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Reference:

Elst Evelyne, de Boeck Hans, Vanmaele Lisa, Verlinden Maya, Dhliwayo Pauline, Nijs Ivan.- Impact of climate extremes modulated by species characteristics and richness

Perspectives in plant ecology, evolution and systematics - ISSN 1433-8319 - 24(2017), p. 80-92

Full text (Publishers DOI): <http://dx.doi.org/doi:10.1016/J.PPEES.2016.12.007>

To cite this reference: <http://hdl.handle.net/10067/1389950151162165141>

Cite as: Elst E M, De Boeck H J, Vanmaele L, Verlinden M, Dhliwayo P, Nijs I (2017)

Impact of climate extremes modulated by species characteristics and richness. Perspectives in Plant Ecology, Evolution and Systematics 24: 80-92.

Impact of climate extremes modulated by species characteristics and richness

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Abstract

Drought and heat extremes can inflict substantial damage on plant communities, but the influences of species characteristics and richness on a community's ability to cope with these events are poorly understood.

In this study, we investigate the effects of species characteristics (drought tolerance and functional group) at different richness levels on (1) the ability to maintain green leaf area during a drought (and heat) extreme (resistance) and recover thereafter (resilience) and (2) the capacity of community biomass to recover by the end of the growing season. Our communities consist of drought-resistant and drought-sensitive grasses and nitrogen-fixing legumes (N-fixers) according to Ellenberg's humidity index, planted in different richness levels (1, 2 and 6 species) with 50% grasses and 50% N-fixers per community. This resulted in 28 different plant communities, repeated under six rain-out shelters, with each climate treatment (control, drought or drought with an additional heatwave) allocated to two main plots.

Grasses associated with drier habitats (assumed resistant) tolerated extremes better than grasses from wetter habitats (assumed sensitive), and grasses were generally more resistant and resilient against climate extremes than N-fixers. Species richness did not influence the green fraction in grasses, while N-fixers senesced more in diverse communities. In contrast, after recovery the N-fixers' biomass was independent of richness, while grasses produced more biomass in species-rich communities, probably due to positive effects of nitrogen fixation. Surprisingly, all these richness effects on stability were similar for resistant and sensitive species.

We demonstrated that understanding community responses to climate extremes requires a simultaneous focus on species characteristics and interspecific interactions (richness). Trends at the community level are determined by functional groups and response groups which

influence each other. To obtain generalised universal patterns, more insight at this level of complexity is needed.

Keywords: Climate extremes, Species richness, Species characteristics, Grassland

1. Introduction

Changes in the global climate are not only altering the average characteristics of climate systems, but also the frequency, intensity, spatial extent, duration and timing of climate extremes (IPCC, 2013; Herring *et al.*, 2014). A climate extreme occurs when the value of a weather or climate variable such as temperature or precipitation exceeds (or falls below) a threshold value near the upper (or lower) end of the range of observed values of the variable (IPCC, 2013). These discrete events often have a disproportionate impact on ecosystems relative to the temporal scale over which they occur. Reichstein *et al.* (2013) state that climate extremes can decrease regional ecosystem carbon stocks and therefore reinforce climate change. Despite an increased interest in the ecological impact of climate extremes, many questions remain unanswered especially regarding the significance of individual species characteristics and the importance of biodiversity in mediating the responses (Smith, 2011; Vogel *et al.*, 2012; Bailey and van de Pol, 2016).

Climate models project that summer droughts and heat waves in European grasslands will become longer and more frequent (IPCC, 2007; Evans *et al.*, 2011). Reduced water uptake by roots during drought extremes triggers a cascade of signalling plant hormones (such as abscisic acid, auxin, cytokinins, ethylene, etc.), resulting in stomatal closure and growth inhibition so that further water loss is abated (Chaves *et al.*, 2003; Wahid *et al.*, 2007; Liu *et al.*, 2012; Lipiec *et al.*, 2013; Hasanuzzaman *et al.*, 2013). As such, drought also induces heat stress since lower latent heat dissipation through transpiration increases tissue temperatures (De Boeck *et al.*, 2010). The connection between the water and energy balance also works the other way round: heat results in warmer leaves and higher atmospheric water demand, which will accelerate drought, especially when leaf area is high. Accordingly, heat and drought can reinforce each other (De Boeck and Verbeeck, 2011; Liu *et al.*, 2012; Lipiec *et al.*, 2013, De Boeck *et al.*, 2016), leading to reduced plant productivity and reproduction, early senescence

and eventually death (Chaves *et al.*, 2003; Wahid *et al.*, 2007; Lipiec *et al.*, 2013). Since naturally occurring climate extremes thus often feature combined stressors, it is important to examine their effects on plants and plant communities in conjunction. However, whether a climatic extreme induces an extreme ecological response (Smith, 2011) depends on the system's ability to withstand and/or recover from stress as well as on the degree of acclimation of the organisms, and will thus be species- and ecosystem-specific (Diez *et al.*, 2012).

Species-specific characteristics can have a profound impact on a plant's ability to cope with climate extremes. Some species withstand stress without loss of function (resistance), while others are able to recover rapidly after stress (resilience) (Virginia and Wall, 2001; Van Peer *et al.*, 2004; Lake, 2012). The stability of an ecosystem is determined by the traits of the dominant plant species (MacGillivray and Grime, 1995; Díaz 2001) or the presence of some specific species such as nitrogen-fixing legumes that can facilitate neighbour plants for some types of extremes (Pfisterer and Schmid, 2002; Arfin Kahn *et al.*, 2014; Hoekstra *et al.*, 2015; Hernandez and Picon-Cochard, 2016). In general, productive species with a high growth rate (e.g. grasses) have low resistance and high resilience, while slow growers with long-lived organs and low rates of nutrient turnover (e.g. trees) have high resistance and low resilience, indicating a trade-off (MacGillivray and Grime, 1995; Díaz 2001). Therefore, productive species, like many grasses, are expected to recover quickly after a climate extreme, especially in the presence of nitrogen-fixing legumes.

Besides species-specific characteristics, species richness can determine ecosystem stability, with species-rich communities generally being more stable than species-poor ones (Elton, 1958; Bloor and Bardgett, 2012; Roscher *et al.*, 2013). Multi-species communities are hypothesized to have an 'insured' stability due to the higher probability of containing (for example) drought-adapted species that can buffer ecosystem functioning (Yachi and Loreau

1999). Moreover, species richness is an important factor regulating the productivity of an ecosystem. Firstly, complementarity (including facilitation) in space and time enables more diverse communities to take up more resources compared to less diverse communities, resulting in higher productivity (Hooper, 1998; Nijs and Impens, 2000; Hooper *et al.*, 2005). In addition, species-rich communities have a higher probability (selection or sampling effect) that one or more productive species become dominant (Hooper *et al.*, 2005). Despite these well-defined mechanisms, the complex interplay between biodiversity, ecosystem stability and productivity yields varying results (e.g. Isbell *et al.*, 2015 [positive richness effects on productivity and stability]; Lanta *et al.*, 2012 [positive richness effects on productivity, negative richness effects on stability]; Van Ruijven and Bredense, 2005 [richness effect on resilience, but not on resistance]). Studies that systematically vary both species attributes and community structure may contribute to elucidating this interplay, but to our knowledge, species-specific drought tolerance was never included in experimental studies on diversity and climate extremes.

In this study, we explore effects of species characteristics (drought tolerance and functional group) and species richness on the impact of extreme drought only and extreme drought with an additional heat wave. We investigate the responses of drought-resistant, drought-sensitive and mixed plant communities (species selected according to Ellenberg's humidity index; Ellenberg *et al.*, 1991), composed of one, two or six species with an equal amount of grasses and nitrogen-fixing legumes (hereafter called N-fixers) during and after the imposed extremes. Specifically, effects of these controlled species characteristics (assumed resistant or sensitive, grass vs. N-fixer) at different richness levels are assessed on (1) the capacity to maintain green leaf area during a climate extreme (resistance) and recover thereafter (resilience) and (2) the capacity of community biomass to recover by the end of the growing season. We hypothesize that species associated with on average drier soil conditions will be

more able to keep plant tissue alive during extreme events compared to species that are found more frequently in wetter habitats. On the other hand, dry-habitat species should be less resilient based on their generally lower growth rates. Richness effects on resistance are expected to differ for species assumed resistant and sensitive because differences in productivity between these groups affect water use during drought. N-fixing species are predicted to facilitate grasses especially during recovery after extremes.

2. Materials and methods

2.1. Experimental design

For this experiment we selected 12 species that occur in Belgian grasslands (Table 1): three drought-sensitive grasses (SG), three drought-resistant grasses (RG), three drought-sensitive N-fixers (SN) and three drought-resistant N-fixers (RN). The drought tolerance level of these species was determined with Ellenberg's humidity index (Ellenberg *et al.*, 1991), since this is the most objective value for drought resistance available for all plants used in the experiment. The humidity index gives a value between one and twelve indicating the soil moisture conditions in which a plant prefers to grow (1: very dry - 12: underwater). On April 5 and 6 2012 (DOY 96 and 97), seeds were sown in quickpots containing sandy soil (78 % sand, 19 % loam, 3.0 % clay, pH 5.5, 260 g m⁻³ P, 110 g m⁻³ K, 97 g m⁻³ Mg, 1300 g m⁻³ Ca, 1.4 g m⁻³ Na), with a volumetric water content of 0.201 m³ m⁻³ (pF 2) at field capacity, 0.052 m³ m⁻³ (pF 3.3) at the point of reduced water availability and 0.030 m³ m⁻³ (pF 4.2) at wilting point (Bodemkundige Dienst van België, Leuven, Belgium). Between May 22 (DOY 143) and June 6 (DOY 158) 2012, the seedlings were transplanted into experimental containers (PVC tubes: Ø 40 cm, 50 cm deep) with the same soil. All transplanted seedlings were of comparable size per species and survival was more than 95%, with immediate replacement of individuals that died after transplanting. The containers were buried to keep the soil temperature in the

communities near natural conditions, and a closed lid at the bottom prevented water inflow from the surroundings. Each community consisted of 42 individuals, planted in a hexagonal grid with 4 cm interspace.

