

Simulated heat waves affected alpine grassland only in combination with drought

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Summary

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- The Alpine region is warming fast, and concurrently, the frequency and intensity of climate extremes are increasing. It is currently unclear whether alpine ecosystems are sensitive or resistant to such extremes.
- We subjected Swiss alpine grassland communities to heat waves with varying intensity by transplanting monoliths to four different elevations (2440–660 m above sea level) for 17 d. Half of these were regularly irrigated while the other half were deprived of irrigation to additionally induce a drought at each site.
- Heat waves had no significant impacts on fluorescence (F_v/F_m , a stress indicator), senescence and aboveground productivity if irrigation was provided. However, when heat waves coincided with drought, the plants showed clear signs of stress, resulting in vegetation browning and reduced phytomass production. This likely resulted from direct drought effects, but also, as measurements of stomatal conductance and canopy temperatures suggest, from increased high-temperature stress as water scarcity decreased heat mitigation through transpiration.
- The immediate responses to heat waves (with or without droughts) recorded in these alpine grasslands were similar to those observed in the more extensively studied grasslands from temperate climates. Responses following climate extremes may differ in alpine environments, however, because the short growing season likely constrains recovery.

Introduction

The number of experimental climate extreme studies performed on temperate grassland systems has been growing rapidly over the past years (Grime *et al.*, 2008; Bloor & Bardgett, 2012; Vogel *et al.*, 2012). The same is not true for experiments investigating extreme events in alpine grasslands, although these systems are exposed to the observed rise in climate extremes as well (Rebetez, 2004). Moreover, the increase in temperatures has been higher in the Alpine region than the global average (Ceppi *et al.*, 2012). In lowland grasslands, studies have demonstrated that the effect of heat waves, that is relatively short events with temperatures well above the average, tends to be limited as long as water is available to the plants (De Boeck *et al.*, 2011). Impacts of drought on ecosystem functioning have been shown to be variable, depending on the phenological stage (Dreesen *et al.*, 2012), the species numbers and identities involved (Kreyling *et al.*, 2008) and the interaction with other factors such as temperature (Hoepfner & Dukes, 2012; Xu *et al.*, 2014) and atmospheric CO₂ concentrations (Larsen *et al.*, 2011). Generally though, drought seems to

incite more negative responses on ecosystem structure and function than heat.

Information on responses of grasslands in colder biomes to specific climate extremes such as exceptional warmth or drought is sketchy. In polar regions, heat has been documented to reduce cold limitation in some cases (Marchand *et al.*, 2005), although we assume that this effect may not match the situation in alpine systems at lower latitudes. These systems, like those in the arctic, are short-statured and therefore have a high aerodynamic boundary resistance (Körner, 2003), but benefit from higher radiation in summer. This causes alpine grassland to become significantly warmer than one would assume from weather station data (Scherer & Körner, 2010; Neuner & Buchner, 2012) and basic metabolic processes such as photosynthesis are therefore similar to those in low elevation plants (Körner & Diemer, 1987). On the other hand, higher Q₁₀ values for respiration reported for alpine vegetation could imply a higher sensitivity to increased temperatures (Larigauderie & Körner, 1995). Whether a period of exceptionally warm weather would significantly affect alpine grasslands, is therefore an open question. In one of the few

studies that considered the impact of a heat wave on alpine communities, satellite image analysis by Jolly *et al.* (2005) suggested that the 2003 European heat and drought increased photosynthetic activity in the alpine zone. On the other hand, Abeli *et al.* (2012) observed decreased flowering in their alpine grasslands during that same climate anomaly, while vegetative growth was stable, although it is unknown whether the reported phenological response was triggered by higher temperatures or by drier conditions.

The Alpine region has not been regularly exposed to drought (Van der Schrier *et al.*, 2007), but increases in frequency and intensity are predicted for the near and long-term future (Gobiet *et al.*, 2014). Responses of ecosystems in the alpine zone to drought, like those to heat, have barely been documented. One study that focused on alpine grassland in Switzerland found that primary production was reduced already at moderate drought levels (Schmid *et al.*, 2011). Other drought studies have been mainly constrained to lower elevations and have suggested both negative (Gilgen & Buchmann, 2009, aboveground biomass) and little or no response to drought (Brilli *et al.*, 2011, carbon and water fluxes). Using a species distribution model fed by fine-scale data, Engler *et al.* (2011) stressed the importance of precipitation in alpine systems compared with rising temperatures. As drought and heat are naturally coupled through atmospheric feedbacks (De Boeck & Verbeeck, 2011), we specifically include their interaction here.

Here, we tested the impact of a heat wave as a single factor event and in conjunction with a period of drought on alpine grasslands in Switzerland. To be able to detect nonlinear or threshold responses we used a gradient approach (Kreyling *et al.*, 2014), with three levels of heat wave intensity plus the unwarmed (reference) situation, all crossed with drought. We hypothesize that: negative effects of heat on plant growth and functioning would likely only occur at the hot end of the gradient, where the probability is highest that temperature tolerance limits in tissues are exceeded and respiration is increased; drought would result in increased stress and reduced productivity; negative effects would be most apparent when heat and drought co-occur, as warming speeds up drought and drought increases the possibility of heat stress.

Materials and Methods

Location and set-up

We transplanted swards of alpine grassland including its main rooting horizons (monoliths) along an elevation gradient as a means of warming, a method that circumvents some of the technical and logistic issues that render applying large warming events difficult *in situ*. The ALPFOR research station, situated at 2440 m near the Furka pass in the Swiss central Alps (46°34'N 8°25'E), has a gradient of *c.* 2000 m in elevation in its surroundings which made it possible to create significant temperature differences. We selected three lower-elevation sites located at the West side of the pass, namely Oberwald (1390 m above sea level (asl), 46°32'N 8°21'E), Bister (1040 m asl, 46°21'N 8°04'E) and Visp (660 m asl, 46°17'N 7°53'E), creating a 1770 m vertical

gradient. Using an elevation gradient also results in a CO₂ gradient, as partial pressures of gases in the atmosphere decrease with altitude. The pressure difference between the highest and lowest sites would have been *c.* 20% (Körner, 2003). However, any direct CO₂ 'fertilization' effect would be expected to be small as alpine systems have been shown not to be carbon limited (Körner *et al.*, 1997; Inauen *et al.*, 2012).

The grassland at the reference alpine site is extensively grazed by sheep and characterized by relatively deeply weathered soils of partly podzolized alpine brown earth on siliceous bedrock (Inauen *et al.*, 2013). Two slopes (± 150 m apart) with similar orientation (S–E) and vegetation were selected in the immediate vicinity of the research station. On each slope, 24 monoliths were excavated during the first week of July 2013 in a zone of *c.* 4 × 10 m and adjusted with a knife to fit tightly into buckets of 27.5 cm diameter and 21 cm depth, perforated at the bottom. An anti-rooting mat prevented roots from growing outside of the buckets while still allowing drainage. Monoliths were selected on the basis of the joint presence of five common alpine species: the graminoids *Nardus stricta* and *Carex curvula*, which together made up half or more of the vegetation according to cover estimates (not shown), and *Homogyne alpina*, *Potentilla aurea* and *Geum montanum*, which were never dominant. Other species present in many of the monoliths were *Trifolium alpinum* (the only nitrogen fixing species recorded), *Poa alpina* and *Anthoxanthum alpinum*. After excavation, monoliths were grouped based on similarity of cover and then randomly allocated to treatments with equal representation from each of the two excavation zones. We subsequently clipped senesced shoots (but not any living tissues) to avoid erroneous attribution of leaf mortality. This also means that the clipped phytomass (see later) was for the most part produced during the experimental year.

