

VENTENATA (*VENTENATA DUBIA*) CONTROL TREATMENTS ON
THE CROW RESERVATION

by

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A thesis submitted in partial fulfillment
of the requirements for the degree

of

Master of Science

in

Land Resources and Environmental Sciences

MONTANA STATE UNIVERSITY
Bozeman, Montana

June 2023

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DEDICATION

For my grandparents and mom,

Grady Hunts Arrow
Alice Hunts Arrow
Rosalie Hunts Arrow

ACKNOWLEDGEMENTS

I am very thankful for my advisors, Dr. Jane Mangold and Dr. Scott Powell. Thank you both for the guidance, encouragement, kindness, and patience throughout my research project. This has been an enjoyable learning experience. I would also like to thank my committee member Dr. Jessica Mitchell, for the collaboration and insight on her work mapping ventenata on Crow tribal lands and help with direction for my remote sensing data. Thank you to the Mangold lab for the support, feedback on presentations, discussions, and the laughs. Thank you to the Statistical Consulting and Research Services at MSU for their help. I would also like to thank Robert Demery, David Hopkins, and Jarvis Gust at the Bureau of Indian Affairs for helping me find and set up my research sites. I would like to acknowledge the Montana Noxious Weed Trust Fund, Alfred P. Sloan Foundation, and Hopa Mountain for financially supporting my graduate research. Thank you to my friends and family for all their encouragement while I pursued higher education. Lastly, to my mom and grandparents, Grady and Alice, thank you for your love, prayers, and support.

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ABSTRACT

Ventenata (Ventenata dubia) is a non-native winter annual grass that has been of increasing concern in southeastern Montana. Research has shown that ventenata can increase rapidly, lower forage production, and reduce biodiversity. This project is located in southeastern Montana, in Bighorn County on the Crow Reservation. Two studies were conducted to understand control options and monitoring of those treatments post-treatment. A field study tested two herbicides and a soil amendment for the management of ventenata. At four sites, indaziflam and imazapic at two water carrier rates and two rates of an organic soil nutrient amendment were tested using a split-plot randomized block design. The water carrier rates were meant to mimic aerial and ground applications. Herbicides were applied using a hand-held boom sprayer pressurized by CO₂. Soil amendment was hand-broadcasted. In late June 2022 (first growing season post-treatment), sampling consisted of randomly placing 3, 20 cm x 50 cm frames in each split-plot and estimating cover by species along with litter and bare ground. Imazapic and indaziflam provided the highest reduction of ventenata, regardless of water carrier rate. Across the four sites, imazapic reduced ventenata cover to <1% while indaziflam reduced cover to 4%, compared to the control which was 38%. The soil amendment reduced ventenata to 25% at two sites, suggesting it may not be as promising of a control method as the herbicides. Application rates for all treatments did not differ, suggesting that aerial application of the herbicides may provide just as good of control as ground application. This is encouraging for the prospect of managing ventenata aurally. At one of the sites, a remote sensing time series study using an Unmanned Aerial Vehicle (UAV) with a multispectral sensor was used to understand differences in the Normalized Difference Vegetation Index (NDVI) between herbicide sprayed and non-sprayed plots. Findings indicate that there is a shift in NDVI in late June where sprayed plots peak in NDVI and remain green longer into the season than non-sprayed plots. This study provides control options that land managers in southeastern Montana can consider using for ventenata management.

CHAPTER ONE

INTRODUCTION AND
RESEARCH OBJECTIVESLiterature ReviewRangelands

Rangelands are one of earth's major ecosystems (Lund, 2007). Rangelands are lands on which the indigenous vegetation is predominately grasses, grass-like plants, forbs, and shrubs or dispersed trees, with grazing by domestic livestock and wildlife as the most common ecological management process (Range Resources, NRCS). Existing plant communities can include both native and introduced plants. Rangelands are a primary source of forage for domestic livestock and wildlife (Range Resources, NRCS). Rangelands used for livestock production are primarily managed by cow-calf producers, and operations mainly maintain a herd of beef cows for raising calves (Havstad et al., 2007; Knight, 2018). In the U.S. rangelands cover about 30% of the entire land cover (Havstad et al., 2007; Range Resources, NRCS). In Montana, approximately 70% percent is made up of rangeland and pasturelands, making grazing lands Montana's largest natural resource (NRCS, Montana). In southeastern Montana, grasses typically comprise the greatest canopy cover, and western wheatgrass (*Pascopyrum smithii* (Rydb.) Barkworth & D.R. Dewey) is usually dominant. Other native species include thickspike wheatgrass (*Elymus lanceolatus* (Scribn. & J. G. Sm.) Gould), green needlegrass (*Nassella viridula* (Trin.) Barkworth), blue grama (*Bouteloua gracilis* (Kunth) Lag. Ex Griffiths), and needle and thread (*Hesperostipa comata* (Trin. & Rupr.) Barkworth) (Luna & Vance, 2017). Common forbs within

this area of the state include yarrow (*Achillea millefolium* L.), scarlet globemallow (*Sphaeralcea coccinea* (Nutt.) Rydb.), western sagewort (*Artemisia ludoviciana* Nutt.), boreal sagewort (*Artemisia frigida* Willd.), silver lupine (*Lupinus argenteus* Pursh), fuzzy beardtongue (*Penstemon eriantherus* Pursh), shining penstemon (*Penstemon nitidus* Douglas ex Benth.), prairie cinquefoil (*Potentilla gracilis* Douglas ex Hook.), Missouri goldenrod (*Solidago missouriensis* Nutt.) and dalea (*Dalea* spp.) (Luna & Vance, 2017). Grasses were typically used by large herbivores such as bison (*Bison bison* Linnaeus.), but since European settlement, herbivores such as cattle and sheep have been the primary grazers of the vegetation (Luna & Vance, 2017).

Invasive Plants and Impacts

Invasive species are nonnative species whose introduction is likely to cause economic or environmental harm or harm to human, animal, or plant health (Beck et al., 2008). They are species that arrive with human assistance and have the potential to become successfully established or naturalized, and spread into new localized natural habitats (Kerns, 2012; Simberloff, 2013). An estimated 5,000 nonnative plant species have been introduced and established and now exist in U.S. ecosystems (Pimental et al., 2005; Simberloff, 2013). Not all of these species are invasive. Many nonnative species have recently arrived and presently occupy only a portion of available habitats. Given enough time, they have the potential to spread widely (Kerns, 2012). Familiar examples include the annual grass cheatgrass (*Bromus tectorum* L.) which has invaded significant areas of sagebrush-steppe and dry forests in the western U.S. and the nonnative vine kudzu (*Pueraria montana* var. *lobata*) in the southeastern U.S. (D'Antonio and Vitousek, 1992; Simberloff, 2013). While most definitions of invasive plants only consider

nonnative species, native species may be considered invasive by some (Carey et al., 2012). For example, juniper (*Juniperus* spp.) species in the western U.S. have historically expanded their range and are considered invasive in certain ecosystems (Kerns, 2012; Miller et al., 2005).

Rangeland invasive weeds can have a significant impact on both humans and the environment. Their impact on human activities can be associated with livestock production, including interfering with grazing practices, lowering yield and quality of forage, increasing costs of managing and producing livestock, and slowing animal weight gain (DiTomaso, 2000). In addition, infestations can reduce recreational land values (DiTomaso, 2000). Weed infestations can reduce plant diversity, threaten rare and endangered species, reduce wildlife habitat and forage, alter hydrology of rivers, lakes, wetlands and alter fire frequency and intensity (Beck et al., 2008; Brooks, 2004; Bradley et al., 2018; D'Antonio & Vitousek, 1992; DiTomaso, 2000; Whisenant, 1990). Rangeland weeds have been shown to reduce species richness, plant diversity, and community productivity in a number of areas (DiTomaso, 2000; Jones et al., 2018; Rikard & Cline 1980; Wallace et al., 1992). In spotted knapweed (*Centaurea maculosa* Lam.) infested rangeland, elk (*Cervus canadensis*) use was reduced by 98% compared with bunchgrass-dominated sites (Sheley et al., 1998; Thompson, 1996). By comparison, when spotted knapweed was removed from a historic elk winter range in western Montana, elk use increased (Thompson, 1996). Billions of dollars are spent every year to mitigate invasive plants or control their impacts (Pimental et al., 2005). It is estimated that biological invasions in the U.S. cost the economy approximately \$120 billion annually, with agricultural weeds costing about \$27 billion in losses and management costs (Pimentel et al., 2005; Simberloff, 2013). These estimates include invasive plants beyond rangeland weeds, but one study local to Montana

rangeland estimated that the combined cost of invasive noxious weed management plus forage losses due to these plants on privately owned rangeland was \$7,243 annually for an average size grazing unit (i.e., 2,046 ha) (Mangold et al., 2018).

Invasive Annual Grasses

Annual grass invasions started with the arrival of Europeans to the North American continent (D'Antonio & Vitousek, 1992). Nonnative annual grass invasions are one of the most transformative stressors in rangelands of the western U.S. (D'Antonio & Vitousek 1992; Germino et al., 2020). When nonnative annual grasses and livestock were introduced, native perennial grasses, which do not recover well from intense grazing, were overgrazed and quickly replaced by winter annual grasses (Callihan & Evans, 1991; DiTomaso, 2000; Young & Longland, 1996). Invasives annual grasses can affect Northern Great Plains grasslands with their fall germination and early spring growth, which gives them a head start over native species in competing for moisture and nutrients (Rinella et al., 2020, Symstad et al., 2021), and their early senescence can affect forage availability later in the growing season (Hart and Meador, 2021; Ogle et al., 2003; Symstad et al., 2021).

The expansion of cheatgrass (*Bromus tectorum* L.) throughout the Great Basin in the western U.S. is linked to the introduction of sheep and cattle (D'Antonio & Vitousek, 1992; Mack, 1981). Cheatgrass is especially notorious in the Intermountain West, where it is well known for increasing fire frequency, leading to replacement of native sagebrush-bunchgrass steppe with fire-prone annual grasslands (e.g., Balch et al., 2012; Brooks et al., 2016; D'Antonio & Vitousek, 1992). Cheatgrass is the most widespread invasive annual grass, but other annual grasses such as other species in the *Bromus* genus, medusahead (*Taeniatherum caput-medusae*

(L.) Nevski.), and *ventenata* (*Ventenata dubia*) are serious threats to different regions and are continuing to spread (Germino et al., 2020). In the past decade, the Wyoming Basin, Great Plains, and Rocky Mountains have begun to experience nonnative annual grass invasion.

Ventenata, in particular, has been expanding into new areas and has the potential to increase rapidly. Nicolli et al. (2020) documented a 300% increase in *ventenata* infestation over four years in the John Day Fossil Beds National Monument in central Oregon. The observations support Jones et al. (2018), who stated that *ventenata* invasion is expanding and represents an existential threat to the sagebrush steppe.

Ventenata

Ventenata [*Ventenata dubia* (Leers) Coss], common name *ventenata* or wiregrass, is a nonnative winter annual grass introduced to North America in the 1950's in Washington state. It has since been documented in other western states, as well as southern Canada (Barkworth et al. 1993; Scheinost et al. 2008). *Ventenata* originates from northern Africa and southern Europe. Of the five known species of *ventenata*, *V. dubia* is the only species established in North America (Scheinost, 2008). Its range is rapidly expanding into new ecosystems such as the sagebrush steppe and the Great Plains (Jones et al., 2020). It is now well established in pasturelands, croplands, and a variety of ecosystems including grasslands, sagebrush steppe, ponderosa pine forests, and woodlands (Averett et al., 2016; Fryer, 2017; Innes, 2022; Jones et al., 2018; Tortorelli et al., 2020). In range, pasture, and natural areas, *ventenata* is associated with decreased plant community diversity, low forage production, and increased soil erosion due to the species' shallow root structure (Jones et al., 2018, 2020; Scheinost, 2008; Wallace et al., 2015). It has also caused up to 50% yield loss in croplands across eastern Idaho and Washington

(Wallace & Prather, 2015, 2016). It has only recently been documented in the Great Plains, where its impact on forage production and biodiversity are still uncertain. In northern Wyoming, Hart & Meador (2021) found that species richness and diversity were unaffected by ventenata removal one year after treatment, and there was improved forage quality and quantity. Ventenata was first documented in Montana in the 1990's and has since been documented in 24 Montana counties where it occurs on at least 22,258 hectares (Harvey & Mangold, 2018; Lesica et al., 2012). Ventenata was listed as a noxious weed in Montana in 2019 (Montana Department of Agriculture, 2019).