Experimental communities were either monocultures (S1, 42 individuals per species), or communities with two (S2, 21 individuals per species) or six (S6, 7 individuals per species) species composed of 50% grasses and 50% N-fixers. In addition, all species from the same functional group had the same drought tolerance according to Ellenberg *et al.* (1991). This resulted in S2 and S6 communities that could be classified as either drought-sensitive (50 % sensitive grasses and 50% sensitive N-fixers), drought-resistant (50 % resistant grasses and 50% resistant N-fixers) or mixed (50 % sensitive grasses and 50% resistant N-fixers or vice versa). Combining the species in this way resulted in twelve different monocultures, twelve different S2 and four different S6 combinations (Table 2). The arrangement of the different species within the hexagonal grid can be found in supplementary file 1. The communities were set up in six main plots of which two (main plots 4 and 6) served as controls (C), two (main plots 1 and 3) were exposed to a drought extreme (D) and two (main plots 2 and 5) were subjected to a drought extreme with an additional heat wave (DH). Therefore, this is a split-plot experiment with main plot nested within climate treatment. The treatments at subplot level are unbalanced, with three replicates in each climate treatment of all S1 and S2 communities (two replicates in one main plot and one replicate in the other main plot), while the S6 communities had three replicates in every main plot i.e. six replicates per climate treatment. This resulted in 288 experimental plant communities applied to subplots (containers) distributed across six main plots with 48 communities per main plot.

Before the start of the climate treatments, all communities were exposed to natural rainfall, which was quite abundant in June (133 mm compared to a long term average of 72 mm, Royal Meteorological Institute of Belgium). Between July 18 and 20 (DOY 200-202), the

main plots were covered by individual transparent polycarbonate rain-out shelters, in the shape of a hexagonal pyramid with the base at 180 cm high and a diameter of 5 m, ensuring quasi-unperturbed air exchange with the surroundings. After the shelters were installed, all communities were equally watered every three to four days with the aim of keeping the soil water content (SWC) in all containers in the upper third (i.e. between 0.14 and 0.20 m³ m⁻³) of the potential range. This watering continued throughout the whole experiment in C, while the D and DH treatments did not receive any water from 3 - 30 August 2012 (DOY 216 - 243). The DH treatment was subjected to an additional heat wave from 24 - 30 August 2012 (DOY 237 - 243). This heat wave was induced by 12 infrared heat lamps (2000 W each) per DH shelter, with two lamps attached to each side of the hexagonal frame of the rain shelters at a height of 130 cm. The heaters were switched on during daytime (9 am - 6 pm GMT+2) and were turned off at night, since during natural heat waves increases in minimum air temperatures are smaller than increases in maximum temperatures (De Boeck *et al.*, 2010). After the D and DH treatment, the communities were watered again. During the first nine days, seven rain events were simulated to enable the soil to rewet near field capacity, hereafter the plants were watered every three to four days with the aim of keeping the soil water content (SWC) in the upper third (i.e. between 0.14 and 0.20 m³ m⁻³) of the potential range. Plants could grow back until the end of September to allow short-term recovery and study the influence of species richness and species characteristics on resilience. Note that the plant communities in this experiment were in their establishment phase, but a climate extreme can also occur when a community is not yet fully established.

2.2. Measurements

During the experiment, the SWC was measured hourly with 0.3 m long Campbell CS616 Time Domain Reflectometry (TDR) probes (Campbell Scientific Ltd, Loughborough, UK) in

24 communities (one community of A, C, D, F, H, I, K, L, 2Ma, 2Mc, 2Md, 2Mf, 2Ra, 2Rc, 2Sb, 2Sc; two communities of 6Ma, 6Mb, 6R, 6S; letter codes see Tables 1 & 2) in one D (main plot 3) and one DH (main plot 5) main plot. In addition, the soil (T-thermocouple, 5 cm depth), surface (K-thermocouple OS36-01-K-80F, Omega) and air (T-thermocouple, 10 cm high, with radiation-shelter) temperature were measured from 24 August (DOY 237: start of the heat wave) until 22 September (DOY 266: end of the recovery period) in one C (main plot 6) and one DH (main plot 5) main plot, to compare the heated with the unheated situation. Two photosynthetic active radiation (PAR) sensors (SDEC, Tauxigny, France) were installed, one under a rain shelter (main plot 6) and one out in the open air, to calculate the loss of PAR when sunlight penetrates the shelter. Both the temperatures and the PAR values were logged automatically every thirty minutes (DL2e Data Logger, Delta-T Devices Ltd).

At plant level we measured whether plants were still alive in each community by scoring seven individuals per species (i.e. seven randomly selected individuals in S1, seven randomly selected individuals per species in S2, and all individuals in S6 communities) on four days during the extreme (2, 16, 24 and 31 August 2012 or DOY 216, 229, 237, 244) and by the end of the season on 26 September 2012 (DOY 270) after about one month of recovery. A plant was considered alive as long as there was green biomass visible. To enhance resolution, we also scored three randomly selected plants per species in each community on the same days (excluding the dead individuals, since their mortality was already represented in the first measure) and estimated the percentage leaf area that was still green in 5 % intervals. We multiplied both measures (in percent) for every species in the community to obtain the combined plant and leaf survival, further called 'green fraction'. In line with common management practice biomass was harvested at the beginning of October (1 - 5 October 2012; DOY 275-279). The aboveground biomass was harvested per community, separated by species, dried for 72 hours at 70 °C and weighed (0.01 g precision).

2.3. Analyses and statistics

All statistical analyses were conducted in R (R core team, 2013) and p-values were considered significant below 0.05. Final models were obtained by stepwise model simplification. PAR and air temperature under the rain shelters were compared to the outside conditions using pairwise t-tests. SWC differences between D and DH were tested at the beginning of the drought, before the onset of the heat, and at the end of the climate extremes. The average air, surface and soil temperature during the heat wave were compared between C and DH. The temporal dynamics in green fraction during the experiment were used as an absolute measure of resistance and resilience against drought and drought plus heat. Note that plants that visually appeared totally senesced in earlier observations, were often able to regrow from meristems during recovery, contributing to resilience of the green fraction. First, we tested for differences in resistance and resilience at community level (Com) between the climate treatments (fixed at main plot level), richness levels (fixed at subplot level) and their interactions over time (DOY, fixed), with a linear mixed effect model (used for split-plot experiments) with random factor main plot nested within climate treatment. In this model, differences in resistance or resilience between climate treatments are indicated by an interaction between climate treatment and DOY: a steeper decrease in green fraction over time in the drought and drought plus heat treatments compared to the control indicates a lower resistance, a steeper increase in green fraction afterwards indicates a higher resilience. However, the tolerance levels (Resistant, Sensitive or Mixed) and functional groups (Grasses, N-fixers or Mixed) could not be included in this test as not all possible combinations were present in the experiment (for example, S2 communities were always composed of one grass and one N-fixer, resulting in only mixed functional group communities being present at this richness level, whereas monocultures comprised single functional groups or tolerance levels).

To detect underlying causes of the trends in green fraction observed at community level, the same test was therefore performed on every separate group RG, SG, RN and SN. Though this test did not include a full statistical analysis, it does give information about the evolution of the green fraction over time, which is ecologically relevant. Therefore, this analysis was preferred over an analysis on calculated differences between the green fraction at the end and the beginning of the climate treatment. Due to three-way interactions that involved DOY, we assessed the importance of species richness for green fraction in two separate models, one at the end of the extreme and one at the end of the season. These models for community level and all four species groups include climate treatment as a fixed factor at the main plot level and richness as fixed factor at the subplot level and main plot nested within climate treatment as a random factor. In addition, the same models were used to test differences between monocultures and mixtures, instead of between richness levels. The latter two analyses (including richness and monocultures versus mixtures) were also used to test the influence of climate treatment and richness on biomass after recovery on both the community level and in the four species groups.

Finally, relations between SWC at the beginning or end of the extremes and green fraction (at the end of the extreme or after recovery) or biomass were explored with regression models to test whether SWC can explain or predict impacts on senescence and/or productivity.

3. Results

3.1. Abiotic measurements

The polycarbonate shelters increased air temperature by 0.4 °C while 4.8% of the incoming PAR was lost, indicating modest shelter-related changes. During the imposed heat wave, the daytime mean air, surface and soil temperature were significantly higher in DH than in C (Table 3).

Even though all communities were watered equally before the beginning of the extreme, the average SWC on the first day of the drought period was higher ($p < 0.001$) in DH (SWC=0.273) than in D (SWC=0.242), while both SWC values were above field capacity (Fig. 1). The latter was probably caused by the wet period in June, before the shelters were built. After 21 days of drought, the SWC values were 0.092 in DH and 0.098 in D ($p < 0.001$). Although, these differences are statistically significant, the values are still well above the wilting point, indicating that the ecological difference is probably minor. The subsequent seven-day heat period further reduced SWC ($p < 0.001$) in DH to 0.064 by increased evapotranspiration, leading to a soil water content close to the point of reduced water availability, as opposed to 0.084 in D ($p < 0.001$) where there was no additional heat (Fig. 1). In D, the drop in SWC during the drought extreme was higher in S6 (Δ SWC=0.180) than S1 (Δ SWC=0.138, $p=0.019$) and S2 (Δ SWC=0.140, $p=0.009$), implying that species-richer communities had used more water, while no significant S effects were observed in DH.

3.2. Green fraction

Before the experiment, N-fixers had a lower green fraction compared to grasses ($p < 0.001$), while sensitive and resistant species did not differ (Fig. 2). During the climate extreme, the green fraction over time declined more in D and DH compared to the controls, which were only subject to natural senescence (interaction climate treatment and DOY Table 4, Fig. 2). However, in contrast to expectation, DH did not affect the loss of green fraction more than D. Only in resistant grasses a difference between D and DH was observed, with the green fraction in D even declining faster than in DH ($p < 0.001$). Comparing the species groups separately, we found in C that sensitive N-fixers had greater natural senescence than both groups of resistant species (RG: $p=0.011$; RN: $p=0.037$). Both groups of sensitive species generally did not maintain green leaf area as well as resistant grasses in D (SG: $p=0.007$; SN:

$p < 0.001$), while all other groups lost green leaf area faster than resistant grasses when drought and heat events co-occurred ($p < 0.001$).

Combined effects of differences already present before the onset of the extreme events and a varying influence of the climate treatments, resulted in different green fractions after the extreme. At the community level and in all species groups these green fractions were lower in D and DH than in C ($p < 0.001$), except in resistant grasses where no difference was detected. The treatments D and DH had similar green fractions (Fig. 2), apart from in resistant grasses where they were higher in DH ($p < 0.001$), again illustrating that the additional warming did not increase mortality.

