DISSERTATION

ECOLOGICAL RESPONSES TO CLIMATE EXTREMES IN A MESIC GRASSLAND

Submitted by

David Lewis Hoover

Graduate Degree Program in Ecology

In partial fulfillment of the requirements

For the Degree of Doctor of Philosophy

Colorado State University

Fort Collins, Colorado

Spring 2014

Doctoral Committee:

    Advisor: Alan K. Knapp

    Melinda D. Smith
    William L. Bauerle
    Joseph C. von Fischer

ABSTRACT

ECOLOGICAL RESPONSES TO CLIMATE EXTREMES IN A MESIC GRASSLAND

Climate change threatens ecosystems through altered climate means and by increasing the frequency and intensity of extreme climatic events. Such events may have greater impacts on ecosystems than shifting means alone because they can push organisms beyond critical thresholds. Thus, there is an urgent need to assess the response of ecosystems to climate extremes as well as elucidate the mechanisms underlying the observed responses. My dissertation examined the ecological impacts of two years of experimentally imposed climate extremes (heat waves and drought) followed by a recovery year, on a mesic tallgrass prairie grassland ecosystem. The broad objectives of this research were (1) to assess the resistance and resilience of this ecosystem to the individual and combined effects of heat waves and drought, and (2) to identify the ecological mechanisms driving the responses and (3) to evaluate the sensitivies of key carbon cycling process to heat waves and drought. I measured a range of biotic responses to these treatments including: ecophysiology, community dynamics, primary production, and soil respiration in order to gain a comprehensive understanding how this ecosystem responds to such extremes.

During the first year of the experiment, I examined the ecophysiological and productivity responses of the dominant C_4 grasses to a growing season-long drought and a midsummer, two-week heat wave. Although differential sensitivities were apparent, the independent effects of drought dominated the ecological responses for both species, with only minor direct effects of heat were observed. However, the heat wave treatments had indirect effects via enhanced soil

drying, making it difficult to separate the effects of the heat wave and precipitation treatments on biotic responses. Therefore in the second year of the experiment, I controlled for heat-induced water losses during the heat wave and examined the independent effects of heat on net photosynthesis in both grass species under contrasting soil moisture regimes. Under low soil moisture, heat had no effect on net photosynthesis, while increasing temperatures moderately reduced photosynthesis under high soil moisture. Next I examined the resistance and resilience in ecosystem function (aboveground primary production) of this tallgrass prairie to the two years of extreme treatments and for one subsequent recovery year. I observed high resistance to heat but not drought, as aboveground production dropped below historic levels during the second year of the drought. Despite this extreme ecological response, productivity fully recovered in just one year post-drought due to rapid demographic compensation by the dominant grass offsetting the loss of the dominant forb. Finally, I examined the response of soil respiration to heat and drought across the three years of the experiment. As with aboveground net primary production, soil respiration was more sensitive to drought than heat, but it was less sensitive overall to drought than production.

There are three main conclusions from my dissertation research. First, this tallgrass prairie ecosystem has low resistance but high resilience to extreme short-term drought, which may be an important characteristic for long-term stability in ecosystems with histories of drought. Secondly, the two most abundant species governed both community and ecosystem-level dynamics across this three-year experiment, providing evidence for the central role of dominant species during these short-term events. Finally, my results suggest that three key carbon cycling processes in this mesic grassland – photosynthesis, plant productivity and soil respiration – are all significantly more sensitive to the independent effects of an extreme drought

than heat waves and there were little to no combined effects of heat waves and drought. Overall, these results suggest that in a future with more frequent and extreme heat waves and drought, this mesic grassland will be most vulnerable to water stress, either directly through precipitation deficits or indirectly through warming-induced drying, while the direct ecological effects of midsummer heat waves will be minor.

ACKNOWLEDGMENTS

TABLE OF CONTENTS

Chapter 1

INTRODUCTION

Background

Climate change threatens ecosystems worldwide through alterations in both climate means (e.g. warming temperatures), and by increasing the frequency and intensity of climate extremes, such as heat waves and drought (Easterling et al. 2000, IPPC 2007, Smith 2011, IPCC 2012). Observed increases in summer temperature extremes (Hansen et al. 2012), heavier rainfall events (Trenberth et al. 2003) and droughts (Dai 2012), suggest that extreme events are already impacting ecosystems. Indeed, recent events such as the 2003 European heat wave and the 2012 US drought, highlight the extensive environmental impacts of extreme events (Ciais et al. 2005, Lal et al. 2012). Assessing their ecological effects remains a key challenge for ecologists, because extreme events will likely cause more dramatic impacts on ecosystem structure and function over the next century, rather than subtle shifts in climate means. This is because climate extremes may push organisms beyond critical physiological thresholds (Gutschick and BassiriRad 2003).

Ecological responses to extreme weather events can range from negligible impacts (Van Peer et al. 2004, Kreyling et al. 2008, Jentsch et al. 2011) to major changes in ecosystem structure and function (Weaver 1954, White et al. 2000b, Haddad et al. 2002, Ciais et al. 2005, Breshears et al. 2005). The later should occur if the magnitude, duration and/or timing of the climate extreme exceed the resistance and resilience of a given ecosystem. Therefore it is important to examine climate extremes from both the driver (climate event) and the response (ecological effects) perspectives. Smith (2011) defined an extreme climatic event as "an episode or occurrence in which a statistically rare or unusual climatic period alters ecosystem structure

1

and/or function well outside the bounds of what is considered typical or normal variability."

Extreme ecological responses are predicted to occur if an extreme climate driver exceeds the

physiological tolerance of individual species, and those effects cascade across multiple

hierarchical levels – from the individual to the ecosystems - impacting ecosystem structure and

function (Smith 2011). Given that most ecosystems are dominated by a few species, which

control ecosystem function (Whittaker 1965, Grime 1998, Hillebrand et al. 2008), extreme

ecological responses are likely if the extreme events trigger significant reductions in fitness and

or mortality of the dominant species. Changes in the abundance of the dominant species may

lead to community-level responses such as community reordering of species abundances, species

loss or invasion with such changes in community structure leading to prolonged recovery or

event state changes (Smith 2011).

Much of our knowledge about the ecological impacts of climate extremes is from

observing naturally occurring events, however the rarity, unpredictability and co-occurrence of

multiple extremes limit our understanding. For example, heat waves and drought are often

concurrent events in nature (De Boeck et al. 2010), and given that both impose water stress on

plants, it is difficult to separate the independent and interactive effects these two climate drivers.

Therefore, experimental manipulations are best suited to assess the ecological responses to

climate extreme and identify underlying mechanisms because climate drivers can be isolated and

attributed to ecological responses. Furthermore, experiments explicitly linked to historical

climatic records and interpreted in the context of long-term ecological data provide the best

opportunity to advance our mechanistic understanding of climate extremes, and thus better

predict their ecological consequences (Smith 2011, Reyer et al. 2013).

The impacts of extreme events on individual ecosystems may extend to the regional and global scale through interactions with the carbon cycle. Over the past few decades, the terrestrial biosphere has been an important carbon sink, absorbing a significant portion of anthropogenically produced CO_2, and thus partially mitigating climate change (Houghton 2007, Pan et al. 2011). However, extreme events have the potential to temporarily reduce the sink capacity of certain ecosystems, even turning them into a carbon source (Reichstein et al. 2013). For example, the 2003 European heat wave reduced both gross primary production and respiration with a net effect of producing a strong source of CO_2 to the atmosphere and reversing four years of carbon sequestration (Ciais et al. 2005). There can also be prolonged effects of such events (Reichstein et al. 2013), as seen by Arnone III et al. (2008), where soil respiration was elevated one year following an anomalously warm year in a tallgrass prairie ecosystems resulting in prolonged suppression of CO_2 uptake. Therefore it is important that the sensitivities of key carbon cycling processes such as photosynthesis, respiration and plant production, are examined in ecosystems that are important to global carbon cycles and are predicted to experience more climate extremes.

Grasslands are one of the largest biomes in the world, covering ~ 40% of the terrestrial biosphere, and providing key ecosystems services including herbivore forage, biofuels, and CO_2 sequestration (White et al. 2000a, Gibson 2009). However, these ecosystem services are at risk due to predicted increases in the frequency and intensity of heat waves and drought (IPCC 2012). This is particularly relevant to regional and global carbon cycles, because the total carbon storage of grasslands is comparable to forests (Gibson 2009), and extremes such as drought can reduce the capacity of these ecosystems to sequester carbon or become temporary carbon sources to the atmosphere (Zhang et al. 2010; Shi et al. 2013) with potential feedbacks to global

warming. Therefore it is critical that we examine how these important ecosystems will respond to climate extremes. In addition, grasslands are model systems to examine the effects of climate change because climate inputs can be easily manipulated, responses are dynamic and ecological data can be collected across a wide hierarchical range.

Dissertation overview

My dissertation research examined the ecological effects of climate extremes on a mesic tallgrass prairie ecosystem. I conducted an experiment that imposed two years of growing-season drought and midsummer heat waves over intact, native prairie and then measured one year of recovery. I collected data on a range of biotic response variables – from the leaf to the ecosystem level – to gain a comprehensive understanding of the ecological responses to such extremes. There were three objectives for my dissertation. First, I assessed the resistance and resilience of the tallgrass prairie ecosystem to the independent and combined effects of heat waves and drought. To date, few studies have examined the response of this ecosystem to extreme heat and drought and such information is critical to predict ecosystem responses to a future with more frequent and intense periods of climate extremes. Secondly and more broadly, I aimed to improve our theoretical understanding of how extreme events shape ecosystem structure and function by using the tallgrass prairie as a model ecosystem to examine the mechanisms underlying ecological responses. Finally, I evaluated the sensitivities of three key carbon cycling processes (photosynthesis, aboveground net primary productivity and soil respiration) to the heat waves and drought in order to identify which processes are most vulnerable in a future with more frequent and intense climate extremes.

Study system and experimental approach

This research was conducted in northeastern Kansas at the Konza Prairie Biological Station, which is part of the Long Term Ecological Research (LTER) network. The station is located within the Flint Hills region, the largest continuous tract of unplowed tallgrass prairie remaining in North America (Knapp et al. 1998). The tallgrass prairie lies along the eastern edge of the Great Plains and is the most mesic grassland type in this ecoregion. The climate is characterized as temperature mid-continental with high interannual variability in precipitation (Knapp et al. 1998). Konza receives about 835 mm of precipitation annually, mostly during the growing season, and has mean annual temperature of 13 °C (Knapp et al. 1998). While hot and dry periods during the summer are not uncommon in this region, there are notable periods in the historical record of extreme heat and drought, such as the 1930's Dust bowl and the early 50's (Woodhouse and Overpeck 1998, Burnette et al. 2010, Burnette and Stahle 2012). These events had devastating impacts on the grasslands with large reductions in productivity, loss of species, changes in community structure and prolonged recovery (Weaver 1954).

This tallgrass prairie ecosystem is characterized by high dominance, with several C_4 grasses providing the majority of aboveground net primary productivity (ANPP) and controlling ecosystem function, while hundreds of C_3 forbs constitute the high biodiversity (Knapp et al. 1998). My dissertation focused on three species, two grasses (*Andropogon gerardii* and *Sorghastrum nutans*) and one forb (*Solidago canadensis*), which together account for about 90% of the ANPP in this lowland community. As previously mentioned, extreme ecological responses are predicted if climate extremes exceeded thresholds for dominant species (Smith 2011). Therefore it was critical to examine the physiological, demographic, community and productivity

responses of these three species in order to identify mechanisms behind any changes in ecosystem structure and function in response to the extreme heat waves and drought.

My dissertation research consisted of a three-year experiment, with each chapter focused on a different set of ecological response variables. The Climate Extremes Experiment was a full factorial split-plot design with two levels of precipitation and four levels of heat (Fig. 1.1). During the first two years (2010 and 2011), I imposed precipitation and heat wave treatments to examine the ecological resistance of the tallgrass prairie to the independent and combined effects of heat and water stress. Precipitation treatments were imposed during the growing season using either passive rainout shelters that removed 66% of ambient rainfall (drought) or net-covered shelters that received all ambient rainfall plus supplemental irrigation (control; Fig. 1.2). The heat wave treatments occurred late July/early August and consisted of passive warming chambers with additional thermal inputs from infrared heat lamps to create four distinct temperature levels (Fig. 1.3). Then in 2012, no treatments were imposed in order to assess ecosystem resilience.

Summary of chapters

This dissertation consists of four complementary chapters, each of which focused on a different aspect of the Climate Extremes Experiment. In Chapters 2 and 3, I examined the responses of the two dominant grasses, *A. gerardii* and *S. nutans* to heat waves and drought. Although treated as functional equivalents, these two species have purported differential sensitivities to heat and water stress (Weaver and Fitzpatrick 1932, Brown 1993, Silletti and Knapp 2002, Swemmer et al. 2006, Nippert et al. 2009). But, no studies have compared how these two species respond to climate extremes, which may have important consequences for

ecosystem structure and function. In Chapter 2, I focused on the first year of the experiment

(2010), and contrasted the sensitivities of the two dominant grasses to heat waves and drought

using ecophysiological and productivity data. To mimic natural conditions (De Boeck et al.

2010), we did not control for heat-induced water loss from control plots during the simulated

heat wave during 2010. As a result, there were strong effects of heat on soil moisture as well as

interactions between heat and precipitation treatments. This indirect effect of heat on soil drying

has been observed in several other studies (Milbau et al. 2005, Marchand et al. 2006, Reichstein

and Ciais 2007, Arnone III et al. 2008, Boeck and Dreesen 2011), which makes it difficult to

separate the independent effects of the two treatments. Therefore, in Chapter 3, this interacting

factor was eliminated by adding water to offset water lost from the heating in the control

treatments in 2011 (while still keeping drought plots dry). This enabled me to examine the

effects of heat on photosynthesis in the dominant grasses under contrasting soil moisture

regimes. In Chapter 4, I used the tallgrass prairie as a model system to test Smith's (2011)

extreme climatic event theoretical framework and hypothesized mechanisms. For this chapter I

assessed resistance and resilience of ecosystem function (ANPP) during two years of extremes,

and then one year post-extreme. Then I used community and demographic data from the three

years to identify mechanisms driving the ecosystem-level dynamics. Finally, in Chapter 5, I

explored the sensitivity of soil respiration to heat and drought across all three years and

compared the responses with those of ANPP. The sensitivities of these two key carbon cycling

processes to such climate extremes will govern the capacity of this ecosystem to sequester

carbon, and therefore may feedback on climate change.

Figures
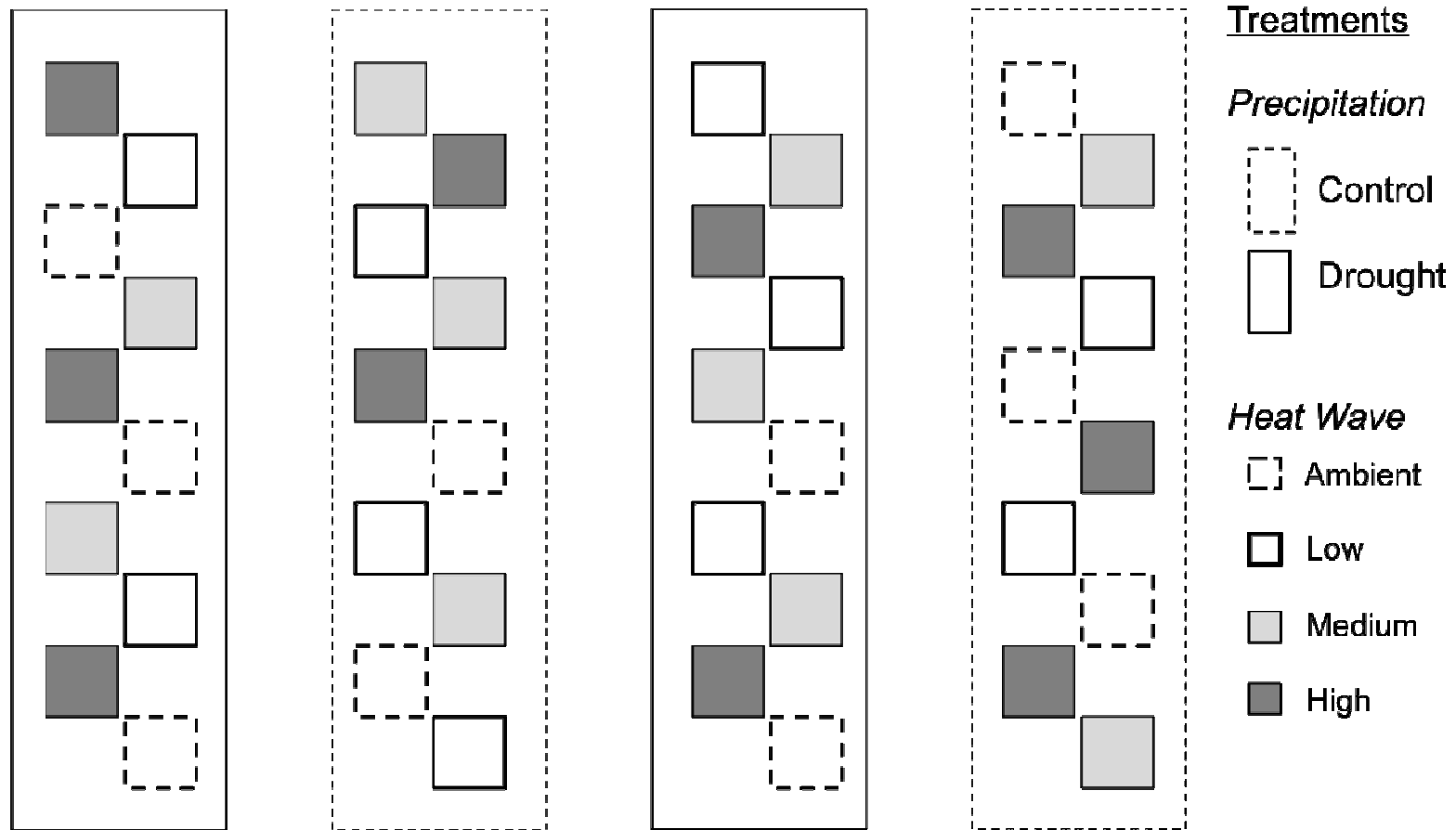
Figure 1.1 Plot layout for the Climate Extremes Experiment. The experiment was a split-plot design with four heat wave treatments nested within two precipitation treatments (n=5 for each precipitation x heat combination). During 2010 and 2011, precipitation treatments were imposed during the growing seasons (April – August) and the heat treatments were applied for two weeks in July.

Figure 1.2 The Climate Extremes Experiment, established in 2010 at the Konza Prairie Biological Station over an intact, native, annually burned lowland tallgrass prairie community. During the 2010 and 2011 growing seasons, four large shelters were used to impose two treatments: control (ambient rainfall plus supplemental irrigation – shelters with netting) and drought (partial roofs on shelters reduced ambient rainfall by 66%). Nested within the rainfall shelters, heat wave treatments were imposed using passive heat chambers combined with infrared lamps during two-weeks mid-summer. All treatments were removed during the 2012 recovery year.

Figure 1.3 Close-up of a heat chamber (2 x 2 x 1.5 m) with a yellow meter stick in the foreground for reference. Infrared lamps were placed within passive warming cambers, which were placed above and slightly into the canopy. However, a gap below the camber and along the edges at the top allowed for air circulation. Different combinations of 2000 W infrared lamp inputs were used to achieve four treatment levels for the heat wave: *ambient* = no lamp (partial heat chamber, not shown), *low* = one lamp at ½ power (+250 W/m^2), *medium* = one lamp at full power (+500 W/m^2), and *high* = two lamps at full power (+1000 W/m^2).

References

Arnone, J. A., P. S. J. Verburg, D. W. Johnson, J. D. Larsen, R. L. Jasoni, A. J. Lucchesi, C. M. Batts, C. von Nagy, W. G. Coulombe, D. E. Schorran, P. E. Buck, B. H. Braswell, J. S. Coleman, R. A. Sherry, L. L. Wallace, Y. Luo, and D. S. Schimel. 2008. Prolonged suppression of ecosystem carbon dioxide uptake after an anomalously warm year. Nature 455:383–6.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. Global Change Biology 16:1992–2000.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. The New phytologist 189:806–17.

Breshears, D. D., N. S. Cobb, P. M. Rich, K. P. Price, C. D. Allen, R. G. Balice, W. H. Romme, J. H. Kastens, M. L. Floyd, J. Belnap, J. J. Anderson, O. B. Myers, and C. W. Meyer. 2005. Regional vegetation die-off in response to global-change-type drought. Proceedings of the National Academy of Sciences of the United States of America 102:15144–15148.

Brown, D. A. 1993. Early Nineteenth-Century Grasslands of the Midcontinent Plains. Annals of the Association of American Geographers 83:589–612.

Burnette, D. J., and D. W. Stahle. 2012. Historical perspective on the dust bowl drought in the central United States. Climatic Change 116:479–494.

Burnette, D. J., D. W. Stahle, and C. J. Mock. 2010. Daily-Mean Temperature Reconstructed for Kansas from Early Instrumental and Modern Observations. Journal of Climate 23:1308–1333.

Ciais, P., M. Reichstein, N. Viovy, and A. Granier. 2005. Europe-wide reduction in primary productivity caused by the heat and drought in 2003. Nature 437:529–533.

Dai, A. 2012. Increasing drought under global warming in observations and models. Nature Climate Change 3:52–58.

Easterling, D. R., G. A. Meehl, C. Parmesan, S. A. Changnon, T. R. Karl, and L. O. Mearns. 2000. Climate extremes: observations, modeling, and impacts. Science 289:2068–2074.

Gibson, D. J. 2009. Grasses and Grassland Ecology. Oxford University Press, Inc. New York.

Grime, J. 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. Journal of Ecology 86:902–910.

Gutschick, V. P., and H. BassiriRad. 2003. Extreme events as shaping physiology, ecology, and evolution of plants: toward a unified definition and evaluation of their consequences. New Phytologist 160:21–42.

Haddad, N. M., D. Tilman, and M. H. Knops. 2002. Long-term oscillations in grassland productivity induced by drought. Ecology letters 5:110–120.

Hansen, J., M. Sato, and R. Ruedy. 2012. Perception of climate change. Proceedings of the National Academy of Sciences of the United States of America 109:1–9.

Hillebrand, H., D. Bennett, and M. Cadotte. 2008. Consequences of dominance: A review of evenness effects on local and regional ecosystem processes. Ecology 89:1510–1520.

Houghton, R. A. 2007. Balancing the Global Carbon Budget. Annual Review of Earth and Planetary Sciences 35:313–347.

IPCC. 2012. Managing the Risks of Extreme Events and Disasters to Advance Climate Change Adaptation. (C. Field, V. Barros, T. Stocker, D. Qin, D. Dokken, K. Ebi, M. Mastrandrea, K. Mach, G. Plattner, S. Allen, M. Tignor, and M. PM, Eds.) A Special Report of Working Groups I and II of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, England.

Jentsch, A., J. Kreyling, and C. Beierkuhnlein. 2007. A new generation of climate-change experiments: events, not trends. Frontiers in Ecology and the Environment 5:365–374.

Jentsch, A., J. Kreyling, M. Elmer, E. Gellesch, B. Glaser, K. Grant, R. Hein, M. Lara, H. Mirzae, S. Nadler, L. Nagy, D. Otieno, K. Pritsch, U. Rasher, M. Schadler, M. Schloter, B. Singh, J. Stadler, J. Walter, C. Wellstein, J. Wollecke, and C. Beierkuhnlein. 2011. Climate extremes initiate ecosystem‐regulating functions while maintaining productivity. Journal of Ecology 99:689–702.

Knapp, A. K., J. M. Briggs, D. C. Hartnett, and S. L. Collins. 1998. Grassland Dynamics: Long-Term Ecological Reserach in Tallgrass Prairie. Oxford University Press, Inc., New York.

Kreyling, J., M. Wenigmann, C. Beierkuhnlein, and A. Jentsch. 2008. Effects of Extreme Weather Events on Plant Productivity and Tissue Die-Back are Modified by Community Composition. Ecosystems 11:752–763.

Lal, R., J. Delgado, and J. Gulliford. 2012. Adapting agriculture to drought and extreme events. Journal of Soil and Water Conservation 67:162–166.

Marchand, F. L., M. Verlinden, F. Kockelbergh, and B. J. Graae. 2006. Disentangling effects of an experimentally imposed extreme temperature event and naturally associated. Functional Ecology 20:917–928.

Milbau, A., L. Scheerlinck, D. Reheul, B. De Cauwer, and I. Nijs. 2005. Ecophysiological and morphological parameters related to survival in grass species exposed to an extreme climatic event. Physiologia Plantarum 125:500–512.

Nippert, J. B., P. A. Fay, J. D. Carlisle, A. K. Knapp, and M. D. Smith. 2009. Ecophysiological responses of two dominant grasses to altered temperature and precipitation regimes. Acta Oecologica 35:400–408.

Pan, Y., R. A. Birdsey, J. Fang, R. Houghton, P. E. Kauppi, W. a. Kurz, O. L. Phillips, A. Shvidenko, S. L. Lewis, J. G. Canadell, P. Ciais, R. B. Jackson, S. W. Pacala, a. D. McGuire, S. Piao, A. Rautiainen, S. Sitch, and D. Hayes. 2011. A Large and Persistent Carbon Sink in the World's Forests. Science 333:988–993.

Van Peer, L., I. Nijs, D. Reheul, and B. De Cauwer. 2004. Species richness and susceptibility to heat and drought extremes in synthesized grassland ecosystems: compositional vs physiological effects. Functional Ecology 18:769–778.

Reichstein, M., M. Bahn, P. Ciais, D. Frank, M. D. Mahecha, S. I. Seneviratne, J. Zscheischler, C. Beer, N. Buchmann, D. C. Frank, D. Papale, A. Rammig, P. Smith, K. Thonicke, M. van der Velde, S. Vicca, A. Walz, and M. Wattenbach. 2013. Climate extremes and the carbon cycle. Nature 500:287–295.

Reichstein, M., and P. Ciais. 2007. Reduction of ecosystem productivity and respiration during the European summer 2003 climate anomaly□: a joint flux tower , remote sensing and modelling analysis. Global Change Biology 13:634–651.

Reyer, C. P. O., S. Leuzinger, A. Rammig, A. Wolf, R. P. Bartholomeus, A. Bonfante, F. de Lorenzi, M. Dury, P. Gloning, R. Abou Jaoudé, T. Klein, T. M. Kuster, M. Martins, G. Niedrist, M. Riccardi, G. Wohlfahrt, P. de Angelis, G. de Dato, L. François, A. Menzel, and M. Pereira. 2013. A plant's perspective of extremes: terrestrial plant responses to changing climatic variability. Global change biology 19:75–89.

Shi, Z., M. L. Thomey, W. Mowll, M. Litvak, N. A. Brunsell, S. L. Collins, W. T. Pockman, M. D. Smith, A. K. Knapp, and Y. Lou. 2013. Differential effects of extreme drought and respiration: Synthesis and modeling analysis. Biogeosciences Open Discu:1–39.

Silletti, A., and A. Knapp. 2002. Long-term responses of the grassland co-dominants Andropogon gerardii and Sorghastrum nutans to changes in climate and management. Plant Ecology 163:15–22.

Smith, M. D. 2011. An ecological perspective on extreme climatic events: a synthetic definition and framework to guide future research. Journal of Ecology 99:656–663.

Swemmer, A., A. Knapp, and M. Smith. 2006. Growth responses of two dominant C4 grass species to altered water availability. International Journal of Plant Sciences 167:1001–1010.

Trenberth, K. E., A. Dai, R. M. Rasmussen, and D. B. Parsons. 2003. The Changing Character of Precipitation.

Weaver, J. 1954. North American prairie. Johnsen Publishing Company, Lincoln, Nebraska.

Weaver, J. E., and T. J. Fitzpatrick. 1932. Ecology and Relative Importance of the Dominants of Tall-Grass Prairie. Botanical Gazette 93:113.

White, R., S. Murray, and M. Rohweder. 2000a. Pilot Analysis of Global Ecosystems: Grasslands Ecosystems.

White, T. A., B. D. Campbell, P. D. Kemp, and C. L. Hunt. 2000b. Sensitivity of three grassland communities to simulated extreme temperature and rainfall events. Global Change Biology 6:671–684.

Whittaker, R. H. 1965. Dominance and diversity in land plant communities: Numerical relations of species express the importance of competition in community function and evolution. Science 147:250–260.

Woodhouse, C., and J. Overpeck. 1998. 2000 years of drought variability in the central United States. Bulletin of the American Meteorological Society 79:2693–2714.

Zhang, L., B. K. Wylie, L. Ji, T. G. Gilmanov, and L. L. Tieszen. 2010. Climate-Driven Interannual Variability in Net Ecosystem Exchange in the Northern Great Plains Grasslands. Rangeland Ecology & Management 63:40–50.

Chapter 2

CONTRASTING SENSITIVITES OF TWO DOMINANT C$_4$ GRASSES TO HEAT WAVES AND DROUGHT

Overview

Heat waves and droughts are predicted to increase in frequency and intensity with climate change. However, we lack a mechanistic understanding of the independent and interactive effects of severe heat and water stress for most ecosystems. In a mesic tallgrass prairie ecosystem, we used a factorial experimental approach to assess ecophysiological and productivity responses of two dominant C$_4$ grasses, *Andropogon gerardii* and *Sorghastrum nutans*, to a season-long drought and a mid-summer heat wave at four intensities. We hypothesized that drought would have greater impacts than heat waves, that combined effects would be greater than either factor alone, and that the dominant grasses would differ in their responses to heat and water heat stress. We detected significant reductions in photosynthesis, leaf water potential and productivity with drought in both species, but few direct responses to the heat waves. Surprisingly, there was no additive effect of heat and water stress on any plant response. However, *S. nutans* was more sensitive than *A. gerardii* to drought. In this grassland, water stress will dominate photosynthetic and productivity responses caused by discrete drought and heat wave events, rather than direct or additive effects of heat stress, with differential sensitivity in these grasses altering future ecosystem function.

15

Introduction

Climate extremes, such as heat waves and drought, are projected to increase in frequency and intensity in the future, and thus there is a clear need to understand how they will impact ecosystems (Easterling et al. 2000, Meehl and Tebaldi 2004, IPPC 2007, Hansen et al. 2012). Despite being relatively short-term events, climate extremes have the potential to cause significant and long-term ecological change, and thus can have impacts disproportionate to their duration (Albertson and Weaver 1946, Tilman and Haddi 1992, Jentsch et al. 2007). Furthermore, heat waves and drought typically co-occur (Trenberth and Shea 2005, De Boeck et al. 2010), so understanding their individual and interactive effects by observation alone is challenging. However, an experimental approach that manipulates both climate factors can improve our mechanistic understanding of the effects of such short-term events on ecological process.