Identification

Ventenata has a shallow root system (Figure 1.1), with one to few tillers, and produces 15 to 35 seeds per plant. The plant has slim, erect culms from 10 to 46 cm (4 to 18 in) tall with microscopic hairs that give the appearance of being smooth (Scheinost, 2008). Seedling leaves are in-rolled or lengthwise folded and appear very narrow. It has a long, narrow, membranous ligule and dark red to black nodes (Figure 1.1). The inflorescence is an open panicle, appearing silvery green but rapidly maturing to a yellowish-tan color (Scheinost, 2008). The seed has a bent and twisted awn, and the spikelet contains three to four florets (Innes, 2022; Scheinost, 2008).



Figure 1.1. Ventenata when mature with shallow roots (left), Photo by Inna Smith. Reddish-black node and membranous ligule (right), Photo by Pamela Scheinost.

Biology and Ecology

Seeds of ventenata are produced May through June, depending on location, and about one month following annual *Bromus* species (Scheinost, 2008). Seeds of ventenata typically germinate in the fall about two weeks after cheatgrass (Innes, 2022; Pavek et al., 2011; Wallace et al., 2015). Most seedlings emerge within six weeks once germination begins (Wallace et al., 2015). This generally corresponds to an October to November time period (Wallace et al., 2015). Seedling emergence rates decline in November and are negligible in the following three months. On Conservation Reserve Program (CRP) land in Washington and Idaho, 95% of the seedlings emerged in the fall (Wallace et al., 2015). Seedlings can emerge in the spring in areas where fall germination is not favorable, such as in more highly managed systems like Timothy (*Phleum pratense* L.) hay fields (Wallace et al., 2015). Inflorescences develop and flower in June and produce seeds by early July (Wallace et al., 2015). The seeds mature by early August, which is typically about one month later than seeds of cheatgrass and other annual bromes (Fryer, 2017;

Innes, 2022; Prather, 2014; Scheinost, 2008). *Ventenata* dries out earlier in the season than native perennial grasses but later than associated nonnative annual grasses (Fryer, 2017; Kerns et al., 2015). Seed shatter and plant death occurs in July or August, when awns bend and twist (Fryer, 2017; Scheinost, 2008). Recent studies into the life history traits of *ventenata* have taken place in the grassland ecosystems of the Pacific Northwest (PNW). One study found that the species had a greater seedling survival rate when under complete (100%) litter cover, revealing positive feedback with thatch-maintained microsite moisture (Wallace et al., 2015). Higher *ventenata* litter levels can increase *ventenata* seedling emergence and survival when compared with bare surface during drier or colder fall growing seasons (Wallace et al., 2015). A small fraction of *ventenata* seed banks (< 1%) may remain persistent, or germinable, for up to three years at shallow soil depths (2 cm). Wallace et al. (2015) found that one month after burial, a significant portion of an artificial seed bank was lost to germination; total germination was 82% and 79% at burial depths of 2 and 8 cm, respectively. They found a small but measurable (< 1% of seed bank) level of seed persistence at the 2 cm depth 13, 25, and 37 months after burial, whereas no seed persistence was observed at the 8 cm depth (Wallace et al., 2015).

Chemical Control of Rangeland Invasive Plants

Herbicides are the most common tool for weed control (DiTomaso, 2000; Vallentine, 1981). Most herbicides kill plants by disrupting or altering one or more of their metabolic processes (Tu et al., 2001). Timing of herbicide applications can determine the effectiveness of the treatment. Pre-emergent herbicides are those applied to the soil before the plant germinates, and either disrupt germination or kill the germinating seedling. Post-emergent herbicides are those that are applied directly to already established plants and/or soil. Some herbicides are

effective both before (pre-emergent) and after (post-emergent) annual grass germination (Clark et al. 2019; Koby et al., 2019; Mangold et al., 2013; Sebastian et al., 2016, 2017; Tu et al., 2001; Wallace & Prather 2016).

Herbicides can be applied to rangelands by fixed-wing aircraft, helicopter, ground applicators, and backpack sprayers (Vallentine, 1981). Broadcast spray application has been the most used method on rangelands. Broadcast spray applications can be made by ground or by aerial application, and water is the most used carrier for an herbicide (Vallentine, 1981). When herbicides are applied by ground, a spray volume of 26 L/ha (10 gal/acre) is common but may vary from 13 to 104 L/ha (5 to 40 gal), depending upon need (Vallentine, 1981). With aerial application, spray volume is reduced to 3 to 8 L/ha (1 to 3 gal/acre) to minimize weight being carrier by the aircraft (Vallentine, 1981). Ground application of herbicides provide more water when compared to aerial applications (Butts et al., 2020; Vallentine, 1981). The advantages of using ground applications include more water applied with the herbicide to improve coverage, less drifting with wind, smaller acreages, commercial equipment not often required, and it is safer for applicators. The advantages of aerial application include faster coverage, adapted to rough ground and steep slopes, lower cost per acre on most large acreages, better coverage of tall or dense brush, and no mechanical disturbance of soil or vegetation (Vallentine, 1981).

Chemical Control of Ventenata

Two herbicides proposed for controlling annual grasses, including ventenata, are indaziflam (Rejuvra®) and imazapic (Plateau®). Indaziflam controls winter annual grasses in rangeland and natural areas by inhibiting seedling establishment (Sebastian et al., 2017). It is a root inhibiting herbicide that remains near the top of the soil. It inhibits germinating seedlings,

leaving desirable perennial grasses, forbs, and shrubs unaffected (Bayer, 2020). Indaziflam could be applied alone pre-emergent; however, having limited post-emergent activity, indaziflam would need to be used in combination with another herbicide to control already emerged annual grasses in the late fall or early spring (Sebastian et al., 2017). In two different studies in Colorado, indaziflam controlled cheatgrass for up to three years and did not impact perennial species richness or abundance (Clark et al. 2019, Sebastian et al., 2016). In southwestern Montana, indaziflam alone provided 67 – 98% control of ventenata 8 – 23 months after treatment (Harvey, 2019). In that same study, indaziflam combined with each of four other commonly used herbicides, including imazapic, provided longer term control ranging from 89 – 100% (Harvey, 2019). Indaziflam provides soil residual control for three or more years compared with imazapic, which only provides one or two-years of control (Sebastian et al., 2016, 2017). Benefits of indaziflam application include consistent long-term control and being approved for application on lands that are grazed by domestic livestock (Bayer, 2020).

Imazapic kills plants by inhibiting the production of branched chain amino acids, which are necessary for protein synthesis and cell growth (Tu et al., 2001). Imazapic is commonly recommended for control of annual grasses, however performance has been inconsistent (Mangold et al., 2013), and it can injure desirable perennial grasses (Sebastian et al., 2016). Imazapic can provide post-emergent winter annual grass control when applied at the seedling stage (Mangold et al, 2013; Sebastian et al., 2016; Wallace & Prather 2016). Specifically, imazapic applied early post-emergent (one to two leaf stage) provided the most consistent, short-term control of cheatgrass (Mangold et al., 2013). However, imazapic does not provide consistent control beyond one year after treatment, resulting in rapid reinvasion of treated areas

through the soil seed bank (Harvey, 2019; Sebastian et al., 2016; Wallace & Prather, 2015). In Colorado, imazapic provided similar control of cheatgrass when compared to indaziflam one year after treatment, then control was reduced to about 60% and 10% two and three years after treatment, respectively (Sebastian et al., 2016). In southwestern Montana, imazapic provided 63% control of ventenata eight months after treatment, before control was reduced to 20% and 11% 11 and 23 months after treatment, respectively (Harvey, 2019).

In a study from 2021 in northern Wyoming, aerial application of imazapic and indaziflam by helicopter was shown to reduce ventenata cover to nearly 0% one year after application (Hart & Meador, 2021). Herbicides were delivered in a total solution of 46.8 L/ha. In studies with higher water carrier rates typical for ground applications, both imazapic and indaziflam controlled ventenata with total solutions ranging between 182 and 187 L/ha, (Harvey, 2019; Koby et al., 2019; Sebastian et al., 2016, 2017; Wallace & Prather, 2016). Spraying herbicide where ventenata covers a large area, aerial application may be more economical than ground application.

While herbicides like indaziflam and imazapic are commonly recommended to control invasive annual grasses, their performance may be inconsistent (Sebastian et al., 2016), and some managers prefer non-synthetic chemical alternatives. The soil amendment NutraFix™ is a new non-chemical option that is piquing the interest of weed managers and is therefore being explored in this project along with herbicides. NutraFix™ is composed of micronutrients used to promote soil health and growth of desirable vegetation (NutraFix Soils, 2020). Case studies conducted in Montana have had mixed results, but some treated areas demonstrated increased perennial grass cover (NutraFix Soils, 2020). This soil amendment may be slower acting than an

herbicide and may need at least a couple of years post-treatment to see positive results (NutraFix Soils, 2020). There is a lack of published, replicated field studies on the performance of this organic product. However, it may potentially provide land managers with a non-chemical option for controlling annual grass-invaded sites, and further testing is warranted.

Role of Remote Sensing in Invasive Plant Management

Remote sensing is the process of detecting and monitoring the physical characteristics of an area on the earth's surface by measuring its reflected and emitted radiation at a distance (typically from satellite or aircraft) (USGS, 2017). Remote sensors can be either active or passive. Active sensors use internal stimuli to collect data about earth, such as a laser-beam remote sensing system which projects a laser onto the surface of earth and measures the time that it takes for the laser to reflect back to its sensor. Passive sensors record natural energy that is reflected or emitted from the earth's surface, and the most common source of radiation detected by passive sensors is reflected sunlight (NOAA, USGS, 2017).

Remote sensing can be a tool to map vegetation distribution across large portions of rangelands, which is not often achievable with traditional field-based methods (Bateman, 2020; Rocchini et al., 2015). For instance, broad-scale distribution maps of cheatgrass over western rangelands have been developed by researchers (Bradley & Mustard, 2006; Clinton et al., 2010; Boyte et al., 2015). Often used in accordance with field-based data, remotely sensed data have been used in mapping and monitoring the distribution and abundance of invasive plant species, including annual grasses (Bateman et al., 2020; Bradley, 2014; Bradley & Marvin, 2011). A challenge for invasive plant management is identifying and preventing invasions before they expand into new landscapes. To prevent invasions, managers rely on early detection and rapid

response (EDRR) or identifying and eradicating new infestations before they can gain a permanent foothold in a new landscape (Simberloff, 2013). Remote sensing can help with these challenges by detecting and monitoring invasions (Bradley, 2014; Vaz et al., 2018), creating distribution maps to inform EDRR of new invasions (Westbrooks, 2004), and supporting weed management decisions (Shaw, 2005).

Multispectral remotely sensed imagery contains a combination of bands that creates a composite image to be used for interpretation and analysis (Huang et al., 2021). Image band transformations have become a common practice to generate new images from two or more image bands for information extraction. The individual bands in the band composite can be transformed to highlight certain features and patterns, such as vegetation that is healthy or unhealthy using vegetation indices (VIs) (Huang et al., 2021). The new images generated this way enhance representations to ground objects such as vegetation. There are more than one hundred vegetation indices that have been derived from multispectral imagery, the most common one being the Normalized Difference Vegetation Index (NDVI) (Huang et al., 2021; Xue & Su, 2017).

