THE IMPACT OF CHANGING PRECIPITATION ON WATER AND CARBON CYCLING IN SEMIARID GRASSLANDS OF THE COLORADO FRONT RANGE

by

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The impact of changing precipitation on water and carbon cycling in the semiarid grasslands of the Colorado Front Range

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Regional climate models project that precipitation in the Great Plains of North America will become characterized by more intense rainfall events separated by longer dry periods. Changing seasonal precipitation patterns may differentially favor grassland productivity in ecosystems dominated by either cool or warm season grass species, and thus influence carbon uptake and loss in these systems. Furthermore, model estimates of ecosystem respiration based primarily on soil temperature could overestimate respiration by failing to account for the effects from saturated conditions during heavy precipitation events. This research contrasted water and carbon fluxes during two years with different intra-annual precipitation within a cool season mixed grassland and compared to a neighboring warm season grassland in Rocky Flats National Wildlife Refuge, Colorado, USA. Results from this study showed a significant positive relationship between the accumulated April/May precipitation and growing season carbon uptake in the cool season, smooth brome-dominated grassland. In addition, significant rainfall in the autumn of 2013 played a role in the early spring growth and carbon uptake in 2014. Comparisons between eddy covariance and soil flux-gradient observations and model estimates of soil respiration showed that during the extreme precipitation event in September 2013, processed-based models better characterized fluxes as compared to empirical models based on soil temperature. The study also found that the cool season grassland was a net
sink of carbon during the spring and autumn whereas the neighboring warm season tallgrass prairie was a net sink during the summer. In addition, the study found that the grasslands had considerably different sensitivities to water limitations, with grasses in the tallgrass prairie having a higher water use efficiency ($WUE$). The comparison of the adjacent semiarid grasslands at Rocky Flats NWR improves our understanding of the response to changing precipitation between cool season and warm season dominated grasslands. This research underscores the importance of expanding grassland research to understand how the composition of grasses will influence carbon cycling, especially as precipitation patterns shift with changing climate. Moreover, this research will add to observations during extreme precipitation events, which can improve both empirical and process-based models of soil respiration.
Dedication

To my brother, Jerry F. Moore, who not only mentored his big sister, but also had to endure reading about water, soil, and grass. Sorry, bro. And to Dad, who planned my PhD since childhood and would have been proud.
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CHAPTER 1. INTRODUCTION

The hydrologic and carbon cycles are strongly linked in terrestrial ecosystems, and are particularly pronounced in water-limited grassland ecosystems. Grasslands, which cover nearly 30% of land area globally, have the greatest interannual variability in aboveground productivity when compared to other biomes (Knapp and Smith, 2001; Loveland et al., 2000). Grasslands are extensive biomes and the soils in these systems are very carbon rich due to highly productive grasses that add abundant fresh biomass to the soils each season (Knapp and Smith, 2001). Grasslands have the potential to absorb vast amounts of atmospheric CO$_2$ and store large amounts of carbon, but the carbon balance is dependent on the hydrologic cycle (Harper et al. 2000). The uptake of carbon by plants drives water movement on land, where photosynthesis is largely responsible for transferring water between the soil, vegetation, and the atmosphere through transpiration. Carbon loss from soil respiration is regulated by temperature and water, where increases in soil water content improves microbial access to substrates for decomposition. While the timing and magnitude of water movement through a grassland ecosystem tells us a lot about the carbon movement, there remains significant uncertainty in how water inputs from precipitation are linked to carbon cycling, especially ecosystem respiration.

Extreme precipitation events in the US are forecasted to increase with climate change; more intense/larger events will be separated by longer dry periods (Christensen et al., 2013; Dai, 2011). Terrestrial carbon cycling is largely controlled by water availability in roughly half of the world’s land area (Heimann and Reichstein, 2008; Knapp and Smith, 2001; Sala et al., 1988), including many grassland ecosystems. Water-limited ecosystems tend to respond strongly to changes in soil water content. However, as
precipitation events become larger, soil water could be recharged in deeper soil layers, becoming available to plants longer in the growing season and/or stimulating deeper soil microbial respiration.

Grass species of differing photosynthetic types also respond differently to changes in air temperature and precipitation throughout the season due to cool or warm season phenological responses (Ehleringer, Cerling, and Helliker, 1997; Epstein et al., 1997; Seastedt et al., 1994). Cool-season (C_3) grasses are more productive at lower temperatures, during the spring and autumn, and warm-season (C_4) grasses prosper at higher temperatures found in the summer. The seasonal timing of precipitation is therefore a key determinant of productivity if grasslands are dominated by grass species belonging to one of these photosynthetic types (Epstein et al., 1997).

Although there have been many studies on grasslands and their response to changes in precipitation and water availability (Bahn et al., 2010; Davidson, Belk, and Boone, 1998; Harper et al., 2005; R Vargas et al., 2010), the temporal resolution is frequently very coarse (daily or less often sampling), likely missing short-term fluctuations in soil water and respiration. In addition, few studies compare the response of plants from different photosynthetic types in the same area, and since the timing of precipitation events may determine the grassland’s net uptake or loss of carbon for a given year, these differences could be significant. Finally, past observations are largely inadequate for estimating how arid and semiarid grassland will respond to more extreme precipitation events and measurements of soil respiration responses to extremely large precipitation events are rare. Therefore, there remains significant uncertainty as to how precipitation changes will affect different grasslands in the US Great Plains in the future.
Given that the two grassland communities that were the focus of the study at Rocky Flats National Wildlife Refuge (NWR) represent ecosystems dominated by contrasting photosynthetic types, the objectives of this research were to answer the following questions:

1. How does the timing and intensity of precipitation affect soil water and carbon exchange in semiarid grasslands?
2. How do extreme precipitation events impact carbon exchange in semiarid grasslands and how do these impacts differ from those of typical rainfall events?
3. How do soil water and carbon exchange compare between neighboring C₃ and C₄ dominated semiarid grasslands?

In order to answer these questions, this study examined the seasonality of water inputs from different precipitation patterns, the magnitude and duration of changes in soil volumetric water content (θ), aboveground changes in biomass and leaf area index (LAI), changes in evapotranspiration (ET) and NEE, the impact of changing θ on soil [CO₂], and how these observations compare to modeled estimates based on soil temperature (Ts). Additionally, contrasting the seasonal water and carbon fluxes between a C₃-dominated, mixed grassland and a neighboring C₄-dominated, native tallgrass prairie showed how the timing of precipitation events lead to soil water availability that produced different responses between the grasslands, likely due to the nature of the different photosynthetic types of the grass species.

The first data chapter (Chapter 2) examines the temporal dynamics of water and carbon at a smooth brome-dominated (C₃-dominated) mixed grassland in Rocky Flats
NWR during calendar years 2013-2014. This chapter contrasted the substantial differences in net ecosystem exchange of CO$_2$ in the same grassland between two consecutive years with different intra-annual precipitation and examined the specific seasonal drivers that may explain these differences. The study utilized short-term field and lab experiments to develop a water balance that characterized in-flows and out-flows of water throughout each year and the associated effect on fluxes of CO$_2$ – uptake by photosynthesis and loss through ecosystem respiration. Net ecosystem exchange was measured with the eddy covariance method and belowground fluxes were measured through the soil gradient method. Sampling of biomass and data obtained from NASA’s MODIS Land Product Subsets were used to analyze changes in aboveground biomass.

Chapter 3 examined the fluxes of water and ecosystem respiration ($R_{eco}$) that took place during the extreme precipitation event in the Colorado Front Range in September 2013. The growing season leading up to the event had a lower than average rainfall and the system, while not stressed, was water-limited and was a net source of carbon to the atmosphere. During the week of heavy rains, the relationship between precipitation, $\theta$, soil [CO$_2$] and $T_s$ were examined on an hourly basis. A week of heavy rainfall during July 2014 was used to compare these responses. The DAMM process-based model was adapted for the week of the extreme events, and all observations were compared to this and empirical model estimates.

Chapter 4 compared seasonal variations in gross primary productivity ($GPP$) and water use efficiency ($WUE$) between two grassland ecosystems to different precipitation patterns in 2013 and 2014. Analysis of the temporal relationships between precipitation, $\theta$, $T_s$, $LAI$, biomass, $NEE$, and $GPP$ were utilized to demonstrate the contrasting seasonal
productivity and water sensitivities between a cool-season (C₃) dominated grassland and a neighboring warm-season (C₄) dominated tallgrass prairie.

This thesis concludes in chapter 5 with the central findings of this study, policy implications and suggestions for further research. In particular, there are many opportunities for land management strategies through US Fish and Wildlife (the agency managing Rocky Flats NWR) and, more broadly, for other governmental bodies that develop or maintain grasslands within the U.S. Great Plains region.
CHAPTER 2. PRECIPITATION DRIVEN CARBON CYCLING IN A SEMIARID SMOOTH BROME-DOMINATED MIXED GRASSLAND

2.1 Introduction

Semiarid grasslands of the U.S. Great Plains include a broad range of plant species compositions which in turn have complex carbon exchange responses to rain events of different duration and size (Cherwin and Knapp, 2012; Gilmanov et al., 2005; Knapp et al., 2002; Tieszen et al., 1997). A nonnative grass, smooth brome (*Bromus inermis*), was seeded extensively in the U.S. and Canada in the late 1800s for a variety of land management objectives (e.g., revegetation, soil stabilization, forage) and has subsequently overrun many native prairies and grasslands, creating low diversity systems (Otfinowski, 2007; Vinton and Goergen, 2006). In addition, decades of cultivating and grazing in North America have not only changed plant compositions but have altered the soil texture, carbon and nitrogen content substantially. Complicating matters, this region is forecasted to undergo seasonal shifts in precipitation (Zeppel, Wilks, and Lewis, 2013), which may further alter water and carbon responses in semiarid grasslands and their associated feedbacks to climate.

The legacy of land-use change in North American grasslands is considerable. A study in the Pawnee National Grasslands in northeastern Colorado found that soil organic matter in abandoned cultivated fields took about 50 years to return to comparable levels of fertility, but were still lower in total organic content than the adjacent native grasslands (Burke, Lauenroth and Coffin 1995). In addition, recent efforts to restore native prairies confound the relationship between carbon exchange and changing precipitation regimes as new species compositions and the various management techniques used in the Great
Plains contribute to the ecosystem response (Dobson et al., 1997; Grman et al., 2013; Phillips and Beeri, 2008).

Regional climate change projections suggest there will be both an increase in drought conditions and more extreme precipitation events in some areas of the U.S. (Christensen et al., 2013; Dai, 2011). Intra-annual variability in precipitation has been projected to increase, with large rain events separated by longer dry intervals. As warming causes an increase in evaporation and vegetation responds by utilizing ever more soil water reserves, precipitation becomes increasingly crucial for recharging water storage in soils (Porporato, Daly and Rodriguez-Itrbe, 2004). Furthermore, the timing and magnitude of precipitation affects soil water pulses in arid and semiarid ecosystems that are linked to nitrogen cycling, which is vital to vegetation and soil microbial productivity (Dijkstra et al., 2012). Thus, water availability in these systems controls net ecosystem exchange (NEE) of CO₂, which is a balance between inputs from gross primary productivity (GPP) and losses from ecosystem respiration (Reco) (Reichstein et al., 2005):

\[ NEE = \text{R}_{\text{eco}} - \text{GPP} \]  \hspace{1cm} (2.1)

thus, GPP values greater than R_{eco} result in negative values of NEE, which represent CO₂ uptake. Productivity in central U.S. grasslands is broadly correlated with mean annual precipitation – the lowest aboveground primary productivity occurs in the western region where the annual rainfall is also lowest (Sala et al., 1988). Studies by Epstein et al. (1997) and Tieszen et al. (1997) also found an increase in productivity that was strongly influenced by seasonal, abiotic factors (precipitation, temperature, humidity, etc.) that
favor the photosynthetic type of grasses that dominate regions of the south (warm-season or C4) and the north (cool-season or C3).

However, an increase in the intra-annual variability of precipitation has distinctly different impacts on net primary production (NPP, GPP – autotrophic respiration) from mesic (moderately moist) to arid grasslands in the Great Plains. A rainfall pattern consisting of less frequent but larger precipitation events caused a reduction in aboveground net primary production (ANPP or NPP of aboveground biomass), in a mesic tallgrass prairie in Kansas (Knapp et al., 2002). Yet a study conducted by Heisler-White et al. (2008) in a northern Colorado shortgrass steppe ecosystem showed that a similar change in rainfall pattern resulted in higher rates of ANPP. Furthermore, a study by Parton et al. (2012) at the same site in Colorado determined that large precipitation events (> 10 mm day⁻¹), specifically during May to June, were crucial for optimizing ANPP and resulted in an annual carbon sink. However, precipitation reductions did not always result in less ANPP in three shortgrass steppes from Colorado to northern New Mexico, indicating how difficult it is to link precipitation to soil water stores and, ultimately, to how grasses utilizes available water in dry ecosystems (Cherwin and Knapp, 2012).

Grasslands are capable of sizable respiratory losses in response to large precipitation events, though when balanced with an increase in primary productivity, the net annual exchange can be a carbon sink (Gilmanov et al., 2005; Knapp and Smith, 2001). A study by Zhou, Talley, and Luo (2009) found increasing mean annual precipitation (MAP) across Oklahoma caused an increase in respiration in three grassland types - short-grass steppe, mixed-grass prairie, and tallgrass prairie. Likewise, daily respiration rates were positively correlated with winter snow depth and summer rain
amounts in a mixed-grass prairie in southern Wyoming (Chimner et al., 2010; Chimner and Welker, 2005). Furthermore, soil respiration was reduced by lower growing season precipitation amounts and increased rainfall variability in the mesic tallgrass prairie site in Kansas (Harper et al., 2005; Knapp et al., 2002). Respiration was found to increase with increasing precipitation event size, even after a fire burned an arid grassland in the northern Chihuahuan Desert in New Mexico (Vargas et al., 2012).

Nevertheless, few studies exist that measured net ecosystem exchange of carbon in semiarid grasslands dominated by smooth brome (Phillips and Beeri, 2008; Polley et al., 2010), particularly in the western Great Plains, and there is interest in understanding the carbon sequestration potential of these grasslands by land managers, especially the Conservation Reserve Program (CRP) (Gelfand et al., 2011). The few studies located in semiarid grasslands that utilized rainfall manipulation experiments altered events equally across the time period of interest (Chimner et al., 2010; Chimner and Welker, 2005; Heisler-White et al., 2008), which make it challenging to relate these results to the response that a nonnative, cool-season grassland will have to both seasonal shifts in precipitation amounts and random extreme precipitation events.

The objective of this research was to determine how the timing of intra-annual precipitation affects the annual carbon exchange in a semiarid mixed grassland. Therefore, answers to two questions were sought: 1) what is the timing for seasonal and monthly precipitation that leads to greater carbon uptake? and; 2) is substantial early season or spring rainfall essential to result in an annual carbon sink? Given that the cool-season perennial grass, smooth brome, currently dominates the semiarid grassland in this study, two hypotheses were proposed:
H1: Primary productivity will be greatest in the cooler growing season periods, spring and autumn; therefore soil water recharge from precipitation will be crucial during these periods.

H2: Large precipitation inputs early in the growing season (March-May) are essential to achieve an annual carbon sink for this ecosystem because the dominant C₃ grass, smooth brome, is most productive in a narrow period in the spring, and thus water demand is highest during this period.

To test these hypotheses, the research utilized continuous aboveground measurements of water and carbon fluxes and discrete sampling of biomass with belowground measurements of soil CO₂ concentration and soil water and temperature during two sequential calendar years with annual precipitation above the 30-year mean, yet with distinct intra-annual precipitation patterns.

2.2 Methods

2.2.1 Study Site

The study was conducted in a semiarid mixed grassland located about 18 km northwest of Denver, CO, USA in the southeastern section of Rocky Flats National Wildlife Refuge (NWR) (39°53'N latitude, 105°12'W longitude) during the 2013 and 2014 calendar years. The refuge (approximately 2500 ha) surrounds an area that once
housed a nuclear weapon components assembly plant, the Department of Energy (DOE) retained area, which is now closed permanently after an extensive environmental cleanup during the late 1990’s and early 2000’s (EPA, 2006, Figure 2.1). The area encompassing the wildlife refuge remained undeveloped for decades as a buffer zone around the industrial area. It is currently managed by the U.S. Fish and Wildlife Service to conserve the natural habitats (USFWS, 2005).

The climate is classified as semiarid, with a mean annual temperature (MAT) of 8.2 °C and mean annual precipitation (MAP) of 426 mm (30-year averages 1981-2010, PRISM Climate Group, Oregon State University, http://prism.oregonstate.edu). The elevation of the site is approximately 1730-m asl and the topography is level, with 0-3 degree slopes. The soils are the Denver-Kutch-Midway complex, derived from colluvium, which consists of surface soils with a loamy texture and clayey soils beneath (USDA, 2011).
The mixed grassland site was part of a broader, ~640 acre region of the refuge which the U.S. Fish and Wildlife Service (USFWS) has classified as a reclaimed mixed grassland since the area was originally cultivated and then re-seeded over a decade ago with a mixture of nonnative/introduced species (USDA, 2014; USFWS, 2005). The
vegetation within the sampling area consists of predominantly perennial, cool season (C₃) grasses, with 70-80% smooth brome (*Bromus inermis* Leyss), 10-20% intermediate wheatgrass (*Thinopyrum intermedium*), and other plant species in varying amounts, including crested wheatgrass (*Agropyron cristatum*), field bindweed (*Convolvulus arvensis*), and sweetclover (*Melilotus officinalis*). The roots of smooth brome are generally concentrated in the top 10 cm of soil (Craine and Wedin, 2002; USDA, 2014), and the vegetation at the site in Rocky Flats NWR reached a maximum height of ~90 cm between June and July during the two years of this study. The grass had a substantial ground cover with a high tiller density, standing dead biomass that endured through the winter months, and a 5-20 cm litter layer covering the soil (Figure 2.2).
Figure 2.2. Photos of the mixed grassland site (top) on February 7, 2013, from ~50 cm above the land surface looking south towards the eddy covariance tower, and (bottom) on May 28, 2013, within the grass canopy, with a soil gas well access valve center frame.

2.2.2 Soil Moisture and Temperature

Two soil moisture and temperature plots were established at the study site – the north plot consisted of two EC-5 soil moisture (capacitance/frequency domain) sensors buried at 5 and 10 cm, and the south plot was instrumented with one EC-5 sensor at 5 cm, one EC-10 sensor, angled at ~45 degrees, from 10 to 15 cm soil depth, and one RT-1 temperature probe buried at 10 cm (Figure 2.3, all sensors and data loggers Decagon Devices, Inc., Pullman, WA 99163). All sensors measured every 60-seconds and the average was recorded every 30-minutes on an Em5b data logger. Where replicated, soil
volumetric water content ($\theta$) measurements for each soil depth were averaged between the two plots.

Soil plot data were filtered for missing or bad records due to sensor disturbance from animals or data logger power issues (13.3% missing records for 2013; 2% for 2014) and gap-filled with available $\theta$ measurements from the eddy covariance station (see section 2.2.5 Eddy covariance below). The integrated (0 – 15 cm) volumetric water content ($\theta_{0-15cm}$) was averaged across the three sensor depths. Soil temperature ($T_s$) reported and used for analysis in this study is the 30-minute averaged measurements from the 10 cm soil depth (°C).

A soil matric potential sensor (227-L, Campbell Scientific, Inc., Logan, UT, USA) was installed to aid in assessing the soil water availability to plants. The sensor was placed ~4 m south of the north plot, buried at 5 cm soil depth, and connected to a separate data logger system (see section 2.2.3 Soil CO$_2$ concentrations below), where measurements were recorded every hour. The lowest value in the measurement range of the sensor, -1 MPa, is approximately equivalent to a $\theta_{0-15cm}$ value of 8% (n = 242, s.d. = 0.03) for the soils at this grassland. It was observed that when $\theta_{0-15cm}$ values dropped below 10% during the growing season, NEE became positive (losing carbon). Additionally, soil matric potential measurements in the range of -0.8 to -1.4 MPa can cause root death in some cool season grass species (Frank et al., 1996). Therefore, $\theta_{0-15cm}$ values of 8% or less were used in this study’s analysis to approximate when soil water stores were becoming depleted and less available to plants.
2.2.3 Soil CO$_2$ concentrations

Hourly measurements of soil CO$_2$ concentrations were obtained from soil gas wells. All soil gas wells and the automated gas analyzer sampler system were designed, constructed, and tested initially at the U.S. Geological Survey (USGS) in Denver, CO., and a subset was installed and tested at the site during September 2012. Final installation
took place during December 2012 and January 2013, where six soil gas wells were divided between two separate plots (north and south plots) ~12 meters apart, and buried at three shallow depths (5, 10 and 15 cm) in each plot. Each soil gas well had an approximate volume of 120 cm$^3$ and was constructed from 1/2” diameter schedule 40 polyvinyl chloride (PVC) tubing (1.58 cm i.d.) cut to 0.6- m length and with 24 evenly spaced holes (approx. 1 cm dia.) drilled along the length of the PVC tubing and positioned to rest along the bottom of the gas well. Each tube was wrapped in fiberglass screen to exclude large debris while allowing the flow of gasses once installed in the soil. Each gas well was connected to the surface through stainless steel tubing and then polyethylene lined plastic tubing (Bev-a-Line) that ran along the ground inside a conduit to a sampler system.

Measurements of soil CO$_2$ concentration were made on the hour by a gas analyzer system (the auto-sampler) located at the land surface. This “auto-sampler” utilized a Li-800 Infrared Gas Analyzer (LI-COR Biosciences) attached to a multiplexer, a 16-channel AC/DC controller (SDM-CD16AC) and controlled by a CR-10X data logger (Campbell Scientific, Inc., Logan UT, USA). In order to improve the resolution, the IRGA was set up to measure CO$_2$ concentrations from 0-5000 ppm, a trade-off that was decided on during the field test in 2012 when even very moist soil conditions rarely resulted in concentrations above 3500 ppm. Gas wells were sampled one at a time, where the multiplexer controlled solenoids to select the line to be sampled, and a pump and flow meter were attached to the outflow line from the gas analyzer to create positive pressure and a flow rate of approximately 400-450 mL min$^{-1}$ into the analyzer. The soil matric potential sensor was excited, measured and recorded as the last reading in the sampling
cycle. Gas samples were filtered on the intake before entering the analyzer (microfiber filter, 0.1 L internal volume, Balston, DFU – Parker Hannifin Corporation). See APPENDIX 5 for complete details.

2.2.4 The flux gradient method calculations

The gradient method is a flux calculation that is a function of the changing soil CO\textsubscript{2} concentration, and soil moisture and temperature profile - a vertical gradient of measurements at different soil depths. Calculations of soil CO\textsubscript{2} flux to the surface can be made and expressed as surface area fluxes that are comparable with eddy covariance and other carbon flux estimates. The soil CO\textsubscript{2} diffusion rate is obtained from Fick’s Law:

$$F = -D \frac{\partial [CO_2]}{\partial z}$$

where $F$ is the gas flux (µmol m\textsuperscript{-2} s\textsuperscript{-1}), $D$ is the CO\textsubscript{2} diffusion coefficient in soil (m\textsuperscript{2} s\textsuperscript{-1}), $\partial [CO_2]$ is the change in CO\textsubscript{2} concentration (µmol m\textsuperscript{-3}) with a change in soil depth $\partial z$ (m). The soil CO\textsubscript{2} diffusion coefficient for this study was estimated based on the CO\textsubscript{2} diffusion coefficient in free air, adjusted for changes in air pressure and temperature, and the ratio of the soil air-filled pore space to total porosity, which in turn is a function of $\theta$ (Moldrup et al. 1999; Tang et al. 2003). Soil samples were collected from the site from the 0 to 10 cm soil depths and porosity for this soil was obtained by averaging the volume of water needed to completely saturate undisturbed, dried soil samples (porosity = 51%, see 5.1.4APPENDIX 4).
Soil flux gradient values were calculated for each hour using values of soil \([CO_2]\) from the soil gas wells to generate the \(
\frac{\partial [CO_2]}{\partial z}\) ratio and the associated \(\theta\) for each soil depth was used to calculate \(D\). The diffusion coefficient was determined by estimating the ratio of gas diffusion in soil to free air \((D_s/D_{air})\) using several known relationships based on a function of \(\theta\) and porosity to determine the soil air-filled pore space \((\theta_{air})\). Resulting flux calculations were validated against eddy covariance observations and a few chamber measurements made during field-testing. Eddy covariance fluxes that were used for validation were filtered to include only nighttime fluxes (soil respiration) and for values that were not gap-filled. A range of different seasonal conditions and soil moisture values were selected and soil flux gradient calculations were compared to these fluxes. The Millington-Quirk (1960) relationship, based on \(\theta_{air}\) and porosity \((\varphi)\) factor of the \(CO_2\) diffusion in free air \(([\theta_{air}^2/\varphi^{2/3}] \times D_{air})\), was determined to be the best fit for a wide range of seasonal (i.e., \(T_{air}\), humidity, wind, aboveground biomass) and soil temperature and moisture values.

