BELOWGROUND BUD BANK DYNAMICS OF NATIVE, PERENNIAL GRASSES AND INTERACTIONS WITH

FIRE IN THE NORTHERN GREAT PLAINS

A Dissertation
Submitted to the Graduate Faculty
of the
North Dakota State University
of Agriculture and Applied Science

By

Morgan Lee Russell

In Partial Fulfillment of the Requirements
for the Degree of
DOCTOR OF PHILOSOPHY

Major Program:
Range Science

November 2013

Fargo, North Dakota

North Dakota State University
Graduate School

Title

Belowground bud bank dynamics of native, perennial grasses

and interactions with fire in the northern Great Plains

By

Morgan Lee Russell

The Supervisory Committee certifies that this ***disquisition*** complies with North

Dakota State University's regulations and meets the accepted standards for the

degree of

DOCTOR OF PHILOSOPHY

SUPERVISORY COMMITTEE:

Dr. Kevin Sedivec

Chair

Dr. Amy Ganguli

Co-Chair

Dr. Lance Vermeire

Dr. John Hendrickson

Dr. Steve Travers

Approved:

November 1, 2013	Dr. Francis Casey
Date	Department Chair

ABSTRACT

Mixed-grass prairies of the northern Great Plains evolved following frequent disturbances such as drought, fire, and extensive grazing by ungulates. As a result of periodic natural disturbances, native grass reproductive mechanisms have adapted to withstand frequent disturbance. Ninety-nine percent of native perennial grasses reproduction occurs through well-protected and densely structured axillary buds. Since vegetative buds are the primary driver of aboveground growth, bud response to differing seasons and fire return intervals may reveal strategies to enhance bud dynamics. The timing of fire may transition dormant buds into active buds elucidating the mechanism responsible for aboveground growth following fire. My objectives were to: 1) reveal belowground bud characteristics of *Bouteloua gracilis* (Willd *ex*. Kunth) Lag. *ex* Griffiths) (C_4 species)*, Pascopyrum smithii* (Rydb.) A. Löve) (C_3 species), and *Hesperostipa comata* (Trin. & Rupr.) Barkworth) (C_3 species), 2) examine immediate fire effects on belowground bud activity, dormancy, and mortality following seasonal fire and fire return interval treatments, 3) quantify short-term belowground bud dormancy, activity, and mortality trajectories following fire, and 4) determine the effects of soil moisture and soil temperature on bud growth and maintenance. Belowground bud characteristics differed among species, where *B. gracilis* produced the most buds (6 ± 4 buds tiller^{-1}), *P. smithii* produced an intermediate amount (4 ± 2 buds tiller^{-1}), and *H. comata* contained the least amount of buds (3 ± 3 buds tiller^{-1}). Fire treatments did not result in immediate, direct mortality of *B. gracilis*, *P. smithii*, or *H. comata* buds. However, fire treatments enhanced bud activity and reduced dormancy for *B. gracilis* and *P. smithii*. Season of fire and fire return interval directly manipulated bud activity, dormancy, and mortality for these species throughout two or more growing and dormant seasons following fire. Soil moisture was found to be a good predictor for bud growth of cool-season species and soil temperature was a reliable predictor for warm-season species bud growth. Prescribed burning can be used to manipulate bud bank dynamics as a management tool and belowground bud dynamics can also be used to improve post-fire management strategies in wildfire situations.

DEDICATION

I dedicate this work to two of the very best Wildland Firefighters and Range Specialists, my brother and dad, Jake and Randy Russell.

PREFACE

Chapters 2, 3, and 4 of this dissertation were written as manuscripts that will be submitted to peer-reviewed journals. Chapter 2, "Season of fire manipulates bud bank dynamics in mixed-grass prairie," will be submitted to *Journal of Ecology*. Chapter 3, "The role of fire return interval, season of fire, and drought on bud bank dynamics" will be submitted to *Oecologia*. Chapter 4, "Phenology of perennial native grass axillary buds in the northern mixed-grass prairie" will be submitted to *American Journal of* Botany. Each chapter follows the style and guidelines of *Rangeland Ecology and Management*.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1. FIRE EFFECTS ON BUD BANKS: BELOWGROUND MECHANISMS FOR ABOVEGROUND

GROWTH

Introduction

Mixed-grass prairies of the northern Great Plains evolved with frequent disturbances such as,

drought, fire, and grazing by ungulates (Wright and Bailey 1982). As a result of recurring disturbances,

native grasses have adapted their growth and reproductive mechanisms to withstand frequent

disturbance (Hartnett and Keeler 1995). For example, native perennial grasses that dominate grasslands

reproduce primarily through vegetative reproduction (Benson and Hartnett 2006). These vegetative buds

are well-protected against drought, fire, and grazing as a result of their growth forms (Wright and Bailey

1982; Ott and Hartnett 2012). Since vegetative reproduction is the primary driver of aboveground

growth, characterizing bud response following fire can improve the timing of recommended prescribed

burns and grassland assessment for post-fire grazing following wildfire.

Fire effects on plant communities are typically assessed from an aboveground perspective while

belowground aspects that contribute to vegetative reproduction are most often assumed. Since

grasslands are largely characterized by vegetative reproduction, a better understanding of belowground

reproductive dynamics of plant communities could improve understanding of plant community ecology.

Furthermore, we can more accurately assess fire effects in the northern Great Plains if we can

characterize how fire influences vegetative reproduction. With a better understanding of fire effects in

grasslands and how plant communities function via vegetative reproduction, we can equip managers with

predictions of plant community responses to fire, including grassland resistance mechanisms. An

improved assessment of the vegetative reproduction mechanisms that encompasses above and

belowground reproductive dynamics would promote more informed decisions and policies to be

generated following fire.

General belowground bud bank dynamics have been assessed in the tallgrass prairie in response

to frequency of fire (Benson et al. 2004; Dalgleish and Hartnett 2009); however, this type of bud bank

research is in its infancy in the northern Great Plains and is the first of its' kind to incorporate belowground bud production response to differing seasons of fire and fire return intervals. Mixed-grass prairie in the northern Great Plains evolved with frequent fire (5 -10 yrs) (Wright and Bailey 1982) and fire most likely has served as a fundamental aspect of perennial grasses evolutionary history.

Fire

Record-breaking drought and wildfires have occurred in recent history, especially in the western regions of the northern Great Plains. The combined effects of drought and wildfires span across different vegetation types prompting controversial views on fire. Coping with wildfires, while still recognizing fire's use as a tool, can be challenging for range managers, livestock producers, recreationists, wildlife-habitat managers, and the general public. However, prescribed burning can mimic natural fire conditions to prevent devastating wildfires while maintaining the structure and function in many terrestrial ecosystems. In fact, many ecosystems do not merely tolerate fire, but often require it as an ecological and evolutionary process (Denslow 1980; Wright and Bailey 1982; Axelrod 1985).

Fire history of the northern Great Plains

Use of prescribed burning in mixed-grass prairie of the western Great Plains has gradually increased because of interest in restoring historic disturbance regimes (Brockway et al. 2002). Although grazing and occurrence of periodic drought played a role in the evolutionary history of mixed-grass prairie, the development and persistence of grassland ecosystems have been closely linked to recurrent natural fires and fires ignited by humans (Higgins 1984; Axelrod 1985; McPherson 1995).

Native Americans throughout eastern Montana utilized fire to modify their environment and ignited fires for a variety of reasons, including hunting, habitat improvement, crop harvesting, pest reduction, warfare, and clearing areas for home sites, crops, and travel (Pyne 1982; Higgins 1984; Arno 1996). Prior to European settlement, fire served as a cultural and ecological disturbance mechanism for sustaining the structure, diversity, and productivity of prairie ecosystems (Wright and Bailey 1982). Numerous historical observations from fur traders, explorers and fort outposts relay the prevalence of

prairie fires (Bradbury 1819; Stansbury 1852; Perrine 1927). Compiled accounts of pre-settlement fires

suggest northern plains Native Americans did not pattern their use of fire according to lightening-fire

patterns and instead burned during all seasons of the year, favoring April and October (Higgins 1984).

From 1940 to 1975, 70% of lightening-ignited fires started in June, July, and August throughout mixed-

grass prairie and pine-savanna in the northern Great Plains (Higgins 1984). Accounts of fire history from

the late 19th century also show fire return intervals of 3-15 years, depending on vegetation types (Higgins

1984; Umbanhowar 1996). Therefore, based on natural and human ignited fires since European

settlement, northern mixed prairie endured frequent fire, which likely influenced the evolution of fire

resistance mechanisms in native, perennial grasses.

Season of fire

Timing of fire is a critical factor for individual plant and community response due to phenological

differences among species and physiological changes throughout the growing and dormant seasons

(Ewing and Engle 1988). Although the close association between fire and mixed-grass prairies is widely

recognized, uncertainty exists concerning the seasons, frequencies, and methods for effectively using fire

in northern Great Plains grasslands. Even though historical accounts suggest 70% of lightening-ignited

fires started in the summer months, little is known concerning seasonal effects of fire on plant

production, especially fires during the summer (Augustine and Milchunas 2009; Vermeire et al. 2011).

Wildfires typically occur during the summer when warm dry air converges with dry, readily

ignitable fuel (Wright and Bailey 1982). However, this natural summer cycle of wildfire in the northern

Great Plains has been replaced by a pattern of spring and fall prescribed fires when burning conditions

are cooler, less volatile, and less erratic. Conducting prescribed burns outside the natural summer

wildfire season has dominated most of the research throughout the northern Great Plains (Engle and

Bultsma 1984; Steuter et al. 1987; Biondini et al. 1989). Furthermore, summer fire research on C_3-

dominated rangelands is limited to post-wildfire data, which usually lacks adequate replication and does

not involve any pre-fire data (Engle and Bidwell 2001).

Summer prescribed burn research is gaining momentum and has been conducted in eastern Montana (Vermeire et al. 2011). For example, *P. smithii*, a native C_3 perennial, more than doubled biomass on burned sites the year following summer fire (Vermeire et al. 2011). In the same study, *P. smithii* standing crop was 47% greater for burned than non-burned sites following a summer fire with a wet spring (250 mm vs. 150 mm long-term average). Since *P. smithii* reproduces vegetatively, summer fire most likely promoted rapid rhizome development through dormant or pre-existing axillary buds. Increased production of C_3 perennial native grasses following fire is likely due to phenological and morphological adaptations, including rhizomatous growth form, rapid tillering, and tolerance to drought; all mechanisms enhancing *P. smithii* resilience and resistance to fire (Vermeire et al. 2011).

Despite the limited summer fire research in mixed-grass prairie, portions of the western Great Plains, replicated summer fire research has been conducted in tallgrass prairie (Ewing and Engle 1988; Howe 1995; Engle et al. 1998). One year following summer prescribed burns, tallgrass prairie production was similar to non-burned sites due to maintained tiller production and improved microclimate conditions (Ewing and Engle 1988). Season of fire effects also include increased litter removal and canopy openness with late summer fires compared to spring burns (Ewing and Engle 1988; Castellano and Ansley 2007). Summer fire effects in tallgrass prairie also have been shown to favor C_3 over C_4 perennial grasses with plant community shifts lasting for up to 3 years (Engle et al. 1998).

Spring and fall burns affect C_3 and C_4 species differently due to differences in optimal growing temperature range and growth period. As a result, shifts in species composition can occur due to season of burning. For example, aboveground production shifted toward C_4 species after a spring burn in C_3 dominated plant communities in northern mixed prairie (Steuter 1987). Spring prescribed burns are also popular in C_4-dominanted tallgrass prairie to reduce exotic C_3 grasses (Ewing and Engle 1988; Engle et al. 1998; Engle and Bidwell 2001). These spring burns damage cool-season invaders before warm-season dominant natives become active (Grilz and Romo 1994). Since bud transitions and tiller recruitment are closely linked with aboveground growth phenology (Ott and Hartnett 2012), changes in season of fire may alter belowground bud dynamics dictated by photosynthetic pathway.

Timing of burning may manipulate bud bank transitions and tiller recruitment. For example, spring burns reduced forage production by 66% and late-winter burns conducted prior to the initiation of plant growth reduced production by 33% during the first post-burn growing season on the short-grass steppe (Brockway et al. 2002). Additionally, late-winter burns conducted in moderately grazed short-grass steppe did not negatively affect forage production in subsequent growing seasons (Augustine and Milchunas 2009). Reductions or increases in aboveground production in short-grass prairie may illustrate altered tiller emergence from the bud bank. However, it is unknown whether tiller emergence from the bud bank influences production to a similar degree in mixed-grass prairie.

Fire return interval

Fire return interval depends on the rate of fuel replenishment by vegetation growth following a fire (Brown and Davis 1973; Trollope 1984; Williams et al. 1998); however, fire return intervals have been altered by many other factors (Pyne 1982). Mixed-grass prairie most likely burned every 5 to 10 years (Wright and Bailey 1982; Higgins 1984). Nonetheless, the introduction of intensive grazing by cattle and sheep, cultivation, and fragmentation during the late 19th century severely altered fire return intervals (Wright and Bailey 1982).

During the late 20th century, fire suppression programs were extremely successful due to simple, emotional messages that equated suppression with national defense (Pyne 1982). Fire suppression programs such as Smokey Bear in 1945 decreased the frequency and extent of fires (Pyne 2004). Fire suppression replaced medium sized, low-intensity fires with relatively large and intense fires that currently account for 98% of the fires in the western United States (Stocks 1991). The net effects of these changes have altered regimes (both culturally and ecologically) causing increased severity of wildfires due to prolonged fire return intervals (Pyne 1982, 2004; Umbanhowar 1996).

Interest in restoring historical processes, such as fire, raises controversial questions about historical versus current fire return intervals. Return intervals in fire-dependent ecosystems are largely determined by prevailing climatic, physiographic, and vegetation conditions (Brockway et al. 2002). As a

result, variation existing in both the timing and frequency of fires in the past and to what degree to apply modern-day prescribed fire (Engle and Bidwell 2001). Mimicking the interaction between fire, vegetation, and climate is extremely complex due to varying precipitation events and drought. Typically, big prairie fires occur during drought conditions preceded by 1-3 years of above average precipitation in the mixed-grass prairie (Wright and Bailey 1982).

Applying historical fire return intervals to current prescribed fire management is challenging because of wildland-urban interface areas and wildland fragmentation (Erichsen-Arychuk et al. 2002). Increased population and development have altered the ability of fire to operate on historical landscape scales. In addition, fire suppression has increased fuel loading past the point observed with historical fire return intervals (Niklasson and Granstrom 2000). For example, woody plant expansion into grasslands has altered previous low-intensity surface fires into crown fires that are extremely difficult to contain (Pyne 1982; Sheley and Bates 2008). Both fragmentation and decades of fire suppression further modify fire behavior, and exacerbate fire unpredictability and intensity, leaving plant communities adapted to short fire cycles with a longer return interval than in the past (Ryan et al. 2013).

On the other hand, utilizing historical fire return intervals can provide a point of reference for rangeland management and prescribed burning. First, historical fire return intervals relate to plant community structure and function providing ranges of variability in which managers can base future decisions. Second, information can be gleaned from the evolutionary history of specific geographic areas or ecosystems. It is important to know what a system is capable of withstanding in order to maximize plant community resistance and resilience mechanisms (Collins 1992; Engle and Bidwell 2001). Last, considering the underlying disturbance catalysts driving landscape patterns and processes provides a framework for current management direction and strategy.

Fire effects on aboveground growth

Fire reduces litter and in the process influences community composition/processes by altering soil temperature and moisture, microbial populations, nutrient cycling, ground layer light, and humidity

(Willems 1983; Stocks 1991; Knapp et al. 1998). In semiarid environments where water availability is low and decomposition rates are slow, fire reduces aboveground biomass and releases nutrients that were previously immobilized in accumulated organic matter (DeBano et al. 1998). The resulting faster rates of nutrient turnover are essential for sustaining the high primary productivity typical of mixed-grass prairie (Woodmansee and Wallach 1981).

In grassland systems, prescribed burning may control undesirable species that invade grasslands (e.g., woody species and annual grasses). Woody species, such as *Juniperus occidentalis* and *Juniperus virginiana*, are typically stunted by fire (Buehring et al. 1971; Weaver and Plaggemeyer 2004; Sheley and Bates 2008). In addition, plains prickly pear (*Opuntia polycantha*) experienced indirect mortality because of insect and browsing damage following fire (Vermeire and Roth 2011). Reductions of invasive annual species (e.g., *Bromus arvensis* and *Bromus tectorum*) can be achieved with fire through direct seed mortality (Vermeire and Rinella 2009).

Graminoid morphological adaptations can also play a critical role of grassland response following fire. Bunchgrasses can accumulate large amounts of dead plant material within the plant crown and, therefore, sustain high temperatures for long periods of time after the fire front has passed (Wright 1971). Small bunchgrasses containing only a few current-year tillers will usually survive a fast-moving fire, unless the fire occurs at a susceptible phenological stage (Wright and Bailey 1982). Rhizomatous grasses are typically resistant to grassland fires due to deeply buried axillary buds and lack of accumulated, dead plant material to sustain lethal temperatures (Wright and Bailey 1982).

Although fire has played a critical role in the development of vegetative defensive mechanisms, uncertainty in the vulnerability of recently burned areas has led to significant controversy. Wildfires on federally managed lands represent significant costs to producers who must defer grazing on federal grazing allotments following fire. A majority of federal lands managed by the U.S. Forest Service and Bureau of Land Management advise complete rest for 2 years following wildfire to ensure rangeland recovery. Producers are then faced with 3 options: 1) reduce their cattle numbers, 2) rent additional pasture, or 3) purchase harvested feedstuffs. Grazing deferment annually represents a $12.8 million loss

to Montana, Wyoming, North Dakota, and South Dakota producers based on rented pasture costs (NIFC 2013).

The current position of grazing management following fire represents a significant disconnect between grazing and fire management, understanding of grass growth, plant community dynamics, and the natural regime of fire, grazing, and drought under which the northern mixed-grass prairie evolved (Wright and Bailey 1982; Higgins 1984; Pyne 2004). Since vegetative reproduction is the primary driver of aboveground growth for perennial grasses in grasslands (Benson and Hartnett 2006), bud bank response could be better utilized in planning prescribed fire and assessing the aftermath of prescribed burns and wildfire. Incorporating bud bank response may not only align land management policies with reproductive dynamics, but could significantly decrease grazing deferment costs for regional producers in mixed-grass prairie systems.

Fire effects on belowground bud banks

Many grass species are protected against intense heat from fire by storing resources in belowground meristems that are well-protected and insulated by soil (Wright and Bailey 1982; Schimmel and Granstrom 1996). Belowground reserves of meristematic tissue, or axillary buds, allow for vegetative reproduction to replace older or dead tillers. This process of vegetative reproduction is self-reinforcing, where growth responses determine future plant community composition and the intensity and frequency of future fires (Pyne 1982).

Including bud response and bud bank knowledge with fire effects research will provide new insight into prescribed burning and perennial grass adaptation to fire. Below the soil surface, temperatures decrease sharply (Norton and McGarity 1965), allowing rhizomatous grasses to be resistant to fire because the axillary buds are buried below the soil surface (Wright and Bailey 1982). Therefore, temperatures of a grass fire may not have substantial direct effect on soil organic matter, microbial populations, or axillary buds (Norton and McGarity 1965). However, exposed vascular tissue is easily killed by heat. A temperature of only 60 °C for one minute is sufficient to coagulate proteins and cause

8

lethal damage to plant tissue (Wright and Bailey 1982). Lethal temperature can vary tremendously depending on plant moisture, maximum temperature, and duration of exposure. Indirect effects of fire on vegetative buds include the release of nutrients, such as nitrogen, to promote bud elongation and increased sunlight penetration (Wright and Bailey 1982). In addition to changes within the soil, season of burning may influence soils indirectly by the percentage of the soil surface burned and the depth of burn (e.g., litter removal) (Wright and Bailey 1982).

Year-to-year replacement of grass tillers primarily depends on the production and survival of axillary buds (Hendrickson and Briske 1997). Once grasses are forced into heat-or drought-induced quiescence their axillary buds cycle repeatedly between active and dormant stages before becoming fully committed (Shimizu-Sato and Mori 2001). Bud bank knowledge can equip producers and land managers with improved decision-making tools in post-fire management. For producers who have experienced wildfires, understanding short-and long-term fire effects on bud banks can sustain ranch and grass productivity by improving grazing management choices post-fire.

Vegetative reproduction

Introduction

Vegetative reproduction is the primary driver of aboveground production and growth, producing more than 99% of new tiller growth (Murphy and Briske 1992; Benson and Hartnett 2006). Therefore, the belowground bud bank is the primary source of recruitment for new tillers. By recruiting tillers from a bud bank, perennial grasses are able to respond rapidly to their environment and are highly resilient following grazing, drought, fire, or other stresses. Research has shown that bud banks play an important role in aboveground vegetation dynamics in grassland systems. For example, patterns of plant abundance and productivity associated with fire, grazing and climatic variability are mediated through the bud bank (Benson et al. 2004; Benson and Hartnett 2006; Dalgleish and Hartnett 2009). Furthermore, decreased bud bank densities lead to decreased aboveground productivity of perennial native grasses allowing invasions by exotic species to occur (Dalgleish and Hartnett 2009).

Despite the importance of vegetative reproduction in year-to-year replacement of tillers, little is known about the timing of tiller recruitment from the bud bank. Although the roles and dynamics of seed banks and tiller processes have been studied (Baskin and Baskin 1998), knowledge and understanding of bud banks is limited. In seed bank populations, the timing of seedling establishment can have a dramatic effect on the likelihood of seedling survival, with early-emerging cohorts strongly out-competing seedlings that emerge later in the season (Rogers and Hartnett 2001). However, it is unknown whether vegetative reproduction in northern mixed prairie follows a similar timing-dependent survivorship, whether bud bank recruitment occurs throughout the growing season, and how these characteristics are affected by season and frequency of disturbance.

Research on belowground reproductive dynamics of plant communities has been conducted in the tallgrass prairie. However, limited work has been conducted on bud banks in the northern Great Plains. An improved understanding of species response to season and frequency of fire may elucidate mechanisms altering population processes. Determining the crucial link between formation and maintenance of the belowground meristem sources and ecosystem-level processes reveals how species develop throughout an annual growth cycle (Dalgleish et al. 2008). More detailed characterization of bud banks is necessary in order to fully comprehend species response and plant community thresholds.

Morphological development

Tillers formed from vegetative reproduction are initiated from the axillary buds of older parent tillers (Briske 1991). Grass tillers are structural subunits consisting of a series of phytomers from a common apical meristem (Robson 1968; Langer 1979; Briske 1991). Each phytomer generally consists of the leaf blade and sheath, internode, node, and associated axillary bud (Briske 1991). The bud is the final structure of the phytomer formed, following the leaf and internode (Sharman 1942). Buds then accumulate belowground since grasses condense their nodes at the base of the tiller, only exposing leaves aboveground during vegetative growth (Robson 1968; Hyder 1972; Jewiss, 1972).

This belowground reserve of meristems associated with rhizomes or other perennating organs is considered a bud bank for future growth (Harper 1977), which plays a fundamental role in local plant population structure and dynamics. Persistence of perennial grasses is dependent on successful tiller production initiated by axillary buds, compensating for tiller mortality and providing the ability to overwinter (Jewiss 1972). Longevity of existing perennial grasses is contingent upon the consistent development and transition of axillary buds from the bud bank into new tillers.

Tiller replacement and maintenance of tiller populations in response to nutrient availability and disturbance have been thoroughly documented (Kays and Harper 1974; Caldwell et al. 1981; Olson and Richards 1988). Many grass tiller populations are consistently maintained in established populations, where tiller natality and mortality rates are relatively equivalent over the active growth period (Jonsdottir 1991). Grazing and fire can shift tiller dynamics by extending the period of tiller recruitment from the bud bank (Butler and Briske 1988). Typically, grass and sedge tillers live for 1 year (Bernard 1975; McKendrick et al. 1975), but may live for 3 or more years in some species (Jonsdottir 1991: Tamm et al. 2002).

Tiller emergence has traditionally been the accepted criterion for the presence of new growth when analyzing possible mechanisms governing the initiation and growth of wheat (Williams 1975). However, tiller emergence is a much more complex event since buds that initiated tiller growth have been growing for weeks prior to emergence, with a portion of buds never emerging (Williams 1975). Few studies have looked at bud dynamics and considered aspects such as bud phenology and bud fate prior to tiller emergence (Robson 1968; Butler and Briske 1988; Olson and Richards 1988).

Seed production vs. bud production

Although vegetative reproduction plays an integral role in grassland population dynamics, perennial grasses still significantly invest in sexual reproduction through flowering and seed production (Baskin and Baskin 1998; Benson and Hartnett 2006). However, tallgrass prairie research suggests less than 1% of new aboveground growth results from seed bank recruitment (Benson and Hartnett 2006).

11

Nevertheless, both modes of reproduction contribute to population persistence in different ways by influencing genetic diversity, spatial pattern, and plant population dynamics (Benson and Hartnett 2006).

Vegetative offspring have much greater survivorship and growth rates than seedlings (Benson and Hartnett 2006). Vegetatively derived tillers have a greater ability than seedlings to emerge through the low light condition of accumulated litter, resulting in greater competitive ability once they emerge (Baskin and Baskin 1998). This is most likely due to their developed root system, stored food reserves, shared resources, and the potential for physiological integration from parent tillers (Hartnett and Keeler 1995).

In comparison to bud bank dynamics, seed bank dynamics are well-studied. For example, few grass seeds persist in the soil beyond 5 years, with seeds of several species surviving less than 1 year (Baskin and Baskin 1998). Although viable seed populations ranging from a few hundred to >6000 seeds·m^{-2} may occur in tallgrass prairie (Rabinowitz 1981), successful seedling establishment is a rare event, by some estimates contributing to <1% of total aboveground tillers (Rogers and Hartnett 2001). However, a seed bank can enhance the persistence of many sub-dominant or rare satellite species and allow rapid occupation of newly available gaps, such as soil disturbances (Reader and Buck 1991). A reserve of dormant seeds in the soil can stabilize plant population dynamics and may influence their resilience to environmental change (Benson et al. 2004). The seed bank can also provide a reserve of genetic variability when long-lived dormant seeds act as a memory of past selection or as a source of new genetic variation through accumulated mutations (Levin 1990). Reproduction by seed enables long-distance dispersal, reduces local intraspecific competition, and generates genetic diversity (Baskin and Baskin 1998).

Meristem limitations

Negative responses of perennial grasses to fire could be predicted by identification of meristems-limited species. Meristem limitations occur when an insufficient amount of reserve or active buds contribute to aboveground tiller populations (Dalgleish and Hartnett 2009). Therefore, the size of the

bud bank and the ability to activate reserves affects the ability of a plant to adequately respond to pulses of high resource availability following fire or precipitation (Dalgleish and Hartnett 2009). Grassland plant communities may experience meristem limitation through a limited number of active buds or a limited total number of buds (Benson et al. 2004). Limitations of the total number of buds also exist across differing precipitation regimes, where more arid rangelands do not maintain a sufficient population of belowground buds to completely replace the aboveground population (Dalgleish and Hartnett 2006). Previous assessments of grass and forb meristem limitations explain variability in net primary productivity among grassland systems and in response to differing fire return intervals (Benson et al. 2004; Dalgleish and Hartnett 2006; Dalgleish and Hartnett 2009).

Assessing plant communities at the onset of tiller emergence without considering bud production, development, and maintenance represents a significant disconnect between understanding of plant community dynamics and the fundamental source of production. Characterizing the demographic mechanisms responsible for tiller recruitment from the bud bank may elucidate meristem limitations.

Previous approaches to studying bud banks

Research involving bud bank response to disturbances is in its infancy (Benson and Hartnett 2006; Dalgleish and Hartnett 2006, 2009). Additionally, approaches to quantify bud banks have varied. For example, previous experiments have strictly assessed community bud bank densities between grasses and forbs (Benson et al. 2004; Dalgleish and Hartnett 2006; Dalgleish and Hartnett 2009). In these types of investigations, multiple species with potentially varying bud production, development, maintenance, and mortality rates are combined for an overall bud bank assessment and broken down into broad functional groups. Buds that are harvested for a functional group assessment are usually taken from soil excavations of quadrats (Dalgleish and Hartnett 2006) or from soil cores (Benson et al. 2004). Studies with soil excavations only assess living buds and not dormant or dead buds (Hartnett et al. 2006).

Previous functional group analyses in the tallgrass prairie showed that plant abundance and productivity in response to fire, grazing, and drought is mediated through the belowground storage of buds (Benson et al. 2004; Dalgleish and Hartnett 2009). Frequent fire increased grass bud bank density and decreased forb bud bank density relative to infrequently burned (20-yr. fire interval) prairie (Benson et al. 2004). Decreased amounts of grass bud banks on infrequently burned sites indicate a much greater potential for meristem limitations on areas excluded from fire (Benson et al. 2004) relative to those burned frequently.

Sexual and asexual vegetative reproduction for plant populations have been compared in annually burned and infrequently burned tallgrass prairie (Benson and Hartnett 2006). Grass and forb tillers produced from belowground buds contributed to more than 99% of all established plant individuals present at the end of the growing season (Benson and Hartnett 2006). Belowground buds consistently increased in annually burned relative to infrequently burned prairie throughout the growing season. Increases in grass and forb bud bank production was observed on infrequently burned sites early during the growing season, but declined steadily throughout the rest of the growing season. Bud bank production has also been assessed from a single census taken during the dormant season. This one time sampling provides a snap-shot of the number of active buds with which a plant will overwinter. One-time inventory sampling of bud bank densities is similar to seed bank techniques used to estimate total densities of grass and forb seeds. Buds that persist throughout dormancy are typically characterized as pre-formed buds for the upcoming growing season (Lehtila and Larsson 2005). One-time inventory sampling is advantageous during plant dormancy since a baseline assessment of future tiller growth for the next growing season can be extrapolated.

Assessing bud numbers from a single census allows minimal inferences to be made concerning bud bank production, development, maintenance and mortality throughout the growing season and consecutive years. For example, dormant buds may be mobilized mid-growing season or the following growing season after drought, fire or increased precipitation (Dalgleish and Hartnett 2006). A dormant

pool of buds that can be mobilized following plant stress is considered a reliable mechanism for resilience to fire, grazing or drought (Lehtila and Larsson 2005).

Tracking bud bank production, development, maintenance, and mortality

Seasonal contributions of the bud bank could explain mechanisms driving vegetative reproduction. These seasonal fluxes include: bud population growth rates, periods of bud sensitivity and dormancy, and potential responses to pulses of high resource availability or prolonged periods of stress (Towne and Owensby 1984). Tracking bud fate including natality, longevity, and mortality of specific species has received limited emphasis, with a majority of research objectives focusing on the dormant season and on functional groups (Benson et al. 2004; Dalgleish and Hartnett 2006; Hartnett et al. 2006). Determining bud bank phenology of established perennial grasses may further describe a species' requirements for bud maintenance and potential plant stressors that lead to bud mortality.

Storing excess bud resources may limit current growth and performance of a species, whereas storing limited amounts increases the risk of mortality, decreases recovery following disturbance and constrains responses to pulses of high resource availability (Iwasa and Kubo 1997 and Russell et al. 2013). Dormant buds represent a relatively large immediate investment in construction and metabolic maintenance costs to the plant (Vesk and Westoby 2004). Despite the upfront costs of dormant buds, relatively low activation costs are associated with these buds (Vesk and Westoby 2004; Lehtila and Larsson 2005). However, buds have been hypothesized to incur a high opportunity cost if they remain dormant for too long and fail to contribute to aboveground production (Vesk and Westoby 2004).

Phenology of bud development, production, maintenance, and mortality are unknown for most grasses. Tallgrass prairie research suggests bud transitions and tiller recruitment of perennial grasses are closely tied to aboveground phenology (Ott and Hartnett 2012). Optimal growing temperature range and growth period in grasses differ by photosynthetic pathway. Most active growth of C_3 species occurs during the cooler conditions in the spring, whereas C_4 species grow most rapidly in the warmer summer months. Therefore, their belowground bud bank phenology and dynamics are likely to differ as well.

A study of tallgrass prairie work showed differences in bud bank dynamics of a C_3 and C_4 grass. The C_4 species (*Andropogon gerardii*) maintained a larger dormant bud bank compared to a C_3 species, maintained consistent bud development and transition to tillers, and its buds lived for multiple years (Ott and Hartnett 2012). It was concluded that C_4 species depend on previous years' buds to consistently produce new tillers. In contrast, *Dichanthelium oligosanthes*, a C_3 species, maintained a smaller dormant bud bank, with inconsistent bud development and active buds present year-round. As a result of over-wintering buds, *D. oligosanthes* buds lived only one year (Ott and Hartnett 2012). The bud phenology of these two species differ dramatically from one another, with C_4 species allowing buds to over-winter and recruit new spring tillers and C_3 species producing active buds throughout the year. These findings in tallgrass prairie illustrate differences in bud bank age structure, phenology, and dynamics. Therefore, differences in bud phenology, may afford some species an inherent buffering potential against timing of disturbance.

The differences found between C_4 and C_3 species in tallgrass prairie are also likely to be found in mixed-grass prairie. Differences between photosynthetic pathways may be further altered due to the compressed growing season for both C_4 and C_3 grasses in the northern Great Plains, where 90% of grass growth is completed by July 1 (Vermeire et al. 2009). In addition, bud bank dynamics differ between caespitose and rhizomatous grasses (Ott and Hartnett 2012).

Outlining the phenology of bud banks will be helpful in building a general understanding of basic bud development. Providing minimum and maximum temperature and soil moisture requirements, along with growing-degree days could establish ranges of conditions which buds require to activate and grow into a tiller. Improved understanding of phenology of bud banks can lead to improved predictions of prescribed burning or wildfire, or how fluctuations in temperature and precipitation may alter bud bank dynamics and tiller emergence.