The NDVI is an index that provides a measure of vegetation greenness and plant health. It is calculated as a ratio between near-infrared (NIR) and red (RED) reflectance values as: $(\text{NIR} - \text{RED}) / (\text{NIR} + \text{RED})$ (Jackson & Huete, 1991; USGS, 2017). NDVI values are unitless and range from -1 to 1. Densely vegetated areas will tend toward positive values, whereas water, rocks, sand, and concrete surfaces will be represented by near zero or negative values (Jackson & Huete, 1991; Huang et al., 2021). Healthy vegetation will absorb most of the visible light and reflect a large portion of the near-infrared light, whereas unhealthy vegetation will reflect more

visible light and less near-infrared light (Ndayisaba et al., 2016). Bare soils reflect moderately in both the red and near-infrared portion of the electromagnetic spectrum (Jackson & Huete, 1991). The use of NDVI in monitoring vegetation is appealing because of its ability to delineate vegetative stress, which has great appeal in commercial agriculture and land-use studies. NDVI has been shown to be positively correlated with plant productivity and has been used to estimate crop yields, pasture performance, and rangeland carrying capacities (Huang et al., 2021; Ndayisaba, 2016; Turvey & McLaurin, 2012; USGS, 2017).

The phenology of a plant during the growing season, including life-cycle events such as emergence, flowering, or senescence, emphasizes the spectral differences between vegetative types and can be detected with remote sensing platforms (Bateman, 2020; Bradley, 2014). Phenological differences can facilitate satellite detection of nonnative invasive plants such as grasses (Bradley, 2014). Annual grasses begin growing, or “green up,” earlier than perennial grasses, producing a distinct pulse of greenness in the early spring. This pulse has been exploited to identify the location of cheatgrass by using a difference of the spring image with peak cheatgrass greenness versus early summer imagery where cheatgrass is senescent (Peterson, 2005). As cheatgrass becomes more abundant, the timing of peak greenness shifts, and this change is observable with satellite image time series (Bradley et al., 2018; Peterson, 2005). Additionally, cheatgrass responds strongly to precipitation (Bradley et al., 2018; Stewart & Hull, 1949), which is highly variable year to year. This inter-annual variability in greenness is also detectable with satellite time series and relates to the distribution of cheatgrass (Bradley & Mustard, 2005; Bradley et al., 2018).

There are tradeoffs with remotely sensed data between spatial extent (size of the image), spatial resolution (pixel size), spectral resolution (number and range of visible and infra-red bands) and temporal resolution (frequency of data acquisition) (Bradley, 2014). Larger spatial extents provided by satellite imaging platforms allow for more extensive mapping of invasive plants and ultimately provide more distribution data to inform spatial models of invasion risk. Satellite platforms have been used for cheatgrass mapping, and time series images have helped to detect greenness (Bradley & Mustard 2005; Bradley et al., 2018; Peterson, 2005). However, spatial resolution is typically coarse with satellite platforms, where small infestations will not be noticed, which can limit detection to only widespread and abundant infestations (Bradley, 2014). Finer spatial resolution makes it more likely that individual species and early infestations can be detected (Bradley, 2014). Satellite platforms such as Landsat enable free access to multispectral data; however, disadvantages with satellite based remote sensing include revisit time, (for Landsat is 16 days), which can present challenges for agricultural monitoring applications (USGS, 2017; Xue & Su, 2017). Furthermore, data capture may be unusable for overcast days (Xue & Su, 2017). Unmanned Aerial Vehicles (UAVs) can help mitigate some of the issues associated with satellite imagery, such as low spatial resolution and longer temporal resolution (frequency of data obtained). Advantages of UAV data are their high spatial resolution and flexibility to capture imagery. UAVs can complement the techniques used in satellite platforms and can be used for image classification (Elkind et al., 2019; Huang et al., 2019; Weisberg et al., 2021). UAVs are an expanding area in remote sensing, driven by both academic and commercial successes (Yao et al., 2019). The rise of NDVI applications is aided by advancements and widespread use of UAVs (Yao et al., 2019).

One study used multitemporal UAV imagery to distinguish multiple co-occurring species of invasive annual grasses from each other and from native vegetation in Nevada, based on phenological differences in the timing of plant life phases over the course of a growing season (Weisberg et al., 2019). Using eight different dates of UAV imagery, a species-specific classification was best using three flight dates: 19 May, 1 June, and 20 July (Weisberg et al., 2021). Imagery derived from readily deployable UAVs offers high-resolution data over carefully timed acquisition dates during the growing season. Species can be separated based on varied phenological stages within the same image, or when phenology differs between images (Singh et al., 2018; Weisberg, 2021). Classification accuracy can also be improved with multi-date imagery. For example, in southwestern Montana, accuracy increased 5-10% between a single and multiple UAV flights over the growing season (Wood et al., 2022). Within herbaceous and sagebrush classes, multi-temporal analyses identified differences in the seasonal timing of green up and senescence within herbaceous and sagebrush classes (Wood et al., 2022). While phenology has proven successful in separating cheatgrass and medusahead in a pixel-based classification (Weisberg et al., 2021), there has not been much remote sensing research on ventenata. Ventenata can be just as problematic as medusahead and has the potential to be as problematic as cheatgrass (Innes, 2022). There is potential to distinguish ventenata from other annual grasses, and further research is needed for detecting and mapping ventenata using remote sensing approaches.

Project Justifications and Objectives

The first goal of this research was to test and provide control options for the invasive annual grass ventenata in southeastern Montana on the Crow Reservation. It is a concern for

livestock producers, landowners, and land managers in Montana where it has been designated as a Priority 2A noxious weed (Montana Department of Agriculture, 2019). Since this species is relatively new to Montana, little information about control options is available, specifically for this part of the state, and more information is needed to contain its spread and protect ventenata-free areas. Chapter 2 describes a field study that tested the effectiveness of indaziflam (Rejuvra®, BAYER, Environmental Science) and imazapic (Plateau®, BASF Corporation), applied at two water carrier rates, and the soil amendment (NutraFix™, Edaphix™), applied at a high and low rate, for reducing ventenata on grasslands in southeastern Montana. Over one growing season post-treatment, canopy cover of ventenata, other vegetation, bare ground and litter was assessed across a split-plot design of the two herbicide treatments, the soil amendment, and a non-sprayed control. Based on previous studies (Harvey, 2019; Hart & Mealor, 2021; Sebastian et al., 2016, 2017), I hypothesized that indaziflam would provide the highest reduction of ventenata canopy cover, followed by imazapic and the soil amendment. I also hypothesized that the higher water carrier rate would reduce ventenata cover more so than the lower rate.

The second goal of this research was to utilize UAV remote sensing to capture and understand vegetation response to treatments using NDVI time series imagery. Chapter 3 describes a field study where an UAV with a multispectral sensor was used to detect differences in NDVI between sprayed and non-sprayed plots to better understand effectiveness of treatments. This was done by conducting weekly drone flights (24 May – 8 August 2022) at one of the study sites described in Chapter 2, focusing on NDVI time series imagery. I hypothesized that sprayed plots would have a higher NDVI longer into the season than non-sprayed plots due to the reduction of ventenata and an increase in perennial grasses, which stay green longer into the

season. Chapter 4 briefly summarizes the findings of my research and discusses implications for management. The fourth chapter also identifies research limitations and possible directions for future research.

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CHAPTER TWO

TESTING INDAZIFLAM, IMAZAPIC, AND A SOIL AMENDMENT FOR
CONTROL OF VENTENATA (*VENTENATA DUBIA*) IN SOUTHEASTERN MONTANAIntroduction

Ventenata (*Ventenata dubia*) is a non-native winter annual grass of increasing concern in southeastern Montana. This species originates from southern Europe and northern Africa. It was first documented in North America in Washington in the 1950's and has since been documented in other western states, as well as southern Canada (Barkworth et al., 1993; Scheinost et al., 2008). *Ventenata* was first documented in Montana in the 1990's and has since been documented in 24 Montana counties where it occurs on at least 22,258 hectares (Harvey & Mangold, 2018; Lesica et al., 2012). *Ventenata* was listed as a noxious weed in Montana in 2019 (Montana Department of Agriculture, 2019). In range, pasture, and natural areas, *ventenata* is associated with decreased plant community diversity, low forage production, and increased soil erosion due to the species' shallow root structure (Jones et al., 2018, 2020; Scheinost, 2008; Wallace et al., 2015). It has also caused up to 50% yield loss in croplands across eastern Idaho and Washington (Wallace & Prather, 2015, 2016). In the Great Plains region, *ventenata* impact is still uncertain. In northern Wyoming, Hart & Meador (2021) found that species richness and diversity were unaffected by *ventenata* removal one year after herbicide treatment, and there was improved forage quality and quantity.

Seeds of *ventenata* typically germinate in the fall about two weeks after cheatgrass (*Bromus tectorum*) (Fryer, 2017; Innes, 2022; Pavek et al., 2011; Wallace et al., 2015). Most

seedlings emerge within six weeks once germination begins (Wallace et al., 2015). This generally corresponds to an October to November time period (Wallace et al., 2015). Seedling emergence rates decline in November and negligible in the following three months. On Conservation Reserve Program (CRP) land in Washington and Idaho, 95% of the seedlings emerged in the fall growing season (Wallace et al. 2015). Seedlings can emerge in the spring in areas where fall germination is not favorable, such as in more highly managed systems like Timothy (*Phleum pratense* L.) hay fields (Wallace et al., 2015). Inflorescences develop in June and produce seeds by early July (Wallace et al., 2015). The seeds mature by early August, which is typically about one month later than seeds of cheatgrass and other annual bromes (Fryer, 2017; Innes, 2022; Scheinost, 2008). Ventenata dries out earlier in the season than native perennial grasses but later than associated nonnative annual grasses (Fryer, 2017; Kerns et al., 2015). Seed shatter and plant death occurs in July or August, when awns bend and twist (Beck, 2014; Fryer, 2017; Scheinost, 2008).

Although most ventenata germination and seedling emergence occurs in fall, some seedlings emerge in spring (Wallace et al., 2015). Less than 1% of seeds stay viable in the seed bank for up to three years (Wallace et al., 2015). Near Pullman, Washington, 82% and 79% of seeds germinated after 30 days of burial at 2 and 8 cm depth, respectively. Germination of seeds buried at two cm depth dropped to <1% after 13, 25, and 37 months of burial, and to 0% after 49 months. No seeds buried at eight cm deep germinated after 13, 25, 37, or 49 months (Wallace et al., 2015). Applying an herbicide that can control ventenata seed banks longer than three years may be key to controlling ventenata.

Herbicides are the most common tool for weed control in rangelands (DiTomaso, 2000; Mangold et al., 2018; Vallentine, 1981). Timing of herbicide applications can determine the effectiveness of the treatment. Most herbicides are applied either pre-emergence, before the weeds emerge from the soil, or post-emergence, after leaves of the weeds have emerged from the soil. Both pre- and post-emergent herbicides have been used on annual grasses, including *ventenata* (Harvey, 2019; Hart & Mealor, 2021; Koby et al., 2019; Mangold et al., 2013; Sebastian et al., 2016, 2017; Wallace & Prather, 2016). A newly approved pre-emergent herbicide that shows promise of multi-year control of invasive annual grasses, including *ventenata*, is indaziflam (Rejuvra®) (Harvey, 2019; Hart & Mealor, 2021; Sebastian et al., 2016, 2017). Indaziflam is typically applied pre-emergence to inhibit seedling root growth. Applications in Colorado showed 80 – 100% control of cheatgrass up to three years after treatment (Sebastian et al., 2016). Indaziflam also did not significantly impact species richness in that same study. In southwestern Montana, indaziflam alone provided 67 – 98% control of *ventenata* 8 – 23 months after treatment (Harvey, 2019). In that same study, indaziflam combined with each of the other commonly used herbicides imazapic, rimsulfuron, glyphosate and propoxycarbazone-sodium provided longer term control ranging from 89 – 100% (Harvey, 2019). Benefits of application include consistent long-term control and being approved for application on lands that are grazed by domestic livestock (Bayer, 2020).