2.2.5 Eddy Covariance station instrumentation

The study site had ongoing eddy covariance-based measurements of surface energy balance, including net ecosystem exchange (\(NEE\)), derived from the turbulent exchange of \(CO_2\) between the atmosphere and land surface. Wind components were measured by a three-dimensional sonic anemometer (CSAT3, Campbell Scientific, Inc., Logan UT, USA), and \(CO_2\) and \(H_2O\) mole densities were measured by an open-path infrared gas analyzer (LI-7500, LI-COR, Lincoln, NE, USA), both instruments were mounted at a height of 1.98 m above the ground, separation distances for the center of the
LI-7500 relative to center of the CSAT path were 12 cm west and 11 cm north, and the CSAT had a north offset of 174 degrees (Figure 2.4). Data were recorded at 10 Hz on a CR-3000 data logger (Campbell Scientific, Inc., Logan, UT, USA) and averaged over 30-minute periods. Air temperature ($T_{air}$) and relative humidity were measured with a shielded sensor mounted at ~ 1.75 m above the ground (HMP45C, Vaisala, Helsinki, Finland), and net radiation was measured with a net radiometer mounted at ~2 m above the ground (model Q*7, Radiation and Energy Balance Systems, Inc., or REBS, Seattle, WA). The soil heat flux was averaged between two heat flux plates (model HFT3, REBS) and thermocouples located 1-m west and east of the tower, buried at 5 cm in the soil.

Figure 2.4. Picture from May 10, 2013 showing the eddy covariance system with soil plots, auto-sampler (rectangle box resting on the ground), and the Em5b datalogger (mounted on a post next to the auto-sampler) in the background, looking towards the east-southeast.
Soil moisture was measured using a TDR-based probe (Time Domain Reflectometry; CS616, Campbell Scientific, Inc., Logan, UT, USA), buried at ~30 degree angle from vertical and integrating soil moisture from the surface to 20 cm soil depth. The eddy covariance station was powered with two deep cycle, flooded, lead acid batteries connected to two 50W solar panels.

Eddy covariance raw data were processed through EddyPro® open source software (https://www.licor.com/env/products/eddy_covariance/eddypro.html, LI-COR, Lincoln, NE) to perform filtering, calibration, and apply algorithms for correcting fluxes (i.e., coordinate system rotation, frequency response, density effects, etc.) (Ivans et al. 2006). Processed eddy covariance data were submitted to Max Planck Institute’s online Eddy covariance gap-filling tool, (http://www.bgc-jena.mpg.de/~MDIwork/eddyproc/) to fill missing NEE and sensible (H) and latent heat (LE) data according to the methods described by Falge et al. (2001). Missing and poor eddy covariance data were caused by a variety of conditions. Short segments of missing data (hourly timescales) were usually due to debris, insects, or rain droplets on the windows of the open-path sensor or were due to measurements made when the wind direction interfered with the tower or mounted instruments and thus created a distortion of the natural airflow. There were larger segments of missing data in late November and throughout December 2013 and January 2014 due to power failures and freezing conditions. Since these winter months typically have the smallest NEE fluxes and were not part of the seasonal periods that are the focus of this study, their estimated fluxes served only to obtain an annual carbon budget. In addition,
the Max Planck flux partitioning calculations were not used in this study (see section 2.2.6 below).

The energy balance closure, or the ratio of available energy, the sum of net radiation \((Rn)\) and soil heat flux \((G)\), to the sum of turbulent fluxes \((H + LE)\), is equal to 1.0 when measurements are perfect. Each 30-minute measurement period was identified as nighttime for solar radiation values \(<= 20 \text{ Wm}^{-2}\). The mean 30-minute energy balance closure for 2013 was 0.987, with a daytime average of 0.80 and a nighttime average of 1.27 \((n = 8506)\). The mean 30-minute energy balance closure for 2014 was 0.91, with a daytime mean of 0.86 and a nighttime mean of 0.95 \((n = 9678)\). These ratios are within the range of values that are considered acceptable (Blanken et al., 1997; Ivans et al., 2006; Wilson et al., 2002). See 5.1.4APPENDIX 2 for more detail.

2.2.6 Partitioning NEE

Partitioning NEE into GPP and \(R_{eco}\) can be determined by first estimating \(R_{eco}\) using the empirical relationship between nighttime NEE and \(T_s\). Nighttime NEE is an approximation of \(R_{eco}\) because at night there is no solar radiation and thus photosynthesis ceases and respiration can be assumed to be the sole flux. The procedure commonly used to estimate \(R_{eco}\) from eddy covariance measurements of nighttime NEE is to determine a threshold of friction velocity \((u^*)\), above which air movement is strong enough to mix the boundary layer above the land surface, reaching the instruments and making turbulent fluxes measurable (Ivans et al., 2006). However, a regression of the nighttime NEE rates as a function of \(u^*\) found that NEE was insensitive to \(u^*\) \((r^2 = 7.9 \times 10^{-5}, n = 209\) for
2013; $r^2 = 0.023$, $n = 192$ for 2014, 5.1.4APPENDIX 3). Therefore, subsequent regressions of nighttime $NEE$ and $Ts$ were not filtered for a threshold $u^*$. Mean nighttime eddy covariance measurements of $NEE$ were regressed against mean nighttime $Ts$ and the regression equation was subsequently used to calculate daytime $R_{eco}$ using the 30-minute averaged measurements of $Ts$ (Lloyd and Taylor, 1994). Nighttime $GPP$ was set to zero and daytime $GPP$ was calculated from $NEE + R_{eco}$. Net ecosystem exchange ($NEE$), $GPP$, and $R_{eco}$ were accumulated over the 30-minute measurement period and expressed as g C m$^{-2}$, and cumulative values were generated from summing all of the 30-minute accumulated measurements for a selected time period.

2.2.7 Precipitation and the Water Balance

Regional precipitation measurements were obtained from the Measurement and Instrumentation Data Center (MIDC) of the National Renewable Energy Lab (NREL), collected continuously from a heated tipping bucket rain gauge (320b, Belfort Instrument, Baltimore, MD) mounted on a tower at the 2-m height ($39^\circ 54' 36.0''N$ $105^\circ 14' 06.0''W$) (Jager and Andreas, 1996), located approximately 5 km northwest of the study site. The NREL observations were used for rainfall analysis and were preferred to the tipping bucket rain gauge located at the grassland study site because the annual precipitation records for 2013-2014 were complete at NREL, and additional meteorological records dating back to 2000 were available for long-term analysis. Given that snowfall is difficult to measure, and winter precipitation amounts were crosschecked against time-lapse images recorded at the grassland site, it was determined that the NREL precipitation data
underreported precipitation in the form of snowfall during January, February, November and December for both calendar years. Precipitation data for these months were therefore substituted with records obtained from a NOAA meteorological station south of the grassland (Arvada 3.8 W, CO US station, 39.8171°, -105.1774°, NOAA, 2015).

The average monthly precipitation, average number of precipitation events, and the number of days between events were all computed. The maximum precipitation event sizes were determined, 30-year historical data obtained, and a plot comparing the current years to past rainfall was generated (Figure 2.5).

![Figure 2.5](image)

Figure 2.5. Plot of monthly precipitation for 2013, 2014, and 30-year mean for the region. Note that the most significant monthly rainfall amount occurred in September 2013 due to the extreme precipitation event that year. May and July 2014 also recorded monthly rainfall totals that were considerably higher than 2013 or the 30-year mean.

A water balance at daily time steps was calculated for the grassland to compare the timing of inflows and outflows of water between calendar years. The volume used for the water balance calculation included the spatial extent of the grassland that contributed
the most to measurements directly related to the flow or storage of water during the study - bounded horizontally by an approximately 625-m$^2$ area centered at the eddy covariance tower (extent of soil sampling) and vertically from 2 meters above the land surface (the height of the tower and above the plant canopy) to a depth of 0.15 m below the land surface (the depth of soil instruments). The following equation was used to quantify inflows and outflows of water to this volume:

$$P - ET = \Delta S + L \quad (2.3)$$

where

- $P$ is precipitation,
- $ET$ is evapotranspiration,
- $\Delta S$ is the change in soil water storage (0 – 150 mm),
- $L$ is loss due to runoff or infiltration below the measured soil depth

Evapotranspiration ($ET$) was calculated from tower measurements of the latent heat fluxes (30-minute measurements) and averaged per day (24-hours). To calculated soil water storage ($S$), the product of $\theta$ (m$^3$m$^{-3}$) at each sensor depth and the thickness of the vertical soil section measured by the sensor, expressed in millimeters, were summed:

$$S = [\theta_{0.5cm} \times 50 \, mm] + [\theta_{5-10cm} \times 50 \, mm] + [\theta_{10-15cm} \times 50 \, mm] \quad (2.4)$$

The change in soil water storage ($\Delta S$) was calculated by using the difference in $S$ between the start and end of the period of interest (month, season, year). Loss ($L$)
represents any runoff or the amount of water permeating the soil layers below 150 mm, beyond the measured depth (15 cm). Runoff is usually assumed to be negligible for this study site, however the extreme precipitation event in September 2013 generated significant amounts of runoff when infiltration was unable to accommodate the water inputs.

2.2.8 Biomass and Leaf Area Index

To characterize changes in aboveground biomass, vegetation samples were collected and analyzed on an approximately bi-weekly schedule during the growing season for each calendar year. Each collection included one sample each from six different 1-m$^2$ grids within a 6,400 m$^2$ area (80 × 80 m) centered around the eddy covariance tower. Sampling grids were selected, a priori, using a custom random number generator algorithm in Microsoft Excel (2010), with each unique grid location identified by two cardinal directions (90 degrees from each other) and two distances, in units of meters (i.e. N30W80). The algorithm was run until all four quadrants of the sampling area (NE, SE, SW, NW) had at least one sample location defined. The sampling area was marked by stakes, placed at 10-m intervals from 10 to 80, on one of four lines radiating out from the eddy covariance tower, in each of the four cardinal directions (N, E S, W, Figure 2.6). A 0.2-m diameter metal ring (area = 0.0314 m$^2$) was randomly placed within each of the six 1-m$^2$ sampling grids at the site, from which vegetation samples were clipped, bagged, and transported in a cooler back to the laboratory. Approximately 0.2-m$^2$ composite were sampled every 2 weeks.
Laboratory analysis of vegetation started with measuring and recording the average grass height in each sample. Each sample was examined to identify the grass species and then separated into visibly green leaf fragments and three replicates of one-sided leaf area were measured with a leaf area meter (LI-3100C, LI-COR, Lincoln, NE). Leaf area index (LAI) is a dimensionless metric (leaf area to ground area, m² m⁻²) used as a reference for vegetation canopy and changes in LAI over time are related to plant growth and senescence. To determine dry plant biomass, vegetation samples were weighed, oven dried for over 12 hours at 75 °C, and reweighed at regular intervals until there was no significant change in mass.
To assess long term trends, remotely-sensed $LAI$ records for a wider area ($3 \times 3$-km) centered at the study site were obtained from NASA’s MODIS Land Product Subsets (NASA, 2015), at a time step of ~8 days for the period from 1/1/2005 – 12/31/2014. Estimates of $LAI$ from destructive sampling and $LAI$ from MODIS were associated using dates and MODIS $LAI$ values between dates were interpolated.

2.2.9 Accumulated growing degree days (GDD)

Plant metabolic activity relies on energy inputs in addition to water and nutrients. Growing degree days, or thermal units, are a method of accumulating the amount of energy used for growth over a period of time. The calculation utilizes daily maximum and minimum air temperatures, a threshold-base air temperature at which a species begins to grow, and an upper limit air temperature, above which most organisms cannot grow any faster. Since smooth brome was the dominant grass species at the study site, growing degree days solely for this grass species were calculated, with a base $T_{air}$ of 0 °C (Romo and Eddleman, 1995). The calculation for daily GDD is:

$$GDD = \frac{(Daily \ max \ T \ °C + \ Daily \ min. \ T \ °C)}{2} - Base \ T \ °C$$

(2.5)

The daily GDD value was set to zero for any days where the difference between the daily maximum and minimum air temperature is less than the base air temperature (GDD can only be a positive number). Daily GDD values were summed together for different time periods to attain the accumulated GDD.
2.2.10 Growing season and early and late season periods

For the purpose of this study, the “growing season” will refer to the period from March 1 – October 31 of each year. Given that the dominant grass at the study site was a cool-season grass species, comparisons between the two years focused on specified periods within the growing season where conditions were favorable for vegetation growth (i.e. lower air temperatures). Two 90-day periods were selected based primarily on the historical precipitation and \( T_{air} \), and an examination of the accumulated daily (24-hour) \( NEE \) for the two calendar years of this study. During March-April, carbon uptake caused \( NEE \) to trend towards negative values (i.e. a carbon sink), and August-September can see this trend occur again if conditions are favorable (e.g., sufficient soil moisture, moderate air temperatures). Therefore, the “early season” period was established as April 1 – June 30, and the “late season” period was set to August 15 – Nov 13, both 90-days long and encompassing the highest potential carbon uptake periods.

2.2.11 Calculations / Statistics

Filtered and gap-filled eddy covariance, meteorological, biomass and soil data were all combined using the associated date and time for each record to synchronize temporal observations, and time series plots of these variables were generated. A Pearson product-moment correlation coefficient (Pearson’s r) was computed to assess the relationship between nighttime \( NEE \) and \( Ts \). A simple and step-wise multiple linear, time-lagged regressions of MODIS derived \( LAI \) as a function of monthly and seasonal precipitation were performed using the NREL derived long-term records for 2005 – 2014. Analysis of residuals exhibited random patterns indicating that the employed models
were appropriate. For all statistical analyses, the R statistical software package was used (version 3.2.2; R Development Core Team, 2015).

2.3 Results

2.3.1 Water Balance and Meteorological Data

Air temperatures at Rocky Flats NWR can vary considerably on daily timescales, and the range of $T_{air}$ values was quite broad throughout both years and in all seasons during this study. However, mean daily $T_{air}$ and $T_s$ were similar between the two years and the early and late season periods and a pair-samples t-test showed no significant difference between the mean $T_{air}$ and $T_s$ for the two years of this study. There were more frost-free days in 2014, both annually and in the early and late season periods, yet minimum daily $T_{air}$ dipped briefly, but substantially, lower during the late season in 2014 as compared to 2013. While air and soil temperatures did not significantly differentiate the two years, analysis of water cycling at the site yielded more interesting results. A summary of the relevant water balance and meteorological data are shown in Table 2.1.
Table 2.1. Meteorological and water balance summary for calendar years 2013 and 2014 (annual), early season, and late season periods. Note that the precipitation total for the late season in 2013 was largely due to one-week of extreme precipitation events in September. Change in soil water storage ($\Delta S$) was computed as the difference between the soil water storage ($S$) at the start and end of the period.

<table>
<thead>
<tr>
<th>Dates</th>
<th>Year</th>
<th>Annual</th>
<th>Early Season</th>
<th>Late Season</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of days</td>
<td>2013</td>
<td>365</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>Frost-free days</td>
<td>2013</td>
<td>224</td>
<td>69</td>
<td>73</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>242</td>
<td>78</td>
<td>81</td>
</tr>
<tr>
<td>Mean daily $T_{air}$ (°C)</td>
<td>2013</td>
<td>9.47 (10.2)</td>
<td>12.37 (8.9)</td>
<td>13.75 (7.6)</td>
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<td></td>
<td>2014</td>
<td>9.95 (9.6)</td>
<td>12.88 (6.3)</td>
<td>13.84 (7.7)</td>
</tr>
<tr>
<td>Max daily $T_{air}$ (°C)</td>
<td>2013</td>
<td>34.63</td>
<td>34.62</td>
<td>34.63</td>
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<tr>
<td></td>
<td>2014</td>
<td>34.62</td>
<td>32.60</td>
<td>31.45</td>
</tr>
<tr>
<td>Min daily $T_{air}$ (°C)</td>
<td>2013</td>
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<td>-18.26</td>
<td>-3.68</td>
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<tr>
<td>Mean daily $T_{s}$ (°C)</td>
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<td>11.07 (9.2)</td>
<td>14.46 (7.0)</td>
<td>14.71 (7.0)</td>
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<td></td>
<td>2014</td>
<td>10.98 (8.2)</td>
<td>14.53 (5.4)</td>
<td>14.51 (5.4)</td>
</tr>
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<td>Total precipitation (mm)</td>
<td>2013</td>
<td>676.52</td>
<td>117.71</td>
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<td></td>
<td>2014</td>
<td>537.29</td>
<td>157.51</td>
<td>131.51</td>
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<tr>
<td>Total $ET$ (mm)</td>
<td>2013</td>
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<td>125.41</td>
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<td>2014</td>
<td>680.76</td>
<td>276.79</td>
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<tr>
<td>Change in soil water storage ($\Delta S$, mm)</td>
<td>2013</td>
<td>15.86</td>
<td>-25.36</td>
<td>8.44</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>-0.84</td>
<td>-20.85</td>
<td>1.45</td>
</tr>
<tr>
<td>Days soil water is below 8%</td>
<td>2013</td>
<td>25</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Mean temperatures followed by standard deviation in parenthesis. A paired-samples t-test was conducted to compare mean, maximum and minimum daily $T_{air}$ and mean daily $T_{s}$ between 2013 and 2014. There was no significant difference in any of the mean temperatures for all periods.

Historically, most precipitation for this area falls early in the growing season; the highest monthly rainfall totals occur in the months of March through June, peaking in May with a 30-year average monthly total precipitation of 64 mm. The timing of precipitation at the grassland study sites varied considerably from the 30-year average and between years for the 2013 and 2014 early and late season periods. April (62 mm)
and September (384 mm) had the most monthly rainfall in 2013 while May (104 mm) and July (142 mm) had the highest monthly totals for 2014. Further details regarding the frequency and distribution of precipitation for each month and the associated changes in soil water are specified in Table 2.2.

Table 2.2. Monthly precipitation ($P$), mean daily $P$ and the number of days it rained, mean interval (number of days) between rain events, and soil water changes. The mean interval is computed from the number of precipitation free days divided by the number of dry periods (consecutive days with no rain) for that month. Starting $\theta$ is the soil moisture content at the beginning of the month, and change in soil water storage ($\Delta S$) is the difference between the $S$ from the beginning to the end of the month - positive numbers denote a gain in soil water stores, while negative numbers indicate a loss.

<table>
<thead>
<tr>
<th>Month</th>
<th>Year</th>
<th>Monthly $P$ (mm)</th>
<th>Mean daily $P$ &amp; days (#)</th>
<th>Mean interval (days)</th>
<th>Starting $\theta$ (m$^3$ m$^{-3}$)</th>
<th>$\Delta S$ (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>January</td>
<td>2013</td>
<td>6.86</td>
<td>1.14 (6)</td>
<td>8.33</td>
<td>0.07</td>
<td>3.09</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>34.80</td>
<td>3.87 (9)</td>
<td>7.33</td>
<td>0.18</td>
<td>7.50</td>
</tr>
<tr>
<td>February</td>
<td>2013</td>
<td>28.70</td>
<td>2.61 (11)</td>
<td>2.83</td>
<td>0.12</td>
<td>-4.92</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>17.27</td>
<td>1.73 (10)</td>
<td>3.00</td>
<td>0.23</td>
<td>2.94</td>
</tr>
<tr>
<td>March</td>
<td>2013</td>
<td>19.97</td>
<td>6.66 (3)</td>
<td>9.33</td>
<td>0.09</td>
<td>26.62</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>3.81</td>
<td>1.27 (3)</td>
<td>9.33</td>
<td>0.25</td>
<td>-5.21</td>
</tr>
<tr>
<td>April</td>
<td>2013</td>
<td>61.64</td>
<td>4.74 (13)</td>
<td>4.25</td>
<td>0.26</td>
<td>-2.39</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>36.82</td>
<td>4.60 (8)</td>
<td>5.50</td>
<td>0.21</td>
<td>-4.93</td>
</tr>
<tr>
<td>May</td>
<td>2013</td>
<td>42.10</td>
<td>3.83 (11)</td>
<td>3.33</td>
<td>0.25</td>
<td>-11.43</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>104.17</td>
<td>8.68 (12)</td>
<td>3.17</td>
<td>0.17</td>
<td>12.65</td>
</tr>
<tr>
<td>June</td>
<td>2013</td>
<td>13.97</td>
<td>1.75 (8)</td>
<td>2.75</td>
<td>0.17</td>
<td>-11.18</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>16.51</td>
<td>1.83 (9)</td>
<td>2.63</td>
<td>0.24</td>
<td>-21.02</td>
</tr>
<tr>
<td>July</td>
<td>2013</td>
<td>39.12</td>
<td>3.26 (12)</td>
<td>2.38</td>
<td>0.10</td>
<td>6.03</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>142.36</td>
<td>9.49 (15)</td>
<td>2.00</td>
<td>0.10</td>
<td>28.41</td>
</tr>
<tr>
<td>August</td>
<td>2013</td>
<td>41.85</td>
<td>3.22 (13)</td>
<td>2.25</td>
<td>0.13</td>
<td>1.51</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>55.02</td>
<td>3.24 (17)</td>
<td>1.75</td>
<td>0.27</td>
<td>-18.75</td>
</tr>
<tr>
<td>September</td>
<td>2013</td>
<td>384.06</td>
<td>27.43 (14)</td>
<td>3.20</td>
<td>0.14</td>
<td>18.31</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>56.08</td>
<td>5.10 (11)</td>
<td>3.80</td>
<td>0.14</td>
<td>12.08</td>
</tr>
<tr>
<td>October</td>
<td>2013</td>
<td>18.19</td>
<td>3.64 (5)</td>
<td>6.50</td>
<td>0.24</td>
<td>-1.10</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>25.67</td>
<td>5.13 (5)</td>
<td>6.50</td>
<td>0.22</td>
<td>-9.94</td>
</tr>
<tr>
<td>November</td>
<td>2013</td>
<td>7.37</td>
<td>2.46 (3)</td>
<td>13.50</td>
<td>0.23</td>
<td>-7.44</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>9.97</td>
<td>1.42 (7)</td>
<td>11.50</td>
<td>0.15</td>
<td>5.53</td>
</tr>
<tr>
<td>December</td>
<td>2013</td>
<td>12.70</td>
<td>1.59 (8)</td>
<td>5.75</td>
<td>0.18</td>
<td>-0.59</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>34.80</td>
<td>4.97 (7)</td>
<td>6.00</td>
<td>0.18</td>
<td>-1.21</td>
</tr>
</tbody>
</table>
The pattern of average intervals between individual precipitation events were similar for both 2013 and 2014 calendar years; intervals were greater than nine days in March, decreasing to three to five days in April to May, falling to just over two days from June through August, increasing to over three days in September, increasing in October and November, and finally decreasing again with snowfall in December. The average number of precipitation days varied somewhat between the two years, however the mean daily precipitation was larger during the late season period in 2013, while the converse was true during the early season period in 2014. Changes in daily $S$ were positively correlated with daily precipitation for both 2013 ($r = 0.559$) and 2014 ($r = 0.581$).