My main objective is to understand vegetative reproductive dynamics through analysis of bud banks and associated plant community dynamics of three native perennial grasses of the northern Great Plains in response to differing seasons and frequencies of fire. It is important to determine the effects of

fire characteristics (e.g., maximum temperature, heat duration, and heat dosage) on belowground buds. Belowground buds are often well protected from fire effects (Raunkiaer 1934; Wright and Bailey 1982; Dalgleish and Hartnett 2009), but fire may have indirect effects on seasonal fluctuations in active and dormant buds, manipulating dormant overwintering densities of buds and tiller emergence. Determining bud response and bud bank trajectory will be helpful when planning prescribed fire and assessing the aftermath of prescribed burns and wildfire in mixed-grass prairie.

Greater characterization of bud phenology will provide an in-depth view of grassland structure and potential response to fire. Bud phenology work will also be helpful toward building baseline knowledge of basic bud development. Providing minimum and maximum temperature and soil moisture requirements, can establish ranges of conditions required to activate and grow buds into tillers. Improved understanding of bud bank phenology will lead to improved predictions of plant community response following prescribed burning or wildfire and climate fluctuations.

I conducted three experiments. Experiments 1 and 2 focused on the effects of season of fire and fire return interval on *Bouteloua gracilis*, *Pascopyrum smithii* and *Hesperostipa comata* bud banks. Experiment 3 was designed to reveal bud phenology by describing the general life cycle of buds for the three target species in the northern Great Plains.

Literature cited

Arno, S.F. 1996. The seminal importance of fire in ecosystem management. *In*: The use of fire in forest restoration. USDA Forest Service General Technical Report INT-GTR-341. p. 16-33.

Augustine, D.J., and D.G. Milchunas. 2009. Vegetation responses to prescribed burning of grazed shortgrass steppe. *Rangeland Ecology and Management* 62: 89-97.

Axelrod, D. 1985. Rise of the grassland biome, central North America. *The Botanical Review* 51:163-201.

Baskin, C.C., and J.M. Baskin. 1998. Seeds, ecology, biogeography, and evolution of dormancy and germination. San Diego, CA, USA: San Diego: Academic Press. 666 p.

Benson, E.J., and D.C. Hartnett. 2006. The role of seed and vegetative reproduction in plant recruitment and demography in tallgrass prairie. *Plant Ecology* 187: 163-178.

Benson, E. J., D.C. Hartnett, and H. Mann. 2004. Belowground bud banks and meristem limitation in tallgrass prairie plant populations. *American Journal of Botany* 91: 416-421.

Bernard, J.M. 1975. The life history of shoots of *Carex lacustris*. *Canadian Journal of Botany* 53: 256-260.

Biodini, M.E., A.A. Steuter, and C.E. Grygiel. 1989. Seasonal fire effects on the diversity patterns, spatial distribution and community structure of forbs in northern mixed prairie, USA. *Vegetatio* 85: 21-31.

Bradbury, J. 1819. Travels in the interior of America, in the years 1809, 1810, and 1811, Vol. V. In: R.G. Thwaites [EDS.]. Early western travels, 1748-1846. Cleveland, OH, USA: Arthur H. Clark Co. p. 351-406.

Briske, D.D. 1991. Developmental morphology and physiology of grasses. In: R.K. Heitschmidt and J.W. Stuth [EDS.]. Grazing management: An ecological perspective. Portland, OR: Timber Press. p. 85-108.

Brockway, D.G., R.G. Gatewood, and R.B. Paris. 2002. Restoring fire as an ecological process in shortgrass prairie ecosystems: initial effects of prescribed burning during the dormant and growing seasons. *Journal of Range Management* 65: 135-152.

Brown, J.K., and K.P. Davis. 1973. Forest fire: control and use. New York, New York, USA: McGraw-Hill Book Co, Inc. p. 686.

Buehring, N., P.W. Santlemann, H.M. Elwell. 1971. Responses of eastern red cedar to various control procedures. *Journal of Range Management* 24: 378-380.

Butler, J.L., and D.D. Briske. 1988. Population structure and tiller demography of the bunchgrass *Schizachyrium scoparium* in response to herbivory. *Oikos* 51:306-312.

Caldwell, M.M., J.H. Richards, D.A. Johnson, R.S. Nowak, and R.S. Dzurec. 1981. Coping with herbivory: Photosynthetic capacity and resource allocation in two semiarid *Agropyron* bunchgrasses. *Oecologia* 50: 14-24.

Castellano, M.J., and R.J. Ansley. 2007. Fire season and simulated grazing differentially affect the stability and drought resilience of a C_4 bunchgrass, C_3 bunchgrass and C_4 lawngrass. *Journal of Arid Environments* 69: 375-384.

Collins, S.L. 1992. Fire frequency and community heterogeneity in tallgrass prairie vegetation. *Ecology* 73: 2001-2006.

Dalgleish, H.J., and D.C. Hartnett. 2006. Below-ground bud banks increase along a precipitation gradient of the North American Great Plains: a test of the meristem limitation hypothesis. *New Phytologist* 171: 81-89.

Dalgleish, H.J., and D.C. Hartnett. 2009. The effects of fire frequency and grazing on tallgrass prairie productivity and plant composition are mediated through bud bank demography. *Plant Ecology* 201: 411-420.

Dalgleish, H.J., A.R. Kula, D.C. Hartnett, and B.K. Sandercock. 2008. Responses of two bunchgrasses to nitrogen addition in tallgrass prairie: The role of bud bank demography. *American Journal of Botany* 95: 672-680.

Debano, L.F., D.G. Neary, and P.F. Ffolliott. 1998. Fire's effects on ecosystems. New York, NY, USA: John Wiley & Sons. 331 p.

Denslow, J.S. 1980. Patterns of plant species diversity during succession under different disturbance regimes. *Oecologia* 46: 18-21.

Engle, D.M., and T.G. Bidwell. 2001. Viewpoint: The response of central North American prairies to seasonal fire. *Journal of Range Management* 54: 2-10.

Engle, D.M., and P.M. Bultsma. 1984. Burning of northern mixed-prairie during drought. *Journal of Range Management* 37:398-401.

Engle, D.M., R.L. Mitchell, and R.L. Stevens. 1998. Late growing-season fire effects in mid-successional tallgrass prairies. *Journal of Range Management* 51: 115-121.

Erichsen-Arychuk, C., E.W. Bork, and A.W. Bailey 2002. Northern dry mixed prairie responses to summer wildlife and drought. *Journal of Range Management* 55: 164-170.

Ewing, A.L., and D.M. Engle. 1988. Effects of late summer fire on tallgrass prairie microclimate and

    community composition. *American Midland Naturalist* 120: 212-223.

Grilz, P.L., and J.T. Romo. 1994. Water relations and growth of *Bromus inermis* Leyss. (smooth brome)

    following spring or autumn burning in fescue prairie. *American Midland Naturalist* 132: 340-348.

Harper, J.L. 1977. Population biology of plants. New York, NY, USA: Academic Press. 81 p.

Hartnett, D.C., and K.H. Keeler. 1995. Population processes. *In*: A. Joern, K.H. Keeler [EDS.]. The

    changing prairie: North American grasslands. New York, New York: Oxford University Press. p.

    82-99.

Hartnett, D.C., M.P. Setshogo, and H.J. Dalgleish. 2006. Bud banks of perennial savanna grasses in

    Botswanna. *African Journal of Ecology* 44: 256-263.

Hendrickson, J.R. and D.D. Briske. 1997. Axillary bud banks of two semiarid perennial grasses:

    occurrence, longevity, and contribution to population persistence. *Oecologia* 110: 584-591.

Higgins, K.F. 1984. Lightening fires in North Dakota grasslands and in pine-savanna lands of South

    Dakota and Montana. *Journal of Range Management* 37: 100-103.

Howe, H.F. 1995. Succession and fire season in experimental prairie plantings. *Ecology* 76: 1917-1925.

Hyder, D.N. 1972. Defoliation in relation to vegetative growth. *In*: V.B. Youngner and C.M. McKell [EDS.].

    The biology and utilization of grasses. New York, NY: Academic Press. p. 302-317.

Iwasa, Y. and T. Kubo. 1997. Optimal size of storage for recovery after unpredictable disturbances.

    Evolutionary Ecology 11: 41-65.

Jewiss, O.R. 1972. Tillering in grasses: its significance and control. *Journal of the British Grassland*

    Society 27:65-82.

Jonsdottir, L.S. 1991. Effects of grazing on tiller size and population dynamics in a clonal sedge *Carex*

    bigelowii. *Oikos* 62: 177-188.

Kays, S., and J.L. Harper. 1974. The regulation of plant and tiller density in a grass sward. *Journal of*

    Ecology 62: 97-106.

Knapp, A.K., S.L. Conrad, and J.M. Blair. 1998. Determinants of soil CO_2 flux from a sub-humid grassland:

    Effect of fire and fire history. *Ecological Applications* 8: 760-770.

Langer, R.H.M. 1972. How grasses grow. London, United Kingdom: Edward Arnold Press. 150 p.

Lehtila, K., and A.S. Larsson. 2005. Meristem allocation as a means of assessing reproductive allocation.
In: E.G. Reekie and F.A. Bazzaz [EDS.]. Reproductive allocation in plants. San Diego, CA:
Elsevier. p. 51-75.

Levin, D.A. 1990. The seed bank as a source of genetic novelty in plants. *American Naturalist* 135: 563-572.

McKendrick, J.D., C.E. Owensby, and R.M. Hyde. 1975. Big bluestem and indiangrass vegetative
reproduction and annual reserve carbohydrate and nitrogen cycles. *Agro-Ecosystems* 2: 75-93.

McPherson, G.R. 1995. The role of fire in desert grasslands. *In*: M.P. McClaran and T.R. Van Devender
[EDS.]. The desert grassland. Tucson, AZ: University of Arizona Press. p. 130-151.

Murphy, J.S., and D.D. Briske. 1992. Regulation of tillering by apical dominance: chronology, interpretive
value, and current perspective. *Journal of Range Management* 45:419-429.

NIFC. 2013. National Interagency Fire Center. Available at: http://www.nifc.gov/nicc/sitreprt.pdf
Accessed 1 August 2013.

Niklasson, M., and A. Granström. 2000. Numbers and sizes of fires: long term spatially explicit fire history
in a Swedish boreal landscape. *Ecology* 81: 1484-1499.

Norton, B.E., and J.W. McGarity. 1965. The effect of burning of native pasture on soil temperature in
northern New South Wales. *Journal of British Grassland Society* 29: 101-105.

Olson, B.E., and J.H. Richards. 1988. Annual replacement of the tillers of *Agropyron desertorum* following
grazing. *Oecologia* 76: 1-6.

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

Perrine, F.S. 1927. Hugh Evan's journal of Colonel Henry Dodge's expedition to the Rocky Mountains in
1835. *Mississippi Valley Historical* 14: 192-214.

Pyne, S.J. 1982. Fire in America: A cultural history of wildland and rural fire. Seattle, WA, USA: University
of Washington Press. 654 p.

Pyne, S.J. 2004. Tending fire: Coping with America's wildland fires. Washington DC, USA: Island Press. 237 p.

Rabinowitz, D. 1981. Buried viable seeds in a North American tallgrass prairie: the resemblance of their abundance and composition to dispersing seeds. *Oikos* 36: 191-195.

Raunkiaer, C. 1934. The life forms of plants and statistical plant geography. London, United Kingdom: Clarendon Press. 632 p.

Reader, R.J., and J. Buck. 1991. Community response to experimental soil disturbance in a mid-successional, abandoned pasture. *Vegetatio* 92: 151-159.

Robson, M. J. 1968. Changing tiller population of spaced plants of S.170 tall fescue (*Festuca arundinacea*). *Journal of Applied Ecology* 5: 575-590.

Rogers, W.E., and D.C. Hartnett. 2001. Vegetation response to different spatial patterns of soil disturbances in burned and unburned tallgrass prairie. *Plant Ecology* 155: 99-109.

Russell, M.L., L.T. Vermeire, N.A. Dufek, and D.J. Strong. 2013. Fire, defoliation, and competing species alter *Aristida purpurea* biomass, tiller, and axillary bud production. *Rangeland Ecology and Management* 66: 290-296.

Ryan, K.C., E.E. Knapp, and J.M. Varner. 2013. Prescribed fire in North American forests and woodlands: history, current practice, and challenges. *Frontiers in Ecology* 11:e15-e24.

Schimmel, J., and A. Granstrom. 1996. Fire severity and vegetation response in the Boreal Swedish forest. *Ecology* 77:1436-1450.

Sharman, B.C. 1942. Developmental anatomy of the shoot of *Zea mays* L. *Annals of Botany* 6: 245-283.

Sheley, R.L., and J.D. Bates. 2008. Restoring western juniper- (*Juniperus occidentalis*) infested rangeland after prescribed fire. *Weed Science* 56: 469-476.

Shimizu, S.S., and H. Mori. 2001. Control of outgrowth and dormancy in axillary buds. *Plant Physiology* 127: 1405-1413.

Stansbury, H. 1852. An expedition to the valley of the Great Salt Lake or Utah. London, England: Sampson Low, Son, & Co. 223 p.

Steuter, A. A. 1987. C_3/C_4 composition shift on seasonal burns: northern mixed prairie. *Journal of Range Management* 40: 27-31.

Stocks, B.J. 1991. The extent and impact of forest fires in northern circumpolar countries. *In*: J.S. Levine [EDS.]. Global biomass burning: atmospheric, climatic, and biospheric implications. Cambridge, MA: MIT Press. p. 197-202.

Tamm, A., K. Kull, and M. Sammul. 2002. Classifying clonal growth forms based on vegetative mobility and ramet longevity: a whole community analysis. *Evolutionary Ecology* 15: 383-401.

Towne, G., and C. Owensby. 1984. Long-term effects of annual burning at different dates in ungrazed Kansas tallgrass prairie. *Journal of Range Management* 37: 392-397.

Trollope, W.S.W. 1984. Fire Behavior. *In*: P.D. Booysen, and N.M Tainton [EDS.]. Ecological effects of fire in South African Ecosystems. Berlin, Germany: Springer-Verlag. p. 199-218.

Umbanhowar, C.E., Jr. 1996. Recent fire history of the northern Great Plains. *American Midland Naturalist* 135: 115-121.

Vermeire, L. T., J. L. Crowder, and D. B. Wester. 2011. Plant community and soil environment response to summer fire in the northern Great Plains. *Rangeland Ecology and Management* 64:37-46.

Vermeire, L.T., R.K. Heitschmidt, and M.J. Rinella. 2009. Primary productivity and precipitation use efficiency in mixed-grass prairie: a comparison of Northern and Southern U.S. Sites. *Rangeland Ecology and Management* 62: 230-239.

Vermeire, L.T., and M.J. Rinella. 2009. Fire alters emergence of invasive plant species from soil surface-deposited seeds. *Weed Science* 57: 304-310.

Vermeire, L.T., and A.D. Roth. 2011. Plains prickly pear response to fire: Effects of fuel load, heat, fire weather, and donor site soil. *Rangeland Ecology and Management* 64: 404-413.

Vesk, P.A., and M. Westoby. 2004. Funding the bud bank: a review of the costs of buds. *Oikos* 106: 200–208.

Weaver, T., and J. Plaggemeyer. 2004. Ecosystem effects of clonal weed invasion of mixed grass prairie. In: Fire and adaptive management of northern prairies: Montana State University Extension Biology Report #42. Montana State University, Bozeman, MT.

Willems, J.H. 1983. Species composition and aboveground phytomass in chalk grassland with different

management. *Plant Ecology* 53: 171-180.

Williams, R.F. 1975. The shoot apex and leaf growth. Cambridge, United Kingdom: Cambridge University

Press. 206 p.

Williams, R.J., A.M. Gill, and P.H.R. Moore. 1998. Seasonal changes in fire behavior in a tropical savanna

in northern Australia. *International Journal of Wildland Fire* 8: 227-239.

Woodmansee, R.G., and L.S. Wallach. 1981. Effects of fire regimes on biogeochemical cycles. *In*: F.E.

Clark and T. Rosswall [EDS.]. Terrestrial nitrogen cycles: processes, ecosystem strategies and

management impacts. Stockholm, Sweden: Sweden University Press. p. 649-669.

Wright, H.A. 1971. Why squirreltail is more tolerant to burning than needle-and-thread. *Journal of Range

Management* 24: 277-284.

Wright, H. A., and A. W. Bailey. 1982. Fire ecology: United States and southern Canada. New York, NY,

USA: John Wiley & Sons. 501 p.

CHAPTER 2. SEASON OF FIRE MANIPULATES BUD BANK DYNAMICS IN NORTHERN MIXED-GRASS

PRAIRIE

Abstract

In perennial grassland dominated systems, belowground bud banks determine plant community dynamics. In fact, plant community responses to disturbance are largely driven by the ability to generate future aboveground growth originating from belowground axillary buds. This study examined bud bank dynamics for *Bouteloua gracilis, Pascopyrum smithii, and Hesperostipa comata*, following fire in northwestern mixed-grass prairie. Belowground axillary buds were counted and classified for three growing seasons to determine immediate and short-term effects of summer, fall, and spring prescribed burns on patterns of bud bank activity, dormancy, and mortality. Summer, fall, or spring prescribed burns did not result in immediate mortality of *B. gracilis*, *P. smithii*, or *H. comata* buds ($P > 0.05$). Spring prescribed burns immediately increased *B. gracilis* bud activity ($P < 0.01$), resulting in fewer dormant buds ($P < 0.01$). In contrast, summer fire immediately reduced *B. gracilis* active buds ($P < 0.01$) and increased dormant buds ($P < 0.01$). Fall burns immediately activated *P. smithii* buds ($P < 0.01$). However, fire did not influence any immediate bud dynamics for *H. comata* ($P > 0.05$). Season of fire directly manipulated bud activity, dormancy, and mortality for these species throughout the growing and dormant seasons following fire. Using season of fire to manipulate bud bank dynamics illustrates potential to improve post-fire management strategies based on known bud development trajectories and bud dynamics following fire.

Keywords: axillary bud, dormancy, grassland, meristem limitation, prescribed burn, tiller

Introduction

One of the most crucial drivers of aboveground growth and persistence of plant communities in perennial grasslands is the below-ground population of vegetative buds. More than 99% of new tiller growth originates from belowground vegetative bud banks and less than 1% from seed (Benson and Hartnett 2006). Since bud banks serve as an essential mechanism for future tiller growth, the size of a

25

species' bud bank plays a decisive role in species and plant community response following disturbances, such as fire, grazing, and drought (Benson et al. 2004; Dalgleish and Hartnett 2009; Carter et al. 2012;).

The ability of native, perennial grass species to respond to fire is linked to belowground bud densities (Benson et al. 2004; Dalgleish and Hartnett 2009). Almost all plant species produce and maintain vegetative buds belowground, aboveground, or both (Klimesova and Klimes 2007). But, the size of the bud bank can vary both within and among species and plant communities (Lehtila 2000). The size of the bud bank available for tiller recruitment can be meristem-limited, when there are not enough buds to completely replace the aboveground tiller population, constraining primary production (Dalgleish and Hartnett 2006).

The rate of tiller recruitment and patterns of bud dormancy and viability determine both, the dynamics of individual species and species composition changes in response to fire and species specific levels of adaptations to fire (Olson and Richards 1998). Although belowground bud viability declines substantially over time (Hendrickson and Briske 1997), some species like *Andropogon gerardii*, have buds that can persist for three or more years in tallgrass prairie (Ott and Hartnett 2011). Therefore, bud banks are an important source for perennial grass regeneration for three or more years following fire (Rogers and Hartnett 2001; Dalgleish and Hartnett 2006; Klimesova and Klimes 2007).

Research evaluating fire effects on bud banks have mostly focused on tallgrass prairie with little emphasis placed on mixed-grass prairie. Few studies, if any, have examined bud bank response following different seasons of fire. Previous research in tallgrass prairie compared annual burns and 20-year burn intervals on bud bank densities (Benson et al. 2004). Annually burned prairie maintained greater grass bud bank density compared to 20-year return intervals. Similar results were found comparing annually burned prairie and four-year fire return interval prescribed burns (Dalgleish and Hartnett 2009). However, knowledge of immediate fire effects on bud bank mortality, maintenance, and emergence are limited. Furthermore, knowledge of bud demography for individual species and the timing of bud transitions in response to differing seasons of fire are relatively unknown for most dominant native perennial grasses in the northern Great Plains, despite the importance of vegetative reproduction.

Even though knowledge surrounding the effects of fire on bud banks in mixed-grass prairie is limited, the aboveground effects are well documented (Dix 1960; Engle and Bultsma 1984; Steuter 1987; Redmann et al. 1993; Shay et al. 2001; Erichsen-Arychuk et al. 2002; Vermeire et al. 2011). For example, spring burning stimulated *Pascopyrum smithii* and *Bouteloua gracilis* production by mid- and late-June (White and Currie 1983). Summer fire more than doubled *P. smithii* production the year following fire and standing crop was 47% greater for burned than non-burned sites with a wet spring in eastern Montana (Vermeire et al. 2011).

Although comparisons have been made between spring and fall fire effects in northern Great Plains plant communities (Engle and Bultsma; Steuter 1987; Biodini 1989), few studies have compared summer fire to spring and fall fires and little is known about summer fire effects. Over 70% of lightening fires in mixed-grass prairie grasslands occur during July and August from accumulations of dry fuel, increased temperatures, and decreased humidity (Higgins 1984). Due to the natural fire season corresponding with periods of increased wildfire danger and logistic challenges, summer fire research is limited in mixed-grass prairie. As a result, research has focused mostly on prescribed burns conducted outside the natural fire season.

Seasonal timing of fire may reinforce bud transitions dictated by differing photosynthetic pathways. Bud transitions and tiller recruitment in a species are inherently linked to its aboveground growth phenology based on photosynthetic pathways (Ott and Hartnett 2012). Therefore, aboveground production shifts due to seasonal prescribed burns may reflect possible timing differences in belowground recruitment from the bud bank (Steuter 1987 and Ott and Hartnett 2012). Using different seasons of fire may accelerate bud bank transitions for increased tiller recruitment from certain species, illustrating the mechanism responsible for aboveground production shifts. Furthermore, using bud banks as a quantifiable response to different seasons of fire may reveal the belowground mechanisms underlying potential plant community shifts due to appropriately timed disturbances.

This study examined bud production and bud bank dynamics of three native perennial grass species with contrasting photosynthetic pathways and growth forms in mixed-grass prairie. The

objectives of this study were to (1) assess immediate bud mortality of the C_4 grass *B. gracilis*, the C_3 caespitose grass *Hesperostipa comata*, and the C_3 rhizomatous grass *P. smithii* following summer, fall, and spring prescribed burns; (2) compare active and dormant bud abundance following summer, fall, and spring prescribed burns for each species; and (3) determine the short-term seasonal fluxes in bud dormancy, bud activity, and bud mortality following summer, fall, and spring prescribed burns. We expected bud bank dynamics to differ among these species based on photosynthetic pathway and growth form differences.

Materials and methods

Site description

Research was conducted at the USDA-ARS Fort Keogh Livestock and Range Research Laboratory (LARRL) in eastern Montana near Miles City, Montana (lat 46°24′N, long 105°56′W). This site is mixed-grass prairie located in the Palouse Dry Steppe Province of the Great Plains and has a semi-arid climate at 815 m elevation. Annual precipitation averages 343 mm, with a majority occurring from mid-April to mid-September. Average daily temperatures range from 23 °C in July to -8 °C in January and the frost-free growing season generally ranges from 125 - 150 days (Western Regional Climate Center, Reno, NV, 2013).

The research area was previously burned during the late summer of 2007 and heavily grazed in 2008, leaving approximately 300kg · ha^{-1} of remaining herbaceous stubble. After 2008, the study area was fenced off to exclude livestock grazing. Topography of the site is flat with minimal slopes (0 – 4%) and characterized as upland plains. The study site is dominated by Pinehill loams (fine, smectitic, frigid Aridic Haplustalfs) including a complex of Kobase clay loams (fine, smectitic, frigid Torrertic Haplustepts) and Gerdrum clay loams (fine, smectitic, frigid Torrertic Natrustalfs). The vegetation is dominated by perennial, native C_3 species including, needle-and-thread (*Hesperostipa comata* (Trin. & Rupr.) Barkworth), western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Löve), and threadleaf sedge (*Carex filifolia* Nutt. var. *filifolia*) and C_4 species blue grama (*Bouteloua gracilis* (Willd *ex*. Kunth) Lag. *ex* Griffiths), and to a lesser extent, buffalograss (*Bouteloua dactyloides* (Nutt.) J.T. Columbus). Annual

grasses include six week fescue (*Vulpia octoflora* (Walter) Rydb.), field brome (*Bromus arvensis*), and

cheatgrass (*Bromus tectorum L.*). The primary shrub species on the site is Wyoming big sagebrush

(*Artemisia tridentata* Nutt. Subsp*. wyomin gensis* Beetle & Young*)* and half shrub fringe sage (*A. frigida*

Willd.). Common forbs include perennial legume, silverleaf Indian breadroot (*Pediomelum argophyllum*

(Pursh.) J. Grimes) and biennial forb, yellow salsify (*Tragopogon dubius* Scop.). Annual forbs include

field cottonrose (*Logfia arvensis* (L.) Holub), woolly plantain (*Plantago patagonica* Jacq.), and rough false

pennyroyal (*Hedeoma hispida* Pursh.). Plant nomenclature follows the USDA PLANTS database (USDA,

NRCS 2013).

Experimental design and fire measurements

Fire treatments were arranged in a completely randomized design with four fire treatments

(summer, fall, spring, and no fire) and four replications applied to 16, 20 × 20-m plots. Alleyways

between plots were 3-m. Fuel load at the study site was approximately 1500 kg $\cdot$ ha^{-1} and all fires were

set using the ring-fire method (Wright and Bailey 1982). Summer fires were applied following quiescence

of *B. gracilis*, *H. comata*, and *P. smithii* plants. We applied summer fire on 7 September 2011 with

ambient temperatures 20-26 °C, winds 7-10 km $\cdot$ h^{-1}, and relative humidity 30-36%. Fall fires were

applied 3 November 2011, after the first killing frost, with ambient temperatures 8-11 °C, winds 8-12 km

$\cdot$ h^{-1}, and relative humidity 37-41%. Lastly, spring fires were applied 29 March 2012 during late

emergence phenology stage of C$_3$ plant species with ambient temperatures 15-18 °C, winds 13-20 km $\cdot$

h^{-1}, and relative humidity 39-43%.

We utilized HOBO$^{®}$ U12 J, K, S, T Thermocouple Data Loggers (Onset Computer Corporation,

Bourne, MA) with K-type Thermocouples (Omega Engineering, Inc., Stanford, CT) to create time-

temperature profiles at the plot level (Table 2.1). We placed thermocouples at the base of a perennial

grass (1-2 cm above the soil surface) and within a 10 × 10-m square in the center of the plot (4

thermocouples $\cdot$ plot $^{-1}$) to avoid edge effects. We programmed the data loggers to record temperatures

at one-second intervals. Maximum temperature was identified by finding the greatest value for each

time-temperature profile. Heat duration was calculated as time (seconds) of heat greater than 60 °C and

heat dosage was the sum of the degrees > 60 °C for the duration of elevated temperatures (degree-seconds). We used these measurements to derive the mean maximum temperature, heat duration, and heat dosage at the plot level.

Table 2.1. Thermocouple observations at the plot level for summer and fall prescribed fires near Miles City, MT.

Thermocouple measurements[1]	Mean ± SE	Maximum	Minimum
Summer fire			
Maximum temperature (C)	172 ± 15	216	148
Heat duration (s)	110 ± 20	147	67
Heat dosage (C · s)	6287 ± 663	8193	5366
Fall fire			
Maximum temperature (C)	213 ± 30	453	135
Heat duration (s)	91 ± 18	161	118
Heat dosage (C · s)	6336 ± 1443	17601	4589
Spring fire			
Maximum temperature (C)	253 ± 24	399	117
Heat duration (s)	135 ± 16	270	43
Heat dosage (C · s)	7801 ± 961	13707	1861

n=16 for summer, fall, and spring fire
[1] Heat duration and dosage of heat were assessed using 60 °C as a base temperature. Heat duration was calculated as time (seconds) of heat greater than 60 °C and heat dosage was the sum of the degrees > 60 °C for each second (degree-seconds).

Field and lab methods

Four random 5-m transects at 20-cm intervals were measured using the point-intercept transect method to quantify canopy cover, basal cover, bare ground, litter, fecal cover, and relative composition. Plant density (# of individuals within a 0.25-m^{-2} quadrat) estimates were performed at the end of the growing season for 2011, 2012, and 2013. Plant density was measured by counting plant individuals rooted within a 0.25-m^{-2} quadrat. *B. gracilis* was considered an individual when gaps of 3-cm or greater were separating neighboring *B. gracilis* individuals. *P. smithii* plant density measurements were based on number of tillers within a 0.25-m^{-2} quadrat. Individuals of *H. comata* were readily distinguished due to their bunchgrass growth form and were counted as an individual when rooted within the 0.25-m^{-2} quadrat.

Tiller counts and plant density measurements were conducted simultaneously. Tiller counts were performed at the end of growing season for 2011, 2012, and 2013 on six permanently marked plants per species and plot. Tillers were classified as vegetative or reproductive within each target plant. Definitions of an individual followed the same rules outlined for plant density measurements. Tiller counts for *P. smithii* were conducted within a 0.25 –m^{-2} quadrat and results for *P. smithii* tiller counts were reported as tillers per 0.25-m^{-2} quadrat instead of per individual plant.

Tillers were harvested from two individuals of each grass species for immediate and seasonal bud assessments. Tillers were harvested from randomly determined individuals 12 to 24 hours before and following prescribed burns to determine immediate bud mortality or activation. Tillers were harvested on 7 and 8 September 2011 (summer burns), 3 and 4 November 2011 (fall burns), and 29 March 2012 (spring burns). In order to address seasonal fluctuations for each species' bud bank, two individuals for each species were sampled on each plot throughout two years (30 July 2011, 15 March 2012, 21 July 2012, 4 November 2012, 25 January 2013, 24 February 2012, 27 March 2013, 5 May 2013, and 21 July 2013).

Buds were counted and classified as living or dead using a dissecting microscope. Active buds, dormant buds, and dead buds were confirmed using Tetrazolium and Evans Blue staining procedures. Tillers were submerged in Tetrazolium solution at room temperature for 24-hr following initial classification. Active buds stained pink and dormant and dead buds retained a white or yellowish color. If dormant buds were present on a tiller, that tiller was submerged into Evan's Blue solution at room temperature following the Tetrazolium staining for 20-min. Upon completion of Evan's Blue staining, dormant buds maintained their white pigment, whereas, dead buds stained dark blue.

Statistical analysis

Data was analyzed by species using analysis of variance (MIXED procedure of SAS, Littell et al. 2006) to quantify bud bank response immediately before and after summer, fall, and spring prescribed burns compared to non-burned plots. The model included burn treatment (fire, no fire) and season of

prescribed burning and their interactions as fixed effects with plots as a random effect. Active, dormant, and dead buds were used as response variables and the experimental unit was plot.

Data was collected prior to fire treatments and throughout one and two growing seasons post-fire to determine bud dynamics and plant community response. These data were analyzed using analysis of variance (MIXED procedure of SAS, Littell et al. 2006) with tiller harvest date as a repeated measure. The model included tiller harvest date and season of fire and their interactions as fixed effects for bud bank dynamics.

For aboveground dynamics, the model included year, fire, and their interactions as fixed effects. Response variables consisted of plant density, vegetative tillers, flowering tillers, total tillers, basal cover, bare ground, litter, and relative composition. Pre-treatment data taken from 2011 were utilized as a covariate. If pre-treatment measurements were not significant ($P > 0.05$), they were eliminated from the model. We set statistical significance at $P < 0.05$ for all models.

Results

Growing conditions prior to the start of treatments in spring of 2011 (April –June) were extremely wet, with a record (250% of average) amount of spring precipitation (Fig. 2.1). The following winter, spring, and summer were 34% below average precipitation (Fig. 2.1). Spring 2012 was the second driest on record and drought conditions persisted throughout eastern MT and the northern Great Plains (Western Regional Climate Center, Reno, NV 2013). Spring of 2013 (two growing seasons following fire) brought near-average spring and summer precipitation, enabling growth of cool- and warm- season grasses following severe drought of 2011-2012.

□1937-2013 ■2010-2011 ▫2011-2012 ■2012-2013
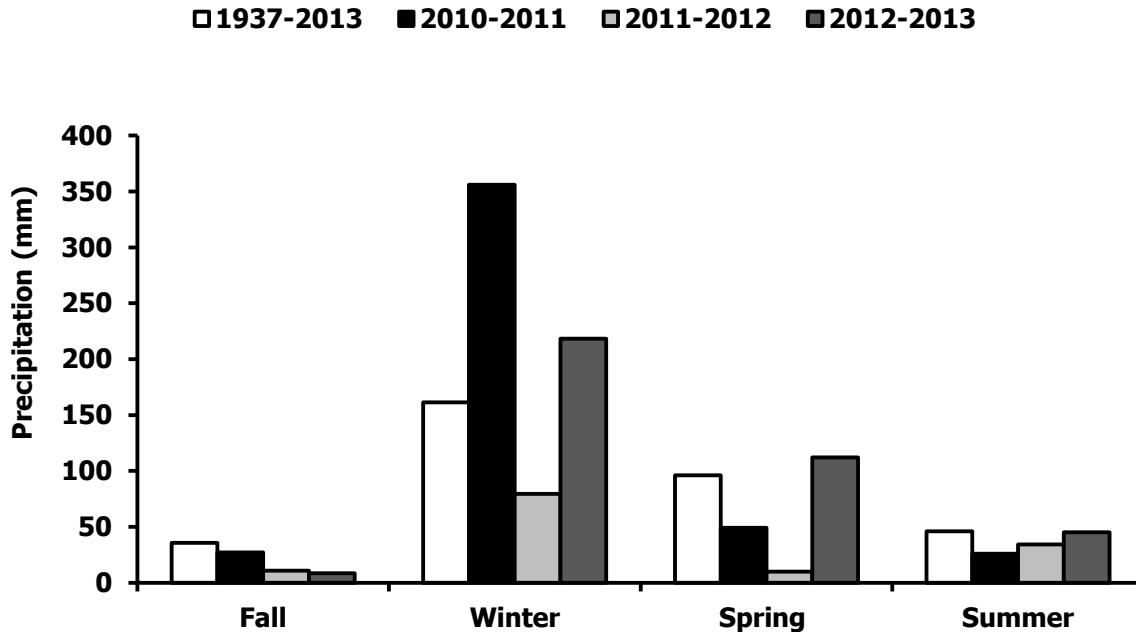
Figure 2.1. Study site 76 yr mean seasonal precipitation, fall (Oct-Dec), winter (Jan-Mar), spring (Apr-Jun), and summer (Jul-Sep) precipitation year (Oct-Sep) near Miles City, MT.