Ventenata may be controlled with post-emergence herbicide applications in fall. However, this application timing is often a narrow window because of unpredictable weather conditions. Imazapic (Plateau®) is a post-emergent herbicide that kills plants by inhibiting the production of branched chain amino acids, which are necessary for protein synthesis and cell

growth (Tu et al., 2001). Imazapic can provide post-emergent winter annual grass control when applied at the seedling stage (Mangold et al., 2013; Sebastian et al., 2016; Wallace & Prather, 2016). Imazapic is commonly recommended for control of annual grasses, however performance has been inconsistent. For example, for cheatgrass, imazapic provided short-term (i.e., one growing season) control, depending on application period (pre-emergent, early post-emergent, post-emergent) (Mangold et al., 2013). Specifically, imazapic applied early post-emergent (one to two leaf stage) provided the most-consistent, short-term control of cheatgrass (Mangold et al., 2013). However, imazapic does not provide consistent control beyond one year after treatment, resulting in rapid reinvasion of treated areas through the soil seed bank (Harvey, 2019; Sebastian et al., 2016; Wallace & Prather, 2015). Furthermore, imazapic can injure desirable perennial grasses (Sebastian et al., 2016). In Colorado, imazapic provided similar control of cheatgrass when compared to indaziflam one year after treatment, then control was reduced to about 60% and 10% two and three years after treatment, respectively (Sebastian et al., 2016). In southwestern Montana, imazapic provided 63% control of ventenata eight months after treatment, before control was reduced to 20% and 11% 11 and 23 months after treatment, respectively (Harvey, 2019). Consistent long-term control is not achievable unless applied annually.

The most common formulation of rangeland herbicides is a concentrated liquid, with water being the common carrier for rangeland applications when sprayed on the target species (DiTomaso, 2000; Frost, 2022; Vallentine, 1981). Ground application of herbicides delivers more water with the herbicide than aerial applications (Butts et al., 2020; Frost, 2022). In a study in northern Wyoming, aerial application of imazapic and indaziflam applied alone by helicopter

was shown to reduce *ventenata* cover to nearly 0% one year after application (Hart & Mealor, 2021). Herbicides were delivered in a total solution of 46.8 L/ha. In studies with higher water carrier rates typical for ground applications, both imazapic and indaziflam controlled *ventenata* with total solutions ranging between 182 and 187 L/ha, (Harvey, 2019; Koby et al., 2019; Sebastian et al., 2016, 2017; Wallace & Prather, 2016). Spraying herbicide where *ventenata* covers a large area, aerial application may be more economical, but further investigation of the relationship between water carrier rate and herbicide performance, especially for indaziflam, is needed.

While herbicides such as glyphosate, imazapic, and rimsulfuron are commonly recommended to control invasive annual grasses, their performance is inconsistent (Sebastian et al., 2016) and some managers may prefer non-synthetic chemical alternatives. The soil amendment NutraFix™ is a new non-chemical option that is piquing the interest of weed managers and is therefore being explored along with herbicides. NutraFix™ is composed of micronutrients used to promote soil health and growth of desirable vegetation (NutraFix Soils, 2020). Case studies conducted in Montana have had mixed results, but some treated areas demonstrated increased perennial grass cover (NutraFix Soils, 2020). This soil amendment may be slower acting than an herbicide and may need a couple of years post-treatment to see positive results (NutraFix Soils, 2020). There is a lack of published, replicated field studies on the performance of this organic product. However, it may potentially provide land managers with a non-chemical option for controlling annual grass-invaded sites, and further testing is warranted.

This study tested the effectiveness of indaziflam (Rejuvra®, BAYER, Environmental Science) and imazapic (Plateau®, BASF Corporation), applied at two water carrier rates, and the

soil amendment (NutraFix™, Edaphix™), applied at a high and low rate, for reducing ventenata on grasslands in southeastern Montana. Over one growing season post-treatment, canopy cover of ventenata, other vegetation, bare ground and litter was assessed across a split-plot design of two herbicide treatments, a soil amendment, and a non-sprayed control. Based on previous studies (Harvey, 2019; Hart & Meador, 2021; Sebastian, et al., 2016, 2017), I hypothesized that indaziflam would provide the highest reduction for ventenata, followed by imazapic and the soil amendment. I also hypothesized that the higher rates would reduce ventenata cover more so than the lower rates.

Methods

This study was located across four sites in southeastern Montana on the Crow Reservation (Figure 2.1). Site one (Lodge Grass) is 29 km southwest of Lodge Grass, MT, (45°13'48.9"N, 107°37'36.3"W) at elevation of 1,020 m and mean annual precipitation of 35 to 45 cm; site two (Highway 212) is 25 km east of Crow Agency, MT, (45°31'44.1"N, 107°11'16.8"W) at elevation of 935 m and mean annual precipitation of 35 to 43 cm; site three (Shack Ranch) (45°25'27.2"N, 108°18'30.0"W) is 16 km east of Pryor, MT, at elevation of 1,244 m and mean annual precipitation of 30 to 35 cm; and site four (McCarty's) is 8 km east of Pryor (45°26'03.0"N, 108°24'29.6"W) at an elevation of 1,244 m and mean annual precipitation of 30 to 45 cm, (NRCS; Montana State Library) (Figure 2.1). These sites were selected from among known areas where ventenata was present and accessible. Therefore, we can only truly draw inference from our study to the specific sites we sampled and not the entire Crow Reservation. However, treatments at each site were randomly assigned, allowing inferences on herbicide and soil amendment effects on canopy cover (hereafter referred to as “cover”) of the vegetative

community. Each site is densely populated with ventenata, with pre-treatment cover ranging from 22% to 53%. Desirable grasses at these sites include western wheatgrass (*Pascopyrum smithii* (Rydb.) Barkworth & D.R. Dewey), interspersed with Kentucky bluegrass (*Poa pratensis* L.), green needlegrass (*Nassella viridula* (Trin.) Barkworth), Idaho fescue (*Festuca idahoensis* Elmer), and Timothy.

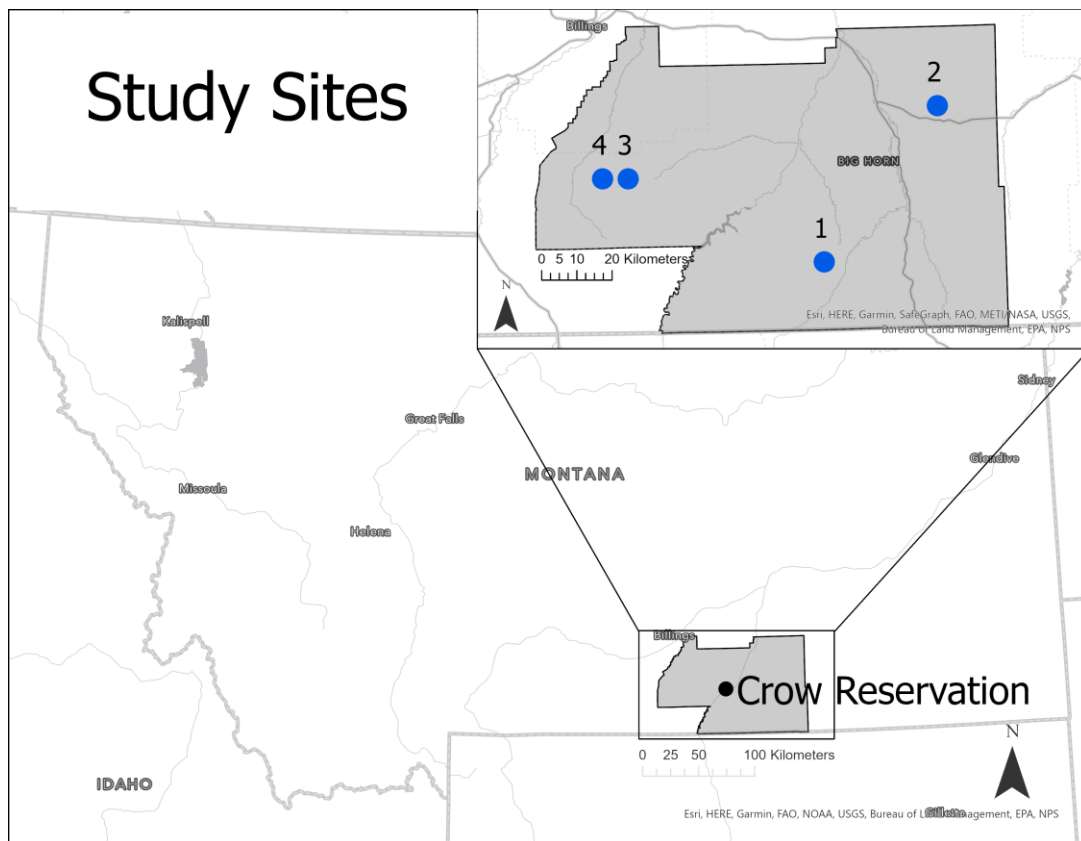


Figure 2.1. Map showing location of the four study sites within the Crow Reservation in southeastern Montana.

Two herbicides, indaziflam at 73.1 g active ingredient (a.i.)/ha (Rejuvra® at 5 oz/A) and imazapic at 105.1 g a.i./ha (Plateau® at 6 oz/A), at two water carrier rates and an organic soil nutrient amendment (NutraFix™) at two applications rates were tested alongside a non-treated

control using a split-plot randomized block design with four replications at each site. The whole plot factor was herbicides and soil nutrient amendment; these plots were 3 m by 18 m. The split plot factor was low versus high water carrier rates for herbicides (46.8 L/ha, 140.3 L/ha, respectively) and low versus high rates of soil nutrient amendments (187 kg/ha, 308 kg/ha, respectively) (Table 2.1). The low and high water carrier rates mimicked aerial and ground applications, respectively. Herbicide treatments were applied using a 3 m wide, hand-held boom sprayer pressurized to 3 kg/cm² using carbon dioxide (CO₂). Soil amendment was hand-broadcasted evenly over each plot. All treatments were assigned randomly within each block. Randomization of treatments was conducted using a random number generator, and randomization of split plots was conducted using a coin flip. Herbicide and soil amendment treatments were applied according to label directions to achieve best timing for annual grass control. Across the four sites, the indaziflam treatment was applied 17-18 August 2021, when *ventenata* had not yet emerged; the imazapic treatment was applied 3-4 November 2021, when *ventenata* was at the one to two-leaf stage; and the organic soil nutrient amendment was applied 17-18 August 2021.

Table 2.1. Treatments applied at the four study sites.

Treatment	Common Product Name	Application Rate (high and low)
Control	Not applicable	Not applicable
Indaziflam	Rejuvra®	73.1 g a.i/ha (water carrier rates: 140.3L, 46.8L)
Imazapic	Plateau®	105.1 g a.i/ha (water carrier rates: 140.3L, 46.8L)
Soil Amendment	NutraFix™	308 kg/ha (high rate); 187 kg/ha (low rate)

Vegetation sampling at each of the four sites was completed during summer 2021 (pre-treatment) and summer 2022 (post-treatment). Pre-treatment sampling occurred on 10-11 June at sites three and four, while sites one and two were completed on 24-25 June 2021. Post-treatment sampling across all four sites occurred 27-30 June 2022. Sampling consisted of randomly placing 3, 20 cm x 50 cm Daubenmire frames (Daubenmire, 1959) in each split-plot and estimating canopy cover by species to the nearest percent along with bare ground and litter. Random placement of frames was achieved by tossing the frame backward without looking.