On July 15, that is 3–4 wk after snowmelt, the monoliths were translocated to the three lower sites (12 monoliths remained at the origin site Furka) and placed into 27.5 cm wide, 30 cm deep outer buckets that were dug 20 cm into the soil. Monoliths were placed together in two groups of six on all four sites, always on a flat terrain that was unshaded by trees or buildings. A 1 × 1.5 m roof made of transparent PVC (4 mm thick, 90% visible light transmission) was placed 40 cm above each group of six buckets (plant height was only a few centimetres). One group (*n* = 6) was watered during the treatment period, the other (*n* = 6) was not. Irrigation was equivalent to a rate of 80 mm per month (an average value for July in Ulrichen, 1350 m asl, located between the highest and the lowest sites in our gradient) and applied in four events during the 17-d treatment period (45 mm in total). On 1 August, all buckets were transported back to the reference site (Furka), placed in a common garden and watered (copiously in drought-exposed monoliths, and resupplying subsequent seepage water from the outer buckets). The monoliths were finally reinstalled into the soil on their original locations on 19 August.

Measurements

Micro-climate Measurements of air temperature (T_{air}), relative humidity (RH) and photosynthetic photon flux density (PPFD)

were recorded every minute at each of the four sites, underneath the shelters. Sensors (all Hobo Data Loggers; Onset Computer Corp., Bourne, MA, USA) were placed at 40 cm height, and the T_{air} -RH-sensor was shielded from the sun by a thin wooden panel. Additional meteorological measurements were made by an automatic weather station at the Furka site (equipped with standard sensors) that contributes to the network of the Swiss Federal Office of Meteorology and Climatology (MeteoSwiss). We also received data from the official weather stations located in Ulrichen (4 km from the Oberwald site) and Visp, operated by MeteoSwiss. Wind speed underneath the shelters and outside (at the same height) was checked on several occasions with a pocket wind meter (Kestrel 3000; Nielsen Kellerman, Boothwyn, PA, USA) at each site to quantify wind reduction by the shelter. Soil temperatures (T_{soil}) at 5 cm depth inside the buckets were recorded once at each site in all monoliths (HANNA Instruments, Woonsocket, RI, USA).

Leaf-level responses As the number of measurements was too high to be able to collect readings at all sites on the same day, we opted to pair sites: Furka–Oberwald and Bister–Visp. We made physiological measurements (fluorescence and stomatal conductance (g_s), see further) twice per day: before noon as well as in the afternoon at each of the paired sites. This strategy was used because plant functioning (e.g. stomatal responses) is likely to differ during the day, especially when stress starts affecting the plants. We repeated the procedure each time at the other two paired sites the following day. The statistical analyses take into account that measurements were not collected on the same days at each site by focusing on the trends in time instead. After the monoliths had been brought back to the alpine site and placed in a common garden, measurements were made on two more days (3 and 10 August, i.e. days 19 and 26) to monitor legacy effects immediately after the climate extreme. The weather during the 17-d treatment period was generally stable with sunny and dry conditions on 10 out of 12 measurement days.

Leaf measurements were made on a subset of three of the five species that were present in every community, namely those forbs with the largest leaves (completely filling the leaf chambers): *H. alpina*, *P. aurea* and *G. montanum*. Measurements were made on randomly chosen leaves (in randomly chosen monoliths) of three plants per species per measurement period, located > 2–3 cm from the bucket rim to minimize edge effects. This means that six measurements per species and per measurement day (three before noon and three in the afternoon) were carried out for each treatment at each site. Stomatal conductance was recorded with a Decagon SC-1 porometer (Decagon Devices, Pullman, WA, USA), a device that allows for fast measurements (30 s per reading) with minimal disturbance of the leaf boundary layer. Chlorophyll fluorescence was used as a stress indicator by determining the F_v/F_m , a ratio that is usually $c. 0.8$ under non-stressed conditions across many plant species and ecotypes (Bjorkman & Demmig, 1987). Lower values indicate deactivation of photosystem II as incoming radiation can no longer be optimally accepted and transferred by open reaction centres and needs to be dissipated through fluorescence. Measurements were

made with a Plant Efficiency Analyser (Hansatech Ltd, King's Lynn, UK) after 30 min of dark adaptation to allow the reaction centres to discharge fully. Realized effects, that is those observed under light-adapted conditions, sometimes differ from potential effects measured after dark adaptation, so that our measurements may over- or underestimate the amount of actual stress to an extent. Nevertheless, using dark adapted leaves reduces unwanted effects of fluctuating light conditions between measurements and measurement days, and provides a good compromise between comparability and stress detection (Maxwell & Johnson, 2000).

Ecosystem and community level responses Because leaf measurements such as those mentioned above can be misleading with regards to whole plant functioning (cf. Kull, 2002 on the upscaling of photosynthesis) and because they only consider some of the species present, we also took a number of community scale measurements. Evapotranspiration (ET) was estimated by lysimetry, based on weighing always (0.1 g precision) the same four monoliths per treatment per site (Mettler-Toledo Inc., Greifensee, Switzerland). Because a transportable (battery-operated) scale became available only after 5 d, only monoliths at Furka could be weighed on the first day of the treatment (Precisa IBK 2400D scale, Obrecht, Switzerland). On 2 days (days 10 and 11), monoliths were weighed before sunrise and after sunset at the Furka and Oberwald sites simultaneously to assess the daily evapotranspiration rates more precisely.

The percentage of green cover (between 0 and 100%), used as an indicator of leaf growth, expansion and senescence, was visually estimated on each measurement day for every monolith by the same observer and without looking at previously recorded data to avoid bias. Canopy temperatures were recorded at each site on two occasions with an infrared high resolution camera (TH9260; NEC Avio Ltd, Tokyo, Japan) under sunny conditions. One image of each block of six monoliths (after removing the shelter) was made from a 45° angle facing away from the sun. Emissivity was set at 0.97. Data were processed by manually selecting desired areas (those of the canopy inside the buckets) to derive average canopy temperatures per monolith using irMotion software (Atus GmbH, Hamburg, Germany), that is six average values per treatment. Aboveground plant biomass was collected on 13 August. Standing plant matter (phytomass) was clipped at $c. 2$ cm height, separated per functional group (graminoids, herbs and nitrogen fixers) and into living (green; biomass) and dead phytomass (brown; necromass), dried at 70°C for 2 d and then weighed.

Statistics

All statistical analyses were performed with the R statistical package (version 2.12.1; R Development Core Team, 2010). Two different measures were used as explanatory variables: VPD and T_{air} . In both cases we used the average daytime values across the whole study period (Table 1). Separate models were fitted for each of these variables because the two are correlated ($r=0.95$). We consider vapour pressure deficit (VPD) as the most relevant explanatory variable, as it combines temperature and humidity,

Table 1 Description of the sites where monoliths were transported to during the treatment period (15 July–1 August 2013)

	Site			
	Furka	Oberwald	Bister	Visp
Coordinates	46°34'N 8°25'E	46°32'N 8°21'E	46°21'N 8°04'E	46°17'N 7°53'E
Elevation (m)	2440	1390	1040	660
T_{air} (°C) 24 h	11.9	16.8	21.4	20.9
T_{air} (°C) day	14.1	19.8	24.5	23.8
T_{air} (°C) night	8.1	11.8	15.9	16.0
RH (%) 24 h	77	73	67	75
RH (%) day	72	64	60	68
RH (%) night	87	89	79	87
VPD (kPa) 24 h	0.40	0.75	1.14	0.85
VPD (kPa) day	0.55	1.08	1.56	1.19
PPFD ($\mu\text{mol m}^{-2} \text{s}^{-1}$) day	708	681	727	675

Mean values of 1 min measurements made throughout the treatment period underneath the rainout shelters, at 40 cm height. Vapour pressure deficit (VPD) was determined from air temperature (T_{air}) and relative humidity (RH). Photosynthetic photon flux density (PPFD) is the average of measurements during daylight ($\pm 15 \text{ h d}^{-1}$).

two important parameters in assessing heat and drought effects (De Boeck *et al.*, 2011).

