER and a regression line fitted through the plot. The regression line had almost one-to-one slope (0.98), with a lower root mean square error (RMSE = 0.32) than that of the multiplicative model (slope = 0.38; RMSE = 1.43; Figure A2). We therefore used Equation 6 for modelling ER. The coefficients of Equation 6 derived for each of our soil cores were applied to the daily set of soil temperature and moisture data that we recorded. Generated daily ER from 1st July 2017 to 30th September 2017 were summed to get the ER for the treatment period.

$$ER = aT + bM + c \qquad 6$$

where T is the soil temperature; M is the soil moisture content; a and b are slopes; c is the intercept.

2.5 Plant biomass measurement and soil analysis

At the end of the treatment period, the experiment was dismantled and the cores were taken to the laboratory for biomass and soil analysis. Aboveground biomass (AGB) of each core was cut with scissors to the soil surface and was sorted into dead biomass (brown and yellow, DAGB) and live biomass (yellowish green and green, LAGB). The intact bulk soil in each core was carefully extruded into an empty semi-cylindrical core of similar diameter as the cores used for the experiment. The extruded soil was split vertically into two equal halves using a knife with serrated edge. Half of the soil removed after the split was used for the determination of soil properties such as MBC. The other intact half in the semi-cylindrical core was used for root or belowground biomass (BGB) determination. The soil sample for BGB determination was washed on sieves of different mesh sizes (50 µm to 2 mm) to separate roots from the soil, and the roots were collected on the sieves. The roots and AGB were dried in an oven at 65 °C for 48 hours and then weighed.

Soil MBC was determined using the chloroform fumigation-extraction method (Vance et al., 1987) after visible roots were removed. Fresh soil samples (25 g dry weight equivalent) from each of the 60 soil cores were fumigated for 24 hours with ethanol-free chloroform ($CHCl_3$). After fumigation, residual $CHCl_3$ vapour was removed by repeated evacuations using water aspirator pump and two-stage rotary oil pump. Additional un-fumigated soil samples of similar weight as the fumigated samples were used as controls. Fumigated and un-fumigated samples were extracted with 75 mL of 0.5M K_2SO_4 (1:3 soil/extractant ratio). The samples were shaken for 1 hour and filtered through Whatman GF 934-AH filter paper. The soluble C in the fumigated and un-fumigated samples were then determined using an aqueous analyzer (Analytik Jena Multi N/C 2100). MBC was calculated as the difference between extractable C in the fumigated and un-fumigated samples divided by a conversion factor, K_{EC}, of 0.35.

2.6 Data analysis

For each soil core, we calculated the total DOC flux and NEE for the three months of the experiment. Total NEE was calculated from modelled GPP and ER as explained in Section 2.4. Total DOC flux was calculated by summing the weekly fluxes obtained by multiplying the concentration of DOC in analyzed aliquots by the volume of leachates collected. The normality and homogeneity of variance of the total DOC and NEE as well as MBC and plant biomass were established using Shapiro-Wilk normality test and Levene's test. For each of the soil core type (LNF and HNF), a split-plot analysis of variance (ANOVA; with temperature as main plot factor and precipitation as sub-plot factor) was used to compare the means of DOC, NEE, MBC and plant biomass between treatments. The differences in mean values were separated using Tukey HSD post hoc test. Multivariate ANOVA (MANOVA) could not be used to assess the overall effect of each treatment on the C pools (MBC and plant biomass) and fluxes (DOC and NEE) due to multicollinearity. Moderate correlations between dependent variables are ideal for MANOVA, however, the correlations between our dependent variables were either too low ($r < 0.3$) or too high ($r > 0.8$). Other statistical analyses related to modelling were as described in Section 2.4, and all statistical analyses and modelling were carried out in Microsoft Excel (2010 version) and SPSS Statistics (version 22).

3. Results

3.1 Effect of warming and altered precipitation on C pools

Plant biomass and MBC were significantly affected by experimental warming and altered precipitation (Tables 2 and 3). The response of plant biomass to the climate manipulation was similar across HNF and LNF cores, whereas the response of MBC to climate manipulation differed between the two core types (Figure 2). There was a warming-induced decline in BGB and TAGB by -26% and -14% in the LNF, and -29% and -22% in the HNF respectively. Across both LNF and HNF, warming plus drought led to significant reductions (-73% and -77%) in LAGB and a significant increase (+192% and +215%) in DAGB. Drought resulted in a significant increase (+75) in the DAGB and a significant decline (-63%) in the MBC of the LNF but had no significant effects on any of the C pools in the HNF. Warming alone and warming plus supplemental precipitation significantly enhanced MBC by +125% and +171% respectively in the HNF, with no significant effects in the LNF.

3.2 Effects of warming and altered precipitation on C fluxes

The NEE was the only component of the C flux that was significantly affected by climate manipulation as the DOC flux was not significantly affected in either HNF or LNF cores (Table 3). All the temperature and precipitation treatments, except supplemental precipitation in the LNF, resulted in the decline of NEE. In the LNF, supplemental precipitation led to a significant increase (+103%) in NEE, whereas in the HNF, drought significantly reduced NEE by -66% (Figure 3). In the LNF and HNF, warming plus drought reduced NEE by -29% and -13% respectively. Similarly, warming plus supplemental precipitation reduced NEE by -14% and -24% respectively in the LNF and HNF, although this was not statistically significant.