Data were compiled into an Excel spreadsheet and uploaded to RStudio Desktop version 2022.7.2.576 (R Core Team, 2022), where calculations, analyses, and plots were completed. To determine treatment effects on cover of *ventenata*, western wheatgrass (the most dominant species after *ventenata*), bare ground, and litter, a mixed model analysis of variance (ANOVA) was used and fit using the `lmer` function from the `lme4` package (Bates et al., 2015). The first model fit was $\mu \{ \text{Ventenata cover} \mid \text{Treatment} * \text{High_Low} \} = \text{Treatment}$. A further look into the four sites resulted in a model $\mu \{ \text{Ventenata cover} \mid \text{Treatment} * \text{Site} \} = \text{Treatment}$. Total cover of *ventenata* by treatment was visualized in R studio with a pirate plot from the `yarr` package (Phillips, 2017). The summary statistics were obtained from the `mosaic` package (Pruim et al., 2017). An effects plot was created using the `effects` package (Fox et al., 2020). The ANOVA function from the `car` package was used to provide the type III F-test for the model. Western wheatgrass, litter, and bare ground were also analyzed separately in a similar fashion as described above. A Tukey HSD post hoc test was used on statistically significant treatment responses to determine all pairwise differences for cover of *ventenata*, western wheatgrass, bare ground, and litter.

Results

Ventenata

Differences in ventenata cover across all four sites were evident across treatments ($p < 0.0001$) (Table 2.2). The split-plot effect of rate (water carrier rate for herbicides, application rate for soil nutrient amendment) was not significant ($F = 0.84$, $p = 0.3727$) as well as the interaction with treatment ($F = 0.96$, $p = 0.4119$) (Table 2.2), suggesting that both high and low application rates provided the same degree of ventenata control. Herbicide treatments reduced ventenata cover the most followed by the soil amendment. Mean ventenata cover in the herbicide treatments across all sites was lowest at $1\% \pm 5\%$ and $4\% \pm 9\%$ for the imazapic and indaziflam treatments, respectively (Table 2.3). The soil amendment reduced ventenata cover to $27 \pm 17\%$, which was lower than the non-treated control but higher than either of the herbicides (Table 2.3).

Table 2.2. Analysis of variance (ANOVA) table for treatment * High_Low interaction Bold numbers are significant at $\alpha = 0.05$.

Main Effects	df	F-Value	P-Value
Treatment	3	203.73	<0.001
High_Low	1	0.84	0.3727
Treatment:High_Low	3	0.96	0.4119

Table 2.3. Mean ventenata canopy cover ($\% \pm SD$) across all four sites as affected by treatments. There was no difference in high vs. low application rates, so data were pooled across rates. Similar letters following means indicate no difference between those means ($\alpha = 0.05$).

Treatment	Ventenata cover (%)
Control	38 ± 17 c
Imazapic	1 ± 5 a
Indaziflam	4 ± 9 a
Soil Amendment	27 ± 17 b

The ANOVA type III F-test resulting from the treatment and site interaction model show differences in ventenata cover were evident across the treatments ($p < 0.0001$), treatment and site

interaction ($p < 0.0001$) and site ($p = 0.05$) (Table 2.4). The split-plot effect of rate was removed in all models as it was not significant (Table 2.2).

Table 2.4. Analysis of variance (ANOVA) table for treatment * site interaction across all sites and replications. Bold numbers are significant at $\alpha = 0.05$.

Main Effects	df	F-Value	P-Value
Treatment	3	203.73	<0.0001
Site	3	2.77	0.05089
Treatment:Site	9	13.87	<0.0001

The effect of sites was partly due to differences in ventenata cover between sites. For example, sites one and two had lower cover of ventenata in the control treatment at 22% and 29%, respectively, when compared to sites three and four, which had the highest cover of ventenata at 52% and 48%, respectively (Table 2.5). At site one, ventenata cover was reduced to nearly zero in both the indaziflam and imazapic treatments (Table 2.5). The soil amendment did not reduce ventenata cover and was similar to the control (Table 2.5). At site two, ventenata was reduced to nearly zero in the indaziflam treatment and to 4% in the imazapic treatment, resulting in similar reductions (Table 2.5). Like at site one, the soil amendment did not result in a reduction in ventenata cover and was similar to the control (Table 2.5). At site three, both indaziflam and imazapic similarly reduced ventenata cover to 6% and zero, respectively (Table 2.5). The soil amendment reduced ventenata cover to about half (23%) of the control (52%) at site three, however, not as low as the herbicides (Table 2.5). At site four, each treatment reduced ventenata cover differently compared to the control. The imazapic treatment had the highest reduction (0% cover), followed by indaziflam (10% cover), soil amendment (28% cover) and control (48% cover) (Table 2.5).

Table 2.5. Mean ventenata cover (% \pm SD) at each site. Similar letters following means indicate no difference between those means ($\alpha=0.05$).

Treatment	Site 1	Site 2	Site 3	Site 4
Control	22 \pm 13 b	29 \pm 12 b	52 \pm 11 c	48 \pm 13 d
Indaziflam	1 \pm 3 a	0.5 \pm 1 a	6 \pm 12 a	10 \pm 11 b
Imazapic	0 \pm 0.5 a	4 \pm 10 a	0 \pm 0 a	0 \pm 0 a
Soil Amendment	21 \pm 13 b	36 \pm 11 b	23 \pm 18 b	28 \pm 19 c

Western Wheatgrass

The split-plot effect of rate (water carrier rate for herbicides, application rate for soil nutrient amendment) was not significant ($F=0.99$, $p=0.3349$) nor was the interaction with treatment ($F=0.32$, $p=0.8083$) (Table 2.6). This suggests that the high and low rate did not affect western wheatgrass cover. Results from the treatment and site interaction model showed differences in western wheatgrass cover across treatments ($p<0.0001$) and the treatment and site interaction ($p=0.0016$) (Table 2.7).

Table 2.6. Analysis of variance (ANOVA) table for treatment * high_low interaction across all sites and replications. Bold numbers are significant at $\alpha=0.05$.

Main Effects	df	F-Value	P-Value
Treatment	3	38.04	<0.0001
High_Low	1	0.99	0.3349
Treatment:High_Low	3	0.32	0.8083

Table 2.7. Analysis of variance (ANOVA) table for western wheatgrass treatment * site interaction across all sites and replications. Bold numbers are significant at $\alpha=0.05$.

Main Effects	df	F-Value	P-Value
Treatment	3	40.3	<0.0001
Site	3	0.86	0.461
Treatment:Site	9	3	0.0016

At site one, western wheatgrass increased the most in the indaziflam treatment (22%) and imazapic treatment (21%) when compared to the control (13%) (Table 2.8). Western wheatgrass

cover in the soil amendment did not differ from the control (Table 2.8). At site two, only the indaziflam treatment increased western wheatgrass cover (17%) compared to the control (10%) (Table 2.8). At site three, western wheatgrass cover was highest in the imazapic (35%) and indaziflam (31%) treatments, followed by the soil amendment (24%), all of which were higher than the control (12%) (Table 2.8). At site four, western wheatgrass cover increased only in the imazapic (26%) and indaziflam (25%) treatments when compared to the control (15%) (Table 2.8). Overall, both imazapic and indaziflam increased western wheatgrass cover at all sites, with the exception of site two where imazapic was not different than the control. The soil amendment only increased western wheatgrass cover compared to the control at site three (Table 2.8).

Table 2.8. Mean western wheatgrass cover (% \pm SD) at each site. Similar letters following means indicate no difference between those means ($\alpha=0.05$).

Treatment	Site 1	Site 2	Site 3	Site 4
Control	13 \pm 6 a	10 \pm 5 a	12 \pm 5 a	15 \pm 7 a
Indaziflam	22 \pm 12 c	17 \pm 11 b	31 \pm 13 bc	25 \pm 11 bc
Imazapic	21 \pm 12 bc	16 \pm 12 ab	35 \pm 12 c	26 \pm 13 c
Soil Amendment	15 \pm 6 ab	13 \pm 7 ab	24 \pm 10 b	19 \pm 9 ab

Litter

Similar to ventenata and western wheatgrass cover, litter was not affected by the high and low rates for the split-plot, so it was left out of the model. Results from the treatment and site interaction model show differences in litter cover that were evident across treatments ($p<0.0001$) and the interaction of treatments and site ($p=0.0022$) (Table 2.9).

Table 2.9. Analysis of variance (ANOVA) table for litter treatment * site interaction across all sites and replications. Bold numbers are significant at $\alpha=0.05$.

Main Effects	df	F-Value	P-Value
Treatment	3	58.69	<0.0001
Site	3	1.04	0.4804
Treatment:Site	9	2.94	0.0022

At site one, litter cover increased in the indaziflam (39%) and imazapic (35%) treatments when compared to the control (21%) (Table 2.10). Litter cover in the soil amendment treatment (28%) at this site was not different from the control. At site two, litter increased in both the indaziflam (41%) and imazapic (45%) treatments when compared to the control (25%) (Table 2.10). Litter in the soil amendment treatment (24%) at this site was not different from the control. At site three, litter cover increased compared to the control in all treatments to 50%, 49%, and 46% in the indaziflam, imazapic, and soil amendment treatments, respectively (Table 2.10). At site four, both the imazapic (57%) and indaziflam (55%) treatments increased litter cover the most, followed by the soil amendment (46%), when compared to the control (28%) (Table 2.10).

Table 2.10. Mean litter cover (% \pm SD) at each site. Similar letters following means indicate no difference between those means ($\alpha=0.05$).

Treatment	Site 1	Site 2	Site 3	Site 4
Control	21 \pm 9 a	25 \pm 14 a	32 \pm 12 a	28 \pm 12 a
Indaziflam	39 \pm 15 c	41 \pm 10 b	50 \pm 17 b	55 \pm 12 c
Imazapic	35 \pm 11 bc	45 \pm 10 b	49 \pm 15 b	57 \pm 11 c
Soil Amendment	28 \pm 12 ab	24 \pm 12 a	46 \pm 15 b	46 \pm 16 b

Bare Ground

Bare ground was not affected by high and low rates for the split-plot, so rate was removed from the model. It also did not have a site and treatment interaction, and site was not significant ($p=0.7091$) (Table 2.11). The model included treatment and site and resulted in differences in bare ground cover that were evident across the treatments ($p<0.0001$) (Table 2.11).

Table 2.11. Analysis of variance (ANOVA) table for bare ground treatment + site across all sites, replications. Bold numbers are significant at $\alpha=0.05$.

Main Effects	df	F-Value	P-Value
Treatment	3	7.32	<0.0001
Site	3	0.46	0.7091

At all sites bare ground cover increased in the herbicide treatments when compared to the control treatment (Table 2.12). Bare ground cover in the soil amendment treatment at all sites did not differ from the control treatment (Table 2.12). Site one had an increase in bare ground cover in the indaziflam (6%) and imazapic (8%) treatments when compared to the control (3%) (Table 2.12). At site two, both the indaziflam and imazapic treatments increased bare ground (2%) when compared to the control (1%) (Table 2.12). At site three, bare ground increased in both indaziflam (10%) and imazapic (7%) treatments, when compared to the control (2%) (Table 2.11). At site four, bare ground increased in both indaziflam (4%) and imazapic (5%) treatments, when compared to the control (2%) (Table 2.12).

Table 2.12. Mean bare ground cover (% \pm SD) at each site. Similar letters following means indicate no difference between those means ($\alpha=0.05$).