Here, we present results from an experiment in which a season-long drought and a mid-season heat wave of differing magnitudes were imposed independently and in combination on an intact, mesic grassland ecosystem in the central US – the tallgrass prairie. Plant communities in these grasslands are characterized by strong dominance (Collins et al. 1998), with a few C_4 grass species regulating ecosystem function. Thus, we focused on the responses of two dominant C_4 grasses in tallgrass prairie, *Andropogon gerardii* and *Sorghastrum nutans*, which together account for almost half of plant canopy cover, and the majority of total aboveground productivity (Silletti and Knapp 2002, Smith and Knapp 2003). These grasses have been broadly viewed as functional equivalents, with a greater research effort focused on *A. gerardii* (Knapp et al. 1998, Silletti and Knapp 2002). While their overlapping geographic distributions suggests similar climatic requirements (Brown 1993), past studies suggest that these two grasses may respond

differently to projected climate changes, with *A. gerardii* more sensitive to warming and *S. nutans* more sensitive to alterations in precipitation (Weaver and Fitzpatrick 1932, Brown 1993, Silletti and Knapp 2002, Swemmer et al. 2007, Nippert et al. 2009). However, due to the nature of these past studies, separating the effects of temperature from water stress, as well as identifying interactive effects of these drivers on the dominant species has not been possible. Such differential sensitivities may lead to divergent responses to climate change and a reordering of species abundances with important consequences for ecosystem function.

To examine the individual and combined effects of short-term heat waves and drought on these dominant C_4 grasses, we experimentally imposed a mid-summer heat wave at four temperature levels (up to +7.7 °C above ambient) in plots receiving either ambient rainfall or a 66% reduction in growing season rainfall to simulate a severe drought. The primary biotic response variables measured were leaf water status, net photosynthesis, and end of season aboveground productivity for each species. We hypothesized that the effects of drought would be greater than heat for both species, since water is the primary limiting factor in this ecosystem (Knapp et al. 1998). In addition, we hypothesized the combined effects of heat waves and drought would be greater than either factor alone (i.e., additive effects, De Boeck et al. (2010)), with the greatest negative effects on both physiology and productivity manifest with the highest temperature heat wave treatment under drought conditions. Finally, as suggested by previous research (Weaver and Fitzpatrick 1932, Brown 1993, Silletti and Knapp 2002, Swemmer et al. 2007, Nippert et al. 2009), we hypothesized that *A. gerardii* would be more sensitive to heat stress, whereas *S. nutans* would be more sensitive to water stress.

Materials and Methods

Study site and focal species

Research was conducted at the Konza Prairie Biological Station, a 3487 ha native tallgrass prairie in NE Kansas, USA (39°05'N, 96°35'W). The site has a temperate mid-continental climate with cold, dry winters and warm, wet summers. This region is characterized by high interannual variability in precipitation and temperature, which can lead to a four-fold variability in aboveground net primary productivity (Knapp et al. 1998). Konza Prairie Biological Station is dominated by perennial C_4 grasses, in particular *A. gerardii* and *S. nutans*, which together account for most standing biomass and productivity (Smith and Knapp 2003) and determine plant community diversity (Collins et al. 1998). Both species are long-lived clonal plants that primarily reproduce vegetatively via belowground buds on rhizomes (Benson and Hartnett 2006). As clones of each species grow, they sever their root and rhizome connections (Benson and Hartnett 2006) forming a dense matrix of tillers (individual stems) that are intermixed.

Experimental design

The Climate Extremes Experiment was established in 2010 on a site with deep (>1 m) silty clay loam soil adjacent to the long-term Rainfall Manipulation Plots experiment (Fay et al. 2000). The experimental treatments of the Climate Extremes Experiment consisted of a season-long drought and a mid-season heat wave at four temperature levels. The drought treatment was imposed using two modified 6 x 24 m cold frame greenhouse structures (Stuppy, Inc., Kansas City, MO, USA) constructed over native grassland plots (Fig. 2.1A). The roofs of these structures were comprised of 15.2 cm wide strips of Dynaglas Plus® clear corrugated

polycarbonate plastic (PALRAM Industries LTD., Kutztown, PA, USA), which covered 75% of the roof surface. This 75% covering of the roof resulted in the exclusion of ~66% of the ambient rainfall from 15-May to 30-Sep-2010. For the control precipitation treatment, two additional greenhouse structures were covered with deer netting (TENAX Manufacturing Alabama, USA) which allowed all ambient rainfall to reach the plots but reduced photosynthetically active radiation by approximately 10% (equivalent to the effects of the drought shelters, Fig. 2.1A). The control precipitation and drought treatments were randomly assigned to each shelter, and the shelters were oriented E-W and arrayed approximately 5 m from each other. Each shelter was hydrologically isolated by trenching to a depth of 1 m around the perimeter of each 6 x 24 m area; the trench was lined with 6 mil plastic, and metal flashing to prevent subsurface and surface water flow.

Within each structure, we established two rows of five 2 x 2 m plots (10 total) arranged diagonally from each other in a checkerboard arrangement, allowing for a 2 m buffer between plots. Plots were randomly assigned to one of four heat wave treatments (ambient, low, medium, and high), which were imposed mid-summer for two weeks (21-Jul to 03-Aug-2010). The timing of the simulated heat wave coincided with the period of greatest sensitivity to high temperature in this grassland (Craine et al. 2012). Heat wave treatments were imposed by placing transparent chambers that combined passive heating with infrared lamps (IR) over the plots (Fig. 2.1A). Chambers were 2 x 2 m wide and 1.5 m tall, with 1 m 6-mil clear polyethylene walls and covered with Dynaglas Plus® clear corrugated polycarbonate roofs. Ventilation was maintained by placing chambers 0.5 m above the ground surface with adjustable gaps between the roofs and walls. In order to achieve four distinct temperature levels, infrared lamps (HS/MRM 2420, 2000 W, Kalglo Electronics, Inc., Bethlehem, PA, USA) were placed within the heat chambers as

19

follows: ambient = no lamp, low heat = one lamp at half power, medium heat = one lamp at full power, and high heat = two lamps at full power. Lamps were suspended 130 cm above the ground to ensure even coverage across the plot. Heated plots were warmed 24 hours per day for the entire two-week heat wave.

Environmental measurements

Soil moisture and canopy temperature were continuously monitored in each plot to evaluate the effectiveness of drought and heat wave treatments. Volumetric water content (VWC) was measured at a depth of 0-15 cm with 30 cm time-domain reflectometery (TDR) probes (Model CS616, Campbell Scientific, Inc., Logan, UT, USA) buried at a 45° angle in the center of each plot. Canopy temperature (CT) was measured with infrared thermometers (Model SI-111, Apogee Instruments, Inc., Logan, UT, USA) mounted in the SE corner of each plot at a height of 1.5 m. Data from all sensors were sampled every 30-seconds and averaged for 30-minute periods (CR10X Datalogger, Campbell Scientific, Inc., Logan, UT, USA).

Plant ecophysiological measurements

Eight intensive sampling campaigns were conducted between 11:00 and 15:00 CDT throughout the growing season to measure leaf gas exchange (A_{net}) and mid-day leaf water potential (Ψ_{mid}) responses of *A. gerardii* and *S. nutans*. In each plot, a tiller (individual stem) of each species with leaves in the upper canopy was permanently tagged for repeated sampling of gas exchange throughout the experiment. Prior to each sampling campaign, an additional tiller, which was morphologically similar to the permanently tagged individual, was selected in each plot for destructive leaf water potential sampling. Thus, for A_{net} and $\Psi_{mid,}$ we sampled a total of

20 individuals per species for each drought treatment and 5 individuals of each species for each drought by heat wave combination. For all individuals, the youngest fully expanded leaf was measured. Plot sampling order was randomized for each sampling campaign, and A_{net} was measured at 5-second intervals for 2 to 6 minutes with a LI-6400 system (LiCOR, Inc., Lincoln, NE, USA) equipped with an LED light source (light intensity was maintained at 2000 µmol m^{-2} s^{-1}, CO_2 concentration at 400 µmol mol^{-1}, and relative humidity at ambient levels). A_{net} was then calculated for each leaf using an objective selection algorithm (Matlab 7.4, The MathWorks, Inc., Natick, MA, USA) to select a 1-minute period of A_{net} when variability was minimal. Ψ_{mid} was measured on a single leaf per individual using a Scholander-type pressure chamber (PMS Instruments, Inc., Corvallis, OR, USA).

Aboveground productivity

Aboveground production of the dominant C_4 grasses was sampled at the end of the growing season (05-Sep-2010) by harvesting all aboveground plant material of each species in three 0.1 m^2 quadrats randomly located within each plot. Samples were oven dried at 60 °C for 48 hours, and weighed to the nearest 0.1 g.

Statistical analyses

The experiment was a randomized block split-plot design with block nested within the drought treatment, heat wave treatments nested within drought treatment, with the block x heat wave treatment interaction as a random effect. We conducted analyses separately for three sampling periods: (1) the entire growing season, (2) the two-week heat wave, and (3) the last day of the heat wave. The growing season and heat wave sample periods were analyzed using a

repeated measures mixed-model ANOVA, while the last day of the heat wave and aboveground production was analyzed with a mixed-model ANOVA. We conducted analyses separately for each species because the two grasses differ significantly in both A_{net} and Ψ_{mid} (higher and lower in *S. nutans,* respectively) under control (non-stressed) conditions. By analyzing each species separately, we focused on the relative magnitude of responses to the drought and heat wave treatments for each species without the confounding effects of the baseline differences in A_{net} and Ψ_{mid}. We also assessed differential sensitivity by examining the relationship between VWC and Ψ_{mid} (excluding measurements from within heated chambers) for each species using nonlinear regression. All analyses were conducted in SAS (version 9.2, SAS Institute Inc., Cary, NC, USA) and significance was set a $p \leq 0.05$.

Results

Effects of drought and heat waves on environmental variables

  The experimental infrastructure was successful in imposing a drought during the 2010 growing season and simulating a two-week heat wave at four distinct temperatures (Fig. 2.1B, C). Between the start of the simulated drought (15-May-2010) and the end of season biomass harvest (05-Sep-2010), plots in the control shelters received 444.1 mm of rainfall while drought plots received 148.0 mm, a 66% reduction. Overall, this reduction in rainfall resulted in a 43% reduction in VWC relative to control plots (Control = 29.5% $\pm$ 0.47, Drought = 16.9% $\pm$ 0.69, *p* = 0.001; Figure 1B). During the two-week heat wave, there was a strong interaction between date and the drought treatments (Table 2.1) due to more rapid and greater reductions in VWC with control precipitation (58%) than with drought (23%, Fig. 2.1B). For the last day of the heat wave, there was a significant interaction between the drought and heat wave treatments on VWC

(Table 2.1), but reductions in soil moisture were only detected under control precipitation when combined with the high heat wave treatment (Fig. 2.2C).

The four heat wave treatments resulted in different mean daily CT's for the two-week period in late July; ambient plots averaged 27.9 °C and the low, medium and high treatments averaged 7, 17, and 28% warmer than ambient plots, respectively (Fig. 2.1C). The maximum daily high CT recorded in the high treatment was 43.1 °C on 03-Aug-2010, the last day of the heat wave. In addition, we found a significant negative correlation between VWC and CT ($r = -0.78$, $p < 0.001$), across all treatments during this period.

Ecophysiological responses to the drought and heat wave treatments

We found little evidence for additive effects of heat and drought on the ecophysiological responses of the dominant grasses. There were significant effects of heat and drought on Ψ_{mid} (drought, heat, drought x date, heat x date) and A_{net} (drought x date) for both species (Table 2.2). But there were few interactions between the two treatments when assessed over the growing season or just during the heat wave, with the exception of A_{net} for *S. nutans* (drought x heat and drought x heat x date interactions; Table 2.2). We focused on the last day of the two-week heat wave to assess the cumulative effects of the heat wave treatments and increase our ability to detect interactions between heat waves and drought. While there were significant interactions between the heat and drought treatments for both species, there were no combined effects of heat waves of any magnitude under drought conditions (Fig. 2.2A, B). Instead, the effects of the heat wave treatments were only significant under control precipitation (Fig. 2.2A, B). In the control rainfall treatment, A_{net} for *A. gerardii* decreased as the average heat wave temperature increased (Fig. 2.2A). In contrast, *S. nutans* was unaffected by the low and medium heat wave treatments

23

but A_{net} was decreased by ~ 80% from ambient levels for the high heat wave treatment (Fig. 2.2B). Consistent with these responses, there was a negative effect of the heat wave treatments on VWC for the control precipitation, but not the drought treatment (Fig. 2.2C).

There were direct effects of the heat wave and drought treatments on the dominant grasses, but these direct effects differed between Ψ_{mid} and A_{net}. In general, Ψ_{mid} was affected by both heat and drought, while A_{net} was only sensitive to drought. The drought and heat wave treatments resulted in similar responses in Ψ_{mid} for both *A. gerardii* and *S. nutans* with significant drought x date and heat x date interactions (Table 2.2). For both species, the effects of drought on Ψ_{mid} were not evident until mid-July, and treatment differences emerged earlier in Ψ_{mid} than A_{net} (Fig. 2.3). Once drought effects were manifest, they persisted for the reminder of the growing season in both species (Fig. 2.3). These dominant grasses also responded similarly to VWC (Fig. 2.4), at VWC greater than 20%, Ψ_{mid} did not vary in response to changes in VWC in either species, but below this threshold Ψ_{mid} decreased sharply in both grasses with reduced VWC (Fig. 2.4). There were significant heat wave x date interactions for both species (Table 2.2, growing season). The greatest decrease in Ψ_{mid} occurred with the high heat treatment, and differences among the heat wave treatments persisting nine days after the heat wave ended (Fig. 2.5). In contrast, A_{net} for both species was only sensitive to drought as evident by drought x date interactions (Table 2.2).

While both species were sensitive to heat and water stress, there was evidence for differential sensitivity to drought, but not heat. A_{net} differed between the two species in specific ways during the drought. First, drought-induced reductions in A_{net} occurred earlier in *S. nutans* (29-Jul-2010) than *A. gerardii* (03-Aug-2010). Secondly, there was a larger relative decrease in A_{net} for *S. nutans* in the drought plots before vs. after the heat wave (94% in *S. nutans,* 81% in *A.*

gerardii; Fig. 2.3). Finally, the difference in A_{net} between the control precipitation and drought treatment immediately after the heat wave was greater in *S. nutans* (85% reduction) than in *A. gerardii* (64% reduction, Fig. 2.3).

Effects of the drought and heat wave treatments on aboveground production

Drought significantly reduced end-of-season aboveground production in *S. nutans* by 37%, but had no effect on production in *A. gerardii* (Fig. 2.6 and Table 2.3). Surprisingly, the heat wave treatments did not reduce aboveground production for either species despite clear visual signs of heat-induced foliar senescence, and thus there were no significant interactions or additive effects of heat and drought for either species (Table 2.3).

Discussion

Forecasts of more frequent and severe drought and heat waves portend ecological responses from individual to ecosystem scales (Smith 2011), particularly if the magnitude or combination of climate stressors pushes species beyond response thresholds (Gutschick and BassiriRad 2003). Given that the attributes of dominant species strongly influence most ecosystem processes (Whittaker 1965, Grime 1998), we focused on the ecophysiological and aboveground productivity responses of two dominant C_4 grasses in central US grasslands as key to predicting ecosystem responses to drought and heat waves of different magnitudes and their interactions. While previous research suggested that these species respond differentially to heat and water stress (Silletti and Knapp 2002, Swemmer et al. 2006, Nippert et al. 2009), our experimental design permitted us to directly test this purported differential sensitivity. Moreover, because we imposed heat waves of different magnitudes, we could further determine if there are

species-specific response thresholds. Overall, we found (1) no additive effect of heat waves and drought in this grassland, (2) that both species were affected more by a season-long drought than a two-week heat wave regardless of magnitude, and (3) evidence for differential sensitivity to drought between the dominant C_4 grasses, with *S. nutans* more sensitive than *A. gerardii*.

The levels of drought and heat waves imposed in this experiment were severe, but not outside the range of recent climate of this site (1984-2008, Konza Prairie LTER data set AWE012). Over this 25- year period, mean annual precipitation was 840.3 ± 38.9 mm, and the driest year (1988) received 481.5 mm. These values were similar to our two precipitation treatments: (control = 836.9, drought = 476.6 mm). The simulated heat wave occurred for two weeks in late July, when mean daily temperatures over the 25-year period were 26.4 ± 0.4 °C and the warmest year (1999) was 29.7 ± 0.6 °C. Mean daily canopy temperature in this study (Fig. 2.1C), overlapped and exceeded this range in temperature (ambient = 27.2 ± 0.4 °C and high = 36.6 ± 0.3 °C), and the maximum daily canopy temperature (high = 43.1 °C) is virtually identical to the 25-year record high air temperature measured at this site (43.2 °C, 1983).

Greater sensitivity to drought in *S. nutans* relative to *A. gerardii* is consistent with past studies (Weaver and Fitzpatrick 1932, Brown 1993, Silletti and Knapp 2002, Swemmer et al. 2006, Nippert et al. 2009). However, none of these studies imposed drought at this magnitude under controlled experimental conditions. *S. nutans* responded earlier to the drought and with greater absolute and relative decreases in A_{net} than *A. gerardii*. Consistent with this response was a significant decrease in aboveground productivity for *S. nutans* while *A. gerardii* did not respond to drought. These divergent responses occurred despite similar responses in Ψ_{mid}. This suggests greater resistance to drought in *A. gerardii* than *S. nutans* and that a drought of greater magnitude may be required before a loss of productivity occurs in *A. gerardii*. Indeed, earlier

experiments found significant reductions in aboveground productivity in response to drought for
A. gerardii, but at almost twice as negative relative to Ψ_{mid} measured in this study (Knapp 1984).
The reduction in aboveground productivity in *S. nutans* but not *A. gerardii* could have important
implications for plant community structure by potentially increasing the competitive advantage
of *A. gerardii* in this grassland.

The imposed heat wave had no direct effects on aboveground production for either
species nor when combined with drought, suggesting that both species were resistant to the
timing, magnitude and the combination of the two treatments. Nonetheless, physiological
responses to the two-week heat wave treatments were evident (e.g., both species had reduced
Ψ_{mid} in response to the heat wave treatments), but there was no support for the hypothesis that *A.
gerardii* is more sensitive to heat than *S. nutans.* Furthermore, when the cumulative effects of the
heat wave and drought treatments were examined for the last day of the two-week heat wave
period, significant interactions were detected for A_{net} in both species, with heat wave effects only
observed in the control rainfall treatment (Fig. 2.2A, B). What was particularly striking was the
appearance of a threshold response to heat in *S. nutans*. In this species, A_{net} was maintained at
the same level as the control treatment for the low and medium heat wave treatments, but was
dramatically reduced (by 78%) with the high heat wave treatment. In contrast, A_{net} gradually
declined in *A. gerardii* with increasing heat wave temperature. Contrary to expectations, A_{net} did
not differ with the heat wave treatments when combined with drought for either species nor did
we observe an additive effect on aboveground productivity. This lack of response was surprising,
given that in a similar study, De Boeck et al. (2011) reported additive effects of drought and heat
waves. Instead, it appears in this system, that the high degree of water stress imposed by the
drought treatment negated any additional effects of the heat wave treatments.

What remains unclear is whether the heat wave effects on A_{net} were direct or if they occurred indirectly through water stress as noted in several experimental and observational studies (Milbau et al. 2005, Marchand et al. 2006, Reichstein and Ciais 2007, Arnone et al. 2011, De Boeck et al. 2011). The direct effects of heat could have lead to thermal damage to the photosynthetic machinery. If there had been significant thermal damage to the photosynthetic capacity of these grasses, we would have expected differences in A_{net} among the heat wave treatments to persist after the treatments ended, as well as a negative response in end of season productivity; neither of these occurred. Also, thermal damage may have been avoided since the maximum temperature imposed in this experiment was within the measured range of thermal tolerance for *A. gerardii* (Knapp 1985). However, we cannot rule out the potential direct effects increasing temperature on metabolic processes, such as respiration, which could decrease A_{net} if respiration and assimilation had different responses to temperature (positive for respiration, neutral or negative for assimilation). Additionally, there are several lines of evidence to suggest that the heat wave treatments indirectly affected A_{net} via water stress. First, there were significant differences in VWC among the heat wave treatments with control precipitation; these differences became more pronounced as the heat wave progressed. This would be expected during a naturally occurring heat wave, which typically co-occur with low precipitation (Trenberth and Shea 2005, De Boeck et al. 2010), and when combined with high evapotranspiration rates due to heat, decreases VWC. For these grasses, we saw evidence for water stress emerging once VWC drops below ~20%, where Ψ_{mid} declines rapidly (Fig. 2.4). By the last day of the heat wave, several of the heat treatments had dropped below this VWC (Fig. 2.4), and we observed direct effects of heat on Ψ_{mid} for both species throughout the heat wave, suggesting that heat may have reduced plant water status indirectly through enhanced vapor pressure deficits, decreased VWC

or both. Overall, these results show that under the control precipitation, the heat wave treatments imposed both heat and water stress, and thus we could not separate the interactive effects of heat and attribute the decline in photosynthesis to a single driver. In order to separate the direct and indirect effects of heat, water would have to be added to keep soil moisture above limiting levels (for example, greater than 20% VWC in this ecosystem).

Although this mesic grassland ecosystem was subjected to severe drought and a range of heat wave intensities, its response was modest overall and primarily related to the ability of the dominant grass, *A. gerardii*, to successfully cope with water stress. However, greater sensitivity to water stress and a strong threshold response to heat were clearly evident in the dominant grass, *S. nutans*. This suggests that a future climate that includes more frequent droughts and heat waves may have greater initial impacts on community structure relative to ecosystem processes. Such mechanistic information is critical for enhancing our ability to forecast ecosystem responses to a future with more frequent and intense periods of climate extremes (Easterling et al. 2000, IPCC 2007).

Tables

Table 2.1 Effects of the drought and heat wave treatments and sampling date on mean daily volumetric water content (VWC) and canopy temperature (CT) during the two-week period in which the heat wave treatments were applied. F-statistics and p-values from mixed-model repeated measures ANOVAs are reported. Bold text indicates significance at $p \leq 0.05$.

Effect	VWC		CT	
	F	p	F	p
Drought	**214.3**	**0.004**	8.3	0.102
Heat	**3.6**	**0.029**	**209.9**	**<.001**
Date	**675.7**	**<.001**	**941.8**	**<.001**
Drought x Heat	1.0	0.402	1.0	0.396
Drought x Date	**384.7**	**<.001**	**9.6**	**<.001**
Heat x Date	**9.6**	**<.001**	**14.2**	**<.001**
Drought x Heat x Date	**6.3**	**<.001**	1.4	0.069

Table 2.2 Effects of the drought and heat wave treatments on mid-day leaf water potential (ψ_{mid}) and net photosynthesis (A_{net}) of the dominant C_4 grasses (*Andropogon* gerardii and *Sorghastrum nutans*) over the growing season (top) and during the two-week period in which the heat wave treatments were applied (bottom). F-statistics and p-values from mixed-model repeated measures ANOVAs for each species separately are reported. Bold text indicates significance at $p \leq 0.05$.

| | *A. gerardii* | | | | *S. nutans* | | | |
| | Ψ_{mid} | | A_{net} | | Ψ_{mid} | | A_{net} | |
Effect	F	P	F	p	F	p	F	p
Growing Season								
Drought	**19.7**	**0.048**	3.8	0.195	**71.7**	**<.001**	6.4	0.130
Heat	4.0	0.070	2.0	0.116	**5.4**	**0.027**	4.0	0.077
Drought x Heat	0.6	0.647	0.6	0.602	0.9	0.499	**5.7**	**0.041**
Date	**97.6**	**<.001**	**32.7**	**<.001**	**97.6**	**<.001**	**50.0**	**<.001**
Drought x Date	**10.3**	**<.001**	**3.2**	**0.004**	**10.5**	**<.001**	**6.3**	**<.001**
Heat x Date	**1.81**	**0.023**	0.6	0.894	**2.5**	**0.001**	1.3	0.219
Drought x Heat x Date	1.32	0.170	0.4	0.987	0.9	0.551	**181**	**0.022**
Heat Wave								
Drought	33.7	**0.030**	3.6	0.205	92.3	**<.001**	8.9	0.099
Heat	6.0	**0.033**	1.0	0.408	14.4	**0.002**	4.2	0.066
Drought x Heat	1.2	0.395	1.3	0.297	1.6	0.281	3.4	0.099
Date	33.6	**<.001**	24.8	**<.001**	38.3	**<.001**	38.0	**<.001**
Drought x Date	0.9	0.426	3.3	**0.043**	1.0	0.368	7.3	**0.001**
Heat x Date	1.8	0.109	0.3	0.932	0.8	0.559	1.0	0.408
Drought x Heat x Date	1.5	0.200	1.0	0.433	0.7	0.648	3.2	**0.009**

Table 2.3 Effects of the drought and heat wave treatments on aboveground productivity for each species separately. F-statistics and p-values from mixed-model repeated measures ANOVAs are reported. Bold text indicates significance at p ≤ 0.05.

Effect	*A. gerardii*		*S. nutans*	
	F	p	F	p
Drought	0.1	0.747	**4.9**	**0.034**
Heat	0.06	0.989	0.28	0.840
Drought x Heat	0.40	0.753	0.78	0.516