Table 2: Mean ± standard error of plant biomass, microbial biomass carbon, net ecosystem exchange (NEE) and dissolved organic carbon after experimental warming and altered precipitation in cores of low (LNF) and high (HNF) nitrogen status (n = 5 for all parameters except NEE with n = 4).

Soil core	Temperature	Precipitation	Belowground biomass (g/m^2)	Dead aboveground biomass (g/m^2)	Live aboveground biomass (g/m^2)	Total aboveground biomass (g/m^2)	Microbial biomass carbon (mg/kg)	NEE* (g CO_2/m^2)	Dissolved organic carbon* (mg C)
LNF	Ambient	Ambient	1403±53	394±93	2330±200	2724±227	163±32	-1256±436	5±1
		Drought	1804±157	690±106	2166±154	2856±145	60±19	-1238±90	5±1
		Supplemental	1585±238	392±120	2809±156	3201±224	76±26	-2546±574	9±3
	Warming	Ambient	1038±152	326±63	2020±171	2346±130	151±28	-967±121	4±1
		Drought	1001±126	1151±89	623±231	1775±183	85±55	-891±94	4±0
		Supplemental	1056±185	404±78	1969±208	2373±165	239±31	-1078±151	4±0
HNF	Ambient	Ambient	1752±314	487±93	2387±73	2874±139	83±32	-1312±320	7±1
		Drought	2012±413	656±91	2145±111	2801±126	49±23	-451±117	11±3
		Supplemental	1988±224	439±53	2662±262	3100±258	91±42	-816±104	8±1
	Warming	Ambient	1241±144	484±67	1751±310	2234±252	187±49	-1120±103	5±1
		Drought	1110±235	1533±74	548±118	2081±187	72±23	-1147±111	5±1
		Supplemental	1292±449	491±113	1602±207	2093±137	225±75	-997±135	8±1

* = sum of the daily values for the experimental period (1st July to 30th September 2017).

Table 3: Split-plot ANOVA result showing the individual and interactive effects of temperature and precipitation on plant biomass, microbial biomass C (MBC), net ecosystem CO_2 exchange (NEE) and dissolved organic C (DOC) flux in cores of low (LNF) and high (HNF) nitrogen status ($n = 5$ for all parameters except NEE with $n = 4$).

Soil core	Source of variation	BGB		DAGB		LAGB		TAGB		MBC		NEE*		DOC*	
		F	Sig.	F	Sig.	F	Sig.	F	Sig.	F	Sig.	F	Sig.	F	Sig.
Low N	Temp.	14.18	**0.01**	1.93	0.20	40.38	**<0.01**	33.50	**<0.01**	3.42	0.10	4.26	0.09	2.76	0.15
	PPT.	0.74	0.49	32.55	**<0.01**	14.24	**<0.01**	3.00	0.08	5.16	**0.02**	7.48	**0.01**	2.00	0.18
	Temp. × PPT	1.09	0.36	6.76	**0.01**	4.97	**0.02**	1.71	0.21	4.51	**0.03**	5.55	**0.02**	1.76	0.21
High N	Temp.	8.60	**0.02**	13.24	**0.01**	42.04	**<0.01**	20.40	**<0.01**	7.88	**0.02**	2.90	0.14	5.04	0.07
	PPT.	0.10	0.91	48.80	**<0.01**	9.92	**<0.01**	0.41	0.67	2.27	0.14	3.29	0.07	0.91	0.43
	Temp. × PPT	0.18	0.84	23.15	**<0.01**	3.02	0.08	0.59	0.57	0.72	0.50	3.48	0.06	1.54	0.25

Temp. = temperature, PPT. = precipitation, BGB = belowground biomass, DAGB = dead aboveground biomass, LAGB = live aboveground biomass, TAGB = total aboveground biomass, * = sum of three months' daily fluxes, Sig. = significant value at 5% probability level. Bold font indicates significant effect at $p < 0.05$.