For the data that were collected during the course of the experiment (evapotranspiration, stomatal conductance, fluorescence and green cover), we fitted and evaluated linear mixed models with Gaussian error distributions, using the function 'lmer' (lme4 package; Bates *et al.*, 2010). All response variables except ET were transformed (square root, arcsin or logarithmic) to achieve normal distribution of the residuals. The models included irrigation treatment (irrigation vs no irrigation) as a categorical fixed factor, the average daytime VPD or T_{air} across the study period and time (measuring day) as continuous fixed factors, as well as all interactions. Random effects were fitted for site, monolith nested within site and census (categorical, 1–5 measurements performed on 2 consecutive days). Models explaining physiological measurements (fluorescence and stomatal conductance) further included random effects for species and session (before noon vs afternoon measurements, nested within census). Full models were simplified by removing nonsignificant parameters to obtain minimum adequate models. The significance of fixed factors was assessed by posteriori likelihood ratio tests. Using autocorrelation function plots we checked for autocorrelation in the residuals of the final models, at various time lags, but did not observe any significant temporal autocorrelation.

The analyses for data recorded repeatedly in time during the extreme event were done in three steps: (1) an overall analysis, (2) a separate analysis for the irrigated and nonirrigated treatments in the case of a significant drought \times time interaction in step 1 (this was always the case), with time, VPD or T_{air} and their interaction as fixed factors, and (3) in the case of a significant time \times VPD or T_{air} interaction in step 2, a further analysis of the last time step (days 16 or 17) to study cumulative effects was carried out with only VPD or T_{air} as a fixed factor. Phytomass data were tested using ANOVA for each functional group and for biomass and necromass separately, with VPD or T_{air} as a continuous fixed factor, irrigation treatment as a categorical fixed factor, and monolith (nested within site) as a random factor.

Separate analyses were performed to look for changes after the end of the imposed extreme event (recovery or continuing damage). We calculated the difference between the measurement at the end of the treatments (days 16 or 17) and the last measurement day in this legacy phase. For fluorescence and green cover this was on day 26, for stomatal conductance on day 19 as humid conditions prevented stable calibration on day 26. Differences in stomatal conductance and fluorescence were analysed using linear mixed models, with irrigation and heat as fixed factors, and site, species and session as random factors. The difference in green cover was analysed using ANOVA.

Results

Micro-climate

Averages of T_{air} , RH, calculated vapour pressure deficits (VPD, based on 1-min readings of T_{air} and RH) and PPFD measured underneath the shelters and in nearby meteorological stations are given in Table 1 and Supporting Information Table S1. Daytime temperatures were $> 10^\circ\text{C}$ higher and VPD was more than doubled at the warmest and driest site (Bister) compared with the reference site (Furka). In comparison, the 2003 heat wave during the first 15 d of August at the nearby Güttsch meteorological station (2287 m asl) increased the mean T_{air} by 6.7°C compared with the average from 1954 to 2012 for the same period (15.2°C vs 8.5°C). Surprisingly, the site at the lowest elevation (Visp) was not the hottest or the most water-demanding location. One explanation is that the Visp site was located just inside a side valley, sheltering it from the prevailing East and West oriented winds. The result is that average air temperature differed only little between the two lowest sites, while evaporative forcing (VPD), which combines air temperature and humidity, differed clearly between all sites (Table 1). The soil temperature readings (at 5 cm depth) suggest that nonirrigated monoliths were $2\text{--}3^\circ\text{C}$ warmer than irrigated monoliths during daytime at the lower

elevations, and were $< 1^{\circ}\text{C}$ warmer at the reference site, where drying was least pronounced (Table S2).

Wind speed measurements underneath and outside the shelters suggest only minor differences (Table S3), which implies that effects of sheltering on canopy temperatures would have been small (cf. De Boeck *et al.*, 2012). Other warming effects by the shelters (e.g. by disrupting convection, i.e. trapping heat) also seemed limited. We compared the 24 h before shelters were installed in Visp with the 24 h after (both days with clear skies), which were very similar regarding average air temperature (0.35°C difference) and radiation (1% difference) according to the Meteoswiss weather station. Air temperatures measured at our site at 40 cm height did not differ ($< 0.1^{\circ}\text{C}$) between these two 24-h periods, indicating that shelter-induced increases in T_{air} were likely $< 0.5^{\circ}\text{C}$. By comparing PPFD readings just before and after the shelters were removed at the end of the experiment, we found that PPFD reduction was *c.* 12% at all sites. Differences in PPFD between sites, affected by topography influencing sunrise and sunset times, were limited to 7% at maximum (Table 1), with the warmest site also being the brightest.

Leaf-level responses

Data of F_v/F_m reveal that this ratio in general (i.e. across the treatment period) differed significantly between the irrigated and nonirrigated monoliths, with lower values – indicating increased stress – observed in the latter (Fig. 1). We also uncovered a significant interaction between VPD (site) and irrigation treatment, which, when analysing both irrigation treatments separately, was found to result from the fact that the F_v/F_m values were similar in irrigated monoliths across all sites (no VPD effect) while there was a significant VPD effect in nonirrigated monoliths (Fig. 1; Table S4). The VPD \times time interaction was significant in nonirrigated monoliths, and further analysis from data of the last

treatment day (day 17) revealed a significant influence of VPD on the F_v/F_m ratio (Table S4), reflecting stress levels that increased more in some sites than in others during the course of the experiment (Fig. 1). In general, the analysis shows that higher VPD only led to measurable differences in plant stress if plants were not irrigated (Fig. 1). The same patterns were observed with temperature as an explanatory variable (Table S5).

Regarding stomatal conductance, we found significantly lower values in the nonirrigated than in the irrigated communities (Fig. 2), but as for fluorescence the effect differed with site (VPD \times drought interaction, Table S4). Considering each irrigation treatment separately, we observed a similar response of stomatal conductance over time for all sites when no water was added (significant time effect, but no significant VPD or VPD \times time effects, Fig. 2; Table S4). This trend differed between sites when the monoliths were watered (VPD \times time effect). The lower values at Bister, the hottest and driest of all sites, towards the end of the treatment period may have triggered this VPD \times time interaction. As for fluorescence, the statistical analysis of g_s using temperature instead of VPD as the explanatory variable yielded the same results (Table S5). No change in g_s or F_v/F_m was found immediately after the end of the climate extreme, suggesting that recovery was largely absent.

Ecosystem and community level responses

Across the period where monolith weight could be determined at all sites (days 6–17), evapotranspiration (ET) was more than twice as high in irrigated than in nonirrigated monoliths (3.87 vs 1.73 mm d^{-1} , Fig. 3). When considering each irrigation treatment separately, ET significantly decreased with time at all sites for the drought-exposed monoliths, as indicated by the significant time effect but absence of significant VPD or VPD \times time effects (Table S4). In the irrigated communities neither time nor