Figures
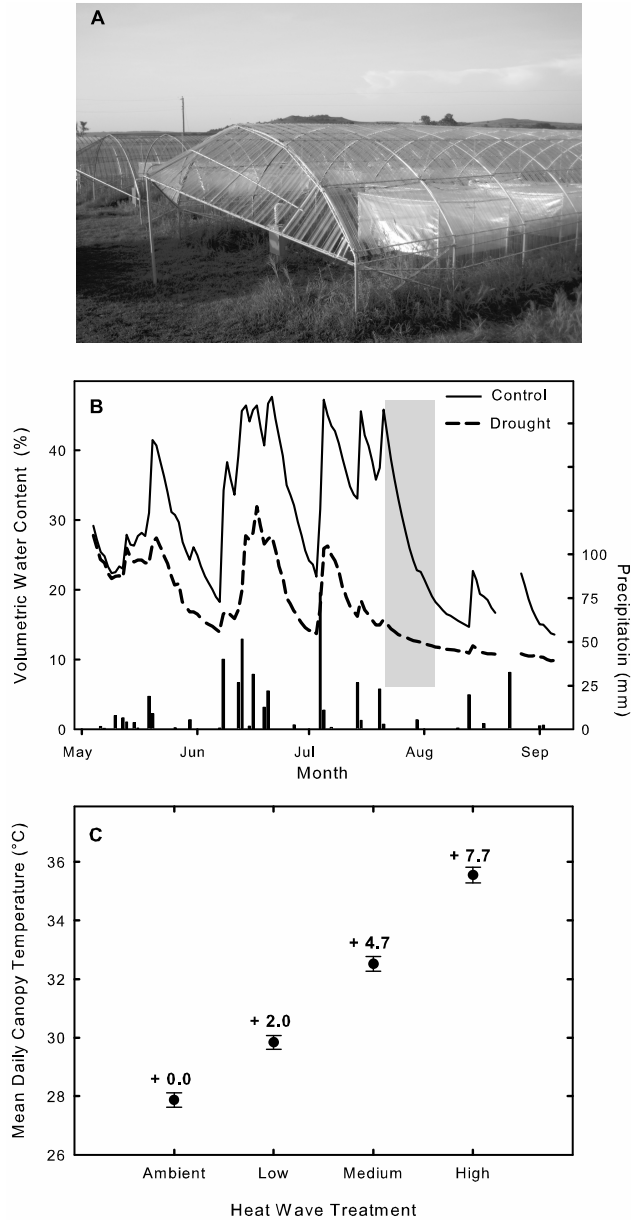
Figure 2.1 Experimental infrastructure during the simulated heat wave (A) and the effects of drought and heat wave treatments on (B) volumetric water content (VWC, %) and (C) canopy temperature (CT, °C). (A) Shown in the foreground is one of the greenhouse structures modified to impose drought. The frame is partially covered with polycarbonate strips to reduce rainfall inputs by 66% during the growing season. Nested within this structure are heat chambers with infrared lamps inside that imposed a two-week heat wave (21-July to 03-Aug-2010). (B) VWC is shown for the control and drought treatments (left x-axis) with a box highlighting the timing of the two-week period that the heat wave treatments were applied. Black bars are ambient precipitation (right x-axis). Note - missing data in late August was due to a power failure. (C) Mean daily canopy temperature (± 1 SE) is shown for the four heat wave treatments.
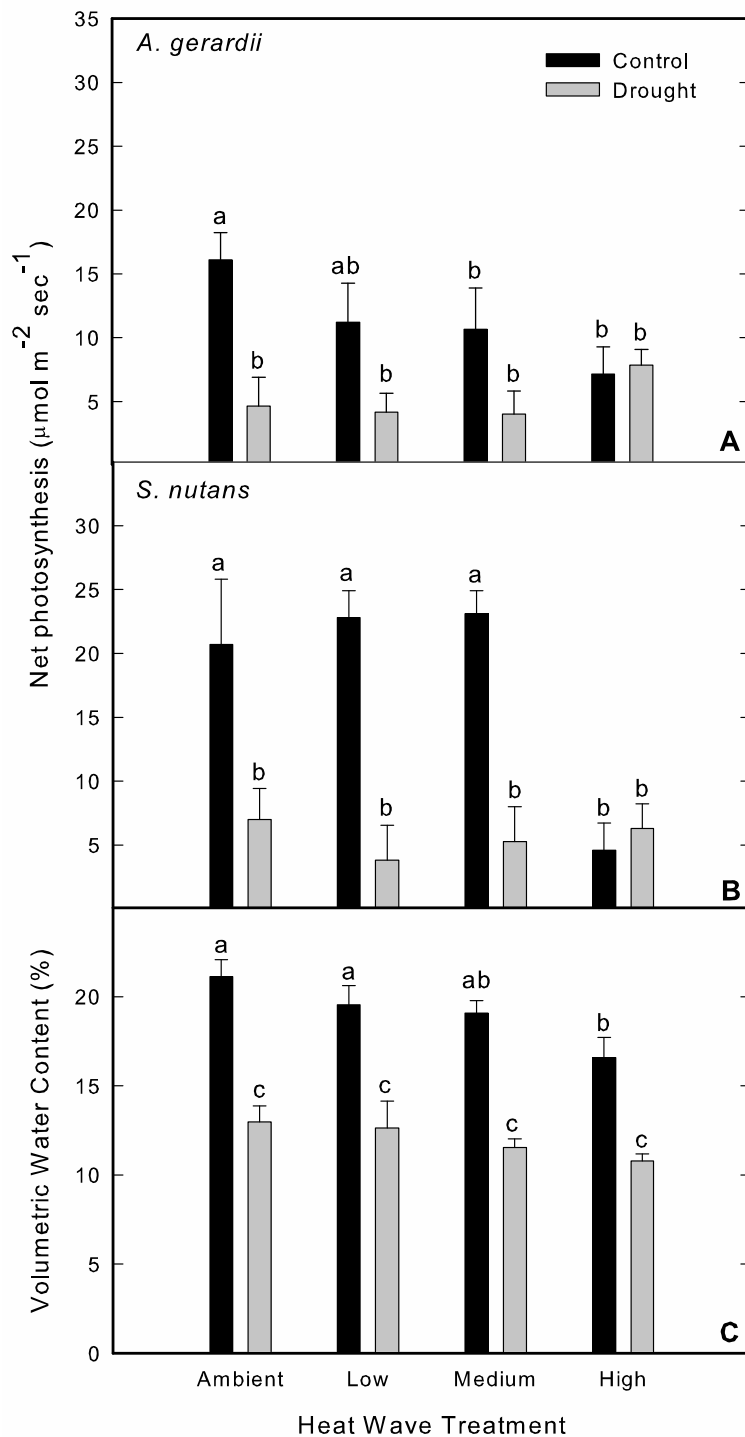
Figure 2.2 Interactive effects of drought and the heat wave treatments on net photosynthesis (A_{net}) of *A. gerardii* (A) and *S. nutans* (B), as well as mean volumetric water content (C) measured on the last day of the simulated heat wave, 03-Aug-2010. Error bars indicate one standard error and different letters denote significant differences among treatments within each of the three independent analyses ($p \leq 0.05$).
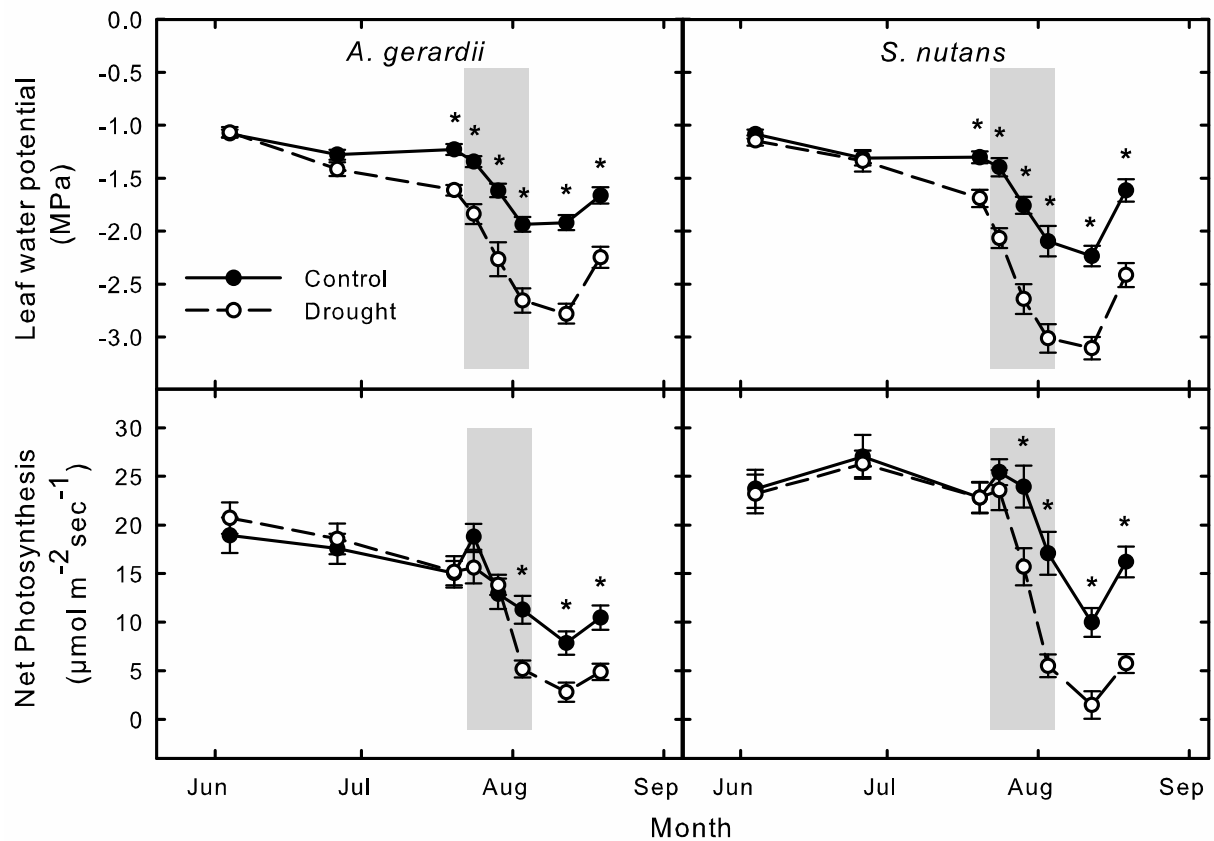
Figure 2.3 Responses of mid-day leaf water potential (Ψ_{mid}) and net photosynthesis (A_{net}) for *A. gerardii* and *S. nutans* to the drought treatment over the growing season. The grey box highlights the two-week period that the heat wave treatments were applied. Error bars indicate one standard error and asterisks denote a significant difference ($p \leq 0.05$) between the control and drought treatments for a given date.
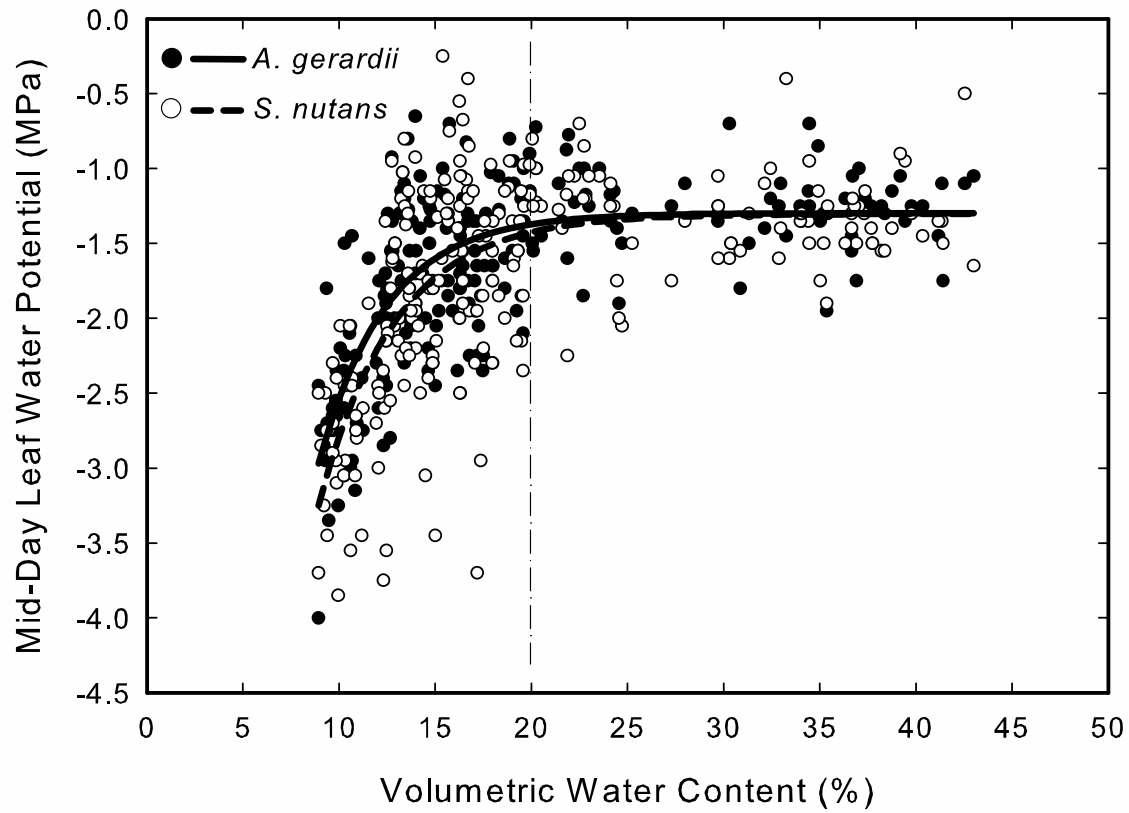
Figure 2.4 Volumetric water content (VWC) vs. mid-day leaf water potential (Ψ_{mid}) for *A. gerardii* and *S. nutans* for all measurements during the growing season (excluding measurements from within heated chambers). Dashed line at 20% VWC, below which Ψ_{mid} declines rapidly with decreasing VWC.
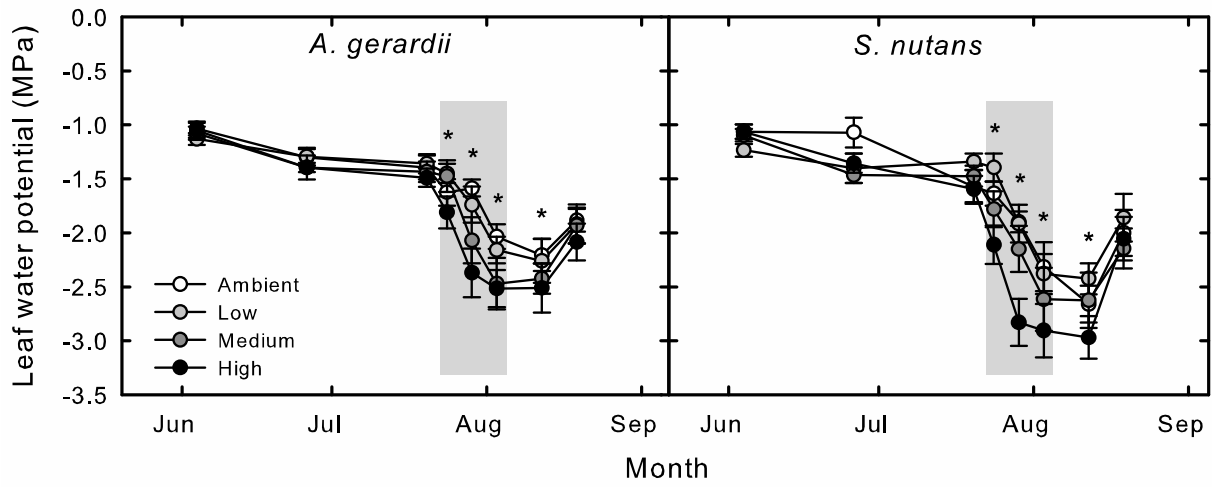
Figure 2.5 Effects of the heat wave treatments on mid-day leaf water potential (Ψ_{mid}) for *A. gerardii* and *S. nutans*. The grey box highlights the two-week period that the heat wave treatments were applied. Error bars indicate one standard error and asterisks denote a significant difference ($p \leq 0.05$) between the heat wave treatments for a given date.
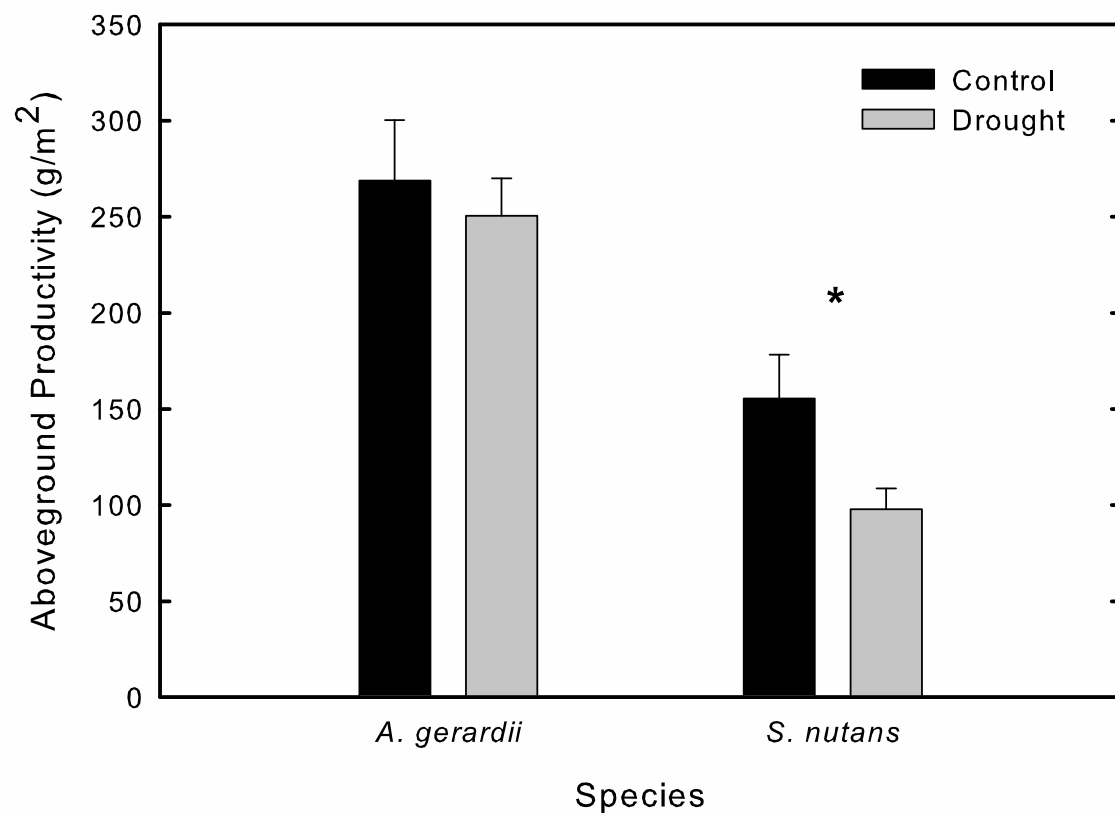
Figure 2.6 Drought effects on aboveground production for *A. gerardii* and *S. nutans*. Error bars indicate one standard error and asterisks denote a significant difference (p ≤ 0.05) between the control and drought treatments for a given species.

References

Albertson, F. W., and J. E. Weaver. 1946. Reduction of ungrazed mixed prairie to short grass as a result of drought and dust. Ecological Monographs 16:449–463.

Arnone, J. A., R. L. Jasoni, A. J. Lucchesi, J. D. Larsen, E. A. Leger, R. A. Sherry, Y. Luo, D. S. Schimel, and P. S. J. Verburg. 2011. A climatically extreme year has large impacts on C4 species in tallgrass prairie ecosystems but only minor effects on species richness and other plant functional groups. Journal of Ecology 99:678–688.

Benson, E. J., and D. C. Hartnett. 2006. The Role of Seed and Vegetative Reproduction in Plant Recruitment and Demography in Tallgrass Prairie. Plant Ecology 187:163–178.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. Global Change Biology 16:1992–2000.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. The New phytologist 189:806–17.

Brown, D. A. 1993. Early Nineteenth-Century Grasslands of the Midcontinent Plains. Annals of the Association of American Geographers 83:589–612.

Collins, S., A. Knapp, J. Riggs, J. Blair, and E. Steinauer. 1998. Modulation of diversity by grazing and mowing in native tallgrass prairie. Science 280:745–7.

Easterling, D. R., G. A. Meehl, C. Parmesan, S. A. Changnon, T. R. Karl, and L. O. Mearns. 2000. Climate extremes: observations, modeling, and impacts. Science 289:2068–2074.

Grime, J. 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. Journal of Ecology 86:902–910.

Gutschick, V. P., and H. BassiriRad. 2003. Extreme events as shaping physiology, ecology, and evolution of plants: toward a unified definition and evaluation of their consequences. New Phytologist 160:21–42.

Hansen, J., M. Sato, and R. Ruedy. 2012. Perception of climate change. Proceedings of the National Academy of Sciences of the United States of America 109:1–9.

IPPC. 2007. Climate Change 2007: The Physical Science Basis. (S. Solomon, D. Qin, M. Manning, Z. Chen, M. Marquis, K. Averyt, M. Tignor, and L. Miller, Eds.) Energy Environment. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

Jentsch, A., J. Kreyling, and C. Beierkuhnlein. 2007. A new generation of climate-change experiments: events, not trends. Frontiers in Ecology and the Environment 5:365–374.

Knapp, A. 1985. Effect of fire and drought on the ecophysiology of andropogon gerardii and panicum virgatum in a tallgrass prairie. Ecology 66:1309–1320.

Knapp, A. K. 1984. Water relations and growth of three grasses during wet and drought years in a tallgrass prairie. Oecologia 65:35–43.

Knapp, A. K., J. M. Briggs, D. C. Hartnett, and S. L. Collins. 1998. Grassland Dynamics: Long-Term Ecological Reserach in Tallgrass Prairie. Oxford University Press, Inc., New York.

Marchand, F. L., M. Verlinden, F. Kockelbergh, and B. J. Graae. 2006. Disentangling effects of an experimentally imposed extreme temperature event and naturally associated. Functional Ecology 20:917–928.

Meehl, G. A., and C. Tebaldi. 2004. More intense, more frequent, and longer lasting heat waves in the 21st century. Science 305:994–997.

Milbau, A., L. Scheerlinck, D. Reheul, B. De Cauwer, and I. Nijs. 2005. Ecophysiological and morphological parameters related to survival in grass species exposed to an extreme climatic event. Physiologia Plantarum 125:500–512.

Nippert, J. B., P. A. Fay, J. D. Carlisle, A. K. Knapp, and M. D. Smith. 2009. Ecophysiological responses of two dominant grasses to altered temperature and precipitation regimes. Acta Oecologica 35:400–408.

Reichstein, M., and P. Ciais. 2007. Reduction of ecosystem productivity and respiration during the European summer 2003 climate anomaly□: a joint flux tower , remote sensing and modelling analysis. Global Change Biology 13:634–651.

Silletti, A., and A. Knapp. 2002. Long-term responses of the grassland co-dominants Andropogon gerardii and Sorghastrum nutans to changes in climate and management. Plant Ecology 163:15–22.

Smith, M. D. 2011. An ecological perspective on extreme climatic events: a synthetic definition and framework to guide future research. Journal of Ecology 99:656–663.

Smith, M., and A. Knapp. 2003. Dominant species maintain ecosystem function function with non-random species loss. Ecology Letters 6:509–517.

Swemmer, A., A. Knapp, and M. Smith. 2006. Growth responses of two dominant C4 grass species to altered water availability. International Journal of Plant Sciences 167:1001–1010.

Swemmer, A. M., A. K. Knapp, and H. a. Snyman. 2007. Intra-seasonal precipitation patterns and above-ground productivity in three perennial grasslands. Journal of Ecology 95:780–788.

Tilman, D., and A. El Haddi. 1992. Drought and Biodiversity in Grasslands. Oecologia 89:257–264.

Trenberth, K. E., and D. J. Shea. 2005. Relationships between precipitation and surface temperature 32:2–5.

Weaver, J. E., and T. J. Fitzpatrick. 1932. Ecology and Relative Importance of the Dominants of Tall-Grass Prairie. Botanical Gazette 93:113.

Whittaker, R. H. 1965. Dominance and diversity in land plant communities: Numerical relations of species express the importance of competition in community function and evolution. Science 147:250–260

Chapter 3

PHOTOSYNTHETIC RESPONSES TO AN EXPERIMENTAL HEAT WAVE ARE MEDIATED BY SOIL WATER STATUS IN A MEISC GRASSLAND

Overview

Climate extremes are predicted to increase in their frequency and intensity, with likely impacts on regional and global carbon cycling. Extremes such as heat waves and drought often co-occur, making it difficult to attribute ecological responses to individual climate drivers using natural observations, and thus experimental manipulations are better suited to study extremes. Past experimental studies have found ecological responses to heat were primarily indirect, through enhanced soil drying. In 2011, we subjected a native mesic grassland to a two week heat wave at four levels under either high or low soil moisture conditions and examined the photosynthetic response of two dominant C_4 grasses. Our goal was to alter these climate drivers independently to assess direct vs. indirect effects of heat. We observed no effect of the heat on photosynthesis under low soil moisture conditions due to stomatal limitations, which resulted in low levels of photosynthesis and apparent insensitivity to increasing levels of heat. On the other hand, there were reductions in photosynthesis with increasing heat under the high soil moisture, which appear to be driven by direct effects of heat. These results suggest that under extreme drought, heat waves may have little detectable effect on photosynthesis, but the effects of increasing heat under non-limiting soil moisture conditions may directly affect photosynthetic capacity. However, as seen in other studies, eventually heat will cause water stress through

enhanced evaporation, causing an additive effect between heat and drought, until severe drought renders photosynthesis insensitive to heat. This dynamic response of photosynthesis to heat and drought could have important implications for regional and global carbon cycles.

Introduction

Climate extremes are expected to increase in their frequency and intensity over the next century (IPCC 2012) with the potential to alter the structure and function of ecosystems more than shifting means alone (Easterling et al. 2000, Jentsch et al. 2007, Smith 2011). If geographically extensive, climate extremes also may impact regional and global carbon cycles, with positive feedbacks on global warming (Ciais et al. 2005, Reichstein et al. 2013). Such feedbacks originate at the ecosystem level primarily as a consequence of climate extremes leading to increased physiological stress (Gutschick and BassiriRad 2003) and in some cases mortality of common (or dominant) plant species (Breshears et al. 2005), which control a high proportion of ecosystem functions (Whittaker 1965, Grime 1998). Thus, assessing how the dominant species in regionally extensive ecosystems, such as grasslands, will respond to climate extremes is important for forecasting the carbon cycling consequences of climate change.

Although much has been learned from the study of naturally occurring climate extremes, employing more mechanistic frameworks to build understanding of their ecological consequences is critical (Smith 2011). Experimental approaches can be particularly valuable in this regard (Jentsch et al. 2007, Smith 2011), however, there are unique challenges to experimentally imposing climate extremes, as heat waves and drought often co-occur naturally. A recent study (De Boeck et al. 2010) on Western European meteorological records found that heat waves coincided with lower precipitation and higher vapor pressure deficits (VPD) than

average. Reduced precipitation and higher VPD could induce plant water stress through combined effects on soil drying, and therefore it is difficult to separate direct and indirect effects of heat on ecological processes using naturally occurring heat waves and drought. Indeed, many warming manipulation experiments, which have not controlled for heat-induced water losses, have found that the effects of heat on ecological processes have been primarily indirect, through enhanced soil drying (Milbau et al. 2005, Marchand et al. 2006, Arnone et al. 2008, De Boeck et al. 2011, Hoover Chapter 2). Separating these may not be an issue as long as past correlations in climate drivers foretell the future. However, there is abundant evidence that this will not be the case with novel and no-analog climates becoming more common (Nippert et al. 2006, Williams et al. 2007). Thus, experiments that alter inputs of heat and water independently can provide the mechanistic understanding needed for modeling ecological responses to future climate extremes.

We examined the physiological responses associated with carbon uptake of two dominant C_4 grasses in a native tallgrass prairie community to a simulated two-week heat wave imposed with contrasting soil moisture conditions: high soil moisture to minimize water limitation vs. low soil moisture conditions that would occur with extreme drought. We focused on leaf-level net photosynthesis because of the important role of carbon uptake processes in regional and global carbon cycles (Houghton 2007), and the thermal sensitivity of photosynthesis (Wahid et al. 2007). Furthermore, the two dominant C_4 grass we examined have purported differential sensitivities to heat and drought (Silletti and Knapp 2002, Swemmer et al. 2006, Nippert et al. 2009). This allowed us to address the following questions: 1) How does soil water availability impact the effect of a simulated heat wave on net photosynthesis in two dominant C_4 grasses? 2) Are their physiological responses driven by direct or indirect effects of heat, or a combination? and 3) Do these grasses differ in their responses to heat?

Methods

Study site

The study was conducted in an intact, annually burned, native tallgrass prairie grassland located in a deep soil, lowland site at the Konza Prairie Biological station in NE Kansas, USA (39°05'N, 96°35'W). The climate is characterized as mid-continental, with cold, dry winters, warm wet summers and high interannual variability in precipitation and temperature (Knapp et al. 1998). In addition to variability, extreme drought and both short- and long-term heat waves are historically significant in this region (Weaver 1954, Woodhouse and Overpeck 1998, Burnette et al. 2010, Burnette and Stahle 2012). The study site was dominated by two perennial C_4 grasses, *Andropogon gerardii* and *Sorghastrum nutans* which control ecosystem function and productivity, accounting for the majority total aboveground primary production (ANPP; Silletti & Knapp, 2002; Smith & Knapp, 2003).

Experimental design

The Climate Extremes Experiment (CEE) was established in 2010 to examine the effects of periods of high temperatures (heat waves) and drought on a tallgrass prairie ecosystem. Treatments were imposed for two consecutive years, with this paper focusing on results from the second year of the experiment (and the most extreme climatically of the two; Hoover Chapter 3). We manipulated precipitation using modified greenhouses and water additions to create two soil moisture conditions (1) high soil moisture, which received ambient rainfall plus supplemental irrigation to maintain soil moisture above limiting levels (>20% volumetric water content; similar to Knapp, Briggs and Koelliker 2001) and (2) low soil moisture, where ambient rainfall was passively removed using partial roofs attached to greenhouse frames (Yahdjian and Sala

45

2002). To impose these soil moisture conditions, we used four modified 6 x 24 m greenhouses frames (Stuppy, Inc., Kansas City, MO, USA) constructed over native grassland plots. The roofs of the two rainfall reduction shelters were partially covered (75% of the surface) by 15.2 cm wide strips of Dynaglas Plus® clear corrugated polycarbonate plastic (PALRAM Industries LTD., Kutztown, PA, USA), which excluded ~66% if ambient rainfall during the growing season (April 1st to August 30th) and caused low soil moisture conditions. Shelters over plots receiving high soil moisture conditions were covered with netting (Cintoflex C, Tenax Corporation, Baltimore, MD, USA) to reduce photosynthetially active radiation by about 10% (equivalent to rainfall reduction shelter effects), but allowing ambient rainfall to pass through. Beneath each shelter, soils and roots were hydrologically isolated by trenching to 1 m and surrounding the soil columns with plastic and metal flashing to prevent above- and belowground lateral flow. Ten 2x2 m plots were established within each shelter and randomly assigned to one of four heat wave treatments (ambient, low, medium, and high). In 2011, the heat wave treatments were imposed mid-summer for two weeks (July 13-26th) using passive heat chambers and infrared heat lamps. The transparent heat chambers were built on ¾" PVC frames, with 1 m 6-mil clear polyethylene walls and Dynaglas Plus® clear corrugated polycarbonate roofs. Ventilation was achieved by placing chambers 0.5 m aboveground with adjustable gaps between the roofs and walls. In order to impose four distinct temperature levels, infrared heat lamps (HS/MRM 2420, 2000 W, Kalglo Electronics, Inc., Bethlehem, PA, USA) were placed within the heat chambers as follows: control = no lamp, low heat = one lamp at ½ power ($+250$ W/m^2), medium heat = one lamp at full power ($+500$ W/m^2), and high heat = two lamps at full power ($+1000$ W/m^2). Lamp heights were adjusted to account for different canopy heights due to the effects of precipitation

treatments on plant growth (control lamps = 150; drought = 120 cm) to ensure even heat coverage across the plot, and remained on 24 hours a day for the duration of the heat wave.

Environmental measurements

Soil moisture, canopy temperature and air temperature/relative humidity were monitored in each plot (n=40) throughout the two-week heat wave. Volumetric water content (VWC) was measured with 30 cm time-domain reflectometery (TDR) probes (Model CS616, Campbell Scientific, Inc., Logan, UT, USA) buried 0-15 cm deep and at a 45° angle. Canopy temperature (CT) was measured with infrared thermometers (Model SI-111, Apogee Instruments, Inc., Logan, UT, USA) mounted in the SE corner of each plot. Data from VWC and CT sensors were sampled every 30-seconds and averaged for 30-minute periods (CR10X Datalogger, Campbell Scientific, Inc., Logan, UT, USA). The vapor pressure deficit (VPD) was calculated for each plot using air temperature and relative humidity data sampled at 30-minute increments using iButtons (Model DS1923, Maxim Integrated, San Jose, CA, USA) placed in the center of each plot at canopy level.

Physiological measurements

Four intensive physiological sampling campaigns (July 15th, 19th, 22nd, 26th) were conducted during the simulated two-week heat wave (July 13-26th), with each campaign conducted between 11:00 and 15:00 CDT. In each plot, a single tiller of both *A. gerardii* and *S. nutans* were permanently tagged and repeatedly sampled for gas exchange. For each individual sampling campaign, another tiller was selected per plot for each species for destructive mid-day leaf water potential (Ψ_{mid}) sampling. Therefore, each precipitation x heat treatment combination

had 5 replicates for each species, and plot sampling order was randomized prior to each sampling campaign. The youngest fully expanded leaf was measured for both gas exchange and Ψ_{mid}. Gas exchange was measured at 5-second intervals for 2 to 6 minutes with a LI-6400 system (LiCOR, Inc., Lincoln, NE, USA) equipped with an LED light source (light intensity was maintained at 2000 μmol m^{-2} s^{-1}, CO_2 concentration at 400 μmol mol^{-1}, and relative humidity at ambient levels). Net photosynthesis (A_{net}) was then calculated for each leaf using an objective selection algorithm (Matlab 7.4, The MathWorks, Inc., Natick, MA, USA) to select a 1-minute period of A_{net} when variability was minimal. Ψ_{mid} was measured on a single leaf per individual using a Scholander-type pressure chamber (PMS Instruments, Inc., Corvallis, OR, USA). Finally, we measured light-adapted ($\Delta F/F_{m'};\ \Phi_{PSII}$) and dark-adapted ($F_v/F_{m'}$) chlorophyll fluorescence on the last day of the heat wave, using the LiCOR LI-6400 system. Light-adapted measurements were taken during the day (1200-1400 CDT) with the following settings: measurement (intensity = 5; modulation = 20; filter = 1; and gain = 10) and flash (duration = 0.8; intensity = 8; modulation = 20; and filter = 50), while dark-adapted measurements were taken at night (2200-2300 CDT) with the following settings: measurement (intensity =1; modulation = 0.25; filter = 1; and gain = 10) and flash (duration = 0.8; intensity = 7; modulation = 20; and filter = 50).

Statistical analyses

The analyses focused exclusively on the two-week heat wave period in mid-July and the sampling campaigns within it. The experimental design was a randomized block split-plot design with block nested within precipitation treatment, heat wave treatments nested within precipitation treatment and the block by heat interaction as a random effect. We used repeated measures mixed model ANOVAs to assess precipitation and heat wave treatment effects over

time. Due to strong interactions between heat and precipitation treatments on environmental variables, we also employed a regression-based approach, analyzing precipitation treatments separately. Linear regressions were performed to examine the relationship between CT and VWC. We also used a stepwise linear regression model to evaluate the effects of three predictor variables (CT, VWC, VPD) on A_{net} for both species, with a parameter selection criteria of $\alpha = 0.05$. Fluorescence measurements were analyzed using a mixed model ANOVA where species and precipitation were analyzed independently. All analyses were conducted in SAS (version 9.3, SAS Institute Inc., Cary, NC, USA) and significance was set a $p \leq 0.05$.