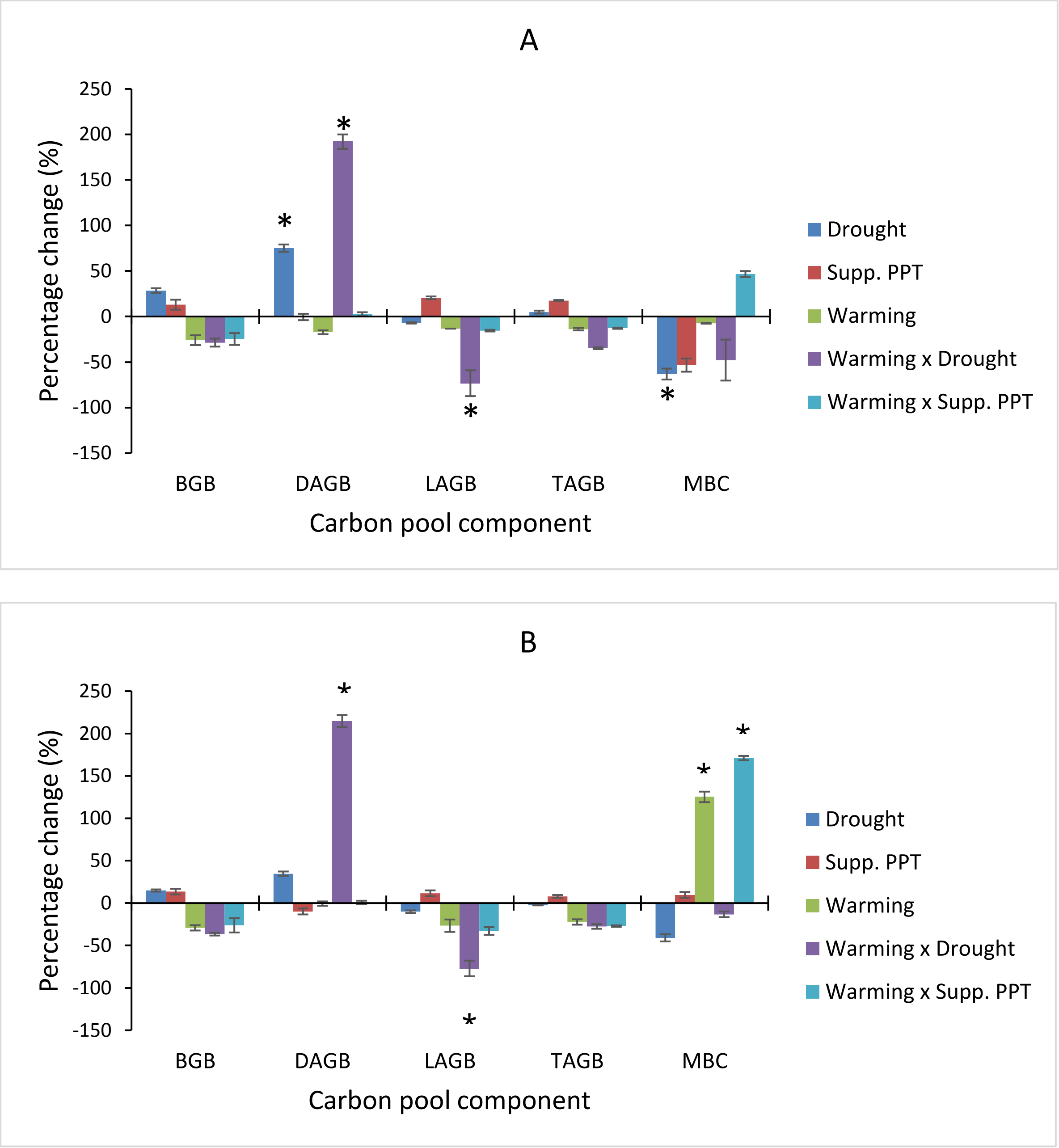

Figure 2: The effects of warming, drought, supplemental precipitation (Supp. PPT) and their interactions on plant biomass (belowground – BGB, dead aboveground – DAGB, live aboveground – LAGB, total aboveground – TAGB) and microbial biomass C (MBC) in cores of (A) low nitrogen and (B) high nitrogen status (n = 5). Bars with asterisks represent significant change at 5% probability level in the C pool component of soil cores subjected to altered climate treatment compared to cores with ambient climate treatment. Error bars represent standard error (n = 5).

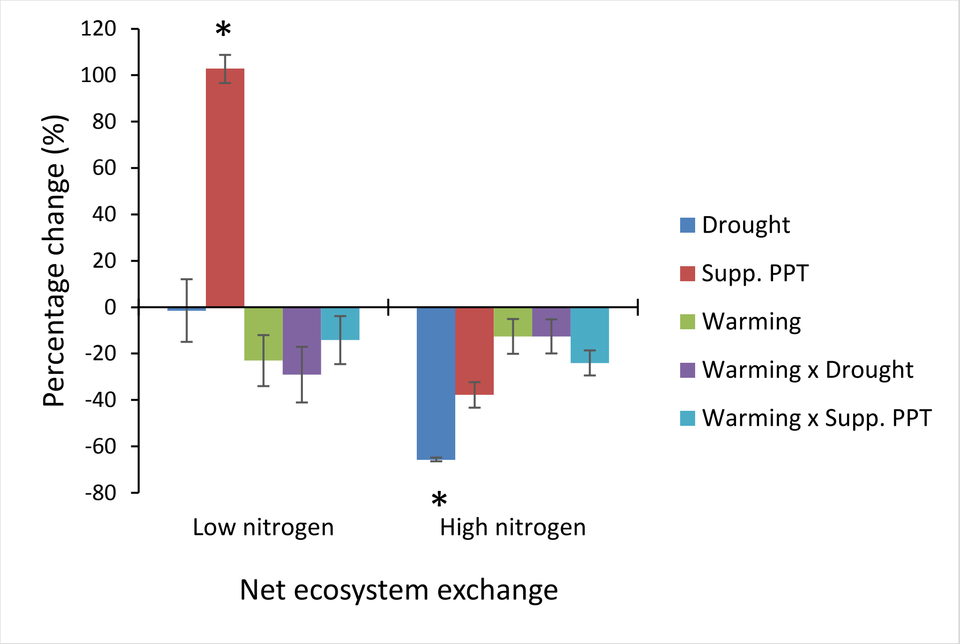

Figure 3: The effects of warming, drought, supplemental precipitation (Supp. PPT) and their interactions on the net ecosystem CO_2 exchange (NEE) in the cores of low nitrogen and high nitrogen status (n = 4). Bars with asterisks represent significant change at 5% probability level in NEE of soil cores subjected to altered climate treatment compared to cores with ambient climate treatment. Error bars represent standard error (n = 4).