After the climate extreme treatments had ended, recovery (i.e. increase in green fraction by the end of the recovery period (DOY 270) compared to after the extremes (DOY 244)) was observed in both D ($p < 0.001$) and DH ($p < 0.001$; SN: $p = 0.030$), whereas in C, natural senescence continued (Fig. 2). As mentioned earlier, plants that visually appeared dead in earlier observations were often able to regrow from meristems, contributing to the increase in green fraction. Recovery was more outspoken at the community level and for grasses than for N-fixers, as indicated by the absence of a climate treatment \times DOY interaction in the latter (Table 4). Within species groups, plants exposed to D and DH recovered similarly, except the sensitive grasses which recovered faster in DH than in D (climate treatment \times DOY: Table 4). When comparing the different species groups within a climate treatment, we found that in the control treatment sensitive grasses declined at a faster rate than N-fixers (RN: $p = 0.006$; SN: $p = 0.012$). In D resistant grasses recovered less than the sensitive N-fixers ($p = 0.046$), while in DH sensitive grasses recovered less than resistant grasses ($p = 0.001$) and sensitive N-fixers ($p = 0.010$).

The end of season green fraction is the result of climate treatment-induced senescence and subsequent recovery. By the end of the growing season, less leaf area had survived in D than

in C ($p < 0.001$) in all species groups (RG: $p = 0.003$; SG: $p = 0.015$; RN: $p < 0.001$; SN: $p = 0.014$), indicating that plants that experienced D were unable to fully recover. At the community level and for grasses, green fraction in DH exceeded that in D (Com: $p = 0.002$; RG: $p < 0.001$; SG: $p = 0.001$). Resistant N-fixers, on the other hand, had lower ($p = 0.019$) green fraction in DH than in C, while the other groups fully recovered from the drought and heat wave.

Additional tests revealed that after the extreme there was no richness effect on green fraction at community level, but sensitive N-fixers had lost a greater fraction of their leaf area when grown in more species-rich communities, and resistant N-fixers followed the same trend (Table 4). When comparing monocultures with mixtures, the growth of resistant N-fixers was significantly different in mixtures compared to monocultures, while this was not the case for sensitive N-fixers. This suggests that the significant richness effect is mostly due to differences between the S2 and S6 communities (Table 4). At the same time, resistant grasses had a greater green fraction in more species-rich communities in DH (climate treatment \times richness interaction Table 4, Fig.3), but not in D. At the end of the growing season, a richness effect on green fraction at the community level and for resistant N-fixers was found, with a lower green fraction in more species-rich communities compared to monocultures (Table 4, Fig. 4). Sensitive N-fixers showed the same trend (Table 4). Richness nor the climate treatments had significant effects on the green fraction in grasses at the end of the growing season (Table 4).

Finally, the effects of drought tolerance on green fraction at community level of mixtures was further explored. Higher green fraction was observed in resistant compared to sensitive communities at the end of the climate extreme and higher green fraction in mixed and resistant communities compared to sensitive communities after recovery (Table 5).

3.3.Biomass

At the end of the season, species-rich communities had produced more biomass than species-poor ones. The biomass-richness curve was slightly flatter when climate extremes occurred, although climate treatment and richness did not interact (Fig. 5). The richness effects were found at the community level and for grasses (Table 4), indicating that when grasses grew together with N-fixers their biomass was higher compared to monocultures. In contrast, the N-fixers' biomass was independent of richness. Resistant N-fixers had lower biomass in D ($p=0.006$) and DH ($p<0.001$) than in C, while there was no D-DH difference. For all other species groups, biomass production was similar in the different climate treatments. The drought tolerance of S2 and S6 community had an influence on the community biomass (Table 5), resulting in a higher biomass in sensitive ($p<0.001$) and mixed communities ($p<0.001$) than in resistant communities.

3.4. Soil water content, survival and biomass

We found a positive relationship (in D and DH) between the SWC before the climate extreme and the communities' green fraction at the end of the extreme (D: $p=0.006$, DH: $p=0.013$), indicating a protective role of higher initial soil water content (Fig. 6A). In addition, in the DH treatment (only a trend in D) the green fraction at the end of the extreme was positively influenced by SWC at the end of the extreme ($p=0.002$), illustrating that senescence was lower if more water was still available at the end of the drought and heat period (Fig 6B). This graph suggests a non-linear relationship, with SWC above about $0.09 \text{ m}^3 \text{ m}^{-3}$ resulting in almost no senescence, and a steep decline in green fraction below this threshold. Finally, biomass production after the recovery period was negatively correlated with SWC at the end of the extreme in D ($p=0.002$) and DH ($p=0.001$) (Fig. 6C).

4. Discussion

In this study, we explored if and how the impact of climate extremes is modulated by species characteristics and richness. The imposed drought induced stress-related senescence and mortality, but surprisingly, additional heat at the end of the drought period did not exacerbate the effect. Moreover, this heat wave even stimulated recovery in those grass species deemed sensitive to drought on the basis of Ellenberg's indicator values. Possibly, the drought which developed was abated by the wet period before the shelters were built. This is in line with the soil water data collected in 48 of the experimental communities, indicating a minimum SWC of 8.4 % in drought and 6.4 % in drought with additional heat main plots, while the wilting point of the soil was 3.0 %. Dreesen *et al.* (2012) found that heat did not have a profound effect on plant growth and survival when water availability was high enough to compensate heating by transpirational cooling, which may have occurred in the current experiment as leaf temperature increments were not as high as expected. In addition, we know that climatic extremes not always induce an extreme ecological response (Smith, 2011). The occurrence of an extreme ecological response depends on many factors such as the previous experienced abiotic conditions (Dreesen *et al.*, 2014), the season (De Boeck *et al.*, 2011), the species composition (Grant *et al.*, 2014), etc.

During the climate treatment, resistant grasses (according to Ellenberg) were able to cope better with the imposed extremes than sensitive species (according to Ellenberg) in D, and better than all other species groups in DH, making them the most resistant group in this experiment. This suggests that Ellenberg values, which are normally used to indicate the preferred soil moisture for species (Ellenberg *et al.*, 1991), could also be used as a proxy for the resistance to climate extremes in grasses. This is in line with Backhaus *et al.* (2014), who reported that recurrent mild drought stress tends to improve drought resistance. In addition, Thompson *et al.* (1993) and Liu *et al.* (2000) found that species occupying dry habitats (low

Ellenberg humidity value) are capable of responding to drought by increasing their rooting depth, while species with higher Ellenberg values for humidity are less capable of such a response (Bartelheimer and Poschlod, 2015). This would explain the higher resistance to drought in resistant grasses in our experiment. However, in N-fixers no such link between the Ellenberg values and the resistance to climate extremes was found, nor did we observe consistent differences between species classified as sensitive and resistant species according to Ellenberg values during the recovery. This may imply limited use of Ellenberg values with regards to species resilience to climate extremes.

Overall, grasses were affected less during the extreme events than N-fixers, in line with Pfisterer and Schmid (2002) who found that N-fixing legumes generally lower an ecosystem's resistance. Likewise, whereas our grasses had higher resilience and fully recovered to the control level after a drought that also featured a heat wave, and slightly less after experiencing drought only, our N-fixers had lower resilience and most were unable to recover fully after all climate extremes. Grasses were consequently not only more resistant but also more resilient to perturbation compared to N-fixers, contradicting the trade-off between resistance and resilience predicted by MacGillivray and Grime (1995). Based on their traits, however, this finding is not complicated to explain, as grasses are typically deep rooting and productive and dispose of basal meristems. The deep roots guarantee prolonged access to water during drought, ensuring high resistance, while favourable conditions enable them to grow back quickly from their meristems, ensuring high resilience (Díaz 2001; Van Peer *et al.*, 2004). At the end of the climate extremes, N-fixers had lost a greater fraction of their green area in the more species-rich communities, indicating that they were adversely affected by interspecific neighbours during stress. This species richness effect persisted during recovery. In contrast, grasses did not respond to richness at the end of the extremes (except resistant grasses in DH) and no climate treatment or richness effect was found after recovery. This is in

contrast with Kreyling *et al.* (2007) who found that more diverse grassland communities exhibited less tissue die-back. However, Van Peer *et al.* (2004) found that more species rich communities typically have higher water use, as we also observed in our drought treatment, and have therefore lower water availability during drought and heat extremes, resulting in higher senescence. Probably, the negative effects of this increased soil drying in more species rich communities were offset in the grasses. As grasses' roots tend to grow deeper than N-fixers' roots, this could enable them to reach sufficient water even during the extremes (Craine *et al.*, 2002). In addition, root growth is typically promoted during drought (Gargallo-Garriga *et al.*, 2014), and in the grasses possibly stimulated by supplementary nitrogen availability due to the N-fixers' presence, which further increased the grasses' water uptake (Hooper, 1998; Loreau and Hector, 2001; De Boeck *et al.*, 2008, Hoekstra *et al.*, 2015). In contrast to the lack of richness effects on the green fraction of the grasses, the biomass versus richness curve for the grasses (and also the community level) showed a typical pattern, with more biomass in species-richer communities (Hector *et al.*, 1999; van Ruijven and Berendse, 2005; De Boeck *et al.*, 2008). Such a curve was not found in the N-fixers, which did not benefit as a group from increased biodiversity and also showed lower survival in more diverse communities (see above). Also in this response, grasses probably benefited from the nitrogen fixing capacity of N-fixers (Temperton *et al.*, 2007; De Boeck *et al.*, 2008; Arfin Khan *et al.*, 2014; Walter *et al.*, 2015; Hernandez and Picon-Cochard, 2016), explaining why grasses in species-richer communities (i.e. together with N-fixers) were able to produce more biomass while the N-fixers themselves were not stimulated by increased species richness. Kreyling *et al.* (2008) and Arfin Khan *et al.* (2014) found that legumes facilitated productivity in ambient conditions, however, they did not find a buffering effect of legumes against extreme drought. Surprisingly, the influence of species richness on the responses to extreme events as described in this paragraph was generally similar for species assumed resistant or sensitive based on

their Ellenberg humidity values, in spite of anticipated differences in productivity and thus water use between these groups. This clearly warrants further research on the role of species traits on resistance and resilience of complex communities.