Treatment	Site 1	Site 2	Site 3	Site 4
Control	3 \pm 4 a	1 \pm 1 a	2 \pm 4 a	2 \pm 2 a
Indaziflam	6 \pm 6 b	2 \pm 1 b	10 \pm 21 b	4 \pm 5 b
Imazapic	8 \pm 10 b	2 \pm 1 b	7 \pm 14 b	5 \pm 7 b
Soil Amendment	4 \pm 6 a	1 \pm 1 a	4 \pm 5 a	2 \pm 2 a

Discussion

Both imazapic and indaziflam significantly reduced ventenata cover at all four sites, regardless of water carrier rate. The imazapic treatment was most consistent, reducing ventenata cover to less than 4% across all sites. However, the indaziflam treatment also provided good control, reducing ventenata to <1% to <10% cover across the four sites, and only differed from

imazapic at site four. I hypothesized that indaziflam would provide the highest reduction of ventenata, however imazapic performed similarly to indaziflam and even better at one site. This may change over time as imazapic is less persistent and typically provides a shorter window of control, for example one to two years of annual grass control (Harvey, 2019; Hart & Mealor, 2021; Mangold et al., 2013; Sebastian et al., 2016, 2017; Wallace & Prather, 2016). At two of the four sites (Site 3 and 4), the soil amendment reduced ventenata cover by about half when compared to the control, and its effectiveness was not dependent upon application rate. These two sites had the highest cover both prior to (2021) and during (2022) the study at about 50%. One year after application, the soil amendment did not reduce ventenata abundance enough to justify its use for controlling ventenata. More time may be needed to see positive results, however, as case studies in Montana have shown decreases in annual grasses and increases in perennial vegetation two to three years after application (NutraFix Soils, 2020).

Western wheatgrass cover increased in response to herbicide treatments, nearly doubling in cover when compared to the control at all four study sites, except for the imazapic treatment at site two. This suggests western wheatgrass is recovering after eliminating ventenata. This is promising for the rangeland resource, as we see the desired grass recovering well after herbicide treatments. This is also encouraging for those managing invasive annual grasses who may be concerned about vegetation dynamics after removal of ventenata. Annual grasses such as ventenata grow quickly and senesce early in the growing season, whereas perennial grasses continue growing later into the growing season. Controlling ventenata can provide better quality of forage which leads to a longer, green, grazable forage window (Hart & Mealor, 2021). My study showed that the desired perennial grasses can increase in response to removal of ventenata.

On rangeland about 100 km south of my study area, Hart & Meador (2021) concluded that one-year after removal of *ventenata* with the herbicides imazapic and indaziflam, there was a positive response from perennial grasses, and that this perennial forage base was of higher quality. They also found that species richness and biodiversity did not change as a result of herbicide applications. Controlling *ventenata* with herbicides can improve forage resources for livestock and wildlife, and monitoring and management efforts to prevent spread should be prioritized (Hart & Meador, 2021).

In a study in Colorado, removal of cheatgrass with indaziflam, imazapic, and mixtures with glyphosate and rimsulfuron resulted in no significant differences in species richness, although there was some injury to perennial wheatgrass species from these herbicides (Sebastian et al., 2016). In another study in Colorado, indaziflam controlled cheatgrass for multiple years without a reduction in perennial species richness or abundance (Clark et al., 2019). Perennial grass cover also did not decrease in the indaziflam treatments, suggesting native species tolerance to indaziflam (Clark et al., 2019). In a study near Pullman, WA, and Moscow, ID, indaziflam mixed with either glyphosate or rimsulfuron reduced *ventenata* cover 6 and 16 months after treatment and resulted in an increase in perennial grass cover at 16 months after treatment (Koby et al., 2019). These studies along with my study suggest indaziflam may be a tool to control *ventenata* while minimally impacting desired vegetation (Clark et al., 2019; Koby et al., 2019; Sebastian et al., 2016, 2017).

In my study, bare ground increased at all sites in the herbicide treatments, however cover averaged about 5% for both herbicides when compared to the control which averaged 2%. Overall, litter cover increased in both the imazapic and indaziflam treatments at all sites, with

neither treatment differing from one another (Table 2.10). There was an increase in litter cover in the soil amendment treatment at sites three and four only (Table 2.10). Litter was nearly doubled when compared to the control at all sites (Table 2.10), indicating *ventenata* was reduced by indaziflam and imazapic causing litter to accumulate. This can be negative for future herbicide treatments where the herbicide may have trouble reaching the soil or *ventenata* seedlings. Winter annual grasses can accumulate large quantities of litter, or thatch, on the soil surface as plants senesce yearly and decompose slowly (Evans & Young, 1970), and litter can facilitate invasions by promoting winter annual grass germination and suppressing native plants (Evans and Young, 1970). Kessler et al. (2015) reported improved performance with herbicides when litter had been eliminated by fire. A study by Clark et al. (2019) investigated the interception and desorption of herbicides applied to litter from three invasive winter annual grass species with simulated rainfall. Imazapic, rimsulfuron, and indaziflam were applied to the three annual grasses (*ventenata*, cheatgrass, medusahead). Cheatgrass herbicide interception was 84%, while *ventenata* and medusahead averaged 76% herbicide interception. Simulated rainfall at zero days after application recovered 100% of the intercepted rimsulfuron and imazapic from cheatgrass litter, while recovery decreased to 65% with rainfall at one or seven days after application. Only 54% of indaziflam could be recovered at zero days, and recovery decreased to 33% when rainfall was applied at one or seven days after application. Applying soil-active herbicides before forecasted rain or tank mixing with a post-emergent herbicide to provide initial control could potentially increase the amount of herbicide reaching the soil and provide more consistent invasive annual grass control (Clark et al., 2019). In a study in Colorado, nearly 75% of imazapic and tebuthiuron (Alligare®) were intercepted when applied over high amounts of cheatgrass

litter, and only 69% of the intercepted herbicide could be desorbed from the litter with 15 mm of rainfall seven days after treatment (Kessler et al. 2015). Depending on the amount of litter, interception of the herbicide may occur which can decrease annual grass control. Adequate rainfall may be needed for the herbicides to reach the soil surface and any emerging seedlings of the annual grass. My sites saw an increase in litter, which may mean that future applications of imazapic may not be as effective; this may be less of a concern with indaziflam treatments since it can provide control for a couple of years.

Current methods to control *ventenata* focus on herbicide applications, with varying degrees of longevity of control based upon the active ingredient used (Harvey, 2019; Sebastian et al., 2016; Wallace & Prather 2016). The imazapic treatment is likely to become less effective over time based on findings by Harvey (2019). In this study near Bozeman, MT, imazapic provided 63% control 8 months after treatment, but then control decreased to 20% at 11 and 20 months after treatment. In that same study, indaziflam provided 70% control 8 months after treatment and 67% after 11 months, but then increased to 98% after 20 months. In a study conducted in northern Colorado, fall application of indaziflam provided 89 to 100% control of cheatgrass two years after treatment and 83 to 100% control three years after treatment; this was compared to the more commonly used herbicides glyphosate, imazapic, and rimsulfuron, whose effectiveness dropped from about 80% in the first year after treatment to 30% by the third year (Sebastian, et al., 2016). This suggests indaziflam may provide multiple years of control while imazapic may lessen in its effectiveness after the first year.

The goal of this research was to provide control options for *ventenata* in southeastern Montana on the Crow Reservation. I tested imazapic and indaziflam at a high and low water

carrier rate to mimic ground and aerial application and to compare the two simulated applications to each other. Water carrier rate did not make a difference in reducing ventenata cover for both imazapic and indaziflam, and this was consistent across all four sites. Just south of our study sites, in northern Wyoming, aerial application of imazapic and indaziflam by helicopter reduced cover of ventenata to nearly 0% one year after application (Hart & Meador, 2021). Herbicides were applied at a water carrier rate of 46.8 L/ha, which is similar to the low rate used in our study. Collectively, these results are encouraging as the Crow Reservation extends south to the Wyoming line, near areas treated in northern Wyoming (Hart & Meador, 2021). Applying by helicopter may be more expensive than ground applications on a per hectare basis, however, aerial application is quicker and may be necessary to cover large areas of infestation. On the Crow Reservation, ventenata covers large areas, many of which are inaccessible by vehicle, suggesting aerial application may be the most efficient and effective way to apply these herbicides. My study in southeastern Montana showed that indaziflam and imazapic controlled ventenata effectively at both ground and aerial application rates. This suggests that aerial application may provide just as good of control as ground application. This is encouraging for the prospect of managing ventenata on challenging landscapes like those on the Crow Reservation.

Long-term ventenata management on rangelands depends on control options that reduce cover of mature plants and thus reduce seed production in the current year of control and reduce the seed bank over multiple years. In this study, I showed that imazapic and indaziflam provided a high reduction in ventenata cover (ranging from 0% to < 10% cover) concurrent with an increase in the desirable native grass, western wheatgrass, at four sites one growing season after

treatment. Further sampling is needed to compare effectiveness of the two herbicides multiple years after treatment. Results from southwestern Montana have shown that an application of indaziflam plus imazapic provided three years of annual grass control (Harvey, 2019), so I am optimistic that ventenata cover will remain low in the treated plots for at least another growing season. Although litter and bare ground did increase in herbicide treatments, western wheatgrass also increased two-fold when compared to the control treatment, suggesting that removal of ventenata can result in recovery of desired perennial grasses. People managing ventenata in southeastern Montana and on the Crow Reservation can use the information from this study to take proactive measures to minimize populations of ventenata, reduce impacts associated with this invasive annual grass, and prevent further spread.

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CHAPTER THREE

USING UNMANNED AERIAL VEHICLE (UAV) TIME SERIES TO MONITOR RESPONSE
OF VENTENATA (*VENTENATA DUBIA*) TO HERBICIDE TREATMENTS IN
SOUTHEASTERN MONTANAIntroduction

Nonnative annual grass invasions are one of the most transformative stressors in rangelands of the western United States (Germino & Lazarus, 2020; D'Antonio & Vitousek 1992). Cheatgrass (*Bromus tectorum* L.) is the most widespread nonnative annual grass, but other annual grasses such as other species in the *Bromus* genus, medusahead (*Taeniatherum caput-medusae* (L.) Nevski), and ventenata (*Ventenata dubia* (Leers) Coss.) are serious threats to different regions and are continuing to spread (Germino & Lazarus, 2020). In the past decade, the Wyoming Basin, Great Plains, and Rocky Mountains have begun to experience annual grass invasion (Germino & Lazarus, 2020). Ventenata has been documented to increase rapidly (Nicolli et al., 2020), and in Montana ventenata is documented in 24 counties (Harvey & Mangold, 2018). Herbicides are the most common tool for weed control in rangelands (DiTomaso, 2000; Mangold et al., 2018; Vallentine, 1981). Most herbicides are applied either pre-emergence, before the weeds emerge from the soil, or post-emergence, after leaves of the weeds have emerged from the soil. Both pre- and post-emergent herbicides have been used on annual grasses, including ventenata (Harvey, 2019; Hart & Meador, 2021; Koby et al., 2019; Mangold et al., 2013; Sebastian et al., 2016, 2017; Wallace & Prather, 2016).

A challenge for invasive plant management is identifying and preventing invasions before they expand into new landscapes. To prevent invasions, managers rely on early detection and rapid response (EDRR) or identifying and eradicating new infestations before they can gain a permanent foothold in a new landscape (Simberloff, 2013). Remote sensing technology can help with these challenges by detecting and monitoring invasions (Bradley, 2014; Vaz et al., 2018), creating distribution maps to inform EDRR of new invasions (Westbrooks, 2004), and supporting weed management decisions (Shaw, 2005).

Remote sensing is the process of detecting and monitoring the physical characteristics of an area on the earth's surface by measuring reflected and emitted radiation (USGS, 2017). Remote sensing can be used to map broad scale vegetation distributions across rangelands, which is often not achievable with traditional field-based methods (Bateman et al., 2020; Rocchini et al., 2015). Remotely sensed data collection has also been used in detecting biological invasions and supporting early detection of invasion (Bradley, 2014; Vaz et al., 2018). As an example, broad-scale distribution maps of cheatgrass over western rangelands have been created from satellite platforms (Bradley & Mustard, 2006; Clinton et al., 2010; Boyte et al., 2015). Often used in accordance with field-based data, remotely sensed data have been used in mapping and monitoring the distribution and abundance of invasive plant species and annual grasses (Bateman et al., 2020; Bradley, 2014; Bradley & Marvin, 2011).