Soil water storage in the top 150 mm of soil declined during the early season in both years, however there was more soil water lost during this period in 2013. While accumulated January through March precipitation was very similar between the two years - 2013 was 55.53 mm and 2014 was 55.89 mm – changes in soil water content throughout this period contrasted sharply. Soil water content for 2013 started the year below the threshold established for this study at about 7%, was below this threshold for a total of 7 days during January, and the average soil water content was only 16%. In contrast, soil water content was approximately 17% at the start of 2014, with no days below the threshold, with an average soil water content of 23%.

The annual precipitation in 2013 was higher than in 2014, however this can be attributed to the 2013 late season period, which was 325% higher than the 2014 late season period. Further, extreme rain events during one-week in September 2013 account for 86% of the 2013 late season precipitation (see Chapter 3 for a discussion of the impacts of this event). Conversely, the 2014 early season rainfall was 34% higher than
2013 early season, and ET was 83% higher during this period in 2014. In fact, ET was considerably higher throughout 2014, which is consistent with more available water for that year, ignoring the one-week of extreme precipitation events in 2013.

2.3.2 Daily NEE

The initiation of net carbon uptake for each year was estimated by observing when the trend in daily accumulated NEE (g C m⁻²) became consistently negative, or where daily GPP consistently exceeded daily R_eco (Figure 2.7).

![Figure 2.7. Daily NEE and the 1-week running mean (black lines) during a) 2013 and b) 2014. Positive values are a carbon source while negative values are a sink. The dashed vertical lines indicate periods that are contrasted between the two years.](image)

Carbon exchange began to diverge between the two years at the beginning of March and ecosystem uptake of carbon started at the beginning of April in 2014, as
compared to 2013, where carbon uptake started ~40 days later, during the second week of May. The rate of carbon uptake was strong and consistent throughout the 2014 early season period, indicated by a sharp downward slope in cumulative NEE (Figure 2.8).

![Cartoon of crocodile in Martian landscape](https://via.placeholder.com/150)

**Figure 2.8.** The cumulative NEE for 2013 and 2014 for the mixed grassland. The slope of the line indicates the rate of exchange of carbon between the atmosphere and ecosystem; positive slopes are a net loss from the ecosystem, while negative slopes indicate a net uptake of carbon from the atmosphere. The shaded portion at the bottom labeled “Uptake of carbon” marks the area where cumulative NEE becomes negative and is therefore a net uptake of carbon by the ecosystem, as seen from May to November 2014.

Primary productivity slowed and respiration losses once again dominated the NEE around the end of June for both years. This resulted in a net loss of approximately 82 g C m$^{-2}$ by July 1, 2013, compared to a net uptake of approximately 73 g C m$^{-2}$ during the same period in 2014 - a difference of 155 g C m$^{-2}$ between the two years. In contrast, July and August were carbon loss periods for both years, though the loss of approximately 104 g C m$^{-2}$ in 2013 was nearly twice the amount in 2014, with a loss of about 55 g C m$^{-2}$ during this period.

In the late season period, the rate of carbon loss decreased during both years. In 2013, the extreme precipitation in early September 2013 was followed by a strong carbon
sink period with an uptake of about 14.5 g C m\(^{-2}\) from September 15 to October 31, 2013 (see Chapter 3). This resulted in a carbon loss of approximately 36 g C m\(^{-2}\) during the late season period in 2013, as compared to a loss of approximately 45 g C m\(^{-2}\) during the same period in 2014. However, the difference of about 150 g C m\(^{-2}\) between the two years was well established at the end of the early season period and this gap remained or increased for the last half of the year.

Total annual accumulated \(NEE\) differed greatly between the two years, with 2013 finishing as a considerable carbon source at approximately 200 g C m\(^{-2}\), as compared to 2014, which had a modest loss of only about 12 g C m\(^{-2}\) for the year. Thus, the grassland was a net source of carbon throughout 2013, yet it was a net carbon sink from May to November 2014.

2.3.3 Water Balance and \(NEE\)

Time series graphs of precipitation, daily \(ET\), \(\theta_{0-15cm}\), and daily \(NEE\) (g C m\(^{-2}\)) are presented for 2013 (Figure 2.9) and 2014 (Figure 2.10).
Daily NEE during the early season period in 2013 did not become negative (carbon sink) until the end of April and beginning of May, concurrent with a peak in ET and declining $\theta_{0-15\text{cm}}$. By the end of the early season period in 2013, soil moisture had declined to 12%, and this corresponded to a decrease in daily ET along with a positive daily NEE, a switch from a sink to a source of carbon.

Precipitation events in July and August 2013 caused short-term increases in $\theta$, but only to 19%, along with a low daily ET and a positive daily NEE (carbon source). Although the late season period started slow, extreme precipitation events beginning on September 9, 2013 caused the soil to become completely saturated. At this point, the late season switched to increasing ET and negative NEE that lasted throughout October,
resulting in water and carbon fluxes that were comparable to the early season period.

In contrast, 2014 saw high antecedent soil water conditions (due to the previous autumn flood and a wet winter) and entered the early season period with $\theta_{0-15\text{cm}}$ of about 20%. Precipitation events were spread out over the year with the highest rainfall in July. This resulted in many peaks in soil moisture, often reaching saturated conditions that slowly receded, making water available to plants nearly throughout the growing season.

![Figure 2.10](image)

Figure 2.10. The 2014 time series plots of daily a) precipitation and $ET$, b) $\theta$ (0-15 cm depth), and c) $NEE$ with one-week running mean (black line).

Evapotranspiration during the early spring and summer of 2014 was higher than in 2013 and this coincided with a period of substantial carbon uptake that lasted until early July. Although soil water continued to be sufficient, carbon losses persisted through the remainder of the growing season in 2014. The threshold used in this study to
approximate when soil water stores are becoming depleted is further supported by a notable reduction in both \( ET \) and carbon uptake (\( NEE \) becomes more positive) that was observed to correspond to \( \theta_{0.15cm} \) of ~10-12% in both years.

2.3.4 Accumulated growing degree days

The accumulated thermal energy inputs, or growing degree days, at the study site during the calendar years 2013 and 2014 are presented in Figure 2.11.

![Figure 2.11. Accumulated GDD for the mixed grassland during 2013 and 2014. Differences in thermal energy inputs to the grassland site occurred very early or late in the year, so that for most of the growing season (from May to September) GDD maintained a similar profile.](image)

The accumulated \( GDD \) for growth of the dominant grass species (\textit{Bromus inermus} Leyss) is comparable between the two years. Although there was more thermal energy available during 2014, the difference was established during February and late April and
did not vary considerably again until the end of October. Much of the growth and uptake of CO₂ from the atmosphere occurred during May and June, when accumulated GDD was very similar between the two years (Table 2.3).

Table 2.3. Monthly and annual accumulated GDD and the percent change from 2013 to 2014. Shaded rows highlight months where 2014 GDD was notably higher (about 50% or greater) than 2013 GDD.

<table>
<thead>
<tr>
<th>Month</th>
<th>2013</th>
<th>2014</th>
<th>Difference from 2013</th>
</tr>
</thead>
<tbody>
<tr>
<td>January</td>
<td>92.10</td>
<td>123.73</td>
<td>34.34%</td>
</tr>
<tr>
<td>February</td>
<td>41.78</td>
<td>90.73</td>
<td>117.17%</td>
</tr>
<tr>
<td>March</td>
<td>137.56</td>
<td>165.64</td>
<td>20.41%</td>
</tr>
<tr>
<td>April</td>
<td>169.05</td>
<td>247.97</td>
<td>46.69%</td>
</tr>
<tr>
<td>May</td>
<td>403.73</td>
<td>382.87</td>
<td>-5.17%</td>
</tr>
<tr>
<td>June</td>
<td>587.16</td>
<td>530.75</td>
<td>-9.61%</td>
</tr>
<tr>
<td>July</td>
<td>660.98</td>
<td>658.52</td>
<td>-0.37%</td>
</tr>
<tr>
<td>August</td>
<td>678.14</td>
<td>622.19</td>
<td>-8.25%</td>
</tr>
<tr>
<td>September</td>
<td>526.98</td>
<td>505.78</td>
<td>-4.02%</td>
</tr>
<tr>
<td>October</td>
<td>241.77</td>
<td>375.62</td>
<td>55.36%</td>
</tr>
<tr>
<td>November</td>
<td>163.70</td>
<td>163.82</td>
<td>0.08%</td>
</tr>
<tr>
<td>December</td>
<td>103.81</td>
<td>98.80</td>
<td>-4.82%</td>
</tr>
<tr>
<td>Annual</td>
<td>3806.76</td>
<td>3966.41</td>
<td>4.19%</td>
</tr>
</tbody>
</table>

The largest percentage increase in monthly thermal energy occurred in February, which had similar average daily T_{air} (2013 = -0.86 °C, 2014 = -0.44 °C) and February 2014 had only three more frost-free days during this month. More relevant to the growth of biomass and increase in carbon uptake in 2014 may be the higher available thermal energy in April, which had 10 more frost-free days in 2014 as compared to 2013. Nevertheless, the increase in thermal energy for 2014 was ultimately less than 5% greater than the previous year, and lower in a majority of the months during which the greatest amount of primary productivity occurred.
2.3.5 GPP and $R_{eco}$

There were two separate equations initially derived for the estimate of $R_{eco}$ in 2013. For $Ts$ less than or equal to 17 °C, 72% of the variation in nighttime NEE was explained by the changes in $Ts$ ($n = 212, r^2 = 0.7186, p < 0.0001$). However, the relationship was weaker ($n = 108, r^2 = 0.1345, p < 0.0001$) and reversed, as NEE declined for $Ts$ above 17 °C. However, applying the second equation for temperatures above 17 °C resulted in an underestimation of cumulative $R_{eco}$, so this equation was discarded in favor of applying the equation for $Ts$ less than or equal to 17 °C (Figure 2.12a, left) to estimate $R_{eco}$ for all of 2013.

In 2014, changes in $Ts$ explained 38% of the variation in nighttime NEE for the entire range of soil temperatures ($n = 296, r^2 = 0.3795, p < 0.0001$), so partitioning NEE into $GPP$ and $R_{eco}$ for this year utilized the equation in Figure 2.12b. Given that NEE became inversely related to $Ts$ for values above 17 °C, $θ_{0-15cm}$ values associated with these fluxes were provided, and the relationship between $θ_{0-15cm}$ and $Ts$ can also be found in 5.1.4APPENDIX 1.
Figure 2.12. Plots of the mean nighttime $NEE$ as a function of mean nighttime $Ts$ during a) 2013, where the equation on the left represents flux rates for soil temperatures $\leq 17 \, ^\circ C$ (grey squares), and the equation on the right represents rates $> 17 \, ^\circ C$ (black dots). And in b) 2014, the relationship between flux rate and $Ts$ appeared to be roughly linear. Selected soil gradient flux rates (open circles) are included in both plots. For some $NEE$ values related to $Ts$ above $17 \, ^\circ C$, the associated range of $\theta_{0-15cm}$ values are given (black arrows).
While estimates of annual, early season, and late season accumulated $R_{eco}$ were quite similar between the two years, accumulated $GPP$ differed considerably. Cumulative $GPP$ was 64% higher in the early season period in 2014 as compared to 2013, whereas the late season period in 2013 was 73% higher than the same period in 2014. The magnitude of the early season flux was such that annually 2014 had 22% more carbon uptake, which resulted in a nearly break-even annual $NEE$ as compared to a substantial net loss for 2013. Plots of $NEE$ partitioned into $R_{eco}$ and $GPP$ for both years and the partitioned fluxes for the annual, early season, and late season periods are presented in Figure 2.13. and Table 2.4.

![Figure 2.13](image)

**Figure 2.13.** Cumulative $NEE$, $R_{eco}$ and $GPP$ during 2013 and 2014. Total cumulative values are labeled at the end of the lines.
Table 2.4. Cumulative annual NEE, $R_{eco}$ and GPP for 2013 and 2014, and early season (April 1 – June 30), and late season (August 15 – November 13) periods.

<table>
<thead>
<tr>
<th></th>
<th>2013</th>
<th>Early Season</th>
<th>Late Season</th>
<th>2014</th>
<th>Early Season</th>
<th>Late Season</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Annual</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NEE</td>
<td>200.55</td>
<td>-6.46</td>
<td>36.33</td>
<td>12.22</td>
<td>-151.27</td>
<td>45.34</td>
</tr>
<tr>
<td>$R_{eco}$</td>
<td>502.49</td>
<td>152.74</td>
<td>156.97</td>
<td>454.53</td>
<td>156.76</td>
<td>138.65</td>
</tr>
<tr>
<td>GPP</td>
<td>-344.83</td>
<td>-163.97</td>
<td>-126.78</td>
<td>-420.30</td>
<td>-269.06</td>
<td>-73.37</td>
</tr>
</tbody>
</table>

2.3.6 Biomass, LAI, and precipitation

Individual samples of dry weight biomass (g) and sample LAI from biweekly destructive sampling were well correlated for both 2013 (n = 63, r = 0.80) and 2014 (n = 90, r = 0.91), and sample LAI values were positively correlated with associated MODIS derived LAI values on or near the sampling dates (n = 19, r = 0.88). A time series plot (Figure 2.14) shows all of the LAI (destructive sampling and MODIS) and biomass measurements for both calendar years.
There were two growth peaks that occurred during both years – one in the early period, around the beginning of June, and then again sometime just before or during the late season, varying from early August, in 2014 to late September and early October in 2013. Changes in biomass followed the changes in LAI fairly well, though peaks in biomass lagged slightly behind peaks in LAI.

Several relationships emerged as a result of the regression analysis between MODIS-derived LAI and NREL precipitation from 2005 – 2014. A time lag of one month resulted in the best fit for regressions of monthly precipitation predicting leaf area (Figure 2.15; n =118, $r^2 = 0.1794$, $p < 0.001$), and nearly 40% of the variation in maximum monthly LAI restricted to April, May, and June (early season period) could be explained by the prior month’s rainfall (n = 30, $r^2 = 0.3954$, $p < 0.01$).
Figure 2.15. Goodness of fit for time-lagged regressions of the maximum monthly \textit{LAI} as a function of monthly accumulated precipitation for 2005 - 2015, lagged from 0 to 12 months. The best predictor for \textit{LAI} is for the number of months lagged = 1, where 18\% of the current \textit{LAI} is explained by the previous month’s precipitation.

Even more predictive was the accumulated prior two-month’s precipitation solely on June’s peak \textit{LAI} for the years 2005 to 2014 (n = 10, $r^2 = 0.6857$, p < 0.001), which explained 69\% of the variation in \textit{LAI} (Figure 2.16).
Figure 2.16. Maximum LAI (June or August) as a function of the previous two months’ accumulated precipitation (April/May or June/July) at the semiarid mixed grassland in Rocky Flats NWR from 2005 to 2014.

Accumulated precipitation from the beginning of the year improved the relationship slightly. Total rainfall for the first five months, from January to May, explained 72% of the peak June LAI for the years 2005 to 2014 (n = 10, $r^2 = 0.7177$, $p < 0.001$).

A similar relationship exists for the second peak in leaf area in the late season period with maximum monthly LAI for August, September, and October as a function of monthly rainfall for July, August, and September (n = 30, $r^2 = 0.26$, $p < 0.01$). A multiple regression with the previous and current month’s rainfall improves the ability to predict peak growth during the late season, explaining 34% of the variation in maximum monthly LAI (n = 30, $r^2 = 0.34$, $p < 0.01$), and accumulated June/July rainfall explained about 54%
of the variation in peak August LAI for the ten-year period from 2005 to 2014 (Figure 2.16; \( n = 10, r^2 = 0.5387, p < 0.001 \)).

2.4 Discussion

The timing of intra-annual precipitation made a substantial difference in how carbon cycling responded in the semiarid, smooth brome-dominated mixed grassland at Rocky Flats NWR. Annual rainfall for 2013 was 30% higher than the annual total for 2014, however carbon losses were greater than carbon uptake in 2013, while the opposite was true during 2014, thus the link between precipitation and carbon cycling is more complex. Based on the most recent 10-year period of precipitation and LAI, the grassland at Rocky Flats NWR is strongly dependent on short-term, accumulated precipitation from April and May to achieve robust early season growth that is necessary to result in a carbon sink for the year. In addition, analysis of precipitation, LAI, and carbon fluxes in 2013 and 2014 indicate that the significant rainfall in the autumn of 2013 played a role in the early spring growth and carbon uptake in 2014. Conversely, it appears that the grassland was a sizeable carbon source in 2013 because the timing of precipitation events was not synchronized with the phenology (life cycle events) of the dominant grass species.

Daily carbon uptake was large during the early season period during both years of this study however, very small positive or negative daily NEE values indicate that respiratory losses were of a similar magnitude. A much greater carbon uptake as compared to respiratory loss was achieved once daily NEE became consistently and strongly negative. A time series plot of the ratio of cumulative GPP to cumulative \( R_{eco} \)
summarizes the relationship between these opposing fluxes throughout the year (Figure 2.17).

![Graph showing cumulative GPP to cumulative Reco for each year]

Figure 2.17. Changes in the ratio of cumulative GPP to cumulative Reco for each year; the proportion of carbon uptake to loss at any point in the year. Any portion of the line above one, in the shaded area of the graph, indicates when the grassland is accumulating more carbon than it is losing at that point in the year.

The ratio of total carbon added to that lost at any point in the year indicates when the ecosystem switches from a source to a sink. As this ratio becomes greater than one, the grassland is accumulating more carbon than it is losing. During the early season period in 2014, the grassland’s carbon uptake outpaced the losses by a factor of 1.5 for all of June.

Concurrent with the strong carbon uptake periods are increases in aboveground vegetation, represented by LAI and biomass measurements. Analysis of the temporal changes in LAI and the relationship between precipitation and changes in LAI from the long term regional records were sufficient to establish three characteristics of changes in vegetation at this grassland: 1) there was frequently a peak in LAI in June, 2) there was a second, usually smaller peak in LAI for August or later, and 3) there is a strong positive
relationship between short-term, accumulated monthly precipitation and LAI peak values. The timing of the peaks in LAI is explained by the dominant grass species phenology and cool-season vegetation growth behavior, while the magnitude of aboveground growth is related to soil water availability, which is dependent on the timing of precipitation.

2.4.1 Soil water availability

The timing of intra-annual precipitation in semiarid ecosystems impacts soil water fluctuations and, ultimately, the timing of biologically available soil water. An estimated ¾ of the biomass in semiarid grasslands exists belowground, and the roots of smooth brome are concentrated in the top 10 cm of the soil (Craine et al., 2007). The soils at this grassland site appeared to retain water in the top 15 cm after recharge for only about a month or two, most notable in the changes in soil moisture after the significant rains in September 2013. Perhaps this is why the accumulated April/May precipitation explained nearly 70\% of the variation in LAI in June at this study site, which is historically the month of highest LAI values and peak biomass for both years of the study. Great Plains rangeland studies have previously shown that forage yields are greatest during this early season period and were also positively affected by increased April/May precipitation (White, 1985).

Soil water content was drawn down while ET increased during both early season periods, which is consistent with plant water uptake and loss through transpiration as the vegetation was growing. However, more soil water was lost in 2013 than in 2014 even though both carbon uptake and ET were higher throughout this period in 2014. Although mean $\theta_{0.15\text{cm}}$ in 2013 was higher (24.8\% ± 0.08) than in 2014 (20.6\% ± 0.06), total
precipitation during the early season period was higher in 2014. Furthermore, $R_{eco}$ was similar between the two years during this period, yet $GPP$ was 64% higher in 2014. It appears that additions to soil water during the early season period in 2014 were utilized for carbon uptake that was well established at the start of the period.

As long as there is sufficient soil water available early in the growing season synchronized with air temperatures that are consistently above freezing, the conditions are optimal for soil microbes to initiate mineralizing organic matter and releasing nitrogen that will be used by plants (Dijkstra et al., 2012). The plants in the grassland have increasing levels of leaf nitrogen content as rates of plant growth increase throughout the spring. Rubisco (Ribulose-1,5-bisphosphate carboxylase/oxygenase), the nitrogen-rich enzyme responsible for the first step in photosynthesis, has a temperature and concentration affinity for $O_2$. $CO_2$ solubility decreases with increasing temperature and photorespiration increases (lowering photosynthetic efficiency), decreasing nitrogen use efficiency and, since water is essential for this process, water use efficiency with increasing temperatures (Ehleringer et al., 1997; Sage et al., 1987). High nitrogen content is associated with both high growth and high maintenance rates, and therefore respiration rates are also elevated (Dijkstra et al., 2012). Thus, it appears that abundant resources, especially soil water and soluble nitrogen, are crucial from March to April to achieve the kind of productivity in June that leads to a much greater uptake to loss ratio needed for a net gain of carbon.

Since January to March precipitation was approximately equal between the two calendar years at the study site and soil water content was very high by the beginning of April 2013, soil water status at the beginning of the year must also be important. Soil
water content started 2013 below the established threshold for this study (8%) and few rain events were able to increased soil moisture consistently above these low water-content levels until February and March, when both precipitation and snow melt recharged soil water stores. Conversely, the water content started well above water-stressed levels in 2014, precipitation was greater in January and February, and soil water content never fell below the threshold although rain events were few in March. In arid and semiarid ecosystems, microbial processes that generate the supply of nitrogen and plant nutrient uptake operate at different water content levels, so they can become uncoupled when there are frequent wetting/drying cycles in the soil (Austin et al., 2004; Dijkstra et al., 2012; Giese et al., 2010). It is therefore possible that the fluctuating nature of soil water combined with water stress during the beginning of 2013 caused nitrogen losses and negatively impacted nutrient supplies to the vegetation by April, when they were crucial for early season growth.

The extent to which water availability impacts carbon cycling during the late season period is less substantial. A second peak in growth was observed during the late season period for both years, however the peak growth in September/October 2013 was fueled by a significant supply of water from extreme precipitation events. Historically, the accumulated two-month summer or early autumn rainfall explained between about 34 and 54% of the variation in peak late season LAI at this grassland, thus while it is still important, dependence on precipitation is reduced compared to the early season period. Although carbon uptake was stronger during this period in 2013, carbon losses were higher and ET was lower than in 2014 and the magnitude of the net gains were considerably lower as compared to the early season period in 2014. Smooth brome does
not generally achieve the same yields at this time of the year, and the availability of soil nitrogen may be further compromised by saturated conditions (Dijkstra et al., 2012; Otfinowski et al., 2007). The extreme precipitation events could have inundated soils and washed away nutrients or created an anaerobic environment where denitrification occurs (Austin et al., 2004). Further, the substantial aboveground growth did not peak until October, which is quite late in the year, and was likely cut short by the onset of winter and freezing temperatures.

Notwithstanding the significantly larger annual rainfall for 2013, LAI in 2014 was consistently higher during every month, with the exception of October. Substantial early season period growth and carbon exchange was clearly more effective at sequestering carbon than comparable growth in the late season period.