We also explored relations between soil water content on the one hand and green fraction and biomass production on the other hand. The observed positive correlation between SWC before and green fraction at the end of the climate extremes indicates that even without knowledge of the number and identity of the species in a grassland community, the impact of drought and heat is to some extent predictable from measurements of the initial water content.

Communities with inherently greater water use, due to high leaf area and productivity, survived the extremes less well than more conservative communities (note that all communities received the same amount of precipitation and irrigation before the extremes). This high water use, which results in faster soil drying before and during the extreme event (Wang *et al.*, 2007), consequently brings low resistance but high regrowth (resilience) when conditions are favourable again (MacGillivray and Grime, 1995; Díaz 2001). This was confirmed by the relationship in DH (a trend in D) between SWC and green fraction at the end of the climate extremes and explains the negative correlation between biomass after recovery and SWC at the end of the extremes. Other mechanisms could also contribute to this high biomass production during recovery. In particular, the augmented stress that communities with a low SWC at the end of the extreme events likely experienced, may alleviate inter- and intraspecific competition through reduced plant survival (De Boeck *et al.*, 2008), promoting subsequent regrowth. In addition, the Birch effect can also play a role in triggering vigorous recovery (Birch, 1958; Jarvis *et al.*, 2007; Borken and Matzner, 2009; Dreesen *et al.* 2012). This arises from a short-term boost of microbial activity and organic matter mineralization upon rewetting after drought, giving rise to increased nutrient availability in the soil (Borken and Matzner, 2009).

5. Conclusions

With this study, we demonstrated that understanding community responses to climate extremes requires a simultaneous focus on species characteristics and species richness (i.e. interspecific interactions). Underlying the trends observed at the community level are divergent reactions in different functional groups or response groups, which mutually influence each other. More insight at this level of complexity is needed to generalize from individual studies to universal patterns that can eventually also advance ecosystem models.

Funding

The set-up was funded in part by FWO-Vlaanderen [grant 1506612N]

Acknowledgements

We thank M. Wellens and F. Kockelbergh for technical support.

References

- Arfin Kahn, M.A.S., Grant, K., Beierkuhnlein, C., Kreyling, J., Jentsch, A. 2014. Climatic extremes lead to species-specific legume facilitation in an experimental temperate grassland. *Plant soil* 379, 161-175.
- Backhaus, S., Kreyling, J., Grant, K., Beierkuhnlein, C., Walter, J., Jentsch, A. 2014. Recurrent mild drought events increase resistance toward extreme drought stress. *Ecosystems* 17, 1068-1081.
- Bailey, L.D., van de Pol, M. 2016. Tackling extremes: challenges for ecological and evolutionary research on extreme climatic events. *J. Anim. Ecol.* 85, 85-96.

- Bartelheimer, M., Poschlod, P. 2015. Functional characterizations of Ellenberg indicator values – a review on ecophysiological determinants. *Funct. Ecol.* 30, 506-516.
- Birch, H.F. 1958. The effect of soil drying on humus decomposition and nitrogen availability. *Plant Soil* 10, 9–31.
- Bloor, J.M.G., Bardgett, R.D. 2012. Stability of above-ground and below-ground processes to extreme drought in model grassland ecosystems: Interactions with plant species diversity and soil nitrogen availability. *Perspect. Plant Ecol.* 14, 193-204.
- Borken, W., Matzner, E. 2009. Reappraisal of drying and wetting effects on C and N mineralization and fluxes in soils. *Glob. Change Biol.* 15, 808–824.
- Chaves, M.M., Maroco, J.P., Pereira, J.S. 2003. Understanding plant responses to drought - from genes to the whole plant. *Funct. Plant Biol.* 30, 239–264.
- Craine, J.M., Wedin, D.A., Chapin III, F.S., Reich, P.B. 2002. Relationship between the structure of root systems and resource use for 11 North American grassland plants. *Plant Ecol.* 165, 85-100.
- De Boeck, H.J., Bassin, S., Verlinden, M., Zeiter, M., Hiltbrunner, E. 2016. Simulated heat waves affected alpine grassland only in combination with drought. *New Phytol.* 209, 531-541.
- De Boeck, H.J., Dreesen, F.E., Janssens, I.A., Nijs, I. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. *Global Change Biol.* 16, 1992-2000.
- De Boeck, H.J., Dreesen, F.E., Janssens, I.A., Nijs, I. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. *New Phytol.* 189, 806–817.
- De Boeck, H.J., Lemmens, C.M.H.M., Zavalloni, C., Gielen, B., Malchair, S., Carnol, M., Merckx, R., Van den Berge, J., Ceulemans, R., Nijs, I. 2008. Biomass production in experimental grasslands of different species richness during three years of climate warming. *Biogeosciences* 5, 585–594.

- De Boeck, H.J., Verbeeck, H. 2011. Drought-associated changes in climate and their relevance for ecosystem experiments and models. *Biogeosciences* 8, 1121–1130.
- Díaz, S., Cabido, M. 2001. Vive la différence: plant functional diversity matters to ecosystem processes. *Trends Ecol. Evol.* 16, 646–655.
- Diez, J.M., D'Antonio, C.M., Dukes, J.S., Grosholz, E.D., Olden, J.D., Sorte, C.J., Blumenthal, D.M., Bradley, B.A., Early, R., Ibáñez, I., Jones, S.J., Lawler, J.J., Miller, L.P. 2012. Will extreme climatic events facilitate biological invasions? *Front Ecol. Environ.* 10, 249–257.
- Dreesen, F.E., De Boeck, H.J., Horemans, J.A., Janssens, I.A., Nijs, I. 2015. Recovery dynamics and invasibility of herbaceous plant communities after exposure to experimental climate extremes. *Basic Appl. Ecol.* 16, 583-591.
- Dreesen, F.E., De Boeck, H.J., Janssens, I.A., Nijs, I. 2012. Summer heat and drought extremes trigger unexpected changes in productivity of a temperate annual/biannual plant community. *Environ. Exp. Bot.* 79, 21–30.
- Dreesen, F.E., De Boeck, H.J., Janssens, I.A., Nijs, I. 2014. Do successive climate extremes weaken the resistance of plant communities? An experimental study using plant assemblages. *Biogeosciences* 11, 109-121.
- Ellenberg, H., Weber, H.E., Düll, R., Wirth, V., Werner, W., Paulissen, D. 1991. Zeigerwerte von Pflanzen in Mitteleuropa. *Scripta Geobotanica* 18, 1–248.
- Elton, C.S. 1958. *The ecology of invasions by plants and animals*. Methuen, London, UK.
- Evans, S.E., Byrne, K.M., Lauenroth, W.K., Burke, I.C. 2011. Defining the limit to resistance in a drought-tolerant grassland: long-term severe drought significantly reduces the dominant species and increases ruderals. *J. Ecol.* 99, 1500–1507.
- Gargallo-Garriga, A., Sardans, J., Pérez-Trujillo, M., Rivas-Ubach, A., Oravec, M., Vecerova, K., Urban, O., Jentsch, A., Kreyling, J., Beierkuhnlein, C., Parella, T., Peñuelas, J. 2014. Opposite metabolic responses of shoots and roots to drought. *Sci. Rep.* 4, 6829.

- Grant, K., Kreyling, J., Heilmeyer, H., Beierkuhnlein, C., Jentsch, A. 2014. Extreme weather events and plant-plant interactions: shifts between competition and facilitation among grassland species in the face of drought and heavy rainfall. *Ecol. Res.* 29, 991-1001.
- Hasanuzzaman, M., Nahar, K., Alam, M.M., Roychowdhury, R., Fujita, M. 2013. Physiological, biochemical, and molecular mechanisms of heat stress tolerance in plants. *Int. J. Mol. Sci.* 14, 9643–9684.
- Hector, A., Schmid, B., Beierkuhnlein, C., Caldeira, M.C., Diemer, M., Dimitrakopoulos, P.G., Finn, J.A., Freitas, H., Giller, P.S., Good, J., Harris, R., Högberg, P., Huss-Danell, K., Joshi, J., Jumpponen, A., Körner, C., Leadley, P.W., Loreau, M., Minns, A., Mulder, C.P.H., O'Donovan, G., Otway, S.J., Pereira, J.S., Prinz, A, Read, D.J., Scherer-Lorenzen, M., Schulze, E.D., Siamantziouras, A.S.D., Spehn, E.M., Terry, A.C., Troumbis, A.Y., Woodward, F.I., Yachi, S., Lawton, J.H. 1999. Plant diversity and productivity experiments in European grasslands. *Science* 286, 1123–1127.
- Hernandez, P., Picon-Cochard, C. 2016. Presence of *Trifolium repens* promotes complementarity of water use and N facilitation in diverse grass mixtures. *Front Plant. Sci.* 7, 538.
- Herring, S.C., Hoerling, M.P., Peterson, T.C., Stott, P.A. 2014. Explaining extreme events of 2013 from a climate perspective. *B. Am. Meteorol. Soc.* 95, S1–S96.
- Hoekstra, N.J., Suter, M., Finn, J.A., Husse, S., Lüscher, A. 2015. Do belowground vertical niche differences between deep- and shallow-rooted species enhance resource uptake and drought resistance in grassland mixtures? *Plant soil* 394, 21-34.
- Hooper, D.U. 1998. The role of complementarity and competition in ecosystem responses to variation in plant diversity. *Ecol.* 79, 704-719.
- Hooper, D.U., Chapin III, F.S., Ewel, J.J., Hector, E., Inchausti, S., Lavorel, S., Lawton, J.H., Lodge, D.M., Loreau, M., Naeem, S., Schmid, D., Satälä, H., Symstad, A.J., Vandermeer, J.,

Wardle, D.A. 2005. Effects of biodiversity on ecosystem functioning: a consensus of current knowledge. *Ecol. Monogr.* 75, 3-35.

IPCC, 2007: Climate Change 2007: Mitigation. Contribution of Working Group III to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change [B. Metz, O.R. Davidson, P.R. Bosch, R. Dave, L.A. Meyer (eds)], Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA, 851 pp.

IPCC, 2013: Climate Change 2013: The physical science basis. Contribution of working group I to the fifth assessment report of the intergovernmental panel on climate change [Stocker, T.F., Qin, D., Plattner, G.K., Tignor, M., Allen, S.K., Boschung, J., Nauels, A., Xia, Y., Bex, V., Midgley, P.M., *et al*]. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA, 1535 pp.