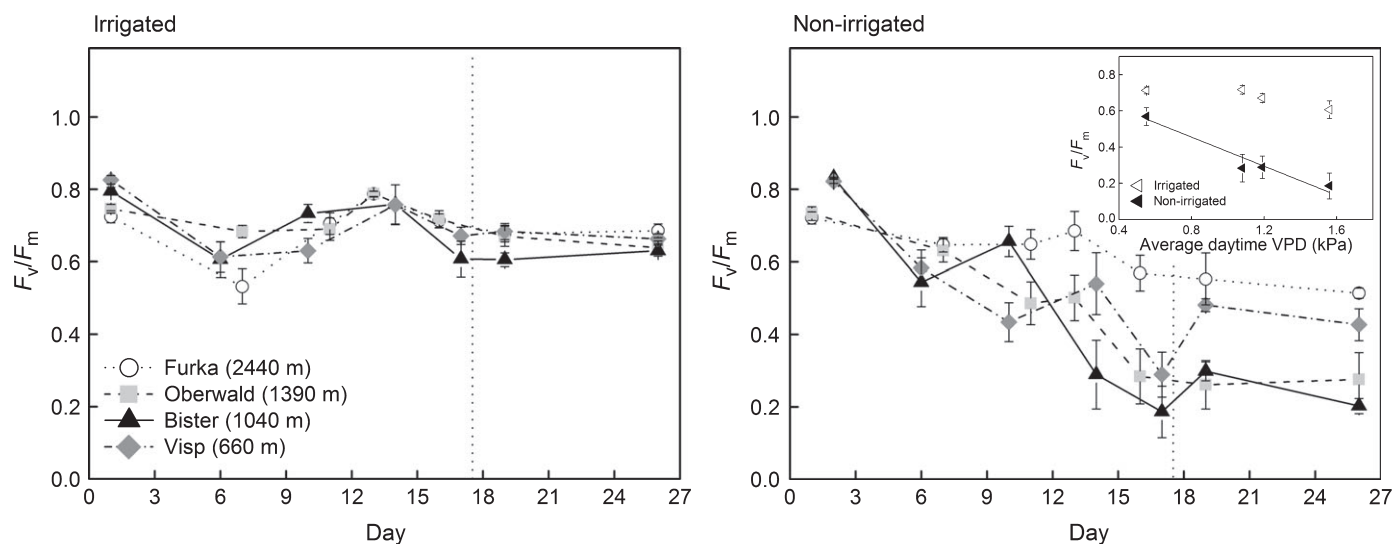


Fig. 1 Maximal efficiency of PSII photochemistry (ratio of F_v/F_m , a stress indicator) during and after the imposed climate extremes (day 1 = 15 July, end of extreme indicated by dotted line) at the four different sites. Measurements made on three dicot species always both before and after noon and data pooled per treatment (mean \pm SE, $n = 18$). Inset graph shows F_v/F_m at the last day of the extreme, in function of average daytime vapour pressure deficit (VPD; during the extreme) of the four sites, with the line depicting a significant trend.

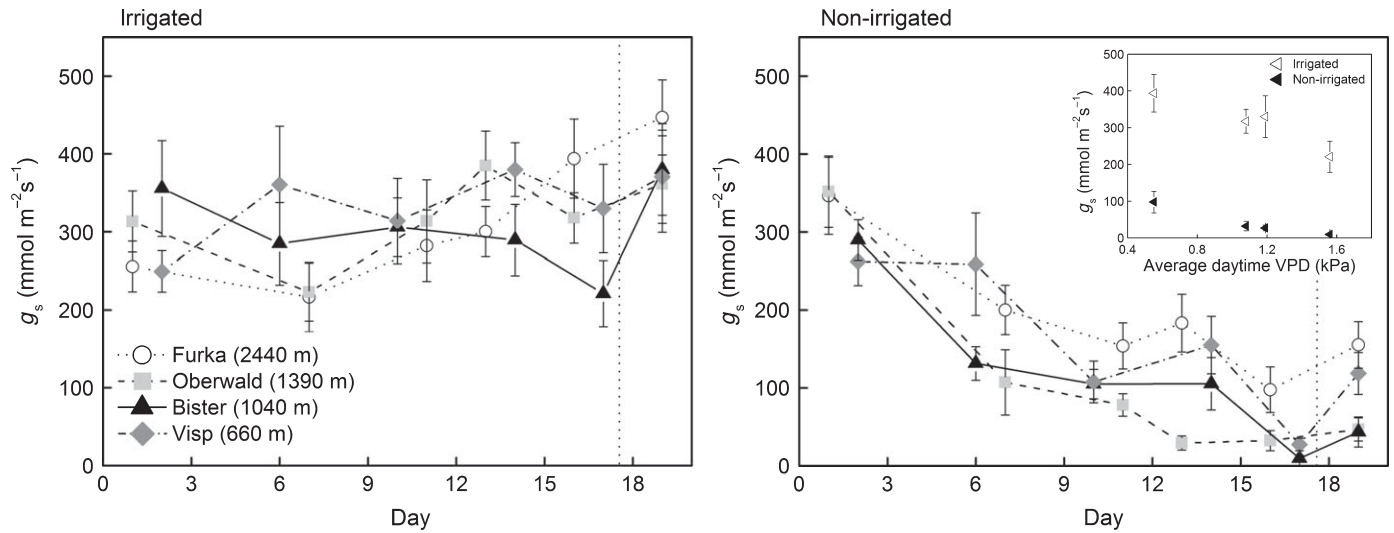


Fig. 2 Ratio of stomatal conductance (g_s) during and after the imposed climate extremes (day 1 = 15 July, end of extreme indicated by dotted line) at the four different sites. Measurements made on three dicot species always both before and after noon and data pooled per treatment (mean \pm SE, $n = 18$). Inset graph shows g_s at the last day of the extreme, in function of average daytime vapour pressure deficit (VPD; during the extreme) of the four sites.

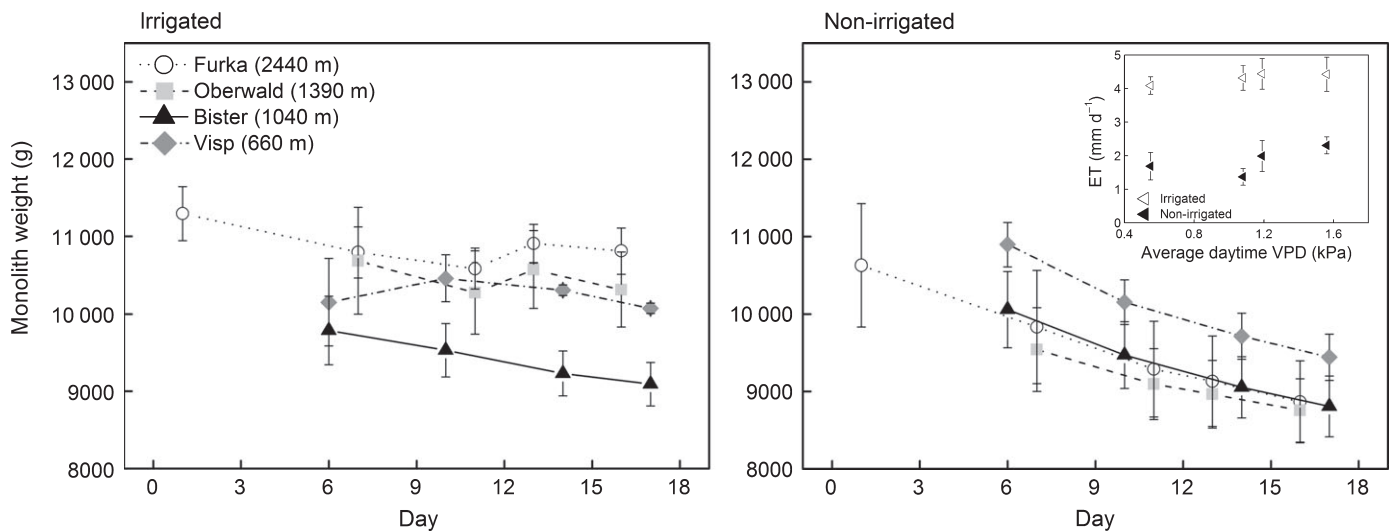


Fig. 3 Monolith weight (mean \pm SE, $n = 4$) during the imposed climate extremes (day 1 = 15 July) at the four different sites. Inset graph shows average evapotranspiration (ET) from days 6 to 17, calculated from weights and irrigation quantities, in function of average daytime vapour pressure deficit (VPD; during the extreme) of the four sites.

VPD was significant. The analyses using temperature as the explanatory variable revealed similar results, with the exception that the overall analysis revealed a significant time \times VPD \times drought interaction, caused by a significant positive relationship between temperature and ET at the last treatment day in the irrigated monoliths only (Table S5). The more detailed assessment of ET during two consecutive days and one night (Table 2), measured simultaneously at the Furka and the Oberwald site, confirmed the significantly lower evapotranspiration in nonirrigated monoliths compared with irrigated monoliths ($P < 0.001$).