Results

The magnitude of the precipitation and heat wave treatments imposed in this experiment was near or exceeded records for drought and high air temperatures for the region. While the growing season (April 1-August 30) precipitation inputs (ambient + supplemental irrigation) was slightly above average in high soil moisture conditions based on this long-term record, (high = 607.3; long-term average = 521.6 ± 16.1 mm) simulated rainfall inputs for low soil moisture conditions were the second lowest on record, with the lowest occurring during the driest year of the 1930's Dust Bowl (236.3 vs. 209.9 mm in 1934; NCDC). Average maximum July air temperatures for this location are 32.5 ± 0.14 °C, with record of 46.1 °C set in 1936 (NOAA). Heat wave treatments led to canopy temperatures that spanned this range of temperatures. Daily maximum CT in the high soil moisture plots ranged from 34.2 to 40.8 °C across the four heat treatments, whereas maximum CT for the low soil moisture plots ranged from 42.9 to 53.9 °C.

The heat wave and precipitation treatments impacted both environmental and physiological variables. During the two-week heat wave, under low soil moisture, there was

greater than a two-fold decrease in VWC relative to high soil moisture (Fig. 3.1, inset). While there was no interaction between heat and precipitation treatments on VWC, both CT and VPD showed strong interactions (Table 3.1, Fig. 3.2). Within a given heat treatment, CT for low soil moisture condition was about 7-12 °C warmer than high soil moisture conditions (Fig. 3.2).

The precipitation and heat wave treatments also impacted several physiological variables (A_{net}, g_s and Ψ_{mid}; Table 3.2), however, the responses to the treatments were not consistent across all three physiological variables using this ANOVA-based approach. The precipitation treatment had strong main effects on all variables, while the heat treatment impacted A_{net} and Ψ_{mid}, and there were no interactions between heat and precipitation treatments on any variables (Table 3.2). In addition to direct responses to treatments, we also observed significant effects of species and precipitation x species interaction for both A_{net} and g_s (Table 3.2; Fig. 3.3).

Although we applied the same thermal inputs within a given heat treatment, interactions with the precipitation treatments led to different levels of CT and VPD under high vs. low soil moisture conditions (Fig. 3.2). Under low soil moisture conditions, we observed a strong negative relationship between VWC and CT, while a much weaker, but significant, negative relationship when soil moisture was high (Fig. 3.4). There was an apparent threshold around 15% VWC where the relationship between VWC and CT changed dramatically (Fig. 3.4). Thus, we used a regression analysis to further assess the effects of heat on Ψ_{mid}, Anet, and gs. Similar to the relationship observed for VWC and CT, a non-linear pattern between VWC and Ψ_{mid} was apparent for both C_4 grasses, with a threshold around 15% VWC (Fig. 3.5). It should also be noted that unlike low soil moisture, there was no significant relationship between VWC and Ψ_{mid} for either species (*A. gerardii*, p = 0.667; *S. nutans*, p = 0.608) under high soil moisture (Fig. 3.5).

Given the strong interaction between the precipitation treatments on both environmental and physiological responses, we used stepwise multiple regression with VWC, CT and VPD as independent variables to assess the relative importance of each of these variables for affecting Anet for each species and the high and low soil moisture conditions separately, CT was the strongest predictor for all models, with VWC and VPD being eliminated from all but one model because they exceeded the minimum cutoff ($\alpha < 0.05$). A_{net} had a significant relationship with CT for both species, but only under high soil moisture conditions (Fig. 3.6). *S.* nutans had no relationship between any of the three predictor variables and A_{net} under low soil moisture, while *A. gerardii* had a weak but significant relationship between VPD and A_{net} ($r^2 = 0.06$, $p = 0.042$).

There were no significant effects of heat on fluorescence for either species, regardless of soil moisture level. Under high soil moisture, there were no effects of heat on Φ_{PSII} (*A. gerardii*: p = 0.224; *S. nutans*: p = 0.847) or $F_v'/F_{m'}$ (*A. gerardii*: p = 0.609; *S. nutans*: p = 0.939). In addition, we did not detect any effects of heat on either Φ_{PSII} (*A. gerardii*: p = 0.171; *S. nutans*: p = 0.185) or $F_v'/F_{m'}$ (*A. gerardii*: p = 0.103; *S. nutans*: p = 0.442) under low soil moisture.

Discussion

In this study, we examined ecophysiological responses of two dominant C_4 grasses to an experimentally imposed two-week heat wave in mid-July under two contrasting soil moisture conditions. The objectives of this study were 1) to assess the role of water availability on photosynthetic responses to heat waves of varying magnitude, 2) to identify indirect and/or direct effects of heat driving the responses and 3) to determine if the dominant C_4 grass species can be treated as functional equivalents in their responses to the heat and drought. We found that soil water status mediated the photosynthetic response to heat inputs in both C_4 grasses; heat only

reduced photosynthesis under high soil moisture conditions even though canopy temperatures were lower than under low soil moisture conditions where increasing CT had no measurable effect on leaf level photosynthesis. Our results suggest that severe plant water stress with low soil moisture conditions likely rendered these grasses insensitive to changes in CT since photosynthetic rates were already very low. However there were moderate declines in photosynthesis under high soil moisture conditions, which appear to have been driven by direct heat effects. Finally, with regard to species-specific responses, the dominant grasses differed in the magnitude of their responses, but not direction.

Response of A_{net} to heat under contrasting soil moisture conditions

We were successful in establishing two contrasting soil moisture conditions during the two-week heat wave. During the heat wave, the precipitation treatments resulted in VWC of ~20% for the high and ~10% for the low soil moisture (Fig. 3.1). These soil moisture conditions were above and below what appears to be a critical threshold for water limitation in this ecosystem. Below ~15% VWC we observed both a rapid decline in Ψ_{mid} for both species (Fig. 3.5) and increase in CT (Fig. 3.4), while above this threshold, there is little effect of VWC on both Ψ_{mid} and CT. These results suggest that below this threshold, water stress is reducing evapotranspiration, leading to decreased evaporative cooling and higher canopy temperature, as seen in other studies (De Boeck et al. 2010). Therefore, since the soil moisture conditions resulting from the two precipitation treatments were clearly above and below this threshold, we were able to impose our heat treatment under limiting and non-limiting soil moisture conditions.

Three key environmental variables differed in their responses to the direct effects of heat treatments, as well as the interaction between heat and precipitation treatments. VWC was not

affected by heat, or the interaction between precipitation and heat treatments, while CT and VPD were both significantly impacted (Table 3.1, Fig. 3.2). Differences in water availability due to the precipitation treatments also led to significant main effects on CT (Table 3.1), with high and low soil moisture conditions differing by 8.6 °C under ambient heat (no heat added; Fig. 3.2). As a result, there was a strong interaction between heat and precipitation treatments on CT. As a consequence, the same infrared heat inputs to each heat treatment (low, medium, high) resulted in different CT temperatures dependent upon soil moisture conditions (e.g. CT in high heat treatments: high soil moisture = 40.8 ±0.4 °C and low soil moisture = 53.9 ± 0.9 °C). VPD also showed a similar interaction between heat and precipitation treatments, which was likely driven by the CT response.

Due to these strong interactions between precipitation and heat wave treatments for both CT and VPD, we decided to use a stepwise regression based-approach in which soil moisture conditions were analyzed independently for each species. Our goal was to determine whether A_{net} was more strongly affected by CT, VWC or VPD, given that the former would be indicative of direct effects of heat whereas the latter two would be indicative of indirect effects being important. For both species, variation in A_{net} was not related to CT, VWC or VPD under low soil moisture conditions. In contrast, both species had significant negative relationships between CT and A_{net} under non-limiting soil moisture conditions. Our experimental design aimed to reduced soil moisture variability within a given precipitation treatment, so it is not too surprising that when we analyzed high and low soil moisture conditions separately, VWC failed to have a significant relationship with A_{net}. These results suggest water stress mediates the response to heat; under the water-limited conditions, A_{net} does not respond to increasing CT, while under non-water-limiting conditions, A_{net} decreases with increasing CT.

Direct vs. indirect effects of heat

The negative effect of increasing CT on A_{net} under non-limiting soil moisture conditions could have been driven by direct or indirect effects of heat, or a combination. One key indirect effect of heat waves on ecosystems is inducing water stress through increased evapotranspiration rates (Milbau et al. 2005, Marchand et al. 2006, Arnone et al. 2008, De Boeck et al. 2011). This indirect heat effect would be observed in factors governing the net water balance of the plant: water source (soil moisture) and water sink (VPD). We eliminated this first indirect effect of heat on the soil moisture as evidenced by the lack of significant main effects of heat or interaction between heat and precipitation on VWC (Table 3.1, Fig. 3.2). Isolating the effects of VPD on plant water status was more challenging because CT and VPD have a strong linear relationship ($r^2 = 0.88$, $p < 0.001$). Both species had a weak, negative relationship between VPD and Ψ_{mid} under high soil moisture conditions (*A. gerardii*: $r^2 = 0.20$, $p < 0.001$; *S. nutans*: $r^2 = 0.10$, $p = 0.005$). However, when we examined whether this decrease in leaf water potential affected stomatal conductance, we found no relationship between Ψ_{mid} and g_s for either species (*A. gerardii*: $p = 0.654$; *S. nutans*: $p = 0.335$), suggesting that reductions in Ψ_{mid} were not related to stomatal closure. To further support the lack of indirect effects of heat, the stepwise regression model also removed VWC and VPD as significant predictors of A_{net}, in favor of CT. These results suggest that the direct effect of heat was the variable driving declines in A_{net} under high soil moisture.

There are several possible direct effects of heat related to non-stomatal limitations that could result in declines in A_{net} under the non-limiting soil moisture conditions. First, heat can directly damage photosystems II in the photosynthetic apparatus, which is particularly heat sensitive (Wahid et al. 2007). To examine this heat effect, we measured light (Φ_{PSII}) and dark

54

(F_v'/F_m') adapted fluorescence on the last day of the heat wave, when the greatest cumulative effects of heat stress should be apparent. Under the high soil moisture conditions, we saw no effects of heat on either Φ_{PSII} or F_v'/F_m', suggesting that heat did not impair the quantum yield or maximum quantum yield of photosystems II. Secondly, heat can effect enzymatic kinetics and thereby alter metabolic processes. Enzymatic activities increase with temperature, and therefore dark respiration should increase with temperature and cause a decrease in A_{net} if gross assimilation rates do not have a corresponding increase (Salvucci and Crafts-brandner 2004). Additionally, reductions in A_{net} have been correlated with a decrease in activation state of Rubisco (Salvucci and Crafts-brandner 2004). We hypothesize that reductions in A_{net} under high soil moisture conditions were a result of heat induced respiration increases.

Unlike high soil moisture conditions, photosynthesis in these C_4 grasses was insensitive to increasing heat under low soil moisture conditions. C_4 photosynthesis is sensitive to declines in soil moisture, with decreased leaf water potential resulting in reduced stomatal conductance, limiting assimilation rates (Ghannoum 2009). It is likely that stomatal limitations caused the lack of heat response in low soil moisture. For both species, low soil moisture conditions had strong negative effects on A_{net} (*A. gerardii*: -54.1%, *S. nutans*: -82.3%) and g_s (*A. gerardii*: -50.5%, *S. nutans*: -60.7%; Fig. 3.3). Given the low VWC and Ψ_{mid}, water was limiting, and thus low stomatal conductance was restricting gas exchange. In addition, the high water stress prevented evaporative cooling, and therefore plants were exposed to higher temperatures than under the high soil moisture conditions. Despite these extreme temperatures, we did not detect any effects of heat on either Φ_{PSII} or F_v'/F_m', suggesting that combined heat and water stress did not damage the cellular machinery in photosystems II. This high resistance to heat damage was surprising given the temperature exceed the thermal optima for *A. gerardii* (33 – 41 °C; Knapp, 1985). It is

possible that the plants were acclimated to these higher temperatures, due to the drought-induced warming of the canopy in the period preceding the heat wave.

Comparing the responses of the dominant C$_4$ grasses

Overall, the two dominant C$_4$ grasses did not differ considerably in their responses to heat and precipitation treatments, although there was some evidence for differential sensitivities. Both species shared similar patterns of responses to the treatments. For example, both species had similar Ψ_{mid} thresholds, both only responded to heat under non-limiting soil moisture, and both did not show any damage to the photosynthetic machinery with heat. Previous work suggests that the two species differ in their responses to heat and water stress, with *A. gerardii* more sensitive to heat and *S. nutans* more sensitive to drought (Silletti and Knapp 2002, Swemmer et al. 2006, Nippert et al. 2009). However in this study, *S. nutans* was more responsive to both heat and precipitation treatments than *A. gerardii*. First, A$_{net}$ decreased more between high and low soil moisture conditions in *S. nutans* than in *A. gerardii* (Fig. 3.6). Secondly, under high soil moisture conditions, we observed a negative effect of CT on A$_{net}$ for both species, but the slope of this relationship for *S. nutans* was greater than in *A. gerardii* (-1.9 vs. -1.1, respectively), suggesting that *S. nutans* was more responsive to increasing temperatures when soil moisture was non-limiting. Since we observed these co-dominant grasses have similar patterns of responses to heat and drought but also had differential sensitivities to these treatments, they can be treated as functional equivalents with regard to direction of response to these climate extremes, but not the magnitude.

Summary

In this study, we determined that soil water availability dominates the photosynthetic response of two C_4 grass species to an experimental heat wave. When soil water was low, severe water stress precluded heat from reducing already low photosynthetic rates, which were limited by stomatal conductance. However, when we eliminated limiting soil moisture conditions as a co-varying factor during the heat wave, photosynthesis was sensitive to increasing CT. Soil moisture also governed the maximum temperatures the leaves experienced during the simulated heat wave; transpirational cooling prevented canopy temperatures in high soil moisture conditions from reaching the extreme levels experienced in low soil moisture conditions. Overall these results suggest that heat waves will have little to no direct or concurrent impacts on photosynthesis under drought, instead only directly effecting photosynthetic rates under non-limiting soil moisture levels. However, prolonged high temperatures will likely cause water stress through enhanced evapotranspiration and concomitant soil drying, driving the system to severe water stress, where heat will no longer affect photosynthesis.

Tables

Table 3.1 Summary of F and p-values from a repeated measures mixed model ANOVA for volumetric water content (VWC), canopy temperature (CT) and vapor pressure defect (VPD) during the two-week heat wave. Bold values indicate significance ($p<0.05$).

| Effect	VWC		CT		VPD	
	F	p	F	p	F	p
Precipitation	**74.3**	**0.013**	**27.4**	**0.035**	**240.8**	**0.005**
Heat	1.3	0.288	**95.6**	**<0.001**	**45.3**	**<0.001**
Precipitation*Heat	0.0	0.996	**5.7**	**0.039**	**9.0**	**<0.001**
Date	**40.1**	**<0.001**	**18.2**	**<0.001**	**7.3**	**0.000**
Precipitation*Date	**27.9**	**<0.001**	**3.4**	**0.020**	**3.9**	**0.011**
Heat*Date	0.8	0.656	0.3	0.976	0.4	0.957
Precipitation*Heat*Date	0.5	0.839	0.5	0.883	0.6	0.832

Table 3.2 Summary of F and p-values from a repeat measures mixed model ANOVA for net photosynthesis (A_{net}), stomatal conductance (g_s), and mid-day leaf water potential (Ψ_{mid}) during the two-week heat wave. Bold values indicate significance ($p<0.05$).

Effect	A_{net} F	A_{net} p	g_s F	g_s p	Ψ_{mid} F	Ψ_{mid} p
Precipitation	**86.**	**<0.001**	**86.1**	**<0.001**	**72.2**	**0.014**
Heat	2.9	0.110	0.5	0.727	**8.4**	**0.014**
Precipitation*Heat	1.9	0.213	2.1	0.183	3.2	0.108
Date	**4.0**	**0.009**	2.2	0.088	**14.6**	**<0.001**
Precipitation*Date	0.5	0.686	2.0	0.116	**13.0**	**<0.001**
Heat*Date	**2.0**	**0.044**	0.9	0.565	1.1	0.334
Precipitation*Heat*Date	1.5	0.141	0.9	0.539	1.1	0.382
Species	**5.4**	**0.024**	**7.5**	**0.008**	1.9	0.172
Precipitation*Species	**27.**	**<0.001**	**4.7**	**0.035**	0.4	0.526
Heat*Species	0.9	0.438	0.1	0.939	0.7	0.549
Precipitation*Heat*Species	2.3	0.088	0.2	0.918	0.8	0.512
Species*Date	2.1	0.107	0.7	0.531	0.6	0.632
Precipitation*Species*Date	**5.1**	**0.002**	1.6	0.189	0.2	0.903
Heat*Species*Date	1.0	0.458	0.6	0.795	0.7	0.694
Precip*Heat*Species*Date	1.1	0.391	0.9	0.534	0.9	0.560
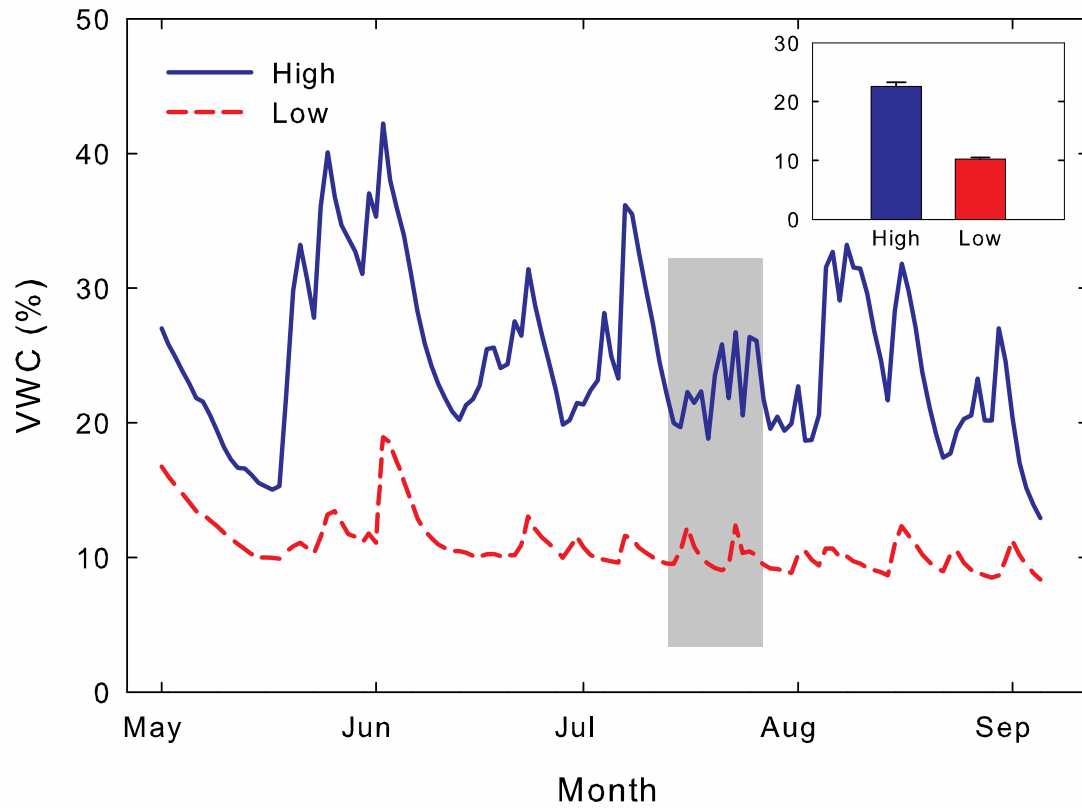
Figure 3.1 Volumetric water content (VWC) during the 2011 growing season for high and low soil moisture conditions. Grey box indicates when the two-week heat treatments were imposed. Inset – mean VWC during the two-week heat wave for high and low soil moisture conditions (error bars ±1 SE).
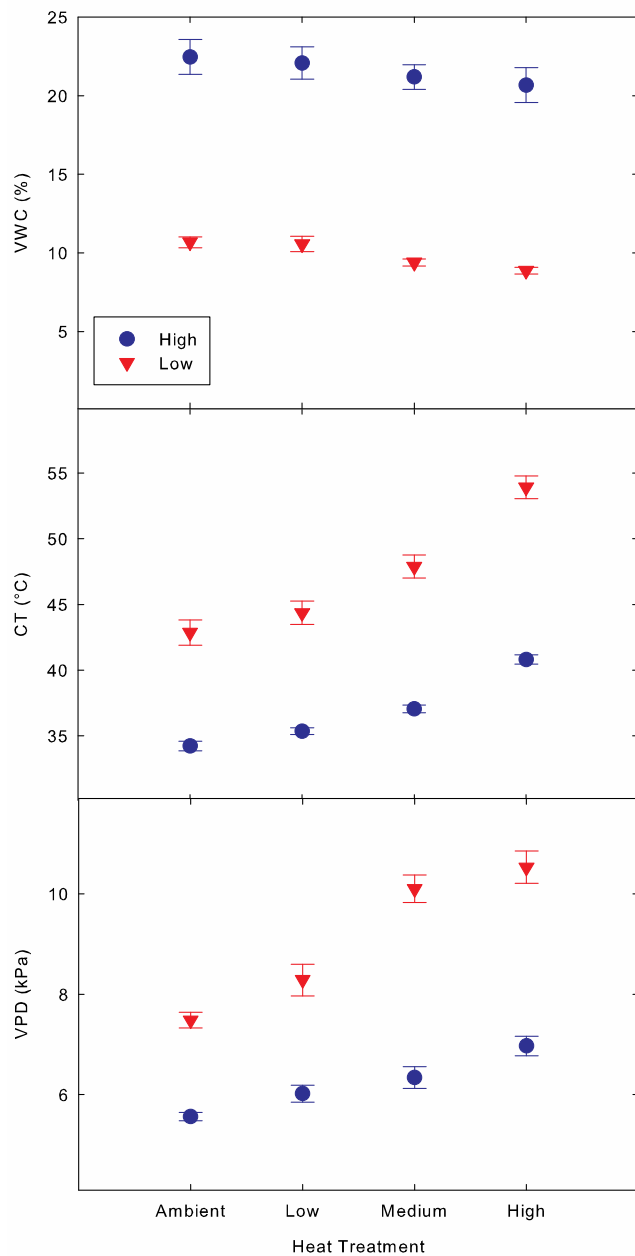
Figure 3.2 Mean values (±1 SE) for each soil moisture condition (high and low) and heat treatment combination for volumetric water content (VWC), canopy temperature (CT) and vapor pressure defect (VPD) during the two-week heat wave. Values were calculated over the four sampling campaigns for n=5 plots/treatment combination.
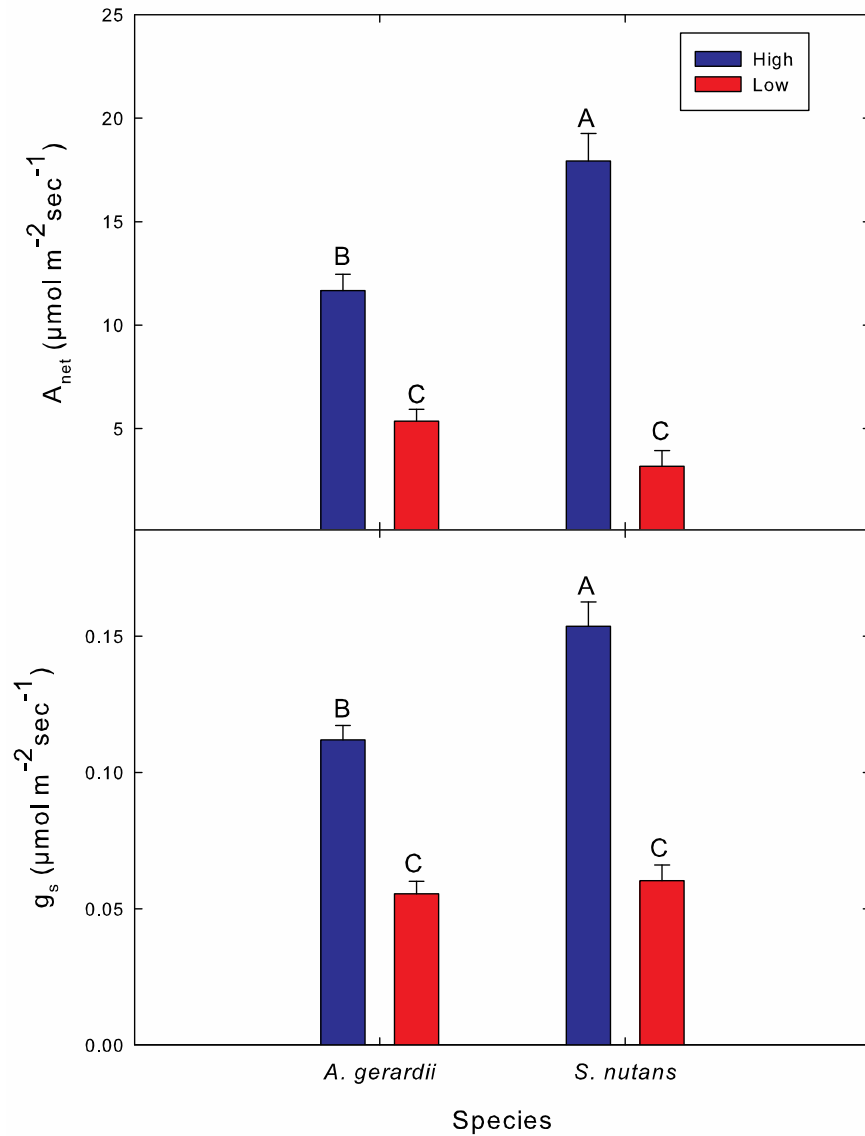
Figure 3.3 Net photosynthesis (A_{net}) and stomatal conductance (g_s) under high and low soil moisture conditions for the co-dominant C_4 grass species. Letters indicate significant differences from pairwise comparisons ($p < 0.005$).
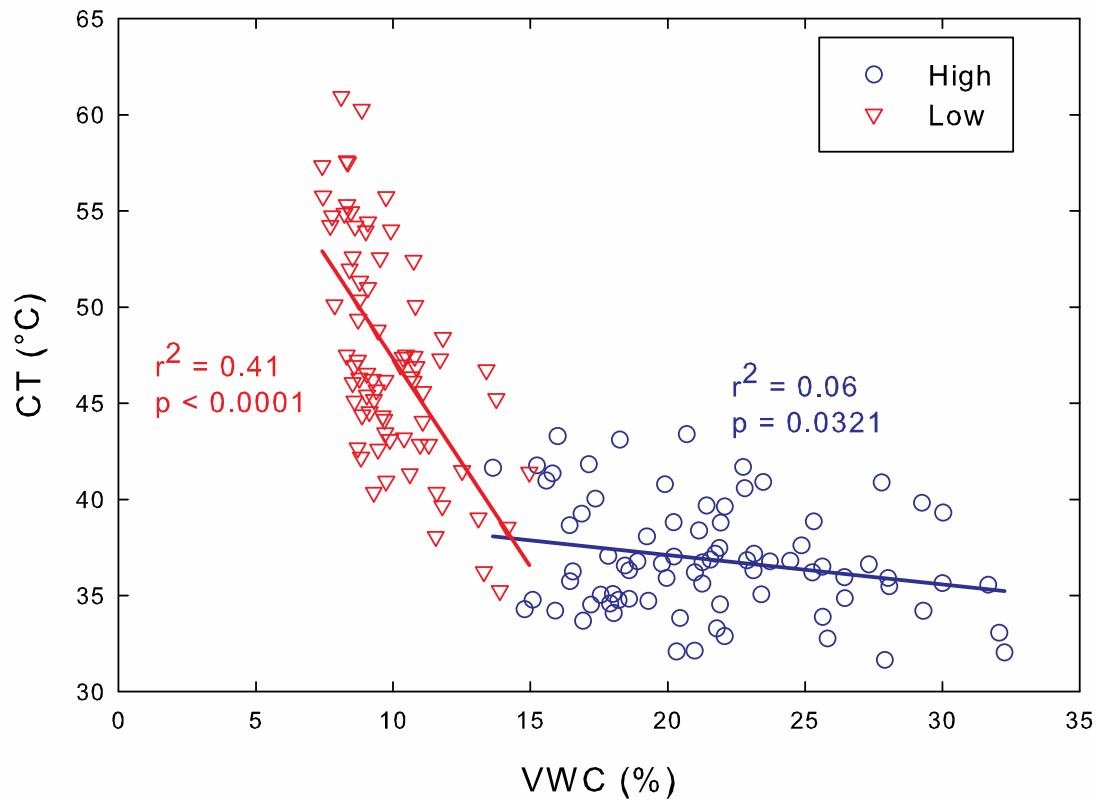
Figure 3.4 Relationship between volumetric water content (VWC) and canopy temperature (CT) for during the two-week heat wave for high and low soil moisture conditions (includes all heat treatments). Regressions were performed on each soil moisture condition, with r^2 and p-values located next to corresponding regression line.
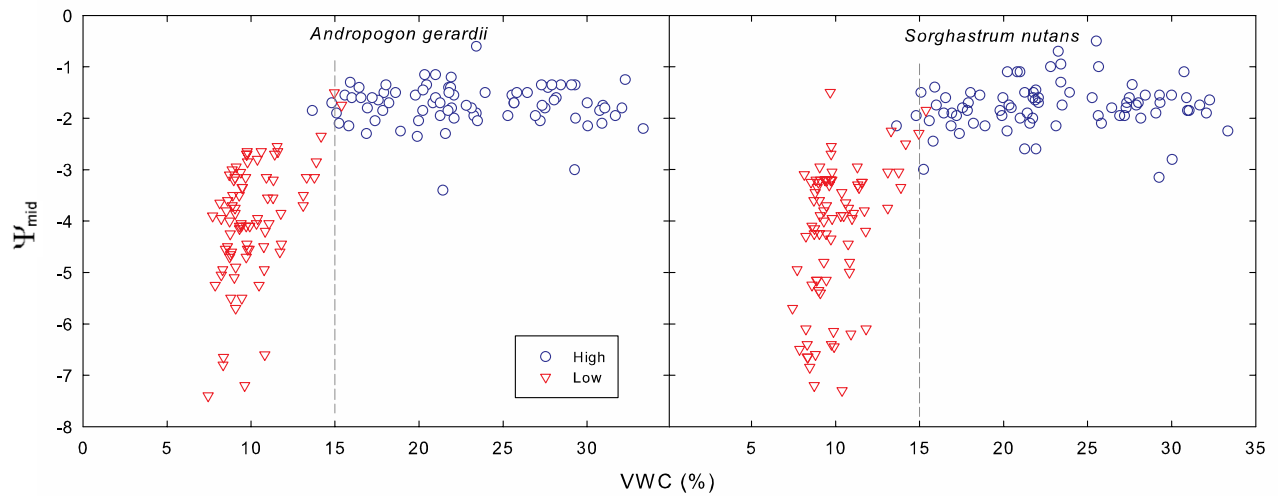
Figure 3.5 Volumetric water content (VWC) vs. mid-day leaf water potential (Ψ_{mid}) during the two-week heat wave for the co-dominant grasses under high and low soil moisture conditions. Dashed line highlights 15% VWC, which appears to be a critical soil moisture threshold for both species.
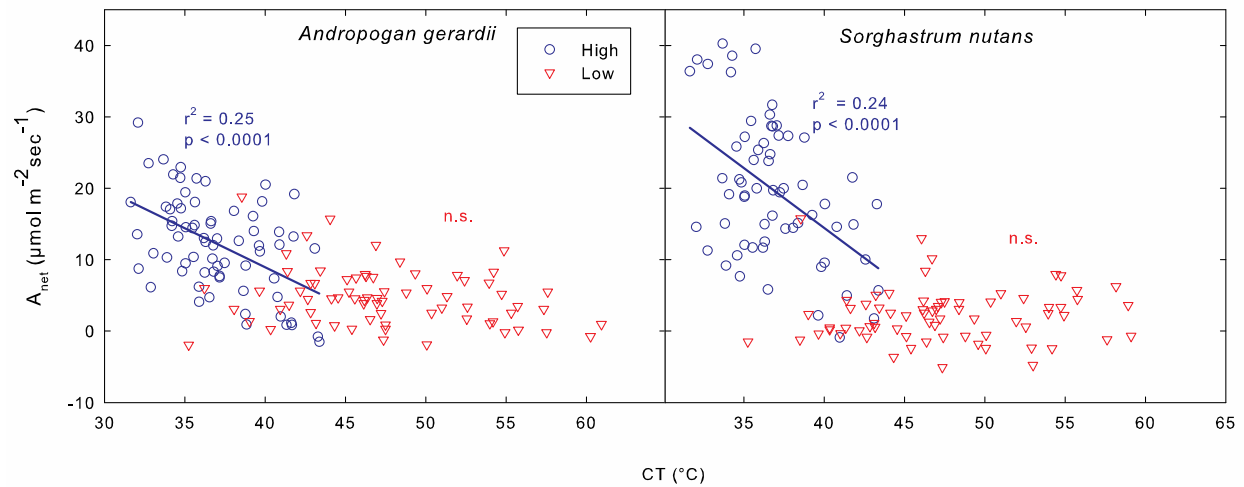
Figure 3.6 For each co-dominant grass species, the relationship between net photosynthesis (A-net) and canopy temperature (CT) for high and low soil moisture conditions. Regressions were performed on each species x soil moisture combination, and if significant (p<0.005) regression lines with corresponding r^2 and p-values (n.s. = non-significant).