The phenology of a plant during the growing season includes life-cycle events such as emergence, flowering, and senescence. Each of these life-cycle events may result in a change in spectral signatures, and these differences may be leveraged to examine phenology from a distance. "Green up" of annual grasses is earlier than "green up" of perennial vegetation,

producing a distinct pulse of greenness in the early spring. For example, cheatgrass becomes productive in spring, earlier than native shrubs and grasses. As it becomes more abundant, the timing of peak greenness shifts, and this change is observable with satellite image time series (Bradley et al., 2018; Peterson, 2005). Additionally, cheatgrass responds strongly to precipitation (Stewart and Hull, 1949). This can be highly variable year to year. The inter-annual variability in greenness can be detected with satellite time series and may be a better approach than a single date image (Boyte et al., 2019; Bradley & Mustard, 2005, 2008; Bradley et al., 2018). For example, differences in the Normalized Difference Vegetation Index (NDVI) between wet and dry years have been used previously to identify areas where cheatgrass is present (Bradley & Mustard 2005, 2008). Peterson (2005) distinguished cheatgrass from other vegetation using satellite data on two different dates (early spring and early summer) within a single year.

With remote sensing data, there are tradeoffs between spatial extent (size of the image), spatial resolution (pixel size), spectral resolution (number and range of visible and infra-red bands) and temporal resolution (frequency of data obtained) (Bradley, 2014). Broader spatial extents allow for more extensive mapping of invasive plants and ultimately provide more distribution data to inform spatial models of invasion risk. However, spatial resolution is typically low, making only abundant and widespread infestations potentially detectable (Bradley, 2014). Finer spatial resolution makes it more likely that individual species and early infestations can be detected, however, spatial extents and repeat temporal coverage are typically limited (Bradley, 2014). The advantages of satellite based remote sensing include high temporal resolution, which makes possible the extraction of time series of consistent and comparable data, which can be cost effective (Xue & Su, 2017). Satellite platforms such as Landsat also enable

free access to multispectral data, however, disadvantages with such passive satellite-based remote sensing platforms are the revisitation time, which for Landsat is 16 days, which can make agricultural applications difficult to monitor (USGS, 2017; Xue & Su, 2017). Furthermore, passive optical satellite imagery cannot penetrate clouds, and data may be unusable on overcast days (Xue & Su, 2017). Unmanned Aerial Vehicles (UAVs) can help mitigate some of the issues associated with satellite imagery.

While satellite imagery may have been the most common use of remote sensing to date, UAVs are an expanding area of research driven by both academic and commercial successes (Yao et al., 2019). In the western Great Basin, Weisberg et al. (2021) developed a multitemporal classification approach that used unmanned aerial vehicle (UAV) imagery to map two invasive annual grasses (cheatgrass and medusahead) to distinguish these from key functional types of native vegetation, based upon differences in plant phenology (Weisberg et al., 2021). Classification accuracy can also be improved with multi-date imagery. For example, in southwestern Montana, accuracy increased 5-10% between a single and multiple UAV flights over the growing season (Wood et al., 2022). Within herbaceous and sagebrush classes, multi-temporal analyses identified differences in the seasonal timing of green up and senescence within herbaceous and sagebrush classes (Wood et al., 2022).

Thus far, most remote sensing research on invasive annual grasses has been focused on cheatgrass or medusahead, but there has been minimal research on ventenata in the above aspects. Current knowledge gaps include developing more accurate and efficient methods for identifying and mapping invasive annual grasses, particularly ventenata, as well as developing better models for predicting the impacts to inform management strategies. The types of remote

sensing analyses done for other annual grasses can help with risk assessments, distribution maps, and can improve management decisions for ventenata (Bradley, 2014; Shaw, 2005; Vaz et al., 2018). While there are maps of other invasive annual grass distributions, there has not been much created for ventenata. Furthering remote sensing analyses focused on ventenata will help mitigate knowledge gaps.

Many remote sensing platforms use multispectral sensors. With multispectral imagery, individual bands can be transformed with spectral indices to highlight certain features and patterns. For information extraction, vegetation indices are commonly used to generate new images from two or more image bands (Huang et al., 2021), enhancing features such as vegetation for further analysis. There are more than one hundred vegetation indices (Vis) that can be derived from multispectral imagery, with NDVI being the most commonly used one (Huang et al., 2021; Xue and Su, 2017). The number of NDVI papers from the Web of Science Core Collection increased from 795 in the 1990s, to 3361 in the 2000s, and to 12,618 in the 2010s (Huang et al., 2021). The rise of NDVI applications is aided by advancements and widespread use of UAVs (Yao et al. 2019).

NDVI provides a measure for vegetation greenness and plant health; it has been shown to be positively correlated with plant productivity and has been used to estimate crop yields, pasture performance, and rangeland carrying capacities (Huang et al., 2021; Ndayisaba, 2016; Turvey & Mclaurin, 2012; USGS, 2017). Calculated as a ratio between near-infrared (NIR) and red (RED) reflectance values, $NDVI = (NIR - RED) / (NIR + RED)$ (Jackson & Huete, 1991; USGS). Generally, healthy vegetation will absorb most of the visible light and reflect a large portion of the NIR light, whereas unhealthy, including senescing, vegetation will reflect more visible light

and less near-infrared light (Ndayisaba et al, 2016). Bare soils reflect moderately in both the red and near-infrared portions of the electromagnetic spectrum (Jackson & Huete, 1991). NDVI values range from -1 to 1, and the NDVI of a densely vegetated area will tend toward positive values, whereas water, rocks, sand, and concrete surfaces will be represented by near zero or negative values (Jackson & Huete, 1991; Huang et al, 2021). Typically, in temperate environments, NDVI values from healthy vegetation will increase as plant cover increases at the beginning of the growing season, reach a peak during the middle of the growing season, and will then decrease at the end of the season as vegetation senesces (Mkhabela et al. 2005; Turvey & Mclaurin, 2012).

The use of NDVI in monitoring vegetation is common because of its ability to delineate vegetative stress, which has great appeal in commercial agriculture and land-use studies (Huang et al., 2021). Observing NDVI differences with a time series after ventenata has been treated with an herbicide may offer a way in which land managers can monitor effectiveness of treatments as well as monitor spread, which is important for ventenata management. For example, a study in southern Wyoming created an accurate map of cheatgrass distribution using Landsat imagery in a post-wildfire area; this map was used for monitoring and targeted cheatgrass management such as aerial herbicide spraying (West et al., 2017). Furthering remote sensing analyses focused on ventenata will further our basic understanding of this species.

In chapter two, I tested the effectiveness of indaziflam (Rejuvra®, BAYER, Environmental Science), imazapic (Plateau®, BASF Corporation) and a soil amendment (NutraFix™, Edaphix™) for reducing ventenata on grasslands in southeastern Montana on the Crow Reservation. Both herbicides significantly reduced ventenata cover to <5% and increased

cover of western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Love) and litter (Chapter two). Therefore, the objectives of this chapter were to examine differences in NDVI time series in response to the treatments from the field study at one of the sites. I hypothesized that there would be a difference in NDVI between the herbicide sprayed plots and the non-sprayed control and that the relationship would change over the course of the growing season due to the different abundances and green up times of annual grasses vs perennial grasses. I expected the sprayed plots to have lower cover of annual grasses, mainly ventenata, and higher cover of the dominant perennial grass western wheatgrass, therefore, I expected NDVI to be higher and stay greener longer when compared to the non-sprayed control.

Methods

Study Area

The study area is located in southeastern Montana on the Crow Reservation, about 25 km east of Crow Agency, MT (45°31'44.1"N, 107°11'16.8"W). The elevation here is 935 m and mean annual precipitation is 35 to 43 cm (Figure 3.1).

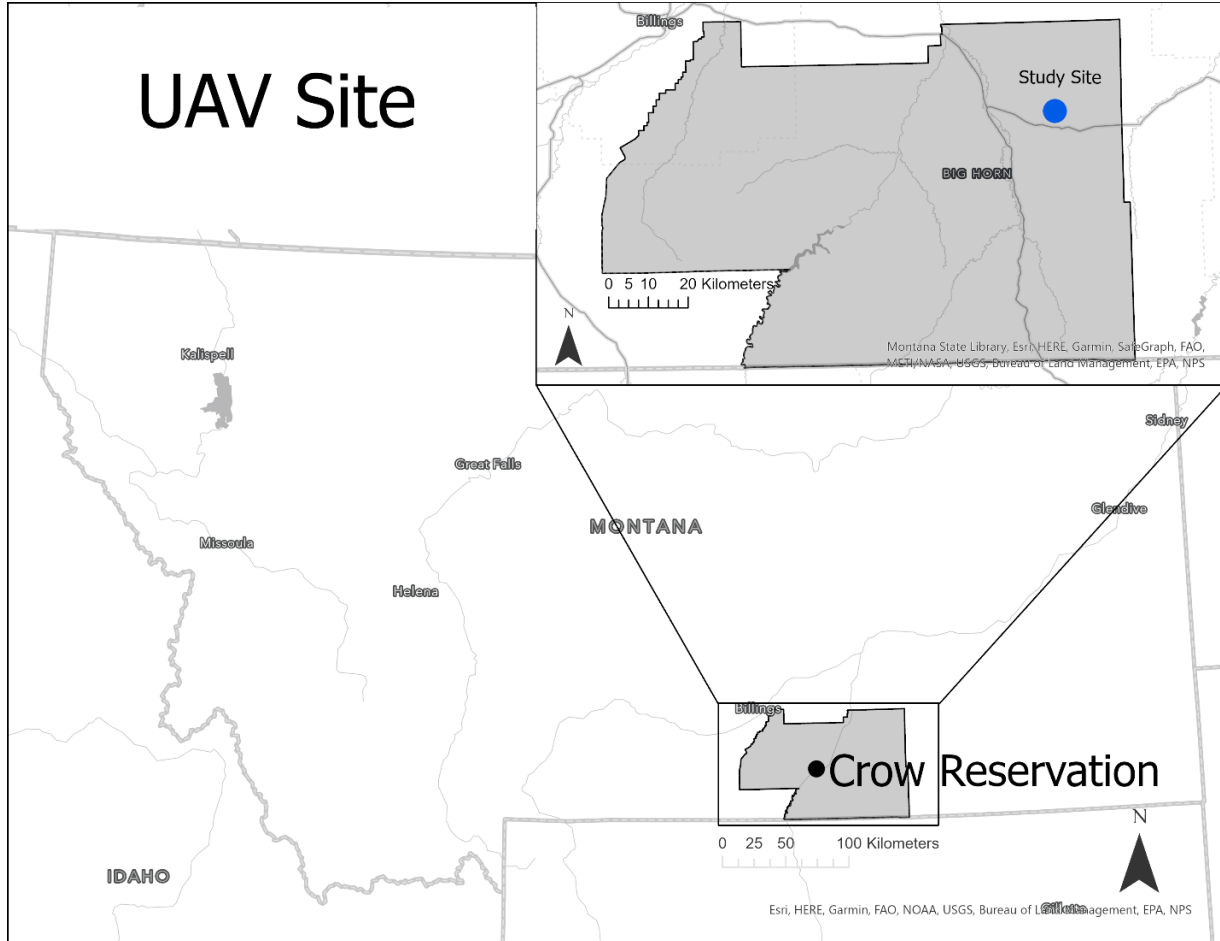


Figure 3.1. Location of the study site on the Crow Reservation in southeastern Montana.

Multispectral imagery was collected using a fixed-winged eBee drone (SenseFly) with an attached multispectral sensor (MultiSPEC 4C, Airinov) (green, red, red-edge, near-infrared) to collect the imagery between spring green up and summer dry out from 24 May – 8 August 2022, for a total of 10 flight dates. NDVI imagery were used to assess response of vegetation to the treatments, which included a non-sprayed control; two different herbicides, indaziflam (Rejuvra®) and imazapic (Plateau®); and a soil amendment (NutraFix™). The treated plots were 3 m x 18 m and replicated four times across the site with 1.5 m buffers between each replication; therefore, total site area was approximately 17 m x 18 m. Before executing each flight,

radiometric calibration images were acquired to obtain reflectance measurements. Calibration of digital numbers to surface reflectance was achieved by holding the drone and sensor over a calibrated reflectance target prior to each flight. The multispectral data were collected at an altitude of approximately 30 m. Flights were typically completed within a ten-minute window centered around solar noon. Flights were conducted using pre-programmed flight plans with eMotion 3 software version (3.5) (SenseFly). Photographs were captured using a 70% lateral and longitudinal overlap, and data were captured at 5 cm spatial resolution. Five ground control points (GCPs) were set out during the duration of the study and used to align the data from the different flight dates. The GCP locations were obtained using an Emlid Reach RS2 GPS receiver (Emlid) and used to georeference the images into WGS 1984 UTM Zone 13N. Images were processed using Pix4Dmapper version (4.8.2) (Pix4D, Lausanne, Switzerland).