The analysis of percentages of green cover broadly confirms visual trends in Fig. 4. The amount of green cover was significantly lower in the monoliths where irrigation was withheld compared with the irrigated monoliths, while the time \times irrigation

treatment interaction suggests different impacts of the irrigation treatments on greenness trends (Table S4; Fig. 4). The response over time seen in irrigated communities was not related to VPD. Raw data suggest a peak in green cover *c.* 1 August in irrigated monoliths (Fig. 4), which corresponds to the normal peak biomass date in these alpine grasslands. Such a peak could not be identified when drought was applied, even at the Furka site, which seems to indicate that drought suppressed normal plant development even under normal temperatures. Specifically, green cover in nonirrigated monoliths varied in time across sites (significant VPD \times time interaction), with differences between the sites growing bigger during the treatment period (significant VPD effect at last measurement day, Table S4). No significant differences between values recorded on the last day of the climate extreme and 9–10 d later were found. Statistical analysis using

Table 2 Evapotranspiration (ET) determined by weighing monoliths ($n = 6$) before and after sunrise on two consecutive days (days 10 and 11 of the treatment) simultaneously at two sites (Furka and Oberwald)

Treatment	Site			
	Furka (2440 m)		Oberwald (1390 m)	
	Irrigated	Nonirrigated	Irrigated	Nonirrigated
ET (mm d^{-1}) \pm SE (day 1)	3.35 \pm 0.29	2.11 \pm 0.55	3.52 \pm 0.16	1.75 \pm 0.32
ET (mm d^{-1}) \pm SE (night)	0.32 \pm 0.02	0.22 \pm 0.05	0.23 \pm 0.03	0.12 \pm 0.03
ET (mm d^{-1}) \pm SE (day 2)	3.38 \pm 0.41	1.80 \pm 0.46	3.19 \pm 0.19	1.47 \pm 0.27

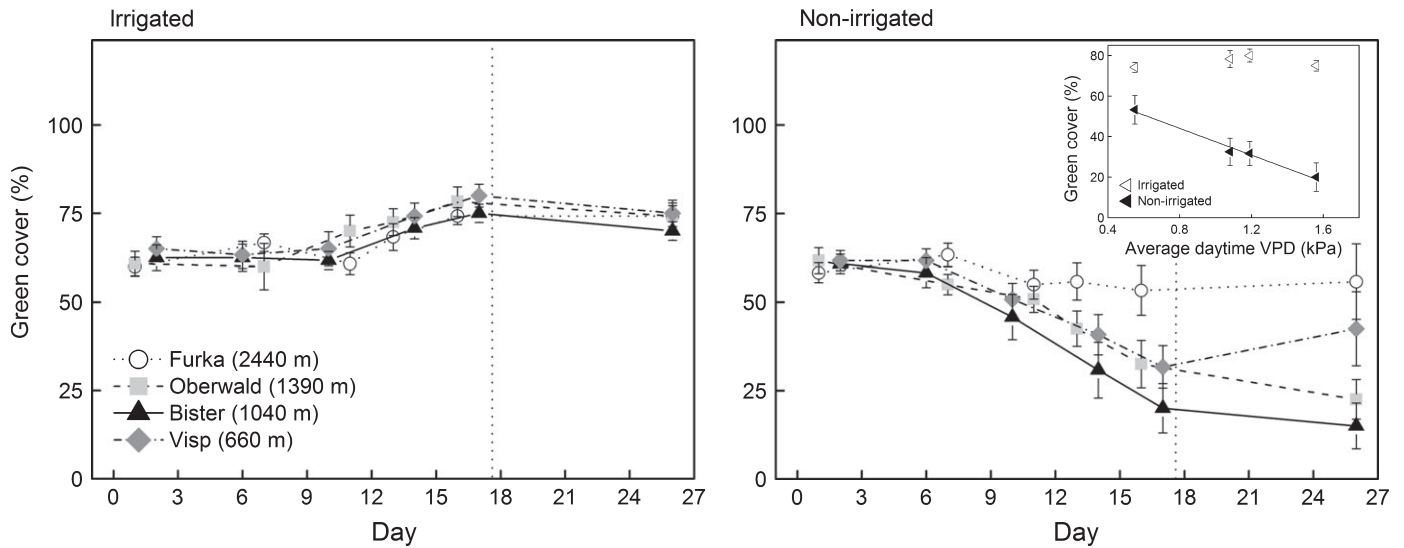


Fig. 4 Visual estimates of green cover (mean \pm SE, $n = 6$) during and after the imposed climate extremes (day 1 = 15 July, end of extreme indicated by dotted line) at the four different sites. Inset graph shows green cover at the last day of the extreme, in function of average daytime vapour pressure deficit (VPD; during the extreme) of the four sites, with the line depicting a significant trend.

temperature instead of VPD as the explanatory variable showed the same results (Table S5).

The importance of drought for community development was confirmed by the analyses of the aboveground phytomass. In general, the phytomass was reduced by 34–49% and the biomass by 40–76% due to drought and any of its indirect effects (Fig. 5, Table S6). Significant effects of drought on biomass production were found for both graminoids and non-nitrogen fixing herbs, while there was a trend towards lower production for nitrogen-fixing (*Trifolium*) species ($P = 0.06$), whose contribution to the overall biomass was marginal (Fig. 5). In irrigated monoliths, no biomass differences between sites were discernible for any of the three functional groups (Table S6). The same was found when the nonirrigated monoliths were analysed separately, with the exception of the nitrogen fixing species (VPD: $P < 0.05$). The amount of necromass (pooled across functional groups because of the small quantities) was significantly higher in nonirrigated communities than in irrigated communities, with some differences between sites (VPD effect) but only a trend ($P = 0.07$) of a VPD \times irrigation treatment interaction (Table S6). The same patterns were found when temperature was used as an explanatory variable (Table S6).

Canopy temperatures were recorded via infrared imaging on two occasions at each site: at days 10 or 11 of the treatment, and at days 16 or 17 (Table 3). Drought led to a clear temperature increase when comparing irrigated and nonirrigated monoliths, which likely stems from the lower values of stomatal conductance observed (Fig. 2). The increase was already high at the lower elevation sites at day 10/11 (+7.5 to 9.2°C), while at that time it was still below 5°C at the Furka site, increasing towards the end of the treatment (+9.1°C). Together with leaf fluorescence and greening data (Figs 1, 4), this suggests that drought stress developed more slowly at the highest (coolest and most humid) site. In irrigated systems, canopy temperatures were above air temperatures at all sites as well (a consequence of the high irradiation), but this relative difference ($c. 6\text{--}8^\circ\text{C}$ at the three lower sites) seemed substantially more pronounced at Furka, the highest and coolest site (12–14°C). This means that inter-site differences in tissue temperatures were likely smaller than would be expected from air temperatures under nondrought conditions.

Discussion

Intuitively, one would expect that exposing plant communities that usually grow under midday air temperatures of 10–15°C to

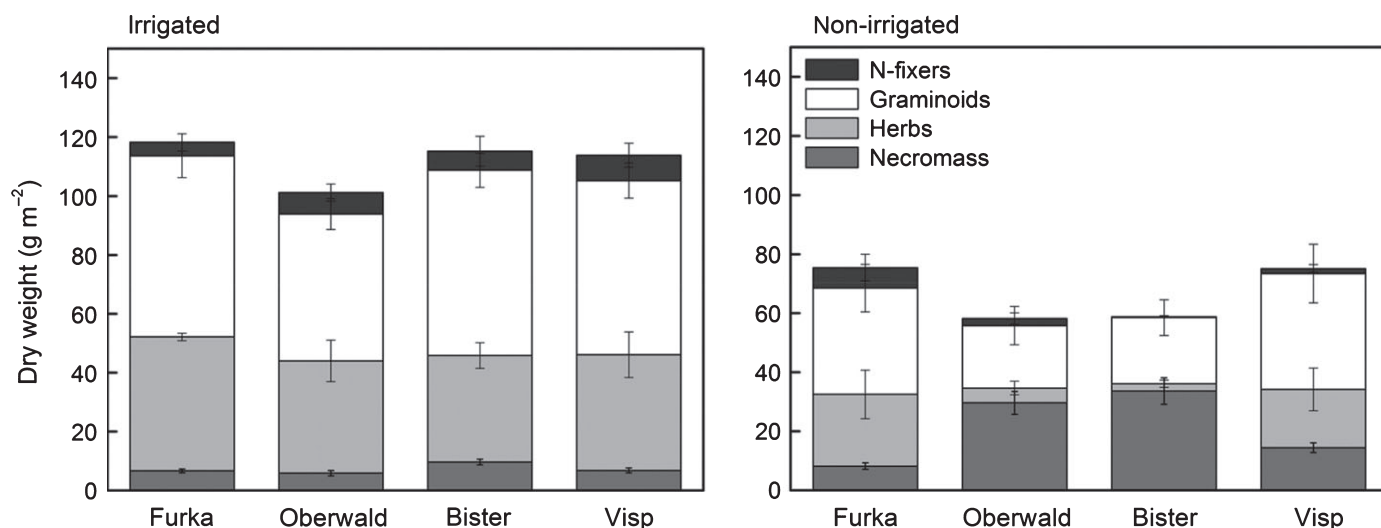


Fig. 5 Aboveground phytomass of all monoliths collected on 13 August (12 d after the end of the climate extreme) and separated per functional group and into living (biomass) and dead (necromass). Error bars depict the standard error (six monoliths per site and per irrigation treatment).