2.4.2 Soil water content, Ts and $R_{eco}$

Soil moisture appeared to have a strong influence on the relationship between $R_{eco}$ and $Ts$. Although a decline in respiration is expected at high soil temperatures, a sharp decline in respiration during 2013 occurred at 17 °C, a temperature too low to cause a decrease in respiratory processes via thermal energy (i.e. denatured proteins). Although the relationship is less clear during 2014, there was a decline in $R_{eco}$ for $Ts$ greater than ~20 °C. Soil temperature was inversely related to $\theta_{0.15cm}$ at this grassland during both years (Figure A - 1). Therefore respiratory decreases strongly coincided with decreasing soil moisture for $Ts$ values greater than 17 °C during 2013, and weakly declined with $Ts$ values greater than ~20 °C during 2014.
2.4.3 The phenology and cool-season behavior of smooth brome

As a cool-season perennial grass, smooth brome’s growth in the spring usually consists of a combination of new vegetation and regrowth of shoots that started before winter (Otfinowski et al., 2007). Therefore the early season period vegetation growth has a dependence on the prior year’s late season productivity. The late season period in 2012 was characterized by low rainfall and low leaf area values, therefore it is unlikely that grasses were productive late in the year and consequently added little to the early season period of 2013. In contrast, the extreme precipitation events in the late season period of 2013 resulted in a strong peak in LAI through October, and thus this late productivity contributed to the early season period of 2014 via regrowth of shoots from late 2013. This is evident because soil moisture remained high throughout the winter and spring 2014 growth and carbon uptake began in late March/early April, well before the effects of April/May precipitation that year.

Cool season plants thrive in air temperatures between approximately 15 and 25 °C, which occurs at the start and end of the growing season and accounts for the double peak in LAI for both years at this grassland site. Vegetation growth in the early season period is therefore also dependent on optimal temperatures. Growing degree days in April 2014 were 47% greater than 2013, one of the biggest differences in thermal energy inputs between the two calendar years. Thermal inputs remained comparable during much of the year until October, when 2014 received 55% more than in 2013. As noted previously, the addition of significant amounts of rain in September 2013 overwhelmed any advantage that 2014 gained from more thermal energy inputs as the late season in 2013 had a higher peak in LAI and carbon inputs during the late season period.
2.4.4 The impact of forecasted changes in precipitation

Grasslands are the most variable ecosystem in aboveground productivity (Knapp and Smith, 2001), with the potential to accumulate carbon for the long term. In the short term, strong productivity in semiarid cool-season grasslands, like the smooth brome-dominated system studied here, will always be balanced by losses due to autotrophic respiration and by stimulating heterotrophic respiration through root exudates or surface litter additions. Soil texture influences the water and nutrient availability to plants and is modulated by precipitation inputs.

The majority of primary productivity in cool-season (C₃) grasslands occurs in the spring, when temperatures are cooler (Ehleringer et al. 1997). Arid and semiarid grasslands in the U. S. Great Plains dominated by the cool-season grass smooth brome are dependent on the timing of precipitation to make water available during these optimal time periods, yet models have forecasted shifts in the seasonal timing and event size of precipitation (Christensen et al., 2013). This could have consequences for carbon cycling in a broad regions of North America where this nonnative grass is still planted or where it has invaded native prairie ecosystems, at times changing the vegetation from a warm-season to a cool-season dominated system (Vinton and Goergen, 2006). Forecasts of future climate could be improved by considering the seasonal feedbacks that these ecosystems will have on both the water and carbon cycles.
CHAPTER 3. THE IMPACT OF AN EXTREME PRECIPITATION EVENT ON RESPIRATION IN A SEMIARID GRASSLAND AT ROCKY FLATS NATIONAL WILDLIFE REFUGE

3.1 Introduction

The historic Colorado floods of 2013 were caused by significantly heavy precipitation events that occurred during the week of September 9-15, 2013. In many areas along the Colorado Front Range affected by the floods, the duration and intensity of rainfall resulted in new daily, multi-day, and monthly rainfall records, and approximately half of that year’s total precipitation fell in this one week (Gochis et al., 2015; Hoerling et al., 2014). Due to the rarity of events such as this, little is known about how ecosystems are impacted by rainfall that is comparable in duration and size to the events that occurred in Colorado that year. However, since large rainfall events may become more frequent in the future (Christensen et al., 2013) and past observations are inadequate for estimating how carbon cycling in these systems will respond to unusually large precipitation events, ecosystem observations made during the Colorado Flood event are extremely valuable.

Precipitation has already increased across the contiguous U.S. by 10% since 1910, and it is noteworthy that it is due in large part to an rise in extreme and heavy rain events (Karl and Knight, 1998). Further, Global Circulation Models (GCMs) have projected an increase in the variability of intra-annual precipitation in North America (Christensen et al., 2013; Trenberth et al., 2003). Aboveground Net Primary Productivity (ANPP) in grassland biomes is highly responsive to rainfall (Knapp and Smith, 2001), and there is evidence that arid and semiarid ecosystems respond positively to extreme precipitation
regimes (Knapp et al., 2008; Zeppel et al., 2013). A study conducted by Knapp et al. (2008) found that xeric ecosystems responded well to less frequent and larger intensity rainfall because soil water stress was relieved. However, that study did not include extremely heavy rain events. Semiarid grasslands frequently experience droughts and long periods between rain events that lead to broad fluctuations in soil water content ($\theta$) over short periods of time (Austin et al., 2004; Newman et al., 2006). Soil water dynamics and the related impacts on carbon cycling in these systems are fairly well characterized for a range of vegetation types, soil textures, and precipitation regimes (Austin et al., 2004; Cherwin and Knapp, 2012; Gao et al., 2010; Ryel et al., 2010). While rainfall manipulation studies have tested conditions under large precipitation pulses, none of these has been close to the duration and intensity of the accumulated precipitation during September 2013, which was an order of magnitude larger than the 30-year average for that month (Figure 3.1).

![Figure 3.1. Total monthly precipitation for every September since the year 2000. The dashed line indicates the mean monthly precipitation for the past 30 years. Precipitation for the week of September 9-15, 2013 made up nearly 95% of the total for September 2013.](image-url)
During the summer and early autumn leading up to the extreme precipitation, ecosystem respiration ($R_{eco}$) at the mixed grassland in Rocky Flats National Wildlife Refuge (NWR) was greater than gross primary productivity ($GPP$), yet the grassland responded with late season growth and a period of carbon uptake in the weeks following the event (see Chapter 2). In addition, results from that study indicate that $R_{eco}$ becomes insensitive to soil temperature ($T_s$) above ~17 °C and is controlled by $\theta$ instead.

Early studies used empirical relationships to estimate soil respiration ($R_s$) from $T_s$ (Lloyd and Taylor, 1994; Raich and Schlesinger, 1992; Singh and Gupta, 1977), which is often modeled by a simple exponential function known as the $Q_{10}$, where the respiration rate increases by a factor for every 10-degree increase in temperature. However, pronounced shallow wet and dry cycles that are typical in semiarid grassland soils affect the temperature sensitivity of $R_s$ (Austin et al., 2004; Burke et al., 1998; Wynn et al., 2006). Further, continuous measurements show that a diel hysteresis between $R_s$ and $T_s$ (or soil [CO$_2$] and $T_s$) is common, where the temperature sensitivity of $R_s$ during warming is different from cooling, resulting in distinct $R_s$ values for the same temperature (Oikawa et al., 2014; Riveros-Iregui et al., 2007). In addition, low soil volumetric water content ($\theta$) limits substrate availability (because water is needed for nutrient accessibility and enzymatic activity) and high $\theta$ limits oxygen diffusion (Davidson et al., 2006), thus narrowing the range of $\theta$ values over which $T_s$ controls $R_s$ (Curiel Yuste et al., 2007). To adjust for the effect of $\theta$, a series of four logarithmic soil respiration functions were evaluated by Vicca et al. (2014), which estimates $R_s$ as a function of $T_s$ and a progressively linear to non-linear relationship with $\theta$. Moving towards process-based concepts, Davidson et al. (2012) developed the Dual Arrhenius
Michealis-Menten (DAMM) model, which goes further and merges the reactions that produce soil CO$_2$ with rate-limiting conditions – $\theta$, $T_s$, soluble soil carbon and oxygen substrates, and the temperature sensitivity of enzymes involved in decomposition.

Pulses of water influence $R_s$ in other ways as well. Drying soils can expose previously protected soil organic matter due to changes in pore size and connectivity (Borken and Matzner, 2009; Van Gestel et al., 1993). Thus, soils that have been dry and recently rewetted have resulted in pulses of CO$_2$ at the land surface (efflux) immediately following a rain event (Birch, 1958; Keift et al., 1987; Orchard and Cook, 1983), and the duration and source of the CO$_2$ has been shown to vary with the size of the precipitation event and depth of water infiltration into the soil (Huxman et al., 2004). Finally, $\theta$ indirectly affects the efflux of CO$_2$ at the surface as water-filled pore spaces inhibit soil gas diffusion, decoupling surface pulses of CO$_2$ from the source of production belowground on short timescales (Moldrup et al., 2000; Pacific et al., 2008; Riveros-Iregui et al., 2007). The amount of carbon lost from the soil during a CO$_2$ pulse after an extremely large rain event has been shown to be quite significant in grasslands in California, where one such release of CO$_2$ equaled nearly 10% of the annual gross primary production (Xu et al., 2004).

In the short term, the production of CO$_2$ in the soil and the efflux of CO$_2$ from the surface to the atmosphere are distinct processes linked by the manner in which CO$_2$ is transported through the soil (Fang and Moncrieff, 1999; Vargas et al., 2011). The soil flux gradient method is based on how this variation in soil CO$_2$ concentration at different soil depths - a vertical gradient - drives the surface efflux of CO$_2$. Therefore, an advantage of measuring soil CO$_2$ flux using the gradient method is that the measurements
are made frequently (i.e., hourly) and with little disturbance from surface processes, like solar radiation and wind (Pingintha et al., 2010; Roland et al., 2015). In a flux gradient method study located in an alpine meadow, Roland et al. (2015) demonstrated that diffusive transport was enhanced by higher soil water content because it buffered shallow soil from disturbance at the land surface. Therefore, increasing soil water content may act to decouple belowground CO₂ production from gas transport by cutting of soil air filled pores from the influence of wind and pressure pumping.

The objective of this study was to assess how significant inputs of water affected soil water and carbon cycling in a semiarid mixed grassland during the extreme precipitation in September 2013. It was unknown how large the carbon losses would be from heterotrophic decomposition in saturated conditions and if carbon gains from late season vegetation growth could balance these losses. Observations were gathered from regional meteorological stations along with hourly measurements of above and belowground water and carbon fluxes from soil plots and eddy covariance instruments at the grassland site to assess the impacts and to answer the following questions: 1) How does the impact of this extreme precipitation event on soil respiration differ from the respiration response of a typical rainfall event at this site? and; 2) How well do models estimate CO₂ efflux during rain events of this intensity and duration? Given that extensive saturation occurred during the week of extreme precipitation and resulted in air-filled pore space and gaseous diffusion in the soil being restricted, the following hypothesis was proposed:
H3: During an extreme precipitation event, soil respiration at the mixed grassland in Rocky Flats NWR will be insensitive to soil temperature and therefore poorly modeled by empirical relationships primarily based on this variable.

To test this hypothesis, model estimates of carbon fluxes were generated from empirical and process-based models and compared to observations made at the mixed grassland site in Rocky Flats NWR for September 9-15, 2013.

3.2 Methods

3.2.1 Study Site

The study was conducted in a semiarid mixed grassland located about 18 km northwest of Denver, CO, USA in the southeastern section of Rocky Flats National Wildlife Refuge (NWR) (39°53'N latitude, 105°12'W longitude). The climate is classified as semiarid with a mean annual temperature (MAT) of 8.2 °C and a mean annual precipitation (MAP) of 426 mm (30-year averages 1981-2010, PRISM Climate Group, Oregon State University, [http://prism.oregonstate.edu](http://prism.oregonstate.edu)), and the elevation of the site is approximately 1730-m asl. The study site was located within an area that received some of the heaviest rainfall during the extreme event (Figure 3.2). See Chapter 2 for complete site details (section 2.2.1).
Figure 3.2. The research location (small black box) within the map of Annual Exceedance Probabilities for worst-case 7-day rainfall during the extreme precipitation event, and within a subset (call out) of a local area topographic map. (Maps were modified from NOAA 2015 and the USGS Louisville Quadrangle topographic map, U.S. Geological Survey [cartographer], 2015).
3.2.2 Precipitation

Regional precipitation measurements were obtained from the Measurement and Instrumentation Data Center (MIDC) of the National Renewable Energy Lab (NREL), collected continuously from a heated tipping bucket rain gauge (320b, Belfort Instrument, Baltimore, MD) mounted on a tower at the 2-m height (39°54'36.0"N 105°14'06.0"W) (Jager and Andreas, 1996), which is located approximately 5 km northwest of the study site. The NREL observations were used for rainfall analysis in preference to the tipping bucket rain gauge located at the grassland study site because the intensity and duration of rainfall during September 2013 exceeded the capacity of the study site’s instrument.

3.2.3 Soil Plots

Two soil plots were established at the study site, designated as north and south and located ~12 meters apart, and were equipped to measure soil moisture, soil temperature, and soil CO₂ concentrations at three shallow soil depths - 5, 10 and 15 cm. All θ and Ts sensors took a measurement every 60-seconds and the average was recorded every 30-minutes. Mean daily θ and Ts values are calculated from the 30-minute values in each 24-hour period. Soil [CO₂] values are recorded once an hour, and mean daily or nighttime (see 3.2.4 Eddy covariance below) values are determined by averaging the hourly measurements. Further details regarding the soil plots and instruments may be found in Chapter 2 (sections 2.2.2, 2.2.3 and 2.2.4).
3.2.4 Eddy Covariance

The study site had ongoing eddy covariance-based measurements of surface energy balance, including net ecosystem exchange (NEE), derived from the turbulent exchange of CO₂ between the atmosphere and land surface. Wind components were measured by a three-dimensional sonic anemometer (CSAT3, Campbell Scientific, Inc., Logan UT, USA), and CO₂ and H₂O mole densities were measured by an open-path infrared gas analyzer (IRGA, Li-7500, LI-COR, Lincoln, NE, USA), both instruments were mounted at a height of 1.98 m above the ground. Each 30-minute measurement period was identified as nighttime for solar radiation values ≤ 20 Wm⁻². A complete description of the eddy covariance system and data processing can be found in Chapter 2 (section 2.2.5). Given that the IRGA was an open path gas analyzer, the eddy covariance measurements included substantial gap-filling during precipitation events in the week of September 9-15, 2013 (71% of the 30-minute measurements were gap-filled).

3.2.5 Soil texture, porosity and organic content

Soil samples were collected at the study site during the summer of 2014. Soils were collected at eight random locations with three samples at each location (n = 24, within 625-m² area centered at the eddy covariance tower). Samples were carefully excavated from beneath the litter layer, at 0 to 10 cm soil depths. Soil samples were oven dried at 100 °C for a minimum of 24-hours. Bulk density (BD) was calculated from the mass of the dry soil and the volume of the soil sample (BD = 1.4 g/cm³). Six soil samples were set aside, undisturbed, and dried for an additional 24-hours. Porosity (φ) for this soil
was obtained by averaging the volume of water needed to completely saturate each of the six soil samples.

All other soil samples were sieved to 2 mm particle size and aliquots of each were set aside. The remaining soil was combined using a soil splitter, portions of which were agitated in a 3% solution of sodium hexametaphosphate (HMP) and passed through a series of sieves to establish an estimate for texture (Kettler et al., 2001), which was determined to be a loam (sand = 49%, silt = 34%, clay = 17%).

The aliquots that were set aside were powdered in a mortar and pestle and sieved to 500 µm. Powdered soil samples were weighed into ceramic crucibles (approximately 5 g each) and placed in a Fisher Scientific Isotemp muffle furnace at 430 °C for a minimum of five hours to determine total soil organic content or organic matter (SOM) by loss on ignition (LOI) following standard procedures (USDA, 1996). The soil organic carbon (SOC) content was calculated by dividing the total soil organic content by a factor of 1.72 (SOM/1.72) (USDA, 1996). Details of each soil test and a table of soil properties can be found in APPENDIX 3.

### 3.2.6 Empirical Models

For this study, respiration was assumed to be predominantly soil respiration for the period of the extreme precipitation event because it was raining and cloudy and therefore photosynthesis was minimal. Respiration from the eddy covariance method and Rs from the flux gradient calculations were compared to estimates of Rs from empirical models based on Ts (Lloyd and Taylor, 1994) and soil moisture (Vicca et al., 2014). The
relationship developed by Lloyd and Taylor (1994) is based on an Arrhenius-type equation:

\[ R_s = R_{10} e^{308.56 \left( \frac{1}{56.02 - \frac{1}{Ts - 227.13}} \right)} \] (3.1)

where \( R_{10} \) is the respiration rate at 10 °C and \( Ts \) is the soil temperature. While temperature changes can predict \( R_s \) over a wide range, \( \theta \) can confound this relationship and \( R_s \) can become insensitive to \( Ts \). A series of four logarithmic \( R_s \) functions, progressively incorporating a linear and then non-linear relationship with \( \theta \), were evaluated in a review by Vicca et al. (2014), and tested in this study:

\[
\begin{align*}
log(R_s) &= a + bTs + c\theta \\
log(R_s) &= a + bTs + \log(c + d\theta) \\
log(R_s) &= a + bTs + \log(c + d\theta + e\theta^2) \\
log(R_s) &= a + bTs + c\theta + d\theta^2
\end{align*}
\] (3.2) (3.3) (3.4) (3.5)

where \( R_s \) is soil respiration, \( Ts \) is soil temperature, \( \theta \) is soil water content, and \( a, b, c, d \) and \( e \) are coefficients.

Equations used to determine model parameters were fitted using nonlinear least squares (nls function) in the R statistical software package (version 3.2.2; R Development Core Team, 2015), utilizing abiotic variables from the extreme precipitation week to calibrate the coefficients.
3.2.7 DAMM model

The Dual Arrhenius Michaelis-Menten (DAMM) model utilizes several equations that contribute to a central Michaelis-Menten kinetics equation for two soil substrates, soluble carbon (Sx) and oxygen (O2):

\[ R_{Sx} = V_{max} \times \frac{[Sx]}{kM_{Sx} + [Sx]} \times \frac{[O_2]}{kM_{O2} + [O_2]} \] (3.6)

where \( R_{Sx} \) is the reaction velocity, \( V_{max} \) is the maximum rate of the enzymatic reaction for unlimited substrate concentrations, \([Sx] \) and \([O_2] \) are the concentrations of soluble soil carbon and oxygen, respectively, and \( kM_{Sx} \) and \( kM_{O2} \) are the respective Michaelis constants, the concentration for each of the substrates at which the reaction rate is half of \( V_{max} \). An Arrhenius function is applied to the calculation of \( V_{max} \):

\[ V_{max} = \alpha_X \times e^{-Ea/RT} \] (3.7)

where \( \alpha_X \) is the pre-exponential factor, \( Ea \) is the activation energy, \( R \) is the universal gas constant, and \( T \) is the temperature in Kelvin. The computation of the Michaelis constant for \( Sx \) is expressed by a linear temperature dependence:

\[ kM_{Sx} = c_{kM_{Sx}} + m_{kM_{Sx}} \times Tc \] (3.8)
where $c_{kM_{Sx}}$ is the intercept, $m_{kM_{Sx}}$ is the slope, and $Tc$ is temperature in Celsius. The concentration of soluble carbon is a function of both available organic material and $\theta$, with $\theta$ directly affecting the diffusion of both enzymes and substrates to reaction sites within the soil. Following the example from the field test in Davidson et al. (2012) and based on the available data and conditions that existed at the grassland site during the period of the study, there were several simplifying assumptions used to determine $[Sx]$. A fixed percentage ($p$) of the mean soil organic carbon (SOC) content $[Sx_{total}]$ is assumed to make up the total available soluble carbon pool at peak soil saturation $([Sx_{soluble}] = p \times [Sx_{total}])$. The percentage was estimated from typical dissolved organic content relationships to SOC (Frank et al., 1996; Neff and Asner, 2001). The $[Sx]$ at each time step is a function of $[Sx_{soluble}]$, $\theta$, and diffusivity:

$$[Sx] = [Sx_{soluble}] \times D_{liq} \times \theta^3$$  \hspace{1cm} (3.9)

where $D_{liq}$ is a dimensionless diffusion coefficient, determined by setting the boundary condition $[Sx] = [Sx_{soluble}]$ for fully saturated soil. The concentration of $O_2$ is a simple relationship from Millington (1959):

$$[O_2] = D_{gas} \times 0.209 \times \varepsilon_a^{4/3}$$  \hspace{1cm} (3.10)

where $D_{gas}$ is the diffusion coefficient for $O_2$ in air, 0.209 is the proportion of air that is made up of $O_2$, and $\varepsilon_a$ is the soil air-filled pore space, which is computed from porosity and $\theta$ at each time step.
Models were calibrated using the 2013 growing season (April 1 – September 30) nighttime net ecosystem exchange (respiration) values from the eddy covariance system, and $T_s$ (10 cm depth) and $\theta$ (integrated over the 0-15 cm depth) from the soil plots. Soil water content was used as a proxy for $[S_x]$ (g C m$^{-2}$) by applying equation (3.9. Mean nighttime $R_s$ (a proxy for $V_{max}$) were binned by $\theta$ and $T_s$ and used in equations (3.7 and (3.8 to determine $\alpha_x$, $E_a$, $c_{kM_{Sx}}$, and $m_{kM_{Sx}}$. Although the eddy covariance nighttime $R_s$ represented a combination of autotrophic and heterotrophic respiration, as well as CO$_2$ produced from labile and recalcitrant soil carbon pools, the DAMM model estimates were used solely for comparisons during the extreme precipitation period when solar radiation was reduced, labile carbon pools were most active, and conditions similar to the nighttime calibrations likely dominated CO$_2$ production. In addition, due to increasingly restricted gas transport, efflux measurements were more offset from production in time for $\theta$ greater than 30% and therefore these fluxes were not included in the calibrations.

As stated in Davidson et al. (2012), the explicit process model requires many more parameters in addition to the $T_s$ and $\theta$ used by the empirical models, and some of these parameters can be prescribed as long as its provides realistic boundary conditions. Since the model was used for a short time frame, many of the soil properties (porosity, soil organic carbon, etc.) can reasonably be assumed to not change over the model period and therefore fixed values were used. Other parameters were fitted using nonlinear least squares (nls function) in the R statistical software package (version 3.2.2; R Development Core Team, 2015).

Table 3.1 lists all of the fixed and calibrated parameters used in the DAMM model for estimating $R_s$ during the week of September 9 – 15, 2013.
Table 3.1. A list of all of the fixed and estimated parameters used in the DAMM model, and the coefficients (a – d) used in the Vicca empirical equation. Estimates and associated p-values for values that were calibrated by the Arrhenius and linear temperature dependent equations that contribute to the Michaelis-Menten equations used in the DAMM model. All parameters were fitted by non-linear least squares (nls) and linear regressions.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bulk density ($BD$, g cm$^{-3}$)</td>
<td>1.40</td>
<td></td>
</tr>
<tr>
<td>Total porosity (φ)</td>
<td>0.51</td>
<td></td>
</tr>
<tr>
<td>$S_{x_{total}}$ (g C cm$^{-3}$)</td>
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<td></td>
</tr>
<tr>
<td>p (%)</td>
<td>0.50</td>
<td></td>
</tr>
<tr>
<td>$S_{x_{soluble}}$ (g C cm$^{-3}$)</td>
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<td></td>
</tr>
<tr>
<td>$D_{liq}$</td>
<td>10.829</td>
<td></td>
</tr>
<tr>
<td>$D_{gas}$</td>
<td>1.67</td>
<td></td>
</tr>
<tr>
<td>$k_{M_{O_2}}$ (g C cm$^{-3}$)</td>
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<td></td>
</tr>
<tr>
<td>R (kJ k$^{-1}$ mol$^{-1}$)</td>
<td>0.008314</td>
<td></td>
</tr>
<tr>
<td>$a_{sx}$ (mg C cm$^{-3}$ h$^{-1}$)</td>
<td>$1.33 \times 10^5$</td>
<td>$P &gt; 0.1$</td>
</tr>
<tr>
<td>$E_{a_{sx}}$ (kJ mol$^{-1}$)</td>
<td>42.6</td>
<td>$P &lt; 0.001$</td>
</tr>
<tr>
<td>$c_{kM_{sx}}$ (g C cm$^{-3}$)</td>
<td>$4.0 \times 10^{-5}$</td>
<td>$P &gt; 0.1$</td>
</tr>
<tr>
<td>$m_{kM_{sx}}$ (g C cm$^{-3}$)</td>
<td>$-1.0 \times 10^{-7}$</td>
<td>$P &gt; 0.1$</td>
</tr>
</tbody>
</table>

*Vicca equation*

- Coefficient a: -0.6193
- Coefficient b: 0.0157
- Coefficient c: 3.4875
- Coefficient d: -5.6803

Bulk density ($BD$), φ, and SOC for the soils at the study site were determined by the soil sampling and subsequent lab analysis ($BD$, n = 12, s.d. = 0.034; φ, n = 6, s.d. = 2.46%). Soil organic carbon (SOC) for the 0-10 cm depth was determined to be 5.99% (n
= 18, s.d. = 0.74%, CV = 12.28%), and $p$ was determined from a conservative estimate of 0.5% of SOC upon full saturation, thereby resulting in a maximum $S_{\text{xsoluble}}$ concentration of $0.00042 \text{ g C cm}^{-3}$. The value for $D_{\text{gas}}$ assumes that $[O_2]$ in dry soil is equivalent to that in free air.