References

Arnone, J. A., P. S. J. Verburg, D. W. Johnson, J. D. Larsen, R. L. Jasoni, A. J. Lucchesi, C. M. Batts, C. von Nagy, W. G. Coulombe, D. E. Schorran, P. E. Buck, B. H. Braswell, J. S. Coleman, R. A. Sherry, L. L. Wallace, Y. Luo, and D. S. Schimel. 2008. Prolonged suppression of ecosystem carbon dioxide uptake after an anomalously warm year. Nature 455:383–6.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. Global Change Biology 16:1992–2000.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. The New phytologist 189:806–17.

Breshears, D. D., N. S. Cobb, P. M. Rich, K. P. Price, C. D. Allen, R. G. Balice, W. H. Romme, J. H. Kastens, M. L. Floyd, J. Belnap, J. J. Anderson, O. B. Myers, and C. W. Meyer. 2005. Regional vegetation die-off in response to global-change-type drought. Proceedings of the National Academy of Sciences of the United States of America 102:15144–15148.

Burnette, D. J., and D. W. Stahle. 2012. Historical perspective on the dust bowl drought in the central United States. Climatic Change 116:479–494.

Burnette, D. J., D. W. Stahle, and C. J. Mock. 2010. Daily-Mean Temperature Reconstructed for Kansas from Early Instrumental and Modern Observations. Journal of Climate 23:1308–1333.

Ciais, P., M. Reichstein, N. Viovy, and A. Granier. 2005. Europe-wide reduction in primary productivity caused by the heat and drought in 2003. Nature 437:529–533.

Easterling, D. R., G. A. Meehl, C. Parmesan, S. A. Changnon, T. R. Karl, and L. O. Mearns. 2000. Climate extremes: observations, modeling, and impacts. Science 289:2068–2074.

Fay, P., J. Carlisle, A. Knapp, J. Blair, and S. Collins. 2000. Altering rainfall timing and quantity in a mesic grassland ecosystem: Design and performance of rainfall manipulation shelters. Ecosystems 3:308–319.

Ghannoum, O. 2009. C4 photosynthesis and water stress. Annals of Botany 103:635–644.

Grime, J. 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. Journal of Ecology 86:902–910.

Gutschick, V. P., and H. BassiriRad. 2003. Extreme events as shaping physiology, ecology, and evolution of plants: toward a unified definition and evaluation of their consequences. New Phytologist 160:21–42.

Houghton, R. A. 2007. Balancing the Global Carbon Budget. Annual Review of Earth and Planetary Sciences 35:313–347.

IPCC. 2012. Managing the Risks of Extreme Events and Disasters to Advance Climate Change Adaptation. (C. Field, V. Barros, T. Stocker, D. Qin, D. Dokken, K. Ebi, M. Mastrandrea, K. Mach, G. Plattner, S. Allen, M. Tignor, and M. PM, Eds.) A Special Report of Working Groups I and II of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, England.

Jentsch, A., J. Kreyling, and C. Beierkuhnlein. 2007. A new generation of climate-change experiments: events, not trends. Frontiers in Ecology and the Environment 5:365–374.

Knapp, A. 1985. Effect of fire and drought on the ecophysiology of andropogon gerardii and panicum virgatum in a tallgrass prairie. Ecology 66:1309–1320.

Knapp, A. K., J. M. Briggs, D. C. Hartnett, and S. L. Collins. 1998. Grassland Dynamics: Long-Term Ecological Reserach in Tallgrass Prairie. Oxford University Press, Inc., New York.

Knapp, A. K., J. M. Briggs, and J. K. Koelliker. 2001. Frequency and extent of water limitation to primary production in a mesic temperate grassland. Ecosystems 4:19–28.

Marchand, F. L., M. Verlinden, F. Kockelbergh, and B. J. Graae. 2006. Disentangling effects of an experimentally imposed extreme temperature event and naturally associated. Functional Ecology:917–928.

Milbau, A., L. Scheerlinck, D. Reheul, B. De Cauwer, and I. Nijs. 2005. Ecophysiological and morphological parameters related to survival in grass species exposed to an extreme climatic event:500–512.

Nippert, J. B., P. a. Fay, J. D. Carlisle, A. K. Knapp, and M. D. Smith. 2009. Ecophysiological responses of two dominant grasses to altered temperature and precipitation regimes. Acta Oecologica 35:400–408.

Nippert, J. B., A. K. Knapp, and J. M. Briggs. 2006. Intra-annual rainfall variability and grassland productivity: can the past predict the future? Plant Ecology 184:65–74.

Reichstein, M., M. Bahn, P. Ciais, D. Frank, M. D. Mahecha, S. I. Seneviratne, J. Zscheischler, C. Beer, N. Buchmann, D. C. Frank, D. Papale, A. Rammig, P. Smith, K. Thonicke, M. van der Velde, S. Vicca, A. Walz, and M. Wattenbach. 2013. Climate extremes and the carbon cycle. Nature 500:287–295.

Reyer, C. P. O., S. Leuzinger, A. Rammig, A. Wolf, R. P. Bartholomeus, A. Bonfante, F. de Lorenzi, M. Dury, P. Gloning, R. Abou Jaoudé, T. Klein, T. M. Kuster, M. Martins, G. Niedrist, M. Riccardi, G. Wohlfahrt, P. de Angelis, G. de Dato, L. François, A. Menzel, and M. Pereira. 2013. A plant's perspective of extremes: terrestrial plant responses to changing climatic variability. Global change biology 19:75–89.

Sala, O., W. Parton, L. Joyce, and W. Lauenroth. 1988. Primary Production of the Central Grassland Region of the United States. Ecology 69:40–45.

Salvucci, M. E., and S. J. Crafts-brandner. 2004. Minireview Inhibition of photosynthesis by heat stress□: the activation state of Rubisco as a limiting factor in photosynthesis:179–186.

Silletti, A., and A. Knapp. 2002. Long-term responses of the grassland co-dominants Andropogon gerardii and Sorghastrum nutans to changes in climate and management. Plant Ecology 163:15–22.

Smith, M. D. 2011. An ecological perspective on extreme climatic events: a synthetic definition and framework to guide future research. Journal of Ecology 99:656–663.

Smith, M., and A. Knapp. 2003. Dominant species maintain ecosystem function function with non-random species loss. Ecology Letters 6:509–517.

Swemmer, A., A. Knapp, and M. Smith. 2006. Growth responses of two dominant C4 grass species to altered water availability. International Journal of Plant Sciences 167:1001–1010.

Tilman, D., and J. Downing. 1994. Biodiversity and stability in grasslands. Nature 367:363–365.

Wahid, A., S. Gelani, M. Ashraf, and M. Foolad. 2007. Heat tolerance in plants: an overview. Environmental and Experimental Botany 61:199–223.

Weaver, J. 1954. North American prairie. Johnsen Publishing Company, Lincoln, Nebraska.

Whittaker, R. H. 1965. Dominance and Diversity in Land Plant Communities: Numerical relations of species express the importance of competition in community function and evolution. Science 147:250–260.

Williams, J. W., S. T. Jackson, and J. E. Kutzbach. 2007. Projected distributions of novel and disappearing climates by 2100 AD. Proceedings of the National Academy of Sciences of the United States of America 104:5738–5742.

Woodhouse, C., and J. Overpeck. 1998. 2000 years of drought variability in the central United States. Bulletin of the American Meteorological Society 79:2693–2714.

Yahdjian, L., and O. Sala. 2002. A rainout shelter design for intercepting different amounts of rainfall. Oecologia 133:95–101.

Chapter 4

RESISTANCE AND RESILIENCE OF A GRASSLAND ECOSYSTEM TO CLIMATE EXTREMES

Overview

Climate change forecasts of more frequent climate extremes suggest that such events will become increasingly important drivers of future ecosystem dynamics and function. Because the rarity and unpredictability of naturally occurring climate extremes limits assessment of their ecological impacts, we experimentally imposed extreme drought and a midsummer heat wave over two years in a central US grassland. While the ecosystem was resistant to heat waves, it was not resistant to extreme drought, which reduced aboveground net primary productivity (ANPP) below the lowest level measured in this grassland for almost thirty years. This extreme reduction in ecosystem function was a consequence of reduced productivity of both C_4 grasses and C_3 forbs. However, the dominant forb was negatively impacted by the drought more than the dominant grass, and this led to a reordering of species abundances within the plant community. Although this change in community composition persisted post-drought, ANPP recovered completely the year after drought due to rapid demographic responses by the dominant grass, compensating for loss of the dominant forb. Overall, our results show that an extreme reduction in ecosystem function attributable to an extreme climate event (e.g., low resistance) does not preclude rapid ecosystem recovery. Given that dominance by a few species is characteristic of most ecosystems, knowledge of the traits of these species and their responses to climate extremes will be key for predicting future ecosystem dynamics and function.

Introduction

Global change threatens ecosystems worldwide through chronic alterations in climate (temperature and precipitation) and resources (increasing atmospheric CO_2 and nitrogen-deposition), as well as by increasing the frequency and intensity of climate extremes, such as drought, floods, and heat waves (Easterling et al. 2000, IPPC 2007, Smith et al. 2009, IPCC 2012). Despite prominent examples of the severe impacts of climate extremes (e.g., the central US Dust Bowl of the 1930s, the 2003 European heat wave and the 2012 US drought; Weaver 1954, Ciais et al. 2005, Lal et al. 2012), global change research has been dominated by the study of chronic environmental changes rather than discrete climate extremes (Jentsch et al. 2007, Smith 2011). Indeed, assessing the ecological consequences of climate extremes, as well as the mechanisms determining ecosystem response and recovery, remains a key challenge for ecologists today (Smith 2011).

Ecological responses to climate extremes are highly variable (Smith 2011), ranging from little to no impact (Van Peer et al. 2004, Kreyling et al. 2008, Jentsch et al. 2011) to major effects on ecosystem structure and function with prolonged recovery (Weaver 1954, White et al. 2000, Haddad et al. 2002, Ciais et al. 2005, Breshears et al. 2005). Such variability in ecological resistance (capacity to withstand change) and resilience (capacity for recovery of function; Pimm 1984, Tilman and Downing 1994) may be due to differences in ecosystem attributes, as well as a result of the magnitude, duration and timing of the climate extreme. Thus, it is critical that both the driver (climate event) and the ecosystem response (ecological effects) are evaluated with respect to their extremity.

Smith (2011) defined an extreme climatic event (ECE) as "an episode or occurrence in which a statistically rare or unusual climatic period alters ecosystem structure and/or function

70

well outside the bounds of what is considered typical or normal variability." Determining if an ECE has occurred, therefore, requires both long-term climatic and ecological data, with the former available more often than the latter. In addition, Smith (2011) provided a mechanistic framework for assessing ecological responses to climate extremes. In brief, this framework depicts how impacts of climate extremes at the species level have the potential to have large impacts on ecosystem function, depending on the role and abundance of the species impacted. For example, a period of climatic extremity that results in mortality or significant loss in fitness of dominant species (i.e., crossing of an ecological response threshold) may lead to community-level responses that include species re-ordering and compositional changes. Such alterations in community composition are predicted to have the greatest impact on ecosystem function, leading to an extreme ecological response (Smith 2011).

Much of our current understanding of the ecological effects of climate extremes is based on opportunistic studies of naturally occurring events (Weaver 1954, Breshears et al. 2005, Reichstein et al. 2007, Lal et al. 2012), with attendant difficulties in the attribution of specific climate drivers to ecosystem response and recovery (Déry and Wood 2005, De Boeck et al. 2010, Smith 2011). Experimental approaches are better suited to study climate extremes because climate drivers can be directly attributed to ecological responses (Reyer et al. 2013). Indeed, manipulative experiments explicitly linked to historical climatic records and interpreted in the context of long-term ecological data provide the best opportunity to advance our understanding of climate extremes (Smith 2011, Reyer et al. 2013). Finally, a wide range of biotic responses across several hierarchal levels must be measured during the event and the subsequent recovery period in order to identify the mechanisms governing ecosystem resistance and resilience and the potential for ECEs.

Here we directly test the ECE framework in a central US grassland where we imposed extreme drought and a two-week heat wave in a fully factorial experiment over two years, and monitored subsequent recovery one year post-drought. In 2010 and 2011, we imposed a drought treatment by passively reducing growing season precipitation by 66% using rainout shelters, while control treatments received ambient rainfall plus supplemental irrigation to alleviate water stress. Nested within the drought and control treatments, we imposed a two-week heat wave midsummer with four levels of infrared radiation inputs. Heat waves and drought typically co-occur in nature, thus our experimental design allowed us to examine the independent and interactive effects of extreme drought and heat on ecosystem structure and function during the two-year extreme period and for one-year post-drought.

Our objectives were to test whether an ECE was imposed by placing the drought and heat wave treatments and the ecosystem response observed within the long-term record, and to assess proposed mechanisms underpinning ecosystem response (resistance) and recovery (resilience) to extreme drought and heat, and thus explicitly link alterations in ecosystem function and plant community composition to the imposed extremes. We hypothesized that: (1) ecosystem function (aboveground net primary production, ANPP) would be less resistant to drought and heat waves combined than either factor alone, with the independent effects of drought greater than heat; (2) the response of dominant species would govern the extremity of the ecological responses, with the expectation that if the dominant species were negatively impacted then large ecosystem responses would be observed; and (3) recovery from an ECE would be prolonged if community composition and diversity were substantially altered by these climate extremes.

72

Methods

Study site

This study was conducted at the Konza Prairie Biological Station, a 3487 ha native

tallgrass prairie in northeastern Kansas, USA (39°05'N, 96°35'W). The region is characterized

by a temperate mid-continental climate, with average annual precipitation of 835 mm and mean

July air temperature of 27 °C (Knapp et al. 1998). In 2010, we established the Climate Extremes

Experiment in an intact, native Kansas grassland, with deep (>1 m) silty clay loam soils, which

was burned annually (as is typical for this region; see Fay et al. 2000 for more site details). This

site is a good representative for the tallgrass prairie region because it is dominated by the two

most abundant plant functional types (PFT's) of this ecosystem; a C_4 perennial, rhizomatous

grass, *Andropogon gerardii,* which dominates much of the historic range of tallgrass prairie

(Brown 1993) and a C_3 perennial, clonal forb, *Solidago canadensis*, which is the locally most

abundant species (unlike the grasses, no single forb species dominates throughout this

ecosystem).

Treatments

During the 2010 and 2011 growing seasons (April 1st – August 30th) we imposed the

drought and control (ambient precipitation) treatments using four passive rainout shelters (based

on a design by Yahdjian and Sala 2002) established over native grassland communities (Fig.

4.1a). Across the central US grassland region, the majority of annual precipitation occurs during

the growing season and extended drought periods often occur during this time, and thus a

reduction in rainfall during this period is highly relevant (Knapp et al. 1998). For the drought

treatment, we reduced each growing season rainfall event by ~66% using two 6 x 24 m cold

frame greenhouse structures (Stuppy, Inc., Kansas City, MO, USA) partially covered (75%; Fig. 4.1a) with strips of Dynaglas Plus® clear polycarbonate plastic (PALRAM Industries LTD., Kutztown, PA, USA). For the control (ambient precipitation) treatment, two shelters were covered with deer netting (TENAX Manufacturing Alabama, USA) to reduce photosynthetically active radiation by about 10% (equivalent to light reduction in drought shelters), but allow all ambient rainfall to pass through (Fig. 4.1a). Our goal was to have non-limiting soil moisture levels in control plots to contrast with low soil moisture in the drought plots, which necessitated adding supplemental water during extended dry periods to the control treatments. This was not required in 2010, but in 2011 control treatments received supplemental irrigation (~12.7 mm event) when soil moisture dropped below a critical threshold for plant water stress (~20% volumetric water content of the top 15 cm; Hoover Chapter 2). The soil within the 6 x 24 footprint of each shelter was hydrologically isolated by trenching to a depth of 1 m, lining it with two layers of 6 mil plastic belowground, and placing metal flashing around the perimeter aboveground to prevent surface and subsurface water flow into the plots.

Nested within each rainout shelter we established ten 2 x 2 m plots that were randomly assigned to one of four heat wave treatments (ambient, low, medium and high). Heat was added for two weeks in mid-summer (21-July to 03-August-2010; 13-July to 26-July-2011) using infrared heat lamps within passive warming chambers (Fig. 4.1a). The 2 x 2 m passive warming chambers were constructed of PVC frames, with 1 m high walls covered with 6-mil transparent polyethylene and clear corrugated polycarbonate roofs (Dynaglas Plus®). These chambers were designed to minimize convective cooling. Ventilation was maintained by placing chambers 0.5 m above the ground surface with an adjustable gap between the roof and sidewalls to decrease or increase airflow if needed. Four heat input levels were imposed with 2000 W infrared heat lamps

(HS/MRM 2420, Kalglo Electronics, Inc., Bethlehem, PA, USA) as follows: control = no lamp, low heat = one lamp at ½ power (+250 W/m^2 output), medium heat = one lamp at full power (+500 W/m^2), and high heat = two lamps at full power (+1000 W/m^2).

In the recovery year (2012), no drought or heat wave treatments were imposed and all plots received ambient rainfall plus supplemental irrigation. All plots were watered weekly by hand if rainfall totals during that week were less than long-term averages (in which case the deficit was added). This allowed us to maintain precipitation inputs near the long-term monthly mean totals.

Abiotic data

Daily precipitation data were obtained from a nearby (~250 m) US Climate Reference Network station (www.ncdc.noaa.gov/crn/, KS Manhattan 6 SSW) and combined with supplemental irrigation and excluded rainfall amounts to calculate precipitation treatment totals each year of the study (Table A4.1). Soil moisture and canopy temperature were continuously monitored in each plot to evaluate the effectiveness of drought and heat wave treatments. Soil moisture was measured in the center of each plot at a depth of 0-15 cm with 30 cm time-domain reflectometery probes (Model CS616, Campbell Scientific, Inc., Logan, UT, USA) inserted at a 45° angle. Canopy temperature was measured using infrared thermometers (Model SI-111, Apogee Instruments, Inc., Logan, UT, USA) mounted in the SE corner of each plot at a height of 1.5 m. Data from both sensors were recorded every 30-min on a CR10X Datalogger (Campbell Scientific, Inc., Logan, UT, USA).

Biotic data

ANPP was estimated at the end of each growing season (first week in September) by harvesting all aboveground plant material in three 0.1 m^2 quadrats located within each 4 m^2 plot. Harvesting at this time of year has been shown to capture peak biomass, and since the site was annually burned (i.e., no previous years dead material), this provides a reliable estimate of ANPP (Knapp et al. 2007). For each year, the locations of quadrats were different to prevent resampling of the same quadrat. Samples were field sorted by growth form, oven dried at 60°C for 48 hours, and then weighed to the nearest 0.1 g. Individual stems (tillers) were censused within a permanent 0.1 m^2 quadrat located within each plot to estimate stem density per m^2. Community composition was assessed twice each year (early June, late August) in one permanent 1 m^2 subplot per 4 m^2 plot by visually estimating percentage aerial cover for each species separately. Maximum cover values of each species were used to determine relative cover of each species and to calculate species richness, Shannon's diversity (H′) and evenness.

Long-term data

Long-term (1900-2012) precipitation and air temperature data for Manhattan, KS (~10 km from the experimental site at the Konza Prairie Biological Station, KPBS) were obtained from the National Climate Data Center's Global Historical Climatology Network (www.ncdc.noaa.gov/oa/climate/ghcn-daily/index.php). These data were used to calculate mean growing season precipitation and temperatures and to estimate their probability density functions (pdfs). Long-term (27-year) aboveground net primary production (ANPP) data from a site comparable to our study system (annually burned lowland) were obtained from the Konza Prairie

Biological Station (www.konza.ksu.edu/knz/; data set PAB01). These data were used to calculate mean annual ANPP and to estimate ANPP pdf.

Statistical analyses

Precipitation totals and ANPP means for the control and drought treatments were compared to estimated pdfs of long-term growing season precipitation and ANPP for the site. Values were considered extreme if they exceeded the 5th percentiles of the pdfs. The experiment was a randomized block split-plot design, and we analyzed biotic responses during the drought (2010-2011) using a repeated measure mixed-model ANOVA with the heat wave treatments (random effect) nested within the drought treatments and year as a repeated effect. Biotic responses during the recovery year (2012 ANPP and stem densities) were analyzed with a mixed-model ANOVA with the heat wave treatments nested within the drought treatments. For each year, we used non-metric multi-dimensional scaling (NMDS) followed by analysis of similarity (ANOSIM) using relative abundance data to assess community-level divergence between treatments (divergence criteria was determined at $p < 0.05$). To assess how much each individual species contributed to the treatment divergence in each year, we calculated similarity percentages using SIMPER analysis based on a euclidian distance matrix for each year separately. All mixed-model ANOVAs were conducted in SAS (v9.3) and all community composition analyses were conducted using Primer v6.

Results

Historical context of precipitation and heat treatments

Within the context of a 112-year climate record for this location, the magnitude of the precipitation and temperature treatments ranged from near average to extreme during the first two years of the experiment (Fig. 4.1b,c). Growing season precipitation inputs were reduced in 2010 and 2011 below the 10th and 5th percentiles of historic amounts, respectively (Fig. 4.1b). These amounts contrasted sharply with the slightly above average rainfall inputs to control plots during these years (Fig. 4.1b; Table A4.1). The cumulative two-year period of growing season precipitation input to the drought plots was 533.4 mm, or 28.0 mm less than the driest consecutive two-year period (1933-34) during the historic 1930s Dust Bowl drought. The drought treatment reduced mean growing season soil moisture by 43 and 56% in 2010 and 2011, respectively (Fig. A4.1). The two-week heat wave imposed a gradient in maximum canopy temperatures that ranged from average (near the 50th percentile) to extreme (well beyond the 95th percentile) based on long-term means of air temperature (Fig. 4.1c). Although control and drought plots received the same thermal inputs within a given treatment and across both years, canopy temperatures were much higher in drought than the control plots (Fig. 4.1c) due to interactions between the precipitation and heat treatments.

ANPP and plant community responses

Despite the extremity of the heat waves imposed, the drought treatment dominated all ecological responses. Across all ANPP and plant community metrics, there were no significant effects of the heat treatments or interactions with the drought treatment during either the drought or recovery periods (Table 4.1). On the other hand, drought significantly impacted total, grass

78

and forb ANPP, and there was a significant interaction between drought and year for evenness and H′ (Table 4.1). Because there were no significant direct or interactive effects of even the highest heat wave treatment on ANPP or plant community composition, we focus hereafter on plant community and ecosystem responses to the drought treatment.

Total ANPP was reduced by 20% in the first year of drought and by ~60% during the second year (Fig. 4.2a). The large reduction in total ANPP during the second year was driven by reductions in ANPP for both grass (-45%) and forb (-76%) plant functional types (Fig. 4.2a). To assess whether the total ANPP response to drought was statistically extreme, we compared our results with long-term (27-year) ANPP data. After two consecutive years of drought, total ANPP was well below the 5th percentile of the statistical distribution of ANPP for the study site (Fig. 4.2b). Further, when placed in the context of the long-term functional relationship between growing season precipitation and ANPP, both the growing season precipitation and ecological response were statistically extreme in 2011 (i.e., both below 5th percentile, Fig. 4.2c).

While we detected no overall effects of extreme drought on plant species richness, there were significant effects of drought over time for evenness and H' (Table 4.1b). In addition, community composition was altered substantially in the second year of the drought (Fig. A4.2). Community divergence between control and drought treatments was driven by shifts in dominant species abundances (Fig. 4.3a; Fig. A4.2; Table A4.2). Large reductions in the abundance of *S. canadensis* during the second year of the drought was the primary reason for this divergence (Table A4.2).

Recovery from drought.

After the two-year drought, all plots received growing season rainfall inputs similar to the long-term average, permitting us to assess the initial extent of ecosystem recovery and quantify drought legacy effects. We observed complete recovery in ecosystem function (total ANPP; Fig. 4.3b) just one year post-drought. Although forb ANPP continued to be dramatically reduced (by 80%) post-drought due to reduced stem densities and abundance of the formerly dominant *S. canadensis* (Fig. 4.3a,c), there was a concurrent 46% increase in grass ANPP (Fig. 4.3b). This compensation by the grasses was accompanied by a 60% increase in tiller density (Fig. 4.3c) and an increase in relative abundance of *A. gerardii* (Fig. 4.3a inset).

Discussion

In this study, we exposed a native tallgrass prairie ecosystem to extreme drought and a two-week heat wave for two years, and then evaluated short-term recovery. Our objectives were to determine if an ECE occurred in response to the treatments based on long-term climate and productivity records for the site, and then to assess mechanisms underpinning ecosystem resistance and resilience to extreme heat and drought. We imposed drought and heat treatments that were statistically extreme, based on over a century of climate records for this location. Drought effects dominated community and ecosystem responses, with no effect of the heat treatments on ANPP or the plant community, nor any interactive effects with drought, despite the magnitude of the heat treatments and the resultant canopy temperatures. The ecosystem response to the imposed drought was extreme during 2011, with total ANPP reduced well below the 5th percentile of the historical distribution of ANPP values for this site. Thus, our experimental approach allowed us to explicitly attribute an extreme ecological response to an imposed climate

extreme, meeting the definition of an ECE (Smith 2011). However, despite this extreme response, we observed complete recovery in ecosystem function (total ANPP) one-year post drought due to a rapid demographic response by the dominant C_4 grass, *A. gerardii*, compensating for the loss of the dominant C_3 forb, *S. canadensis*. Collectively, these results suggest that this ecosystem has relatively low resistance yet high resilience to a two-year extreme drought and that the dominant species governed these responses.

While both drought and heat treatments were statistically extreme from a climatological perspective, only drought significantly impacted ANPP and plant community composition. Thus, contrary to our expectations there were no interactive or additive effects of heat and drought on ANPP and the plant community, despite evidence for these interactive effects from other studies (De Boeck et al. 2011). As previously mentioned, little to no ecological responses to a climate extreme may be due to the ecosystem's resistance to the magnitude, duration or timing of the climate driver. Given that canopy temperatures exceeded the 95th percentile for past air temperatures in both precipitation treatments, the resistance to the simulated heat waves was not likely a result of too low of magnitude of IR added. In addition, while the timing of the heat wave coincided with the purported greatest sensitivity to heat for this ecosystem (Craine et al. 2012), it was also near the time of peak production (Knapp et al. 2001), which could have minimized the impact on end of season total ANPP. Thus, it is possible that if the heat wave occurred over a longer period of time, or if the heat wave had occurred earlier in the growing season, we may have observed a greater response to the heat treatments.

We used the ECE framework to assess potential mechanisms by which an extreme climate driver may elicit an extreme ecological response, and we hypothesized that dominant species would govern the extremity of the ecological responses. While total ANPP was reduced

in both years of drought relative to the control, there was a three-fold difference in total ANPP

reductions between the first and second year of the drought. This difference in the impact of

drought on ANPP occurred with only a modest difference in precipitation between years (60.8

mm; Table A4.1). During the first year of the drought much of the reduction in ANPP was driven

by equivalent reductions in both grass and forb production (Fig. 4.2a), and thus the primary

mechanism of response was physiological (reduced growth). Indeed, no shift in community

structure (species richness, evenness or H') or composition was detected in the first year of the

drought (Table 4.1b; Fig. A4.2). Extremity in ecological responses is predicted to occur when

systems cross extreme response thresholds, in which the tolerance of one or more species in a

community is exceeded (Smith 2011, Kardol et al. 2012). This would lead to a significant

decrease in abundance of a species due to reduced growth, reproduction and/or mortality of

individuals, and a subsequent shift in plant community composition (via species re-ordering).

Depending on which species are impacted, effects on ecosystem function could be small if only

rare species are affected or large if dominant species are affected (Smith and Knapp 2003,

Hillebrand et al. 2008). In the second year of the drought, the extreme reduction in ANPP was

driven by significant reductions in grass and forb biomass, however the dominant forb *S.*

canadensis exhibited greater sensitivity than the dominant grass *A. gerardii* to the drought. This

resulted in a significant shift in species composition and reordering of species abundances (*A.*

gerardii increased in dominance but *S. canadensis* became much less abundant, Fig. 4.3a). It

appears that an increase in drought intensity in 2011 and/or the cumulative effects of two

consecutive years of drought may have exceeded an extreme response threshold for the dominant

C_3 forb. Previous work in this ecosystem has shown that forbs rely on deep soil moisture to avoid

water stress during dry periods, while grasses rely mostly on shallow soil moisture, tolerating the

dry periods (Nippert and Knapp 2007). It is possible that the cumulative effects of two years of drought and the extremity of drought in 2011, depleted these deeper soil moisture layers, leading to increased mortality of the dominant C_3 forb. Thus a drought avoidance strategy failed for the forb in the second year of the drought, while a drought tolerance strategy allowed persistence of the dominant grass.