Table 3 Averages of canopy temperatures ($T_{\text{canopy}} \pm \text{SE}$) recorded with an infrared camera at two different dates per site: days 11 and 16 for Furka and Oberwald, days 10 and 17 for Bister and Visp

	Day	Treatment	Site			
			Furka	Oberwald	Bister	Visp
T_{canopy} (°C)	10 or 11	Irrigated	33.8 ± 1.4	38.3 ± 0.9	38.3 ± 0.9	40.7 ± 1.0
T_{canopy} (°C)	10 or 11	Nonirrigated	38.2 ± 2.4	47.5 ± 1.4	45.8 ± 1.7	48.8 ± 2.0
T_{air} (°C)	10 or 11	Both	21.5	30.5	33.6	34.0
PPFD ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	10 or 11	Both	1869	1445	2151	2139
VPD (kPa)	10 or 11	Both	1.08	2.74	3.29	3.03
T_{canopy} (°C)	16 or 17	Irrigated	32.3 ± 0.7	32.5 ± 0.6	42.7 ± 0.8	38.3 ± 1.2
T_{canopy} (°C)	16 or 17	Nonirrigated	41.4 ± 1.0	43.0 ± 0.7	52.0 ± 1.0	48.2 ± 2.2
T_{air} (°C)	16 or 17	Both	18.2	25.3	34.1	31.5
PPFD ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	16 or 17	Both	1650	1924	1877	1806
VPD (kPa)	16 or 17	Both	0.82	1.93	3.24	2.43

Air temperature ($T_{\text{air}} \pm \text{SE}$), photosynthetic photon flux density (PPFD) and vapour pressure deficit (VPD) recorded at the time (average of ten 1 min readings) of T_{canopy} measurements is also given (at 40 cm height). Temperatures partly reflect dried (dead) leaves and minor fractions of bare soil related to stress-induced plant senescence.

temperatures that occasionally reached 35°C would cause them harm. However, our data showed that, as long as the soil did not dry out, the direct effects of temperature increases on fluorescence (a stress indicator), canopy greenness and aboveground production were insignificant. None of our measurements indicated gradual changes with increasing temperature, which suggests that short-term warming without concomitant drought affects these alpine grasslands via thresholds (which were not exceeded here) rather than progressively. It is important to note that differences between canopy and air temperatures were more pronounced at the highest site (12–14°C) than at the lowest three sites (6–8°C), meaning that canopy temperatures differed less between the highest and lowest sites than air temperatures (cf. Cernusca & Seiber, 1981). Higher heat dissipation through evapotranspiration at all the lower sites, triggered by substantially higher atmospheric water demand compared with the reference site may be the reason (cf. Van den Bergh *et al.*, 2013). Such a mitigating effect on

tissue temperatures has been demonstrated for temperate systems (De Boeck *et al.*, 2011), but depends on the availability of soil water reserves.

This also highlights the notion that air temperatures can be misleading (Körner, 2003; Scherrer & Körner, 2010; De Boeck *et al.*, 2012) and that actual tissue temperatures should be used to judge whether excessive heat occurs in plants. Tissue temperatures also explain why the imposed heat wave did not alleviate any growth limitation by cool temperatures. The abovementioned large differences between canopy and air temperatures at the highest site (Furka) led plant tissues to be warmed above 25°C and even above 30°C during daytime. The possibility of a heat wave mitigating any low-temperature related growth restrictions, as observed by Marchand *et al.* (2005) in tundra ecosystems and suggested in the warming study by Cavieres & Sierra-Almeida (2012) in the Andes, therefore seems unlikely here and was not supported by any changes in aboveground dry matter

production. Also, unlike in Marchand *et al.* (2005), fluorescence measurements after the imposed heat wave did not reveal any increase in stress resulting from the return to cooler conditions.

Alpine grasslands thus seem stable in the face of heat waves varying in intensity, but this no longer proved true when heat coincided with drought. In that case, aboveground production decreased and the proportion of necromass to biomass increased, with two measurements directly related to stress, fluorescence and greenness of the vegetation, changing linearly with VPD (Figs 1, 4), suggesting that drought effects get progressively worse when the air gets warmer and drier. This is likely caused by both direct and indirect effects of drought. Direct effects were demonstrated by drought impacts being significant also without warming, at the reference site. The fact that a heat wave, regardless of intensity, had little directly measurable effects on plant functioning as a single factor, whereas drought did, is similar to findings for lowland grassland species (De Boeck *et al.*, 2011; Poirier *et al.*, 2012; Hoover *et al.*, 2014). Earlier studies on drought in alpine systems were not conclusive, with for example, Wieser *et al.* (2008) suggesting that even during dry years the importance of water stress in grassland ecosystems in the Austrian Alps was limited, whereas others did observe negative single factor drought effects (Gilgen & Buchmann, 2009; Schmid *et al.*, 2011). Common direct effects include lower photosynthesis, growth reduction as meristems become less active, decreased leaf expansion, and tissue and plant mortality. Reductions in photosynthesis can occur either through direct limitation of CO₂ through closure of stomates (Cornic, 2000; Chaves *et al.*, 2002) or by metabolic constraints (Flexas *et al.*, 2008), with the second process likely more important when drought stress is intense. Yet, photosynthesis as such was probably not a key factor in propagating negative drought effects due to the low sensitivity to carbon of these alpine systems (Körner *et al.*, 1997; Inauen *et al.*, 2012). Growth reduction, earlier senescence and tissue mortality was evident from greenness and aboveground phytomass data.

The observation that drought impacts became progressively stronger with higher temperatures and atmospheric water demand, likely relates to more than merely an increase in drought stress. One important indirect effect of intense drought is a higher probability of high-temperature stress as heat mitigation through transpiration fails when water reserves are depleted (De Boeck *et al.*, 2011). Also in the current study, drought increased the likelihood of tissue temperatures reaching damaging levels (cf. Neuner & Buchner, 2012; Hasanuzzaman *et al.*, 2013). Indeed, above 40–45°C surface temperatures were almost exclusively recorded in the nonirrigated communities. Photosystems are thought to be fairly heat-sensitive (Larcher, 2003), and the functioning of photosystem II decreased as the drought progressed and associated tissue temperatures increased. This loss of function could not be attributed specifically to direct drought effects or indirect ones such as overheating, however, as fluorescence is a universal stress indicator. Another potential indirect effect of drought is nutrient limitation as nutrients become immobilized in the dry soil (e.g. Joslin *et al.*, 2000). The role of nutrient starvation in the current experiment may not have been substantial, however. The period in which nutrient transport may have been

severely obstructed was likely limited, as data of stomatal conductance (Fig. 2) and evapotranspiration data (Fig. 3) demonstrate that water was still being cycled at significant rates at least until half of the 17-d treatment period.

The interplay between heat and drought also led to different responses between functional groups. At Oberwald, the graminoid proportion of total aboveground biomass in droughted vs irrigated monoliths was 74% vs 52%, at Bister 89% vs 60% and at Visp 65% vs 55%. This suggests that the broadleaved species present in our systems suffered more than graminoids from the combination of drought and heat. Their anatomy (broader leaves with an approximate horizontal orientation) could have caused excess heat stress because they capture more midday radiation and dissipate the heat more slowly than graminoids (which have narrow leaves with a more vertical orientation, such as *N. stricta* and *C. curvula*). Resistance did not differ for single-factor droughts (i.e. at the reference site), with graminoids making up 54% and 55% of (living) aboveground biomass in nonirrigated and irrigated monoliths, respectively. This contrasts with the findings of a drought study by Gilgen and Buchmann (2009) at a subalpine grassland site, who observed that grasses suffered most from drought.