Isbell, F., Craven, D., Connolly, J., Loreau, M., Schmid, B., Beierkuhnlein, C., Bezemer, T.M., Bonin, C., Bruelheide, H., de Luca, E., Ebeling, A., Griffin, J.N., Guo, Q., Hautier, Y., Hector, A., Jentsch, A., Kreyling, J., Lanta, V., Manning, P., Meyer, S.T., Mori, A.S., Naeem, S., Niklaus, P.A., Polley, H.W., Reich, P.B., Roscher, C., Seabloom, E.W., Smith, M.D., Thakur, M.P., Tilman, D., Tracy, B.F., van der Putten, W.H., van Ruijven, J., Weigelt, A., Weisser, W.W., Wilsey, B., Eisenhauer, N. 2015. Biodiversity increases the resistance of ecosystem productivity to climate extremes. *Nature* 526, 574-577.

Jarvis, P., Rey, A., Petsikos, C., Wingate, L., Rayment, M., Pereira, J., Banza, J., David, J., Miglietta, F., Borghetti, M., Manca, G., Valentini, R. 2007. Drying and wetting of Mediterranean soils stimulates decomposition and carbon dioxide emission: the “Birch effect”. *Tree Physiol.* 27, 929–940.

Kreyling, J., Wenigmann, M., Beierkuhnlein, C., Jentsch, A. 2008. Effects of extreme weather events on plant productivity and tissue die-back are modified by community composition. *Ecosystems* 11: 752-763.

- Lake, P.S. 2012. Resistance, resilience and restoration. *Ecol. Manage. Restor.* 14, 20-24.
- Lanta, V., Doležal, J., Zemková, L., Lepš, J. 2012. Communities of different plant diversity respond similarly to drought stress: experimental evidence from field non-weeded and greenhouse conditions. *Naturwissenschaften* 99, 473–482.
- Lipiec, J., Doussan, C., Nosalewicz, A., Kondracka, K. 2013. Effect of drought and heat stresses on plant growth and yield: a review. *Int. Agrophysics* 27, 463–477.
- Liu, Z.M., Thompson, K., Spencer, R.E., Readeer, R.J. 2000. A comparative study of morphological responses of seedling roots to drying soil in 20 species from different habitats. *Acta Bot. Sin.* 42, 628–635.
- Liu, T., Zhang, L., Yuan, Z., Hu, X., Lu, M., Wang, W., Wang, Y. 2012. Identification of proteins regulated by ABA in response to combined drought and heat stress in maize roots. *Acta Physiol. Plant* 35, 501–513.
- Loreau, M., Hector, A. 2001. Partitioning selection and complementarity in biodiversity experiments. *Nature* 412, 72–76.
- MacGillivray, C.W., Grime, J.P., The Integrated Screening Programme (ISP) Team. 1995. Testing predictions of the resistance and resilience of vegetation subjected to extreme events. *Funct. Ecol.* 9, 640-649.
- Nijs, I., Impens, I. 2000. Biological diversity and probability of local extinction of ecosystems. *Funct. Ecol.* 14, 46-54.
- Pfisterer, A.B., Schmid, B. 2002. Diversity-dependent production can decrease the stability of ecosystem functioning. *Nature* 416, 84–86.
- R Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org/>.
- Reichstein, M., Bahn, M., Ciais, P., Frank, D., Mahecha, M.D., Seneviratne, S.I., Zscheischler, J., Beer, C., Buchmann, N., Frank, D.C., Papale, D., Rammig, A., Smith, P., Thonicke, K.,

- van der Velde, M., Vicca, S., Walz, A., Wattenbach, M. 2013. Climate extremes and the carbon cycle. *Nature* 500, 287-295.
- Roscher, C., Fergus, A.J.F., Petermann, J.S., Buchmann, N., Schmid, B., Schulze, E.D. 2013. What happens to the sown species if a biodiversity experiment is not weeded? *Basic Appl. Ecol.* 14, 187-198.
- Smith, M.D. 2011. An ecological perspective on extreme climatic events: a synthetic definition and framework to guide future research. *J. Ecol.* 99, 656–663.
- Temperton, V.M., Mwangi, P.M., Scherer-Lorenzen, M., Schmid, B., Buchmann, M. 2007. Positive interactions between nitrogen-fixing legumes and four different neighbouring species in a biodiversity experiment. *Oecologia* 151, 190–205.
- Thompson, K., Hodgson, J., Grime, J.P., Rorison, I.H., Band, S.R., Spencer, R.E. 1993. Ellenberg numbers revisited. *Phytocoenologia* 23, 277–289.
- Van Peer, L., Nijs, I., Reheul, D., De Cauwer, B. 2004. Species richness and susceptibility to heat and drought extremes in synthesized grassland ecosystems: compositional vs physiological effects. *Funct. Ecol.* 18, 769–778.
- Van Ruijven, J., Berendse, F. 2005. Diversity-productivity relationships: initial effects, long-term patterns, and underlying mechanisms. *P. Natl. Acad. Sci. USA* 102, 695–700.
- Virginia, R.A., Wall, D.H. 2001. Principles of ecosystem function. *Encyclopedia of Biodiversity*, Vol. 2 (ed. S.A. Levin), pp. 345–352. Academic Press, London.
- Vogel, A., Scherer-Lorenzen, M., Weigelt, A. 2012. Grassland resistance and resilience after drought depends on management intensity and species richness. *Plos One* 7, e36992.
- Wahid, A., Gelani, S., Ashraf, M., Foolad, M. 2007. Heat tolerance in plants: an overview. *Environ. Exp. Bot.* 61, 199–223.

Walter, J., Kreyling, J., Singh, B.K., Jentsch, A. 2015. Effects of extreme weather events and legume presence on mycorrhization of *Plantago lanceolata* and *Holcus lanatus* in the field. *Plant Biol.* 18, 262-270.

Wang, Y., Yu, S., Wang, J. 2007. Biomass-dependent susceptibility to drought in experimental grassland communities. *Ecol. Lett.* 10, 401-410.

Yachi, S., Loreau, M. 1999. Biodiversity and ecosystem productivity in a fluctuating environment: the insurance hypothesis. *Proc. Natl. Acad. Sci. USA* 96, 1463–1468.

Tables

Table 1: Grassland species used in this experiment with their scientific and English name, code used in the experiment, functional group, drought tolerance level and Ellenberg's humidity value.

Code	Species name	English name	Functional group	Drought tolerance	Ellenberg value
A	<i>Agrostis stolonifera</i> L.	Fiorin	Grass	Sensitive	7
B	<i>Deschampsia cespitosa</i> (L.) P. Beauv.	Tussock grass	Grass	Sensitive	7
C	<i>Festuca rubra</i> L.	Red fescue	Grass	Sensitive	6
D	<i>Dactylis glomerata</i> L.	Orchard grass	Grass	Resistant	5
E	<i>Lolium perenne</i> L.	English ryegrass	Grass	Resistant	5
F	<i>Festuca filiformis</i> Pourr.	Hairy fescue	Grass	Resistant	4
G	<i>Lathyrus pratensis</i> L.	Meadow vetchling	N-fixer	Sensitive	6
H	<i>Trifolium repens</i> L.	White clover	N-fixer	Sensitive	5*
I	<i>Vicia cracca</i> L.	Tufted vetch	N-fixer	Sensitive	6
J	<i>Lotus corniculatus</i> L.	Bird's-foot trefoil	N-fixer	Resistant	4
K	<i>Medicago lupulina</i> L.	Black medic	N-fixer	Resistant	4
L	<i>Trifolium medium</i> L.	Zigzag clover	N-fixer	Resistant	4

**T. repens* is considered a drought-sensitive species in literature (Dreesen *et al.*, 2015)

Table 2: Species composition of the twelve monocultures (S1), twelve S2-communities and four S6-communities and number of repeats of each community type per climate treatment.

See Table 1 for species codes.

Richness	Sensitive	Mixed	Resistant
Monocultures (3 replicates per climate treatment)	1A 1B 1C 1G 1H 1I		1D 1E 1F 1J 1K 1L
S2-communities (3 replicates per climate treatment)	2Sa = A-G 2Sb = B-H 2Sc = C-I	2Ma = A-J 2Mb = B-K 2Mc = C-L 2Md = D-G 2Me = E-H 2Mf = F-I	2Ra = D-J 2Rb = E-K 2Rc = F-L
S6-communities (6 replicates per climate treatment)	6S = A-B-C-G-H-I	6Ma = A-B-C-J-K-L 6Mb = D-E-F-G-H-I	6R = D-E-F-J-K-L

Table 3: Mean air, surface and soil temperature \pm se during daytime (i.e. when heat lamps were on, 9 am – 6 pm GMT+2) in control (C) and drought + heat (DH) treatments during the heat period from 24 to 31 August 2012 (DOY 237 - 243), with temperature difference between climate treatments and p-value (significant in bold).

	T _{mean} C (°C)	T _{mean} DH (°C)	Δ T (°C)	p-value
Air	21.1 \pm 0.4	23.7 \pm 0.4	2.7	<0.001
Surface	19.5 \pm 0.2	23.1 \pm 0.3	3.6	<0.001
Soil	17.8 \pm 0.2	20.9 \pm 0.3	3.1	<0.001

Table 4: F- and p-values (significant in bold) of the best models after stepwise model simplification of the linear mixed-effect models conducted in this study. The model testing green fraction over time during the climate extreme and the recovery includes the fixed factors treatment (T), richness (R) and day of the year (D) and their interactions (x). The effect of climate treatment (T) and monocultures vs. mixed (M) communities or richness levels (R) and their interactions (x) on green fraction and biomass production were tested at the end of the climate extremes (DOY 244) and after recovery (DOY 270). The degrees of freedom of every factor are indicated next to the factor between brackets. For every model, the variance (Var) and the standard error of the error terms is given. Error (T/P) is the error of the random factor where main plot (P) is nested within climate treatment (T), Error (Res) is the residual error term.