Aboveground plant community and ground cover

Relative composition of *B. gracilis* did not differ among summer, fall or spring burns ($P = 0.07$). However, *B. gracilis* composition varied one and two years post-fire (13 vs. 21 ± 3%; $P < 0.05$). Cool-season grasses (*H. comata* and *P. smithii*) relative composition were similar following summer, fall, or spring burns ($P = 0.20$, 0.25 and 0.31) and were similar between one and two growing seasons after fire ($P = 0.20$). In general, all fires reduced shrub composition more than 90% the second growing season (12.75 vs. 1.25 ± 0.7%; $P < 0.01$) and reduced annual grasses more than 86% the second growing season post fire (26.8 vs. 3.8 ± 4.6%; $P < 0.01$).

Across seasons of burning, basal cover for *B. gracilis* decreased 45% two years post-fire (9.4 ± 2.3%; $P < 0.05$). Basal cover of *H. comata* varied by season of burning and year, with summer and spring burns increasing basal cover one year post-fire compared to non-burned plots ($P < 0.05$). *P. smithii* basal cover was similar among seasons of burning and between growing seasons following fire ($P = 0.44$; 0.07). Bare ground increased following summer, fall and spring burns, regardless of year, in comparison to non-burned plots (41, 39, and 38 vs. 15 ± 5%; $P < 0.01$). In addition, summer, fall and spring fires reduced litter amounts compared to non-burned plots (11, 14, and 10 vs. 57 ± 3%; $P <$

33

0.01), but litter cover increased 29% the second post-fire growing season, suggesting a steady recovery. Percent fecal ground cover following summer, fall and spring fires increased the second growing season compared to the first growing season and non-burned control plots (24, 17 and 9 ± 2.8%; $P < 0.01$).

Bouteloua gracilis dynamics

Pre-treatment plant density was similar between 2012 and 2013, despite the drought effects in 2012 ($P = 0.35$). Plant density was similar in summer burned (24.8 ± 7.6 plants · m^{-2}), fall burned (35.2 ± 7.6 plants · m^{-2}), spring burned (25.2 ± 7.6 plants · m^{-2}), and non-burned plots (22.0 ± 7.6 plants · m^{-2}; $P = 0.15$).

Drought reduced vegetative tillers one growing season post-fire, but tillers doubled in the second growing season post-fire ($P < 0.01$; Table 2.2). Summer, fall, and spring fire increased reproductive tillers during the second growing season compared to non-burned plots and first year growing season responses ($P < 0.01$).

Table 2.2. Fire treatment and year effects on *B. gracilis* vegetative and reproductive tillers plant^{-1} near Miles City, MT.

Measurement	Treatment and Year Effects				SEM	*P*-value
	------------------------Fire------------------------					
	Summer	Fall	Spring	No fire		
Vegetative tillers	42.8	33.3	38.8	34.9	6.4	0.73
Reproductive tillers						
2012	0.3d	0.3d	0.4d	0.4d	1.4	< 0.01
2013	16.9b	19.8a	15.8b	6.9c		
	----------Year----------					
	2012	2013				
Vegetative tillers	25.3b	49.6a			4.3	< 0.01

[1] **Means within measurement are similar when followed by a common letter (*P* > 0.05).**

Bud mortality did not occur immediately following any season of fire ($P = 0.41$; Table 2.3). Surprisingly, active bud development increased 71% following spring burns compared to fall and summer fires, resulting in fewer dormant buds following spring fires ($P < 0.01$). In contrast to spring burns, summer fire decreased active buds 35% ($P < 0.01$). Consequently, dormant buds increased 77% immediately following summer fire ($P < 0.01$). Fall fire did not affect active or dormant buds ($P > 0.05$).

Table 2.3. Immediate (< 24 hr) summer, fall, and spring fire effects on active, dormant, and dead axillary bud means belonging to *B. gracilis* tillers near Miles City, MT.

Measurement	Summer		Fall		Spring		SEM	P-value
	No fire	Fire	No fire	Fire	No fire	Fire		
B. gracilis	--------------------------buds tiller^{-1}--------------------------							
Active	3.9a	2.5b	4.9	4.3	2.4b	8.3a	0.7	<0.01
Dormant	0.3b	1.1a	0.3	0.3	1.5a	0.3b	0.3	0.01
Dead	0.0	0.0	0.0	0.3	0.0	0.0	0.1	0.41

[1] **Means within bud classification and season of fire are similar when followed by a common letter ($P > 0.05$).**

Season of fire did not affect bud mortality throughout the two growing seasons post-fire ($P = 0.15$) and, overall, the number of dead buds was very low. Dead buds were most abundant during late winter ($P < 0.01$; Fig. 2.2). Dead buds peaked during the first post-fire winter and during the second post-fire winter. Surprisingly, bud mortality was similar between dry and wet years.
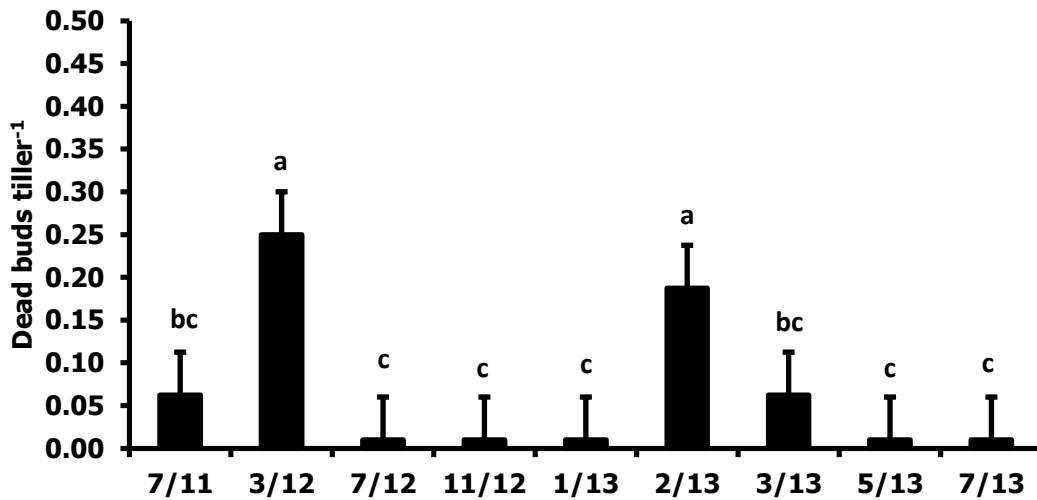
Figure 2.2. Mortality of *B. gracilis* buds tiller^{-1} (± SEM) throughout two growing seasons near Miles City, MT. Means marked with the same letter are similar ($P < 0.01$).

Season of fire manipulated bud bank dynamics differently throughout the two growing seasons after fire ($P < 0.01$; Fig. 2.3). Summer burns resulted in 35% more active buds than fall or spring burned plots throughout winter dormancy ($P < 0.01$). Development of active buds following fall burns tripled by the end of the second growing season post-fire compared to non-burned plots ($P < 0.01$). Spring burns increased active buds 77% at the beginning of the second growing season post-fire

compared to non-burned plots and all buds present were active ($P < 0.01$). Interestingly, summer, fall

and spring burns increased active buds compared to non-burned plots during the drought of the first

growing season post-fire (6.0, 5.4 and 5.5 vs. 2.3 ± 0.6 buds· tiller $^{-1}$; $P < 0.01$).
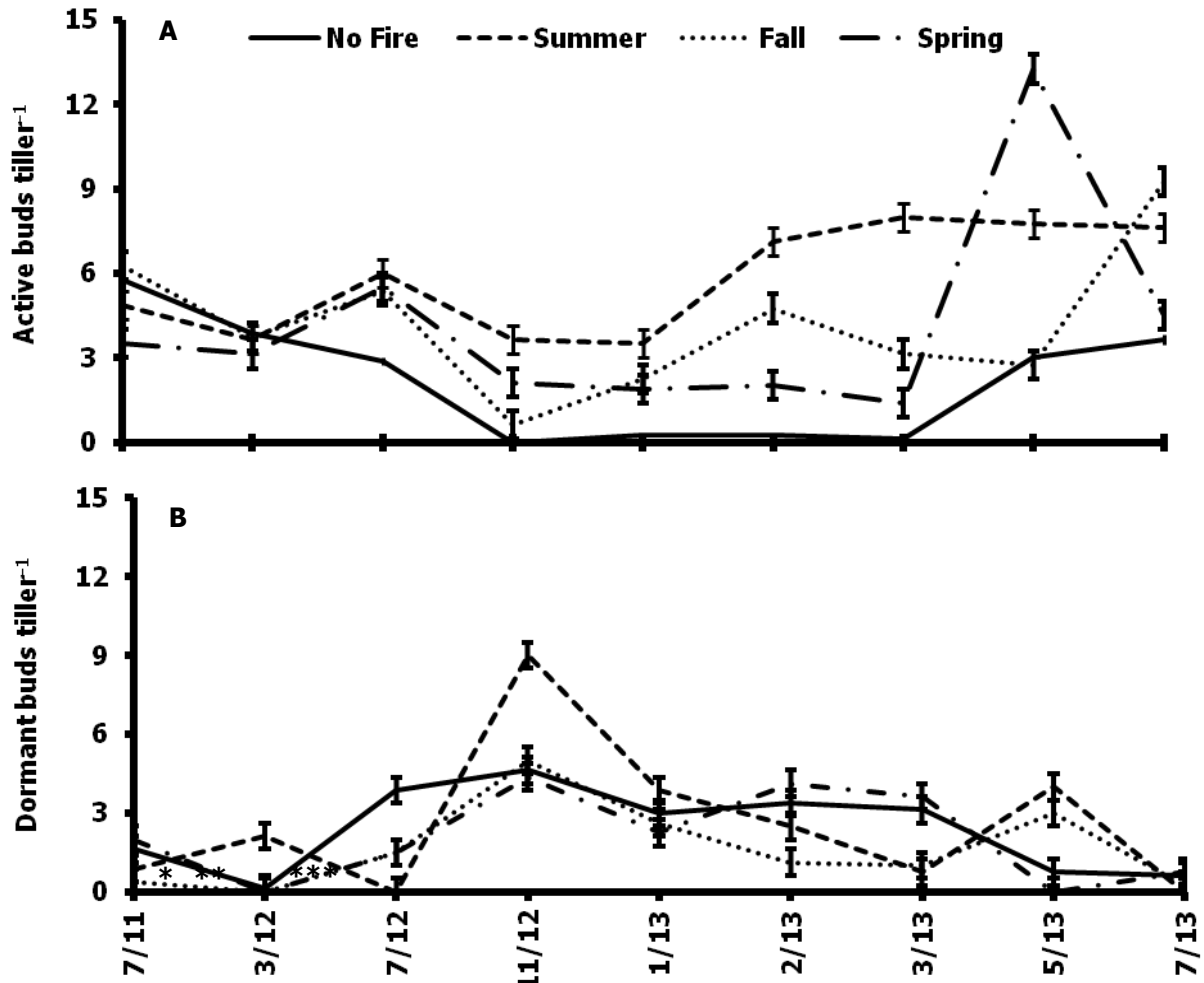
**Figure 2.3 Bud dynamics of *B. gracilis* from July 2011 to July 2013 following no fire,
summer, fall, and spring prescribed fire near Miles City, MT on *B. gracilis* A) active buds tiller^{-1} ($P < 0.01$) (± SEM) and B) dormant buds tiller^{-1} ($P < 0.01$) (± SEM). * = summer fire
treatment 8 September 2011, ** = fall fire treatment 3 November 2011, and *** = spring
fire treatment 29 March 2012.**

Dormant buds decreased the first growing season following summer, fall, and spring burns

compared to non-burned plots ($P < 0.01$; Fig. 2.3). Summer burned plots increased dormant buds

entering the first winter after fire; whereas, fall and spring burns were similar to non-burned plots ($P <$

0.01). At the initiation of the second post-fire growing season, dormant buds increased with summer and fall burns ($P < 0.01$). Dormant buds transitioned into active buds during May of the second growing season after spring fire and two months later after summer fire.

Hesperostipa comata dynamics

Plant density was similar ($P = 0.50$) in summer burned (14.0 ± 3.2 plants $\cdot$ m^{-2}), fall burned (15.2 ± 3.2 plants $\cdot$ m^{-2}), spring burned (13.6 ± 3.2 plants $\cdot$ m^{-2}), and non-burned plots (20.0 ± 3.2 plants $\cdot$ m^{-2}). In addition, density estimates did not differ between the subsequent two growing seasons post-fire (16.0 and 15.2 ± 2.4 plants $\cdot$ m^{-2}; $P = 0.83$).

Pre-treatment vegetative and reproductive tiller counts did not differ between 2012 and 2013. Both vegetative and reproductive tiller numbers were similar among fire treatments, but each differed between post-fire study years (Table 2.4). Fire had little effects on vegetative and reproductive tillers, regardless of the season in which it was burned ($P = 0.46$ and 0.60, respectively). However, vegetative tillers more than doubled the second growing season ($P < 0.01$). Furthermore, reproductive tillers increased the second growing season post-fire ($P < 0.01$).

Table 2.4. Fire treatment and year effects on *H. comata* vegetative and reproductive tillers plant^{-1} near Miles City, MT.

Measurement	Treatment and Year Effects				SEM	*P*-value
	------------------------Fire------------------------					
	Summer	Fall	Spring	No fire		
Vegetative tillers	20.3	24.8	22.3	29.6	4.2	0.46
Reproductive tillers	2.4	2.6	2.1	3.3	0.6	0.60
	-----------Year-----------					
	2012	2013				
Vegetative tillers	15.1b	33.4a			3.0	< 0.01
Reproductive tillers	0.2b	5.0a			0.5	< 0.01

[1] **Means within measurement are similar when followed by a common letter (*P* > 0.05).**

Neither summer, fall or spring fires had an immediate effect on the number of *H. comata* active or dormant buds ($P = 0.43$ and 0.55; respectively; Table 2.5). Fire also had no immediate effect on bud mortality regardless of season ($P = 0.56$).

Table 2.5. Immediate (< 24 hr) summer, fall, and spring fire effects on active, dormant, and dead axillary buds belonging to *H. comata* tillers near Miles City, MT.

Measurement	Summer		Fall		Spring		SEM	*P*-value
	No fire	Fire	No fire	Fire	No fire	Fire		
H. comata	----------------------------buds tiller^{-1}-----------------------							
Active	1.4	1.0	1.1	1.4	4.3	3.9	0.3	0.43
Dormant	0.0	0.4	0.4	0.8	0.0	0.0	0.2	0.55
Dead	0.1	0.4	0.1	0.3	0.4	0.3	0.2	0.56

[1]**Season of fire treatment means within bud classification and species are similar when followed by a common letter (*P* < 0.05).**

Although there were no immediate fire effects, season of fire played a key role in active bud dynamics throughout two growing seasons after fire (*P* < 0.01; Fig. 2.4A). At the end of the first growing season, summer burns increased active buds compared to fall and spring burns and non-burned plots (*P* < 0.01). At the end of the second winter dormant period (February 2013), active buds were greater following fall and spring burns compared to summer burns and non-burned plots (*P* < 0.01). Active buds were reduced entering the second growing season post-fire by summer and spring burns (*P* < 0.01). These fire effects continued throughout the second growing season for summer, fall and spring burns 81, 85 and 65% reductions, respectively.

Dormant buds after fire were minimal, but varied between winter and growing seasons (*P* < 0.05; Fig. 2.4B). Dormant buds were not present on *H. comata* tillers during winter months of the first year post-fire. The following July, dormant buds increased for spring burns and non-burned plots. During the winter months of the second year, dormant buds did not differ among fire treatments, including non-burned plots (*P* > 0.10). Similar dormant bud densities were maintained among all treatments (*P* = 0.18) at the start of the second growing season. Mortality of buds occurred only during the beginning of the first growing season following spring burns (*P* < 0.05; Fig. 2.4C). Spring burns increased dead buds 72% compared to fall burns and non-burned plots during March 2012. No bud mortality was observed at the conclusion of the first and second growing seasons (*P* = 0.12).
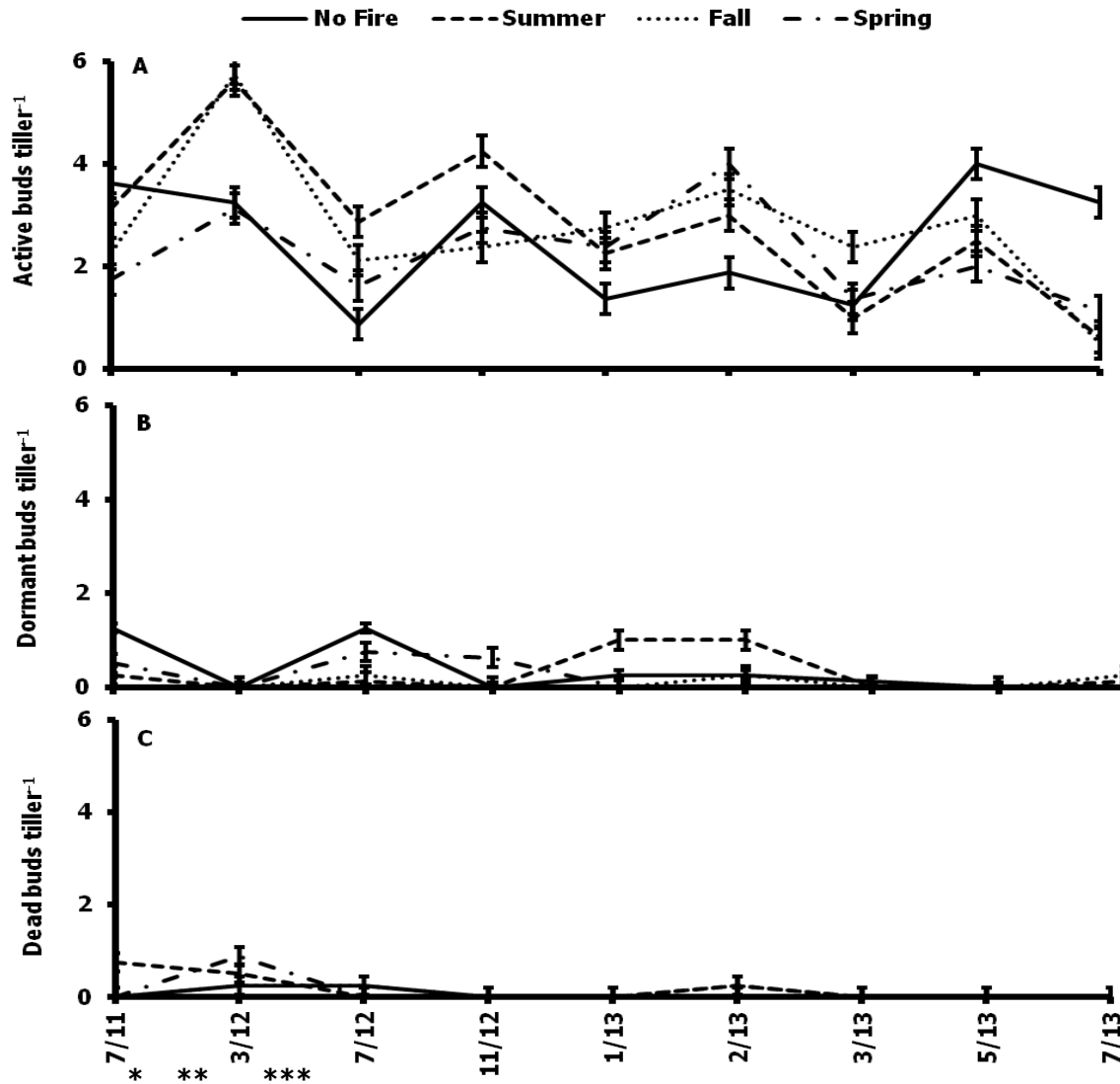
Figure 2.4. Short-term bud dynamics of _H. comata_ from July 2011 to July 2013 following no fire, summer, fall, and spring prescribed fire near Miles City, MT on _H. comata_ A) active buds tiller⁻¹($P < 0.01$) (± SEM), B) dormant buds tiller⁻¹ ($P < 0.05$) (± SEM) and C) dead buds tiller⁻¹ ($P < 0.05$) (± SEM). * = summer fire treatment 8 September 2011, ** = fall fire treatment 3 November 2011, and * = spring fire treatment 29 March 2012.**

Pascopyrum smithii dynamics

Plant density was similar ($P > 0.53$) following summer burned (58.4 ± 9.6 plants · m⁻²), fall burned (54.0 ± 9.6 plants · m⁻²), spring burned (49.2 ± 9.6 plants · m⁻²), and non-burned plots (48.8 ± 9.6 plants · m⁻²$, respectively). Density estimates did not differ between one and two growing seasons post-fire (57.6 and 41.6 ± 10.0 plants · m⁻²$; $P > 0.07$).

Pre-treatment tiller counts were similar between 2012 and 2013, despite drought during the first growing-season post-fire. Vegetative tillers increased following summer and fall fire ($P < 0.05$; Table 2.6), but were unaffected afterwards ($P > 0.10$). Reproductive tillers were more abundant the second than first growing season following summer and fall fires (8.0 and 6.7 vs. 0.4 and 0.0 ± 1.1 tillers · 0.25-m^{-2}; $P < 0.01$).

Table 2.6. Fire treatment and year effects on *P. smithii* vegetative and reproductive tillers 0.25-m^{-2} near Miles City, MT.

Measurement	Treatment and Year Effects				SEM	*P*-value
	---------------------------Fire---------------------------					
	Summer	Fall	Spring	No fire		
Vegetative tillers	15.4a	14.6a	11.5b	8.9c	1.5	< 0.05
Reproductive tillers						
2012	0.4d	0.1d	0.5d	0.3d	1.10	< 0.01
2013	8.0a	6.7b	2.4c	1.0d		
	------------Year----------					
	2012	2013				
Vegetative tillers	11.2	14.0			1.1	0.10

[1] **Means within measurement are similar when followed by a common letter ($P > 0.05$).**

Summer, fall, and spring fire had no immediate effect on dormant or dead buds for *P. smithii* ($P = 0.57$ and 0.43, respectively; Table 2.7). However, development of active buds increased 37% immediately following fall fire ($P < 0.05$).

Table 2.7. Immediate (< 24 hr) summer, fall, and spring fire effects on active, dormant, and dead axillary buds belonging to *P. smithii* tillers near Miles City, MT.

Measurement	Summer		Fall		Spring		SEM	*P*-value
	No fire	Fire	No fire	Fire	No fire	Fire		
P. smithii	--------------------------buds tiller^{-1}--------------------------							
Active	3.4	2.9	3.6b	5.8a	3.9	3.8	0.5	0.05
Dormant	0.0	0.3	0.3	0.3	0.0	0.1	0.1	0.57
Dead	0.0	0.4	0.3	0.1	0.3	0.3	0.2	0.43

[1] **Season of fire treatment means within bud classification and species are similar when followed by a common letter ($P < 0.05$).**

Development of active buds primarily occurred during the second year post-fire ($P < 0.01$; Fig. 2.5). Active, overwintering buds increased following summer and fall fire compared to spring burns and

non-burned plots ($P < 0.01$). Increased activity of buds continued through the winter months and summer, fall, and spring burns increased active buds by 84, 82, and 80% compared to non-burned plots ($P < 0.01$). By the beginning of the second post-fire growing season, only fall and summer fires increased bud activity ($P < 0.01$), and active bud counts became similar across all fire treatments by the middle of the second growing season ($P = 0.25$).
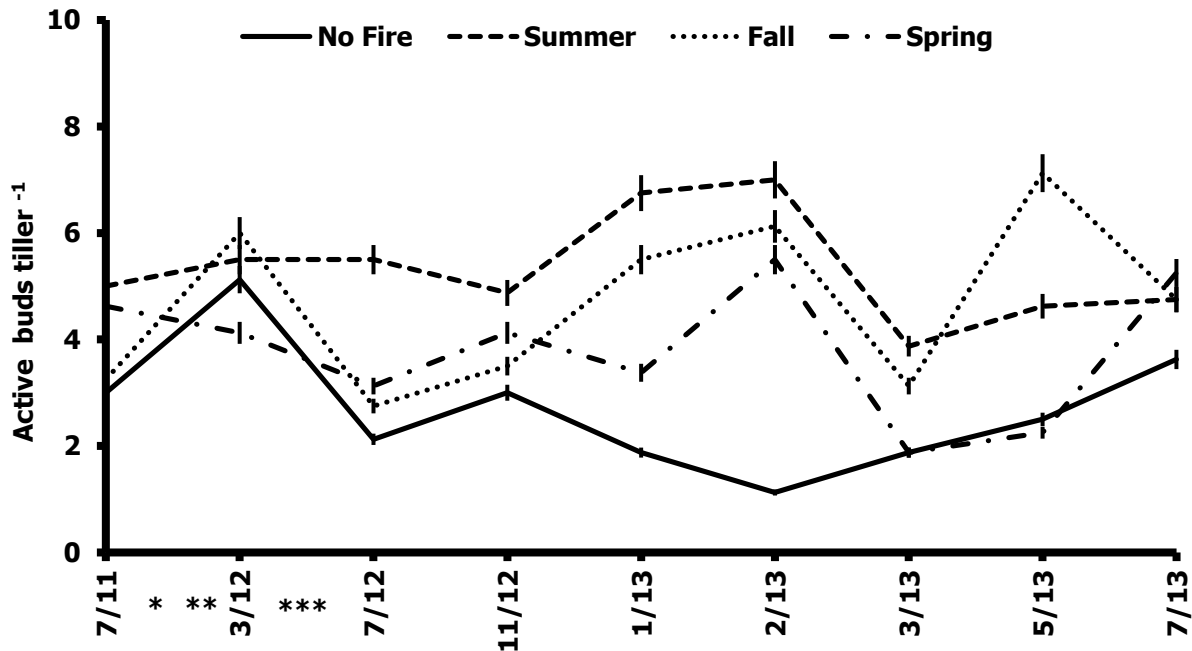
Figure 2.5. Active buds tiller^{-1} ($P < 0.01$) for *P. smithii* from July 2011 to July 2013 following no fire, summer, fall, and spring prescribed fire active buds tiller^{-1} near Miles City, MT (± SEM). * = summer fire treatment 8 September 2011, ** = fall fire treatment 3 November 2011, and * = spring fire treatment 29 March 2012.**

Maintenance of dormant buds differed among fire treatments ($P < 0.05$; Fig. 2.6) and throughout growing and dormant seasons ($P < 0.01$; Fig. 2.7). In general, dormant buds were reduced by fire ($P < 0.05$). Non-burned plots had 64% more dormant buds than summer, fall, or spring burned plots. Dormant buds were more abundant during July of the first growing season and maintained a greater dormant bud bank throughout the winter of the second year ($P < 0.01$). Mortality of buds increased during initiation of the second growing season ($P < 0.01$; Fig. 2.8) with no observed fire effects ($P = 0.74$).

41
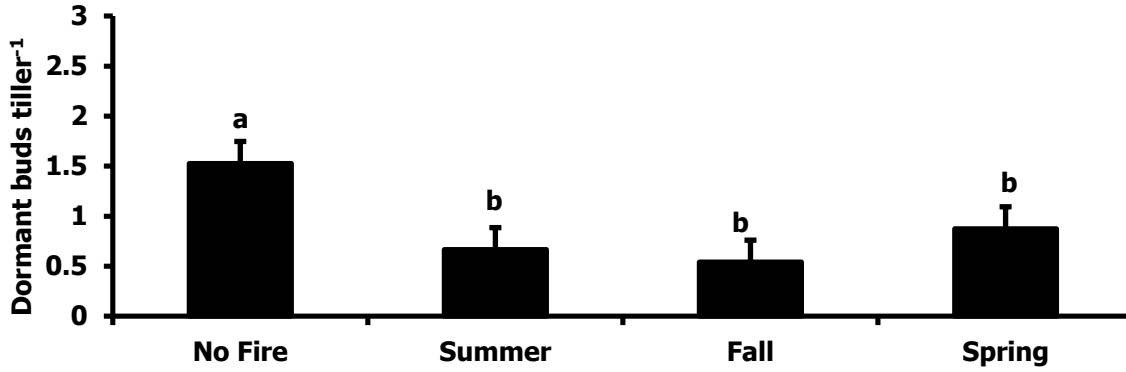
Figure 2.6. Dormant buds tiller^{-1} of *P. smithii* (± SEM) following no fire, summer, fall and spring fire treatments for two growing seasons near Miles City, MT. Means marked with the same letter are similar (*P* < 0.05).
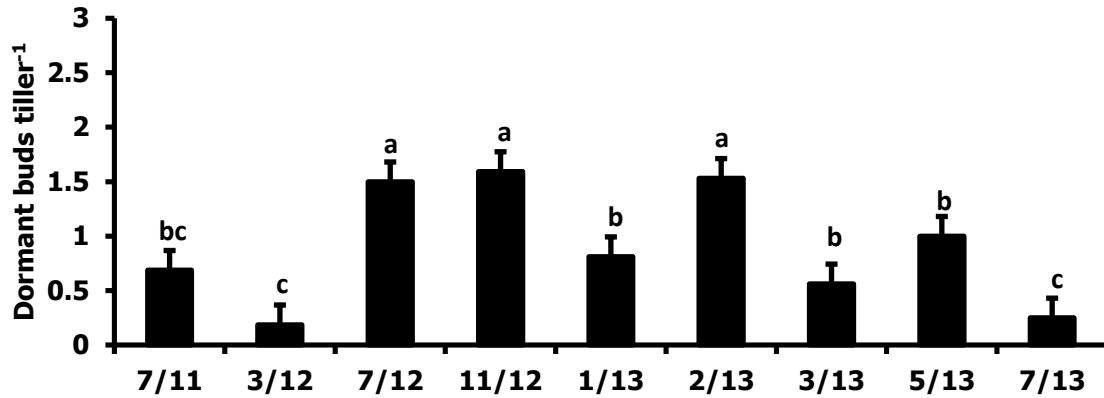
Figure 2.7. Dormant buds tiller^{-1} of *P. smithii* (± SEM) throughout two growing seasons near Miles City, MT. Means marked with the same letter are similar (*P* < 0.01).
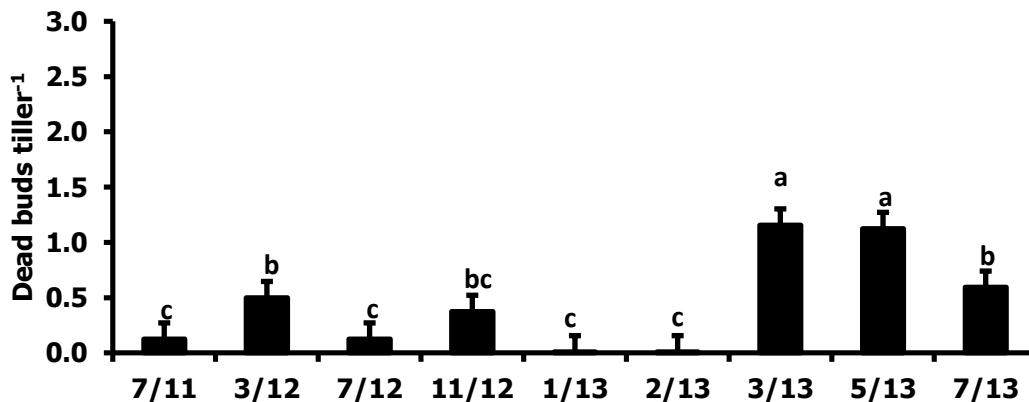
Figure 2.8. Dead buds tiller^{-1} of *P. smithii* (+SEM) throughout two growing seasons near Miles City, MT. Means marked with the same letter are similar (*P* < 0.01).

Discussion

Aboveground dynamics

      Growing conditions varied among study years. The drought conditions during spring 2012 altered below and aboveground responses. Spring precipitation in 2013 was above average; therefore, any prolonged drought effects of 2012 were minimal and isolated within 2012. Prescribed fire reduced annual grass and woody shrub abundance during the first post-fire growing season, with further reductions in abundance two years following fire. Observed decreases of annual grasses and shrubs are similar to earlier observations from northern mixed prairie (Vermeire et al. 2011) and from tallgrass prairie (Heisler et al. 2003). Fire reductions in litter and increases in bare ground during the first post-fire growing season are similar to previous mixed prairie research on summer wildfire effects in southeastern Alberta (Erichsen-Arychuk et al. 2002) and summer prescribed burn effects in eastern Montana (Vermeire et al. 2011).