4. Discussion

A combination of warming and drought resulted in a significant reduction in plant biomass production regardless of the N status of the soil cores. This is consistent with our first hypothesis where we expected a decline in C uptake and storage due to warming and drought. The reduction in aboveground plant biomass (-28 to -35% or -793 to -949 g m^{-2}) and NEE (-13 to -29% or -165 to -365 g CO_2 m^{-2}) resulting from warming plus drought in our study is consistent with the findings of previous experimental climate change studies in the UK (Table A1), and a Europe-wide C flux study carried out after a heat wave and drought in 2003 (Ciais et al., 2005). Our results are also consistent with the results of modelling studies (e.g. Thornley and Cannell, 1997) which indicate a decrease in net ecosystem C uptake and plant biomass production under warming scenarios in temperate grasslands.

In temperate ecosystems where plant growth is limited by low temperature (Wingler and Hennessy, 2016), particularly in upland environments, one would expect an increase in biomass production under elevated temperature. However, the results from our study indicate that the response of the temperate grassland ecosystem to rising temperature will depend on the level of stress imposed by a combination of warming and altered precipitation. Detailed interactive effects of warming and drought as well as warming and supplemental precipitation, and the implications of these for future grassland management are discussed in the following sections.

4.1 Effects of warming and drought on net ecosystem C uptake and plant biomass production

Consistent with our findings, previous studies have reported that drought conditions under experimental warming suppressed plant growth and reduced MBC (Ganjurjay et al., 2016; Liu et al., 2009). In extreme cases, the survival of some species of plant can be threatened by a combination of warming and drought (Xu et al., 2014). These negative interactive effects of warming and drought are attributable to physiological stress resulting from severe water deficits. By the end of the drought period, the soil cores in our study under warming and drought had less than half (5 – 12%) of the moisture content of soils under field conditions at our site during summer months (54 – 64%; Eze et al., 2018b), indicating that severe physiological stress was the likely cause of increased plant death (Figure 2). Severe moisture deficit has been shown to cause low stomatal conductance (Sanaullah et al., 2011), a decrease in the mobility of nutrients and reduced microbial access to substrates (Fuchsluege et al., 2014). These impose stress on plants and soil microbes resulting in a significant reduction in C uptake by plants (Hasibeder et al., 2015), plant biomass (Hartmann and Niklaus, 2012), microbial activities (Liu et al., 2009), MBC (Shi et al., 2012), and an increase in the death of plants and microbes (Sanaullah et al., 2011).

Our findings suggest that under future summer climate change, particularly elevated temperature and drought, the C sink capacity of the grasslands we studied will be reduced, with likely similar effect on other temperate upland grasslands. This may lead to the ecosystem switching from being a net C sink to a net C source. It is however important to be cautious in extrapolating our results because other factors may dampen the negative effects reported. For example, the suppression of photosynthetic activity and plant growth, and increased senescence and mortality, resulting from the combination of warming and drought has been found to be partly alleviated by elevated atmospheric CO_2 in greenhouse studies (Van De Velde et al., 2015; Xu et al., 2014). In some grassland field manipulative experiments where the climate change was simulated, elevated atmospheric CO_2 alongside experimental warming led to an increase in the net C uptake by plants (Mueller et al., 2016; Ryan et al., 2017). In addition, we considered only summer conditions thus reflecting the current short growing season for mid-to-high latitude grasslands, but the effects of climate extremes such as drought can be carried over to other seasons (Niu et al., 2014). Under climate change, not only are lagged changes possible but the growing season may also be extended (Cleland et al., 2007; Xia et al., 2014) due to increased temperatures at either end of the summer. Hence multifactorial manipulative field experiments are needed that extend beyond the current growing season length, in order to determine the extent of ecosystem recovery from and resilience to multi-factor climatic stress (Van De Velde et al., 2015).

4.2 Effects of warming and supplemental precipitation on net ecosystem C uptake and plant biomass productivity

The lack of significant effects of warming plus supplemental precipitation on net ecosystem C uptake and plant biomass production is surprising. Although warming is known to increase the loss of water from grassland ecosystems via increased evapotranspiration, supplemental precipitation recharges the ecosystem's water storage, often leading to a decrease in soil temperature (e.g. -0.5°C in this study) due to the high heat capacity of water (Lal and Shukla, 2004; Liu et al., 2009). As a result, we had expected supplemental precipitation in this study to remove any water limitation imposed by elevated temperature, and at the same time minimize any direct warming-induced stress on plants and microbes possibly leading to increased C uptake. This was however not the case as only MBC increased under warming and supplemental precipitation whereas net C uptake and plant biomass productivity showed a non-significant decline. This indicates that supplemental precipitation may not sufficiently offset the negative effects of elevated temperature under severe warming-induced plant stress, at least in the short term.

Seasonal precipitation in the UK is highly variable (Jenkins et al., 2009) and this makes the prediction of seasonal distribution of precipitation difficult. Although summer precipitation in the grasslands we studied is expected to decline in the future, including supplemental precipitation in this study helped to show that even if precipitation should increase in the future, it will not significantly reverse the negative effects of summer warming on net C uptake. A limitation of our work, however, is the short duration imposed by limited resources. The results reported here might have been different if the experiment was conducted for a longer period of time rather than restricted to growing season duration.