3.2.8 Comparison to week of July 29-August 4, 2014.

The week of July 29 – August 4, 2014 (DOY 210 – 216) was selected for comparison with the week of the extreme precipitation event. During the study period (2013-2014), this week experienced the second largest precipitation events after the September 2013 events, matched the meteorological and soil conditions fairly well (similar $T_{\text{air}}$ and $T_s$, and starting $\theta_{0-15 \text{ cm}}$), and the aboveground biomass was at a similar seasonal stage in phenology, occurring after the peak growth and during a period of slow or no growth.

Time series plots were generated that showed changes in soil $[\text{CO}_2]$ at each depth over the week of the extreme precipitation events (September 9 – 15, 2013) and the week of July 29 – August 4, 2014. In addition, a small number of profile plots were created to show soil $[\text{CO}_2]$ changes at each depth with reference to the surface between two consecutive, noteworthy measurements over 1-hour. Plots of soil $[\text{CO}_2]$ at the three soil depths and $T_s$ for the extreme precipitation week and the comparison week in 2014 were generated to show the progression of the relationship as soils wetted up and to evaluate any diel hysteresis.
3.2.9 Statistical analysis

A Pearson’s r was computed to assess the relationship between all model estimates of $R_s$ and both the nighttime eddy covariance $R_s$ and the soil flux gradient calculations, utilizing the 2013 growing season, but excluding the extreme precipitation week. In addition, a Pearson’s r was computed to assess the relationship between $R_s$ and $T_s$ and between soil [CO$_2$] and $T_s$. For all statistical analyses, the R statistical software package was used (version 3.2.2; R Development Core Team, 2015).

3.3 Results

3.3.1 Precipitation, $\theta$, $T_s$, and soil [CO$_2$] during the event

Cumulative rainfall measurements for the period September 9-15, 2013 were approximately 200 mm, about half of the average annual rainfall for this area. Historic records for the region around Rocky Flats National Wildlife Refuge (NWR) showed that total annual precipitation for 2013 was about 50% higher than the 30-year average (NOAA, 2015; Figure 3.3).
It is noteworthy that the monthly precipitation for 2013 was at or below average for every month except February and September, therefore this grassland experienced below average rainfall for the majority of the 2013 growing season. The details of rainfall during the week of September 9-15 (DOY 252 – 258) are presented along with the associated changes in $\theta$, $T_s$, and soil $[CO_2]$ in time series plots in Figure 3.4.
Figure 3.4. The progression of meteorology and soil dynamics during September 9 -15, 2013 (DOY 252 to 258). Time series plots include a) hourly precipitation, b) mean hourly $T_s$ and $T_{air}$, and c) mean hourly soil $[CO_2]$ at 5 cm, 10 cm, and 15 cm, and $\theta_{0-15cm}$. Note that measurements of soil $[CO_2]$ reach the upper measurement limit of the gas analyzer at 5000 ppm during the fully saturated periods.

September 9 (DOY 252) began with high temperatures, both $T_{air}$ and $T_s$ reaching about 30 °C, and low soil moisture, with the $\theta_{0-15cm}$ at 10%, and the shallow soil moisture (0-5 cm $\theta$) at about 3%. A small precipitation event started at 15:00 MST, with more substantial rainfall occurring from 16:30 through 23:30 MST, and rainfall for that day totaling approximately 40 mm. Rainfall increased daily and peaked on September 12
(DOY 255), with a 15 hour rain event and a total of 165.4 mm of precipitation. Table 3.2 details the daily hydrology and range of temperatures during the week.

Table 3.2. Daily hydrology, *T_s* and *T_air* for the week of the extreme precipitation event - September 9 -15 (DOY 252 – 258) 2013. Precipitation indicates accumulated rainfall for the 24-hour period. Each day had from 1 to 7 events of varying duration, however the “Peak P duration” is the single longest rainfall event given in units of hours. For example, the peak rainfall duration on DOY 258 was 15 hours from start to finish, and θ for the shallow 0-5 cm depth increased from 25% to 45% for that day.

<table>
<thead>
<tr>
<th>DOY</th>
<th>Precipitation (mm)</th>
<th>Peak P duration (hrs)</th>
<th>θ_0-5cm low (m^3 m^-3)</th>
<th>θ_0-5cm high (m^3 m^-3)</th>
<th>θ_0-15cm low (m^3 m^-3)</th>
<th>θ_0-15cm high (m^3 m^-3)</th>
<th><em>T_air</em> low (°C)</th>
<th><em>T_air</em> high (°C)</th>
<th><em>T_s</em> low (°C)</th>
<th><em>T_s</em> high (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>252</td>
<td>40.1</td>
<td>4.5</td>
<td>3%</td>
<td>16%</td>
<td>3%</td>
<td>20%</td>
<td>11.3</td>
<td>27.7</td>
<td>19.2</td>
<td>30.2</td>
</tr>
<tr>
<td>253</td>
<td>15.0</td>
<td>3.5</td>
<td>16%</td>
<td>22%</td>
<td>20%</td>
<td>24%</td>
<td>13.8</td>
<td>17.5</td>
<td>18.8</td>
<td>21.6</td>
</tr>
<tr>
<td>254</td>
<td>102.4</td>
<td>4.5</td>
<td>21%</td>
<td>32%</td>
<td>24%</td>
<td>37%</td>
<td>14.5</td>
<td>17.5</td>
<td>17.0</td>
<td>20.9</td>
</tr>
<tr>
<td>255</td>
<td>165.4</td>
<td>15.0</td>
<td>25%</td>
<td>46%</td>
<td>29%</td>
<td>43%</td>
<td>14.1</td>
<td>16.4</td>
<td>17.2</td>
<td>18.7</td>
</tr>
<tr>
<td>256</td>
<td>8.6</td>
<td>3.5</td>
<td>42%</td>
<td>46%</td>
<td>43%</td>
<td>45%</td>
<td>13.6</td>
<td>22.5</td>
<td>16.8</td>
<td>23.6</td>
</tr>
<tr>
<td>257</td>
<td>0.3</td>
<td>0.5</td>
<td>34%</td>
<td>42%</td>
<td>37%</td>
<td>42%</td>
<td>12.0</td>
<td>23.4</td>
<td>17.2</td>
<td>23.2</td>
</tr>
<tr>
<td>258</td>
<td>30.0</td>
<td>5.5</td>
<td>32%</td>
<td>45%</td>
<td>35%</td>
<td>45%</td>
<td>11.8</td>
<td>15.2</td>
<td>16.5</td>
<td>18.7</td>
</tr>
</tbody>
</table>

During the first four days of the event, θ_0-15 cm increased from about 10% to approximately 45% (completely saturated) and *T_air* and *T_s* decreased, both in magnitude and the range between the high and low values. Although soil water responded rapidly to the water inputs, soil [CO₂] remained steady and low throughout the first day and into the early hours of September 10 (DOY 253). Thirteen hours after the first rainfall (accumulated 41.4 mm) and once θ_0-15 cm reached about 20%, soil [CO₂] responded very quickly at all depths - the 15 cm soil depth went from about 1300 ppm to approximately
4200 ppm between the 4:00 - 5:00 MST measurement period. Increasing soil water content continued to increase soil [CO$_2$] until all soil depths reached the peak of 5000 ppm midway through September 11 (DOY 254), when $\theta_{0-15\text{ cm}}$ climbed above 30%.

The soil [CO$_2$] values briefly decreased at various depths just after the largest rain events on September 12 (DOY 255). Although it is possible that the measurements were compromised by the saturated conditions, there is some evidence that these values were valid as respiratory pulses aligned in time with the eddy covariance measurements, indicating a large release that is consistent with gas escaping saturated soils as they infiltrate into deeper soil layers.

A period of quiescence from September 13 - 14 (DOY 256 – 257) resulted in a return of a wider range of $T_{air}$ and $T_s$, a slow draining of soil water, and soil [CO$_2$] at 5cm briefly dropped to about 3000 ppm. Relatively modest rains returned on September 15 (DOY 258), resulting in narrowing the range of $T_s$, already wet soils quickly climbing to fully saturated, and soil [CO$_2$] increasing at all depths.

3.3.2 Comparison to July-August, 2014 precipitation event

Time series plots of the comparison week during July 29 – August 4, 2014 (DOY 210 – 216) show the precipitation event and subsequent changes in $T_s$, $\theta$ and soil [CO$_2$] (Figure 3.5).
Figure 3.5. Changes in meteorology and soil dynamics during July 29 – August 4, 2014 (DOY 210 – 216) showing a) hourly precipitation, b) $T_s$, and c) the mean hourly soil $[CO_2]$ at 5 cm (light grey triangles), 10 cm (dark grey squares) and 15 cm (black circles) with $\theta_{0-15cm}$ (thin black line). Measurements of soil $[CO_2]$ reach the upper measurement limit of the gas analyzer at 5000 ppm during the fully saturated periods.

The rain events that started late on July 29, 2014 (DOY 210) resulted in rapid responses in $T_s$, $\theta_{0-15cm}$, and soil $[CO_2]$. Soil temperatures were already decreasing on DOY 210 due to nighttime decreases, but remained low throughout July 30 (DOY 211), when the bulk of the precipitation fell. Soil water content (0-15 cm) increased from about 12% to approximately 30% within four hours of the first rainfall, and soil $[CO_2]$ increases were nearly synchronized with increases in $\theta$, with the 5cm soil $[CO_2]$ increasing within
the first hour of rainfall, just ahead of the soil [CO₂] increases in the deeper soil layers. Diel variations in soil [CO₂] and Tₛ were apparent before the rain started, and returned within hours after precipitation ended on July 31, 2014 (DOY 212). In contrast, during the week in September 2013, soil [CO₂] displayed very little daily cycling before or after the extreme precipitation events and soil [CO₂] did not increase for many hours after the start of rainfall, when all three soil depths increased together (Figure 3.4).

Depth profiles of soil [CO₂] measurements of four, 1-hour time spans for September 2013 (Figure 3.6), and two, 1-hour time spans for July-August, 2014 (Figure 3.7) explore the processes that occurred belowground during the precipitation events, and contrasts the difference between these two weeks.
Figure 3.6. Depth profile plots of soil [CO2] changes during selected days and times - each plot shows the values for the sampling at the start of one hour (white circles) to the next (black circles). The vertical axis shows the elevation in cm, 0 is the value at land surface (from ambient intake line at the auto-sampler), and the horizontal axis denotes the soil [CO2] in ppm. Soil water content ($\theta$) for 0-15 cm is denoted in the center of the plot. The direction of change from the start of one hour to the next is indicated by grey arrows. Selected periods are a) 9/10 (DOY 253) from 4:00 to 5:00 MST, b) 9/12 (DOY 255) from 12:00 to 13:00 MST, c) 9/13 (DOY 256) from 7:00 to 8:00 MST, and d) 9/15 (DOY 258) from 11:00 to 12:00. Although soil water content had begun to rise during the previous 8 hours, this 1-hour time span was the first notable response in soil [CO2].
The first substantial change in soil [CO₂] change for the week of the extreme precipitation event was on September 10, 2013 (DOY 253) between 4:00 and 5:00 MST. The largest change was at the 10 cm depth, which increased about 3000 ppm in one hour. During the rainfall with the longest duration and intensity on September 12, 2013 (DOY 255), before soils were completely saturated, the vertical soil [CO₂] depth profile developed negative soil gradients (diffusion downward) were created when the 15 cm soil depth was nearly 1500 ppm lower than the 10 cm soil depth above it (Figure 3.6 b). Once the soils were completely saturated, unstable and negative gradients developed at various times. An extensive and large negative gradient developed on September 13, 2013 (DOY 256) when the difference between the 5 cm and 15 cm soil [CO₂] was about -3300 ppm at 7:00 MST, followed by a smaller stable positive profile at 8:00 MST (Figure 3.6 c). At the end of the extreme precipitation week, the soil profile shifted between high and moderate soil [CO₂] values in a uniform manner with little apparent vertical gradient (Figure 3.6 d).

Figure 3.7. Depth profile plots of soil [CO₂] changes over a 1-hour time span for a) July 29, 2014 (DOY 210) from 16:00 to 17:00 MST and b) August 3, 2014 (DOY 215) from 11:00 to 12:00 MST.
In contrast, the initial change in soil [CO₂] on July 29, 2014 (DOY 210) was largest at the 5 cm depth, with almost no change at the 10 cm depth and a modest increase at the 15 cm depth. No negative or downward gradients evolved in the soil profile during the rain events, and once a soil gradient emerged on July 31, 2014 (DOY 212), it remained stable throughout the remainder of the week of July 29 – August 4, 2014 (Figure 3.7 b).

3.3.3 Comparison of θ decay characteristics

Starting dates and times were chosen for each the comparison weeks on which rainfall ceased and water was allowed to drain freely from the soil. Table 3.3 gives the accumulated precipitation for the 72-hours before the start time, the average Ts for the 72-hours following the start time, and the \( \theta_{0-5\text{cm}} \) and the \( \theta_{0-15\text{cm}} \) values at the start time and at 72-hours following the start time. A time series plot of the soil water decay, the change in \( \theta_{0-5\text{cm}} \) and \( \theta_{0-15\text{cm}} \) over the next 72 hours, is presented in Figure 3.8.

<table>
<thead>
<tr>
<th>Start of ( \theta ) decay</th>
<th>Accum P Previous 72 Hrs (mm)</th>
<th>Start ( \theta ) 0-5cm (%)</th>
<th>Start ( \theta ) 0-15cm (%)</th>
<th>After 72 hrs 0-5cm (%)</th>
<th>After 72 hrs 0-15cm (%)</th>
<th>Avg Ts (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>July 31, 2014, 9:00 MST</td>
<td>81.9</td>
<td>29</td>
<td>33</td>
<td>18</td>
<td>24</td>
<td>20.32</td>
</tr>
<tr>
<td>September 16, 2013, 8:00 MST</td>
<td>40.8</td>
<td>45</td>
<td>45</td>
<td>29</td>
<td>33</td>
<td>18.50</td>
</tr>
</tbody>
</table>

Table 3.3. Comparison of soil saturation from precipitation events.
Figure 3.8. Time series plot comparing soil volumetric water content ($\theta$) decay over time after peak saturation during the extreme precipitation events of September 2013 and the week of July 29 – August 4, 2014. Boxes indicate the starting date and time, and the x-axis indicates the number of hours that have elapsed since that start time. Solid lines are the $\theta$ for 0-5 cm depths, and dotted lines are the $\theta_{0-15cm}$.

The most notable difference between the soil water decays for the two comparison weeks is the starting values. September 16, 2013 (DOY 259) began with the $\theta_{0-5cm}$ and the $\theta_{0-15cm}$ values equal and at full saturation, whereas the $\theta_{0-15cm}$ starting value on July 31, 2014 (DOY 212) was very wet, nearly 5% lower for $\theta_{0-5cm}$, and the soils were not saturated. For the first six hours after drainage begins, the slope for all of the soil water decays are shallow and very similar, after which the rate increases for the $\theta_{0-5cm}$ value on DOY 212. This shallow soil water drainage does not increase for the week of the extreme precipitation event until after 12 hours have elapsed. After the first six hours of soil water drainage that started on DOY 212 remained fairly constant for all depths measured. However, DOY 259 saw the rate of drainage for the shallow soils increase faster than the full profile, and so a stratification of soil water values emerged after 18 hours, and the soils drained faster in 2013 as compared to the week in 2014.
3.3.4 Hysteresis comparison

The results of the Pearson’s r analysis indicated there was a significant, but weak correlation between the hourly soil \([\text{CO}_2]\) and mean hourly \(T_s\) for September 2013 (5 cm depth, \(n = 168, r = 0.2568, p < 0.001\)) and July-August 2014 (5 cm depth, \(n = 168, r = 0.1145, p < 0.001\)). See APPENDIX 1 for more information regarding the relationship between \(\theta\) and \(T_s\). However, plots of soil \([\text{CO}_2]\) as a function of \(T_s\) for the two comparison weeks are useful in assessing the hysteresis that may exist between these variables (Figure 3.9).

![Figure 3.9](image-url)

Figure 3.9. Plots showing the relationship between hourly soil \([\text{CO}_2]\) (for 5, 10, and 15 cm depths) and mean hourly \(T_s\) for the week of a) September 9-15, 2013 (DOY 252 – 258) and b) July 29-August 4, 2014 (DOY 210 – 216), black arrows indicate the direction of hysteresis. The time progression within each plot was similar for both weeks – the initial conditions are on the lower left, as rains commenced during the first day, soil \([\text{CO}_2]\) values changed little as \(T_s\) increased, the relationship between soil \([\text{CO}_2]\) and \(T_s\) varied left and right and up the plot after the precipitation events caused the soils to wet up during the middle of the week, and as rains ceased soil \([\text{CO}_2]\) decreased while \(T_s\) varied little.
A shallow, counter-clockwise hysteresis between the soil [CO₂] at 5cm and Ts is apparent for the week of July 29-August 4, 2014 (DOY 210 – 216), after θ began to decreased and soil [CO₂] had modest values between 2000 and 3000 ppm, during the end of the week (DOY 213 - 216). During this time, diel variations in both soil [CO₂] and Ts resumed and caused the relationship between the variables to differ between cooling and warming cycles. A counter-clockwise hysteresis developed when the soil [CO₂] values during warm-up were greater than those at the same temperature during cool-down. Hysteresis was missing completely from the week in September 2013, and soil [CO₂] appears to be insensitive to Ts at the start of the rainfall, when soils were still dry.

3.3.5 Soil flux gradient

There was a significant correlation between mean nighttime Rs (for 60-minute measurements with net radiation <= 20 Wm⁻²) calculated from the flux gradient method and mean nighttime Rs from eddy covariance measurements for the 2013-growing season (n = 115, r = 0.3827 p < 0.001). Figure 3.10 is a time series plot of the soil flux gradient and nighttime Rs from eddy covariance over the growing season, with an inset plot showing the regression of the two Rs measurements.
Figure 3.10. Time series plot of the mean nighttime (based on solar radiation $\leq 20$ Wm$^{-2}$) respiration ($Rs$) from eddy covariance and the soil flux gradient method, from April 1 - September 30, 2013. Large grey arrows above and below the graph indicate the extreme precipitation event week. Inset graph (center top) shows the regression between the two variables for the same time period, however excluding the values during the extreme precipitation week.

Respiration from the flux gradient method underestimated the $Rs$ early in the growing season and during the first part of August, but matched the eddy covariance flux variations during these times, and more closely matched the eddy covariance values for the remainder of the growing season. The week of the extreme precipitation event had the least agreement between the flux values.

3.3.6 Empirical models

Hourly $Rs$ calculated from the Lloyd and Taylor (1994) model (3.1) were not well correlated with hourly soil flux gradient measurements for the week of September 9 – 15 ($n = 166, r = 0.01091$ $p > 0.1$). Further, while the Vicca et al. (2014) models estimates of
Rs had significant correlations with the hourly soil flux gradient values for the week of the extreme precipitation events (eq. 3.2, n = 166, r = 0.2386 p < 0.001; eq. 3.3, n = 166, r = 0.1889 p < 0.001; eq. 3.4, n = 166, r = 0.1968 p < 0.001; eq. 3.5, n = 166, r = 0.7812 p < 0.001), the relationships were weak for all but the last model, therefore eq. 3.5 was the only empirical model included in the analysis of cumulative fluxes. This forth empirical model from Vicca et al. (2014) will be referenced as simply “the Vicca equation” for the analysis and graphs from this point forward. The values for the coefficients used in the model are found in Table 3.2.

3.3.7 DAMM model

The range of Vmax and [Sx] relationships for different temperatures for this soil were generated to determine $kM_{Sx}$ and the results of three of the temperature-dependent fitted curves are plotted in Figure 3.11.
Figure 3.11. Mean hourly respiration rates (RSx, umol m\(^{-2}\) s\(^{-1}\)) plotted against the substrate concentrations [Sx] (g C m\(^{-2}\) s\(^{-1}\)) for three specific temperatures (indicated at the end of the lines) for observations (diamonds; black = 13 °C, white = 15 °C, grey = 19 °C) and fitted curves (solid and dashed lines). Inset plot (center bottom) shows the linear relationship between Ts and the estimated Michaelis constants for the soil carbon substrate.

The Michaelis constant for the carbon substrate (kM\(_{Sx}\)) was not very sensitive to temperature, as seen in the inset regression plot in Figure 3.11. This is apparent from the left side of the main plot where the curves for a range of temperatures converge at lower values of [Sx].

For the 2013 growing season, there was a significant, positive relationship between eddy covariance measurements of the mean nighttime Rs and the DAMM model estimates (n = 166, \(r^2 = 0.3132\), \(p < 0.001\)). There was also a positive relationship between soil flux gradient mean nighttime Rs and the DAMM model estimates (n = 115, \(r^2 = 0.2645\), \(p < 0.001\), Figure 3.12).
Figure 3.12. Evolution of the mean nighttime respiration ($Rs$) from the eddy covariance method and the DAMM model from April 1 - September 30, 2013. The large grey arrows above and below the graph indicate the extreme precipitation event week. Inset graph (center top) shows the regression of the two variables during the same time period, however excluding the values during the extreme precipitation week.

The time series of the two $Rs$ values over the 2013 growing season shows very good agreement from April through July, after which the DAMM model overestimates fluxes (by comparison to eddy covariance) in August, when the soils were very dry, followed by underestimating $Rs$ during the extreme precipitation event in September.

During the extreme precipitation event, there was no significant correlation between hourly $Rs$ from eddy covariance and the DAMM model estimates ($n = 168, r^2 = 0.0318, p > 0.10$) or the Vicca eq. 3.9 model estimates ($n = 168, r^2 = 0.0289, p > 0.10$). In contrast, there is a strong positive relationship between hourly $Rs$ from the soil flux gradient method and the DAMM model estimates ($n = 168, r^2 = 0.7158, p < 0.001$), and the $Rs$ estimates generated by the Vicca equation, empirical model ($n = 168, r^2 = 0.7812$, $p < 0.001$).
p < 0.001). A plot of the regressions between hourly Rs from model estimates and the flux gradient shows that the DAMM model’s fitted line is much closer to a 1:1 relationship with the soil flux gradient measurements as compared to the Vicca empirical model Figure 3.13.