Our data consistently indicate that the heat wave impacts in alpine grasslands, much like in temperate grasslands, are likely to be limited unless coinciding with drought. The importance of changes in moisture compared with those in air temperature also in an alpine environment had been hinted upon by Fu *et al.* (2013), who found that the warming effects on the aboveground productivity on the Northern Tibetan Plateau depended largely on soil drying. The importance of soil moisture in systems with a short growing season has furthermore been stressed by Le Roux *et al.* (2013), who observed that fine-scale variation in soil moisture was strongly related to species occurrence patterns. Low elevation studies show that on longer time scales, a single drought event (with associated high temperatures) can be the instigator of long-lasting vegetation change (Kreyling *et al.*, 2011). In a long-term observation study, Stampfli and Zeiter (2004) found that relative cover of the major growth forms present in their study area, graminoids and forbs, changed more in the period following extreme drought than at other times. Changes in recruitment may be key as gaps opened by mortality after the extreme event generate opportunities for colonization (cf. Vittoz *et al.*, 2009), especially if propagule rain is changed in response to a climate extreme, as was observed by Ertl (2013) in alpine areas. Recruitment by seed currently plays a minor role in the system studied, since most species grow clonally and some have been shown to be thousands of years old (De Witte *et al.*, 2012). Like for drought, legacy effects of short but intense warm episodes may be relevant, even if the alpine systems in our study seemed to be very resistant in the short term. Processes that could be affected are altered allocation to storage organs (affecting next year's growth) or to a lesser extent effects on recruitment from seed (cf. Liu *et al.*, 2012). Moreover, ongoing increases in mean temperatures can affect growing season length (Jolly *et al.*, 2005), plant–animal interactions (Liu *et al.*, 2011; Wu *et al.*, 2011) and other factors that change the competitive balance between species. The timing

of warm spells could also be an important factor in alpine environments (Volk *et al.*, 2014).

Like observations made by Scherrer and Körner (2010), our findings suggest that future research into temperature effects in cold biomes should take into account that air and tissue temperatures may deviate substantially, and that the latter parameter is of most significance when trying to determine whether plants may be directly affected by heat or low temperature. Some of the drought levels we exposed the grasslands to are probably very extreme as soil drying would have been accelerated by decoupling the systems from full soil conditions. However, experiments that extend beyond common ranges of environmental conditions can be highly informative (Kayler *et al.*, 2015), and here, our observations clearly suggest that high temperatures have much more potential for immediate, significant effects on plant functioning in alpine grasslands when water is limiting. As we noted, this is similar to earlier observations in temperate grasslands. In contrast to such grasslands, however, where resilience is an important ecosystem trait (Brilli *et al.*, 2011; Dreesen *et al.*, 2014), short-term regrowth in systems where the growing season only lasts 2–3 months and where life strategy is centred on persistence rather than vigour, is inherently constrained, as was confirmed by our observations. A more complete assessment of the sensitivity of alpine systems to climate change will require future studies to take into account both direct impacts and longer term legacy effects of increasingly intense climatic extremes.

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References

- Abeli T, Rossi G, Gentili R, Gandini M, Mondoni A, Cristofanelli P. 2012. Effect of the extreme summer heat waves on isolated populations of two orophitic plants in the north Apennines (Italy). *Nordic Journal of Botany* 30: 109–115.
- Bates D, Maechler M, Bolker B. 2010. *lme4: linear mixed-effects models using Eigen and Eigen++*. [WWW document] URL <http://www.r-project.org/web/packages/lme4/index.html> [accessed 20 April 2014].
- Bjorkman O, Demmig B. 1987. Photon yield of O₂ evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. *Planta* 170: 489–504.
- Bloor JMG, Bardgett RD. 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. *Perspectives in Plant Ecology, Evolution and Systematics* 14: 193–204.
- Brilli F, Hörtnagl L, Hammerle A, Haslwanter A, Hansel A, Loreto F, Wohlfahrt G. 2011. Leaf and ecosystem response to soil water availability in mountain grasslands. *Agricultural and Forest Meteorology* 151: 1731–1740.
- Cavieres LA, Sierra-Almeida A. 2012. Facilitative interactions do not wane with warming at high elevations in the Andes. *Oecologia* 170: 575–584.
- Ceppi P, Scherrer SC, Fischer AM, Appenzeller C. 2012. Revisiting Swiss temperature trends 1959–2008. *International Journal of Climatology* 32: 203–213.
- Cernusca A, Seeber MC. 1981. Canopy structure, microclimate and the energy budget in different alpine plant communities. In: Grace J, Ford ED, Jarvis PG, eds. *Plants and their atmospheric environment*. Oxford, UK: Blackwell, 75–81.
- Chaves MM, Pereira JS, Maroco J, Rodrigues ML, Ricardo CP, Osório ML, Carvalho I, Faria T, Pinheiro C. 2002. How plants cope with water stress in the field. Photosynthesis and growth. *Annals of Botany* 89: 907–916.
- Cornic G. 2000. Drought stress inhibits photosynthesis by decreasing stomatal aperture – not by affecting ATP synthesis. *Trends in Plant Science* 5: 187–188.
- De Boeck HJ, De Groot T, Nijs I. 2012. Leaf temperatures in glasshouses and open-top chambers. *New Phytologist* 194: 1155–1164.
- De Boeck HJ, Dreesen FE, Janssens IA, Nijs I. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. *New Phytologist* 189: 806–817.
- De Boeck HJ, Verbeeck H. 2011. Drought-associated changes in climate and their relevance for ecosystem experiments and models. *Biogeosciences* 8: 1121–1130.
- De Witte LC, Armbruster GF, Gielly L, Taberlet P, Stocklin J. 2012. AFLP markers reveal high clonal diversity and extreme longevity in four key arctic-alpine species. *Molecular Ecology* 21: 1081–1097.
- Dreesen FE, De Boeck HJ, Janssens IA, Nijs I. 2012. Summer heat and drought extremes trigger unexpected changes in productivity of a temperate annual/biannual plant community. *Environmental and Experimental Botany* 79: 21–30.
- Dreesen FE, De Boeck HJ, Janssens IA, Nijs I. 2014. Do successive climate extremes weaken the resistance of plant communities? An experimental study using plant assemblages. *Biogeosciences* 11: 109–121.
- Engler R, Randin CF, Thuiller W, Dullinger S, Zimmermann NE, Araujo MB, Pearman PB, Le Lay G, Piedallu C, Albert CH *et al.* 2011. 21st century climate change threatens mountain flora unequally across Europe. *Global Change Biology* 17: 2330–2341.
- Ertl S. 2013. Positive effects of an extremely hot summer on propagule rain in upper alpine to subnival habitats of the Central Eastern Alps. *Plant Ecology & Diversity* 6: 467–474.
- Flexas J, Ribas-Carbó M, Diaz-Espejo A, Galmés J, Medrano H. 2008. Mesophyll conductance to CO₂: current knowledge and future prospects. *Plant, Cell & Environment* 31: 602–621.
- Fu G, Zhang X, Zhang Y, Shi P, Li Y, Zhou Y, Yang P, Shen Z. 2013. Experimental warming does not enhance gross primary production and above-ground biomass in the alpine meadow of Tibet. *Journal of Applied Remote Sensing* 7: 073505.
- Gilgen AK, Buchmann N. 2009. Response of temperate grasslands at different altitudes to simulated summer drought differed but scaled with annual precipitation. *Biogeosciences* 6: 2525–2539.
- Gobiet A, Kotlarski S, Beniston M, Heinrich G, Rajczak J, Stoffel M. 2014. 21st century climate change in the European Alps – a review. *Science of the Total Environment* 493: 1138–1151.
- Grime JP, Fridley JD, Askew AP, Thompson K, Stevens CJ, Bennett CR. 2008. Long-term resistance to simulated climate change in an infertile grassland. *Proceedings of the National Academy of Science, USA* 105: 10028–10032.
- Hasanuzzaman M, Nahar K, Alam MM, Roychowdhury R, Fujita M. 2013. Physiological, biochemical, and molecular mechanisms of heat stress tolerance in plants. *International Journal of Molecular Sciences* 15: 9643–9684.
- Hoeppe SS, Dukes JS. 2012. Interactive responses of old-field plant growth and composition to warming and precipitation. *Global Change Biology* 18: 1754–1768.
- Hoover DL, Knapp AK, Smith MD. 2014. Resistance and resilience of a grassland ecosystem to climate extremes. *Ecology* 95: 2646–2656.
- Inauen N, Körner C, Hiltbrunner E. 2012. No growth stimulation by CO₂ enrichment in alpine glacier forefield plants. *Global Change Biology* 18: 985–999.
- Inauen N, Körner C, Hiltbrunner E. 2013. Hydrological consequences of declining land use and elevated CO₂ in alpine grassland. *Journal of Ecology* 101: 86–96.
- Jolly WM, Dobbertin M, Zimmermann NE, Reichstein M. 2005. Divergent vegetation growth responses to the 2003 heat wave in the Swiss Alps. *Geophysical Research Letters* 32: L18409.