Another aspect of our study that needs to be considered in extrapolating the results is the possibility that drainage from the cores was enhanced because of their open end. This most likely added to the stress caused by warming-induced evapotranspiration on both plants and soil microbes. Water storage capacity in the soil cores (15 cm in depth) was likely to be less than found in field conditions where soil depth averages 20 cm from the surface down to the subsoil. Even when the top soil layer (e.g. the rooting zone) is dry, water stored deeper in the profile is made available to plants via capillary action (Vervoort and Van Der Zee, 2008).

Also, plant roots grow more extensively during moisture stress which makes it possible for greater soil volume to be explored for water (Briones et al., 2009). These sources of water to plants were absent from our mesocosm experiment. It is also possible that CO_2 might have been lost from the open bottom of the soil cores.

4.3 Implication of experimental warming and altered precipitation for future land management

Mesocosms from the two sites investigated here were both responsive to warming and altered precipitation, suggesting that the level of N addition in the HNF did not increase its sensitivity to climate change. The effects of altered precipitation on some components of the C cycle in the LNF and HNF differed slightly. For example, drought led to a significant increase in the death of AGB and a significant decline in MBC in the LNF, whereas in the HNF, drought significantly reduced NEE. The significant drought-induced death of AGB in the LNF may be connected to the presence of Lolium perenne L., which has been shown to be sensitive to drought (Aper et al., 2014). The death of plants in the LNF might subsequently have affected the rate of rhizodeposition thereby leading to the reduction in MBC (Bloor et al., 2018). On the other hand, the drought-induced reduction in the NEE of the HNF could be attributed to the reduction in GPP and an increase in ER (Figure A3), which limited the net C uptake by the plants. Despite the slight differences in the responses of LNF and HNF to altered precipitation, LNF and HNF had similar responses to both drought and supplemental precipitation when combined with warming. These findings were contrary to our third hypothesis where we expected greater effects of climate manipulation on the HNF that receives inorganic N addition. In an earlier UK study, Grime et al. (2000) reported that fertile grasslands with fast growing species were more sensitive to experimental climate change than less fertile and more matured grasslands. The site studied by Grime et al. (2000) that was considered fertile and more productive was a successional grassland converted from arable land, whereas our fields have been managed for grazing animals for over 100 years. Hence, the differences in sensitivity to climate change reported by Grime et al. (2000) represented land use change and possibly a more extreme comparison than the relatively small differences in N application reported here, representing typical upland grazing management.

Our results indicate that soil water management during the growing season may present a greater challenge to C uptake and biomass production than nutrient addition by the end of the 21st Century. However, as explained earlier, the level of soil moisture loss recorded in our mesocosm experiment is likely to be higher than observed in field conditions. Our study was also short term making it difficult to account for adaptive changes in plant and microbial species' physiology and community composition (Grime et al., 2000), which are detectable in the long term. Multi-year climate manipulative studies under field conditions are called for to inform and devise appropriate strategies for future grassland management that will be climate-smart.

5. Conclusion

A combination of warmer summer (+3 ºC) and drought (-23% precipitation) conditions led to a significant decline in the net ecosystem C uptake (-13 to -29%) and plant biomass production (-29 to -37% belowground biomass and -35 to -77% aboveground biomass) after three months of experimental manipulation. Supplemental summer precipitation (+15% precipitation) could not reverse the negative effects of warming on the net ecosystem C uptake and plant biomass production. Mesocosms representing grassland sites with differing N status (0.54% and 0.37%) were equally responsive to the experimental climate treatments, suggesting that differences in N management may not have significant influence on the response of the grasslands to climate change. The negative effects of the experimental climate change on the net ecosystem C uptake was attributed to physiological stress resulting from severe soil moisture deficits. However, in addition to water losses resulting warming-induced evapotranspiration, the reduction in soil moisture might be partially attributable to open-ended mesocosms used in this experiment. The cores have less volume and capacity to retain water than is possible in the field. This led to the recommendation that future experimental climate change studies in the upland grasslands should be carried out in the field. Also, due to the short term nature of our experiment, it is further recommended that long term studies should be conducted to account for adaptive changes in plant and soil microbial species that may result from changing temperature and precipitation regimes.

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Table A1: Reported responses of plant biomass and C fluxes to experimental warming and altered precipitation in some European grasslands and their site characteristics.