Figure 3.13. Plot of hourly Rs during the extreme precipitation event (September 9 – 15, 2013) showing the relationship between the empirical model estimates (Vicca et al. 2014) and the DAMM model estimates and the soil flux gradient Rs.
Figure 3.14 shows a comparison of the cumulative $Rs$ from four different observations and estimates – the soil flux gradient method, eddy covariance estimates from nighttime $Rs$ and $Ts$, estimates using the Vicca empirical model, and estimates from the DAMM model.

![Graph showing cumulative Rs during the extreme precipitation event, September 9-15, 2013.](image)

There is a distinct point during DOY 255 when the soil flux gradient and the DAMM model diverge from the eddy covariance measurement and the Vicca empirical estimates. Respiration seems to nearly cease for almost 24-hours in the case of the two diverging accumulation lines. In contrast, there is little noticeable change in accumulation rate for the eddy covariance $Rs$ and empirical model estimates, as these lines have a nearly constant slope for the entire week.
3.4 Discussion

The impact of the extreme precipitation events of September 2013 on $\theta$, soil [CO$_2$] and $Rs$ contrasted sharply with that of a typical rain pulse at the grassland study site in Rocky Flats NWR. As soil [CO$_2$] increased with soil water infiltration, the soil [CO$_2$] gradient during the week in July – August 2014 stayed stable and positive (upward) during most of the wet up, while during the extreme precipitation event the soil became fully saturated for several days and appeared to distort the soil [CO$_2$] gradient at times. Soil respiration appeared to be insensitive to $Ts$ during the extreme precipitation event because of a lack of hysteresis and infrequent, large pulses of CO$_2$ that are consistent with saturated conditions in the soil exerting the dominated influence on CO$_2$ production and transportation. Conversely, hysteresis between soil [CO$_2$] and $Ts$ developed shortly after rainfall ceased during the week in July 2014, indicating temperature driven processes had returned. In addition, observations during the week in September 2013 of short-term, large pulses of CO$_2$ from the soil gas wells and eddy covariance tower seemed to indicate that occasional efflux of CO$_2$ at the land surface was caused by physical processes other than diffusion (see section 3.4.1 below). Model estimates and soil flux gradient values of $Rs$ were in agreement with eddy covariance for most of the growing season, however the process-based model estimates (DAMM) and soil flux gradient $Rs$ diverged abruptly from eddy covariance values during the week of the extreme precipitation events, having much smaller fluxes during the longest rainfall duration and highest saturated soil conditions.
3.4.1 Soil [CO₂] gradient changes

During both precipitation weeks, a reversal of the soil [CO₂] gradient occurred at the start of soil wet-up. A positive soil [CO₂] gradient returned quickly during the week in 2014, even as rains continued to increase θ, however the gradient remained unstable throughout the week of September 2013. Under steady-state θ conditions, CO₂ production in the upper soil layers generally creates a concentration gradient that increases with soil depth. Since diffusion moves gases from high to low concentrations, the gradient drives movement of CO₂ upward, towards the land surface. When rainfall occurs, the addition of water in the uppermost soil layers makes the labile pool of carbon available to the soil microbial community, where decomposition proceeds rapidly and generates additional CO₂. The concentration gradient could be reversed temporarily as the uppermost soil layer produces more CO₂ than deeper layers. This seems to be the case at the beginning of the week in 2014, when soil [CO₂] at 5 cm increased while deeper soil layers remained unchanged. The changes in soil [CO₂] went deeper at the start of the extreme precipitation events in 2013, with the three observed soil layers increasing, though at different rates, within the same hour.

While it is possible that some CO₂ could be driven downward due to a negative gradient, a positive soil [CO₂] gradient usually returns when large releases of CO₂ are caused by mass flow, also known as pressure pumping, due to wind at the land surface (Oikawa et al., 2014; Roland et al., 2015). Another physical process that moves CO₂ to the surface is advective gas transport due to decreased air-filled pores as the soil wets up (Birch, 1958; Orchard and Cook, 1983; Ryan and Law, 2005). There is evidence of large gas releases from mass flow or advection during the extreme precipitation event.
Downward spikes in soil [CO$_2$] are seen during the later part of the week in 2013 (Figure 3.4 c), and apparent sharp reverses are seen in soil [CO$_2$] profiles in Figure 3.6 (b, c and d). Since there were no substantial changes in precipitation, $\theta$ or $T_s$ that could explain these sudden changes in soil [CO$_2$], mass flow or advective gas transport processes were the probable physical processes at work (Birch, 1958; Guo et al., 2008).

3.4.2 Model estimates of $R_s$

The model estimates of $R_s$ during the extreme precipitation week that were used in this study indicate that extraordinary field conditions existed during the events and this challenged the validity of the values that were produced. The DAMM model matched the soil flux gradient variations and magnitudes quite well, likely due to the incorporation of soil [O$_2$] changes and process-based dynamics to estimate the production of CO$_2$. However, both the soil flux gradient and DAMM model fail to completely characterize pulse releases of CO$_2$ from physical processes other than diffusion. Conversely, the eddy covariance measurements included substantial gap-filling during heavy rains (71%), and therefore respiratory pulses were included even as meteorological and soil conditions were definitely hostile to decomposition or other CO$_2$ production processes (i.e., heavy rains, saturated and anaerobic soils). The difference in cumulative $R_s$ for this one week demonstrates how these differences impact estimates of carbon cycling during extreme conditions (Figure 3.14). The soil flux gradient and DAMM model computed nearly half the accumulated carbon than the amount that was totaled by the eddy covariance and the empirical model.
While process-based model may be better suited to estimate soil fluxes, they are still difficult to parameterize and this limits their widespread use. On the other hand, simpler empirical model estimates of $Rs$ depend solely on $Ts$ and, in some cases, $\theta$. These estimates, however, do not fully capture fluxes from pulse events because the efflux of soil CO$_2$ and its’ abiotic drivers become decoupled during rapid soil wetting (Oikawa et al., 2014; Porporato et al., 2004; Riveros-Iregui et al., 2007; Ryan and Law, 2005). Although hysteresis between $Rs$ and $Ts$ is usually a challenge to empirical models due to more than one possible range of $Rs$ values for every $Ts$, no apparent hysteresis between $Rs$ and $Ts$ developed during the week of extreme precipitation and the models were still in disagreement. This is likely due to $Rs$ being primarily controlled by $\theta$, with little influence from $Ts$ during this period (see APPENDIX 1 Figure A - 3 and APPENDIX 6). In addition, models may fail to account for shifts in environmental conditions that act as a tipping point – the assumption that gaseous diffusion is the dominant transport mechanism breaks down when the air filled pore spaces become smaller and disconnected from each other and land surface processes influence shallow soil layers (Borken and Matzner, 2009; Roland et al., 2015).

### 3.4.3 Implications of the extreme precipitation event

An extreme precipitation event like the one that occurred in Colorado in September 2013 can be an ecological disturbance, affecting the resilience of the ecosystem (Bengtsson, 2002). The flora and fauna may be unable to cope with the amount of soil water that is added, especially in a semi-arid community (Ryel et al., 2010). Changes in the soil structure and composition are possible since the flush of water
is capable of changing the hydrophobicity of the soil due to disruption of the organo-mineral complexes (aggregates of organic matter and mineral particles), making fresh organic matter available (Christensen, 2001; Lal, 2003). However, disturbances have been known to improve ecosystem function. For example, long-term evaporation at the land surface may concentrate solutes dissolved in the water that pools there, and large precipitation events may be needed to flush these out (Borken and Matzner, 2009).

Species composition changes can occur when disturbances remove competitors or create new niches (Bengtsson, 2002).

The mixed grassland community at Rocky Flats NWR that was the focus of this study had a late season growth burst in 2013 of the dominant plant species, a productive growing season the following year, and there were no obvious short-term negative impacts to the ecosystem (see Chapter 2). However, the long-term influence to ecosystems from changes in the precipitation regime, including an increase in very large precipitation events, are at the early stages of study and opportunities to add to these observations may be forthcoming if GCM predictions are correct. Although model estimates of carbon exchange may be challenged by extreme precipitation events, understanding how to correctly characterize these exchanges is becoming increasingly important.
CHAPTER 4. WATER AND CARBON CYCLING IN NEIGHBORING C₃ AND C₄-DOMINATED SEMIARID GRASSLANDS

4.1 Introduction

The timing of aboveground productivity in grasslands is influenced annually by the composition of plants that are either C₃ (cool season) or C₄ (warm season) photosynthetic types (Epstein et al., 1997; Ode and Tieszen, 1980; Seastedt et al., 1994; Tieszen et al., 1997; von Fischer et al., 2008). Grass species evolved to adapt to warm or cool climates and they exhibit functional traits related to temperature and water availability, though grasses of both photosynthetic types are found in varying proportions throughout the temperate regions of North America. The demand for water is linked to the grass species seasonal affinity and therefore changing intra-annual precipitation patterns (Christensen et al., 2013) and accompanying water availability could impact the magnitude of carbon uptake in a grassland dominated by one photosynthetic type.

The C₃ photosynthetic pathway evolved first and is still used by the vast majority of plants. The C₃ pathway utilizes rubisco, an enzyme that fixes CO₂ but is also prone to fix oxygen in a wasteful process known as photorespiration. Photorespiration losses increase with increasing temperature, therefore C₃ plants are more efficient and productive in cooler climates (Ehleringer and Cerling, 2002). In contrast, C₄ plants initially fix CO₂ with a different enzyme and then complete the carbon assimilation process in another part of the leaf, with no losses due to photorespiration (Ehleringer et al., 1997). While this process requires more energy as compared to C₃ photosynthesis, the C₄ pathway allows the plant to fix carbon at a higher rate in high temperatures and with less water loss (Tieszen et al., 1979). Warm season (C₄) grasses are thought to have
evolved in the tropics and subtropics where the mean annual temperatures are higher on average compared to other terrestrial biomes (Ehleringer et al. 1997). Globally, C_{4} plants account for only about 5% of plant biomass yet they make up approximately 18% of primary production, mostly from grass species (Epstein et al. 1997, Bond et al., 2004). The photosynthetic type of the grass species influences the response to changes in climate as well as future distributions and grassland compositions (Ehleringer et al. 1997, Epstein et al. 1997).

Different environmental factors can stimulate opposing spatial and temporal responses that favor one type of grass over another. All C_{4} species (the vast majority are grasses) across the U.S. Great Plains are strongly correlated to mean annual temperature so that C_{4} grasses decrease in abundance northward with decreasing mean annual temperatures, with a crossover of 50-50% C_{3} - C_{4} plant composition at 43-45° latitude (Tieszen et al. 1997, Epstein et al. 1997, Ehleringer et al. 1997). Primary productivity is linked to seasonal preferences, therefore C_{3} grasses are most productive in the winter-spring growing season when temperatures are cooler and C_{4} grasses drive summer productivity (Ehleringer et al. 1997). Mean annual precipitation can also influence the distribution of C_{4} grasses in areas where the timing of precipitation in the late summer months (July and August) occurs when these plants are most productive. Rising CO_{2} concentrations enhances C_{3} competitiveness by decreasing photorespiration and inducing water-use efficiencies (Epstein et al. 1997, Morgan et al. 2011), however biogeochemical feedbacks generate considerable uncertainty regarding this effect. Free-air Carbon Dioxide Enrichment (FACE) studies have shown that as atmospheric CO_{2} increases, there is a reduction in stomatal conductance and an increase in water-use efficiency in plants,
or the amount of dry biomass produced to water lost through transpiration (Leakey et al., 2009). Although it has been proposed that this response should be strongest when water is limited, several studies that tested this hypothesis have resulted in little agreement (Hovenden et al., 2014). Nevertheless, Epstein et al. (1997) estimates that a warming of 2° C will push C$_3$ species occurrence northward in the Great Plains, decreasing from 35% to 19% of grassland dominance in the U.S. Great Plains.

Few studies compare soil water use between cool season (C$_3$) and warm season (C$_4$) grasses in the same area and so it is unclear how these different grasses will be affected by changes in soil water availability. In a study in an east-central Minnesota grassland, Craine et al. (2002) found that C$_3$ grasses utilized very shallow (0-5cm) soil water during the growing season while the C$_4$ tallgrass species used broader, deeper soil water reserves. In contrast, the Nippert and Knapp (2007) study in Kansas found that C$_4$ grassland species, including one of the same species in the Minnesota study (*Andropogon gerardii*), utilized shallow soil water exclusively. Furthermore, there are few studies comparing soil respiration within systems composed of differing photosynthetic types (Morgan et al. 2011) or within grasslands that were reclaimed after cultivation (Burke et al. 1995). Moreover, many studies use coarse temporal sampling (e.g. weekly, bi-weekly) of soil water and/or respiration that miss short-lived responses, such as episodes of rainfall on dry soil and the belowground wetting cycle that induce pulses of CO$_2$ efflux (Ford et al. 2012).

The objective of this research was to compare the seasonal differences in above and belowground water and CO$_2$ fluxes between two neighboring semiarid grasslands – a C$_3$ dominated grassland composed of introduced grass species on reclaimed land, and a
nearby C$_4$ dominated, native remnant tallgrass prairie. The grasslands represent distinct plant photosynthetic types that should respond differently to changes in seasonal water availability. The dominant photosynthetic type at each site means that these grasslands are most productive during different seasons, and therefore:

*H4:* The C$_3$ grassland will assimilate more carbon during the Spring and Autumn, and by comparison, the C$_4$ prairie will assimilate more carbon during the Summer months.

Given that water demands also vary seasonally between these vegetation types, it is proposed that:

*H5:* The C$_3$ grassland will be dependent on adequate water availability early in the growing season and the C$_4$ prairie will be more productive when summer rainfall and soil water content is abundant.

Because the life cycle requirements of these two types of grasses will utilize pulses of shallow soil water and nutrients at different times of the year, belowground responses at the grasslands are strongly related to aboveground activity. To test these hypotheses, the research utilized continuous aboveground measurements of water and carbon fluxes and discrete sampling of biomass with belowground measurements of soil CO$_2$ concentration and soil water and temperature during two sequential calendar years with annual precipitation above the 30-year mean, yet with distinct intra-annual precipitation patterns. Research compared fluxes of water and CO$_2$ seasonally and between grasslands, examined changes in biomass and estimates of water use efficiency
(WUE), and compared how changes in soil water affected the sensitivity of respiration to varying \( T_s \) differently between the two grassland types.

4.2 Methods

4.2.1 Study area

Water and CO\(_2\) flux observations were measured at two sites in Colorado, within the Rocky Flats National Wildlife Refuge (NWR) approximately 5 km apart - hereafter referred to as the C\(_3\) grassland and the C\(_4\) prairie. The C\(_3\) grassland, located on the southeast section, was a reclaimed, mixed grassland, dominated by cool season grasses, and the neighboring C\(_4\) prairie, located in the southwest section, was a native, tallgrass prairie dominated by warm season grasses (Figure 4.1).
Figure 4.1. Map showing the two grasslands within Rocky Flats National Wildlife Refuge - the C₃ grassland was a reclaimed, mixed grassland dominated by smooth brome and the C₄ prairie was a native, tallgrass prairie dominated by big bluestem (map modified from USFWS, 2005).

The C₃ grassland site was originally cultivated and then re-seeded over 10 years ago with a mixture of primarily smooth brome (*Bromus inermis* Leyss) and intermediate wheatgrass (*Thinopyrum intermedium*), both perennial, cool season C₃ grasses (USFWS 2005). For complete details, see Chapter 2 (section 2.2.1).

The C₄ prairie is a native, tallgrass prairie, a rare vegetation community covering over 600 ha of the refuge, and mainly comprised of perennial, warm season C₄ grasses
(Andropogon gerardii, Panicum virgatum, Schizachyrium scoparium, Muhlenbergia montana, Verbascum thapsus and Sorghastrum nutans) (USFWS 2005). The dominant grass species, big bluestem (Andropogon gerardii), grows in bunches and has deep roots that can extend to ~3 meters (USDA, 2014). All vegetation at the site in Rocky Flats NWR reached maximum heights of ~39 cm in July and August of 2013, and ~85 cm in October of 2014. Vegetation at the C₄ prairie site was sparse, low density, and heterogeneous in comparison to the C₃ grassland, with some bare soil exposed between bunchgrass and a thin litter layer (Figure 4.2). Soils within the tallgrass prairie are very cobbly to very stony loamy. A summary of the site locations, species composition and soils is provided in Table 4.1.
Figure 4.2. Photo of the tallgrass C$_4$ prairie site on March 29, 2014, looking southwest. Soil plots and instruments are seen inside a fence, covered with screening to discourage animal disturbance.
Table 4.1. Attributes for the two grassland sites at Rocky Flats NWR used in the study.

<table>
<thead>
<tr>
<th>Attribute</th>
<th>C3 Grassland</th>
<th>C4 Prairie</th>
</tr>
</thead>
<tbody>
<tr>
<td>Latitude, Longitude</td>
<td>39.8765, -105.1702</td>
<td>39.8752, -105.2184</td>
</tr>
<tr>
<td>Elevation</td>
<td>1730 m asl</td>
<td>1821 m asl</td>
</tr>
<tr>
<td>Ecosystem</td>
<td>Reclaimed, mixed grassland</td>
<td>Remnant tallgrass prairie</td>
</tr>
<tr>
<td>Dominant vegetation</td>
<td><em>Bromus inermis</em> Leyss, <em>Thinopyrum intermedium</em></td>
<td><em>Andropogon gerardii</em>, <em>Schizachyrium scoparium</em></td>
</tr>
<tr>
<td>Soils</td>
<td>Standley-Nunn gravelly clay loams 0 - 5° slope</td>
<td>Flatirons very cobbly sandy loam 0 - 3° slope</td>
</tr>
</tbody>
</table>

asl = above sea level

4.2.2 Precipitation

Regional precipitation measurements were obtained from the Measurement and Instrumentation Data Center (MIDC) of the National Renewable Energy Lab (NREL), collected continuously from a heated tipping bucket rain gauge (320b, Belfort Instrument, Baltimore, MD) mounted on a tower at the 2-m height (39°54'36.0"N 105°14'06.0"W) (Jager and Andreas, 1996), which is located approximately 5 km north of the Rocky Flats NWR. The NREL observations were used for rainfall analysis in preference to the tipping bucket rain gauges located at the study sites because the annual precipitation records for 2013-2014 were complete at NREL, and additional meteorological records dating back to 2000 were available for long-term analysis.
4.2.3 *Eddy Covariance*

Each grassland study site had ongoing eddy covariance-based measurements of surface energy balance, including net ecosystem exchange (*NEE*), derived from the turbulent exchange of CO₂ between the atmosphere and land surface. In July 2010, the U.S. Geological Survey (USGS) installed a 3-m-tall eddy covariance flux tower at an elevation of 1821 m asl in the C₄ prairie (Figure 4.3). Wind components were measured by a three-dimensional sonic anemometer (CSAT3, Campbell Scientific, Inc., Logan UT, USA), and CO₂ and H₂O mole densities were measured by a closed-path infrared gas analyzer or IRGA (LI-7200, LI-COR, Lincoln, NE, USA), and separation distances for the center of LI-7200 relative to center of CSAT path were 12 cm east and 12 cm north, and the CSAT had a north offset of 160°. Air temperature (*Tₐir*) and relative humidity was measured with a shielded sensor (HMP45C, Vaisala, Helsinki, Finland), and net radiation was measured with a net radiometer (model Q*7, Radiation and Energy Balance Systems, Inc., or REBS, Seattle, WA), and Photosynthetically active radiation (*PAR*) was determined by measuring photosynthetic photon flux density using a quantum sensor (LI-190, LI-COR, Lincoln, NE, USA). Soil heat flux was averaged between two heat flux plates (model HFT3, REBS) and thermocouples buried at 5 cm in the soil and located ~1-m southwest and southeast of the tower (see APPENDIX 3 for a description of soil heat flux corrections and energy balance). Soil moisture was measured using two TDR probes (time domain reflectometry; CS616, Campbell Scientific, Inc., Logan, UT), buried at an ~30 degree angle from vertical and integrating soil moisture from the surface to 20 cm soil depth, and located approximately 5-m west-southwest and east-southeast of the
The instruments were powered by two deep cycle, flooded, lead acid batteries connected to three 50W solar panels.

Figure 4.3. Photo of the eddy covariance tower at the C₄ prairie site, looking east-southeast (December 2013).

In December 2012, the USGS installed an eddy covariance flux tower in the C₃ grassland. See Chapter 2 for a complete description of the eddy covariance system at the
C₃ grassland, the data processing and gap-filling methodology, and methods for partitioning NEE into GPP and $R_{eco}$ (section 2.2.5).

The energy balance closure, or the ratio of available energy, the sum of net radiation ($Rn$) and soil heat flux ($G$), to the sum of turbulent fluxes ($H + LE$), is equal to 1.0 when measurements are perfect. Each 30-minute measurement period was identified as nighttime for solar radiation values $\leq 20$ Wm$^{-2}$. The mean 30-minute energy balance closure for the C₃ grassland was 0.987 ($n = 8506$) for 2013, with a daytime average of 0.80 and a nighttime average of 1.27, and an annual closure of 0.91 ($n = 9678$) in 2014, with a daytime mean of 0.86 and a nighttime mean of 0.95. The energy balance closure for the C₄ prairie was 0.94 ($n = 3406$) for 2013, with a daytime average of 0.57 and a nighttime average of 1.86, and an annual closure of 0.70 ($n = 4648$) in 2014, with a daytime mean of 0.77 and a nighttime mean of 0.67. Most of these ratios are within the range of values that are considered acceptable (Blanken et al., 1997; Ivans et al., 2006; Wilson et al., 2002). See 5.1.4APPENDIX 2 for more detail.

4.2.4 Soil Plots

Two soil plots were established at the C₃ grassland study site (see diagram of the plots in Chapter 2, Figure 2.3), and four plots at the C₄ prairie site (see Figure 4.4 for map of two plots). All plots were located within 10-40 meters of the eddy covariance towers and spaced 7-10 meters apart. The plots were equipped to measure soil moisture and temperature at three shallow soil depths - 5, 10 and 15 cm. Table 4.2 lists the plots, instruments and depths for each grassland site. All sensors measured every 60-seconds and the average was recorded every 30-minutes on an Em5b data logger (all soil plot
sensors and data loggers Decagon Devices, Inc., Pullman, WA 99163). Where replicated, soil volumetric water content ($\theta$) and soil temperature ($T_s$) measurements for each soil depth were averaged between the plots.

Table 4.2. Location and soil depths for instruments used to measure soil water content and soil temperature.

<table>
<thead>
<tr>
<th>Instrument</th>
<th>C$_3$ Grassland</th>
<th>C$_4$ Prairie</th>
</tr>
</thead>
<tbody>
<tr>
<td>EC-5, EC-10 (soil water content)</td>
<td>North plot. 5 and 10 cm</td>
<td>RF1 5, 10, 15 cm</td>
</tr>
<tr>
<td></td>
<td>South plot. 5 and 10-15 cm</td>
<td>RF2 5, 10, 15 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RF3 5, 10, 15 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RF4 10 and 15 cm</td>
</tr>
<tr>
<td>RT-1 (soil temperature)</td>
<td>South plot 10 cm</td>
<td>RF1 5 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RF2 10 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RF3 5 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RF4 10 cm</td>
</tr>
</tbody>
</table>
4.2.5 Soil CO₂ Concentrations

Hourly measurements of soil CO₂ concentrations were obtained from soil gas wells. The soil gas wells used in this study were installed in the C₃ grassland during December 2012 and January 2013, and in the C₄ prairie during April – September 2013, and May 2014. Six soil gas wells were divided between two separate plots at each study site. There were three different IRGAs used in the auto-samplers during the study: the C₃ grassland used a Li-800 (LI-COR Biosciences, Lincoln, NE USA), and the C₄ prairie
used a WMA-4 (PP Systems, Amesbury, MA USA) during 2013 and a Li-840 (LI-COR Biosciences, Lincoln, NE USA) during 2014. See Chapter 2 for complete details on soil gas well design, installation, auto-sampler design, the sampling protocol, and soil gradient method CO$_2$ flux calculations (sections 2.2.3 and 2.2.4).