      Plant density was similar among the three species for all seasons of fire and for two years following fire. Similar densities among summer, fall, and spring fire combined with variable spring precipitation suggest that native perennial grasses are well adapted to fire and spring drought. These responses are indicative of their evolutionary history and plant adaptations (Engle and Bultsma 1984; Umbanhowar 1996; Pausas and Schwilk 2012). Mixed-grass prairie most likely burned every 5 to 10 years (Wright and Bailey 1982; Higgins 1984). As a result of recurring disturbances, native grasses have adapted their growth and reproductive mechanisms to withstand frequent disturbance (Hartnett and Keeler 1995).

      B. gracilis and *H. comata* tiller numbers decreased the first post-fire growing season, regardless of season of burning. Drought effects likely stunted vegetative and reproductive tiller emergence, especially for C_3 species, since spring precipitation was 44% below the long-term (1937-2012) average during the first post-fire growing season. Spring precipitation is considered superior to annual or growing season precipitation for predicting peak standing crop in eastern Montana (Vermeire et al. 2008;

Vermeire et al. 2009; Wiles et al 2011). Reductions in tillers during the first year following fire

treatments are similar to drought responses in tallgrass prairie, where tillers were reduced by drought,

but increased the following year due to bud bank recovery (Carter et al. 2012). By the second post-fire

growing season both total and reproductive tillers more than doubled. Similar second year response

following fire has been observed in northern mixed-grass prairie, where perennial grasses made a

substantial recovery the second year following a severe fire event during drought conditions (Engle and

Bultsma 1984). Increased *B. gracilis* and *P. smithii* reproductive tiller emergence the second growing

season post-summer and fall fire is likely a result of increased bud activity.

Immediate season of fire effects on bud bank dynamics

Summer, fall, or spring fire did not affect bud mortality for any species immediately after fire.

Survival of species during a fire event usually depends on available vegetative buds to generate new

growth (Dalgleish and Hartnett 2009). Belowground buds are well-protected from direct effects of fire

due to the buffering capacity of soil and soil moisture (Alexander 1982; Bradstock and Auld 1995). In

contrast, aboveground vegetation is susceptible to fire and a temperature of only 60 °C during one

minute is, in most cases, sufficient to coagulate proteins and cause lethal damage to aboveground plant

tissue (Precht et al. 1973). Both *B. gracilis* and *P. smithii* axillary bud resistance to fire stem from their

geophyte growth form (Raunkiaer 1934).

Ewing and Engle (1988) concluded *H. comata* should be more sensitive to fire based on its

bunchgrass growth form and its elevation of meristems into the crown of the plant as the growing season

progresses. Bunchgrasses are generally more susceptible to fire damage because litter accumulates in

the crown of these species over time, increasing the fuel load and resulting in greater heat duration and

heat dosage (Wright 1971; Wright and Bailey 1982; Engle et al. 1998). Bunchgrasses also grow from the

center of the plant and expand outward (Wright 1971). This type of growth promotes litter accumulation

in the center of the bunchgrass, rather than the outer edges. Bud mortality in bunchgrasses has been

reported to be greater in the middle of the plant compared to the outer edges where litter accumulation

is less (Morgan and Lunt 1999). In contrast to previously observed fire responses of bunchgrasses,

immediate bud mortality did not occur in this study for *H. comata*. This may be due to the dense bunch-form of *H. comata*, which may have extinguished the fire or decreased heat duration due to lack of oxygen at the crown. Some plants like *H. comata*, protect vegetative buds through the sheath of the tiller, where top removal of fine fuels has minimal effects on well-protected axillary buds that are just above the soil surface (Morgan and Lunt 1999).

When a fire occurs, it generally sweeps across the top of vegetation with minimal heat transfer to the base (Wright 1971). Due to *H. comata's* coarse stems and dense plant crown, the base of the plant may have been compacted adequately enough to prevent direct heat transfer to the buds (Haile 2011). Heat threshold conditions required to produce greater than 50% mortality of *H. comata* are 56, 985 °C·s, 10.4 minutes above 60 °C, 628 °C maximum temperature, and 8,500 kg · ha^{-1} fuel load (Haile 2011). The thresholds reported for *H. comata*, are substantially higher than the fire intensities recorded for summer, fall, and spring fire treatments for this study. Therefore, the presumption that fire often damages bunchgrasses, like *H. comata*, is not justified by this study's results because required heat thresholds for *H. comata* mortality represent fire intensities that are not very common in Great Plains grasslands and were not achieved in this study for any seasons of fire treatments.

Activity of *B. gracilis* buds increased immediately following spring fires. Additionally, fall fire immediately increased active bud development of *P. smithii*. Spring and fall fire served as catalysts for both species, stimulating new bud growth, transitioning dormant buds into active buds, and initiating buds into future tillers within 24 hours of burning. Immediate active bud responses were not expected, but may be possible due to improved growing conditions post-fire through the removal of litter and existing vegetation (Wright and Bailey 1982). Removal of dead vegetation increases the quantity of light that reaches the soil surface, warming it (Knapp et al. 1998) altering the incident red:far-red light ratio (Willems 1983). Increased red:far-red light ratios (Tomlinson and O'Connor 2004) and increased soil temperatures (Svejcar 1990) are potential cues for the growth of buds into tillers.

Immediate bud bank responses to fire may also be due to temporary transitions of buds. In many cases, newly formed axillary buds enter transition stages of temporary growth or dormancy (Devitt

and Stafstrom 1995; Shumizu and Mori 2001). Buds cycle through these temporary transition stages until a developmental or environmental cue signals buds to become fully dormant or undergo growth. The timing of spring fire most likely manipulated C_4 buds during temporary transition periods and acted as an environmental signal for buds to enter active growth. Similarly, the timing of fall fire manipulated developing C_3 buds to increase activity and growth. In contrast, summer fires decreased *B. gracilis* bud activity and increased bud dormancy. Axillary buds in temporary transitions can become developmentally arrested if an environmental cue signals heat or drought induced quiescence. These dormant axillary buds resume development at a later time, depending on their developmental program or in response to environmental cues (Stafstrom and Sussex 1992; Shimizu and Mori 1998). This temporary bud strategy is possible since northern mixed-grass prairie evolved under summer fire regimes where temporary dormancy of buds may be advantageous in order to cope with increased summer fire intensity and stress (Higgins 1984; Umbanhowar 1996).

Season of fire effects on short-term bud bank dynamics

B. gracilis bud banks were more active during drought if burned the previous year. Bud banks on non-burned plots increased dormant buds compared to burned plots during summer drought. Bud bank activity of *B. gracilis* likely increased due to increased nitrogen availability following fire (Cui and Caldwell 1997; Enquist et al 1998; Russell et al. 2013). Since *B. gracilis* total buds did not increase during drought conditions, it is likely that B. *gracilis* bud banks were able to recover by transitioning dormant buds into active buds, contributing to plant community persistence in challenging post-drought and fire environments.

By the end of the study, *H. comata* had the smallest bud bank of all species and contained 84% fewer buds on burned plots compared to control plots. When species do not maintain a large population of belowground buds throughout the growing season, meristem limitations occur. Since bud production did not increase as tiller production increased, bud banks of *H. comata* were not replenished and plants became meristem limited. Depleted bud banks of *H. comata* also align with previous bud bank studies in tallgrass prairie that reported C_3 species maintain a smaller bud bank with short-lived buds and tiller

recruitment occurs from current year's buds only (Ott and Hartnett 2012). Therefore, in northern mixed prairie, *H. comata* would be expected to closely track inter-annual environmental changes and demonstrate fluctuating bud populations and potential meristem limitations due to variable environmental conditions.

Unlike bud banks of *H. comata*, *P. smithii*, also a C_3 grass, maintained a large overwintering reserve of dormant and active buds following summer and fall fires. New bud growth and increased bud activity for *P. smithii* during winter dormancy following spring and summer fires are similar to this study's C_4 bud bank results and C_4 bud response in tallgrass prairie. For C_4 species, young buds plus older buds produced in previous years form the overwintering bud bank (Ott and Hartnett 2012). Similar to *P. smithii* bud bank results, overwintering bud banks of C_4 species in tallgrass prairie remain active in order to rapidly transition into new tillers before warmer summer temperatures occur (Ott and Hartnett 2012).

Conclusions

Native, perennial grasses in mixed-grass prairie rely heavily on the maintenance of the bud bank to regenerate and persist in environments dominated by fire, drought and herbivory, common characteristics of the northern Great Plains. Belowground bud banks respond positively to moderate fire intensities due to increases in bud activity and minimal bud mortality following fire. Both *B. gracilis* and *P. smithii* responded to fire with increased bud activity throughout winter dormancy in order to replenish bud bank reserves and capitalize upon early spring resources (e.g., precipitation). This plant strategy is effective in northern mixed prairie due to variation in precipitation patterns. Furthermore, summer, fall, and spring fires manipulated C_3 and C_4 grasses differently, illustrating the potential for flexible resource allocation and growth tradeoffs. Differences in season of fire responses due to photosynthetic pathways can influence future patterns of reproduction and community dynamics of perennial grasses. Manipulating bud bank dynamics by season of fire illustrates potential to improve management strategies following wildfires or prescribed burns based on the trajectory of bud development. Incorporating bud bank dynamics will also inevitably improve overall predictions of grassland vegetation response to fluctuating environmental growing conditions.

Acknowledgements

Literature cited

Alexander, M.E. 1982. Calculating and interpreting forest fire intensities. *Canadian Journal of Botany* 60: 349-357.

Benson, E.J., and D.C. Hartnett. 2006. The role of seed and vegetative reproduction in plant recruitment and demography in tallgrass prairie. *Plant Ecology* 187: 163-178.

Benson, E. J., D.C. Hartnett, and H. Mann. 2004. Belowground bud banks and meristem limitation in tallgrass prairie plant populations. *American Journal of Botany* 91: 416-421.

Biodini, M.E., A.A. Steuter, and C.E. Grygiel. 1989. Seasonal fire effects on the diversity patterns, spatial distribution and community structure of forbs in northern mixed prairie, USA. *Vegetatio* 85: 21-31.

Bradstock, R.A., and T.D. Auld. 1995. Soil temperatures during experimental brushfires in relation to fire intensity: consequences for legume germination and fire management in south-eastern Australia. *Journal of Applied Ecology* 32: 76-84.

Carter, D.L., B.L. VanderWeide, and J.M. Blair. 2012. Drought-mediated stem and belowground bud dynamics in restored grasslands. *Applied Vegetation Science* 15: 470-478.

Cui, M., and M.M. Caldwell. 1997. Shading reduces exploitation of soil nitrate and phosphate by Agropyron desertorum and Artemisia tridentata from soils with patchy and uniform nutrient distribution. *Oecologia* 109: 177-183.

Dalgleish, H.J., and D.C. Hartnett. 2006. Below-ground bud banks increase along a precipitation gradient of the North American Great Plains: a test of the meristem limitation hypothesis. *New Phytologist* 171: 81-89.

Dalgleish, H.J., and D.C. Hartnett. 2009. The effects of fire frequency and grazing on tallgrass prairie productivity and plant composition are mediated through bud bank demography. *Plant Ecology* 201: 411-420.

Dix, R.L. 1960. The effects of burning on the mulch structure and species composition of grasslands in western North Dakota. *Ecology* 41: 49-56.

Devitt, M.L., and J.P. Stafstrom. 1995. Nucleotide sequence of four ribosomal protein L27 cDNAs from growing axillary buds of pea. *Plant Physiology* 107: 1031-1032.

Engle, D.M., and P.M. Bultsma. 1984. Burning of northern mixed-prairie during drought. *Journal of Range Management* 37:398-401.

Engle, D.M., R.L. Mitchell, and R.L. Stevens. 1998. Late growing-season fire effects in mid-successional tallgrass prairies. *Journal of Range Management* 51: 115-121.

Enquist, B.G., J.H. Brown, and G.B. West. 1998. Allometric scaling of plant energetic and population density. *Nature* 395: 163-165.

Erichsen-Arychuk, C., E.W. Bork, and A.W. Bailey 2002. Northern dry mixed prairie responses to summer wildlife and drought. *Journal of Range Management* 55: 164-170.

Ewing, A.L., and D.M. Engle. 1988. Effects of late summer fire on tallgrass prairie microclimate and community composition. *American Midland Naturalist* 120: 212-223.

Haile, K.F. 2011. Fuel load and heat effects on northern mixed prairie and four prominent rangeland graminoids [thesis]. Bozeman, MT, USA: Montana State University. 81 p.

Hartnett, D.C., and K.H. Keeler. 1995. Population processes. *In*: A. Joern, K.H. Keeler [EDS.]. The changing prairie: North American grasslands. New York, New York: Oxford University Press. p. 82-99.

Heisler, J.L., J.M. Briggs, and A.K. Knapp. 2003. Long-term patterns of shrub expansion in a C4-dominanted grassland: Fire frequency and the dynamics of shrub cover and abundance. *American Journal of Botany* 90: 423-428.

Hendrickson, J.R. and D.D. Briske. 1997. Axillary bud banks of two semiarid perennial grasses: occurrence, longevity, and contribution to population persistence. *Oecologia* 110: 584-591.

Higgins, K.F. 1984. Lightening fires in North Dakota grasslands and in pine-savanna lands of South

    Dakota and Montana. *Journal of Range Management* 37: 100-103.

Klimešová, J., and L. Klimeš. 2007. Bud banks and their role in vegetative regeneration – A literature

    review and proposal for simple classification and assessment. *Perspectives in Plant Ecology,*

    Evolution and Systematics 8: 115-129.

Knapp, A.K., S.L. Conrad, and J.M. Blair. 1998. Determinants of soil CO2 flux from a sub-humid

    grassland: Effect of fire and fire history. *Ecological Applications* 8: 760-770.

Lehtila, K. 2000. Modeling compensatory regrowth with bud dormancy and gradual activation of buds.

    Evolutionary Ecology 14: 315-330.

Littell, R.C., G.A. Milliken, W.W. Stroup, R.D. Wolfinger, and O. Schabenberger. 2006. SAS[®] for Mixed

    Models, second edition. Cary, NC: SAS Institute Inc

Morgan, J.W., and I.D. Lunt. 1999. Effects of time-since-fire on tussock dynamics of a dominate grass

    (*Themeda triandra*) in a temperate Australian grassland. *Biological Conservation* 88: 379-386.

Olson, B.E., and J.H. Richards. 1998. Annual replacement of the tillers of *Agropyron desertorum* following

    grazing. *Oecologia* 76: 1-6.

Ott, J.P. and D.C. Hartnett. 2011. Bud production and dynamics of flowering and vegetative tillers in

    Andropogon gerardii (*Poaceae*): the role of developmental constraints. *American Journal of*

    Botany 98: 1293-1298.

Ott, J.P., and D.C. Hartnett. 2012. Contrasting bud bank dynamics of two co-occurring grasses in

    tallgrass prairie: implications for grassland dynamics. *Plant Ecology* 213: 1437-1448.

Pausas, J.G. and D. Schwilk. 2012. Fire and plant evolution. *New Phytologist* 193: 301-303.

Precht, H., J. Christophersen, H. Hensel, and W. Larcher. 1973. Temperature and life. Springer-Verlag,

    Heidelberg, Germany. 87 p.

Raunkiaer, C. 1934. The life forms of plants and statistical plant geography. London, United Kingdom:

    Clarendon Press. 632 p.

Redmann, R.E., J.T. Romo and B.Plypec. 1993. Impacts of buring on primary productivity of *Festuca* and

    Stipa-Agropyron grasslands in central Saskatchewan. *American Midland Naturalist* 130: 262-273.

Rogers, W.E., and D.C. Hartnett. 2001. Vegetation response to different spatial patterns of soil

    disturbances in burned and unburned tallgrass prairie. *Plant Ecology* 155: 99-109.

Russell, M.L., L.T. Vermeire, N.A. Dufek, and D.J. Strong. 2013. Fire, defoliation, and competing species

    alter *Aristida purpurea* biomass, tiller, and axillary bud production. *Rangeland Ecology and*

    Management 66: 290-296.

Shay, J., D. Kunex, B. Dyck. 2001. Short-term effects of fire frequency on vegetation compostion and

    biomass in mixed prairie in south-western Manitoba. *Plant Ecology* 155: 157-167.

Shimizu, S.S., and H. Mori. 1998. Changes in protein interactions of cell cycle-related genes during the

    dormancy-to-growth transition in pea axillary buds. *Plant Cell Physiology* 10: 1073-1079.

Shimizu, S.S., and H. Mori. 2001. Control of outgrowth and dormancy in axillary buds. *Plant Physiology*

    127: 1405-1413.

Stafstrom, J.P., and I.M. Sussex. 1992. Expression of a ribosomal protein gene in axillary buds of pea

    seedlings. *Plant Physiology* 100: 1494-1502.

Steuter, A. A. 1987. C_3/C_4 composition shift on seasonal burns: northern mixed prairie. *Journal of Range*

    Management 40: 27-31.

Svejcar, T.J. 1990. Response of *Andropogon gerardii* to fire in the tallgrass prairie. *In*: S.L. Collins, and

    L.L. Wallace [EDS.]. Fire in North American tallgrass prairies. Norman, OK, USA: University of

    Oklahoma Press. p. 19-27.

Tomlinson, K.W., and T.G. O'Connor. 2004. Control of tiller recruitment in bunchgrasses: uniting

    physiology and ecology. *Functional Ecology* 18: 489-496.

Umbanhowar, C.E., Jr. 1996. Recent fire history of the northern Great Plains. *American Midland*

    Naturalist 135: 115-121.

USDA, NRCS. 2013. The PLANTS database. Available at: http://plants.usda.gov. Accessed 7

    September 2013.

Vermeire, L. T., J. L. Crowder, and D. B. Wester. 2011. Plant community and soil environment response

    to summer fire in the northern Great Plains. *Rangeland Ecology and Management* 64:37-46.

Vermeire, L.T., R.K. Heitschmidt, and M.R. Haferkamp. 2008. Vegetation response to seven grazing treatments in the northern Great Plains. *Agriculture, Ecosystems, and Environment* 127: 66-72.

Vermeire, L.T., R.K. Heitschmidt, and M.J. Rinella. 2009. Primary productivity and precipitation use efficiency in mixed-grass prairie: a comparison of Northern and Southern U.S. Sites. *Rangeland Ecology and Management* 62: 230-239.

Western Regional Climate Center. 2013. Western U.S. climate historical summaries. Available at http://www.wrcc.dri.edu/cgi-bin/cliMAIN.pl?mt5690. Accessed 22 August 2013.

White, R.S., and P.O. Currie. 1983. Prescribed burning in the northern Great Plains: yield and cover response of 3 forage species in the mixed grass prairie. *Journal of Range Management* 36: 179-183.

Wiles, L.J., G. Dunn, J. Printz, B. Patton, and A. Nyren. 2011. Spring precipitation as a predictor for peak standing crop of mixed-grass prairie. *Rangeland Ecology Management.* 64: 215-222.

Willems, J.H. 1983. Species compostion and aboveground phytomass in chalk grassland with different management. *Plant Ecology* 53: 171-180.

Wright, H.A. 1971. Why squirreltail is more tolerant to burning than needle-and-thread. *Journal of Range Management.* 24: 277-284.

Wright, H. A., and A. W. Bailey. 1982. Fire ecology: United States and southern Canada. New York, NY, USA: John Wiley & Sons. 501 p.

CHAPTER 3. THE ROLE OF FIRE RETURN INTERVAL, SEASON OF FIRE, AND DROUGHT ON BUD BANK
DYNAMICS

Abstract

Despite the importance of vegetative reproduction in year-to-year replacement of tillers, little is
known about the patterns and timing of tiller recruitment from the bud bank, especially regarding fire
return intervals, seasons of fire, and drought. This study examined aboveground plant density, temporal
patterns of tiller production, and belowground bud bank dynamics for *Bouteloua gracilis, Pascopyrum
smithii, and Hesperostipa comata* following summer, fall, and spring prescribed fires and 1 yr, 3 yr, and 6
yr fire return intervals, and their interactions. Fire treatments were initiated in 2006 and buds were
assessed July 2011 through July 2013. Density and number of reproductive *B. gracilis* tillers increased in
2013 following drought during 2012, unlike *H. comata*, which decreased reproductive tiller production. In
general (irrespective of fire treatments), *B. gracilis* produced the most buds (8-10 buds · tiller^{-1}) and *H.
comata* produced the least (2-3 buds · tiller^{-1}), with *P. smithii* producing an intermediate amount (6-8
buds · tiller^{-1}). All seasons of fire and fire return intervals stimulated immediate post-fire bud activity and
decreased dormancy of *B. gracilis* and *P. smithii* within hours following fire. Bud mortality of *B. gracilis*
and *P. smithii* did not differ between immediate pre- and post-fire assessments. However, *H. comata* bud
mortality increased immediately following summer and fall prescribed fires. Three-year fire return
intervals increased active buds throughout the 2013 winter and growing season for *B. gracilis* and *P.
smithii* relative to control plots and annual and 6 yr fire return intervals. Fire stimulated bud activity of *B.
gracilis* and *P. smithii* relative to non-burned plots. The above and belowground response of *H. comata*
indicated meristem limitations following fire treatments, illustrating greater vulnerability to fire for that
species than *B. gracilis* and *P. smithii*.

Keywords: dormancy, grassland, meristem limitation, prescribed burning, tiller, vegetative
reproduction

Introduction

Vegetative reproduction is the primary driver of aboveground production and growth, producing more than 99% of new tiller growth (Benson and Hartnett 2006). Bud banks play an important role in aboveground vegetation dynamics in grassland systems by serving as the primary source of recruitment for new tiller formation. Utilizing bud banks, perennial grasses are able to respond rapidly with tiller production, even during fluctuating environmental conditions. For example, bud banks mediated effects of grazing, drought, fire and other stresses on productivity of grassland dominated by perennial grasses (Benson et al. 2004; Benson and Hartnett 2006; Dalgleish and Hartnett 2009).

Dense bud banks provide a safeguard for grassland populations in response to aboveground disturbances such as, grazing, drought, fire, or a combination of these grassland processes (Dalgleish and Hartnett 2009). In systems with less dense bud banks, meristem limitations are revealed both above and below ground through decreased tillering and recruitment from the bud bank (Dalgleish and Hartnett 2009). Periodic fire, grazing, and variable precipitation are important factors governing grassland ecosystems, having large impacts on individual species and plant community structure and function (Fuhlendorf and Engle 2004; Archibald et al. 2005; Castellano and Ansley 2007).

Despite the importance of vegetative reproduction in year-to-year replacement of tillers, little is known about the patterns and timing of tiller recruitment from the bud bank following different fire return intervals and seasons of fire. Because fire intensity and plant physiology vary with season and frequency of fire (Collins 1992; Govender et al. 2006), plant communities can respond differently to seasonal timing of fire (Dix 1960; Steuter 1987; Copeland et al. 2002). In tallgrass prairie, the presence of fire increased bud bank density of perennial grasses compared to non-burned plots (Benson et al. 2004 and Dalgleish and Hartnett 2009). However, it is unclear if northern mixed prairie responses to fire and drought depend on the size of bud banks to mediate disturbance effects. Furthermore, previous fire ecology research in northern mixed-grass prairie typically measured aboveground vegetation response one to two years post-fire and did not include different seasons of fire and their interaction with fire return intervals (Engle and Bidwell 2001; Castellano and Ansley 2007; Vermeire et al. 2011).

In semiarid grasslands, such as northern mixed-grass prairie in the western Great Plains, variability in precipitation has a large effect on grass production (Vermeire et al. 2009; Wiles et al. 2011), presumably by reducing tiller recruitment from the bud bank (Dalgleish and Hartnett 2006; Carter et al. 2012). In these systems, year-to-year variability in precipitation can have a greater effect on annual production than fire (Ansley et al. 2006). Tallgrass prairie bud bank densities increased more than 50% the second year following drought compared to non-drought controls (Carter et al. 2012). Even though the combined effects of fire and drought are common in the northern Great Plains, the level to which they influence bud banks is unknown.

An improved understanding of native, dominant, grassland species responses to the timing of fire and fire return interval may elucidate mechanisms governing population processes and the potential for future tiller recruitment. Determining the links between formation, development, and maintenance of belowground meristems and aboveground processes will help explain annual growth cycles and identify periods of bud bank vulnerability and resistance (Dalgleish et al. 2008).

This study utilized three growing seasons of post-fire belowground and aboveground data to examine the effects of summer, fall, spring, and no-fire paired with fire return intervals of 1, 3, and 6 yr in a factorial design on above and belowground dynamics. A C_4 rhizomatous/caespitose grass (*Bouteloua gracilis* (Willd *ex*. Kunth) Lag. *ex* Griffiths), C_3 rhizomatous grass (*Pascopryon smithii* (Rydb.) A. Löve), and C_3 caespitose grass (*Hesprostipa comata* (Trin. & Rupr.) Barkworth) were selected for study as dominant, native, perennial grasses in northern mixed prairie of the western Great Plains. Additionally, a severe drought during the second year of the study allowed examination of each species' post-drought bud bank response in the third year of the study. The objectives of this study were to quantify: (1) fire seasonality and return interval and potential interacting effects on belowground bud banks and tiller emergence; and (2) bud bank dynamics, tiller emergence, and plant density response to interactions of fire with drought. By using summer, fall, and spring fires paired with varying fire return intervals, we expected bud banks of different photosynthetic pathways to vary in magnitude and timing of tiller recruitment from the bud bank. We hypothesized that the presence of fire would increase the capacity of

species' bud banks to mediate effects of subsequent fire. We expected that fall fires would stimulate *P. smithii* bud growth and spring fires would enhance *B. gracilis* bud growth. We also hypothesized that *H. comata* bud banks would decrease in productivity illustrating bud bank vulnerability (Wright 1971).

Materials and methods

Study site

Research was conducted in semi-arid mixed-grass prairie at USDA-ARS Fort Keogh Livestock and Range Research Laboratory near Miles City, MT (lat 46°24′N, long 105°56′W; 815 m elevation) from July 2011 to July 2013. Average annual precipitation for the area is 339 mm, with the majority occurring April through September. Precipitation effects on annual biomass production are typically greatest during April and May (Vermeire et al. 2008; Wiles et al. 2011). Average daily temperatures range from 23 °C in July to -8 °C in January and the frost-free growing season generally ranges from 125 - 150 days (Western Regional Climate Center, Reno, NV, 2013).

The research area consists of native rangeland on upland plains with minimal slopes (0 – 4%). The study site is dominated by Pinehill loams (fine, smectitic, frigid Aridic Haplustalfs) including a complex of Kobase clay loams (fine, smectitic, frigid Torrertic Haplustepts) and Gerdrum clay loams (fine, smectitic, frigid Torrertic Natrustalfs) (USDA, NRCS Ecological Site Descriptions, 2013).

Vegetation is dominated by perennial, native C_3 species including, *H. comata*, *P. smithii*, and threadleaf sedge (*Carex filifolia* Nutt. var. *filifolia*) and C_4 species, *B. gracilis*, and to a lesser extent, buffalograss (*Bouteloua dactyloides* (Nutt.) J.T. Columbus). Annual grasses include six week fescue (*Vulpia octoflora* (Walter) Rydb.), field brome (*Bromus arvensis* L.), and cheatgrass (*B. tectorum L.*). The primary shrub species on the site is Wyoming big sagebrush (*Artemisia tridentata* Nutt. Subsp. *wyomingensis* Beetle & Young) and half shrub fringe sage (*A. frigida* Willd.). The perennial legume, silverleaf Indian breadroot (*Pediomelum argophyllum* (Pursh.) J. Grimes) and the biennial forb, yellow salsify (*Tragopogon dubius* Scop.) were also present. Annual forbs included field cottonrose (*Logfia arvensis* (L.) Holub), woolly plantain (*Plantago patagonica* Jacq.), and rough false pennyroyal (*Hedeoma*

hispida Pursh.). Plant nomenclature follows the USDA PLANTS database (USDA, NRCS; Plants Database 2013)

Fire measurements

Fire treatments consisted of summer, fall, and spring fire with annual, 3, and 6 yr return interval treatments in a factorial design. Each treatment was replicated three times on 15 × 20-m plots for a total of 36 plots and all fires were set using the ring-fire method (Wright and Bailey 1982). Fire treatments were initiated in 2006 and bud collection started during the fall of 2010. Summer fires were applied following quiescence of *H. comata*, and *P. smithii*. Annual fires for summer, fall, and spring were applied on 7 September, 3 November, and 15 March 2011, respectively. Annual fires were lit on the same dates during 2012 as 3 and 6 yr burns. However, annual fires did not carry through the plots during 2012 because drought and the annual fire regime limited fuel mass and continuity. The 3 and 6 yr fire return interval treatments were applied 16 August 2012 with ambient temperatures 30-34°C, winds 7-10 km $\cdot$ h^{-1}, and relative humidity 30-36%; after the first killing frost on 19 October 2012 with ambient temperatures 18-24°C, winds 8-12 km $\cdot$ h^{-1}, and relative humidity 37-41%; and when *B. gracilis* initiated aboveground growth 6 May 2013 with ambient temperatures 20-25 °C, winds 13-20 km $\cdot$ h^{-1}, and relative humidity 39-43%.

We utilized HOBO$^{®}$ U12 J, K, S, T Thermocouple Data Loggers (Onset Computer Corporation, Bourne, MA) with K-type Thermocouples (Omega Engineering, Inc., Stanford, CT) to create time-temperature profiles at the plot level (Table 3.1). Thermocouples were placed at the base of a plant's crown within 1-2 cm above the soil surface in a 10 × 10-m square within each plot (4 thermocouples $\cdot$ plot $^{-1}$) and were programmed to record temperatures at one-second intervals. Maximum temperature was identified by finding the greatest value for each time-temperature profile. Heat duration was calculated as time (seconds) of heat greater than 60 °C and heat dosage was calculated as the sum of the temperatures exceeding a base temperature of 60 °C (degree-seconds). These measurements were used to derive the mean maximum temperature, heat duration, and dosage of heat imposed at the plot level. Fire behavior measurements were taken during the study years 2011, 2012, and 2013.

Table 3.1. Thermocouple measurements at the plot level for summer, fall, and spring fires at 1, 3, and 6 yr fire return intervals near Miles City, MT.

Summer fire[1]	1 yr	3 yr	6 yr
Maximum temperature (C)	240±10	287±12	306±25
Heat duration (s)	297±16	884±13	438±8
Heat dosage (C · s)	10381±569	27774±621	16878±718
Fall fire			
Maximum temperature (C)	225±8	240±16	261±11
Heat duration (s)	135±16	162±14	214±13
Heat dosage (C · s)	12891±426	13428±589	13775±646
Spring fire			
Maximum temperature (C)	206±12	266±16	186±10
Heat duration (s)	268±5	83±9	100±11
Heat dosage (C · s)	2789±457	5744±368	4837±540

[1] Heat duration and dosage were assessed using 60 °C as a base temperature. Heat duration was calculated as time (seconds) of heat greater than 60 °C and heat dosage was the sum of the degrees > 60 °C for each second (degree-seconds).

Field sampling and lab methods

Plant density estimates were performed at the end of the growing season for 2011, 2012, and 2013. Plant density was measured by counting individual plants rooted within a 0.25-m^{-2} quadrat. *B. gracilis* was considered an individual when gaps of 3-cm or greater were separating neighboring *B. gracilis* tillers. Each *P. smithii* tiller with a unique point of origin from the soil within a 0.25-m^{-2} quadrat was considered an individual plant. Individual *H. comata* used in plant density measurements were readily distinguished due to their bunchgrass growth form. Tiller counts were performed at the end of 2011, 2012, and 2013 growing seasons on permanently marked individual plants with individuals defined as they were for density counts. Tillers were classified as vegetative or reproductive for one individual plant.

Tillers were harvested from two individuals per plot for each target species immediate before and after fire, and seasonally for two years. Tillers were harvested from randomly selected plant individuals within a 10 × 10-m square within the plot, 12 to 24 hours before and following prescribed burns to determine immediate fire effects on bud activity and survival. Therefore, tillers assessed for immediate bud response following annual summer, fall, and spring burns were harvested on 7-8 September 2011, 3-4 November 2011, 28-29 March 2012, and 15-16 August 2012, 19-20 October 2012, and 6-7 May 2013 (3

and 6 yr interval burns). To address seasonal fluctuations for each species' bud bank, two individuals from each plot and target species were sampled 30 July 2011, 15 March 2012, 18 July 2012, 3 November 2012, 9 January 2013, 24 February 2012, 25 March 2013, 1 May 2013 and 22 July 2013.

Buds were counted and classified using a dissecting microscope and assessed to be living or dead. Number of active buds, dormant buds, and dead buds were confirmed using Tetrazolium and Evans Blue staining procedures. Tillers were submerged in Tetrazolium solution at room temperature for 24-hr following initial classification. Active buds stained pink and dormant and dead buds retained their white or yellowish color. If inactive buds were present, the tiller was submerged into Evan's Blue solution at room temperature following the Tetrazolium staining for 20-min. Upon completion of Evan's Blue staining, dormant buds would maintain their white pigment, whereas, dead buds stained dark blue.

Statistical analysis

Data were analyzed using analysis of variance (MIXED procedure of SAS, Littell et al. 2006) to quantify aboveground plant response throughout three growing seasons following summer, fall, and spring fire at annual, 3, and 6 yr fire return intervals. The model included season of fire, fire return interval, year, and all interactions as fixed effects. Vegetative tillers, reproductive tillers, and plant density (# of individuals within a 0.25-m^{-2} quadrat) by species were used as response variables and the experimental unit was plot.

Immediate bud response was assessed following all fire treatments. Data was analyzed using analysis of variance (MIXED procedure of SAS, Littell et al. 2006). The model included season of fire, fire return interval, and their interaction as fixed effects. However, comparisons were only made within the same season of burning to avoid confounding of seasonal and fire seasonality effects. Active, dormant and dead buds by species were used as response variables with plot as the experimental unit.