	COMMUNITY LEVEL		RESISTANT GRASSES		SENSITIVE GRASSES		RESISTANT N-FIXERS		SENSITIVE N-FIXERS	
	F-value	p-value								
Green fraction: resistance										
TxRxD (2)	0.028	0.973	1.706	0.179	1.904	0.146	0.018	0.982	0.064	0.938
RxD (1)	3.387	0.065	1.445	0.227	0.022	0.881	3.697	0.053	3.606	0.057
TxD (2)	20.272	<0.001	22.151	<0.001	23.863	<0.001	9.195	<0.001	7.460	<0.001
TxR (2)	0.848	0.426	12.037	<0.001	1.577	0.206	2.090	0.138	1.741	0.152
R (1)	0.340	0.560	/	/	3.693	0.054	2.298	0.128	1.332	0.242
Error (T/P)	Var: 0.005 \pm 0.068		Var: 0.012 \pm 0.111		Var: 0.009 \pm 0.096		Var: 0.001 \pm 0.032		Var: 0.002 \pm 0.039	
Error (Res)	Var: 0.054 \pm 0.233		Var: 0.016 \pm 0.126		Var: 0.036 \pm 0.191		Var: 0.085 \pm 0.292		Var: 0.116 \pm 0.341	

Green fraction: resilience

TxRxD (2)	0.238	0.782	2.875	0.054	0.469	0.619	0.031	0.969	0.409	0.658
RxD (1)	0.228	0.628	2.440	0.115	0.527	0.464	0.609	0.431	0.004	0.952
TxD (2)	8.346	<0.001	4.982	0.007	14.228	<0.001	1.331	0.260	0.787	0.450
TxR (2)	0.005	0.990	3.154	0.042	0.503	0.595	0.077	0.931	0.298	0.740
R (1)	8.009	0.005	/	/	0.009	0.934	10.988	<0.001	6.696	0.009
D (1)	/	/	/	/	/	/	2.405	0.121	1.730	0.188
T (2)	/	/	/	/	/	/	1.007	0.030	0.977	0.050
Error (T/P)	Var: 0.016 ± 0.125		Var: 0.026 ± 0.162		Var: 0.016 ± 0.125		Var: 0.008 ± 0.090		Var: 0.008 ± 0.089	
Error (Res)	Var: 0.056 ± 0.237		Var: 0.022 ± 0.150		Var: 0.049 ± 0.221		Var: 0.108 ± 0.328		Var: 0.142 ± 0.377	

Green fraction: end extreme

Monocultures-Mixtures

TxM (2)	0.043	0.957	3.506	0.032	0.290	0.756	0.076	0.949	0.393	0.709
T (2)	1.577	0.043	/	/	0.507	0.083	6.270	0.009	1.030	0.017
M (1)	3.169	0.075	/	/	0.005	0.942	4.422	0.031	3.287	0.060
Error (T/P)	Var: 0.013 ± 0.112		Var: 0.025 ± 0.158		Var: 0.022 ± 0.150		Var: 0.003 ± 0.058		Var: 0.004 ± 0.063	
Error (Res)	Var: 0.067 ± 0.259		Var: 0.024 ± 0.156		Var: 0.057 ± 0.239		Var: 0.110 ± 0.331		Var: 0.131 ± 0.362	

Richness

TxR (2)	0.074	0.928	5.736	0.004	0.671	0.495	0.157	0.920	0.101	0.932
T (2)	1.577	0.043	/	/	0.565	0.082	1.888	0.011	6.420	0.018
R (1)	2.336	0.127	/	/	0.139	0.708	2.887	0.067	3.748	0.047
Error (T/P)	Var: 0.013 ± 0.112		Var: 0.025 ± 0.158		Var: 0.022 ± 0.150		Var: 0.006 ± 0.079		Var: 0.003 ± 0.052	
Error (Res)	Var: 0.067 ± 0.259		Var: 0.024 ± 0.153		Var: 0.057 ± 0.239		Var: 0.110 ± 0.332		Var: 0.129 ± 0.360	

Green fraction: end recovery

Monocultures-Mixtures

TxM (2)	0.337	0.711	1.339	0.259	0.265	0.734	0.541	0.587	0.023	0.984
T (2)	0.363	0.419	0.272	0.501	0.369	0.290	0.743	0.087	0.571	0.213
M (1)	3.169	0.042	0.101	0.756	2.241	0.132	13.989	<0.001	1.105	0.284
Error (T/P)	Var: 0.014 ± 0.119		Var: 0.0209 ± 0.145		Var: 0.008 ± 0.090		Var: 0.009 ± 0.094		Var: 0.010 ± 0.099	
Error (Res)	Var: 0.047 ± 0.217		Var: 0.0202 ± 0.142		Var: 0.040 ± 0.200		Var: 0.103 ± 0.321		Var: 0.158 ± 0.398	

Richness

TxR (2)	0.188	0.827	0.177	0.838	0.113	0.900	0.005	0.994	0.620	0.543
T (2)	0.371	0.419	0.285	0.501	0.342	0.308	0.745	0.096	0.569	0.214
R (1)	6.618	0.010	0.253	0.621	0.330	0.571	8.480	0.004	3.130	0.075
Error (T/P)	Var: 0.014 ± 0.119		Var: 0.020 ± 0.145		Var: 0.008 ± 0.090		Var: 0.010 ± 0.098		Var: 0.010 ± 0.099	
Error (Res)	Var: 0.046 ± 0.216		Var: 0.020 ± 0.142		Var: 0.040 ± 0.200		Var: 0.106 ± 0.326		Var: 0.158 ± 0.398	

Biomass: end recovery

Monocultures-Mixtures

TxM (2)	1.985	0.137	0.359	0.693	0.074	0.927	0.087	0.914	0.196	0.817
T (2)	0.680	0.167	0.190	0.671	0.579	0.555	2.437	0.016	1.063	0.335
M (1)	33.148	<0.001	9.859	0.002	6.067	0.014	0.573	0.444	1.550	0.215
Error (T/P)	Var: 63.70 ± 7.981		Var: 0.048 ± 0.219		Var: 0.000 ± 0.000		Var: 0.000 ± 0.000		Var: 0.000 ± 0.000	
Error (Res)	Var: 384.67 ± 19.613		Var: 0.457 ± 0.676		Var: 2.283 ± 1.513		Var: 0.893 ± 0.945		Var: 2.055 ± 1.433	

Richness										
TxR (2)	1.372	0.252	0.675	0.500	0.032	0.967	0.225	0.793	0.071	0.929
T (2)	0.427	0.259	0.176	0.693	0.573	0.558	1.550	0.016	1.440	0.126
R (1)	22.656	<0.001	4.144	0.041	4.098	0.043	0.269	0.600	0.338	0.567
Error (T/P)	Var: 493.7 ± 22.22		Var: 0.053 ± 0.229		Var: 0.000 ± 0.000		Var: 0.000 ± 0.000		Var: 0.000 ± 0.000	
Error (Res)	Var: 5657 ± 75.21		Var: 0.470 ± 0.685		Var: 2.313 ± 1.521		Var: 0.893 ± 0.945		Var: 2263 ± 47.57	

Table 5: F- and p-values (significant in bold) of the best models after stepwise model simplification of the linear mixed-effect models conducted on the S2 and S6 communities.

The models test the influence of climate treatment (T), richness (R) and community drought tolerance (To) and their interactions (x) on the green fraction at the end of the climate extremes (DOY 244) and after recovery (DOY 270) and the biomass after recovery (DOY 270). The degrees of freedom of every factor are indicated next to the factor between brackets. For every model, the variance (Var) and the standard error of the error terms is given. Error (T/P) is the error of the random factor where main plot (P) is nested within climate treatment (T), Error (Res) is the residual error term.

	Green fraction: end extreme		Green fraction: end recovery		Biomass: end recovery	
	F-value	p-value	F-value	p-value	F-value	p-value
TxRxTo (4)	0.612	0.596	0.585	0.648	0.441	0.752
TxR (2)	0.161	0.842	0.260	0.758	0.596	0.533
TxTo (4)	0.337	0.813	0.456	0.737	0.573	0.690
RxTo (2)	0.708	0.463	1.256	0.256	1.777	0.189
T (2)	0.972	0.036	0.367	0.360	0.538	0.174
R (1)	0.329	0.563	3.333	0.067	6.838	0.011
To (2)	5.855	0.003	3.099	0.045	11.615	<0.001
Error (T/P)	Var: 0.010 ± 0.100		Var: 0.010 ± 0.100		Var: 34.057 ± 5.836	
Error (Res)	Var: 0.056 ± 0.236		Var: 0.039 ± 0.196		Var: 282.092 ± 16.796	

Figures

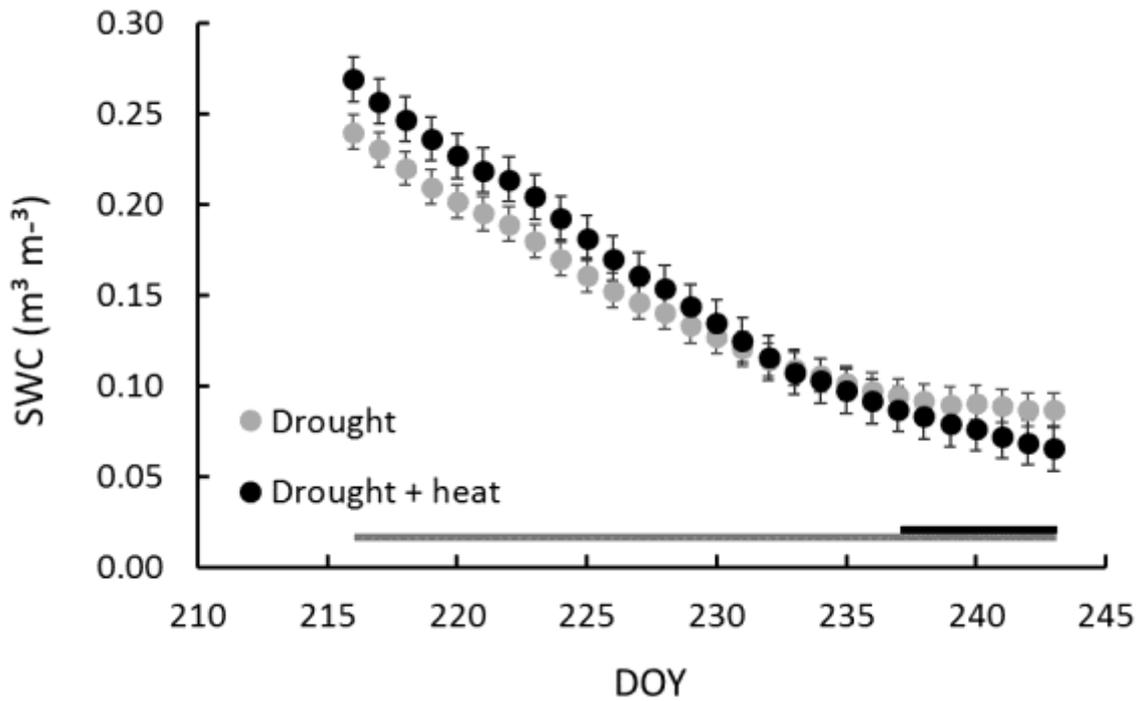


Figure 1: Average daily soil water content (SWC) \pm se in 24 communities exposed to drought and 24 communities exposed to drought with additional heat. The grey (D) and black bar (DH) indicate the timing of the drought extreme and the additional warming. DOY = day of the year.