The auto-sampler / soil gas well system located in the C$_4$ Prairie was frequently disturbed by wildlife biting through sample lines or from saturated conditions that included standing water and overland flow. Observations were therefore limited or missing during these times, which included periods during April – September 2013, April - May 2014, and August 2014.

4.2.6 Biomass and leaf area index (LAI)

To characterize changes in aboveground biomass, vegetation samples were collected and analyzed on an approximately bi-weekly schedule during the growing season for each calendar year. See Chapter 2 for details on sampling and lab protocols and MODIS-derived LAI (section 2.2.8).

4.2.7 Water Use Efficiency (WUE)

Water use efficiency (WUE) is defined as the ratio of water used by plants for metabolism to water lost through transpiration. Ecosystem level WUE may be calculated as the ratio between $GPP$ and $ET$, which represents the amount of carbon taken up by the grassland ecosystem to the amount of water lost through both evaporation and transpiration (Brümmer et al., 2012; Hibbard et al., 2005). For this study, WUE was
computed as the absolute value of cumulative monthly $GPP$ to monthly $ET$ and expressed in units of g C kg H$_2$O month$^{-1}$.

4.2.8 Selection of season periods

Missing or poor data from eddy covariance at the C$_4$ prairie that resulted in uncertainties in gas fluxes during the growing season limited the selection of days used to compare seasons. In addition, preliminary results indicated that there was a more distinct early vegetative influence on carbon cycling as compared to later in the growing season, and the extreme precipitation events in September 2013 had an extraordinary impact on carbon and water cycling in both grasslands. Therefore, the growing season refers to March 1 – October 31, and after examining the time series of daily $NEE$, April 1 – June 30 was assigned the “spring” period for 2014 and July 10 - October 31 was assigned the “summer / autumn” seasonal period for both 2013 and 2014.

4.2.9 Calculations / Statistics

A Pearson’s $r$ was computed to assess the relationship between dry weight biomass and estimates of $LAI$, and between $LAI$ and $GPP$. In addition, the relationship between $GPP$ and $ET$ was analyzed in order to compare the water use between the two grasslands. Further, a Pearson’s $r$ was computed to access the relationship between $Reco$ and $Ts$ at each grassland site and for each of the study years. For all statistical analyses, the R statistical software package was used (version 3.2.2; R Development Core Team, 2015).
4.3 Results

4.3.1 Precipitation and soil water content

Though neither of the study years at Rocky Flats NWR was water limited on an annual time scale, precipitation in 2013 was below average during the spring and summer months and an order of magnitude above normal during the historic extreme precipitation events in September, while 2014 precipitation was abundant throughout the spring and summer. Plots of precipitation, $\theta_{0-15cm}$, and $T_s$ at each grassland for each calendar year are presented in Figure 4.5 (2013) and Figure 4.6 (2014).
Figure 4.5. Calendar year 2013 time series plots of a) daily precipitation, b) mean daily $T_s$ and c) mean daily $\theta_{0-15cm}$ at each grassland study site. Note that the magnitude of daily precipitation values during September 2013 would have caused the vertical scale to obscure other daily precipitation totals and therefore two days of precipitation totals during this time were truncated and the values indicated by call-out boxes to the left and right.

Soil temperature was comparable between the two grasslands for both study years, although the trend was for $T_s$ at the C$_3$ grassland to rise slightly above $T_s$ values at the C$_4$ prairie during the growing season, starting around April-May through September. Mean daily $\theta_{0-15cm}$ was more variable at the C$_4$ prairie (s.d. = 13.4% and 10.9%) as...
compared to the C\textsubscript{3} grassland (s.d. = 8.2\% and 4.9\%), which is apparent from the pronounced wet and dry cycles seen at the C\textsubscript{4} prairie in response to precipitation events.

Table 4.3.

Table 4.3. Annual precipitation, mean daily $\theta_{0-15}$ cm\textsuperscript{-3}, and mean daily $T_s$ (at 10 cm) for each grassland site and for both years.

<table>
<thead>
<tr>
<th></th>
<th>Year</th>
<th>C\textsubscript{3} Grassland</th>
<th>C\textsubscript{4} Prairie</th>
</tr>
</thead>
<tbody>
<tr>
<td>Precipitation (mm)</td>
<td>2013</td>
<td>676.52</td>
<td>676.52</td>
</tr>
<tr>
<td></td>
<td>2014</td>
<td>537.29</td>
<td>537.29</td>
</tr>
<tr>
<td>Mean daily $\theta$ (m\textsuperscript{3} m\textsuperscript{-3})</td>
<td>2013</td>
<td>18.1% (8.2%)</td>
<td>15.6% (13.4%)</td>
</tr>
<tr>
<td>0-15 cm depth</td>
<td>2014</td>
<td>20.1% (4.9%)</td>
<td>16.2% (10.9%)</td>
</tr>
<tr>
<td>Mean daily $T_s$ (°C)</td>
<td>2013</td>
<td>11.07 (9.2)</td>
<td>10.70 (8.5)</td>
</tr>
<tr>
<td>10 cm depth</td>
<td>2014</td>
<td>10.98 (8.2)</td>
<td>10.72 (7.3)</td>
</tr>
</tbody>
</table>

Precipitation values apply to both grassland sites; standard deviation values for $T_s$ and $\theta$ are in parenthesis.

However, despite episodes of saturation, soils at the C\textsubscript{4} prairie were on average drier than soils at the C\textsubscript{3} grassland, with the exception of the period following the extreme precipitation in September 2013 when there was pooling water and overland flow observed at the C\textsubscript{4} prairie for weeks following the flood.

4.3.2 Annual net ecosystem exchange (NEE)

Air temperatures followed similar patterns for both years, with cold, winter months having temperatures varying greatly between mean daily minimums and maximums, and the summer months having mean daily temperatures with less range and the mean remaining below 30 °C. Though there were gaps in observations on an annual
basis, particularly at the C_4 prairie, plots of net ecosystem exchange (NEE) along with air temperatures are presented in Figure 4.7 for calendar year 2013 and Figure 4.8 for 2014.

Figure 4.7. Time series plots of a) daily mean $T_{air}$ (C_3 Grassland shown), and b) NEE (grey bars) with 1-week running mean (black line) for the C_3 grassland and the c) C_4 prairie during 2013. The black bars for daily NEE in the C_4 prairie (April 14 – July 9) indicate days for which values have been gap-filled by Max Planck due to missing or poor original values. Note that data is also missing and not gap-filled for the C_4 prairie from November 15 – December 31, 2013 (inclusive). The time period that is analyzed in more detail is marked by the vertical dashed grey lines (Summer / Autumn 2013).
Figure 4.8. Time series plots of a) daily mean $T_{\text{air}}$ (C$_3$ Grassland shown), and b) $\text{NEE}$ (grey bars) with 1-week running mean (black line) for the C$_3$ grassland and the c) C$_4$ prairie during 2014. Note that data is also missing and not gap-filled for the C$_4$ prairie from January 1 – February 28, 2014 (inclusive). The time periods that are analyzed in more detail are marked by the vertical dashed grey lines (Spring 2014 and Summer / Autumn 2014).

There were distinctive periods of net CO$_2$ uptake by the ecosystem or loss to the atmosphere apparent from the time series plots. Both grasslands begin to take up CO$_2$ during the early part of the growing season, with the C$_3$ grassland beginning in May of 2013 and April of 2014, and the C$_4$ prairie beginning much earlier in 2014 as compared to 2013, perhaps even ahead of the C$_3$ grassland for that year, though uptake was much
stronger in June. There were distinct differences in NEE between the grassland during the late part of the growing season, in the summer and autumn of both years. The C₄ prairie was a sink for carbon during July and August of 2013 while the C₃ grassland was a source during this time. Interestingly, after the extreme precipitation events in early September 2013, the grasslands reversed and the C₃ grassland was a sink while the C₄ prairie was a source. During the following summer/autumn, the C₄ prairie was a sink for carbon from July through October of 2014, and the C₃ grassland was a source throughout this period.

4.3.3 Leaf Area Index (LAI) and biomass

Leaf area index had a bi-modal pattern for both calendar years, with larger values that endured for longer periods observed in 2014. The pattern is consistent with seasonal trends - a spring growth peak and a summer or early autumn growth peak. Values of LAI were regularly larger at the C₃ grassland, with the exception of a short-lived peak at the C₄ prairie during the summer of 2013. MODIS values for the C₃ grassland agreed well with plot scale derived sampling during both years, while MODIS values for the C₄ prairie agreed better in 2014 (Figure 4.9).
Figure 4.9. Time series plot of leaf area index (LAI) for 2013 and 2014 calendar years. The grey (C3 Grassland) and black (C4 Prairie) solid lines are from MODIS observations (interpolated from 8-day intervals), representing a $3 \times 3$ km area centered at the eddy covariance tower for each study site. The grey (C3 Grassland) and black (C4 Prairie) dashed lines are plot level observations from destructive sampling at each grassland site.

Leaf area index was strongly related to biomass measurements from destructive sampling for both the C$_3$ grassland ($n = 26$, $r^2 = 0.7803$, $p < 0.001$) and the C$_4$ Prairie ($n = 26$, $r^2 = 0.7802$, $p < 0.001$). The slope of the linear regression between LAI and biomass was shallower for the C$_3$ grassland, indicating that there is generally a higher leaf area to biomass ratio for this ecosystem, particularly at higher LAI values (Figure 4.10a). There was also a positive linear correlation between the cumulative monthly GPP (absolute value) and the mean monthly LAI from destructive sampling for the C$_3$ grassland ($n = 11$, $r^2 = 0.692$, $p < 0.001$) and the C$_4$ Prairie ($n = 10$, $r^2 = 0.6336$, $p < 0.001$) (Figure 4.10b). Comparison of the slopes of the regression lines indicate that the C$_4$ prairie site had higher monthly GPP for the same values of mean LAI.
Figure 4.10. Regression for the C₃ Grassland (grey circles) and the C₄ Prairie (black diamonds of a) biomass as a function of leaf area index (LAI) for all destructive sampling collections and b) monthly GPP (absolute value) as a function of maximum monthly LAI (from destructive sampling), during the growing season months (March – October) in each year.

Given that biomass and LAI are strongly related, analysis of grassland productivity will utilize either of these metrics when comparing the two grasslands.
4.3.4 GPP, ET and WUE

Time series plots of the evolution of monthly $WUE$ during the two growing seasons and between the two grassland sites shows that the C$_4$ prairie was consistently more water efficient as compared to the C$_3$ grassland (Figure 4.11).

(Figure 4.11. Time series of monthly $WUE$ between the two grassland sites for the growing season during 2013 and 2014.

From March to June of 2013, $WUE$ values were similar between grasslands when precipitation and $\theta_{0-15cm}$ values indicated that sufficient water was available to both ecosystems. However, the C$_4$ prairie began to have higher $WUE$ values during the dry
summer and early autumn, until the extreme precipitation events in September, after which the C₃ grassland’s WUE exceeded the C₄ prairie. In contrast, the C₄ prairie had higher WUE values as compared to the C₃ grassland for the entire 2014 growing season months, with the greatest distinction between the grasslands occurring during the summer and autumn. This can also be seen by examining the relationship between ET and cumulative monthly GPP (absolute value), which indicates that the C₃ grassland generally loses more water for similar levels of productivity as the C₄ prairie (Figure 4.12).

![Graph showing the relationship between ET and cumulative monthly GPP for C₃ Grassland and C₄ Prairie.](image)

Figure 4.12. Monthly ET as a function of cumulative monthly GPP.

The relationship between water loss and monthly GPP was strongly correlated at the C₄ prairie site (n = 16, \( r^2 = 0.9214, p < 0.0001 \)), however this correlation was weaker by comparison at the C₃ grassland (n = 16, \( r^2 = 0.5219, p < 0.0001 \)).
4.3.5 Temperature sensitivity of Rs

Soil temperature was a strong predictor of Reco during the growing season for both grasslands during 2013 (C₃ grassland, n = 235, \( r^2 = 0.6463, p < 0.001 \); C₄ prairie, n = 150, \( r^2 = 0.7137, p < 0.001 \)), and for the C₄ prairie during 2014 (n = 225, \( r^2 = 0.7332, p < 0.001 \)). Abundant soil water seemed to confound the relationship in the C₃ grassland during the growing season in 2014 (n = 238, \( r^2 = 0.2872, p < 0.001 \)). Conversely, when \( \theta \) was less than 10\%, there was a weak, negative relationship between Reco and \( T_s \) in both grasslands during 2013, indicating that Reco is insensitive to \( T_s \) at low water content during the 2013 growing season (Figure 4.13).
Figure 4.13. Growing season (March 1 – October 31) mean nighttime NEE as a function of mean nighttime soil temperature ($T_s$) for the C$_3$ Grassland during 2013 (upper left) and 2014 (lower left), and the C$_4$ Prairie during 2013 (upper right) and 2014 (lower right). Each regression is divided into two groups – dark grey squares indicate values for days when the soil water content ($\theta$) was greater than or equal to 10% with the coefficient of determination ($r^2$) above and left of the relationship; black dots indicate values for soil water content ($\theta$) less than 10% with the $r^2$ below and right of the relationship.

There were few values of $\theta$ less than 10% for the C$_3$ grassland during 2014, so the relationship is unclear. Interestingly, even in dry soils, the relationship between $Reco$ and $T_s$ continued to be a fairly strong, positive one in the C$_4$ prairie during 2014. See APPENDIX 1 for regressions of $T_s$ and $\theta$ for each year at both grasslands.

4.3.6 Spring water and CO$_2$ exchange

The peak in CO$_2$ uptake occurred on May 20, 2014 for the C$_3$ Grassland and was 26 days later for the C$_4$ Prairie, on June 15, 2014 (Figure 4.14).
Figure 4.14. Time series plots from April 1 – June 30, 2014 of daily NEE (grey bars), the 1-week running mean (black lines), and $\theta_{0,15 \text{ cm}}$ (dark blue dashed line) for a) the C$_3$ grassland and b) the C$_4$ prairie. Positive values of NEE represent a net flux of carbon from the grassland to the atmosphere. Large grey arrows at the top of each plot show the direction (up = source, down = sink) and labels give the approximate magnitude of the accumulated flux of carbon over the entire period. Vertical dashed grey lines mark the peak carbon uptake for the C3 Grassland on May 20 and the C4 Prairie on June 15, 26 days later.

Although the net uptake was comparable between the grasslands during the spring of 2014, it was at a fairly consistent rate for the C$_3$ grassland, while the C$_4$ prairie increased daily until the peak in mid-June. This peak occurred after a large fluctuation in $\theta$, and both grasslands began to switch back to a net source when $\theta$ declined substantially.

Both ecosystem respiration (Reco) and gross primary productivity (GPP) was essentially
equal between the two grasslands during this period, though the timing of peak productivity and uptake was skewed later for the C₄ prairie. Both grasslands increased in biomass throughout this period, though the C₄ prairie remained lower than the C₃ grassland at each sampling. Although the C₄ prairie generated less biomass during the same period as compared to the C₃ grassland, the C₄ prairie experienced less water lost through ET throughout this period, even as productivity increased in June (Figure 4.15).
Figure 4.15. Time series plots for spring 2014 comparison of a) cumulative $R_{eco}$ and $GPP$, b) daily evapotranspiration ($ET$), and c) mean biomass from destructive sampling.
4.3.7 Summer / Autumn water and CO₂ exchange

The summer of 2013 had markedly different CO₂ cycling responses between the two grasslands, and before and after the extreme precipitation event in September (Figure 4.16).

Figure 4.16. Time series plots from July 10 – October 31, 2013 of daily NEE (grey bars), the 1-week running mean (black lines), and θ₀–15 cm (dark blue dashed line) for a) the C₃ grassland and b) the C₄ prairie. Positive values of NEE represent a net flux of carbon from the grassland to the atmosphere. Large grey arrows at the top of each plot show the direction (up = source, down = sink) and labels give the approximate magnitude of the accumulated flux of carbon over two periods, before and after September 24 (vertical grey dashed line).
Soil water content was low, especially in the C₄ prairie, from July 10 up to September, yet the prairie was a weak net sink for carbon during this period, whereas the C₃ grassland was a continuous source, losing 125 g C m⁻². After September 24, the C₃ grassland switched from a source to a sink, gaining approximately 24 g C m⁻² by the end of October. In contrast, the C₄ prairie switched over to a weak source, losing 10 g C m⁻². During this time, soils remained saturated at the prairie for an extended period of time.

As in the spring of 2014, Reco was very similar between the two grasslands during this period, however accumulated GPP was larger in the C₄ prairie as compared to the C₃ grassland, despite the weak and intermittent net uptake throughout July and August. Evapotranspiration (ET) was similar between the grasslands until after the extreme precipitation event when the C₃ grassland lost more water through ET. The C₄ prairie gained more in biomass as compared to the C₃ grassland during the prairie growth peak in July and August. However, after the extreme precipitation event in September, the vegetation at the C₃ grassland had additional growth in late September and early October, whereas the C₄ prairie continued to senesce (Figure 4.17c).
Figure 4.17. Time series plots for Summer / Autumn 2013 comparison of a) cumulative $R_{eco}$ and GPP, b) daily evapotranspiration ($ET$), and c) mean biomass from destructive sampling.
In general, the summer and autumn of 2014 had similar productivity and carbon cycling patterns as in 2013. The C₃ grassland was a source of carbon throughout the late growing season period, whereas the C₄ prairie gained approximately 65 g C m⁻² (Figure 4.18).

Figure 4.18. Time series plots from July 10 – October 31, 2014 of daily NEE (grey bars), the 1-week running mean (black lines), and θ₀₋₁₅ cm (dark blue dashed line) for a) the C₃ grassland and b) the C₄ prairie. Positive values of NEE represent a net flux of carbon from the grassland to the atmosphere. Large grey arrows at the top of each plot show the direction (up = source, down = sink) and labels give the approximate magnitude of the accumulated flux of carbon over the entire period.
Figure 4.19. Time series plots for Summer / Autumn 2014 comparison of a) cumulative $R_{eco}$ and $GPP$, b) daily evapotranspiration ($ET$), and c) mean biomass from destructive sampling.
Substantial precipitation during the summer resulted in pulses of $\theta$ and high $ET$ at both sites, and an increase in biomass at the C$_4$ prairie during August as compared to the C3 grassland during this period (Figure 4.19c). Once again, $R_{eco}$ was similar between the grassland sites, but $GPP$ at the C$_4$ prairie (-268 g C m$^{-2}$) was more than twice that at the C$_3$ grassland (-121 g C m$^{-2}$). Biomass declined at both sites beginning in late September, as did $\theta$ and $ET$.

4.3.8 Belowground water and CO$_2$ fluxes

Soil [CO$_2$] profiles for four selected dates and times contrasts the belowground production and distribution of gases between the grassland sites during the spring and summer. During the beginning of May, soils were wet for both years, but much cooler in 2014 (Figure 4.20 a and c). The soil [CO$_2$] gradient was steep for both grasslands on May 10, 2013, with the C$_4$ prairie generating comparably higher concentrations at the 15 cm depth. In contrast, May 11, 2014 was much colder and while the C$_3$ grassland had a steep soil [CO$_2$] gradient from 5 to 15 cm, the gradient at the C$_4$ prairie was very shallow.
Figure 4.20. Profile plots of [CO$_2$] measured at 5 cm above the land surface (ambient air intake line located at the auto-sampler) and soil [CO$_2$] at three soil depths (5, 10 and 15 cm) for the C$_3$ Grassland and the C$_4$ Prairie on a) May 10, 2013 at 15:00 MST, b) July 17, 2013 at 10:00 MST, c) May 11, 2014 at 15:00 MST and d) July 9, 2014 at 10:00 MST. Soil water content ($\theta$) from 0-15 cm and $T_s$ at 10 cm for each site at the time of sampling is denoted at the top of the plot, below the legend for each grassland.
Soil water content contrasted sharply during the 2013 and 2014 summer months and resulted in dynamic [CO$_2$] responses at both grasslands (Figure 4.20 b and d). For example, July 17, 2013 was very dry, yet the C$_4$ Prairie developed a steep gradient, especially at the 10 – 15 cm soil depth. In contrast, the soil [CO$_2$] at the C$_3$ grassland remained shallow unless there was an increase in $\theta$, in which case the gradient was steep throughout the profile, as seen on July 9, 2014. Although the soil [CO$_2$] at the C$_4$ prairie was comparatively shallow at this specific time, the gradient was dynamic and exhibited diel fluctuations.

4.4 Discussion

Water and CO$_2$ cycling differed between the two grasslands seasonally and between the two years of the study. Though analysis of both spring periods was limited by missing data at the C$_4$ prairie site, based on available data, it appeared that the C$_3$ grassland was more productive as compared to the C$_4$ prairie with a strong net sink of carbon from the atmosphere to the ecosystem during this period. While the C$_3$ grassland had late season growth and subsequent carbon uptake in response to the extreme precipitation in September 2013, the C$_4$ prairie had stronger primary production during the summer months for both years. Therefore, the data generally support the first hypothesis for this chapter (H4) given that the C$_3$ grassland took up more carbon in the spring while the C$_4$ prairie was a net sink of CO$_2$ during the summer. However, the differences in carbon cycling between the two grasslands was less conclusive in the autumn months for these two years. Although the autumn of 2013 would seem to support the hypothesis that the C$_3$ grassland was productive at this time, the extreme precipitation
event in September was significant, therefore making it difficult to conclude that the outcomes were representative of the typical grassland responses during this season. In fact, the autumn of 2014 showed that the C₃ grassland did not switch back to a net sink, and instead the C₄ prairie remained a net sink well into the first part of November.

Although both grasslands were largely productive during the predominantly cool or warm air temperatures with which they are associated, the grasses within the C₃ grassland demonstrated strong NEE responses to water availability while NEE at the C₄ prairie was relatively temperature sensitive. For example, \( R_{eco} \) became less sensitive to \( T_s \) at the C₃ grassland site when there was an increase in \( \theta \), yet the C₄ prairie retained the temperature sensitivity of \( R_{eco} \) through a range of \( \theta \) values. Although soils were very dry in the summer of 2013, the C₄ prairie was still a weak net sink for carbon. In addition, abundant rainfall and the resulting changes in \( \theta \) in late July 2014 caused respiratory pulses in the C₄ prairie, and while the prairie responded with increased uptake later in August, the C₃ grassland was also able to take advantage of this increase in water availability and become a net sink briefly that summer. Furthermore, fluctuations in \( \theta \) after precipitation events were much greater in the C₄ prairie and the WUE exceeded that in the C₃ grassland during both summers. Therefore, the data conditionally support the second hypothesis for this chapter (H5) because adequate water availability was important in the spring for the C₃ grassland but was less important and, due to the better WUE, likely not a primary driver of productivity in the summer for the C₄ prairie.
4.4.1 Photosynthetic types, water availability, and light-use efficiency

In a study by Huxman et al. (2004), it was hypothesized that semiarid ecosystems with high production potential are most sensitive to variations in water availability. The production potential of both semiarid grasslands at Rocky Flats NWR was high – *Bromus inermis* has been planted widely across temperate North America because it is very productive and sod forming, and *Andropogon gerardii* is a native to the tallgrass prairies found in eastern, mesic grasslands, with the potential to grow much taller than it does in Colorado. However, maximum rates of photosynthesis differ between C$_3$ and C$_4$ plants even when water was not limiting, and this is because water use and CO$_2$ pathways during photosynthesis differ physiologically.