Location	Altitude (m)	MAT (°C)	MAP (mm)	Soil	Exp. set-up	Trt type	Trt	Duration (years)	Effect on GPP	Effect on ER	Effect on AGB	Effect on BGB	Effect on DOC	Authors
Belgium		9.6	776	Silt loam	Soil cores in sun-lit chambers	W	+3.0 °C	2	-12%	-7% AGR	-30%			De Boeck et al., 2007
Denmark		8.0	600	Sandy loam	Field plots	W	+1.0 °C	1			+10%			Andresen et al., 2009
Denmark		8.0	600	Sandy loam	Field plots	W	+1.0 °C	3			+5%			Kongstad et al., 2012
France	850	8.7	780	Cambisol	Field monoliths	W	+3.5 °C	3			+7%			Bloor et al., 2010
UK (England)	370	8.0	1300	Limestone derived	Field plots	W	+3.0 °C	13			-9%			Grime et al., 2008
UK (England)	150	10.0	680	Limestone derived	Field plots	W	+3.0 °C	5			+2%			Grime et al., 2000
UK (Scotland)	309	8.0	900	Brown earth	Soil cores in exp. garden	W	+3.5 °C	2		+49%	-15%	+37%		Briones et al., 2009
UK (England)	150	10.0	680	Limestone derived	Field plots	P	+20% of summer ppt	5			+33%			Grime et al., 2000
UK (England)	370	8.0	1300	Limestone derived	Field plots	P	+26% of summer ppt	13			-3%			Grime et al., 2008

Denmark		10.0	707	Loamy sand	Field plots	D	-7% of MAP	3	-11% SR				Selsted et al., 2012
Denmark		8.0	600	Sandy loam	Field plots	D	-8% of MAP	3		-5%			Kongstad et al., 2012
UK (England)	150	10.0	680	Limestone derived	Field plots	D	-100% of July – August ppt	5		-6%			Grime et al., 2000
UK (England)	370	8.0	1300	Limestone derived	Field plots	D	-77% of summer ppt	13		-34%			Grime et al., 2008
Switzerland	393	9.8	1232	Cambisol (loamy clay)	Field plots	D	-33% of MAP	1	-17% SR			-58%	Hagedorn and Joos, 2014
Switzerland	982	7.7	1765	Cambisol (loamy clay)	Field plots	D	-33% of MAP	1	-24% SR			-42%	Hagedorn and Joos, 2014
Switzerland	1978	2.3	969	Leptosol (sandy loam)	Field plots	D	-26% of MAP	1	-38% SR			-81%	Hagedorn and Joos, 2014
Switzerland	393	9.6	1103	Cambisol	Field plots	D	-31% of MAP	3		-22%			Prechsl et al., 2015
Switzerland	1978	1.7	948	Cambisol	Field plots	D	-26% of MAP	2		-42%			Prechsl et al., 2015
UK (England)	150	10.0	680	Limestone derived	Field plots	WP	+3.0 °C; +20% of summer ppt	5		+6%			Grime et al., 2000

UK (England)	370	8.0	1300	Limestone derived	Field plots	WP	+3.0 °C; +26% of summer ppt	13		-4%		Grime et al., 2008
Denmark		8.0	600	Sandy loam	Field plots	WD	+1.0 °C; -8% of MAP	3		+4%		Kongstad et al., 2012
Denmark		10.0	707	Loamy sand	Field plots	WD	+0.4 °C; -7% of MAP	3	-20% SR			Selsted et al., 2012
UK (England)	370	8.0	1300	Limestone derived	Field plots	WD	+3.0 °C; -77% of summer ppt	13		-25%		Grime et al., 2008
UK (England)	150	10.0	680	Limestone derived	Field plots	WD	+3.0 °C; -100% of July – August ppt	5		-32%		Grime et al., 2000
France	850	8.7	780	Cambisol	Field monoliths	WD	+3.5 °C; -20% of summer ppt	3		-4%		Bloor et al., 2010

MAP= mean annual precipitation, MAT = mean annual temperature, Exp. = experimental, AGR = aboveground respiration, SR = soil respiration, AGB = aboveground biomass, BGB = belowground biomass, Trt = treatment, ppt = precipitation, W = warming, P= increased precipitation, D = drought, WP = warming and increased precipitation, WD = warming and drought.

Table A2: References for the studies in Table A1.

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Table A3: Rainfall chemistry for Moor House – Upper Teesdale, UK (54° 41'N, 2° 23' W) from 4th June 2003 to 29th August 2012 (Rennie et al., 2015). In the absence of rainfall chemistry data for our site, the artificial rain used for our experiment was prepared in the laboratory using this published rainfall chemistry data from a long-term monitoring site about 60 km away.

Ion	10-year mean (mg/L)	10-year mean (meq/L)
Ca^{2+}	0.2890	0.0144
Mg^{2+}	0.0766	0.0063
Na^{+}	0.6222	0.0271
K^{+}	0.1378	0.0035
Cl^{-}	0.9722	0.02739
SO_4^{2-}	0.9946	0.0207
NH_4^{+}	0.4885	0.0271
NO_3^{-}	0.2860	0.0046
pH 5.36		

Rennie, S., Adamson, J., Anderson, R., Andrews, C., Bater, J., Bayfield, N., Beaton, K., Beaumont, D., Benham, S., Bowmaker, V., Britt, C., Brooker, R., Brooks, D., Brunt, J., Common, G., Cooper, R., Corbett, S., Critchley, N., Dennis, P., Dick, J., Dodd, B., Dodd, N., Donovan, N., Easter, J., Eaton, E., Flexen, M., Gardiner, A., Hamilton, D., Hargreaves, P., Hatton-Ellis, M., Howe, M., Kahl, J., Lane, M., Langan, S., Lloyd, D., McElarney, Y., McKenna, C., McMillan, S., Milne, F., Milne, L., Morecroft, M., Murphy, M., Nelson, A., Nicholson, H., Pallett, D., Parry, D., Pearce, I., Pozsgai, G., Rose, R., Schafer, S., Scott, T., Sherrin, L., Shortall, C., Smith, R., Smith, P., Tait, R., Taylor, C., Taylor, M., Thurlow, M., Turner, A., Tyson, K., Watson, H., Whittaker, M., 2015. UK Environmental Change Network (ECN) precipitation chemistry data: 1992-2012. NERC Environmental Information Data Centre. https://doi.org/10.5285/0cd4abd2-1bc9-48bc-b5c2-cebdeaa23ceb.