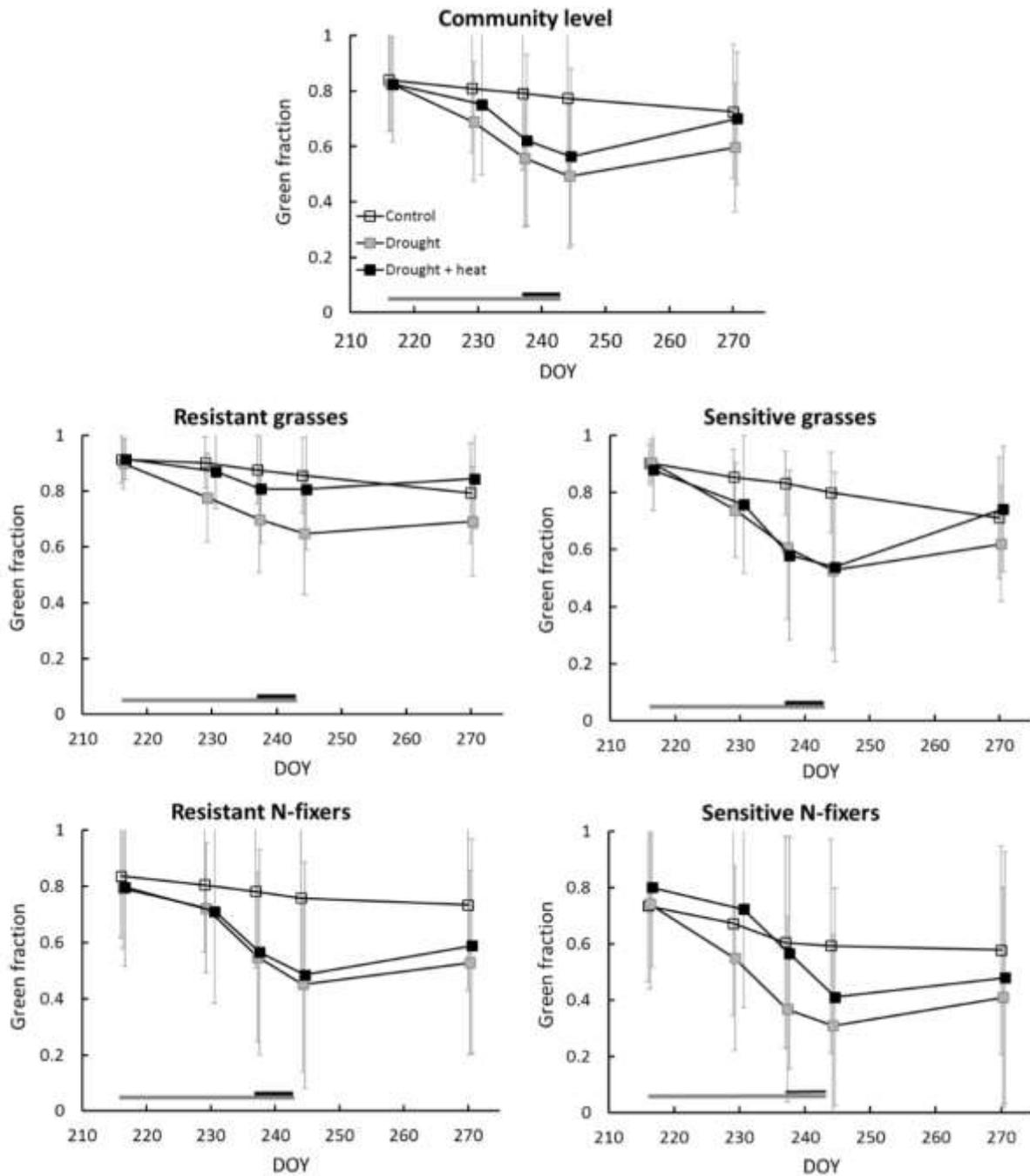


Figure 2: Average green fraction \pm sd over time in the different climate treatments during the climate extreme and the recovery, on community level and in the different species groups.

The grey bar indicates the drought extreme and the black bar the additional heating in the DH treatment. DOY = day of the year. Note that standard deviations are large as multiple richness levels are combined in each data point (for statistical analyses see Tables 4 and 5).

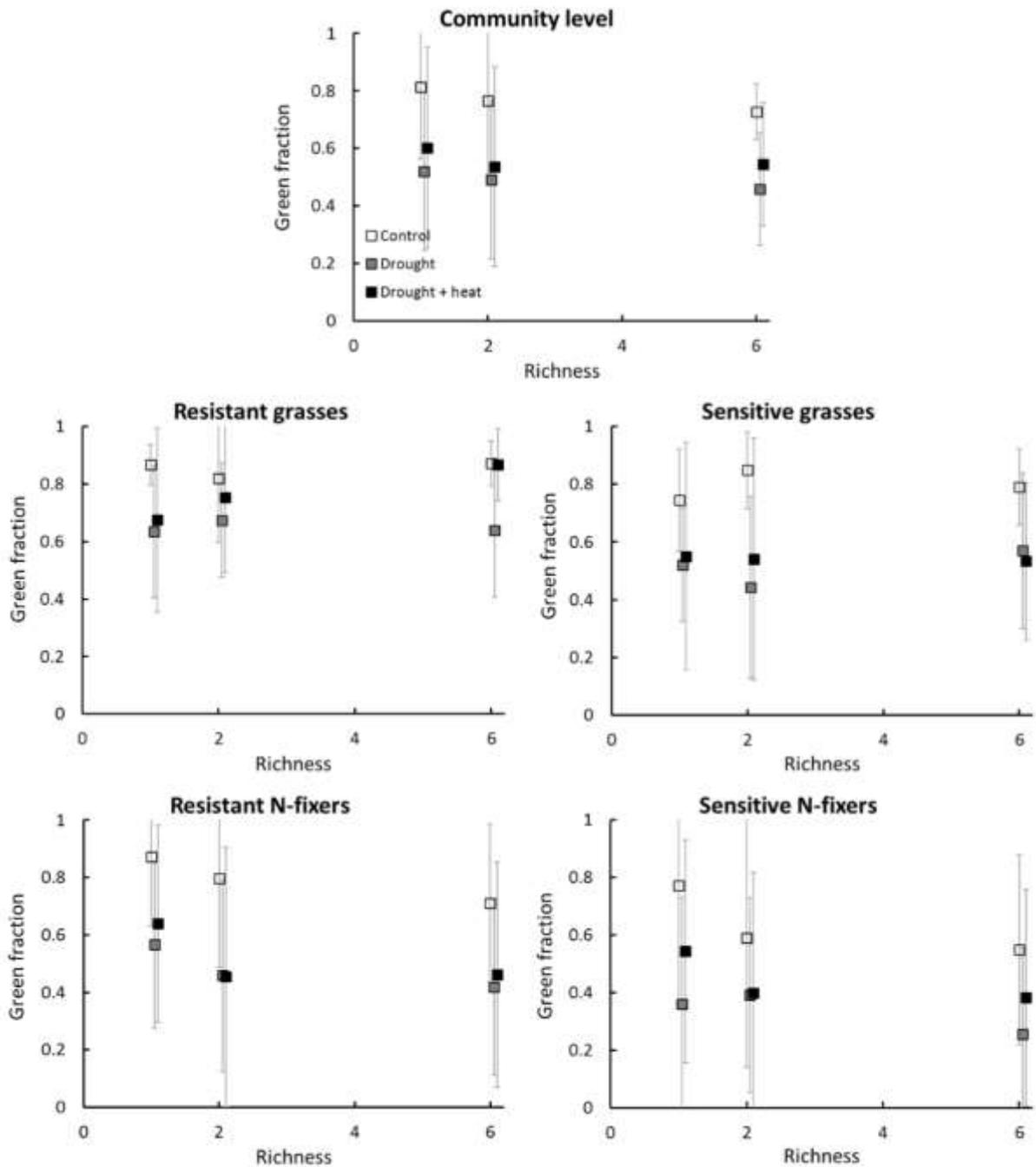


Figure 3: Average green fraction \pm sd at the end of the climate extreme (DOY 244) versus the communities' species richness in the different climate treatments, on community level and in the different species groups.