Bud response following fire treatments were examined during four years (October 2010- July 2013). Data were analyzed using analysis of variance (MIXED procedure of SAS, Littell et al. 2006) with sampling date as a repeated measure. The model included sampling date, season of fire, fire return interval and their interactions. Response variables were active, dormant and dead buds by species. For

59

all analyses, values from non-burned controls were subtracted from fire treatments and the difference was used in the models. Statistical significance was set at $P < 0.05$ for all models.

Results

Growing conditions during the spring of 2011 (April –June) were extremely wet, with a record amount of precipitation (250% of average; Fig. 3.1). The following winter, spring, and summer were dry with drought conditions occurring most of the spring and summer (34% below average). Spring 2012 was the second driest on record and drought conditions persisted throughout eastern MT and the northern Great Plains (Western Regional Climate Center, Reno, NV, 2013). Spring of 2013 (two growing seasons post-fire) brought near-average spring and summer precipitation enabling growth of cool- and warm-season grasses following severe drought of 2011-2012. Spring precipitation (April – June) is considered to be a major driver of aboveground productivity in the northern Great Plains (Heitschmidt and Vermeire 2005; Wiles et al. 2011). Because time since fire varied throughout the study depending on fire return interval treatments accumulations of fuel based on above-average precipitation years (e.g., 2011) may have resulted in differences among fire treatments and years.
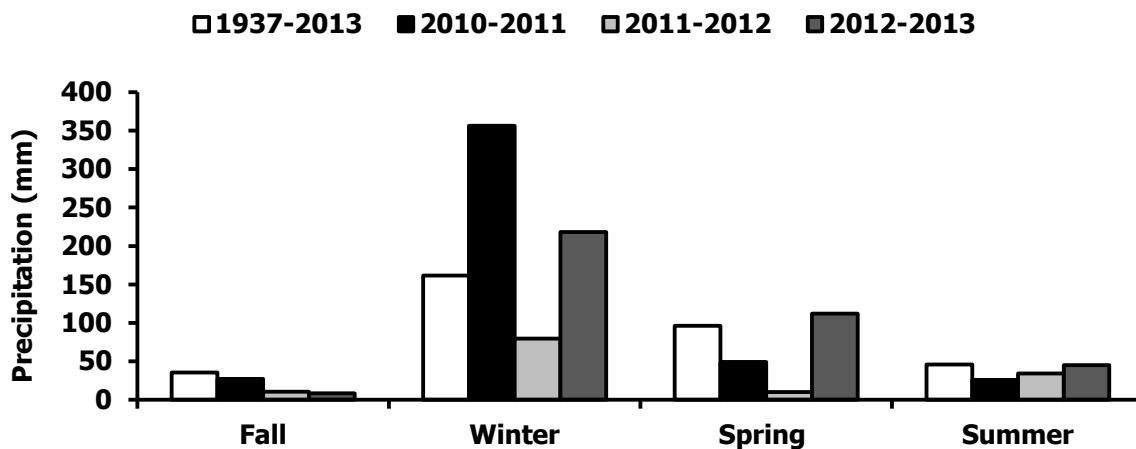
Figure 3.1. Study site 76 yr mean seasonal precipitation, fall (Oct-Dec), winter (Jan-Mar), spring (Apr-Jun), and summer (Jul-Sep) precipitation year (Oct-Sep) near Miles City, MT.

Aboveground response

Plant densities on non-burned plots were similar among years for *B. gracilis* (P = 0.21) and *H. comata* (P = 0.26), but *P. smithii* density decreased during the 2012 drought (P < 0.01; Fig. 3.2). Density of *B. grama* varied among years on burned plots nearly doubling by the end of the study on burned plots compared to nonburned controls (6, 4, 11 vs. 15 ± 3 plants m^{-2}; P < 0.05).
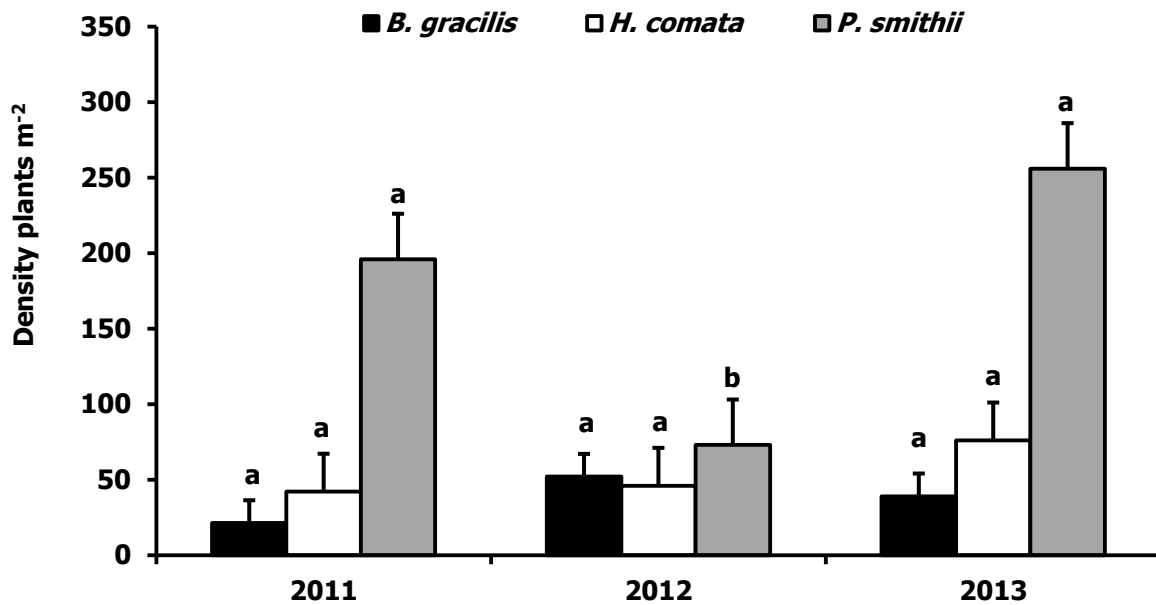
Figure 3.2. Plant density of *B. gracilis*, *H. comata*, and *P. smithii* near Miles City, MT on non-burned plots (+ SEM). Means marked with same letter within species are similar (P < 0.05).

In contrast, *H. comata* density decreased steadily each year (5, 4, 2 ± 1 plants m^{-2}; P < 0.01). Annual fire reduced density of *H. comata* (3 ± 1 plants m^{-2}; P < 0.01) compared to 3 and 6 yr fire return intervals (5 and 10 ± 2 plants m^{-2}). Fire return interval and year interacted in their effects on *P. smithii* density plants m^{-2} (P < 0.01; Fig. 3.3). *P. smithii* density more than tripled during the driest year on annual and 3 yr fire return intervals relative to controls.
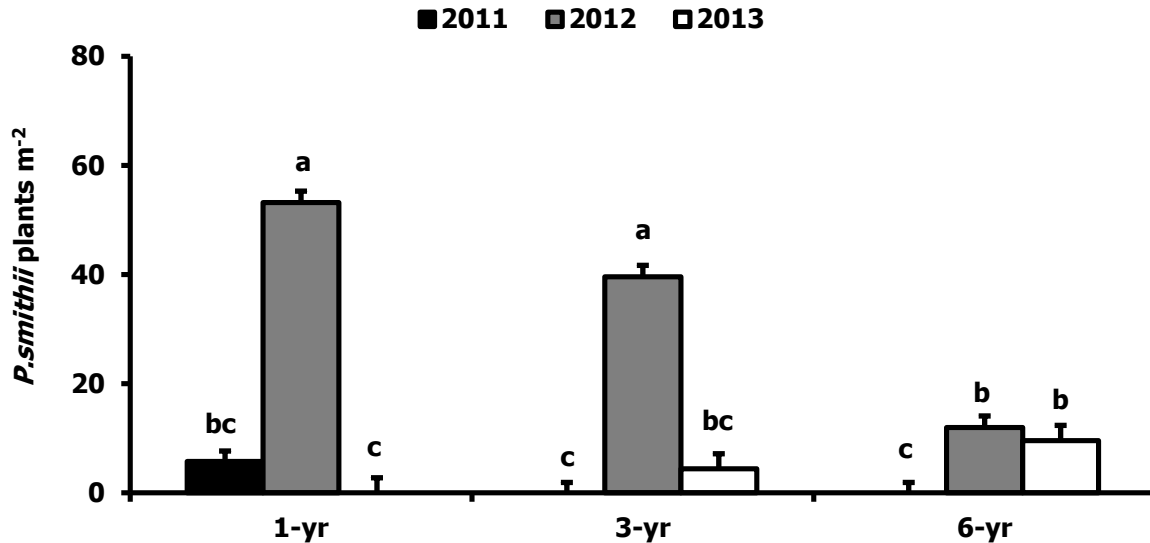
Figure 3.3. Plant density of _P. smithii_ individuals m^{-2} following 1 yr, 3 yr, and 6 yr fire return intervals near Miles City, MT for study years 2011, 2012, 2013 (+ SEM). Means are the difference between fire treatments and controls with no fire. Means marked with same letter are similar (_P_ < 0.05).

Vegetative tillers of _B. gracilis_ and _H. comata_ varied among years on non-burned plots (_P_ < 0.05; Fig. 3.4). However, reproductive tillers of each species on non-burned plots were similar among years (_P_ = 0.56; Fig. 3.4). Vegetative _B. gracilis_ tillers plant^{-1} were similar among fire treatments and years (3 ± 6 plant^{-1}; _P_ =0.23).
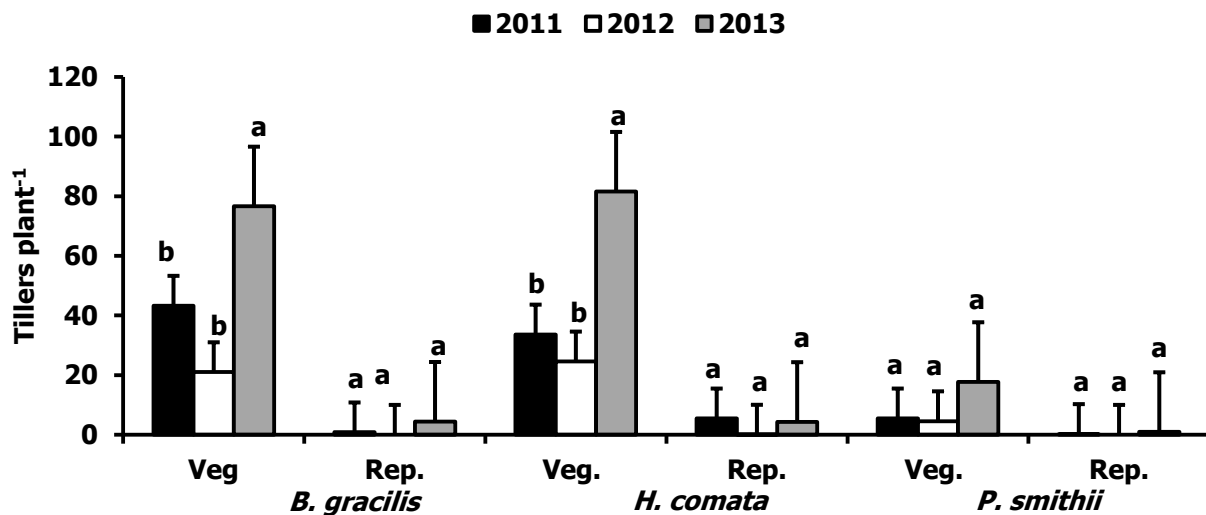
Figure 3.4. _B. gracilis, H. comata_, and _P. smithii_ vegetative and reproductive tillers plant^{-1} on non-burned control plots near Miles City, MT (+SEM). Means marked with the same letter within tiller classification and species are similar (_P_ < 0.05).

B. gracilis reproductive tillers plant[-1] were similar throughout 2011 and 2012; however, in 2013, spring burns every three years tripled reproductive tillers plant[-1] compared to annual and 6 yr fire return intervals and non-burned plots ($P < 0.01$; Table 3.2). Annual and 6 yr fall fires, and 3 and 6 yr summer fires also increased reproductive tillers plant[-1] compared to annual summer fires and non-burned plots.

Table 3.2. *B. gracilis* reproductive tillers plant[-1] and *P. smithii* vegetative tillers plant[-1] following 1 yr, 3 yr, and 6 yr fire return intervals and summer, fall, and spring fire treatments during 2011, 2012, and 2013[1] near Miles City, MT. Means are the difference between fire treatments and controls with no fire.

	2011			2012			2013				
	SU[2]	F	SP	SU	F	SP	SU	F	SP	SE	*P* - value
B. gracilis	----------------------reproductive tillers plant[-1]----------------------										
1 yr return interval	2 cd	0 d	0 d	0 d	0 d	1 cd	4 c	12 b	5 c	3	< 0.01
3 yr return interval	1 d	0 d	1 d	0 d	0 d	0 d	14 b	3 c	20 a		
6 yr return interval	0 d	0 d	0 d	0 d	0 d	0 d	11 b	14 b	6 c		
P. smithii	------------------------vegetative tillers plant[-1]----------------------										
1 yr return interval	2 c	0 cd	-3 d	14 a	10 ab	3 c	-5 d	-2 c	1 c	3	< 0.01
3 yr return interval	-3 d	-3 d	-3 d	8 b	8 b	5 b	8 b	0 c	-1 c		
6 yr return interval	-3 d	3 c	3 c	1 c	-1 c	0 c	9 b	-3 d	5 b		

[1]**Means marked with the same letter within species are similar (*P* < 0.05)**
[2] **SU = Summer fire; F = fall fire; SP = spring fire**

P. smithii vegetative tillers plant[-1] varied among season of fire, fire return interval and year ($P < 0.05$; Table 3.2). *P. smithii* vegetative tillers increased during the drought of 2012, especially on annual summer and fall burned plots relative to controls. By 2013, *P. smithii* vegetative tillers decreased slightly, producing more following 3 and 6 yr interval summer fires. *P. smithii* vegetative tiller growth was least during 2011 on 3 yr intervals for all seasons of fire relative to controls. *P. smithii* averaged 1 reproductive tiller plant[-1] more than non-burned plots during 2011 and 2012, but increased to 5 reproductive tillers plant[-1] during the growing season of 2013 ($P < 0.01$).

Vegetative tillers plant[-1] of *H. comata* depended on season of fire and fire return interval ($P < 0.05$; Fig. 3.5). Annual burns during the spring and summer 3 yr burns increased *H. comata* vegetative tillers plant[-1] relative to controls. However, 3 and 6 yr fire return intervals during the fall and spring reduced *H. comata* vegetative tillers plant[-1] relative to controls. Annual summer fires caused the greatest reduction in *H. comata* vegetative tillers plant[-1] relative to controls.
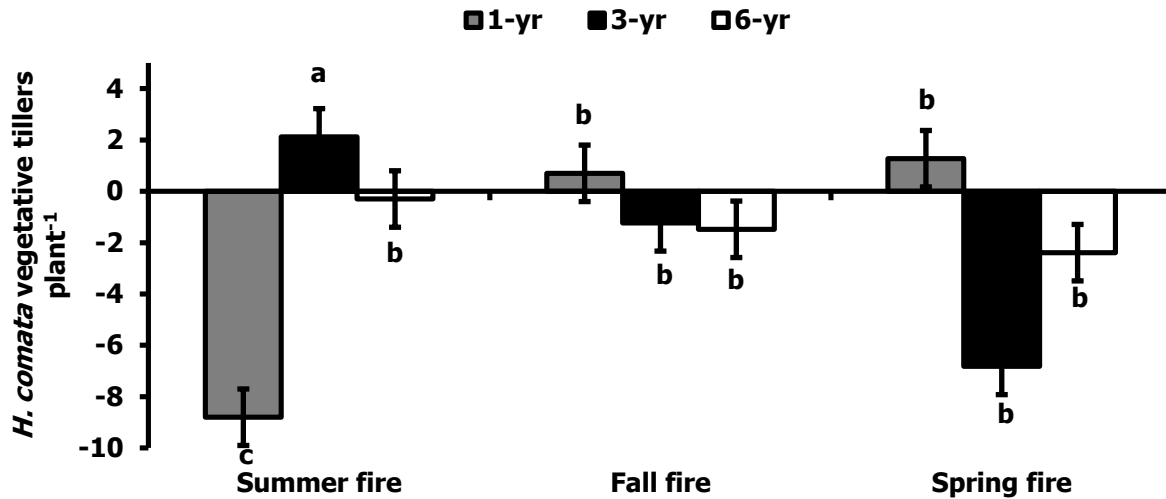
Figure 3.5. *H. comata* vegetative tillers plant^{-1} following summer, fall and spring fire with 1 yr, 3 yr, and 6 yr fire return intervals near Miles City, MT (± SEM). Means are the difference between fire treatments and controls with no fire. Means marked with same letter are similar (*P* < 0.05).

Season of fire and year interacted in their effects on reproductive tillers plant^{-1} of *H. comata* (*P* < 0.05; Fig. 3.6). During 2011, fall fire plots increased *H. comata* reproductive tillers plant^{-1} by 73% or more compared to other seasons of fire and non-burned plots. Summer and spring burned plots decreased reproductive tillers relative to the controls during 2013.
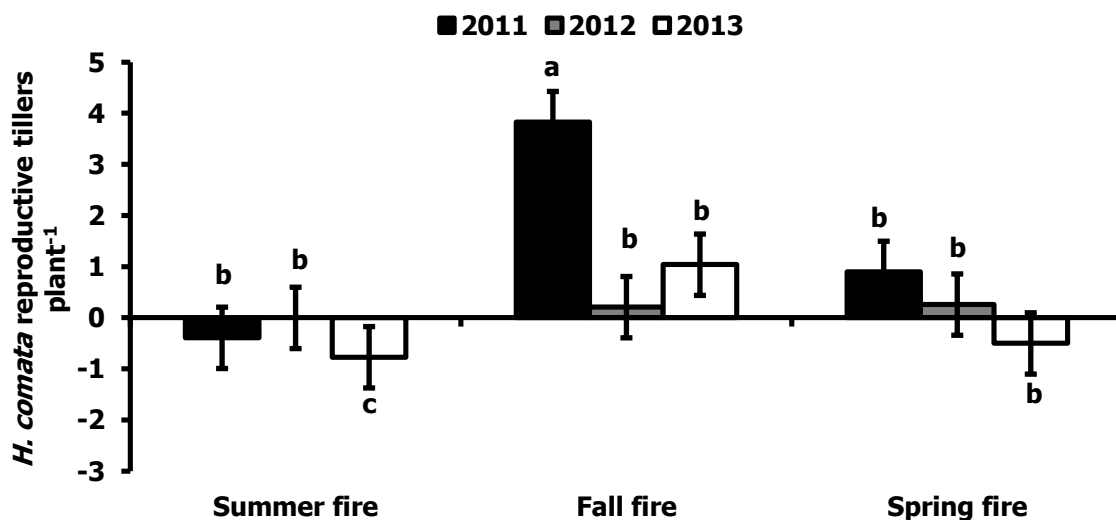
Figure 3.6. *H. comata* reproductive tillers plant^{-1} following summer, fall, and spring fire during 2011, 2012, and 2013 near Miles City, MT (± SEM). Means are the difference between fire treatments and controls with no fire. Means marked with same letter are similar (*P* < 0.05).

Immediate bud response

Active *B. gracilis* buds immediately increased 54% after spring burns at 3 yr fire return intervals (P < 0.01; Fig. 3.7). In contrast, fall fire at 3 and 6 yr fire return intervals immediately reduced active buds by 95%. Summer fire at 6 yr return intervals reduced active buds by 97%. However, annual summer, fall, and spring fire, 3 yr summer fire, and 6 yr spring fire maintained similar amounts of active buds pre-and post-fire.
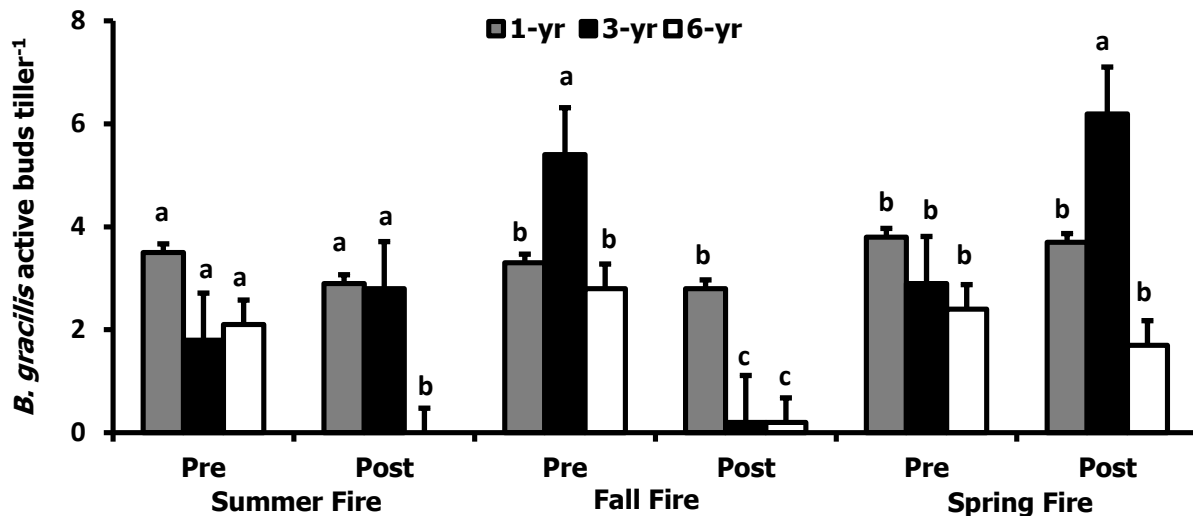
Figure 3.7. Active buds tiller^{-1} of *B. gracilis* immediately following seasonal fires with 1 yr, 3 yr, and 6 yr fire return intervals near Miles City, MT (+ SEM). Means are the difference between fire treatments and controls with no fire. Means marked with same letter within season of fire treatment are similar (*P* < 0.01).

Immediate *B. gracilis* bud mortality was similar following summer, fall, and spring fires (0.07, 0.04, 0.1 ± 0.07; *P* > 0.07) and 1, 3, and 6 yr fire return intervals (0.07, 0.21, and 0.26 ± 0.08; *P* > 0.21). Dormant *B. gracilis* buds decreased 50% immediately after spring 3 yr burns (*P* < 0.01; Fig. 3.8). However, dormant *B. gracilis* buds increased 35% within hours after summer fire at 6 yr return intervals. Fall fire burned at 3 yr return interval increased dormant buds the greatest by transitioning all of its buds into early winter dormancy. All other fire treatments were similar in immediate effects following fire treatments (*P* > 0.05).
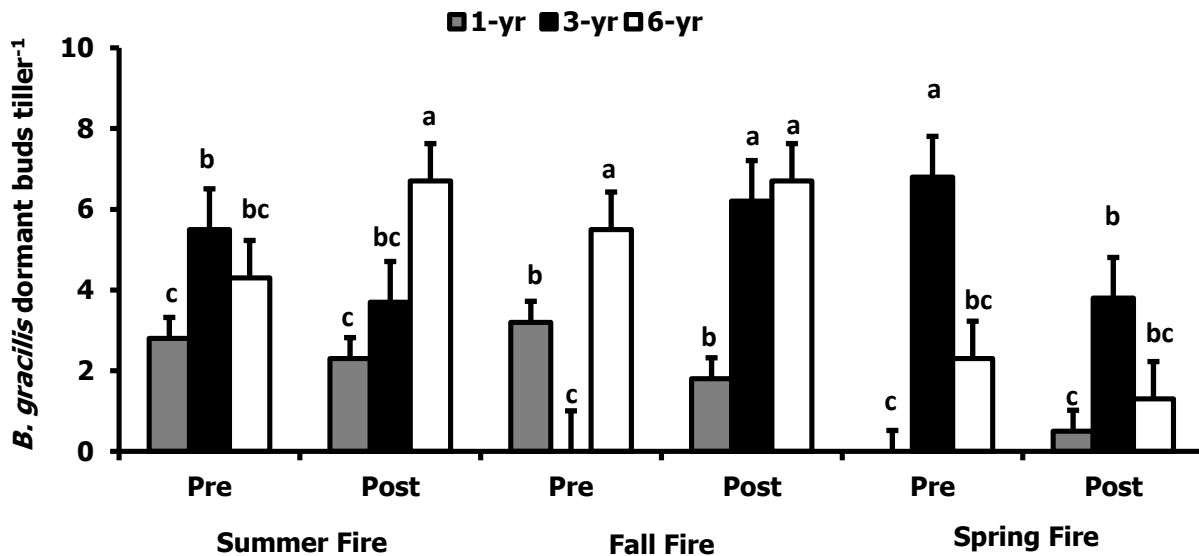
Figure 3.8 Dormant buds tiller^{-1} of *B. gracilis* following summer, spring, and fall fires paired with 1 yr, 3 yr, and 6 yr fire return intervals near Miles City, MT (+ SEM). Means are the difference between fire treatments and controls with no fire. Means marked with same letter within season of fire treatment are similar ($P < 0.05$).

Active and dormant buds of *H. comata* were similar to pre-burn assessments immediately following all fire treatment combinations ($P \geq 0.13$). However, *H. comata* bud mortality increased immediately following summer and fall fire ($P < 0.05$; Fig. 3.9). *H. comata* dead buds were minimal prior to spring fire and were similar to pre-burn assessments ($P > 0.85$).
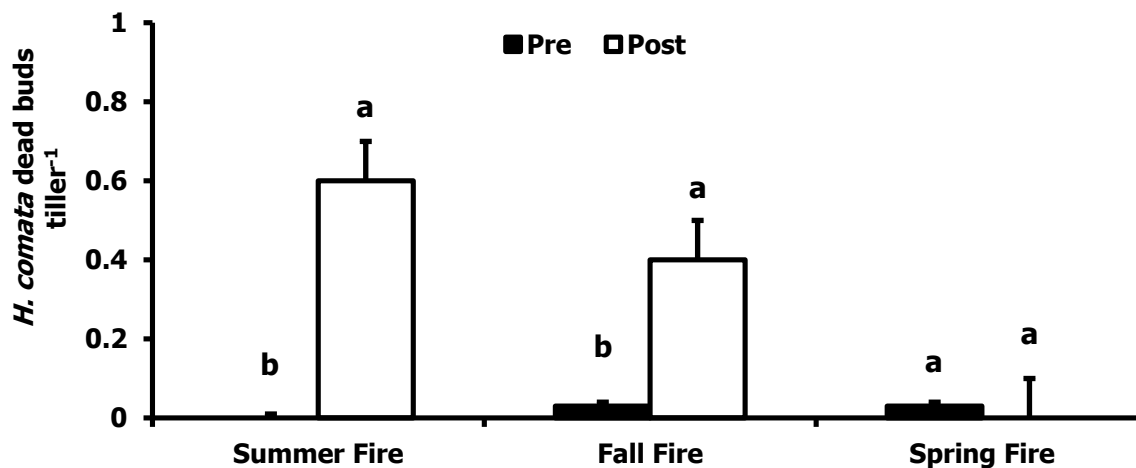
Figure 3.9. *H. comata* dead buds tiller^{-1} immediately following summer, spring, and fall fires near Miles City, MT (+ SEM). Means are the difference between fire treatments and controls with no fire. Means marked with same letter within season of fire treatment are similar ($P < 0.05$).

Active buds of *P. smithii* increased 56% following summer fire every 3 years ($P < 0.05$; Fig. 3.10). Fall fire at 3 yr intervals reduced active buds 52% and spring fire at 3 yr intervals reduced active buds 15%. *P. smithii* active buds were similar to pre-burn assessments for all other treatment combinations ($P > 0.05$). Dormant buds of *P. smithii* were not immediately altered by season of fire, fire return interval, or their interaction ($P \geq 0.08$). Immediate bud mortality did not occur for *P. smithii* buds ($P > 0.19$).
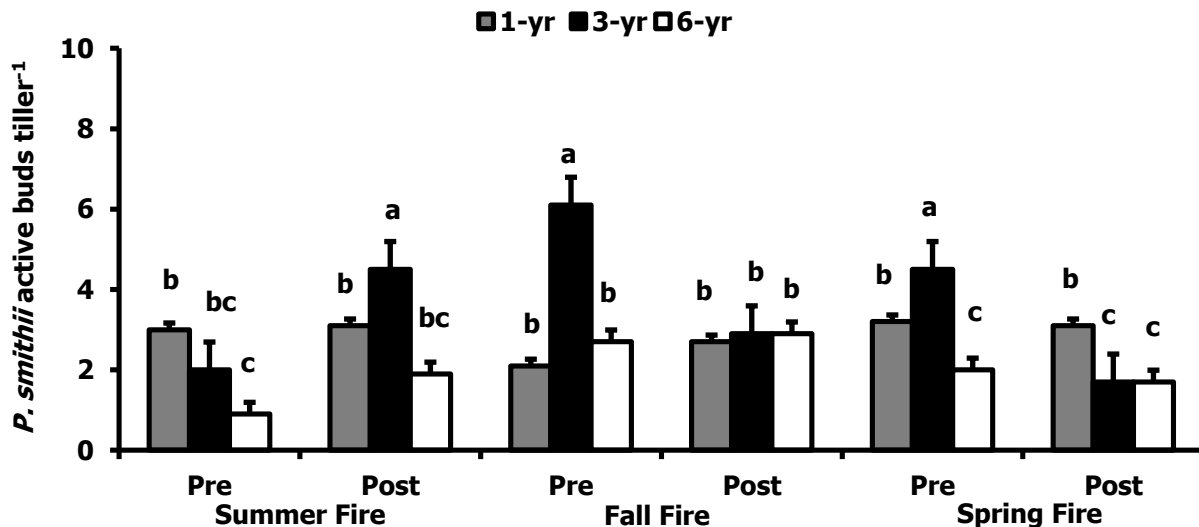
Figure 3.10. Active buds tiller^{-1} of *P. smithii* following summer, spring, and fall fires paired with 1 yr, 3 yr, and 6 yr fire return intervals near Miles City, MT (+ SEM). <eans are the difference between fire treatments and controls with no fire. Means marked with same letter within season of fire treatment are similar ($P < 0.05$).

Short-term bud response

Short-term seasonal fluctuations of bud banks varied according to species and sampling time on non-burned plots ($P < 0.01$; Fig. 3.11). Therefore, short-term bud bank responses to season of fire and fire return interval are discussed relative to non-burned controls. Active and dormant bud bank responses of each species varied throughout the study. Non-burned dormant bud banks of *B. gracilis* peaked during the winter; whereas, active buds peaked during the growing seasons. Active buds were consistently more abundant than dormant or dead buds for C_3 grasses.
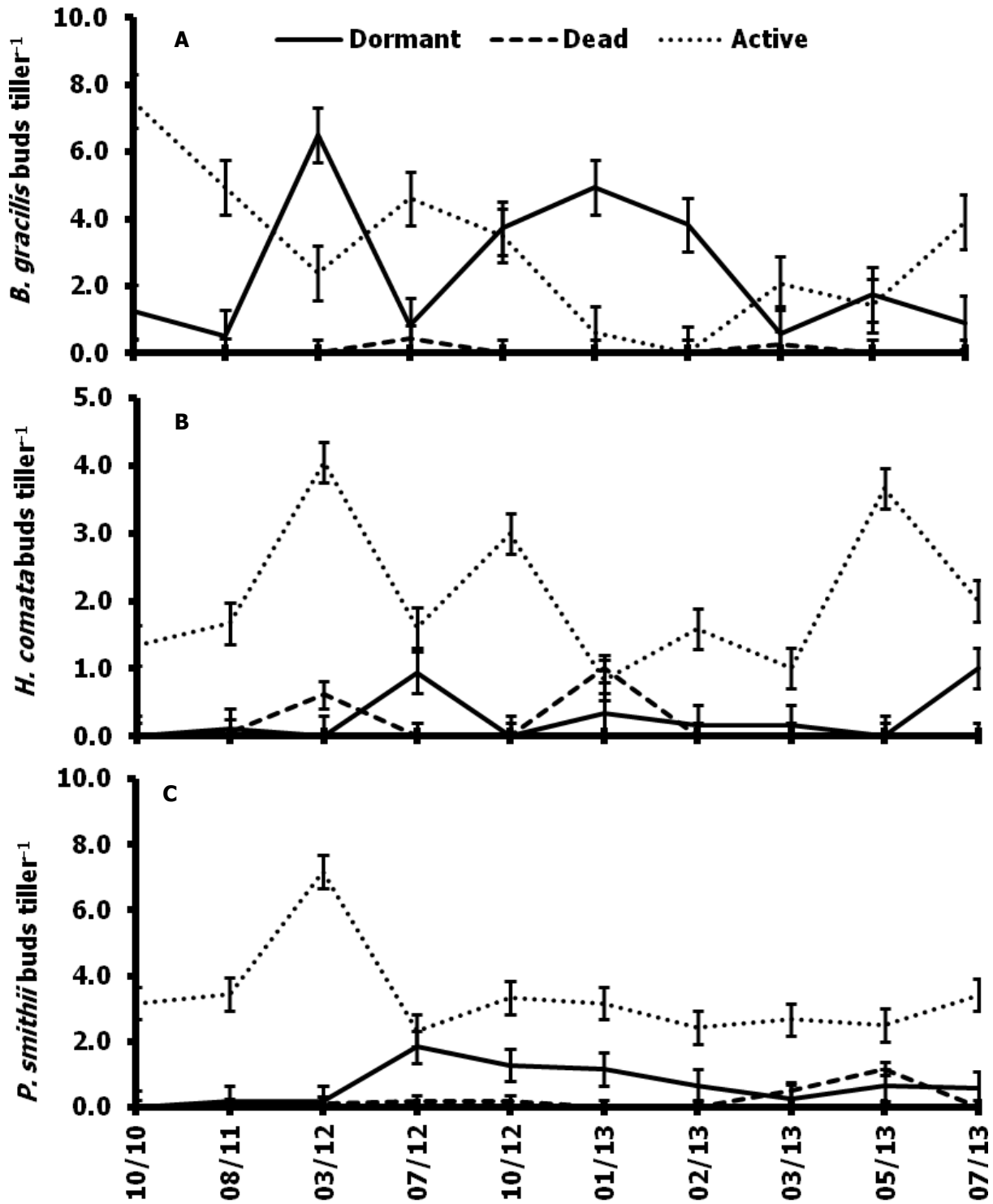
Figure 3.11. Short-term seasonal fluctuations of A) *B. gracilis*, B) *H. comata*, and C) *P. smithii* dormant, dead, and active buds on non-burned control plots near Miles City, MT (± SEM; *P* < 0.01).