Contrary to our hypothesis, the altered community composition that occurred in the second year of drought did not preclude rapid recovery in ecosystem function. Instead, the large reduction in the production of *S. canadensis* was completely compensated by increases in A. gerardii in just one year following drought (Fig. 4.3). Loss of dominant species can have prolonged and significant ecosystem effects (Smith and Knapp 2003, Bershears et al. 2005), however the changes in community composition we observed are consistent with two proposed biotic mechanisms enhancing functional resilience: shifts in the abundance of dominant species and demographic (recruitment) compensation (Hillebrand et al. 2008, Lloret et al 2012). Compensation by *A. gerardii* further increased the divergence in community structure between drought and control plots the year after the drought ended (Fig. A4.2), and this compensation was likely driven rapid demographic recruitment of tillers post-drought (Fig. 4.3c). In this grassland, greater than 99% of aboveground shoots are vegetatively produced from belowground buds (Benson and Hartnett 2006), and therefore bud bank demography is an important mechanism behind responses of this grass to environmental stress and disturbance. *A. gerardii* has a large dormant bud bank, consisting of multiple years of cohorts (Ott and Hartnett 2012), and therefore has the capability to respond rapidly post-drought. This post-drought recruitment response of *A. gerardii* was also observed during the years immediately following the historic 1930's Dust Bowl (Weaver 1954). Given this is a region with a history of severe short-term and

multi-year droughts (Woodhouse and Overpeck 1998, Burnette and Stahle 2012), such a demographic response may be key to the dominance of *A. gerardii* and the high resilience of this ecosystem.

The immediate recovery observed in this study exceeded previous reports of resilience in this grassland ecosystem (Tilman and Downing 1994, Haddad et al. 2002, Sherry et al. 2008) and differs from reported legacy effects (Sala et al. 2012). Although the magnitude of the imposed drought was comparable to the 1930's Dust Bowl, the duration was shorter (2 versus 8 years), which may have been key to the rapid recovery in function. The long duration of the Dust Bowl resulted in much more dramatic community changes than we observed in our study; mesic species died and were replaced by xeric species, resulting in a prolonged, twenty-year recovery, once precipitation levels returned to normal (Weaver 1968). Therefore, while this ecosystem had high resilience to the short-term drought imposed in this experiment, a lengthier event (multi-year to decade-long) may significantly alter community structure (through species losses and additions), thereby reducing resilience.

Summary

We draw three broad insights from our research. First, extremity in a climate driver does not necessarily mean extremity in an ecological response. In our experiment, we observed an ECE associated with drought, while extreme heat had no effect on ecosystem function. Factors such as the intensity, duration and the timing of the extreme event will ultimately determine resistance. Central US grasslands have a long history of both short-term and decadal-scale droughts (Woodhouse and Overpeck 1998, Burnette and Stahle 2012), yet functional resistance was lost after only two years of extreme drought. This emphasizes the need to quantify the

timing of loss of function with climate extremes. To date, this has been difficult due to the rarity of naturally occurring extremes and because they must often be studied retrospectively (after loss of function is evident) with limited temporal resolution. Second, low resistance of ecosystem function to climate extremes does not preclude high resilience. Despite the extreme reduction in ANPP after two years of imposed drought, full recovery of this important ecosystem function required only one growing season post-drought. This raises the intriguing possibility that resilience in function, rather than resistance, might be expected for ecosystems with a history of climate extremes. Finally, the presence of a dominant species capable of rapidly recruiting new individuals and restoring function after an extreme climatic event underpinned high ecosystem resilience in this grassland. This compensatory response was primarily driven by the growth of a single dominant C_4 grass, *A. gerardii,* which is an important species for the resilience in this ecosystem (Weaver 1954, Sherry and Arnone 2012). Given that most ecosystems are dominated by a few species (Whittaker 1965), knowledge of the traits that influence dominant species responses to and recovery from climate extremes will be key for predicting ecosystem dynamics and function in a future with more extreme events.

Tables

Table 4.1. Analysis of variance (ANOVA) for **(a)** three component of aboveground net primary productivity (ANPP) and **(b)** three common metrics of community structure (H' = Shannon's diversity) across all three years of the experiment. Bold values indicate significance (p < 0.05).

a)

Effect	Total		Grasses		Forbs	
	F	p	F	p	F	p
Drought	15.6	0.060	0.1	0.828	24.0	0.043
Heat	0.9	0.511	0.2	0.924	0.3	0.816
Drought x Heat	1.0	0.475	0.1	0.960	0.3	0.851
Year	**74.0**	**<0.001**	**88.3**	**<0.001**	**15.7**	**<0.001**
Drought x Year	**33.1**	**<0.001**	**38.6**	**<0.001**	**11.0**	**<0.001**
Heat x Year	0.4	0.883	0.3	0.949	0.6	0.727
Drought x Heat x Year	2.2	0.055	2.0	0.080	0.9	0.489

b)

| Effect	Richness		Evenness		H'	
	F	p	F	p	F	p
Drought	1.4	0.250	0.1	0.797	0.0	0.892
Heat	0.7	0.548	2.7	0.063	0.5	0.671
Drought x Heat	1.7	0.187	1.6	0.214	1.5	0.233
Year	**5.9**	**0.004**	**3.8**	**0.029**	1.8	0.177
Drought x Year	1.5	0.235	**12.0**	**<.0001**	**4.8**	**0.012**
Heat x Year	0.9	0.500	0.7	0.622	1.7	0.147
Drought x Heat x Year	1.4	0.229	0.6	0.744	0.9	0.528

Figures
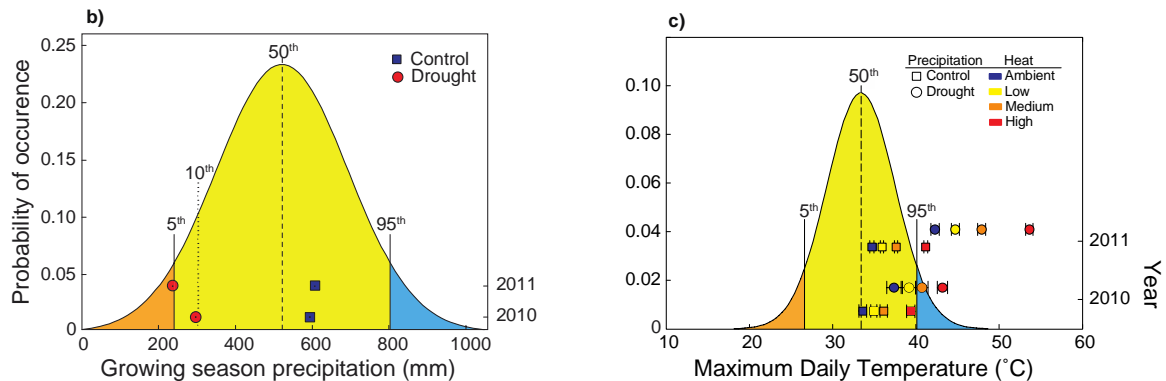
Figure 4.1 The Climate Extremes Experiment established in 2010 in a central US grassland. (**a**) During the 2010 and 2011 growing seasons, four large shelters were used to impose two treatments: drought (partial roofs on shelters reduced ambient rainfall by 66%) or ambient (control) rainfall inputs (no roofs on shelters). Nested within the rainfall shelters, heat wave treatments were imposed using passive heat chambers combined with IR lamps during two-weeks mid-summer. (**b**) The drought treatment resulted in severe drought in 2010 (exceeding the 10th percentile, dotted line) and extreme drought in 2011 (exceeding 5th percentile, solid line) based on an estimated probability function calculated from 112-years of growing season precipitation for the study site. In contrast, growing season precipitation was slightly above average in both years (50th percentile, dashed line) for the control treatments. (**c**) Mean maximum daily canopy temperatures (±1SEM) resulting from the four heat treatments during the two-week heat wave in under control in drought conditions compared to 112-years of maximum daily air temperature. Canopy temperatures ranged from near average (50th percentile, dashed line) to extreme (well beyond the 95th percentile, solid line).
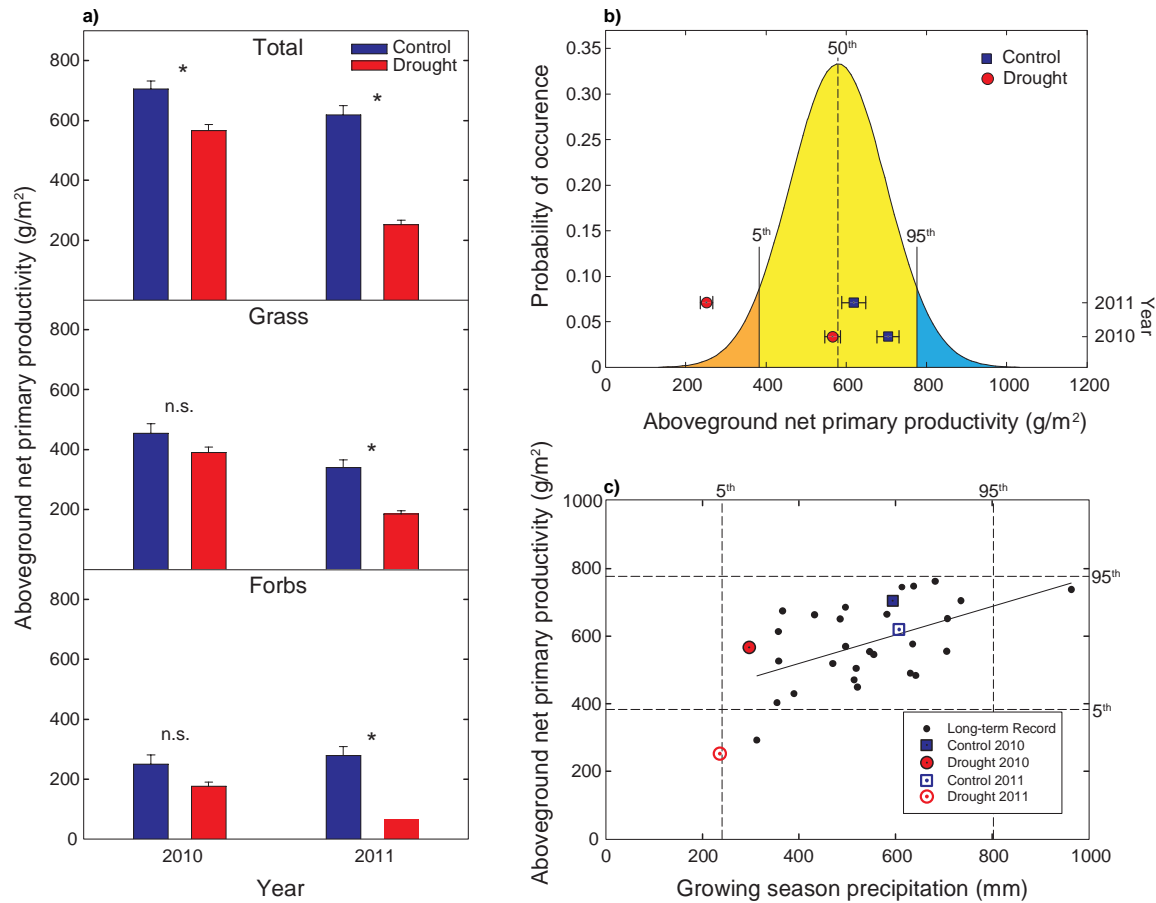
Figure 4.2 Response of aboveground net primary productivity (ANPP) to one (2010) and two years (2011) of experimentally imposed growing season drought in a central US grassland (**a**) Total, grass and forb ANPP during drought years. Bars are means + 1SEM. Asterisks denote significant treatment differences (p <0.05) for each year (n.s. = non-significant differences). (**b**) After two years of growing season drought, reductions in total ANPP exceeded the 5th percentile based on an estimated probability function for 27-years of ANPP measurements for the study site. (**c**) Relationship between growing season precipitation and ANPP ($r^2 = 0.27$, p = 0.005) over a 27-year period at the study site (long-term data sets from Fig. 4.1A, 2B). Total ANPP for control and drought treatments in both years are overlaid on this relationship. Based on this relationship, the second year of drought was considered an extreme climatic event, i.e., both precipitation and ANPP were statistically extreme (dashed lines show 5th and 95th percentiles for each variable).
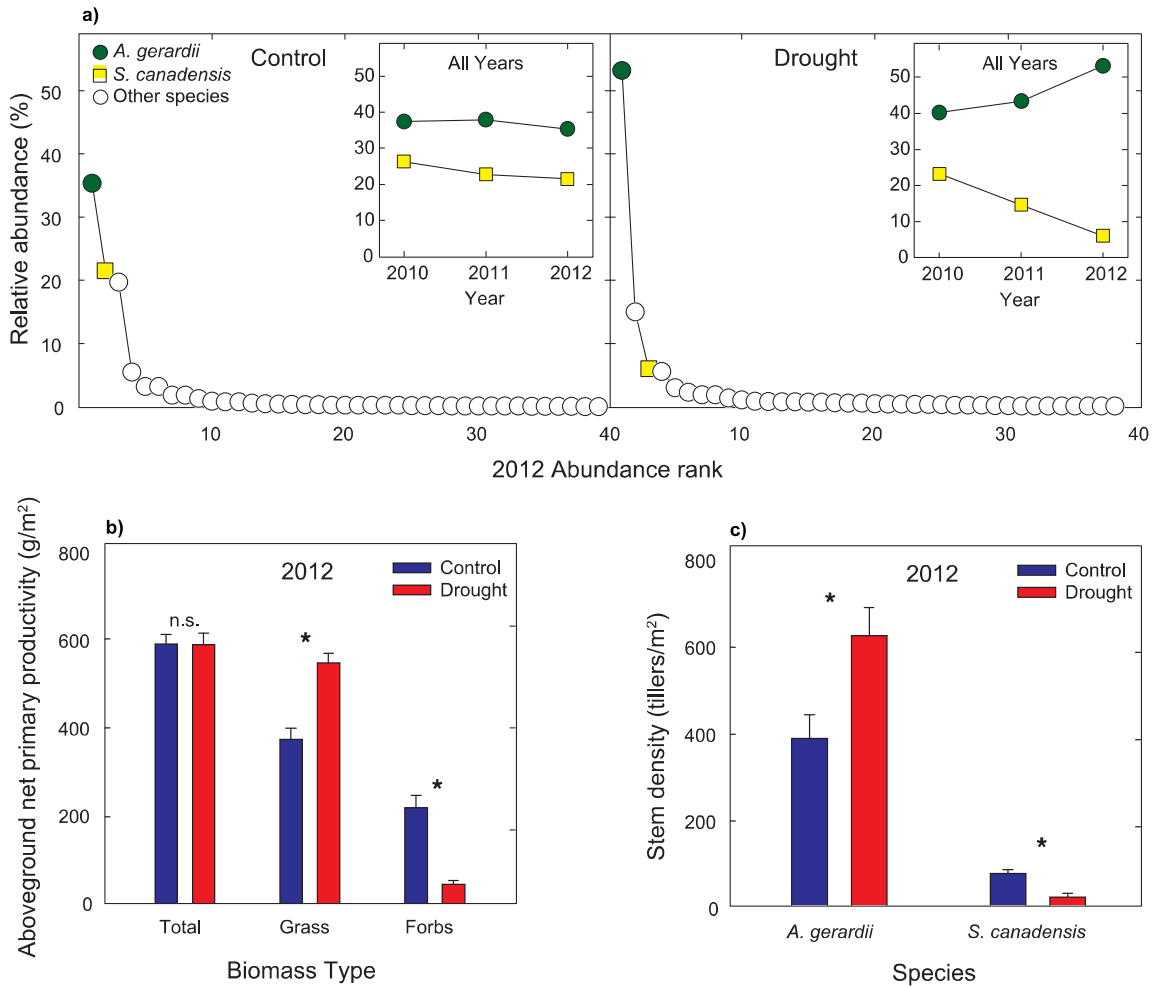
Figure 4.3 Recovery from extreme drought in 2012; both control and previously droughted plots received ambient rainfall plus supplemental irrigation to achieve long-term mean precipitation inputs in 2012. (**a**) Rank abundance curve of all species in control and previously droughted plots one year post-drought (2012). Inset: relative abundance of the dominant C_4 grass (*Andropogon gerardii*) and dominant C_3 forb (*Solidago canadensis*) for control and drought treatments during the two year drought (2010-11), and the subsequent recovery year (2012). (**b**) Response of total, grass and forb aboveground net primary productivity (ANPP) during the 2012 recovery year. (**c**) Stem densities of *A. gerardii* and *S. canadensis* during the 2012 recovery year. For B and C, bars are means +1SEM, and asterisks denote significant differences between precipitation treatments (p ≤0.05) for each biomass type or species (n.s. = non-significant differences)

Appendix

Table A4.1 Growing season inputs (in mm, April 1st – August 30th) for the drought (75% reduction in rainfall inputs) and control treatments. Numbers in parentheses indicate the amount of supplement rainfall added.

Year	Control	Drought
2010	594.0	297.1
2011	607.3	236.3
2012	626.4	626.4

Table A4.2 Percentage contribution of different plant species to community divergence between control and drought treatments based on SIMPER analysis (shown here are species contributing > 5% to divergence). Data from 2010 is not shown because communities were not significantly different (Fig. A4.2).

2011		2012	
Species	Contribution to Divergence (%)	Species	Contribution to Divergence (%)
Solidago canadensis	31.9	*Andropogon gerardii*	40.8
Andropogon gerardii	25.6	*Solidago canadensis*	29.0
Sorghastrum nutans	13.0	*Sorghastrum nutans*	8.4
Bouteloua curtipendula	9.2	*Carex meadii*	8.1
All other species	20.3	*All other species*	13.7
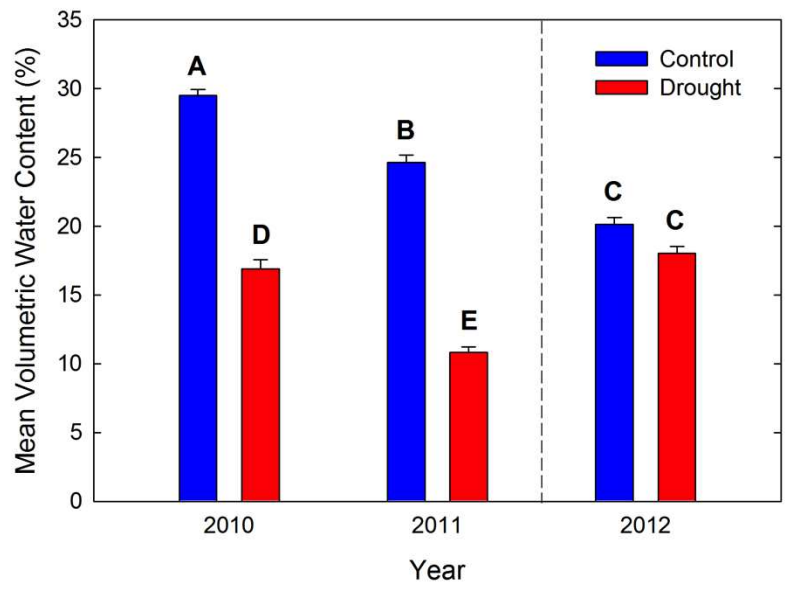
Figure A4.1 Mean growing season soil moisture for control and drought treatments for the two drought treatment years (2010 and 2011) and the recovery year (2012). Letters denote significant differences across treatments and years ($p \leq 0.05$).
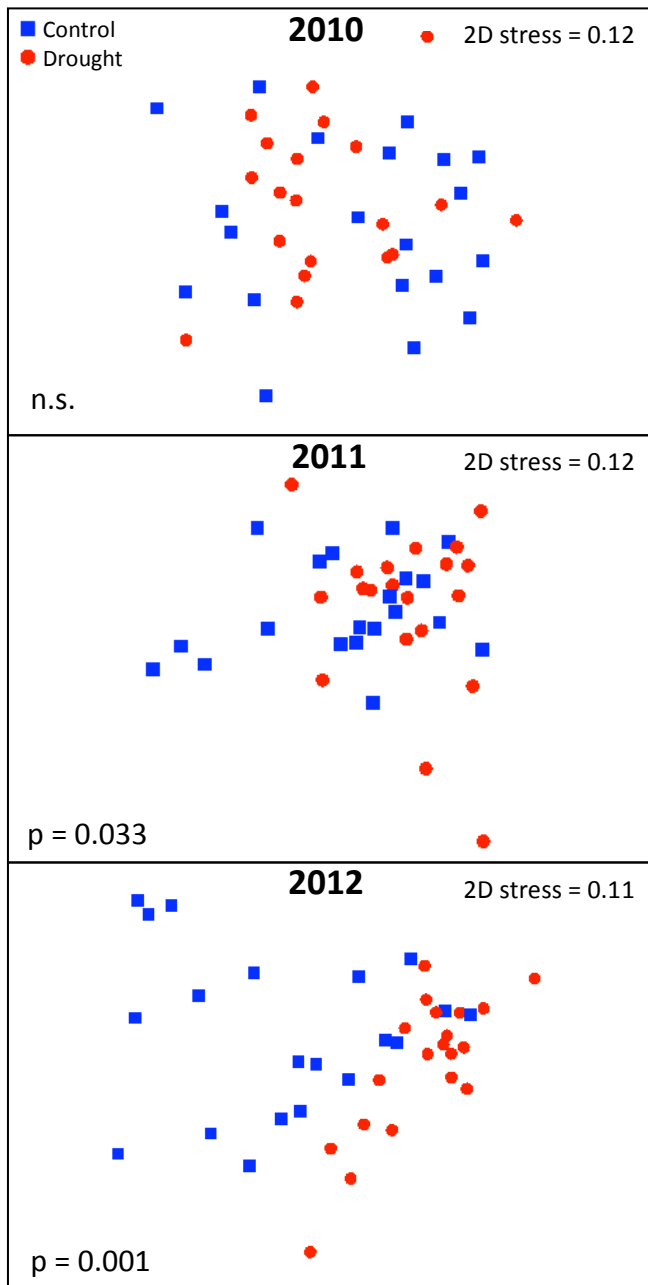
Figure A4.2 Non-metric multidimensional scaling (NMDS) ordination for control and treatments over the two drought years (2010, 2011) and recovery year (2012). Significance from analysis of similarity (ANOSIM) tests (p ≤0.05 for divergence criteria) and 2D stress of the NMDS are included.

References

Benson, E. J., and D. C. Hartnett. 2006. The Role of Seed and Vegetative Reproduction in Plant Recruitment and Demography in Tallgrass Prairie. Plant Ecology 187:163-178.

Breshears, D. D., N. S. Cobb, P. M. Rich, K. P. Price, C. D. Allen, R. G. Balice, W. H. Romme, J. H. Kastens, M. L. Floyd, J. Belnap, J. J. Anderson, O. B. Myers, and C. W. Meyer. 2005. Regional vegetation die-off in response to global-change-type drought. Proceedings of the National Academy of Sciences of the United States of America 102:15144–8.

Brown, D. A. 1993. Early Nineteenth-Century Grasslands of the Midcontinent Plains. Annals of the Association of American Geographers 83:589–612.

Burnette, D. J., and D. W. Stahle. 2012. Historical perspective on the dust bowl drought in the central United States. Climatic Change 116:479–494.

Ciais, P., M. Reichstein, N. Viovy, and A. Granier. 2005. Europe-wide reduction in primary productivity caused by the heat and drought in 2003. Nature 437:529–533.

Craine, J. M., J. B. Nippert, A. J. Elmore, A. M. Skibbe, S. L. Hutchinson, and N. A. Brunsell. 2012. Timing of climate variability and grassland productivity. Proceedings of the National Academy of Sciences of the United States of America 109:3401–3405.

De Boeck, H. J., F. E. Dreesen, I. a Janssens, and I. Nijs. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. The New phytologist 189:806–17.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. Global Change Biology 16:1992–2000.

Déry, S. J., and E. F. Wood. 2005. Observed twentieth century land surface air temperature and precipitation covariability. Geophysical Research Letters 32:L21414–L21418.

Easterling, D. R., G. A. Meehl, C. Parmesan, S. A. Changnon, T. R. Karl, and L. O. Mearns. 2000. Climate extremes: observations, modeling, and impacts. Science 289:2068–2074.

Fay, P., J. Carlisle, A. Knapp, J. Blair, and S. Collins. 2000. Altering rainfall timing and quantity in a mesic grassland ecosystem: Design and performance of rainfall manipulation shelters. Ecosystems 3:308–319.

Haddad, N. M., D. Tilman, and M. H. Knops. 2002. Long-term oscillations in grassland productivity induced by drought. Ecology letters 5:110–120.

Hillebrand, H., D. Bennett, and M. Cadotte. 2008. Consequences of dominance: A review of evenness effects on local and regional ecosystem processes. Ecology 89:1510–1520.

IPCC. 2012. Managing the Risks of Extreme Events and Disasters to Advance Climate Change Adaptation. (C. Field, V. Barros, T. Stocker, D. Qin, D. Dokken, K. Ebi, M. Mastrandrea, K. Mach, G. Plattner, S. Allen, M. Tignor, and M. PM, Eds.) A Special Report of Working Groups I and II of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, England.

IPPC. 2007. Climate Change 2007: The Physical Science Basis. (S. Solomon, D. Qin, M. Manning, Z. Chen, M. Marquis, K. Averyt, M. Tignor, and L. Miller, Eds.) Energy Environment. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

Jentsch, A., J. Kreyling, and C. Beierkuhnlein. 2007. A new generation of climate-change experiments: events, not trends. Frontiers in Ecology and the Environment 5:365–374.

Jentsch, A., J. Kreyling, M. Elmer, E. Gellesch, B. Glaser, K. Grant, R. Hein, M. Lara, H. Mirzae, S. E. Nadler, L. Nagy, D. Otieno, K. Pritsch, U. Rascher, M. Scha, M. Schloter, B. K. Singh, J. Stadler, J. Walter, and C. Wellstein. 2011. Climate extremes initiate ecosystem-regulating functions while maintaining productivity:689–702.

Kardol, P., J. Long, and M. Sundqvist. 2012. Crossing the threshold□: the power of multi-level experiments in identifying global change responses. New Phytologist:2008–2011.

Knapp, A., J. Briggs, and J. Koelliker. 2001. Frequency and extent of Water Limitation to Primary Production in a Mesic Temperate Grassland. Ecosystems 4:19–28.

Knapp, A. K., J. M. Briggs, D. L. Childers, and O. E. Sala. 2007. Estimating aboveground net primary production in grassland and herbaceous dominated ecoystems. Pages 27–48 in T. J. Fahey and A. K. Knapp, editors. Principles and Standards for Measuring Net Primary Production. Oxford University Press, Inc. New York.

Knapp, A. K., J. M. Briggs, D. C. Hartnett, and S. L. Collins. 1998. Grassland Dynamics: Long-Term Ecological Reserach in Tallgrass Prairie. Oxford University Press, Inc., New York.

Kreyling, J., M. Wenigmann, C. Beierkuhnlein, and A. Jentsch. 2008. Effects of Extreme Weather Events on Plant Productivity and Tissue Die-Back are Modified by Community Composition. Ecosystems 11:752–763.

Lal, R., J. Delgado, and J. Gulliford. 2012. Adapting agriculture to drought and extreme events. Journal of Soil and Water Conservation 67:162–166.

Nippert, J. B., and A. K. Knapp. 2007. Soil water partitioning contributes to species coexistence in tallgrass prairie. Oikos 116:1017–1029.

Ott, J. P., and D. C. Hartnett. 2012. Contrasting bud bank dynamics of two co-occurring grasses in tallgrass prairie: implications for grassland dynamics. Plant Ecology 213:1437–1448.

Van Peer, L., I. Nijs, D. Reheul, and B. De Cauwer. 2004. Species richness and susceptibility to heat and drought extremes in synthesized grassland ecosystems: compositional vs physiological effects. Functional Ecology 18:769–778.

Pimm, S. 1984. The complexity and stability of ecosystems. Nature 307:321–326.

Reichstein, M., P. Ciais, D. Papale, R. Valentini, S. Running, N. Viovy, W. Cramer, A. Granier, J. Ogée, V. Allard, M. Aubinet, C. Bernhofer, N. Buchmann, A. Carrara, T. Grünwald, M. Heimann, B. Heinesch, A. Knohl, W. Kutsch, D. Loustau, G. Manca, G. Matteucci, F. Miglietta, J. M. Ourcival, K. Pilegaard, J. Pumpanen, S. Rambal, S. Schaphoff, G. Seufert, J. F. Soussana, M. J. Sanz, T. Vesala, and M. Zhao. 2007. Reduction of ecosystem productivity and respiration during the European summer 2003 climate anomaly: A joint flux tower, remote sensing and modelling analysis. Global Change Biology 13:634–651.

Reyer, C. P. O., S. Leuzinger, A. Rammig, A. Wolf, R. P. Bartholomeus, A. Bonfante, F. de Lorenzi, M. Dury, P. Gloning, R. Abou Jaoudé, T. Klein, T. M. Kuster, M. Martins, G. Niedrist, M. Riccardi, G. Wohlfahrt, P. de Angelis, G. de Dato, L. François, A. Menzel, and M. Pereira. 2013. A plant's perspective of extremes: terrestrial plant responses to changing climatic variability. Global change biology 19:75–89.

Sala, O. E., L. A. Gherardi, L. Reichmann, E. Jobbágy, D. Peters, and E. Jobba. 2012. Legacies of precipitation fluctuations on primary production□: theory and data synthesis 367:3135–3144.

Sherry, R. A., E. Weng, J. a. Arnone Iii, D. W. Johnson, D. S. Schimel, P. S. Verburg, L. L. Wallace, and Y. Luo. 2008. Lagged effects of experimental warming and doubled precipitation on annual and seasonal aboveground biomass production in a tallgrass prairie. Global Change Biology 14:2923–2936.

Sherry, R., and J. Arnone. 2012. Carry over from previous year environmental conditions alters dominance hierarchy in a prairie plant community. Journal of Plant Ecology 5:134–146.

Smith, M. D. 2011a. An ecological perspective on extreme climatic events: a synthetic definition and framework to guide future research. Journal of Ecology 99:656–663.

Smith, M. D. 2011b. The ecological role of climate extremes□: current understanding and future prospects:651–655.

Smith, M. D., A. K. Knapp, and S. L. Collins. 2009. A framework for assessing ecosystem dynamics in response to chronic resource alterations induced by global change 90:3279–3289.

Smith, M., and A. Knapp. 2003. Dominant species maintain ecosystem function function with non-random species loss. Ecology Letters 6:509–517.

Tilman, D., and J. Downing. 1994. Biodiversity and stability in grasslands. Nature 367:363–365.

Weaver, J. 1954. North American prairie. Johnsen Publishing Company, Lincoln, Nebraska.

White, T. A., B. D. Campbell, P. D. Kemp, and C. L. Hunt. 2000. Sensitivity of three grassland communities to simulated extreme temperature and rainfall events. Global Change Biology 6:671–684.