- Joslin JD, Wolfe MH, Hanson PJ. 2000. Effects of altered water regimes on forest root systems. *New Phytologist* 147: 117–129.
- Kayler ZE, De Boeck HJ, Faticchi S, Grünzweig JM, Merbold L, Beier C, McDowell N, Dukes JS. 2015. Experiments to confront the environmental extremes of climate change. *Frontiers in Ecology and the Environment* 13: 219–225.
- Körner C. 2003. *Alpine plant life, 2nd edn*. Heidelberg, Germany: Springer.
- Körner C, Diemer M. 1987. In situ photosynthetic responses to light, temperature and carbon dioxide in herbaceous plants from low and high altitude. *Functional Ecology* 1: 179–194.
- Körner C, Diemer M, Schättli B, Niklaus P, Arnone J. 1997. The responses of alpine grassland to four seasons of CO₂-enrichment: a synthesis. *Acta Oecologica* 18: 165–175.
- Kreyling J, Jentsch A, Beier C. 2014. Beyond realism in climate change experiments: gradient approaches identify thresholds and tipping points. *Ecology Letters* 17: 125–e1.
- Kreyling J, Jentsch A, Beierkuhnlein C. 2011. Stochastic trajectories of succession initiated by extreme climatic events. *Ecology Letters* 14: 758–764.
- 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.
- Kull O. 2002. Acclimation of photosynthesis in canopies: models and limitations. *Oecologia* 133: 267–279.
- Larcher W. 2003. *Physiological plant ecology, 4th edn*. Berlin, Germany: Springer-Verlag.
- Larigauderie A, Körner C. 1995. Acclimation of leaf dark respiration to temperature in alpine and lowland plant species. *Annals of Botany* 76: 245–252.
- Larsen KS, Andresen LC, Beier C, Jonasson S, Albert KR, Ambus P, Andersen KS, Arndal MF, Carter MS, Christensen S *et al.* 2011. Reduced N cycling in response to elevated CO₂ warming and drought in a Danish heathland: synthesizing results of the CLIMAITE project after two years of treatments. *Global Change Biology* 17: 1884–1899.
- Le Roux PC, Aalto J, Luoto M. 2013. Soil moisture's underestimated role in climate change impact modelling in low-energy systems. *Global Change Biology* 19: 2965–2975.
- Liu Y, Mu J, Niklas KJ, Li G, Sun S. 2012. Global warming reduces plant reproductive output for temperate multi-influenced species on the Tibetan plateau. *New Phytologist* 195: 427–436.
- Liu Y, Reich PB, Li G, Sun SC. 2011. Shifting phenology and abundance under experimental warming alters trophic relationships and plant reproductive capacity. *Ecology* 92: 1201–1207.
- Marchand FL, Mertens S, Kockelbergh F, Beyens L, Nijs I. 2005. Performance of High Arctic tundra plants improved during but deteriorated after exposure to a simulated extreme temperature event. *Global Change Biology* 11: 2078–2089.
- Maxwell K, Johnson GN. 2000. Chlorophyll Fluorescence: a practical guide. *Journal of Experimental Botany* 51: 659–668.
- Neuner G, Buchner O. 2012. Dynamics of tissue heat tolerance and thermotolerance of PS II in alpine plants. In: Lütz C, ed. *Plants in alpine regions: cell physiology of adaptation and survival strategies*. Vienna, Austria: Springer-Verlag, 61–74.
- Poirier M, Durand JL, Voltaire F. 2012. Persistence and production of perennial grasses under water deficits and extreme temperatures: importance of intraspecific vs. interspecific variability. *Global Change Biology* 18: 3632–3646.
- R Development Core Team. 2010. *R: a language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing.
- Rebetez M. 2004. Summer 2003 maximum and minimum daily temperatures over a 3300 m altitudinal range in the Alps. *Climate Research* 27: 45–50.
- Scherer D, Körner C. 2010. Infra-red thermometry of alpine landscapes challenges climatic warming projections. *Global Change Biology* 16: 2602–2613.
- Schmid S, Hiltbrunner E, Spehn EM, Lüscher A, Scherer-Lorenzen M. 2011. Impact of experimentally induced summer drought on biomass production in alpine grassland. In: Pötsch EM, Krautzer B, Hopkins A, eds. *Grassland farming and land management systems in mountainous regions*. Gumpenstein, Austria: Proceedings of the 16th Symposium of the European Grassland Federation, 214–216.
- Stampfli A, Zeiter M. 2004. Plant regeneration directs changes in grassland composition after extreme drought: a 13-year study in southern Switzerland. *Journal of Ecology* 92: 568–576.
- Van den Bergh T, Inauen N, Hiltbrunner E, Körner C. 2013. Climate and plant cover co-determine the elevational reduction in evapotranspiration in the Swiss Alps. *Journal of Hydrology* 500: 75–83.
- Van der Schrier G, Efthymiadis D, Briffa KR, Jones PD. 2007. European Alpine moisture variability for 1800–2003. *International Journal of Climatology* 27: 415–427.
- Vittoz P, Randin C, Dutoit A, Bonnet F, Hegg O. 2009. Low impact of climate change on subalpine grasslands in the Swiss Northern Alps. *Global Change Biology* 15: 209–220.
- 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.
- Volk M, Wolff V, Bassin S, Ammann C, Fuhrer J. 2014. High tolerance of subalpine grassland to long-term ozone exposure is independent of N input and climatic drivers. *Environmental Pollution* 189: 161–168.
- Wieser G, Hammerle A, Wohlfahrt G. 2008. The water balance of grassland ecosystems in the Austrian Alps. *Arctic, Antarctic, and Alpine Research* 40: 439–445.
- Wu XJ, Duffy E, Reich PB, Sun S. 2011. A brown-world cascade in the dung decomposer food web of an alpine meadow: effects of predator interactions and warming. *Ecological Monographs* 81: 313–328.
- Xu ZZ, Shimizu H, Ito S, Yagasaki Y, Zou CJ, Zhou G, Zheng Y. 2014. Effects of elevated CO₂, warming and precipitation change on plant growth, photosynthesis and peroxidation in dominant species from North China grassland. *Planta* 239: 421–435.

Supporting Information

Additional supporting information may be found in the online version of this article.

Table S1 Meteorological measurements collected at stations near the sites used in our experiment

Table S2 Soil temperatures recorded in each monolith at the end of the imposed climate extreme

Table S3 Wind speed measured consecutively underneath and outside of the shelters

Table S4 Parameter estimates and significances in mixed-effects models using vapour pressure deficit as the explanatory variable characterizing different sites

Table S5 Parameter estimates and significances in mixed-effects models using air temperature as the explanatory variable characterizing different sites

Table S6 Effects of heat, drought and their interaction on biomass and on necromass

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