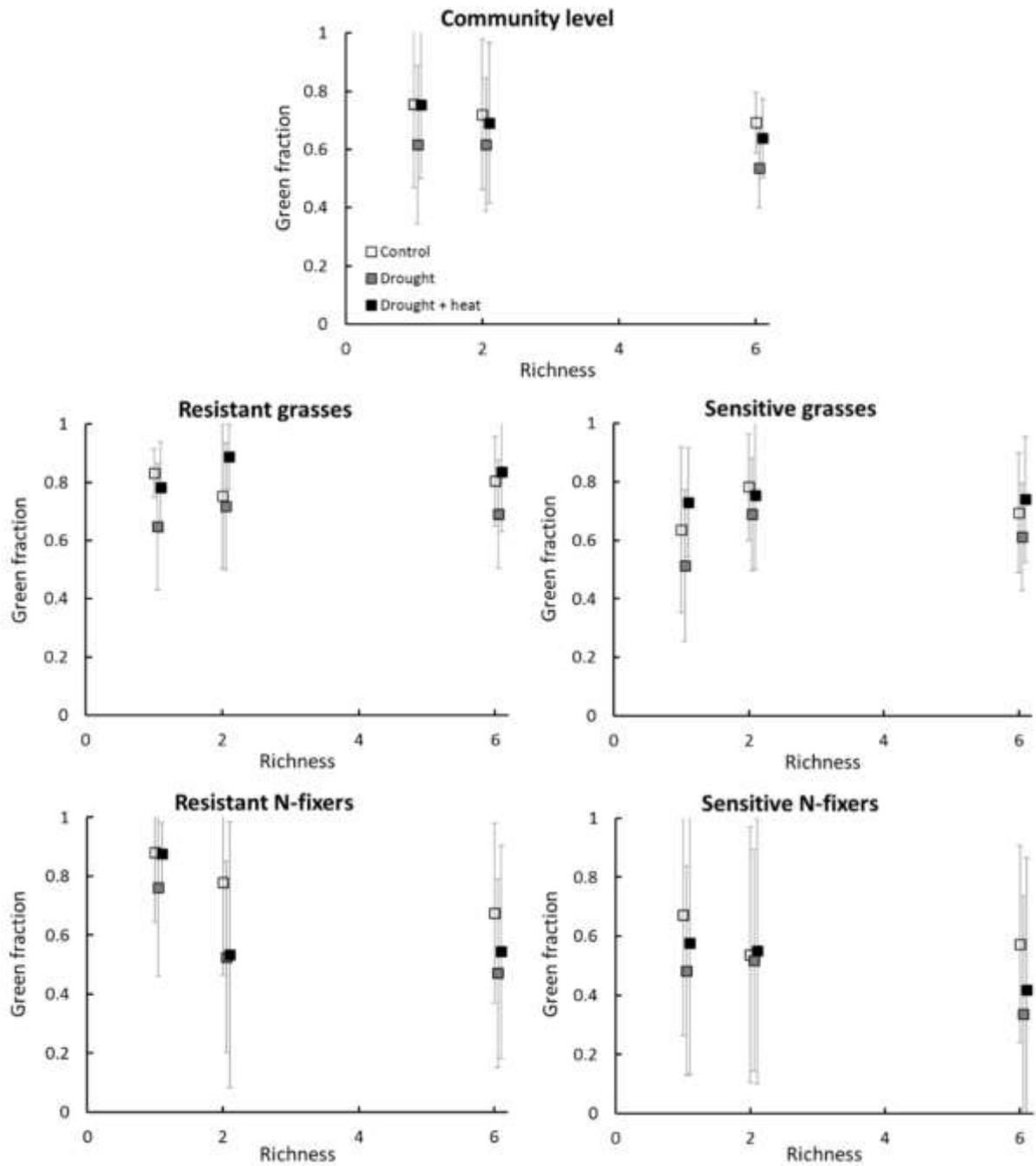


Figure 4: Average green fraction \pm sd at the end of the recovery (DOY 270) versus the communities' species richness in the different climate treatments, on community level and in the different species groups.

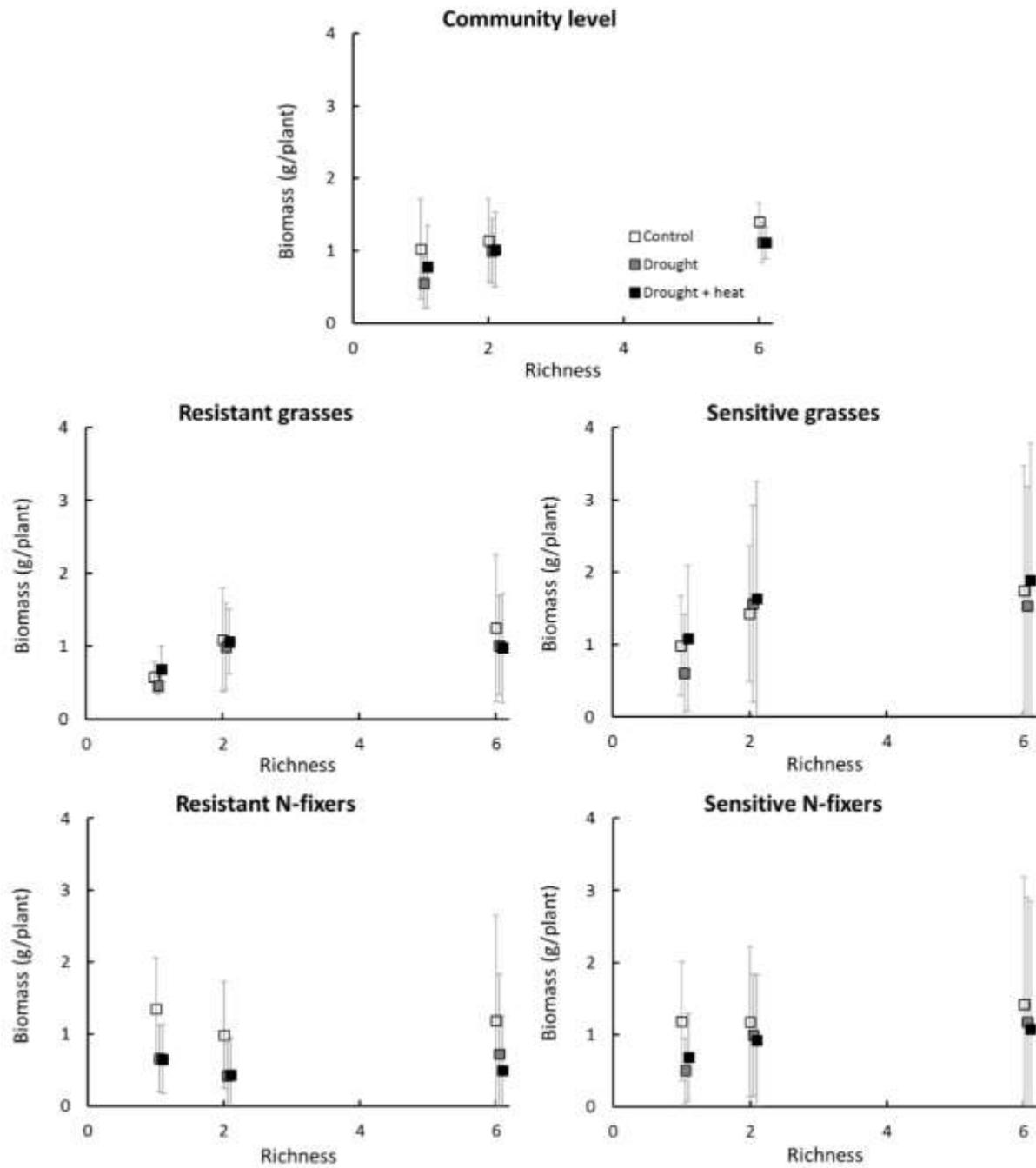


Figure 5: Average biomass production \pm sd per plant at the end of the recovery (DOY 277) versus the communities' species richness in the different climate treatments, on community level and in the different species groups.

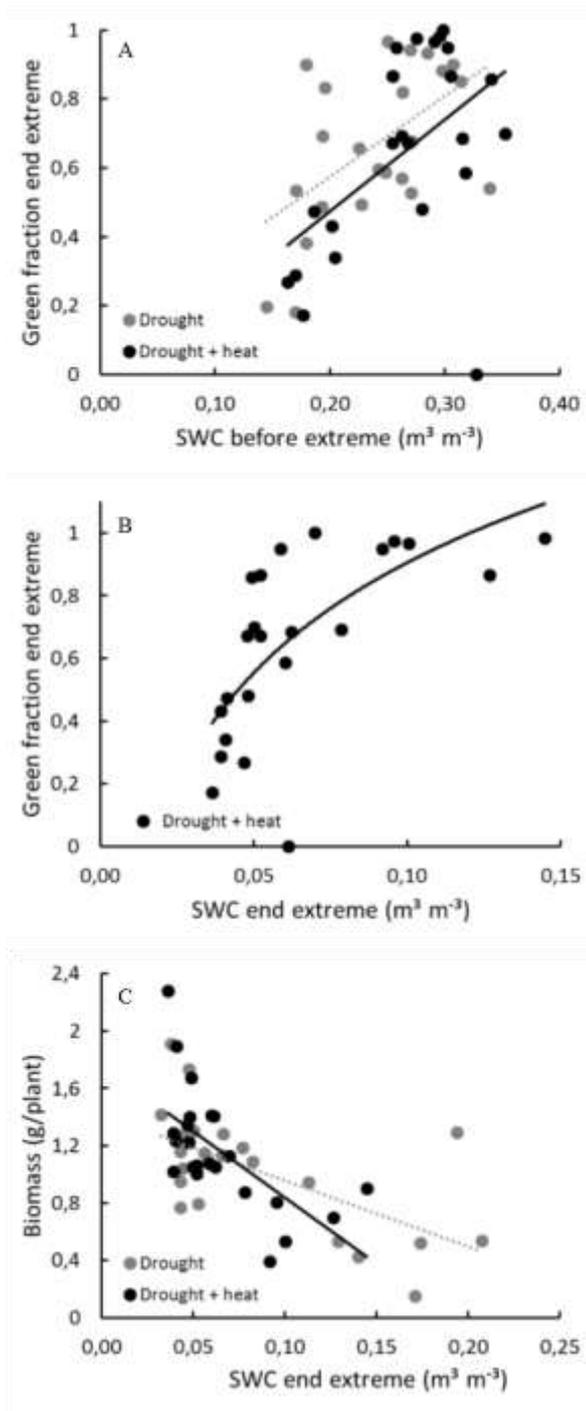


Figure 6: Significant regressions between (A) soil water content (SWC) before (DOY 216) and green fraction at the end (DOY 244) of the extreme events; (B) SWC and green fraction at the end (DOY 244) of the drought + heat treatment (C) SWC at the end of the extreme events (DOY 244) and biomass after recovery (DOY 277). Grey dots and dashed lines indicate drought treatment, black dots and full lines indicate drought with additional heat treatment.

Supplementary files

Supplementary file 1: Arrangement of the different species in the hexagonal grid in S2 and S6 communities. X indicates the absence of a plant, 1-6 are the different species, in the order they can be found in Table 2.

Supplementary file 1: Arrangement of the different species in the hexagonal grid in S2 and S6 communities. X indicates the absence of a plant, 1-6 are the different species, in the order they can be found in Table 2.

S6

		X	3	1	5	4		
		2	4	6	2	1	6	
4	3	5	3	5	3	2		
	1	2	1	6	1	2	4	5
		6	4	2	5	3	5	3
		5	6	4	6	1	6	
			2	3	1	4	X	

S2

		X	1	2	1	2		
		1	2	1	2	1	2	
1	2	1	2	1	2	1		
	2	1	2	1	2	1	2	1
		2	1	2	1	2	1	2
		1	2	1	2	1	2	
			1	2	1	2	X	