Whittaker, R. H. 1965. Dominance and diversity in land plant communities: Numerical relations of species express the importance of competition in community function and evolution. Science 147:250–260.

Woodhouse, C., and J. Overpeck. 1998. 2000 years of drought variability in the central United States. Bulletin of the American Meteorological Society 79:2693–2714.

Yahdjian, L., and O. Sala. 2002. A rainout shelter design for intercepting different amounts of rainfall. Oecologia 133:95–101.

Chapter 5

RESPONSE AND RECOVERY OF SOIL RESPIRATION TO EXTREME DROUGHT AND HEAT IN A MESIC GRASSLAND

Overview

A predicted increase in the frequency and intensity of climate extremes may impact terrestrial carbon fluxes to the atmosphere; changing ecosystems from carbon sinks to sources, with positive feedbacks to climate change. Soil respiration, the second largest terrestrial carbon flux, may be affected by heat waves and drought due to soil moisture and temperature sensitivities. In this study, we investigated the effects of such climate extremes on soil respiration in a mesic grassland by experimentally imposing two years of drought and a midsummer heat wave, followed by a recovery year. Soil respiration was reduced by ~25% during both years of drought but was increased by 6% during the recovery year in formerly droughted plots. Meanwhile, we found little evidence for temperature sensitivity of soil respiration; the heat wave treatments had no effect on soil respiration (alone or when combined with drought) and we found no relationship between soil temperature and soil respiration over the growing season even when controlling for soil moisture. We compared the drought sensitivity of soil respiration to aboveground net primary productivity and found that soil respiration was less sensitive to drought than production. These results suggest soil respiration will be more sensitive to more frequent and intense droughts than heat waves, with potential consequences for regional carbon cycles.

Introduction

The rise in atmospheric CO_2 due to anthropogenic inputs over the past few decades has been partially mitigated by the terrestrial carbon sink, thereby slowing the accumulation this important greenhouse gas in the atmosphere (Houghton 2007, Pan et al. 2011). However, a predicted increase in the frequency and intensity of climate extremes (IPCC 2012) may temporarily reduce the capacity of regions to act as a carbon sinks, or even turn them into a carbon sources. Such a reduction was observed during the 2003 European heat wave, when gross primary production was reduced by 30% changing this region from a net sink to a source of CO_2 (Ciais et al. 2005). The impacts of such extremes on carbon fluxes between the land surface and atmosphere may be concurrent and/or prolonged (Reichstein et al. 2013). While concurrent impacts of extremes on carbon fluxes have been observed (Reichstein et al. 2007, Arnone III et al. 2008, Schwalm et al. 2010), prolonged effects are less well known. For example, tree mortality triggered by events such as drought or super storms could result in prolonged effects on carbon fluxes, because a loss in annual carbon uptake by the dead trees coupled with their decomposition could result in a longer-term net carbon source to the atmosphere (Reichstein et al. 2013). Given the potential for extreme events to alter carbon fluxes, there is a growing need to understand both the responses and recovery dynamics of key carbon cycling processes.

Globally, soil respiration (R_s) is the second largest terrestrial carbon flux to the atmosphere, releasing ~98 Pg per year (Raich et al. 2002, Bond-Lamberty and Thomson 2010), and therefore small changes in this flux could have large impacts on global carbon cycles. Two processes contribute to R_s, autotrophic (plant roots) and heterotopic (microbes/soil fauna) respiration, which are generally limited by three factors: soil temperature, soil moisture and carbon substrate (Lou and Zhou 2006). R_s rates increase exponentially with soil temperature, as

99

higher temperatures decrease the activation energy of respiration-related metabolic processes (Lloyd and Taylor 1994). Therefore climate change associated increases in mean annual temperature or the frequency and intensity of heat waves could enhance this carbon source to the atmosphere. However, the sensitivity of R_s to temperature is also dependent on soil moisture (Mielnick and Dugas 2000). In a tallgrass prairie ecosystem, Harper et al. (2005) found that R_s was most sensitive to temperature at intermediate soil moisture levels, but not under very wet or dry conditions. In addition to climate, substrate supply is a critical factor regulating R_s, with carbon substrates taking many forms including: labile carbon from photosynthesis, above- and belowground plant litter, and soil organic matter (Wan and Luo 2003). These sources will likely differ in their sensitivities to climate extremes and the timescale of response, which in turn may affect R_s.

Although grasslands are typically considered to have a long-term carbon balance near zero (Owensby et al. 2006), recent observations suggest that drought can turn grasslands into temporary carbon sources (Zhang et al. 2010). R_s and aboveground net primary production (ANPP) are two important carbon fluxes between the land surface and atmosphere in grasslands that will likely be affected by climate extremes, but there is some uncertainty in their relative sensitivities to climate extremes such as drought. In many studies, ANPP responds much more negatively to drought than respiration, shifting ecosystems from being a sink to a source of carbon (Ciais et al. 2005, Schwalm et al. 2010, Jongen et al. 2011); however there have been exceptions (Welp et al. 2007, Jentsch et al. 2011). In a recent literature synthesis and modeling analysis, Shi et al. (in review) also found that production was more sensitive to drought than respiration in the short-term, with sensitivity of both dependent on the severity of the drought.

In this study, we investigated the effects of climate extremes – drought and heat waves - on R_s in a productive, mesic grassland. We experimentally imposed extreme drought for two growing seasons and then followed recovery for one year post-drought. In addition to drought, we imposed a short-term heat wave treatment mid-summer during the first two years to examine the independent and combined effects of drought and heat, since they naturally co-occur (De Boeck et al. 2010). This allowed us to determine if there were concurrent and prolonged effects of these climate extremes on R_s. We tested three hypotheses: 1) that growing season drought would have greater effects on R_s than the two-week heat wave; 2) that R_s sensitivity to soil temperature would be dependent on soil moisture; and 3) that ANPP would respond more to these climate extremes than R_s.

Methods

Study site

This study was conducted at the Konza Prairie Biological Station in NE Kansas, USA (39°05'N, 96°35'W), on an intact, native tallgrass prairie ecosystem. The climate is characterized as mid-continental, with cold, dry winters and warm wet summers with mean annual temperature of 13.0 °C and mean annual precipitation of 835 mm (Knapp et al. 1998b). This region also has high interannual variability in precipitation (Knapp et al. 1998b), as well as episodic heat waves and drought (Woodhouse and Overpeck 1998, Burnette and Stahle 2012). The experiment took place on an annually burned lowland site with deep soils classified as Typic Argiustoll, with a silty clay loam texture (8% sand; 32% clay), and a bulk density of 1.5 g m^{-3} (Blecker 2005; Blecker et al., 2006). This grassland is dominated by several C$_4$ grasses, with many C$_3$ forbs accounting for the high diversity for this ecosystem (Knapp et al. 1998b).

Experimental design

In 2010, the Climate Extremes Experiment was established to examine the independent and combined effects of drought and short-term heat waves on the tallgrass prairie ecosystem. To identify the concurrent and prolonged effects of the climate extremes on ecosystem function, treatments were applied during two consecutive years (2010 and 2011), followed by a recovery year (2012) when no treatments were imposed. Precipitation treatments (drought and control) were imposed during the growing season (April 1st - August 30th), beneath four modified 6 x 24 m greenhouse frames, constructed over native grassland. The drought treatment was imposed using passive rainfall removal whereby partial roofs (covered with strips of Dynaglas Plus® clear polycarbonate plastic; PALRAM Industries LTD., Kutztown, PA, USA) removed ~ 66 % of ambient precipitation. The control precipitation treatment received ambient rainfall plus supplemental irrigation (to prevent water limitations during dry periods). To control for the effects of shading by the infrastructure (~10% reduction in PAR) the control greenhouse shelter frames were covered with deer netting (TENAX Manufacturing Alabama, USA), which produced equivalent light reductions, but allowed rainfall to pass through. Within each rainfall shelter, ten 2 x 2 m plots were established and randomly assigned one of four heat treatments (ambient, low, medium and high), which were imposed for two weeks in late July using passive warming chambers and infrared heat lamps. Transparent chambers consisted of a 2 x 2 x 1.5 m polyvinyl chloride (PVC) frame, with 1m, 6-m polyethylene walls and polycarbonate roofs (Dynaglas Plus). Gaps at the base and the top of the chamber allowed for air to circulate between the chamber and ambient environment. To provide four levels of increased heat input, infrared heat lamps (HS/MRM 2420, 2000 W, Kalglo Electronics, Inc., Bethlehem, PA, USA) were paced within in chambers in the following combinations: control = no lamp, low heat = one lamp

at ½ power (+250 W/m^2), medium heat = one lamp at full power (+500 W/m^2), and high heat =

two lamps at full power (+1000 W/m^2). During 2012, no precipitation or heat treatments were

imposed and all plots received ambient precipitation plus supplemental irrigation to maintain

precipitation inputs near long-term monthly averages for the site.

Environmental measurements

In each plot (n=40), we measured three key environmental variables: soil moisture, soil

temperature and canopy temperature. Soil moisture was measured in the top 0-15 cm of the soil

using time-domain reflectometery probes (TDR, Model CS616, Campbell Scientific, Inc., Logan,

UT, USA). We measured soil temperature at a depth of 5 cm using thermocouples (K-type,

OMEGA Engineering Inc., Stamford, CT, USA). Canopy temperature was measured with

infrared thermometers (Model SI-111, Apogee Instruments, Inc., Logan, UT, USA) mounted in

the SE corner of each plot at a height of 1.5 m. Data from all three sensor types were sampled at

30 minute intervals and recorded using data loggers (CR10X Datalogger, Campbell Scientific,

Inc., Logan, UT, USA). To assess post-drought levels of soil nitrogen, we used anion and cation

exchange resin bags (n = 2/plot) that were incubated *in situ* throughout the 2012 growing season

to provide an integrated measurement of $NO_3 + NH_4$ availability (Baer et al. 2003).

Soil CO$_2$ flux measurements

In situ soil CO$_2$ flux measurements (R_s) were sampled bi-weekly during all three growing

seasons using a Li-Cor 8100 (LiCOR, Inc., Lincoln, NE, USA) portable gas exchange system. In

each plot, two PVC collars (10 cm diameter x 8 cm deep, buried 6 cm into the ground) were

placed in opposite corners of each 2 x 2 m plot, and 25 cm from the edge. Living plant material

and litter were carefully removed from within each collar so that we only measured R_s. Due to

slight shifting of collars throughout the summer, we took monthly measurements of the interior

height of each collar and adjusted the flux values to account for changes in aboveground collar

volume. Flux measurements for each collar required ~ 1 minute and were taken between 1100

and 1300 CDT.

Aboveground net primary production

End of season aboveground net primary production (ANPP) was estimated in the first

week in September each year. Within each plot, all aboveground plant material was harvested

within three 0.1 m^2 quadrats (locations were changed each year to prevent resampling). Because

the site was burned each spring, this biomass closely approximates ANPP (Knapp et al. 2007).

Samples were oven dried at 60°C for 48 hours, and then weighed to the nearest 0.1 g.

Statistical analyses

The experimental design was a randomized split-plot; block was nested within the

precipitation treatment, the heat wave treatment was nested within the drought treatment, and the

block x heat wave treatment interaction was a random effect. We analyzed the treatment effects

of drought and heat on R_s with a repeated measures mixed-model ANOVA for each year

separately. We used a stepwise linear regression model to assess the relative effects of soil

temperature and soil moisture (during 2010 and 2011) on R_s. Factors were eliminated from the

model using a cutoff of $\alpha = 0.05$. The correlation between soil moisture on soil temperature over

the whole growing season was analyzed at intermediate (20-40 %) and low (<15%) volumetric

water content during 2010 and 2011. Finally we compared the responses of ANPP and R_s to soil

moisture using linear regression and ANOVAs. Linear regressions were conducted using annual means for the two response variables (ANPP, R_s) against mean growing season soil moisture for all three years. ANOVA's were conducted for both R_s and ANPP by year (same previously described mixed model methods), with significant differences between control and drought treatments for each year were determined using pairwise comparisons (least square means). All analyses were conducted in SAS (version 9.3, SAS Institute Inc., Cary, NC, USA) and significance was set a p ≤0.05.

Results

During 2010 and 2011, the drought treatment reduced mean soil moisture by 43% and 56%, respectively (Table 5.1a). In both years, the timing of the two-week heat wave coincided with existing differences in soil moisture levels (Fig. 5.1). In 2012, the ambient rainfall plus supplemental irrigation applied to both control and drought treatments resulted in similar mean soil moisture (Table 5.1a) and dynamics over the course of the growing season (Fig. 5.1). The heat treatments had minimal impacts on soil temperature within the control treatments in both years (~ 1 °C range; Table 5.1b), and in the drought treatment in 2010 (~ 2 °C range; Table 5.1b). However in 2011, the most extreme drought year, we observed a much larger range in soil temperatures (~ 9.0 °C range; Table 5.1b), despite maintaining constant thermal inputs for each heat treatment and across both years.

We found that the drought treatments dominated responses of Rs (Table 5.2). During the extreme treatment years, we observed significant main effects of drought as well as drought x date interactions (Table 5.2). Drought treatments reduced R_s by 26% (2010) and 25% (2011) relative to the control. However, in the recovery year we observed a slight increase (6%) in soil

105

CO_2 for the formerly drought treatment relative to the control. This increase in R_s rates was most pronounced in the first half of the growing season (Fig. 5.1). We observed no significant effects of the heat waves or any interactions with drought in either of the treatment years, so we focus below on the precipitation treatment effects and interactions with soil temperatures throughout the growing season.

To investigate the effects of soil temperature and soil moisture on R_s, a stepwise regression model was constrained to the two extreme years, given the legacy effects we observed in the former drought treatment in in 2012. Soil temperature was dropped from the model ($\alpha > 0.05$), while volumetric water content was retained ($r^2 = 0.34$, $p < 0.001$; Fig. 5.2). The relationship between volumetric water and R_s was positive throughout the range of soil moisture, with no decrease noted at the highest or lowest soil moisture levels. Porosity in these soils ranges from 47-53% (Blecker 2005; Blecker et al., 2006), suggesting that we did not encounter saturated soil moisture conditions when CO_2 fluxes could be limited by diffusion rates.

As noted earlier, the sensitivity of R_s to temperature may be dependent on soil moisture levels, particularly during extreme years. We examined the relationship between soil temperature and R_s under two soil moisture conditions (values based on Harper et al. 2005), low (volumetric water content $< 15\%$) and intermediate (volumetric water content 20-40%) across the entire growing season. At both low and intermediate soil moisture levels, we observed no relationship between soil temperature and R_s (intermediate: $F = 0.33$, $p = 0.564$; low: $F = 1.25$, $p = 0.264$; Fig. 5.3).

Finally, we compared the drought sensitivities of R_s and ANPP to soil water across all three years of the experiment. Both of these key carbon cycling processes had strong relationships with mean volumetric water content during the growing season (R_s: $r^2 = 0.93$, $p =$

0.002; ANPP: $r^2 = 0.76$, p = 0.023; Fig. 5.4). However, when we examined the impact of drought over the three years we see evidence for differential sensitivities during and following drought. In 2010, both R_s and ANPP had similar relative reductions due to drought, but in the second and more extreme drought year, there was a three-fold decrease in ANPP relative to the first year, while R_s had equivalent decreases with drought each year (Fig. 5.5). In addition, while ANPP completely recovered in 2012 (no significant difference between precipitation treatments), R_s was significantly increased in the drought treatment relative to the control (Fig. 5.5).

Discussion

We subjected a native, intact mesic grassland ecosystem to two years of extreme drought, followed by a recovery year to examine the impacts on R_s, a key carbon flux to the atmosphere. In addition to drought, we exposed the ecosystem to a short-term heat wave each year at varying intensities. We observed significant reductions in R_s with drought, as well as increases in respiration in formerly droughted plots in the recovery year. However, there were no significant main effects of the short-term heat wave or interactive effects with drought. While this lack of sensitivity to heat may have been a result of the minimal heat treatment effects on soil temperature, we also observed no strong effect of changes in growing season soil temperature at any point during this study, even when controlling for soil moisture levels. Finally, although both R_s and ANPP were decreased during the drought, ANPP showed greater sensitivity during the second more extreme year, as well as divergent responses in the recovery year.

Drought was the dominant factor affecting R_s both during the two years with extreme climates and in the subsequent recovery year, while we observed no effects of the heat treatments (Table 5.2). This difference may be due to the duration and/or magnitude of these extreme

treatments. The heat wave lasted two-weeks, while the imposed drought lasted the entire growing season, where 75% of the precipitation occurs (Knapp et al. 1998b). The magnitude of the drought treatment was extreme, exceeding the 10th and 5th percentiles for growing season precipitation in 2010 and 2011, respectively (based on 112-year nearby climate record; Hoover Chapter 4). In contrast, the heat treatments had little effects on soil temperature, with the exception of the 2011 drought treatments (Table 5.1). This contrasted with canopy temperatures, with differences between ambient and high heat treatments ranging between 6 to 11 °C in both years (Hoover Chapter 4). However, it is important to note that even when soil temperatures were increased due to the heat treatments in 2011, we observed no concurrent or lagged effects of heat.

When we examined the effects of soil moisture and soil temperature over the entire growing season in the two extreme years using regression-based approaches, we also observed a greater sensitivity to soil moisture than temperature in this mesic grassland ecosystem. The stepwise regression results suggest that soil moisture is a strong predictor of R_s (Fig. 5.2), which supports the previous ANOVA results with significant effects of drought treatments. The lack of a direct response of R_s to soil temperature was surprising, given that it has been reported to be a very important factor in grasslands (Lloyd and Taylor 1994, Mielnick and Dugas 2000) and in this tallgrass prairie (Knapp et al. 1998a). Recently, Poll et al. (2013) observed that the sensitivity of R_s to soil temperature was dependent on soil moisture. To test this relationship in this ecosystem, we examined the response of R_s to soil temperature under intermediate and dry conditions. These soil moisture levels were based on a response surface analysis by Harper et al. (2005), which found R_s was sensitive to temperature under intermediate but not dry conditions (our experiment was conducted adjacent to the study reported in Harper et al. 2005). However,

we did not observe any sensitivity to soil temperature under either soil moisture level (Fig. 5.3). These results suggest that during the growing season, R_s in this ecosystem is more sensitive to changes in soil moisture than soil temperature, and thus a future with more frequent and intense droughts should impact this important carbon flux to the atmosphere more than increases in growing season temperature.

In addition to strong concurrent impacts of extreme drought on R_s, we also observed prolonged effects of drought in the recovery year. Despite receiving the same precipitation inputs and having similar mean soil moisture during the growing season in 2012 (Table 5.1), we observed a significant increase in R_s (6%) in the formerly droughted plots (Table 5.2; Fig. 5.1). This increase occurred primarily in the early growing season (Fig. 5.1), and diminished by mid-July. We consider three possible mechanisms that may have resulted in this post-drought increase in Rs. First, an increase post-drought nitrogen availability (Yahdjian et al. 2006, Evans and Burke 2013), which may stimulate autotrophic and heterotrophic activities, leading to higher respiration rates. However, we detected no significant difference in nitrogen (NO_3^- and NH_4) in the soil between previously droughted and control plots during the recovery period (F =0.01, p = 0.949). Another possibility is that drought reduced litter decomposition over the two treatment years, leading to a litter build up and a liable source of carbon for the recovery year. This increase in substrate combined with high soil moisture and warm temperatures may have resulted in a transient early season R_s pulse. Finally, during the recovery year there was a large shift in plant community composition in formerly droughted plots, with grasses increasing in abundance and forbs decreasing (Hoover Chapter 4). Given that roots contribute about 80% of total soil respiration in the tallgrass prairie (Ham et al. 1995), and grasses tend to have shallower root

systems than forbs (Knapp et al. 1998b), such a change in plant species composition may contributed to elevated R_s.

In this study we found that two key atmospheric carbon fluxes, R_s (CO_2 source) and ANPP (CO_2 sink), were both affected by drought, however they differed in their sensitivities to the extreme event. During the first year of drought, both R_s and ANPP had similar reductions relative to the control plots (Fig. 5.5). But during the second, more extreme year of drought, ANPP was reduced three fold relative to the previous year, but reductions in R_s were similar to the previous year (Fig. 5.5). This differential sensitivity may be due to different response times of production and respiration to soil moisture limitations. ANPP is strongly controlled by precipitation (Sala et al. 1988, Knapp and Smith 2001), and therefore drought can cause immediate reductions in plant production as well as legacy effects (Sala et al. 2012). However, in this study we saw rapid recovery in ANPP in the year following drought where control and drought treatments did not differ in production (Fig. 5.5). While R_s was also limited by soil moisture in this experiment (Fig. 5.4), Shi et al. (2013) argue that soil carbon supplies are more stable and therefore R_s is more buffered against short-term precipitation variation than plant production. As previously mentioned, we observed increased R_s in formerly droughted plots during the recovery year, which contrasted with the full recovery in ANPP. Combined, the net effect of these differential sensitivities to extreme drought of these two key carbon fluxes will have important consequences on atmospheric CO_2 levels.

Summary

A future with more frequent and intense climate extremes, will impact terrestrial carbon cycles, with potential feedbacks to global climate change (Reichstein et al. 2013). Our study has

110

several important implications for the effects of extreme drought and heat on carbon fluxes in mesic grassland ecosystems. First, to understand the net effect of climate extremes on carbon cycles, it is important to examine concurrent responses and prolonged effects. For example, increased R_s during the recovery year in formerly droughted plots partially offset the decreased fluxes during the two years of drought. Such prolonged effects of a short-term climate anomalies on R_s have been documented in this ecosystem previously (year-long warming, Arnone III et al. 2008). Secondly, short-term heat waves had virtually no effect on R_s in this study; instead soil moisture effects dominated. As hypothesized, we did not expect to see a strong effect of soil temperature under droughted conditions due to soil moisture limitations, however when we specifically examined intermediate soil moisture levels, there also was no effect of soil temperature. One possibility is that the range in temperatures experience under intermediate soil moisture was limited (~17-28 °C). Empirical studies on the relationship between soil temperature and R_s, or calculating Q_{10} values, often use a broader ranged in temperatures by taking measurements throughout the year, not just the growing season (Lloyd and Taylor 1994, Knapp et al. 1998a, Mielnick and Dugas 2000, Harper et al. 2005), which may enhance the importance of temperature. However, our results suggest that during the growing season, R_s may not be as sensitive to the range of temperatures experienced. Finally, given that grasslands are important carbon sinks (White et al. 2000), the differential sensitivity to drought of R_s and ANPP has important consequences for carbon balance of this system. Extreme drought may reduce the capacity of this ecosystem to sequester carbon if ANPP (sink) is more sensitive than R_s (source) during extreme drought, and if respiration rates increase following drought. Overall, these results suggest that that R_s may be less temperature sensitive during the growing season than previously

reported and that extreme drought has strong but differential impacts on two key carbon cycling

processes in this ecosystem.

Tables

Table 5.1 Treatment effects on key environmental factors. (a.) Mean soil moisture during the growing season for control and drought treatments during the extreme years (control = ambient precipitation plus supplemental irrigation; drought = 66% reduction in ambient precipitation) and recovery year (ambient precipitation plus supplemental irrigation for both treatments). (b.) Soil temperature (0-5cm; °C) for each heat x precipitation treatment combination (n=5) during the two week simulated heat wave in 2010 and 2011. Parentheses for both tables indicate ± 1 SE.

a.	Extreme Treatment Years		Recovery Year
Precipitation Treatments	2010	2011	2012
Control	29.5 (0.5)	24.6 (0.5)	20.1 (0.5)
Drought	16.9 (0.7)	10.8 (0.4)	18 (0.5)

b.	2010		2011	
Heat Treatment	Control	Drought	Control	Drought
Ambient	26.4 (0.3)	27.2 (0.3)	26.1 (0.1)	30.4 (0.1)
Low	26.7 (0.3)	27.5 (0.3)	26.5 (0.1)	31.9 (0.2)
Medium	26.7 (0.3)	28.2 (0.3)	26.8 (0.1)	34.5 (0.3)
High	27.5 (0.3)	29.3 (0.3)	27.3 (0.1)	39.3 (0.4)

Table 5.2 Effects of drought and heat wave treatments on soil CO_2 flux during the two consecutive extreme years (2010, 2011) followed by a recovery year (2012). Drought was imposed during the growing season, while the heat wave was imposed for two-weeks mid-summer. F-statistics and p-values from mixed-model repeated measures ANOVAs for each year separately are reported. Bold text indicates significance at $p < 0.05$.