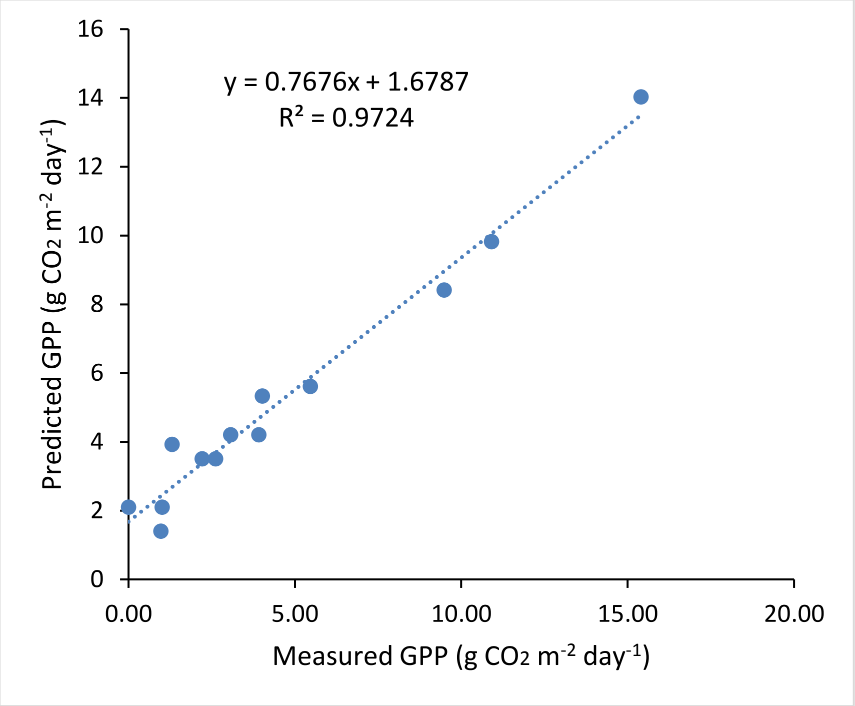

Figure A1: An example of linear relationships between measured and predicted GPP (using data from one soil core).