There was a season of fire by sampling date interaction on active buds of *B. gracilis* ($P < 0.01$; Fig. 3.12A). Activity of *B. gracilis* buds increased following summer burns during January 2012; whereas, during the same time bud activity reached its' lowest point on fall burned plots relative to controls. Fall and spring burns manipulated *H. comata* bud activity similarly throughout the study years ($P \geq 0.18$). However, summer burns increased *H. comata* bud activity relative to controls from July 2012 to January 2013 more so than fall or spring fire treatments ($P < 0.01$; Fig. 3.12B). Active *H. comata* buds decreased sharply for all seasons of fire during the initiation of the 2013 growing season ($P > 0.12$) relative to controls. Season of fire and time of tiller harvest had interacting effects on bud activity for *P. smithii* ($P < 0.01$; Fig. 3.12C). Summer fire increased the 2013 overwintering *P. smithii* bud bank with maintained activity throughout the start of the 2013 growing season compared to fall and spring fire ($P < 0.01$) and control plots.

Fire return interval and sampling date interacted in their effects on active buds for *B. gracilis* ($P < 0.01$; Fig. 3.13A), *H. comata* ($P < 0.01$; Fig. 3.13B), and *P. smithii* ($P < 0.01$; Fig. 3.13C). Active buds were similar for *B. gracilis* and *H. comata* following all fire return intervals until October 2012. During winter 2012, annual and 3 yr return intervals maintained more active buds than controls for *B. gracilis* and *H. comata* until March 2013. At the beginning of the 2013 growing season, *B. gracilis* increased active buds following 3 yr return intervals and *H. comata* decreased active buds relative to controls. Three-year fire return intervals maintained at least 50% more active *B. gracilis* buds than annual and 6 yr fire return intervals relative to controls after March 2013. Active *P. smithii* buds increased for 3 yr return intervals during winter 2013 and remained more abundant through the end of the study. Plots burned at 6 yr return interval decreased bud activity during winter and maintained limited bud activity compared to 3 yr return intervals, but activity was similar to those following annual burns relative to controls.
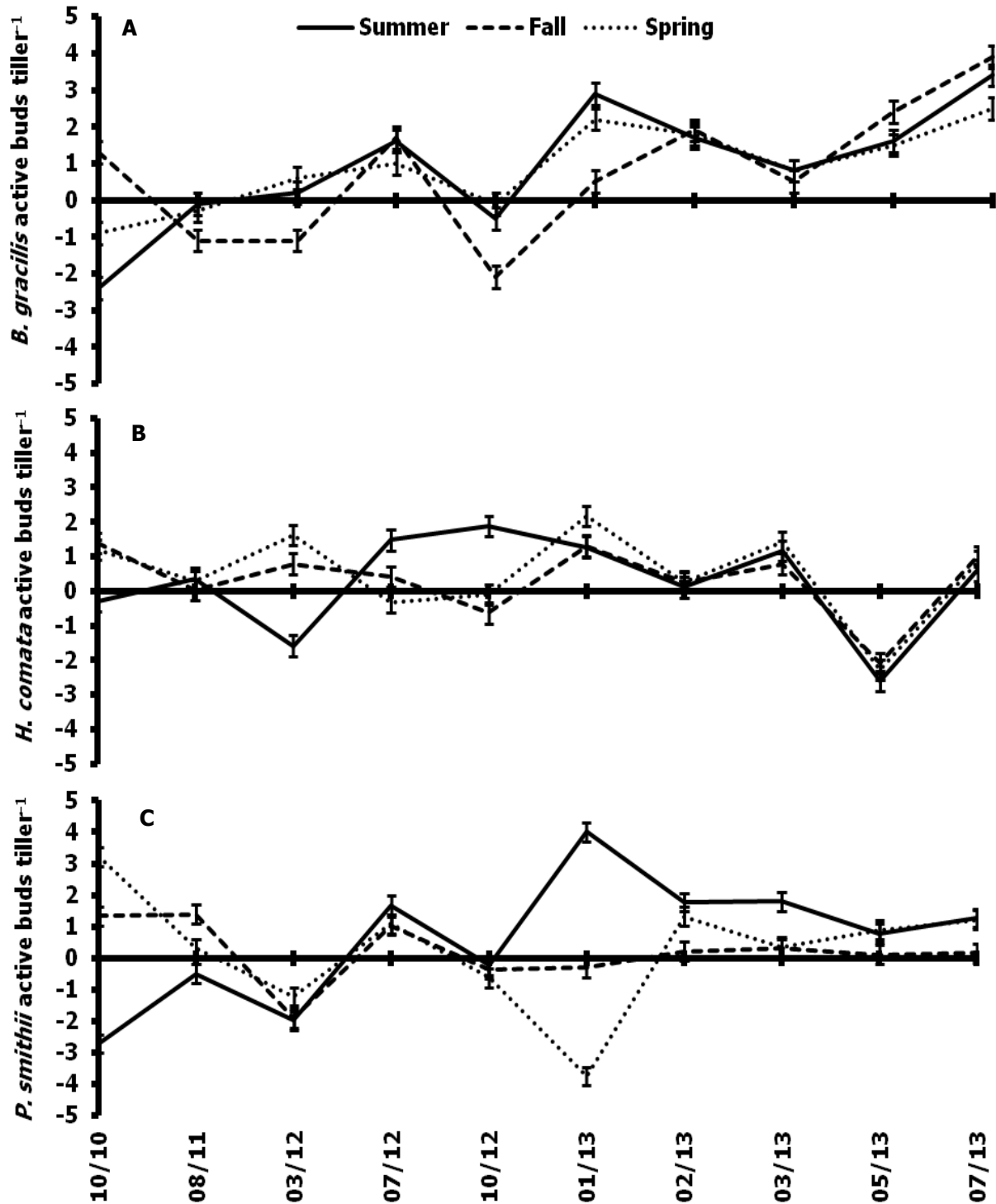
Figure 3.12. Short-term seasonal fluctuations of *B. gracilis*, *H. comata*, and *P. smithii* active buds following season of fire treatments relative to non-burned controls near Miles City, MT (± SEM; *P* < 0.01). Means are the difference between fire treatments and controls with no fire.

70
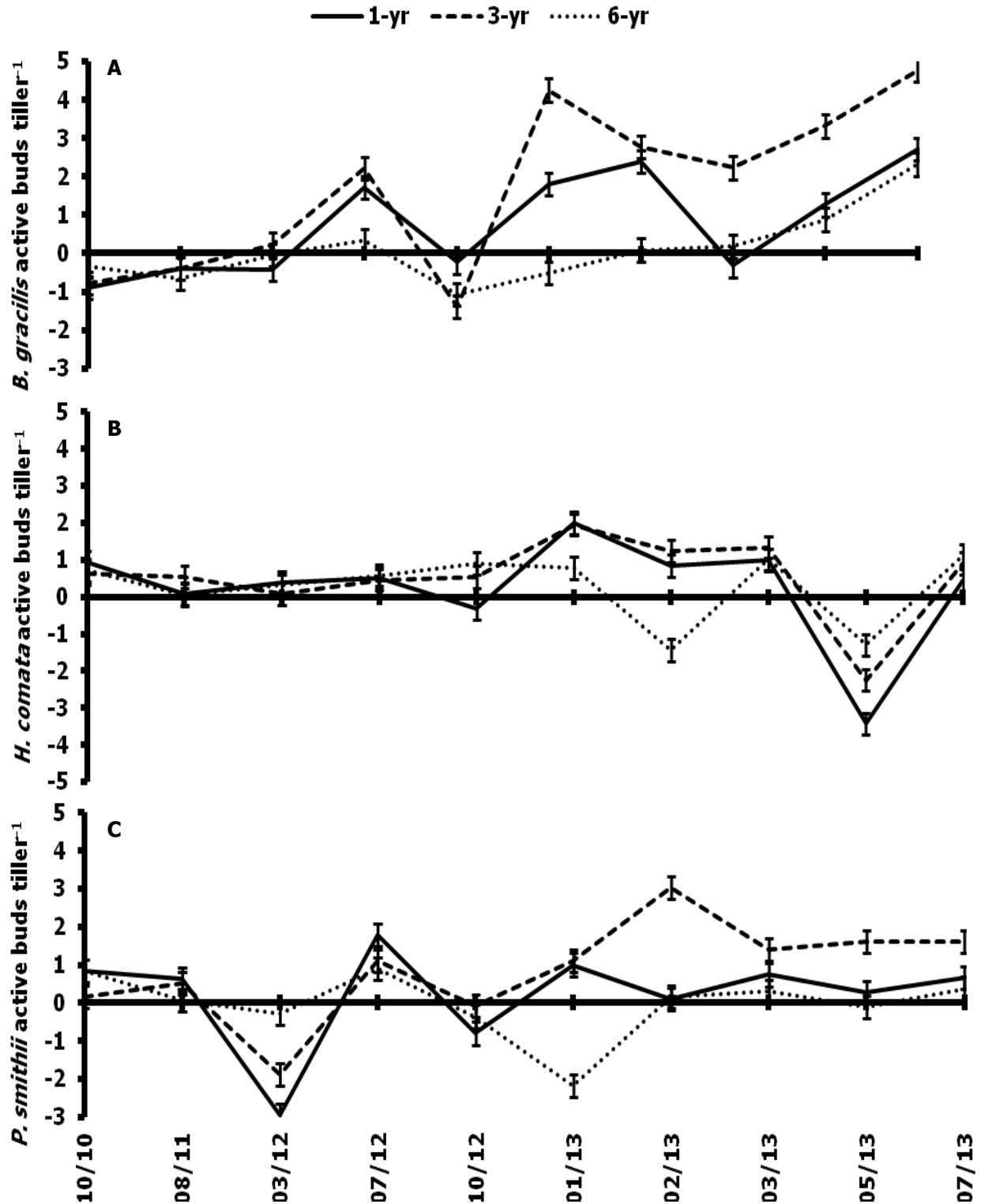
Figure 3.13. Short-term seasonal fluctuations of *B. gracilis, H. comata*, and *P. smithii* active buds tiller^{-1} following 1, 3, and 6 yr fire return intervals near Miles City, MT (± SEM; *P* < 0.01). Means are the difference between fire treatments and controls with no fire.

Season of fire and sampling date interacted in effects on dead buds of *B. gracilis* (*P* < 0.01; Fig. 3.14A) and *H. comata* (*P* < 0.01; Fig. 3.14B). *B. gracilis* on summer burned plots had fewer dead buds than controls during March 2012. Fall fire increased dead *B. gracilis* buds during winter 2012 relative to controls. Fewer dead *H. comata* buds were observed during winter 2012 on fall and spring burns than controls. In January 2013, reductions in dead *H. comata* were even greater for fall and spring burns compared to summer burned and non-burned plots.
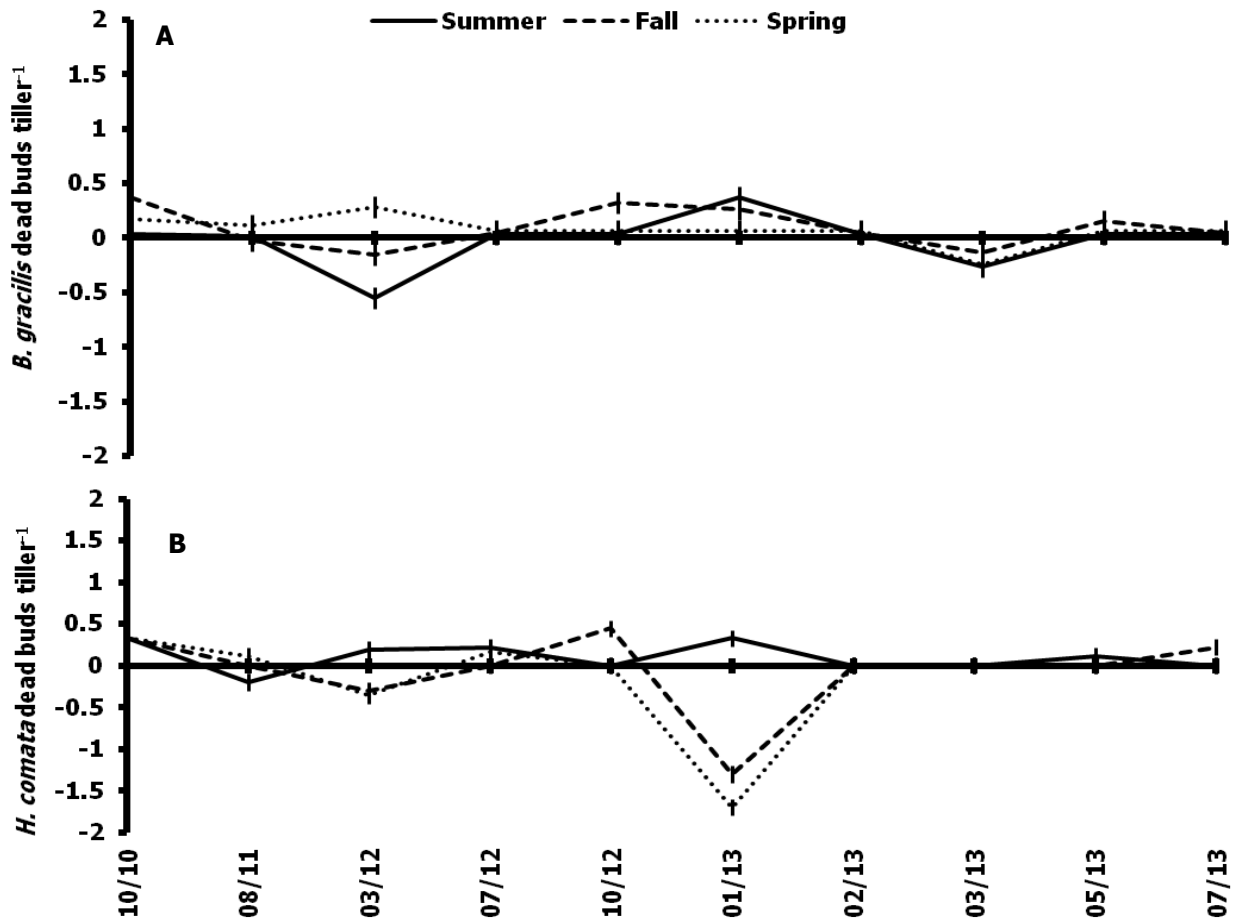
Figure 3.14. Short-term seasonal fluctuations of *B. gracilis* and *H. comata* dead buds tiller^{-1} following summer, fall, and spring prescribed burns near Miles City, MT (± SEM; *P* < 0.01). Means are the difference between fire treatments and controls with no fire.

Dormant *P. smithii* response varied by interacting effects of season of fire and sampling date (*P* < 0.01; Fig. 3.15). Dormant *P. smithii* buds fluctuated tremendously throughout 2012 and 2013. Dormant *P. smithii* buds decreased by 70% or more in July 2012 and January 2013 on summer burned

72

plots compared to fall, spring, and non-burned plots. However, dormant buds increased 63% or more during October 2012 compared to fall and spring burns and increased again by 35% in February 2013 relative to controls. Dormant buds of *P. smithii* became similar between control plots and season of fire treatments by the beginning of 2013 growing season.
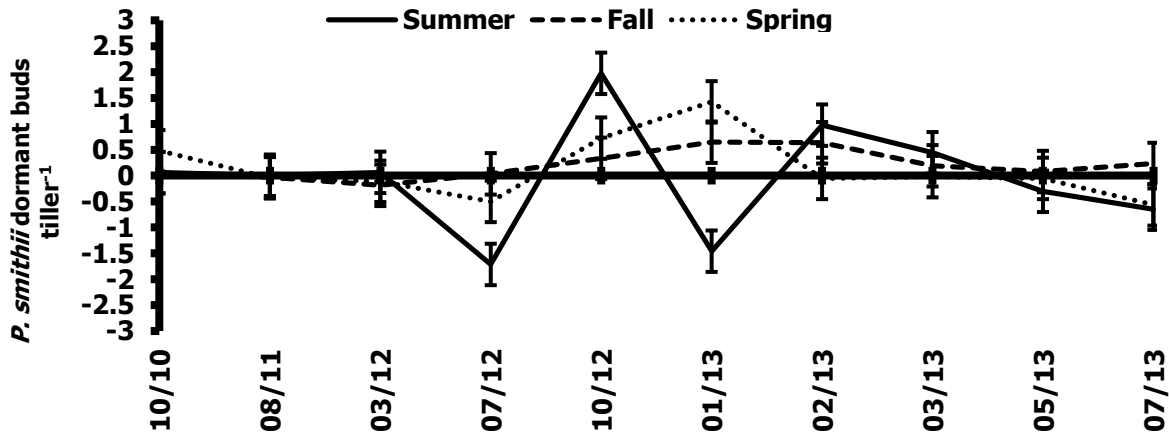
Figure 3.15. Short-term seasonal fluctuations of *P. smithii* dormant buds tiller^{-1} response following summer, fall, and spring burned plots near Miles City, MT (± SEM; *P* < 0.01). Means are the difference between fire treatments and controls with no fire.

Short-term response of *P. smithii* dormant buds also varied by fire return interval (*P* < 0.01). Dormant *P. smithii* buds differed among annual (0.23 ± 0.10), 3 yr (-0.20 ± 0.10), and 6 yr (0.34 ± 0.10) fire return intervals relative to controls. Dead *B. gracilis* buds increased on 6 yr fire return interval plots (0.13 ±0.70) relative to controls, but were similar on annual (0.0 ± 0.70) and 3 yr (0.0 ± 0.70) fire return interval plots relative to controls. Dead buds of *P. smithii* only varied by sampling date (*P* < 0.01). Dead *P. smithii* buds peaked during July 2012 and 2013 (0.59 and 0.45 ± 0.11, respectively) relative to controls.

Dormant buds of both *B. gracilis* (*P* < 0.05; Table 3.3) and *H. comata* (*P* < 0.01; Table 3.4) varied by season of fire, fire return interval and sampling date. Dormant *B. gracilis* were driven largely by the overwintering bud response. Plots burned during the summer and fall at 1 and 3 yr fire intervals increased the overwintering *B. gracilis* bud bank compared to spring burns 6 yr fire returns and controls. Summer and fall fire effects at 1 and 3 yr intervals maintained greater dormant buds throughout the start

73

of the growing season relative to controls. Summer and fall plots at 3 yr return intervals maintained

more dormant buds until the end of the 2013 growing season, unlike annual burn treatments and non-

burned plots.

Table 3.3. Short-term seasonal fluctuations of *B. gracilis* dormant buds tiller^{-1} throughout 10 sampling dates following summer, fall, and spring fire at 1, 3, and 6 yr fire return intervals near Miles City, MT (SEM ± 1.1; *P* < 0.05)[1]. Means are the difference between fire treatments and controls with no fire.

	1 yr			3 yr			6 yr		
Time	Summer	Fall	Spring	Summer	Fall	Spring	Summer	Fall	Spring
Oct. 2010	-0.8	1.0	0.0	-1.2	1.3	0.7	0.8	0.0	0.3
Aug. 2011	-0.2	0.0	0.0	0.1	0.0	0.0	-0.2	0.0	0.1
Mar. 2012	0.0	0.4	-1.5	0.0	-0.1	-1.5	1.2	-0.3	-0.9
Jul. 2012	-0.1 a	-1.3 a	-2.8 a	-3.3 b	-1.7 a	-3.0 a	-1.3 a	-0.2 a	-1.0 a
Oct. 2012	-0.3 d	-2.5 e	-0.9 d	6.5 a	0.0 c	-4.8 e	3.5 b	0.5 bc	-5.2 e
Jan. 2013	2.3 a	0.2 b	-2.9 c	2.0 a	2.0 a	-7.9 e	1.6 a	-0.2 b	-4.4 d
Feb. 2013	-3.5 c	-0.8 b	-2.5 c	4.9 a	-2.0 c	-0.8 b	-0.6 b	-0.4 b	-0.1 b
Mar. 2013	2.6 a	3.1 a	1.4 ab	2.2 ab	0.4 b	-0.6 b	1.4 b	0.9 b	-0.3 c
May 2013	-1.3 d	-1.8 d	-0.9 cd	3.3 b	1.0 c	5.1 a	3.3 b	0.0 c	0.6 c
Jul. 2013	-0.9	-0.9	1.3	-0.2	-0.9	-0.9	-0.9	0.1	-0.6

[1]Means with the same letter within each time period are similar (*P* < 0.05).

Table 3.4. Short-term seasonal fluctuations of *H. comata* dormant buds tiller^{-1} throughout 10 sampling dates following summer, fall, and spring fire at 1, 3, and 6 yr fire return intervals near Miles City, MT (SEM ± 0.6; *P* < 0.01)[1]. Means are the difference between fire treatments and controls with no fire.

	1 yr			3 yr			6 yr		
Time	Summer	Fall	Spring	Summer	Fall	Spring	Summer	Fall	Spring
Oct. 2010	-0.1	-0.1	-0.1	-0.1	0.2	-0.2	-0.1	0.2	0.2
Aug. 2011	-0.1	-0.3	0.0	-0.1	-0.1	-0.4	-0.1	-0.3	-0.3
Mar. 2012	-0.1	0.3	-0.1	-0.1	-0.1	-0.2	-0.1	-0.1	-0.1
Jul. 2012	-2.3 c	-0.8 b	0.0 a	-2.0 c	-0.1 a	0.2 a	-2.3 c	-0.3 ab	0.5 a
Oct. 2012	-0.1 a	-0.1 a	-0.1 d	-0.1 a	0.4 a	-0.2 a	-0.1 a	-0.1 a	-0.1 a
Jan. 2013	0.8 b	-0.4 b	-0.1 b	-0.1 b	-0.4 b	1.2 a	-0.8 b	1.2 a	0.5 a
Feb. 2013	-3.7 d	-1.1 b	-2.8 d	4.6 a	-2.3 c	-1.2 b	-0.8 b	-0.8 b	-0.5 b
Mar. 2013	-0.3	-0.3	-0.3	-0.3	-0.3	-0.4	-0.3	-0.3	-0.3
May 2013	-0.1	-0.1	-0.1	-0.1	-0.1	0.0	-0.1	-0.1	-0.1
Jul. 2013	0.9 a	0.6 a	0.5 a	0.1 a	0.2 a	1.0 a	1.1 a	-0.1 b	-0.1 b

[1]Means with the same letter within each time period are similar (*P* < 0.05).

Dormant buds of *H. comata* responded differently to fire treatments, especially during the 2012 drought and throughout winter periods (Table 3.4). During July 2012, summer and fall burned plots at annual, 3, and 6 yr intervals decreased dormant buds of *H. comata* compared to spring burns at all intervals and non-burned plots. Dormant *H. comata* buds were consistently reduced on annually burned plots compared to the 3 and 6 yr return interval treatments throughout 2012 and 2013. However, annual and 6 yr fire return intervals decreased dormant buds during winter 2013 across all seasons of fire relative to controls.

Discussion

Season of fire and fire return interval effects varied by species and sampling date. Short-term effects of fire treatments were maintained for at least two growing seasons following fire treatments. Belowground bud bank response illustrated the potential for *B. gracilis* and *P. smithii* bud banks to mediate aboveground fire effects by having well-protected vegetative buds and by altering the pattern and timing of tiller recruitment during growing and dormant seasons. These belowground responses illustrate mechanisms that native perennial grasses utilize in order to maintain belowground meristems and aboveground processes.

Aboveground response

Interannual variation in spring precipitation and time since fire were primary drivers in aboveground responses for this study. Aboveground plant density and reproductive tillers of *B. gracilis* exhibited signs of recovery in 2013 following drought effects of 2012. Similar results have been observed regarding *Bouteloua curtipendula*, a C_4 bunchgrass, following summer fire with consecutive droughts (Castellano and Ansley 2007). Species with large bud banks mediate drought effects (Carter et al. 2012). Despite drought, belowground buds were maintained and the large bud bank of *B. gracilis* later contributed to aboveground growth in 2013 when more favorable growing conditions occurred.

Reduced *H. comata* plant density and vegetative tillers following summer annual fire relative to controls was expected since *H. comata* is considered a fire-sensitive species. *H. comata* buds may be

sensitive to heat damage due to the species bunchgrass growth form (Raunkiaer 1934). Plant litter can accumulate in bunchgrass crowns, prolonging heat exposure (Wright 1971; Engle et al. 1998). *H. comata* is also more likely than *B. gracilis* and *P. smithii* to become meristem limited. Summer fires may have exacerbated all of these conditions since summer fire results in more complete fuel combustion (Ansley et al. 2006). In this study, summer fires burned with more intensity than fall or spring burned plots, based on thermocouple results of increased maximum temperature, heat duration, and heat dosage compared to fall and spring burned plots.

P. *smithii* increased plant density and vegetative tillers during the drought following annual and 3 yr fire return intervals relative to controls. Drought effects on *P. smithii* buds support other work indicating the species is drought tolerant and resilient following fire (Engle and Bultsma 1984; Vermeire et al. 2011). The impact of summer drought in cool-season dominated grasslands is minimal since production is completed by late spring/early summer (Eneboe et al. 2002). Additionally, increased production of *P. smithii* during the first growing season following fire has been observed in other studies (White and Currie 1983; Whisenant and Uresk 1990; Vermeire et al. 2011).

Immediate bud response

Season of fire and fire return interval treatments immediately affected bud activity of *B. gracilis* and *P. smithii*. Immediate fire effects on bud activity align with the bud transition hypothesis (Devitt and Stafstrom 1995; Cline 1997; Beveridge 2006) where buds enter different developmental stages that have varying degrees of sensitivity or responses to hormone signals (Stafstrom and Sussex 1992; Shimizu-Sato and Mori 2001; Morris et al. 2005). Bud activity is regulated by hormones that allow previously dormant buds to transition into active buds following environmental cues or development program signals (Shimizu-Sato and Mori 2001; Reece et al. 2007). Buds represent a large investment and are carefully modulated in response to environmental factors such as light quality, soil moisture, nitrogen and carbon availability, and growth and development of other plant parts (Vesk and Westoby 2004; Dun et al. 2006). If environmental conditions are not favorable, buds enter into temporary dormancy until growing conditions have improved or vice versa (Shimizu-Sato and Mori 2001).

This reversible developmental transition from dormancy to growth occurs rapidly (Stafstrom and Sussex 1992; Devitt and Stafstrom 1995; Shimizu and Mori 1998). Dormant axillary buds on Alaska pea (*Pisum sativum* L.) seedlings showed visible growth within 8 hours after defoliation (Devitt and Stafstrom 1995). Three days later, active buds became dormant again, illustrating that pea axillary buds can be stimulated to undergo more than one complete growth-dormancy cycle during the course of a few days (Devitt and Stafstrom 1995). Other studies on *Zea* L. and *Brassica* L. have found ribosomal protein genes increase or decrease during multiple growth-dormancy cycles in order to control bud development (Larkin et al. 1989; Bonham-Smith et al. 1992; Stafstrom and Sussex 1992).

In agreement with the hypotheses, bud mortality of *B. gracilis* and *P. smithii* did not differ between pre- and immediate post-fire assessments. However, *H. comata* bud mortality increased immediately after summer and fall fire. Summer and fall fire thermocouple measurements were consistently greater than those of spring fire. Increased bud mortality, decreased plant density, and decreased vegetative tillers of *H. comata* following summer fire further supports the premise that *H. comata* is a summer fire-sensitive species and is meristem-limited (Distel and Bóo 1996).

Short-term bud response

Short-term fluctuations and dynamics of bud banks varied tremendously according to their fire history. Previous research has emphasized increased bud density of C_4 species during the winter (Ott and Hartnett 2012). However, this study's results indicate summer and spring fire can also increase overwintering active buds relative to controls. Warm-season grasses maintain a larger overwintering bud bank, with long-living buds (> 1 yr) resulting in mixed-aged bud banks capable of tillering (Ott and Hartnett 2012). Summer and fall fire likely accelerated C_4 overwintering bud strategies in order to maximize bud development and storage to prepare for the upcoming growing season. Although *P. smithii* is a C_3 grass, belowground bud development and transitions between growth and dormancy closely resemble the C_4 pathway following fire events. Summer fire increased active *P. smithii* buds throughout the 2013 winter and into the 2013 growing season.

In previous studies, the degree of metabolic activity and outgrowth of axillary buds has been similar on perennial grass plants coming from sites with very different grazing intensities and history (Hendrickson and Briske 1997; Flemmer et al. 2002; Russell et al. 2013). Following all seasons of fire and fire return intervals, *H. comata* consistently decreased active buds during May 2013 relative to controls. All *H. comata* active buds synchronized bud transitions for tiller emergence in the upcoming 2013 growing season. Buds that overwintered provided more mature buds to start transitioning early in the growing season and to complete reproduction and new bud production before experiencing competition from other species. Similar responses were observed with other C_3 species in tallgrass prairie (Ott and Hartnett 2012). Annual and 3 yr interval burns increased overwintering active *H. comata* buds, illustrating the potential to manipulate overwintering strategies.

Three-year fire return intervals increased active buds throughout the 2013 winter and growing season for *B. gracilis* and *P. smithii* compared to annual and 6 yr fire return intervals and controls. The response of active buds to 3 yr fire return intervals illustrates the adaptive nature of theses native species to recurring fire in mixed-grass prairie. Since fire is recognized as a contributing factor in shaping plant communities, the Intermediate Disturbance Hypothesis (IDH) may be a valid model for understanding frequent fire effects on plant community composition and bud bank dynamics.

The IDH outlined by Connell (1961), Paine (1966), and Grime (1973), posits that species richness will be greatest at intermediate temporal spans following disturbance (Connell 1978). According to IDH, the optimal amount of disturbance (neither too rare, nor too frequent) within an ecosystem enables different ecological niches to be filled due to life-history characteristics (i.e., reproductive effort, bud development, tiller emergence) and differing responses to disturbance. Based on the contradicting life history traits of C_3 and C_4 species devoted toward bud outgrowth, intermediate fire return intervals allow both photosynthetic pathways to coexist and to maximize bud activity to maintain mixed-grass prairie dynamics and populations. This finding corresponds to previous research in tallgrass prairie illustrates the potential of intermediate fire return intervals to maximize species richness and grass productivity (Collins and Barber 1985; Collins 1987, 1992).

Summer fire decreased dormant *P. smithii* buds during the 2012 drought and also during

January of 2013 relative to controls. Based on previous drought literature, dormant buds of *P. smithii*

should have increased (Busso et al. 1989; Flemmer et al. 2002; Carter et al. 2012). However, dormant

buds decreased, resulting in increased vegetative tillering during the 2012 drought. Unlike the other C_3

species, *H. comata*, *P. smithii* is not meristem limited and buds appear to thrive following fire and during

drought conditions. This contrasts with previous studies that have reported decreased bud densities and

increased bud dormancy during drought years (Gardner 1942; McIntyre 1976; Carter et al. 2012).

However, those drought studies were not preceded by fire. Summer fire treatments may have provided a

buffer against consecutive-year drought conditions by increasing nutrient availability and signaling buds

to transition out of dormancy. Results of decreased dormant buds during drought and following summer

fire further emphasize the adaptive, resilient nature of *P. smithii* (Engle and Bultsma 1984; Heitschmidt et

al. 1999; Vermeire et al. 2011).

Conclusions

Earlier research suggested bud banks of C_3 species differ from C_4 species, and results were

interpreted to mean overwintering strategies were the primary driver of these differences. However,

these results indicate that bud banks of *P. smithii*, a C_3 species, more closely align with C_4 bud bank

strategies following fire and drought. Additionally, *B. gracilis* and *P. smithii* bud banks are resistant to

fire, perhaps as a function of their evolutionary history and adaptive belowground mechanisms.

Prescribed fire during the spring at 3 yr return intervals immediately increased *B. gracilis* bud

activity and decreased bud dormancy. Summer prescribed fire at 3 yr return intervals increased *P. smithii*

bud activity within hours of burning. The immediate responses of these two dominant perennial grasses

illustrate the importance of incorporating bud responses in planning prescribed fire and assessing the

aftermath of prescribed fire and wildfire in mixed-grass prairies. This aspect may be extremely beneficial

for rangeland managers looking to shift community composition toward a certain species, or in managing

post-fire landscapes.

Since mixed grasslands in the northern Great Plains evolved with frequent fire (5 -10 yr) (Wright and Bailey 1982), fire most likely has served as an environmental cue for tiller recruitment from the bud bank. Three-year fire return intervals appear to be very beneficial for bud banks of two dominant native perennial species. These results suggest recurring fire acts as a stimulant for axillary buds to activate tiller initiation. A single fire alone may not be an adequate environmental cue and frequent fire with return intervals of 3 yr may provide a more positive feedback from the bud bank to initiate growth of perennial grasses. Furthermore, immediate and short-term seasonal fluctuations provide a temporal scale with which to recommend burning and at what rate recovery may occur.

Acknowledgements

Literature cited

Ansley, R.J., M.J. Castellano, and W.E. Pinchak. 2006. Sideoats grama responses to seasonal fires and clipping. *Rangeland Ecology and Management* 59: 258-266.

Archibald, S., W.J. Bond, W.D. Stock, and D.H.K. Fairbanks. 2005. Shaping the landscape: fire-grazer interactions in an African Savanna. *Ecological Applications* 15: 96-109.