CO$_2$ diffuses into the leaf through stomata where it is absorbed in the leaf water and then, in cool season C$_3$ plants, it is initially fixed by the enzyme, rubisco. Since rubisco has a temperature and concentration affinity for O$_2$, and CO$_2$ solubility decreases with increasing temperature, photorespiration increases with increasing temperature. Photorespiration releases both CO$_2$ and NH$_3$, and C$_3$ plants utilize a lot of rubisco, which is a N-rich enzyme. Leaf nitrogen content is relatively high in cool season plant species compared to warm season plants, mostly due to the higher rates of use of rubisco. Therefore, the C$_3$ grassland site, with cool season species, should have low rates of CO$_2$ exchange during hot days because although the grasses may be productive at higher temperatures, photorespiration is high due to inefficient photosynthesis (Gowik and Westhoff, 2011; Leakey et al., 2009; von Fischer et al., 2008).

In contrast, the warm season grasses in the C$_4$ prairie do not utilize the enzyme, rubisco, as the first contact with CO$_2$ after leaf diffusion and so less water is lost trying to
achieve the same photosynthetic rate. Warm season plants are more nitrogen efficient and have lower leaf nitrogen than cool season plants, and therefore should have lower soil nutrient requirements. However, more energy is required to fix CO$_2$ through the C$_4$ photosynthetic pathway, and therefore the C$_4$ grassland site, with warm season species, are generally more efficient at CO$_2$ exchange and have high water use efficiencies during higher temperatures (Ehleringer and Cerling, 2002; Gowik and Westhoff, 2011; Ode and Tieszen, 1980).

In addition to having dissimilar water use strategies, the two photosynthetic types have different light-use efficiencies as a result of the energy that is needed to fix carbon through their distinctive pathways. Light-use efficiency is defined as the amount of carbon gained per photon absorbed by the leaf. Physiologically, the C$_3$ pathway utilizes less energy (in the form of ATP) to fix carbon as compared to the C$_4$ pathway, and therefore C$_3$ plants should have the higher light-use efficiency. However, observations do not support this conclusion. Because the rate of photosynthesis in C$_3$ plants is reduced by photorespiration, and photorespiration increases with increasing temperatures, light-use efficiencies tend to be lower than those in C$_4$ plants as air temperatures increase. In the semiarid climate at Rocky Flats NWR, higher air temperatures are often associated with higher solar radiation during the summer, and thus, light-use efficiencies were likely lower at the C$_3$ grassland as compared to the C$_4$ prairie, further supporting the stronger carbon uptake by the prairie during the summer months.
4.4.2 Grassland structural differences

There are structural differences between the two grasslands that may influence how CO\textsubscript{2} is exchanged between the land surface and the atmosphere. The dominant grass species in the C\textsubscript{3} grassland site is smooth brome (*Bromus inermis* Leyss), which has relatively wide leaf blades for a grass species and a tall profile, the plants begin to grow early in the year and remain fairly large throughout their life cycle, and the plants grow closely together increasing the foliar and canopy cover. In addition, the large shape and size of the leaves cause an extensive and deep litter layer over the soil. Taken together, this canopy could act to disrupt the exchange of gases between the soil surface and the atmosphere, as well as inhibit evaporation and retain soil water.

The microclimate is distinctly different at the C\textsubscript{4} prairie site, where vegetation is spaced further apart, the dominant grasses (*Andropogon gerardii* and *Panicum virgatum*) have a more narrow leaf blade, begin growing later in the year and for a shorter period of the growing season, resulting in an open canopy and less litter covering the soil as compared to the C\textsubscript{3} grassland. Soil texture is coarse and poorly sorted at both grasslands, however the C\textsubscript{4} prairie soils are more heterogeneous and the depth to coarse fragments is shallow compared to the C\textsubscript{3} grassland. All of these characteristics affect the soils water holding capacity, the rate of evaporation from the soil surface, and gas exchange between the land surface and the atmosphere (Raich et al., 2000).

While the soils at the C\textsubscript{4} prairie were on average drier than soils in the C\textsubscript{3} grassland, there were several points during the study when significant precipitation resulted in sustained saturation at the prairie site. A ditch located to the south of the study
site frequently floods onto the areas surrounding this grassland and can be another factor that affects water availability during years with above average precipitation.

Given that \( \theta \) was higher at the \( C_3 \) grassland, it is possible that there was an increase in thermal diffusivity in the soils (Ochsner et al., 2001), reinforcing spatial homogeneity in aboveground productivity during the growing season.

4.4.3 Belowground responses

Analysis of soil [CO\(_2\)] profiles between the grassland sites highlights the seasonal differences in aboveground activity, as well as possible gas transport differences due to the depth of respiratory responses and/or soil texture. Water fluctuations in the soil stimulate the microbial community and generate heterotrophic CO\(_2\) production. Soil water content fluctuations were more prominent at the \( C_4 \) prairie site and produced steep gradients in soil [CO\(_2\)] as compared to the \( C_3 \) grassland, however these response were limited to warmer temperatures. During the wet and cold spring of 2014, the belowground response in CO\(_2\) production was weak in the \( C_4 \) prairie, while the \( C_3 \) grassland had high soil [CO\(_2\)] values below 10 cm depth. Dryer soils permit gasses to diffuse to the land surface unimpeded and also allow shallow soil layers to be influenced by turbulent fluxes at the surface, and so the shallow soil [CO\(_2\)] at the \( C_4 \) prairie had a diel pattern, whereas increases in soil [CO\(_2\)] endured longer at the \( C_3 \) grassland.

4.4.4 Conclusion

Grasslands primarily composed of one photosynthetic type have distinct seasonal responses to water availability at Rocky Flats NWR. While abundant precipitation is
beneficial to most semiarid grasslands, the timing of precipitation is key to eliciting the maximum grassland productivity and resulting carbon uptake between a cool season (C₃) and a warm season (C₄) dominated system. The relationships between LAI, GPP and biomass confirmed that the grassland ecosystems employed different leaf level strategies for carbon uptake and water use, as was expected for the photosynthetic type of the vegetation. The warm season species that dominated the C₄ prairie were productive during periods when water was limited, as long as thermal energy was sufficient, and continued to have high water use efficiency even during periods of water abundance. Interestingly, canopy structures that contribute to ecosystem water conservation (i.e., thick litter layer, dense vegetation) should favor the C₃ grassland, yet higher water requirements as compared to the C₄ prairie appeared to offset this advantage.

This study showed that the smooth brome-dominated C₃ grassland was dependent on water availability early in the year, but grassland productivity also increased throughout the growing season in response to additional water availability. The tallgrass C₄ prairie ecosystem was still productive during more water-limited conditions, and when water did become available, the ecosystem was limited to higher air and soil temperature ranges to take advantage of the resource and increase grassland productivity.
CHAPTER 5. CONCLUSION

The objectives of this research were to 1) determine how the timing and intensity of precipitation affect water and carbon cycling in semiarid grasslands, 2) analyze the impact from an extreme precipitation event, and 3) compare water and carbon cycling between neighboring C₃ and C₄ dominated semiarid grasslands. The research was conducted during two consecutive years that were not water-limited on an annual basis, yet contrasted sharply on a seasonal basis. This allowed the analysis of distinct patterns of water cycling and the impacts to carbon uptake and loss in two types of grassland ecosystems. The use of plot scale soil moisture, soil temperature, and soil [CO₂] along with ecosystem scale eddy covariance methods assisted in evaluating the response of the entire plant community to changes in precipitation.

This study concluded that precipitation early in the growing season, specifically accumulated precipitation during April and May, made a substantial difference in generating a carbon sink in the semiarid, smooth brome-dominated C₃ grassland at Rocky Flats NWR. Although annual rainfall for 2013 was above average and the extreme precipitation event resulted in additional carbon uptake in the grassland during September and October, this was insufficient to overcome the carbon losses earlier in the season when water was more limited. In contrast, higher precipitation amounts during March - May resulted in substantially higher gross primary productivity (GPP) throughout 2014 as compared to the 2013.

Results from the study of the impacts from the extreme precipitation events of September 2013 showed that the fully saturated soil conditions affected both the production and diffusivity CO₂, and that soil respiration (Rs) became insensitive to soil
temperature \((T_s)\). This was demonstrated by a lack of hysteresis between soil \([\text{CO}_2]\) and \(T_s\) and the weak relationships between both soil \([\text{CO}_2]\) and \(T_s\) and volumetric water content \((\theta)\) and \(T_s\) during this week. Consequently, empirical model estimates of respiration based on \(T_s\), in agreement during the rest of the growing season, failed to agree with observations during the extreme precipitation event. However, the process-based model estimates (DAMM) and soil flux gradient \(R_s\), better characterized the diminished fluxes during periods with the longest rainfall duration and highest saturated soil conditions.

The final chapter found that water and carbon cycling differed considerably between the two grasslands of differing photosynthetic types, both seasonally and between the two years of the research. The smooth brome-dominated \(C_3\) grassland was more productive during the spring as compared to the tallgrass \(C_4\) prairie, which had stronger primary production during the summer months for both years. Differences in carbon cycling between the two grasslands was less conclusive in the autumn months for the two years of observations. While the \(C_3\) grassland had late season growth and subsequent carbon uptake in response to the extreme precipitation in September 2013, the water inputs were significant and atypical, making it difficult to conclude that the outcomes were representative of this grassland’s response. Moreover, the \(C_3\) grassland did not switch back to a net sink in the autumn of 2014, while the \(C_4\) prairie remained a net sink during this time.

Finally, the grasses within the \(C_3\) grassland demonstrated strong responses to water availability while the vegetation at the \(C_4\) prairie was more temperature driven. Ecosystem respiration \((R_{eco})\) became less sensitive to \(T_s\) at the \(C_3\) grassland site when
soils were saturated, yet the C₄ prairie retained the temperature sensitivity of $R_{eco}$ through a range of $θ$ values, and was still a weak net sink for carbon with very dry soils. Fluctuations in $θ$ were much greater in the C₄ prairie, however water use efficiency ($WUE$) was better as compared to the C₃ grassland, particularly during the summer months, and therefore $θ$ fluctuations did little to dampen carbon assimilation at the C₄ prairie.

5.1.1 Ecosystem carbon cycling and Federal land management

Federal land management includes developing carbon sequestration strategies, and the remnant tallgrass prairie in the southwest section of Rocky Flats NWR is a rare vegetation community, the preservation of which is desired by federal and state government agencies (USFWS 2005). This research adds insights into the impact that episodic changes in water availability have on the fluxes of water and CO₂ between C₃ and C₄ dominated semiarid grasslands along the Colorado Front Range. This study also informs land managers about the productivity responses to rain events and how the timing of precipitation may impact the long-term resiliency of this ecosystem.

The Natural Resource Conservation Service/U.S. Department of Agriculture (NRCS/USDA) has adapted plans once suitable for Dust Bowl Era concerns to be in line with more recent issues of agricultural runoff, soil degradation, and climate change. The Conservation Reserve Program (CRP) provides incentives for farmers and ranchers in the U.S. Great Plains to allow cultivated or grazed fields to be abandoned and reseeded, eventually reclaimed ecosystems with native grassland vegetation communities (Burke et al. 1995). My research will be among the few that provide observations in a reclaimed
5.1.2 Validation of plant functional groups in ecosystem models

The comparison of C\textsubscript{3} and C\textsubscript{4} grasses in this study can help determine the role that each of these types of grasses plays in cycling water and CO\textsubscript{2} in the soil-plant-atmosphere continuum. Ecosystem models generally have very broad categories of vegetation that can over or under-estimate CO\textsubscript{2} fluxes based on the manner in which these groups are parameterized to use water and nutrients in the model, on the timing and amount of growing season productivity, and the leaf level stomatal conductance that regulates the fluxes of water between the soil and atmosphere. Real world observations provide validation data for model evaluation and future development.

5.1.3 Soil gas well design

Analysis of soil efflux has only recently begun to include continuous belowground measurements. Relatively few studies exist that use soil CO\textsubscript{2} concentrations and fluxes at different depths in the soil profile, however among these few, many techniques and designs have been utilized (Jassal and Black 2006, Riveros-Iregui et al. 2007, Pacific et al. 2008, Riveros-Iregui et al. 2008, Vargas et al. 2010, Vargas et al. 2012). The gas wells developed for this study are a unique design and represent an original approach to measuring soil gasses. The wells have a small vertical footprint but encompass a larger horizontal area that allows soil CO\textsubscript{2} concentrations to be integrated across the length of the well at the depth of interest, capturing soil spatial heterogeneity.
5.1.4 Broader implications

Water-limited ecosystems have developed a unique ability to respond to episodic precipitation events however, the timing of water availability is still a significant determinant of this resilience in some semiarid grassland ecosystems (Bengtsson, 2002; Potts et al., 2006). While significant disturbances impact water and carbon cycling in these terrestrial ecosystems, shifting precipitation patterns may cause similar impacts that are more difficult to predict. For many grassland ecosystems within the US Great Plains, the change in seasonal precipitation has implications for feedback mechanisms to climate change, may influence the annual sink or source of carbon, and could affect plant species composition shifts.
REFERENCES


APPENDIX 1. SOIL WATER CONTENT AND SOIL TEMPERATURE REGRESSIONS

Smooth brome-dominated, mixed (C₃) grassland

Figure A - 1. Relationship between mean daily (24-hour) soil water content (θ) and Ts at the smooth brome-dominated (C₃) grassland for the growing season (March 1 – October 31) during a) 2013 and b) 2014. Note that values during September 10-20, 2013 were discarded due to the extreme precipitation event which caused saturated conditions that skewed the relationship for that season.

Remnant tallgrass (C₄) prairie

Figure A - 2. Relationship between mean daily (24-hour) soil water content (θ) and Ts at the remnant tallgrass (C₄) prairie for the growing season (March 1 – October 31) during a) 2013 and b) 2014. Selected days were discarded due to water pooling at the surface.
and saturated conditions that skewed the relationship - from September 10 through the end of the 2013 growing season, May 22 – June 4, 2014, and July 31 – August 4, 2014.

Figure A - 3. Hourly soil moisture and $T_s$ relationship during September 9-15, 2013.
APPENDIX 2. ENERGY BALANCE FOR THE EDDY COVARIANCE METHOD

Energy balance closure for eddy covariance measurement taken at the two grassland study sites in Rocky Flats NWR during 2013 and 2014 was analyzed using only non-gapfilled data that passed quality control criteria for $H$ and $LE$. All soil heat flux measurements were corrected for heat storage in the soil layers above the heat flux plates using measured soil temperature and soil moisture and according to instrument instructions from Campbell Scientific Inc. (2012).

![Smooth brome-dominated, mixed (C_3) grassland](image)

Figure A - 4. Regression of turbulent fluxes and available energy at the smooth brome-dominated (C_3) grassland for a) 2013 and b) 2014.
Figure A - 5. Average hourly energy and turbulent fluxes for the smooth brome-dominated (C₃) grassland during 2013 and 2014.

Figure A - 6. Regression of turbulent fluxes and available energy at the remnant tallgrass (C₄) prairie for a) 2013 and b) 2014.
Figure A - 7. Average hourly energy and turbulent fluxes for the remnant tallgrass (C₄) prairie during the 2013 and 2014 growing seasons (March 1 – October 31).
APPENDIX 3. NIGHTTIME NEE REGRESSIONS WITH FRICTION VELOCITY

Figure A - 8. Nighttime NEE regressed with friction velocity ($u^*$) from the smooth brome-dominated ($C_3$) grassland during selected nighttime periods in a) 2013 and b) 2014.
Figure A - 9. Nighttime NEE regressed with friction velocity (u*) from the remnant tallgrass (C₄) prairie during selected nighttime periods in a) 2013 and b) 2014.
### APPENDIX 4. SOIL TEXTURE, POROSITY, AND ORGANIC MATTER CONTENT

Soil Organic Carbon for C3 Grassland - Loss on Ignition Method and texture analysis

<table>
<thead>
<tr>
<th>Sample</th>
<th>Crucible mass (g)</th>
<th>Dry soil mass +cruc (g)</th>
<th>Dry soil mass (g)</th>
<th>After + cruc mass (g)</th>
<th>Remain mass (g)</th>
<th>Mass loss (g)</th>
<th>SOM %</th>
<th>SOC %</th>
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<td>39.77</td>
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<th>&lt;500 um (g)</th>
<th>2mm &gt; x &gt; 500 um (g)</th>
<th>&gt; 2 mm (g)</th>
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<th>Clay %</th>
<th>Silt %</th>
<th>Sand %</th>
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<td>29.3%</td>
<td>54.0%</td>
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</table>

Means: 16.19% 33.38% 50.42%
## Soil Porosity

### C3 Grassland

Volume: 380.00 cm³ or mL

<table>
<thead>
<tr>
<th>Sample</th>
<th>Dry soil (g)</th>
<th>Water (mL)</th>
<th>Porosity (%)</th>
<th>Bulk Density g/cm³</th>
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</thead>
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<tr>
<td>1</td>
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<td>517.91</td>
<td>185.00</td>
<td>48.68%</td>
<td>1.363</td>
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<tr>
<td>6</td>
<td>526.36</td>
<td>190.00</td>
<td>50.00%</td>
<td>1.385</td>
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</table>

Mean = 50.7%  StDev = 2.46%  CV = 4.9%

### C4 Prairie

Volume: 380.00 cm³ or mL

<table>
<thead>
<tr>
<th>Sample</th>
<th>Dry soil (g)</th>
<th>Water (mL)</th>
<th>Porosity (%)</th>
<th>Bulk Density g/cm³</th>
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</thead>
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<td>552.93</td>
<td>195.00</td>
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</table>

Mean = 47.1%  StDev = 2.69%  CV = 5.7%
APPENDIX 5. SOIL GAS WELLS AND AUTOSAMPLER SYSTEM

To measure belowground CO$_2$ concentration (soil [CO$_2$]) changes, six soil gas wells were divided between two plots and collocated with the soil moisture and temperature sensors at each grassland study site in Rocky Flats NWR. Soil [CO$_2$] measurements were replicated at the 5, 10, and 15 cm soil depths. The soil gas wells developed for this study had a small diameter and 60-cm length, resulting in a horizontal footprint that allows measurements to be integrated across the span of the well in an effort to capture spatial heterogeneity at each depth (Figure A - 10).

![Figure A - 10. U.S. Geological Survey designed/constructed soil gas well.](image)

Each soil gas well had an approximate volume of 120 cm$^3$ and was constructed from 1/2” diameter schedule 40 polyvinyl chloride (PVC) tubing (1.58 cm i.d.) cut to 0.6-m length and sealed on both ends with 1/2” diameter PVC end caps. There were 24 evenly spaced holes (approx. 1 cm dia.) drilled along the length of the PVC tubing and positioned to rest along the bottom of the gas well. Each tube was wrapped in fiberglass screen to exclude large debris while allowing the flow of gasses once installed in the soil. Each end cap had a hole drilled in the center where a 1/2” diameter elbow connector (Swagelok, 1.27 cm i.d.) was inserted. Extending upward from the connectors was
stainless steel tubing (Swagelok, 1.27 cm o.d.) of varying lengths, with another connector attached to facilitate assembly with the analyzer system at the surface.

Laboratory and field tests of the soil gas wells and gas analyzer system were conducted during 2012 and 2013. Each laboratory test was performed using a prototype gas well, fully assembled, buried in a large container of uniform sand with Bev-a-line tubing attached at both ends, extending outside of the container at the length calculated for field deployment, and attached to a gas analyzer. The sand container used for testing was further equipped with a long, large diameter tube with many holes drilled long the length, winding around the bottom of the container and covered in sand, and connected to gas canisters with known CO\textsubscript{2} concentrations. The sand container was sealed to make it as air tight as possible, and a slight positive pressure was created inside of the sand container by adjusting the flow from the gas canisters to ensure that ambient air did not enter the container during testing. A range of CO\textsubscript{2} gas concentrations were used to fill the sand container on different days and trials of gas well sampling strategies and flow rates were tested. It was determined that a flow rate of 400-500 mL was optimal for measuring the representative concentration inside of the gas well, and a peak measurement during the initial 30 – 60 seconds produced the best value. Gas well prototypes were buried at 5 and 10 cm depths at Rocky Flats during late August, early September 2012 and a prototype sampling system was attached and tested. Sampling strategies, measurement frequency and duration, and soil CO\textsubscript{2} concentration ranges were determined in order to refine the system for deployment.

Once fully tested, the soil gas wells were buried in plots at the grassland field sites. The soil gas wells were installed in the soil horizontally. Care was used to ensure a
consistent depth was excavated and soil was carefully removed and replaced with intact pieces, when possible, to preserve the original soil structure and horizons. This was particularly challenging as the soils contained angular, poorly sorted rocks in the soil matrix. Therefore, it was necessary to rearrange or remove some rocks in order to eliminate preferential flow paths for rainwater and/or voids that could potentially connect the gas well to the surface, compromising the concentration measurements. All wells were extensively tested in the labs at the USGS in Denver, a subset was installed and tested at the site during September 2012, and final installation took place during December 2012 and January 2013.

Figure A - 11. Images of soil gas well auto-sampler at the smooth brome-dominated C₃ mixed grassland site (left) and at the remnant tallgrass C₄ prairie (right) in Rocky Flats NWR.
Hourly, belowground measurements were used to determine the carbon flux by measuring the time rate of change in soil CO$_2$ concentration with depth. This was accomplished by pumping the air from each soil gas well and circulating it through a closed system that was connected to an infrared gas analyzer located at the land surface. The “auto-sampler” system at the surface utilized a LI-800 Infrared Gas Analyzer (IRGA, LI-COR Biosciences) attached to a multiplexer, a 16-channel AC/DC controller (SDM-CD16AC) and controlled by a CR-10X data logger (Campbell Scientific, Inc., Logan Utah). In order to improve the resolution, the IRGA was set up to measure CO$_2$ concentrations from 0-5000 ppm, a trade-off that was decided on during the field test in 2012 when even very moist soil conditions rarely resulted in concentrations above 3500 ppm. Gas wells were sampled through polyethylene lined plastic tubing (Bev-a-Line) that ran from each well along the ground inside a conduit and then into a centrally located weather resistant container, where the multiplexer controlled solenoids to select the line to be sampled by the analyzer. A pump and flow meter were attached to the outflow line from the analyzer to create positive pressure and a flow rate of approximately 400 mL min$^{-1}$, and gas samples were filtered on the intake before entering the analyzer (microfiber filter, 0.1 L internal volume, Balston, DFU – Parker Hannifin Corporation). All soil gas wells and the auto-sampler system were designed, constructed, and tested at the U.S. Geological Survey (USGS) in Denver, CO.

At the start of each hour, the auto-sampler used an 8-minute cycle to sample and record each of the six soil gas wells and the ambient CO$_2$ at the surface. During the first minute and 30 seconds, the pump was turned on and air was drawn into the gas analyzer from an ambient intake line located at 5 cm above the soil surface, attached to the outside
of the auto-sampler container. This allowed the system to clear lines and to establish the ambient CO$_2$ concentration above the soil surface and in the grass canopy. After the stabilization phase, each minute alternated between sampling a gas well for 30 seconds and then the ambient intake line for 30 seconds, with CO$_2$ concentration values being recorded every five seconds. The final 30 seconds of the 8-minute cycle sampled from the ambient intake line again to stabilize the analyzer. The peak CO$_2$ concentration for each soil gas well recorded during the hourly sampling cycle was isolated as the single representative value. To determine a single ambient CO$_2$ concentration for each hour, the last recorded value for each 30-second ambient intake line sample were selected and these values were averaged. After numerous laboratory trials and field tests, it was determined that the representative gas well concentrations and ambient values were best captured using this procedure and a flow rate between 400-450 mL min$^{-1}$. In addition, the gas analyzer was equipped with a zero calibration and the system was also intermittently calibrated using a canister of CO$_2$ reference gas during field visits.
APPENDIX 6. HOURLY SOIL FLUX-GRADIENT RESPIRATION AND $T_s$ REGRRESSIONS DURING THE 2013 FLOOD

VWC increased from 24 to 37% by midnight
VWC > 40%, saturated all day