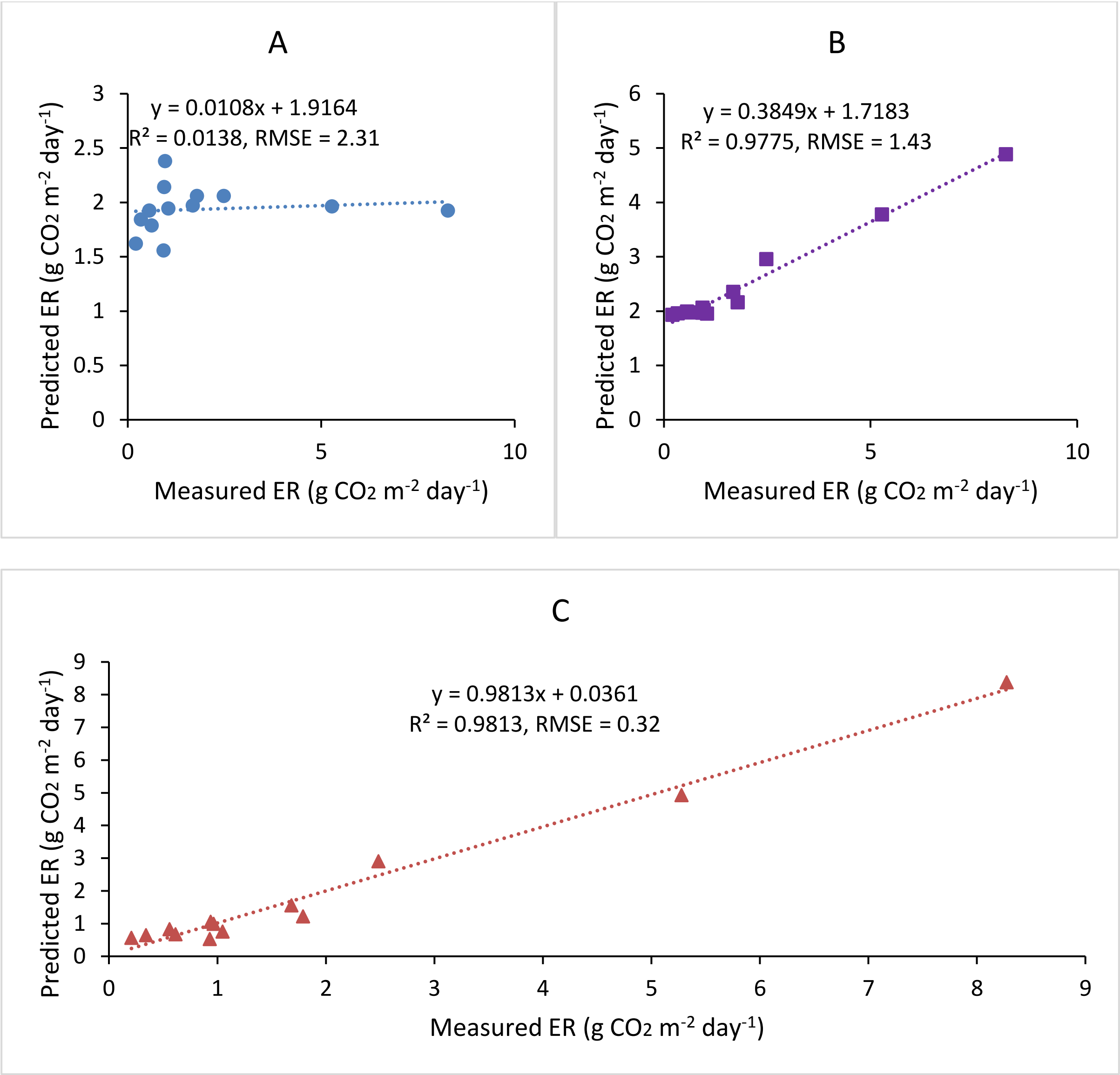

Figure A2: Examples of relationships between measured ER and the ER predicted using Equations 4 (A), 5 (B) and 6 (C).

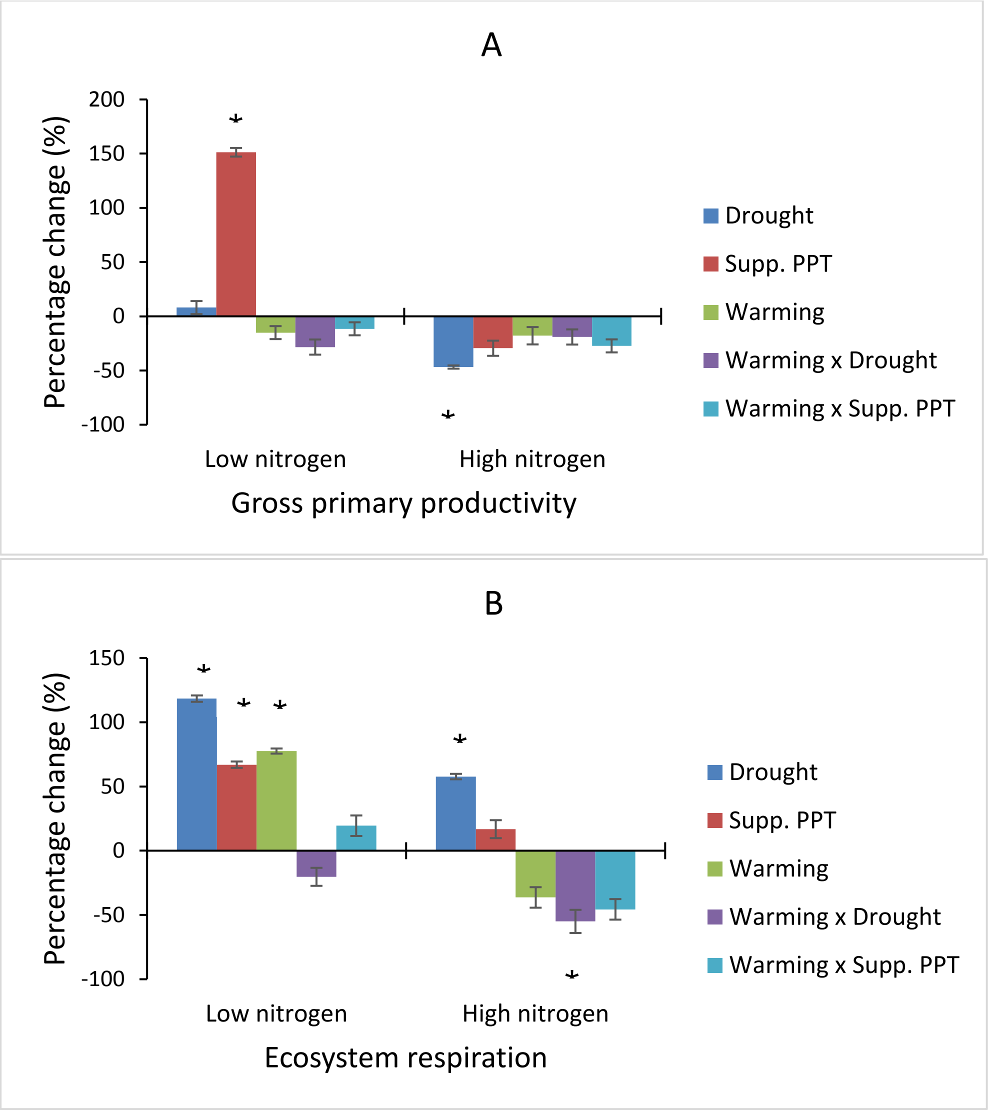

Figure A3: The effects of warming, drought, supplemental precipitation (Supp. PPT) and their interactions on the gross primary productivity (A) and ecosystem respiration (B) in the low nitrogen and high nitrogen cores (n = 4). Bars with asterisks represent significant change at 5% probability level in GPP or ER of soil cores subjected to altered climate treatment compared to cores with ambient climate treatment. Error bars represent standard error (n = 4).