Benson, E.J., and D.C. Hartnett. 2006. The role of seed and vegetative reproduction in plant recruitment and demography in tallgrass prairie. *Plant Ecology* 187: 163-178.

Benson, E. J., D.C. Hartnett, and H. Mann. 2004. Belowground bud banks and meristem limitation in tallgrass prairie plant populations. *American Journal of Botany* 91: 416-421.

Beveridge, C.A. 2006. Axillary bud outgrowth: sending a message. *Current Opinion Plant Biology* 9:35-40.

Bonham-Smith, P.C., T.L. Oancia, and M.M. Moloney. 1992. Cytoplasmic ribosomal protein S15a from *Brassica napus*: molecular cloning and developmental expression in mitotically active tissues. *Plant Molecular Biology* 18: 909-919.

Busso, C.A., R.J.Mueller, and J.H. Richards. 1989. Effects of drought and defoliation on bud viability in two caespitose grasses. *Annals of Botany* 63: 477-485.

Carter, D.L., B.L. VanderWeide, and J.M. Blair. 2012. Drought-mediated stem and belowground bud dynamics in restored grasslands. *Applied Vegetation Science* 15: 470-478.

Castellano, M.J., and R.J. Ansley. 2007. Fire season and simulated grazing differentially affect the stability and drought resilience of a C_4 bunchgrass, C_3 bunchgrass and C_4 lawngrass. *Journal of Arid Environments* 69: 375-384.

Cline, M.G. 1997. Concepts and terminology of apical dominance. *American Journal of Botany* 84: 1064-1069.

Collins, S.L., and S.C. Barber. 1985. The effects of disturbance on diversity in mixed-grass prairie. *Vegetatio* 64: 87-94.

Collins, S.L. 1987. Interaction of disturbances in tallgrass prairie: a field experiment. *Ecology* 68: 1243-1250.

Collins, S.L. 1992. Fire frequency and community heterogeneity in tallgrass prairie vegetation. *Ecology* 73: 2001-2006.

Connell, J.H. 1961. Effects of competition, predation by *Thais lapillus*, and other factors on natural population of the barnacle *Balanus balanoides*. *Ecological Monographs* 31: 61-104.

Connell, J.H. 1978. Diversity in tropical rain forests and coral reefs. *Science* 199: 1302-1310.

Copeland, T.E., W. Sluis, and H.F. Howe. 2002. Fire season and dominance in an Illinois tallgrass prairie restoration. *Restoration Ecology* 10: 315-323.

Dalgleish, H.J., and D.C. Hartnett. 2006. Below-ground bud banks increase along a precipitation gradient of the North American Great Plains: a test of the meristem limitation hypothesis. *New Phytologist* 171: 81-89.

Dalgleish, H.J., and D.C. Hartnett. 2009. The effects of fire frequency and grazing on tallgrass prairie productivity and plant composition are mediated through bud bank demography. *Plant Ecology* 201: 411-420.

Dalgleish, H.J., A.R. Kula, D.C. Hartnett, and B.K. Sandercock. 2008. Responses of two bunchgrasses to nitrogen addition in tallgrass prairie: The role of bud bank demography. *American Journal of Botany* 95: 672-680.

Distel, R.A., and R.M. Bóo. 1996. Vegetation states and transitions in temperate semiarid rangelands of Argentina. In: N.E. West [ED.]. Rangelands in a sustainable biosphere, Proceedings Symposium V[th] International Rangeland Congress 23-28 July. pp 117-118. Salt Lake City, UT.

Dix, R.L. 1960. The effects of burning on the mulch structure and species composition of grasslands in western North Dakota. *Ecology* 41: 49-56.

Devitt, M.L., and J.P. Stafstrom. 1995. Nucleotide sequence of four ribosomal protein L27 cDNAs from growing axillary buds of pea. *Plant Physiology* 107: 1031-1032.

Dun, E.A., B.J. Ferguson, and C.A. Beveridge. 2006. Apical dominance and shoot branching. Divergent opinions or divergent mechanisms? *Plant physiology* 142: 812-819.

Engle, D.M., and T.G. Bidwell. 2001. Viewpoint: The response of central North American prairies to seasonal fire. *Journal of Range Management* 54: 2-10.

Engle, D.M., and P.M. Bultsma. 1984. Burning of northern mixed-prairie during drought. *Journal of Range Management* 37:398-401.

Engle, D.M., R.L. Mitchell, and R.L. Stevens. 1998. Late growing-season fire effects in mid-successional tallgrass prairies. *Journal of Range Management* 51: 115-121.

Eneboe, E.J., B.G. Sowell, R.K. Heitschmidt, M.G. Karl, and M.R. Haferkamp. 2002. Drought and grazing:IV. Blue grama and western wheatgrass. *Journal of Range Management* 55: 197-203.

Flemmer, A.C., C.A. Busso, and O. A. Fernández. 2002. Bud viability in perennial grasses: Water stress and defoliation effects. *Journal of Range Management* 55: 150-163.

Fuhlendorf, S. D., and D. M. Engle. 2004. Application of the fire: Grazing interaction to restore a shifting mosaic on tallgrass prairie. *Journal of Applied Ecology* 41: 604-614.

Gardner, J.L. 1942. Studies in tillering. *Ecology* 23: 162-174.

Govender, N., W.S.W. Trollope, and B.W. Van Wilgen. 2006. The effect of fire season , fire frequency, rainfall and management on fire intensity in savanna vegetation in South Africa. *Journal of Applied Ecology* 43: 748-758.

Grime, J.P. 1973. Competitive exclusion in herbaceous vegetation. *Nature* 242: 344-347.

Heitschmidt, R.K., M.R. Haferkamp, M.G. Karl, and A.L. Hild. 1999. Drought and grazing: I. Effects on quantity of forage produced. *Journal of Range Management* 52: 440-446.

Heitschmidt, R.K., and L.T. Vermeire. 2005. An ecological and economic risk avoidance drought management decision support system. *In:* J. A. Milne [ED.]. Pastoral systems in marginal environments. XXth International Grasslands Congress; 26 June–1 July; Dublin, Ireland. p 178.

Hendrickson, J.R. and D.D. Briske. 1997. Axillary bud banks of two semiarid perennial grasses: occurrence, longevity, and contribution to population persistence. *Oecologia* 110: 584-591.

Larkin, J.C., J.P. Hunsperger, D. Culley, I. Rubinstein, and C.D. Silflow. 1989. The organization and xpression of a maize ribosomal protein gene family. *Gene Development* 3: 500-509.

Littell, R.C., G.A. Milliken, W.W. Stroup, R.D. Wolfinger, and O. Schabenberger. 2006. SAS® for Mixed Models, second edition. Cary, NC: SAS Institute Inc

McIntyre, G.I. 1976. Apical dominance in the rhizome of *Agropyron repens*: the influence of water stress on bud activity. *Canadian Journal of Botany* 54: 2747-2754.

Morris, S.E., M.C.H. Cox, J.J. Ross, S. Krisantini, and C.A. Beveridge. 2005. Auxin dynamics after decapitation are not correlated with the initial growth of axillary buds. *Plant Physiology* 138: 1665-1672.

Paine, R.T. 1966. Food web complexity and species diversity. *The American Naturalist* 100: 65-75.

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

Raunkiaer, C. 1934. The life forms of plants and statistical plant geography. London, United Kingdom: Clarendon Press. 632 p.

Reece, P.E., W.H. Schacht, and J.D. Volesky. 2007. Skillful grazing management on semiarid rangelands. Lincolon, NB: University of Nebraska-Lincoln Extension EC162.

Russell, M.L., L.T. Vermeire, N.A. Dufek, and D.J. Strong. 2013. Fire, defoliation, and competing species alter *Aristida purpurea* biomass, tiller, and axillary bud production. *Rangeland Ecology and Management* 66: 290-296.

Shimizu, S.S., and H. Mori. 1998. Changes in protein interactions of cell cycle-related genes during the dormancy-to-growth transition in pea axillary buds. *Plant Cell Physiology* 10: 1073-1079.

Shimizu, S.S., and H. Mori. 2001. Control of outgrowth and dormancy in axillary buds. *Plant Physiology* 127: 1405-1413.

Stafstrom, J.P., and I.M. Sussex. 1992. Expression of a ribosomal protein gene in axillary buds of pea seedlings. *Plant Physiology* 100: 1494-1502.

Steuter, A. A. 1987. C_3/C_4 composition shift on seasonal burns: northern mixed prairie. *Journal of Range Management* 40: 27-31.

USDA, NRCS. 2013. The PLANTS database. Available at: http://plants.usda.gov. Accessed 7 September 2013.

USDA, NRCS. 2013. Ecological Site Description, Northern Rolling High Plains. Available at: https://esis.sc.egov.usda.gov. Accessed 7 September 2013.

Vesk, P.A., and M. Westoby. 2004. Funding the bud bank: A review of the costs of buds. *Oikos* 106: 200-208.

Vermeire, L. T., J. L. Crowder, and D. B. Wester. 2011. Plant community and soil environment response to summer fire in the northern Great Plains. *Rangeland Ecology and Management* 64:37-46.

Vermeire, L.T., R.K. Heitschmidt, and M.R. Haferkamp. 2008. Vegetation response to seven grazing treatments in the northern Great Plains. *Agriculture, Ecosystems, and Environment* 127: 66-72.

Vermeire, L.T., R.K. Heitschmidt, and M.J. Rinella. 2009. Primary productivity and precipitation use efficiency in mixed-grass prairie: a comparison of Northern and Southern U.S. Sites. *Rangeland Ecology and Management* 62: 230-239.

Western Regional Climate Center. 2013. Western U.S. climate historical summaries. Available at http://www.wrcc.dri.edu/cgi-bin/cliMAIN.pl?mt5690. Accessed 22 August 2013.

Whisenant, S.G., and D.W. Uresk. 1990. Spring burning Japanese brome in a western wheatgrass

  community. *Journal of Range Management* 43: 205-208.

White, R.S., and P.O. Currie. 1983. Prescribed burning in the northern Great Plains: yield and cover

  response of 3 forage species in the mixed grass prairie. *Journal of Range Management* 36: 179-

  183.

Wiles, L.J., G. Dunn, J. Printz, B. Patton, and A. Nyren. 2011. Spring precipitation as a predictor for peak

  standing crop of mixed-grass prairie. *Rangeland Ecology Management* 64: 215-222.

Wright, H.A. 1971. Why squirreltail is more tolerant to burning than needle-and-thread. *Journal of Range*

  Management 24: 277-284.

Wright, H. A., and A. W. Bailey. 1982. Fire ecology: United States and southern Canada. New York, NY,

  USA: John Wiley & Sons. 501 p.

CHAPTER 4. PHENOLOGY OF PERENNIAL NATIVE GRASS AXILLARY BUDS IN THE NORTHERN MIXED-
GRASS PRAIRIE

Abstract

Vegetative reproduction from belowground bud banks is the primary driver of aboveground
production and growth. Despite the importance of vegetative reproduction, the timing of tiller
recruitment from belowground buds is unknown for most dominant, perennial grasses. To address this
gap in knowledge, we assessed patterns of belowground bud development, dormancy, and mortality
associated with three perennial grass species differing in photosynthetic pathways and growth form in the
northern Great Plains. Temperature and soil moisture were measured below the soil surface of the plant
crown to determine development thresholds for belowground buds. Dormant buds increased during
winter for C_4 species ($P < 0.01$); whereas, C_3 species maintained limited dormant buds throughout winter,
with minor fluctuations in dormant and active bud transitions. Soil temperature was a good predictor (C-
statistic = 0.88) for C_4 species bud development and soil moisture was a reliable predictor (C-statistic =
0.86) for C_3 bud development during both years of the study. Photosynthetic pathway of perennial
grasses dictates belowground bud phenology for *B. gracilis* and *H. comata*. Perennial grasses differ in
belowground bud phenological dynamics during dormancy, impacting the following growing season's tiller
recruitment and bud transitions. The ability to predict belowground bud development could enhance the
utilization of management tools to manipulate tiller recruitment from the bud bank based on phenology
patterns. Patterns of bud dormancy and development will also be useful in understanding the underlying
mechanisms by which management practices and fluctuations in precipitation amount and growing
season length can alter mixed-grass prairie plant community dynamics.

Keywords: tiller, dormancy, soil moisture, soil temperature, vegetative reproduction, grassland

Introduction

Vegetative reproduction in perennial grasslands relies on tiller recruitment from belowground bud
banks. In fact, more than 99% of new tiller formation in grasslands originates from vegetative

reproduction (Benson and Hartnett 2006). However, little is known about bud bank phenology. Belowground stages of bud development are unknown for most dominant native grasses, especially in mixed-grass prairie.

Belowground bud development is typically associated with growth form (e.g., caespitose vs. rhizomatous) and photosynthetic pathway (e.g., C_3 vs. C_4) of grasses (Briske 1991). Plant growth form is largely determined by patterns of bud placement and outgrowth (Hendrickson and Briske 1997). Photosynthetic pathway primarily determines optimal growing temperature range of a species, especially growth period, reproductive phenology, and geographic distribution (McIntyre 1967). Since C_3 and C_4 species differ in aboveground phenological patterns, belowground bud development and transitions between activity and dormancy are likely to differ as well (Ott and Hartnett 2012).

Patterns of belowground bud development, dormancy, and mortality are important because new buds, longevity of maintained buds, and overall bud dynamics determine the size of the bud bank during the growing and dormant seasons (Ott and Hartnett 2011). The size and demography of the bud bank are considered ecological drivers in response to typical grassland disturbance processes, such as fire and grazing (Dalgleish and Hartnett 2006; Russell et al. 2013). For example, a reserve population of dormant buds may buffer population dynamics in the face of unpredictable environmental change, such as drought or prolonged growing seasons (Ott and Hartnett 2011).

In semi-arid environments, native, perennial grasses adapt for drought and variable precipitation by altering their bud strategies (Busso et al. 1989; Volaire 1995; Flemmer et al. 2002). For example, drought conditions during early spring may prevent current bud development from reaching a critical bud development threshold, thereby, reducing overall bud production and increasing the importance of dormant bud bank strategy (McKendrick et al. 1975; Van Noordwijk and De Jong 1986; Busso et al. 1989). In addition, limited late-spring/early- summer precipitation may have a greater impact on the maintenance of dormant buds than the growth and development of new buds. Increasing the importance of bud bank phenology strategies, northern mixed prairie has a compressed growing season, with 90% of perennial grass growth completed by 1 July (Vermeire et al. 2009). Therefore, species'

strategies and adaptability regarding belowground bud phenology are important factors optimizing tiller recruitment given the variable growing conditions.

Grass species containing extensive below-ground bud banks may be able to opportunistically capitalize on periods of abundant resource availability such as increased nitrogen following fire or early spring precipitation (Dalgleish and Hartnett 2006). However, some species may be meristem limited, with sparse bud banks that render them unable to respond to pulses of resource availability (Dalgleish and Hartnett 2006). Species that are meristem limited do not contain enough buds in the bud bank during the growing season to completely replace the aboveground tiller population (Dalgleish and Hartnett 2006).

Improved understanding and characterization of perennial grass bud bank phenology will facilitate predictions of tiller recruitment following aboveground disturbances, such as fire, drought, or grazing. Soil temperature and soil moisture, may identify ranges of conditions required to activate buds and grow tillers, enhancing the ability to time management decisions to achieve a desired outcome (e.g., maximized bud activity and development). The objectives of this study were to 1) assess bud development dynamics in dominant perennial grasses of mixed-grass prairie and 2) determine the relationship between bud development and soil moisture and soil temperature. We hypothesized that bud activity, dormancy, and mortality strategies will differ between C_3 and C_4 species due to differences in photosynthetic pathway and that soil moisture and soil temperature will be reliable indicators of bud growth.

Materials and methods

Site description

Research was conducted in semi-arid, mixed-grass prairie near Miles City, Montana, USA (lat 46°24′N, long 105°56′W) from March 2012 through July 2013. Average annual precipitation for the area is 343 mm, with a majority occurring from mid-April to mid-September (Fig. 4.1). Average daily temperatures range from 23 °C in July to -8 °C in January. The frost-free growing season ranges from 125 to 150 days (Western Regional Climate Center, Reno, NV, 2013).
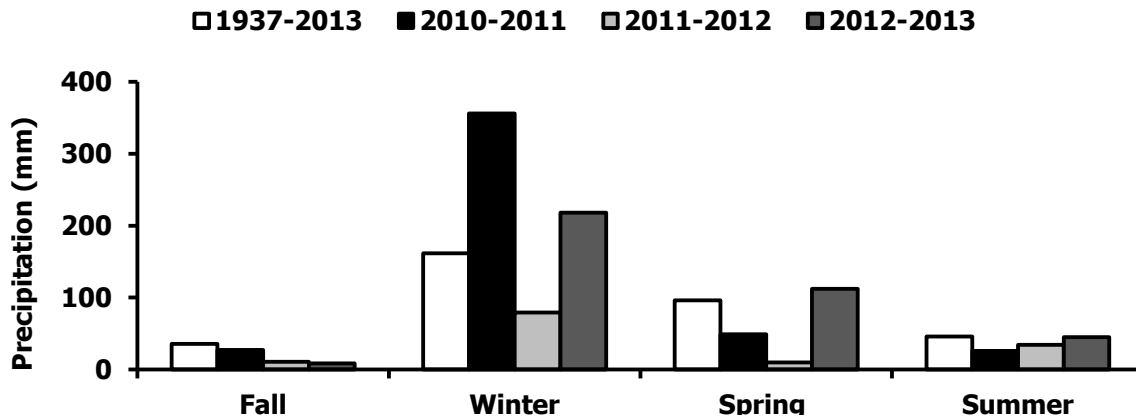
Figure 4.1. Study site 76 yr mean seasonal precipitation, fall (Oct-Dec), winter (Jan-Mar), spring (Apr-Jun), and summer (Jul-Sep) precipitation year (Oct-Sep) near Miles City, MT.

The study site is dominated by Pinehill loams (fine, smectitic, frigid Aridic Haplustalfs) including a complex of Kobase clay loams (fine, smectitic, frigid Torrertic Haplustepts) and Gerdrum clay loams (fine, smectitic, frigid Torrertic Natrustalfs) (USDA, NRCS Ecological Site Descriptions, 2013). Vegetation is dominated by perennial, native C$_3$ species including, needle-and-thread (*Hesperostipa comata* (Trin. & Rupr.) Barkworth), western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Löve), and threadleaf sedge (*Carex filifolia* Nutt.) and C$_4$ species blue grama (*Bouteloua gracilis* (Willd *ex*. Kunth) Lag. *ex* Griffiths), and to a lesser extent, buffalograss (*Bouteloua dactyloides* (Nutt.) J.T. Columbus). Annual grasses include six week fescue (*Vulpia octoflora* (Walter) Rydb.), field brome (*Bromus arvensis* L.), and cheatgrass (*Bromus tectorum L.*). The primary shrub species on the site is Wyoming big sagebrush (*Artemisia tridentata* Nutt. sp*. Wyomingensis* Beetle & Young*)* and half shrub fringed sage (*A. frigida* Wild.). Forbs included the biennial forb, yellow salsify (*Tragopogon dubius* Scop.) and annual forbs field cottonrose (*Logfia arvensis* (L.) Holub), woolly plantain (*Plantago patagonica* Jacq.), and rough false pennyroyal (*Hedeoma hispida* Pursh.). Plant nomenclature follows the USDA PLANTS database (USDA, NRCS 2010)

Field sampling and lab methods

Tiller counts, bud assessments, and classifications of tiller phenological stages were conducted weekly during the growing season and bi-monthly during early spring and early autumn seasons. Tiller

counts and phenological stages were tracked for two growing seasons on 36 permanently marked plants for each target species of *B. gracilis* (C$_4$ species)*, P. smithii* (C$_3$ species), and *H. comata* (C$_3$ species).

Tillers were classified as vegetative or reproductive for one individual plant. Due to the rhizomatous growth form of *B. gracilis* and *P. smithii*, definitions of an individual were outlined prior to the start of the study and implemented throughout. Tiller counts performed on *B. gracilis* plants were considered and counted as part of an individual when gaps between tillers did not exceed 3- cm. *P. smithii* tiller measurements were made by counting all tillers within a 0.25-m^{-2} quadrat. Individual *H. comata* tiller counts were readily distinguished due to their bunchgrass growth form.

Tillers were counted and classified into phenological stages using the continuous numerical index of Moore et al. (1991). Tillers were classified into four primary growth stages of germination, vegetative, elongation, and reproduction. The reproductive primary stage included sub-stages numbered 0-5 which pertained to particular events in the ontogeny of each tiller. The vegetative and elongation sub-stages were open ended, with the number of sub-stages being equivalent to the number of morphological events that occurred for that species.

The germination primary stage encompasses the events following coleoptile emergence from the soil (Moore et al. 1991). The vegetative stage refers to the developmental period comprising leaf growth and development. The vegetative stage begins with the emergence of the first leaf from the prophyll and each successive substage refers to the number of fully emerged live, collared leaves present. The elongation primary stage is during culm elongation and node development. Substages of the elongation stage are defined by the number of nodes that have become either palpable or visible as the result of stem elongation. The elongation stage concludes when the inflorescence is enclosed in the uppermost leaf sheath, which is commonly referred to as the boot stage. This boot stage is the initiation of the primary stage of reproduction. The reproductive stage begins with emergence of the inflorescence and continues through anthesis and fertilization. The substages for the reproductive stage were based on pre-determined morphological descriptors. Numerical indices for each phenological primary and sub-stage mean growth stage (MGS) were determined by summing the numerical indices for tillers within

each growth stage, totaling the sums from each growth stage, and dividing by the total number of tillers staged.

Belowground bud assessments were made from tillers that were similar in phenological primary and sub-stages of target plants for each species. Tillers were harvested from neighboring permanently marked plants that were in a similar phenological stage using the classification system of Moore et al. (1991). Buds were counted and classified using a dissecting microscope and determined to be living or dead. Amount of active buds, dormant buds, and dead buds were confirmed using Tetrazolium and Evans Blue staining procedures. Tillers were submerged in Tetrazolium solution at room temperature for 24- hr following initial classification. Active buds stained pink and dormant and dead buds retained a white or yellowish color. If dormant buds were present on a tiller, that tiller was submerged into Evan's Blue solution at room temperature following the Tetrazolium staining for 20- min. Upon completion of Evan's Blue staining, dormant buds maintained their white pigment, whereas, dead buds stained dark blue.

Soil moisture was measured at each tiller harvest and phenology classification during the growing seasons of 2012 and 2013. Soil moisture was determined with a Field Scout TDR 100 soil moisture meter with 7.5-cm probes inserted at the base of each target plant (Spectrum Technologies, Aurora, IL). Soil temperatures (°C) were determined from hourly measurements each day during the 2012 and 2013 growing seasons using WatchDog B-series button loggers (Spectrum Technologies, Aurora, IL) and the daily soil temperature maximum and minimum were used in analyses. Each temperature button logger was inserted adjacent to the crown of each target plant at approximately 0-1 cm below the soil surface.

Statistical analysis

Tiller count data were analyzed for all species using analysis of variance (MIXED procedure of SAS; Littell et al. 2006). The model included year and species as fixed effects with vegetative and reproductive tillers as response variables. Bud assessments were analyzed using analysis of variance (MIXED procedure of SAS, Littell et al. 2006) with tiller harvest date as a repeated measure. Plant was the experimental unit and statistical significance was set at $P < 0.05$. The independent factor was tiller

harvest date and response variables were active buds, dormant buds, and dead buds. In separate

analyses, soil moisture and soil temperature were tested as predictor variables for *B. gracilis*, *H. comata*,

and *P. smithii* bud development with logistic regression (LOGISTIC procedure of SAS; Littell et al. 2006).

Plant was the experimental unit and statistical significance was set at $P < 0.05$.

Results

The 2012 growing season consisted of three primary phenological stages for all three species

(Table 4.1). Both C_3 species had two fully-emerged live leaves that were collared by 16 April 2012. The

C_4 grass maintained one fully-emerged leaf until 28 May 2012, when vegetative growth increased rapidly

and began elongation by 15 June 2012. All species reached elongation with three visible nodes;

however, the reproductive stage was not achieved as a primary growth stage for any species during

2012.

Table 4.1. Phenology stages of *B. gracilis*, *H. comata*, and *P. smithii* throughout the 2012 growing and dormant seasons near Miles City, MT.

Date	B. gracilis	H. comata	P. smithii
15 March 2012	V1	V1	V1
28 March 2012	V1	V1	V2
16 April 2012	V1	V2	V2
15 May 2012	V1	V2	V2
28 May 2012	V2	V2	V2
5 June 2012	V3	E1	E1
15 June 2012	E1	E2	E3
23 June 2012	E2	E3	E3
28 June 2012	E3	E3	E3
5 July 2012	E3	E3	E3
16 July 2012	E3	E3	E3
30 July 2012	E3	E3	E3
15 August 2012	E3	E3	E3
28 August 2012	E3	E3	E3
10 September 2012	E3	E3	E3
25 September 2012	E3	E3	E3
9 October 2012	E3	E3	E3
24 October 2012	E3	E3	E3

Similar to 2012, the 2013 growing season consisted of three primary phenology stages for all

three species (Table 4.2). Unlike 2012, all three species had three fully-emerged leaves with collars by

28 May 2013. The elongation stage for *P. smithii* began within a week of the third leaf vegetative stage. However, both *B. gracilis* and *H. comata* maintained the primary vegetative phenology stage through July 2013. Vegetative tillers of *B. gracilis* produced a fourth fully-emerged collared leaf by 2 July 2013, which was not achieved in 2012.

Table 4.2. Phenology stages of *B. gracilis*, *H. comata*, and *P. smithii* throughout the 2013 growing season near Miles City, MT.

Date	*B. gracilis*	*H. comata*	*P. smithii*
2 May 2013	V1	V1	V1
13 May 2013	V1	V1	V2
28 May 2013	V3	V3	V3
5 June 2013	V3	V3	E1
11 June 2013	V3	V3	E1
18 June 2013	V3	V3	E1
26 June 2013	V3	V3	E2
2 July 2013	V4	V3	E3
10 July 2013	V4	V3	E3

Soil moisture varied throughout the 2012 and 2013 growing seasons (Table 4.3). Growing conditions during 2012 were extremely dry, with drought conditions occurring most of the spring and summer (34% below average; Fig. 4.1). Spring 2012 was the second driest on record and drought conditions persisted throughout eastern MT and the northern Great Plains (Western Regional Climate Center, Reno, NV). Spring of 2013 brought near-average spring and summer precipitation, enabling growth of cool- and warm-season grasses following severe drought of 2011-2012.

Vegetative tiller production varied by species and year ($P < 0.01$; Fig. 4.2A). Cool-season grass vegetative tiller production increased during 2013; however, *B. gracilis* produced more vegetative tillers during 2012 compared to the other species. *P. smithii* produced 76% more reproductive tillers in 2012 than 2013 ($P < 0.01$; Fig. 4.2B). Reproductive tillers of *B. gracilis* and *H. comata* were similar between study years ($P > 0.05$).

Table 4.3. Average monthly soil moisture (%) and soil temperature (°C) taken at target species during the 2012 and 2013 growing seasons near Miles City, MT (± SE).

|	2012		2013	
Month	SM, %	Soil temp, °C	SM, %	Soil temp, °C
March	17.9 ± 2.3	17.2 ± 3.4	23.3 ± 1.7	9.1 ± 6.3
April	10.6 ± 4.3	20.7 ± 6.2	31.6 ± 6.9	10.7 ± 3.3
May	4.5 ± 1.3	10.6 ± 2.3	29.3 ± 3.6	19.8 ± 6.9
June	2.8 ± 1.2	33.2 ± 6.2	31.0 ± 6.6	30.1 ± 7.6
July	3.6 ± 0.7	35.7 ± 6.2	41.6 ± 9.3	31.8 ± 9.1
August	4.9 ± 2.1	37.0 ± 3.2	-----	-----
September	26.9 ± 4.3	17.4 ± 2.1	-----	-----
October	32.6 ± 2.9	9.4 ± 4.6	-----	-----
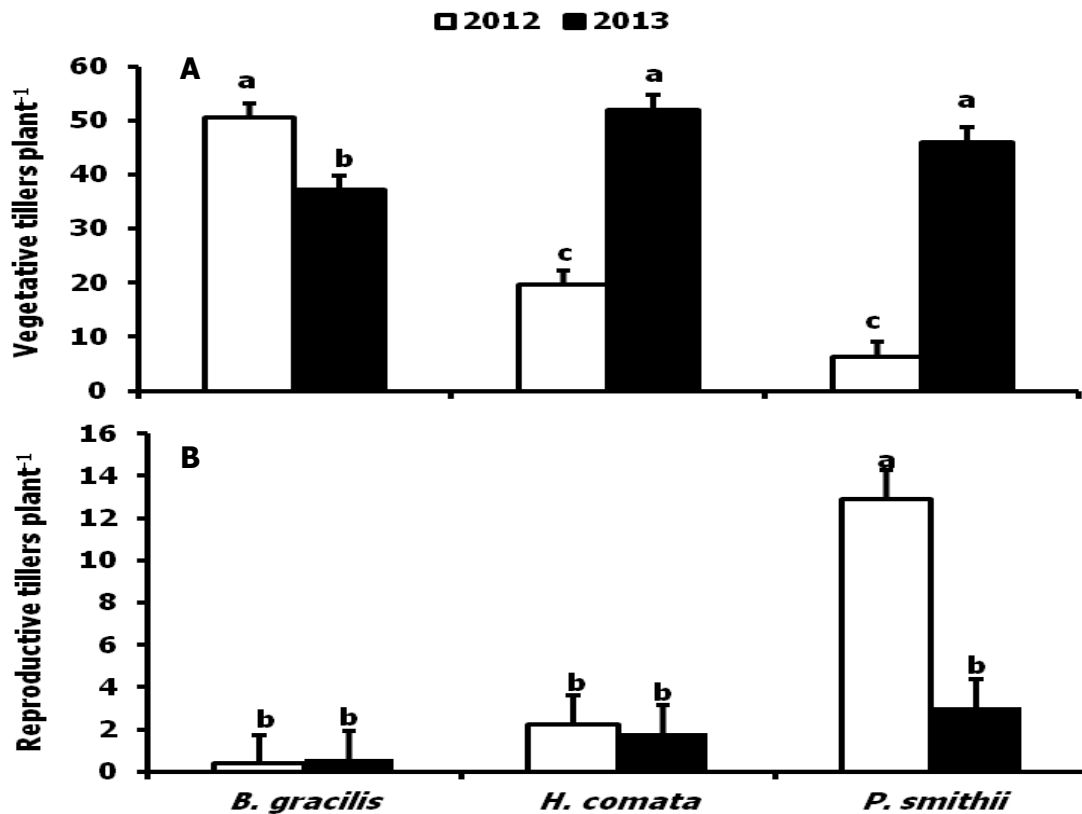
Figure. 4.2. Species and year effects A) vegetative tillers plant^{-1} (+SEM) and B) reproductive tillers plant^{-1} (+SEM) during 2012 and 2013 near Miles City, MT. Means marked with the same letter are similar within response variable ($P < 0.05$).

Active bud production varied according to photosynthetic pathway ($P < 0.01$; Fig. 4.3). Active buds for *B. gracilis* decreased during late summer of 2012, but increased during fall 2012 and the

growing season of 2013 ($P < 0.01$; Fig. 4.3A). Active *H. comata* buds were maintained at similar

numbers with minor fluctuations during late summer of 2012 and May of 2013 ($P < 0.01$; Fig. 4.3B).