| Effect	2010		2011		2012	
	F-value	p-value	F-value	p-value	F-value	p-value
Drought	**73.8**	**<0.001**	**75.4**	**<0.001**	**8.7**	**0.006**
Heat	0.5	0.656	0.8	0.498	2.7	0.061
Drought*Heat	0.6	0.626	0.3	0.829	0.8	0.508
Date	**185.9**	**<0.001**	**22.7**	**<0.001**	**257.7**	**<0.001**
Drought*Date	**39.6**	**<0.001**	**16.4**	**<0.001**	**7.1**	**<0.001**
Heat*Date	1.2	0.270	1.6	0.056	0.6	0.885
Drought*Heat*Date	1.3	0.226	0.8	0.728	1.2	0.302
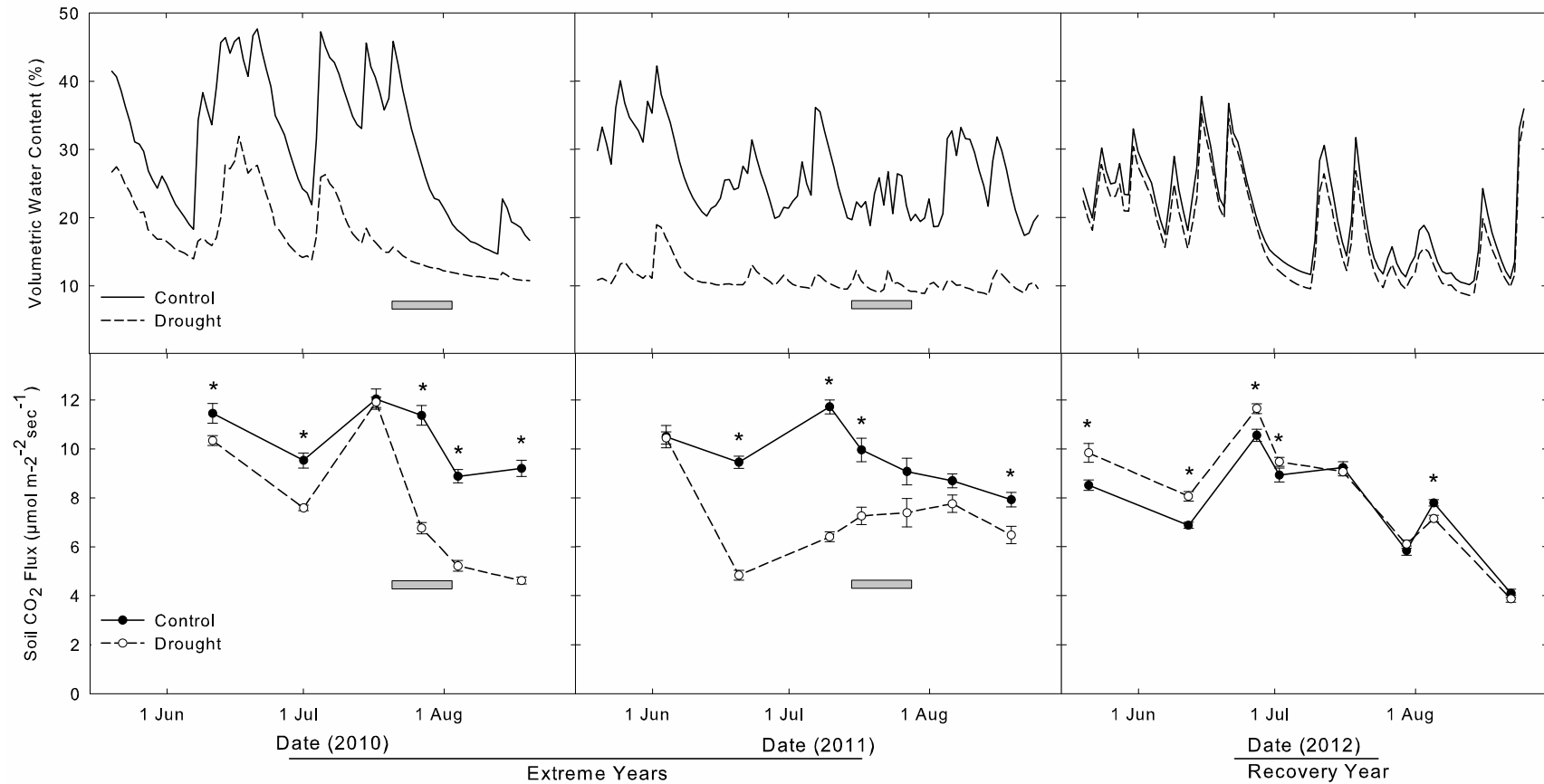
Figure 5.1 Volumetric water content (Top) and Soil CO_2 flux (R_s; Bottom) during the 2010-2012 growing seasons. Grey bar indicates the timing of the two-week heat wave. No treatments were imposed in 2012, with plots receiving ambient rainfall plus supplemental irrigation. Asterisks indicate significant differences ($p < 0.05$) on individual dates between control and drought treatments for soil CO_2 flux.
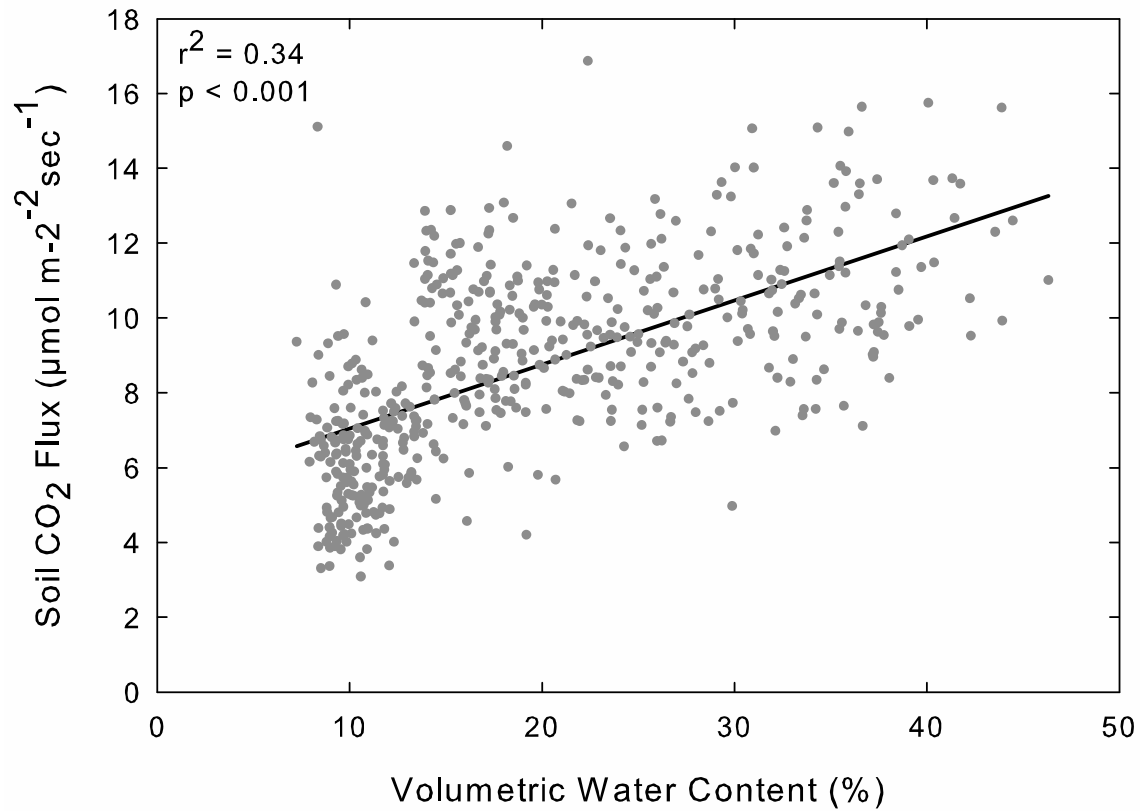
Figure 5.2 The relationship between volumetric water content and soil CO_2 flux (R_s) for all dates, precipitation and heat treatments combined (in 2010 and 2011). Results are from a stepwise linear regression which included volumetric water content and soil temperature as independent variables and soil CO_2 flux as the dependent variable. Soil temperature was removed from the model ($p > 0.05$), but volumetric water content was retained ($p < 0.05$), suggesting that soil moisture was the dominant variable controlling soil CO_2 flux. Statistics from the regression are reported in the figure.
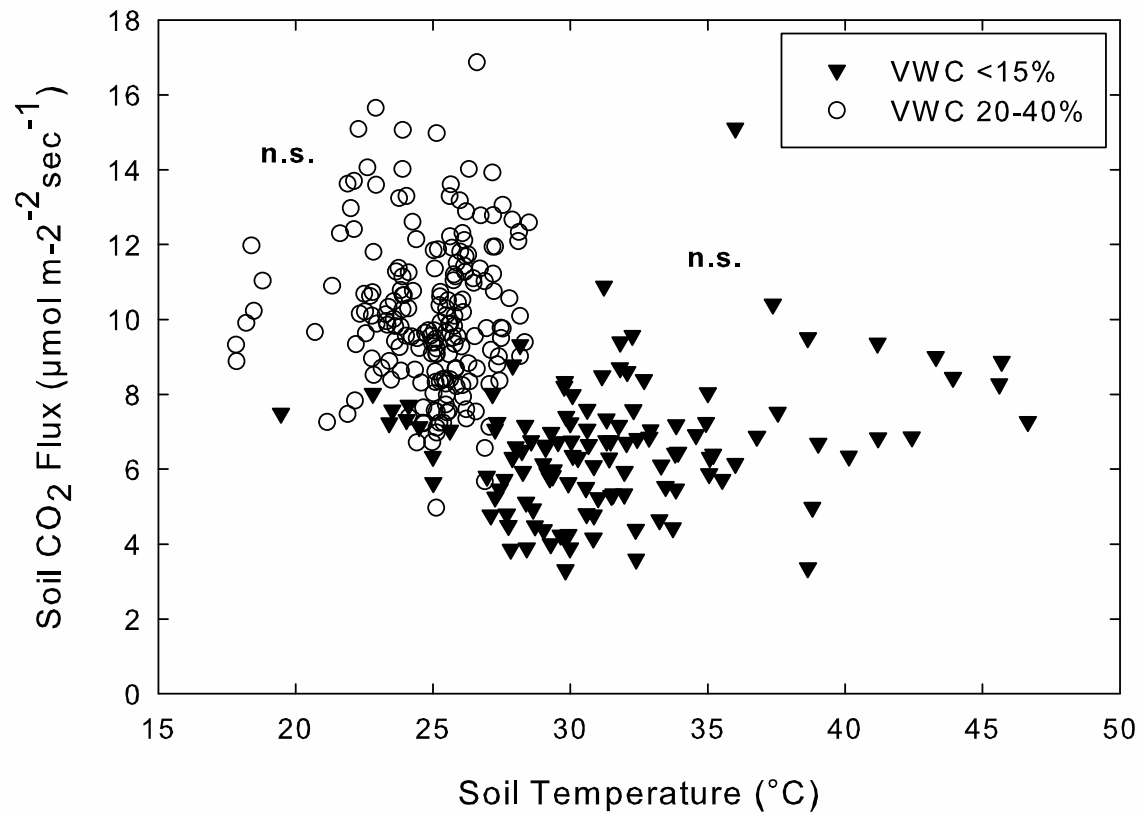
Figure 5.3 Sensitivity of soil CO_2 flux (R_s) to soil temperature at low (volumetric water content < 15%) and intermediate (volumetric water content 20-40%) moisture levels. Linear regressions were run for each soil moisture level independently, however neither resulted in a significant relationship (n.s. = non-significant).
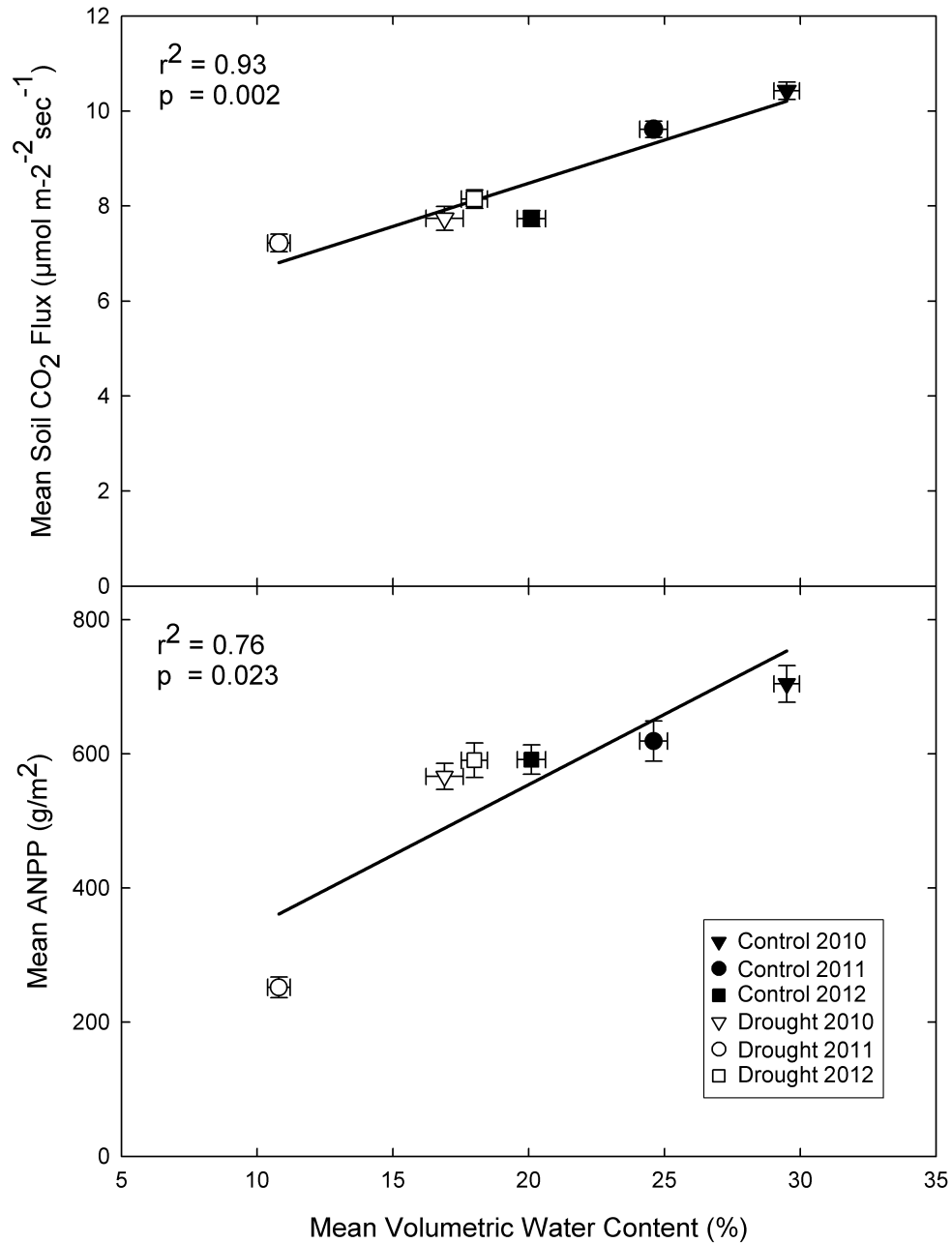
Figure 5.4 The relationship between mean soil CO_2 flux (R_s;Top) and mean aboveground net primary production (ANPP; bottom) and mean volumetric soil water content during the growing season for the two extreme (2010, 2011) and recovery (2012) years. Error bars indicate $\pm$ 1 standard error. Statistics from linear regressions are reported in each figure.
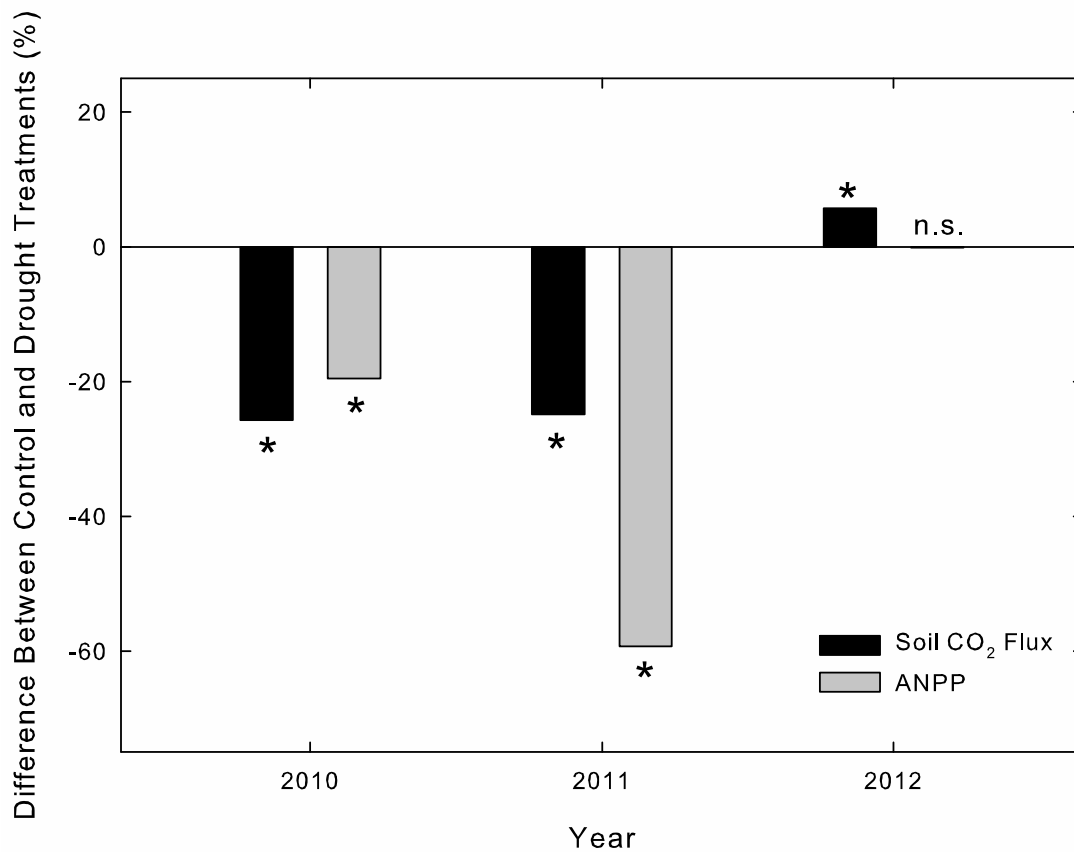
Figure 5.5 Relative differences between control and drought precipitation treatments for soil CO_2 flux (R_s) and aboveground net primary production (ANPP), during the two extreme (2010, 2011) and recovery (2012) years. Asterisks indicate significant differences ($p < 0.05$) between control and drought treatments by year for each response variable, based on pairwise comparisons from mixed model ANOVA's (n.s. = non-significant).

References

Arnone, J. A., P. S. J. Verburg, D. W. Johnson, J. D. Larsen, R. L. Jasoni, A. J. Lucchesi, C. M. Batts, C. von Nagy, W. G. Coulombe, D. E. Schorran, P. E. Buck, B. H. Braswell, J. S. Coleman, R. A. Sherry, L. L. Wallace, Y. Luo, and D. S. Schimel. 2008. Prolonged suppression of ecosystem carbon dioxide uptake after an anomalously warm year. Nature 455:383–6.

Baer, S., J. Blair, S. Collins, and A. Knapp. 2003. Soil resources regulate productivity and diversity in newly established tallgrass prairie. Ecology 84:724–735.

Blecker, S. W., R. L. McCulley, O. A. Chadwick, and E. F. Kelly. 2006. Biologic cycling of silica across a grassland bioclimosequence. Global Biogeochemical Cycles 20:GB3023 1–11.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. Global Change Biology 16:1992–2000.

Bond-Lamberty, B., and A. Thomson. 2010. Temperature-associated increases in the global soil respiration record. Nature 464:579–582.

Burnette, D. J., and D. W. Stahle. 2012. Historical perspective on the dust bowl drought in the central United States. Climatic Change 116:479–494.

Ciais, P., M. Reichstein, N. Viovy, and A. Granier. 2005. Europe-wide reduction in primary productivity caused by the heat and drought in 2003. Nature 437:529–533.

Evans, S., and I. Burke. 2013. Carbon and Nitrogen Decoupling Under an 11-Year Drought in the Shortgrass Steppe. Ecosystems 16:20–33.

Ham, J., C. Owensby, P. Coyne, and D. Bremer. 1995. Fluxes of CO2 and water vapor from a prairie ecosystem exposed to ambient and elevated atmospheric CO2. Agricultural and Forest Meteorology 77:73–93.

Harper, C. W., J. M. Blair, P. A. Fay, A. K. Knapp, and J. D. Carlisle. 2005. Increased rainfall variability and reduced rainfall amount decreases soil CO 2 flux in a grassland ecosystem. Global Change Biology 11:322–334.

Houghton, R. A. 2007. Balancing the Global Carbon Budget. Annual Review of Earth and Planetary Sciences 35:313–347.

IPCC. 2012. Managing the Risks of Extreme Events and Disasters to Advance Climate Change Adaptation. (C. Field, V. Barros, T. Stocker, D. Qin, D. Dokken, K. Ebi, M. Mastrandrea, K. Mach, G. Plattner, S. Allen, M. Tignor, and M. PM, Eds.) A Special Report of Working

Groups I and II of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, England.

Jentsch, A., J. Kreyling, M. Elmer, E. Gellesch, B. Glaser, K. Grant, R. Hein, M. Lara, H. Mirzae, S. Nadler, L. Nagy, D. Otieno, K. Pritsch, U. Rasher, M. Schadler, M. Schloter, B. Singh, J. Stadler, J. Walter, C. Wellstein, J. Wollecke, and C. Beierkuhnlein. 2011. Climate extremes initiate ecosystem□regulating functions while maintaining productivity. Journal of Ecology 99:689–702.

Jongen, M., C. A. Pio, J. S. Pereira, and L. M. I. Aires. 2011. The effects of drought and timing of precipitation on the inter-annual variation in ecosystem-atmosphere exchange in a Mediterranean grassland.

Knapp, A., S. Conard, and J. Blair. 1998a. Determinants of soil CO2 flux from a sub-humid grassland: Effect of fire and fire history. Ecological Applications 8:760–770.

Knapp, A. K., J. M. Briggs, D. L. Childers, and O. E. Sala. 2007. Estimating aboveground net primary production in grassland and herbaceous dominated ecoystems. Pages 27–48 *in* T. J. Fahey and A. K. Knapp, editors. Principles and Standards for Measuring Net Primary Production. Oxford University Press, Inc. New York.

Knapp, A. K., J. M. Briggs, D. C. Hartnett, and S. L. Collins. 1998b. Grassland Dynamics: Long-Term Ecological Reserach in Tallgrass Prairie. Oxford University Press, Inc., New York.

Knapp, A., and M. Smith. 2001. Variation among biomes in temperal dynamcis of aboveground primary production. Science 291:481–484.

Lloyd, J., and J. A. Taylor. 1994. On the temperature dependence of soil respiration. Functional Ecology 8:315–323.

Mielnick, P. C., and W. A. Dugas. 2000. Soil CO2 flux in a tallgrass prairie. Soil Biology and Biogeochemistry 32:221–228.

Owensby, C. E., J. M. Ham, and L. M. Auen. 2006. Fluxes of CO 2 From Grazed and Ungrazed Tallgrass Prairie Fluxes of CO 2 From Grazed and Ungrazed Tallgrass Prairie 59:111–127.

Pan, Y., R. A. Birdsey, J. Fang, R. Houghton, P. E. Kauppi, W. a. Kurz, O. L. Phillips, A. Shvidenko, S. L. Lewis, J. G. Canadell, P. Ciais, R. B. Jackson, S. W. Pacala, a. D. McGuire, S. Piao, A. Rautiainen, S. Sitch, and D. Hayes. 2011. A Large and Persistent Carbon Sink in the World's Forests. Science 333:988–993.

Poll, C., S. Marhan, F. Back, P. A. Niklaus, and E. Kandeler. 2013. Field-scale manipulation of soil temperature and precipitation change soil CO2 flux in a temperate agricultural ecosystem. Agriculture, Ecosystems & Environment 165:88–97.

Raich, J. W., C. S. Potter, and D. Bhagawati. 2002. Interannual variability in global soil respiration, 1980-94. Global Change Biology 8:800–812.

Reichstein, M., M. Bahn, P. Ciais, D. Frank, M. D. Mahecha, S. I. Seneviratne, J. Zscheischler, C. Beer, N. Buchmann, D. C. Frank, D. Papale, A. Rammig, P. Smith, K. Thonicke, M. van der Velde, S. Vicca, A. Walz, and M. Wattenbach. 2013. Climate extremes and the carbon cycle. Nature 500:287–295.

Reichstein, M., P. Ciais, D. Papale, R. Valentini, S. Running, N. Viovy, W. Cramer, A. Granier, J. Ogée, V. Allard, M. Aubinet, C. Bernhofer, N. Buchmann, A. Carrara, T. Grünwald, M. Heimann, B. Heinesch, A. Knohl, W. Kutsch, D. Loustau, G. Manca, G. Matteucci, F. Miglietta, J. M. Ourcival, K. Pilegaard, J. Pumpanen, S. Rambal, S. Schaphoff, G. Seufert, J. F. Soussana, M. J. Sanz, T. Vesala, and M. Zhao. 2007. Reduction of ecosystem productivity and respiration during the European summer 2003 climate anomaly: A joint flux tower, remote sensing and modelling analysis. Global Change Biology 13:634–651.

Sala, O. E., L. A. Gherardi, L. Reichmann, E. Jobbágy, D. Peters, and E. Jobba. 2012. Legacies of precipitation fluctuations on primary production□: theory and data synthesis 367:3135–3144.

Sala, O., W. Parton, L. Joyce, and W. Lauenroth. 1988. Primary Production of the Central Grassland Region of the United States. Ecology 69:40–45.

Schwalm, C. R., C. A. Williams, K. Schaefer, A. Arneth, D. Bonal, N. Buchmann, J. Chen, L. E. Beverly, A. Lindroth, S. Luyssaert, M. Reichstein, and A. D. Richardson. 2010. Assimilation exceeds respiration sensitivity to drought□: A FLUXNET synthesis. Global Change Biology 16:657–670.

Shi, Z., M. L. Thomey, W. Mowll, M. Litvak, N. A. Brunsell, S. L. Collins, W. T. Pockman, M. D. Smith, A. K. Knapp, and Y. Lou. 2013. Differential effects of extreme drought and respiration: Synthesis and modeling analysis. Biogeosciences Open Discu:1–39.

Wan, S., and Y. Luo. 2003. Substrate regulation of soil respiration in a tallgrass prairie: Results of a clipping and shading experiment. Global Biogeochemical Cycles 17:1–12.

Welp, L. R., J. T. Randerson, and H. P. Liu. 2007. The sensitivity of carbon fluxes to spring warming and summer drought depends on plant functional type in boreal forest ecosystems. Agricultural and Forest Meteorology 147:172–185.

White, R., S. Murray, and M. Rohweder. 2000. Pilot Analysis of Global Ecosystems: Grasslands Ecosystems.

Woodhouse, C., and J. Overpeck. 1998. 2000 years of drought variability in the central United States. Bulletin of the American Meteorological Society 79:2693–2714.

Yahdjian, L., O. Sala, and A. Austin. 2006. Differential controls of water inputs on litter decomposition and nitrogen dynamics in the Patagonian Steppe. Ecosystems 9:128–141.

Zhang, L., B. K. Wylie, L. Ji, T. G. Gilmanov, and L. L. Tieszen. 2010. Climate-Driven Interannual Variability in Net Ecosystem Exchange in the Northern Great Plains Grasslands. Rangeland Ecology & Management 63:40–50.

Chapter 6.

CONCLUSION

The objectives of my dissertation were (1) to assess the resistance and resilience of a tallgrass prairie ecosystem to the independent and combined effects of heat waves and drought and (2) to examine the mechanisms behind these dynamics to improve our theoretical understanding of how extreme events can shape ecosystem structure and function and (3) to determine how key carbon cycling processes respond to extreme heat and drought. To accomplish this, I conducted a three-year, field-based study, where I experimentally imposed two years of extreme heat and drought over an intact native grassland community, then followed recovery for one subsequent year. I examined biotic response variables that ranged across several hierarchical levels including: ecophysiology, demography, community, and ecosystem, in order to gain a comprehensive understanding how this ecosystem responds to such extremes.

In Chapter 2, I contrasted the sensitivities of the two dominant grasses, *Andropogon gerardii* and *Sorghastrum nutans* to a growing season-long drought during in which a two-week heat wave occurred at four intensities. I hypothesized that drought would have greater impacts than heat waves on the ecophysiological and productivity responses of the two grasses with the combined effects of the two climate drivers greater than either alone, and that the dominant grasses would differ in their ecophysiological and productivity responses to heat and water stress. My results showed that drought reduced net photosynthesis and aboveground net primary production (ANPP) in both species, however heat had no independent effects. Unlike previous

work, (De Boeck et al. 2010) there were also no additive effects of heat and drought, however there was an interaction between heat and drought whereby heat-induced soil drying may have indirectly reduced photosynthesis in the control treatment. In addition, *S. nutans* was more sensitive to drought than *A. gerardii* but with no observed differential sensitivity to heat between the two species, with the former supporting and the later contradicting previous studies (Silletti and Knapp 2002, Swemmer et al. 2006, Nippert et al. 2009). The broad conclusion of this chapter was that water stress will likely dominate the ecophysiological and productivity responses of these two key species in the future through direct effects of drought and indirect effects of heat.

I further explored the mechanisms driving the photosynthetic responses of *A. gerardii* and *S. nutans* to heat in Chapter 3, by removing the interactive effects of heat-induced water loss during the simulated heat wave, a challenge in past experiments (Milbau et al. 2005, Marchand et al. 2006, De Boeck et al. 2011). This enabled me to explore the independent effects of heat on net photosynthesis under high and low soil moisture regimes. Under low soil moisture, there were no effects of heat on net photosynthesis for either species, despite extremely high temperatures, due to stomatal limitations and already low photosynthetic rates. On the other hand, both species had a significant relationship between heat and net photosynthesis under high soil moisture, which appeared to be driven by the direct effects of heat. These results suggest that heat waves will have little impact on photosynthesis under extreme drought, but cause moderate decreases in photosynthesis under high soil moisture, unless heat-induced water losses eventually lead to stomatal limitations on photosynthesis.

In Chapter 4, I assessed the resistance and resilience of the tallgrass prairie ecosystem to two years of extreme heat and drought and then evaluated resilience one year following. This

study also allowed me to use the tallgrass prairie as a model ecosystem to test Smith's (2011) extreme climatic event theoretical framework and proposed mechanisms. The results demonstrated that the ecosystem was resistant to heat waves but not drought. After two years of extreme drought, total ANPP was reduced below any level observed in almost thirty years. However this ecosystem had high resilience, completely recovering total ANPP in just one-year post-drought. Using long-term records of biotic and climate data, I was able to explicitly attribute the extreme ecological response of low productivity during the second year of drought to an extreme climate driver and thus identifying an extreme climatic event *sensu* Smith (2011). Next, I examined community and demographic responses to determine mechanisms behind the ecosystem-level responses and found that two dominant species played critical roles in the resistance and resilience of this system. The lack of resistance was attributed to the sensitivity of the dominant forb, *Solidago canadensis*, while the dominant grass, *A. gerardii* provided the ecosystem resilience through a rapid demographic response in the year following drought, replacing the loss of the forb. The results from this chapter suggest that this system has low resistance but high resilience to extreme drought, and that the traits of the dominant species were key to community and ecosystem responses.

Finally, in Chapter 5, I examined how another important ecosystem function, soil respiration, responded during the two years of extreme and one year post-extreme. I hypothesized that drought would have greater impacts than heat, that the sensitivity of heat would be dependent on soil moisture, and that ANPP would be more responsive to drought than soil respiration. As with ANPP, there were significant effects of drought but no independent or additive effects of heat on soil respiration. Surprisingly, even when controlling for soil moisture, soil temperature had little to no effects on soil respiration, a result that deviates from previous

126

work at this site (Harper et al. 2005). While both ANPP and soil respiration decreased with drought, ANPP showed greater sensitivity during the second, more extreme year of drought. But while ANPP fully recovered in the year following drought, soil respiration rates were actually higher in the formerly droughted plots relative to the control. Overall, these results suggest that during the growing season, soil respiration may be less sensitive to temperature than previously reported, and that extreme drought has strong but differential impacts on two key carbon cycling process in this ecosystem.

There are three general conclusions from this dissertation. First, low ecosystem resistance to drought does not preclude rapid recovery in function, which may be an important trait for ecosystem with a history of climate extremes. Although the second year of drought elicited an extreme ecological response – productivity was lower than ever recorded – the ecosystem was able to fully recover function the second year due to the rapid demographic response of the dominant C_4 grass. Secondly, these results emphasize the importance of dominant species responses to climate extremes in governing the resistance and resilience of ecosystems to such events. Many related experimental manipulations studies, impose extreme climate events in artificial communities that are designed with high evenness (Van Peer et al. 2004, Jentsch et al. 2011, De Boeck et al. 2011). Such approaches may overemphasize the importance of species that would naturally be rare in the community, while underemphasizing the importance of dominant species, as seen in studies on natural communities (Kardol et al. 2010, Arnone et al. 2011, Sherry and Arnone 2012). Finally, in this tallgrass prairie ecosystem, the ecological effects of drought exceed those of heat waves across several key carbon cycling processes (photosynthesis, ANPP and soil respiration), and many cases heat had no effect alone or when combined with drought. This may be a consequence of the interactions between heat and drought treatments – high soil

moisture in the control precipitation treatment provided resistance to heat, while low soil

moisture in the drought treatment prevented any measurable responses to heat stress. Overall,

these results suggest that the tallgrass prairie is more vulnerable to short-term extreme drought

than heat waves, and that dominant species play a key role in the resistance and resilience of this

ecosystem.

References

Arnone, J. A., R. L. Jasoni, A. J. Lucchesi, J. D. Larsen, E. A. Leger, R. A. Sherry, Y. Luo, D. S. Schimel, and P. S. J. Verburg. 2011. A climatically extreme year has large impacts on C4 species in tallgrass prairie ecosystems but only minor effects on species richness and other plant functional groups. Journal of Ecology 99:678–688.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2010. Climatic characteristics of heat waves and their simulation in plant experiments. Global Change Biology 16:1992–2000.

De Boeck, H. J., F. E. Dreesen, I. A. Janssens, and I. Nijs. 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. The New phytologist 189:806–17.

Harper, C. W., J. M. Blair, P. A. Fay, A. K. Knapp, and J. D. Carlisle. 2005. Increased rainfall variability and reduced rainfall amount decreases soil CO_2 flux in a grassland ecosystem. Global Change Biology 11:322–334.

Jentsch, A., J. Kreyling, M. Elmer, E. Gellesch, B. Glaser, K. Grant, R. Hein, M. Lara, H. Mirzae, S. Nadler, L. Nagy, D. Otieno, K. Pritsch, U. Rasher, M. Schadler, M. Schloter, B. Singh, J. Stadler, J. Walter, C. Wellstein, J. Wollecke, and C. Beierkuhnlein. 2011. Climate extremes initiate ecosystem☐regulating functions while maintaining productivity. Journal of Ecology 99:689–702.

Kardol, P., C. E. Campany, L. Souza, R. J. Norby, J. F. Weltzin, and A. T. Classen. 2010. Climate change effects on plant biomass alter dominance patterns and community evenness in an experimental old-field ecosystem. Global Change Biology 16:2676–2687.

Marchand, F. L., M. Verlinden, F. Kockelbergh, and B. J. Graae. 2006. Disentangling effects of an experimentally imposed extreme temperature event and naturally associated. Functional Ecology 20:917–928.

Milbau, A., L. Scheerlinck, D. Reheul, B. De Cauwer, and I. Nijs. 2005. Ecophysiological and morphological parameters related to survival in grass species exposed to an extreme climatic event. Physiologia Plantarum 125:500–512.

Nippert, J. B., P. a. Fay, J. D. Carlisle, A. K. Knapp, and M. D. Smith. 2009. Ecophysiological responses of two dominant grasses to altered temperature and precipitation regimes. Acta Oecologica 35:400–408.

Van Peer, L., I. Nijs, D. Reheul, and B. De Cauwer. 2004. Species richness and susceptibility to heat and drought extremes in synthesized grassland ecosystems: compositional vs physiological effects. Functional Ecology 18:769–778.

Sherry, R., and J. Arnone. 2012. Carry over from previous year environmental conditions alters dominance hierarchy in a prairie plant community. Journal of Plant Ecology 5:134–146.

Silletti, A., and A. Knapp. 2002. Long-term responses of the grassland co-dominants Andropogon gerardii and Sorghastrum nutans to changes in climate and management. Plant Ecology 163:15–22.

Smith, M. D. 2011. An ecological perspective on extreme climatic events: a synthetic definition and framework to guide future research. Journal of Ecology 99:656–663.

Swemmer, A., A. Knapp, and M. Smith. 2006. Growth responses of two dominant C4 grass species to altered water availability. International Journal of Plant Sciences 167:1001–1010.

Tilman, D., and J. Downing. 1994. Biodiversity and stability in grasslands. Nature 367:363–365.