Active *P. smithii* buds showed strong intra-and inter-annual variation ($P < 0.01$; Fig. 4.3C). Similar to *B.*

gracilis and *H. comata* results, *P. smithii* active buds also decreased during mid-summer of 2012.
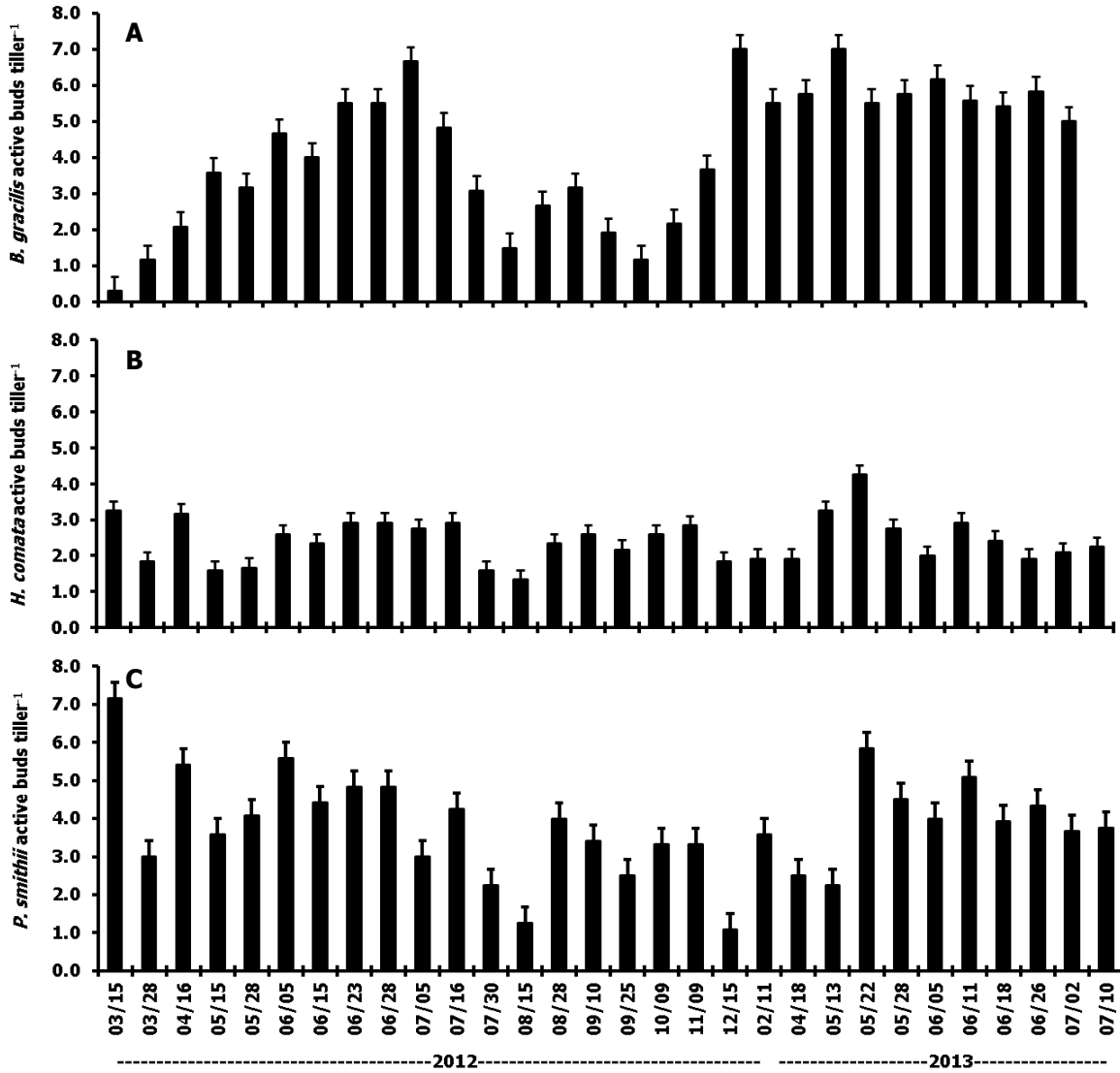
**Figure 4.3. Seasonal fluctuations in active buds tiller^{-1} for A) *B. gracilis*, B) *H. comata*, and C)
P. smithii near Miles City, MT (± SEM; *P* < 0.05).**

Dormant *B. gracilis* buds increased during summer, but decreased at the beginning of 2013 ($P <$

0.01; Fig. 4.4A). Dormant *H. comata* buds kept much smaller reserves throughout the study years with

slight increases (15-20%) occurring from July 2012 to September 2012 and November 2012 to May 2013

(P < 0.05; Fig. 4.4B). Dormant *P. smithii* buds began increasing during June 2012 and did not decrease

until February 2013 (P < 0.01; Fig. 4.4C). Similar amounts of dormant buds were observed during
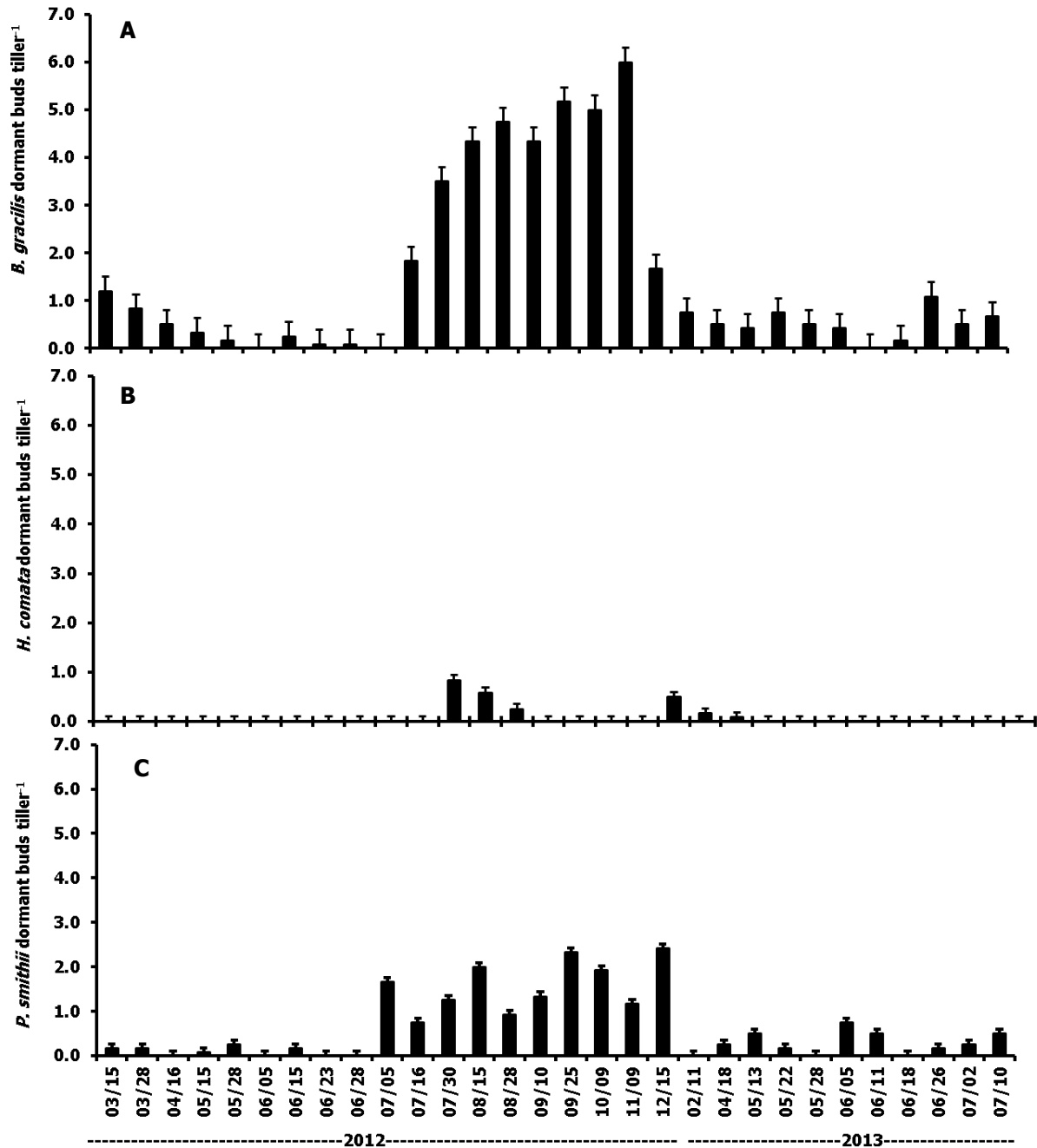
growing seasons of both study years for *P. smithii*.

Figure 4.4. Seasonal effects on dormant buds tiller^{-1} for A) *B. gracilis* and B) *H. comata*, and C) *P. smithii* for two growing seasons near Miles City, MT (± SEM; *P* < 0.01).

Dead *B. gracilis* buds increased 50% from July 2012 to February 2013, with peak dead buds occurring during November 2012 ($P < 0.01$; Fig. 4.5A). Within this period, B. gracilis bud mortality spiked well over a six-fold increase. However, by the start of 2013, bud mortality rates were similar to 2012. Dead buds of *H. comata* were minimal, except for 5-10% increases during March, May, and November of 2012 ($P < 0.01$; Fig. 4.5B). Bud mortality was minimal during 2013 and did not peak at similar times compared to 2012. Dead *P. smithii* buds were similar throughout 2012, but increased 70% at the beginning of 2013 ($P < 0.01$; Fig. 4.5C). After June 2013, P. smithii bud mortality decreased similar to 2012 rates.
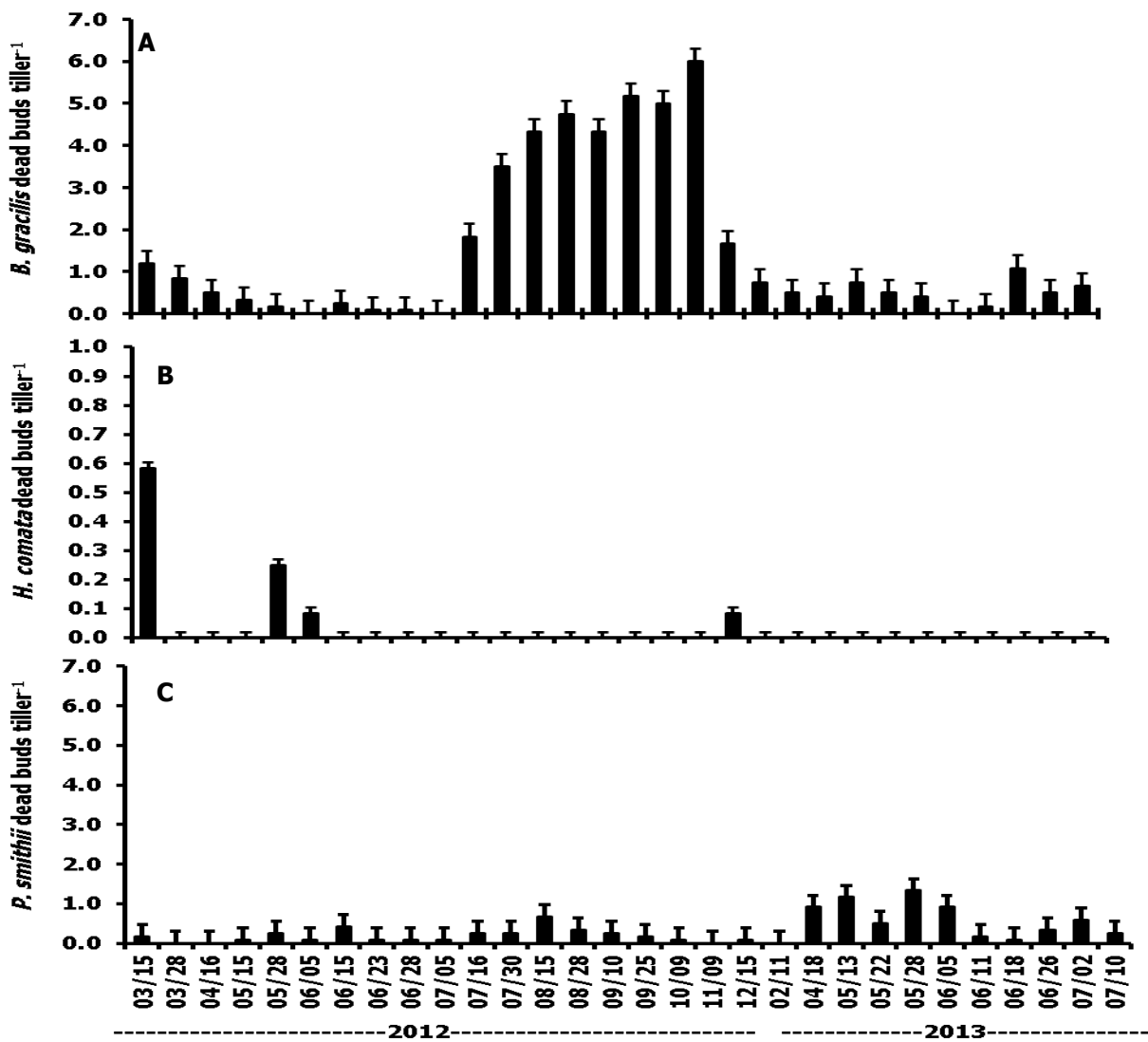

Figure 4.5. Dead buds tiller^{-1} for A) *B. gracilis* and B) *H. comata,* and C) *P. smithii* for two growing seasons one winter dormancy period near Miles City, MT (± SEM; $P < 0.01$).

Mean growth-stage (MGS) counts of *H. comata* and *P. smithii* differed during the first tiller harvest at the beginning of the 2012 growing season, but demonstrated a similar MGS by the second tiller harvest (Fig. 4.6). *P. smithii* matured earlier than *H. comata* and was approximately 2 weeks ahead until 31 May when both species entered a rapid transition phase through elongation. Both cool season grasses produced the most active buds by mid-March, with *H. comata* producing 3.3 ± 0.4 and *P. smithii* producing 7.2 ± 0.4. In contrast, *B. gracilis* initiated reproduction at the end of May and experienced phenological development during the first week in July, producing the most active buds (6.7 ± 0.4) by 5 July and decreased active bud production by 16 July (4.8 ± 0.4).
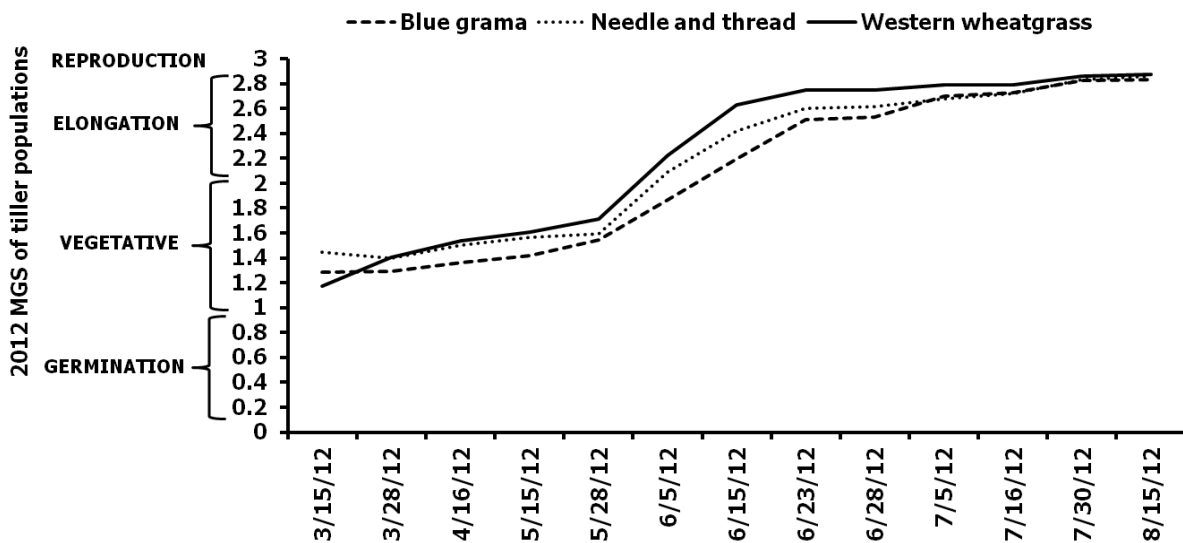
Figure 4.6. Mean growth stage (MGS) of tiller populations throughout the 2012 growing season near Miles City, MT.

During 2013, tiller populations of *B. gracilis* did not begin to phenologically advance until 14 May 2013 in comparison to cool-season grasses (Fig. 4.7). After 14 May 2013 all species in the study advanced similarly in phenology stages until *P. smithii* reached elongation and reproductive primary stages starting 3 June 2015. Phenology stages of *P. smithii* kept advancing until 3 July 2013, when a majority of *P. smithii* tillers reached reproductive status in comparison to *B. gracilis* and *H. comata* tiller populations.

98
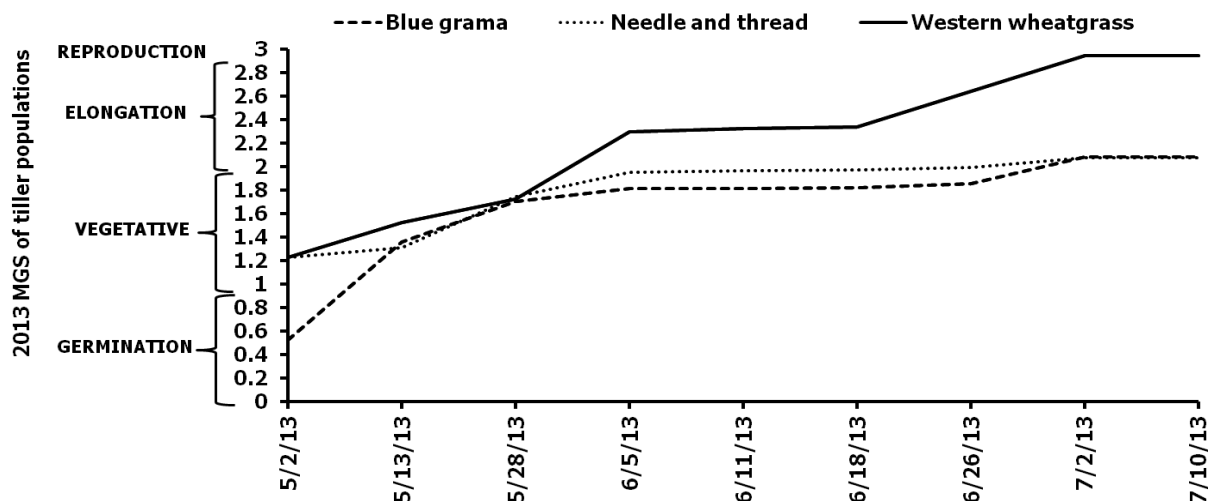
Figure 4.7. Mean growth stage (MGS) of tiller populations throughout the 2013 growing season near Miles City, MT.

Soil temperature was the best predictor for *B. gracilis* bud development; whereas, soil moisture was not a significant predictor of *B. gracilis* bud development (Table 4.4). On the other hand, soil moisture was a reliable indicator of bud development for both cool-season grasses, and soil temperature was not a significant predictor.

Table 4.4. Predictive ability of variables used in logistic regression model to test for bud development near Miles City, MT.

Predictor variable	Model information		
	C-statistic[a]	H-L test[b]	$P > $ Chi^{-2}
B. gracilis			
Soil moisture (%)	0.65	0.15	0.83
Soil temperature (°C)	0.88	< 0.01	0.03
H. comata			
Soil moisture (%)	0.86	0.41	0.04
Soil temperature (°C)	0.65	0.04	0.44
P. smithii			
Soil moisture (%)	0.93	0.40	0.03
Soil temperature (°C)	0.71	0.64	0.30

[a]**C-statistic is the predictive ability of the model.**
[b]**Hosmer-Lemeshow goodness of fit test (higher value = better fit).**

When soil temperatures rose above 50 °C, the probability of *B. gracilis* bud development exceeded 0.1 (Fig. 4.8). Probability of bud development increased rapidly for *B. gracilis* when temperatures were between 55 and 65 °C. With cool-season grasses, the probability of bud development

reached 0.1 when soil moisture was greater than 15% (Fig. 4.9A and 4.9B). However, probability of bud

development increased faster for *P. smithii* than *H. comata* after the 15% soil moisture threshold.
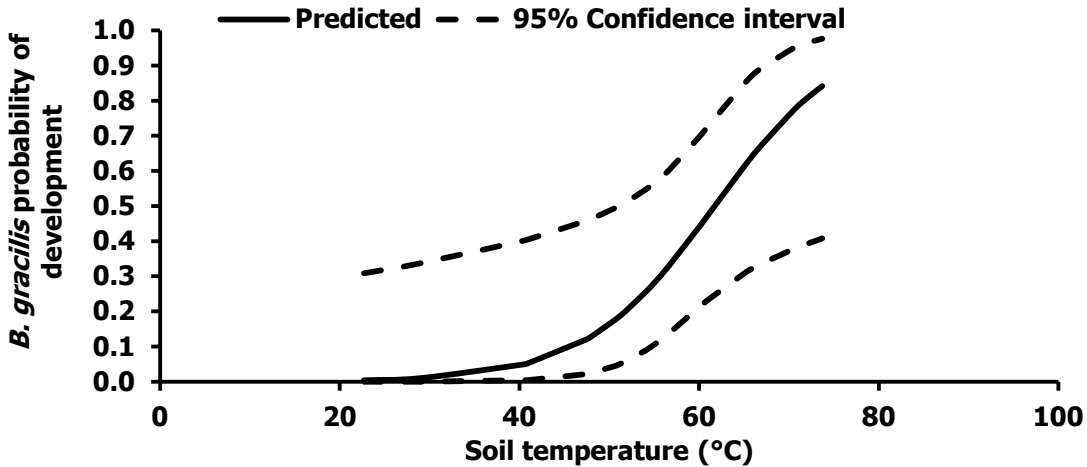

Figure 4.8. Predicted bud development and 95% confidence interval for *B. gracilis* bud development as a function of soil temperature (°C) near Miles City, MT.
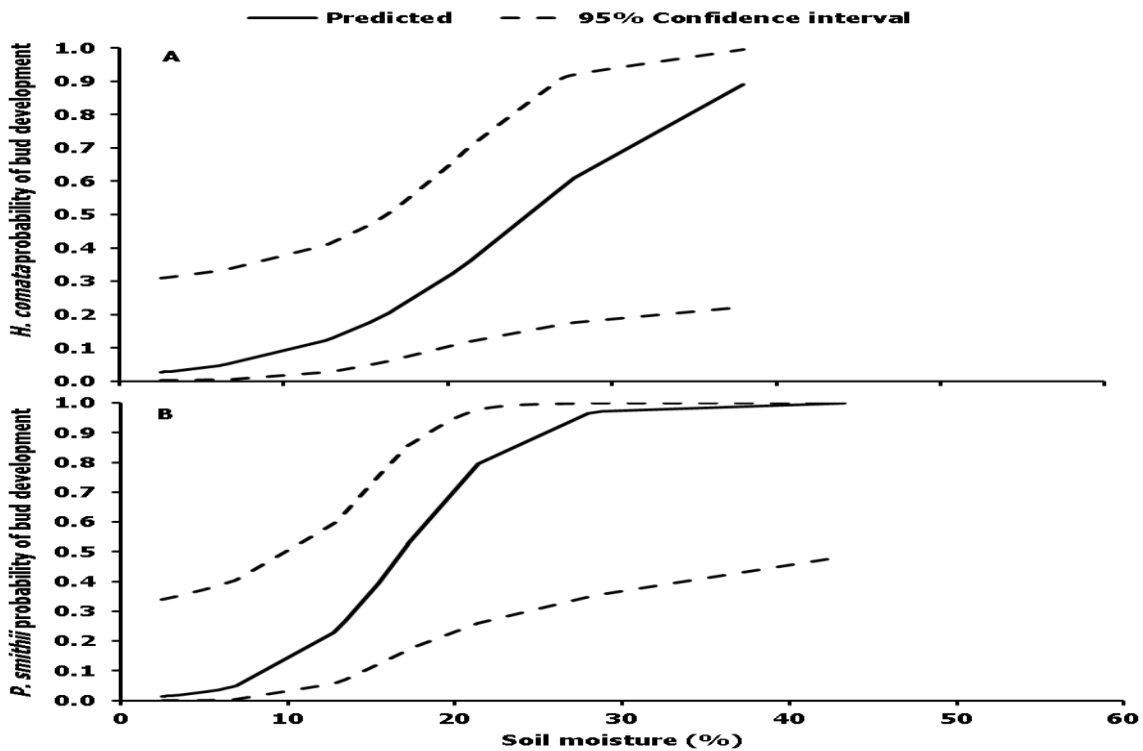

Figure 4.9. Predicted bud development and 95% confidence interval for A) *H. comata* and B) *P. smithii* as a function of soil moisture near Miles City, MT.

Discussion

Phenological patterns of belowground bud development, maintenance, and transitions resemble aboveground phenological patterns for *B. gracilis* and *H. comata* as a consequence of their photosynthetic pathways. Outlining these similarities and differences in bud development provides foundational knowledge of belowground bud dynamics of species with different growth forms and photosynthetic pathways. These results are similar to observations in tallgrass prairie (Ott and Hartnett 2012); where phenological patterns of C_3 and C_4 species differ dramatically during growing and dormant seasons. However, in this study, *P. smithii* had characteristics of both C_3 and C_4 phenological patterns. These results are the first highlighting bud phenology patterns of a resistant, native species that is capable of altering belowground bud transitions based on aboveground stimuli.

Reproductive *P. smithii* tillers increased during drought conditions of the 2012 growing season. Spring precipitation (April – June) is considered a major driver of aboveground productivity in the northern Great Plains (Vermeire et al. 2009; Wiles et al. 2011). Observations are similar to phenology experiments in semi-arid environments (Aronson et al. 1992), where desert plants increased reproductive tillers and transitioned earlier to reproductive development compared to matched species from Mediterranean environments (Herms and Mattson 1992). Observed increases in *P. smithii* reproductive tillers may be an effective strategy for *P. smithii* during drought since previous research shows reproductive tillers contain more mature, active buds than vegetative tillers (Ott and Hartnett 2011). Results of others suggest drought served as an environmental cue to advance tillers to reproduction in order to activate greater numbers of buds and increase bud size (Angus and Moncur 1977; Ott and Hartnett 2011). Due to their larger size, buds on reproductive tillers have a higher survival rate and, thus, spend more time in the bud bank (Ott and Hartnett 2011). Larger buds may also display more well-developed and active vascular connectivity with the polar auxin transport system, increasing auxin export and nutrient import into the bud (Waldie et al. 2010).

Active *B. gracilis* buds transitioned into dormancy during the late summer of 2012, increasing the dormant bud bank going into winter. Results were similar to those for *Andropogon gerardii* (Vitman) bud density during winter in tallgrass prairie (Ott and Hartnett 2012). Bud longevity is > 2.5 years in *A. gerardii*, resulting in an accumulation of dormant bud banks (Hendrickson and Briske 1997; Ott and Hartnett 2012). Results of others suggest C_4 species depend on large dormant bud banks to survive winter and recruit new tillers in the spring. Species like *B. gracilis* may maintain high population dominance because their high-density dormant bud banks enable them to respond rapidly to a resource pulse or sustain their population during drought years (Dalgleish and Hartnett 2006; Ott and Hartnett 2012).

Mortality of *B. gracilis* buds increased immediately following drought. Bud mortality observations support previous bud mortality research in tallgrass prairie that examined drought effects on bud densities of C_4 grasses (Carter et al. 2012). Therefore, data indicate that occasional droughts of short duration (less than one year) may do little to obstruct the recovery of bud bank densities. This may also be why C_4 grasses maintain a large dormant bud bank during winter (Ott and Hartnett 2012; Dalgleish and Hartnett 2006).

Active and dormant bud dynamics for *P. smithii* were similar to other C_3 species in tallgrass prairie, with small accumulations of dormant bud banks and large seasonal fluctuations of overall bud dynamics (Ott and Hartnett 2012). These results suggest bud banks of C_3 species closely track intra- and inter-annual environmental changes and show more dynamic tiller recruitment than C_4 species under variable environmental conditions.

Data supported the hypothesis that soil moisture and soil temperature are driving factors behind bud development and support previous findings of significant growth depending on soil moisture and temperature thresholds (Cable 1975; Kaspar and Bland 1992; Ott and Hartnett 2012). Furthermore, these results indicated that soil temperature more accurately predicted bud development of C_4 species, whereas, soil moisture more accurately predicted bud development of C_3 species. Even though the soil surface reached extreme temperatures, bud development progressed rapidly after 40 °C, which aligns

with previous soil temperature threshold work on corn (*Zea mays*, L.) (Weaich et al. 1996), sorghum (Harris 1996; Townend et al. 1996), and rangelands (Townsend and Fuhlendorf 2010). The majority of data was on the upper end of the soil temperature range and on the lower end of the soil moisture range. Therefore, that type of distribution indicates other factors (e.g., exogenous vs. endogenous growth cues) were also contributing to bud development. Endogenous growth cues include hormonal control over bud development that involve complex interactions among auxin, cytokinin, strigolactone, and bud development stages to determine bud activity, dormancy, and tiller emergence (Dun et al. 2006; Waldie et al. 2010).

Understanding the patterns and factors influencing of axillary bud development dynamics is important to developing a mechanistic and predictive understanding of grassland response to disturbances such as fire, grazing, and drought. Descriptions of bud bank phenology can provide insight to the timing and action of disturbances and its potential impacts on tiller population dynamics. Linking aboveground phenology with belowground bud phenology illustrates potential mechanisms regulating bud development and maintenance, which may ultimately determine plant community populations.

Acknowledgements

Literature cited

Angus, J.F., and M.W. Moncur. 1977. Water stress and phenology in wheat. *Australian Journal of Agricultural Research* 28: 177-181.

Aronson, J., J. Kigel, A. Shmida, and J. Klein. 1992. Adaptive phenology of desert and Mediterranean populations of annual plants with and without water stress. *Oecologia* 89: 17-26.

Benson, E.J., and D.C. Hartnett. 2006. The role of seed and vegetative reproduction in plant recruitment and demography in tallgrass prairie. *Plant Ecology* 187: 163-178.

Briske, D.D. 1991. Developmental morphology and physiology of grasses. In: R.K. Heitschmidt and J.W. Stuth [EDS.]. Grazing management: an ecological perspective. Portland, OR, USA: Timber Press. p. 85-108.

Busso, C.A., R.J.Mueller, and J.H. Richards. 1989. Effects of drought and defoliation on bud viability in two caespitose grasses. *Annals of Botany* 63: 477-485.

Cable, D.R. 1975. Influence of precipitation on perennial grass production in the semidesert Southwest. *Ecology* 56: 981-986.

Carter, D.L., B.L. VanderWeide, and J.M. Blair. 2012. Drought-mediated stem and belowground bud dynamics in restored grasslands. *Applied Vegetation Science* 15: 470-478.

Dalgleish, H.J., and D.C. Hartnett. 2006. Below-ground bud banks increase along a precipitation gradient of the North American Great Plains: a test of the meristem limitation hypothesis. *New Phytologist* 171: 81-89.

Dun, E.A., B.J. Ferguson, and C.A. Beveridge. 2006. Apical dominance and shoot branching. Divergent opinions or divergent mechanisms? *Plant Physiology* 142: 812-819.

Flemmer, A.C., C.A. Busso, and O. A. Fernández. 2002. Bud viability in perennial grasses: Water stress and defoliation effects. *Journal of Range Management* 55: 150-163.

Harris, D. 1996. The effects of manure, genotype, seed priming, depth, temperature, and date of sowing on the emergence and early growth of Sorghum bicolor (L.) Moench in semi-arid Botswana. Soil and Tillage Research 40: 73-88.

Hendrickson, J.R. and D.D. Briske. 1997. Axillary bud banks of two semiarid perennial grasses: occurrence, longevity, and contribution to population persistence. *Oecologia* 110: 584-591.

Herms, D.A., and W.J. Mattson. 1992. The dilemma of plants: To grow or defend. *The Quartely Review of Biology* 67: 283-335.

Kaspar, T.C., and W.L. Bland. 1992. Soil temperature and root growth. *Soil Science* 154: 290-299.

Littell, R.C., G.A. Milliken, W.W. Stroup, R.D. Wolfinger, and O. Schabenberger. 2006. SAS® for Mixed Models, second edition. Cary, NC: SAS Institute Inc

McIntyre, G.I. 1967. Studies on bud development in rhizome of Agropyron repens. 1. Influence of

      temperature, light intensity, and bud position on pattern of development. *Canadian Journal of*

      Botany 48: 1903-1909.

McKendrick, J.D., C.E. Owensby, and R.M. Hyde. 1975. Big bluestem and Indiangrass vegetative

      reproduction and annual reserve carbohydrate and nitrogen cycles. *Agro-Ecosystems* 2: 75-93.

Moore, K.J., L.E. Moser, K.P. Vogel, S.S. Waller, B.E. Johnson, and J.F. Pedersen. 1991. Describing and

      quantifying growth stages of perennial forage grasses. *Agronomy Journal* 83: 1073-1077.

Ott, J.P., and D.C. Hartnett. 2011. Bud production and dynamics of flowering and vegetative tillers in

      Andropogon gerardii (Poaceae): The role of developmental constraints. *American Journal of*

      Botany 98: 1293-1298.

Ott, J.P., and D.C. Hartnett. 2012. Contrasting bud bank dynamics of two co-occurring grasses in

      tallgrass prairie: implications for grassland dynamics. *Plant Ecology* 213: 1437-1448.

Russell, M.L., L.T. Vermeire, N.A. Dufek, and D.J. Strong. 2013. Fire, defoliation, and competing species

      alter *Aristida purpurea* biomass, tiller, and axillary bud production. *Rangeland Ecology and*

      Management 66: 290-296.

Townend, J., P.W. Mtakwa, C.E. Mullins, and L.P. Simmonds. 1996. Soil physical factors limiting

      establishment of sorghum and cowpea in two contrasting soil types in the semi-arid tropics. *Soil*

      and Tillage Research 40: 89-106.

Townsend, E.E. II, and S.D. Fuhlendorf. 2010. Evaluating relationships between spatial heterogeneity and

      the biotic and abiotic environments. *The American Midland Naturalist* 163: 351-365.

USDA, NRCS. 2013. The PLANTS database. Available at: http://plants.usda.gov. Accessed 7

      September 2013.

USDA, NRCS. 2013. Ecological Site Description, Northern Rolling High Plains. Available at:

      https://esis.sc.egov.usda.gov. Accessed 7 September 2013.

Van Noordwijk, A.J., and G. De Jong. 1986. Acquisition and allocation of resources: Their influence on

      variation in life history tactics. *American Naturalist* 128: 137-142.

Vermeire, L.T., R.K. Heitschmidt, and M.J. Rinella. 2009. Primary productivity and precipitation use efficiency in mixed-grass prairie: a comparison of Northern and Southern U.S. Sites. *Rangeland Ecology and Management* 62: 230-239.

Volaire, F. 1995. Growth, carbohydrate reserves and drought survival strategies of contrasting *Dactylis glomerata* populations in a Mediterranean environment. *Journal of Applied Ecology* 32: 56-66.

Waldie, T., A. Hayward, and C.A. Beveridge. 2010. Axillary bud outgrowth in herbaceous shoots: How do strigolactones fit into the picture? *Plant Molecular Biology* 73: 27-36.

Weaich, K., A. Cass, and K.L. Bristow. 1996. Pre-emergent shoot growth of maize (*Zea mays*, L.) as a function of soil strength. *Soil and Tillage Research* 40: 3-23.

Western Regional Climate Center. 2013. Western U.S. climate historical summaries. Available at http://www.wrcc.dri.edu/cgi-bin/cliMAIN.pl?mt5690. Accessed 22 August 2013.

Wiles, L.J., G. Dunn, J. Printz, B. Patton, and A. Nyren. 2011. Spring precipitation as a predictor for peak standing crop of mixed-grass prairie. *Rangeland Ecology Management* 64: 215-222.