Processed images and derived indices were imported into ArcGIS Pro version 2.9.0 for further analysis. To extract the NDVI values from the treatment plots, shapefiles were created by delineating polygons within the perimeters of the treatment plots which were approximately 3 m by 18 m. Zonal statistics from the ArcGIS Pro toolbox were used to obtain summary NDVI statistics on the treatment polygons for each of the 10 flight dates. Mean NDVI for each date and treatment were then imported into Excel to analyze the NDVI values across the 10 flight dates, focusing on the non-sprayed and herbicide sprayed plots; the soil amendment plots were not included because they did not reduce ventenata canopy cover relative to the non-sprayed control.

Results

There were differences in NDVI values between the treatments starting with the first flight date on 24 May through the last flight date on 8 August. At the beginning of the flight

dates, the two herbicide treatments had lower NDVI values than the non-sprayed treatments, but this relationship switched on 7 July, at which point the non-sprayed treatments had lower NDVI values than either of the two herbicide treatments (Figure 3.2). On the first flight date (24 May), the lowest NDVI was the imazapic treatment (0.39), followed by the indaziflam treatment (0.49) and non-sprayed treatment (0.56) (Figure 3.2).

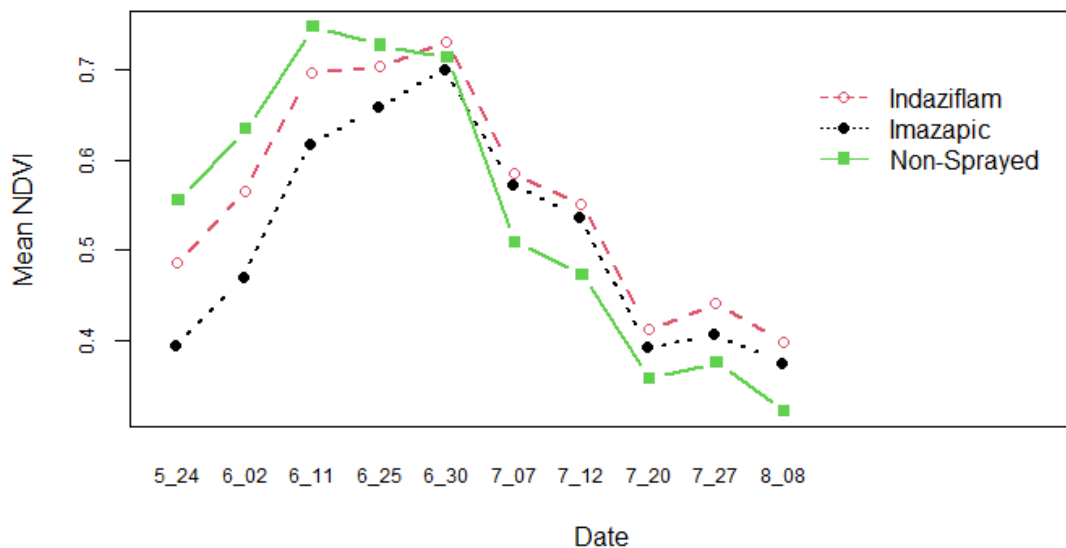
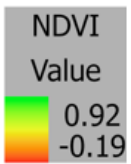
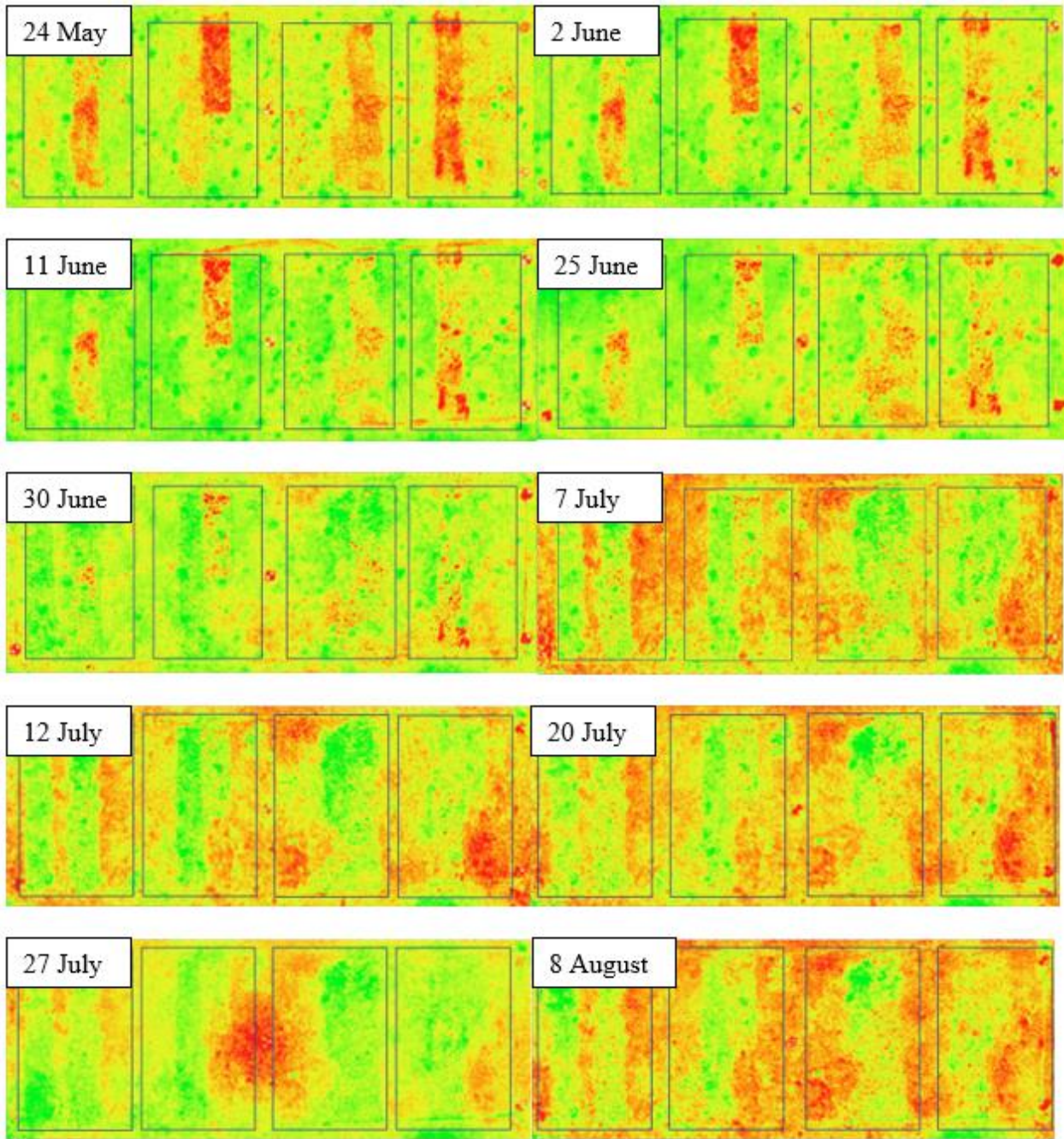


Figure 3.2. Mean NDVI for imazapic, indaziflam and non-sprayed treatments for the 10 weekly flight dates (24 May- 8 August 2022).

The NDVI for all treatments continued to increase further into June before decreasing. The non-sprayed treatment started with the highest NDVI on the first flight date (0.56) and continued to increase until June 11 where it peaked at 0.75 before decreasing slightly to 0.73 on 25 June and to 0.71 30 June. The following flight date on 7 July had the biggest decrease between dates where the NDVI was reduced to 0.51 for the non-sprayed treatment (Figure 3.2). From July 7 until the end of the flight dates, the non-sprayed treatment had a lower NDVI than

either of the herbicide treatments, decreasing to its lowest NDVI value of 0.32 on the last flight date on August 8 (Figure 3.2). The imazapic treatment started with the lowest NDVI on the first flight date on 24 May (0.39) and continued to increase until 30 June where it peaked at 0.70 before decreasing to 0.57 on 7 July and then continued to decrease until the last flight date on 8 August when the NDVI was 0.37 (Figure 3.2). The indaziflam treatment had an NDVI of 0.49 at the first flight date and continued to increase until 30 June where it peaked at 0.73 before decreasing to 0.59 on 7 July and then continued to decrease until the last flight date when NDVI was 0.40, similar to the imazapic treatment (Figure 3.2).



Arrows = Sprayed Plots

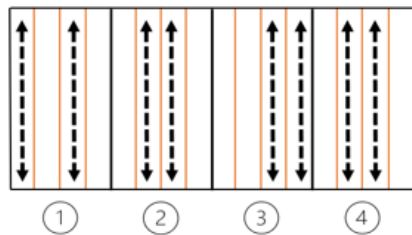


Figure 3.3. Flight dates 24 May- 8 August 2022. Reps 1-4 are shown. Starting with flight one on top from left to right (24 May-8 August). Sprayed plots start with a low NDVI value (red) then gradually increase (green) throughout the season. There is a shift in NDVI between 30 June and 7 July, when sprayed plots begin to stand out in greenness and non-sprayed plots turn red.

The herbicides significantly reduced *ventenata* cover to <5% when compared to the non-sprayed plots which had 30% *ventenata* cover (Chapter 2). There was also an increase in perennial grass cover, primarily due to western wheatgrass, and an increase in litter cover in the sprayed plots (Chapter 2). Perennial grasses had approximately 30% cover in herbicide sprayed plots and about 45% litter cover (Chapter 2). The reduction in *ventenata* and increases in perennial grass cover and litter played a part in the differences in NDVI values between treatments throughout the season. At the beginning of the season, sprayed plots had a lower NDVI than the non-sprayed sites, and this difference is seen in the dates leading up to 30 June (Figure 3.3). On 7 July there was a noticeable reduction in NDVI in the non-sprayed plots, whereas the sprayed plots exhibited higher NDVI values (Figure 3.3).

The two critical dates on which there was the biggest change in NDVI between treatments were 30 June and 7 July (Figure 3.2; Figure 3.3). During this period, the sprayed plots had a higher NDVI than the non-sprayed plots, which up until this point had a higher NDVI (Figure 3.2). From 7 July until the last flight date, the sprayed plots remained higher in NDVI than the non-sprayed plots. (Figure 3.2).

Discussion

Results from this study indicate that multi-date NDVI may serve as a useful indicator of vegetative change after application of an herbicide to control the annual grass *ventenata*. The indaziflam and imazapic treatments started with a lower NDVI and ended with a higher NDVI

than the non-sprayed control, which supports my hypothesis of a difference in NDVI between the herbicide sprayed plots and the non-sprayed control due to the different green up times of annual grasses vs perennial grasses. The herbicides reduced the annual grasses, mainly *ventenata*, and created space for the perennial grasses such as western wheatgrass to recover. This is similar to a study in northern Wyoming where *ventenata* was removed with the same herbicides and perennial grasses showed a positive response (Hart & Meador, 2021). Annual grasses green up and senesce earlier in the season than perennial grasses, but perennial grasses remain green for longer in the summer, senescing in late summer. Plots with more *ventenata* cover, such as the non-sprayed control, can be expected to have a higher NDVI initially but then experience a reduction in NDVI by late June or early July when annual grasses senesce. In contrast, plots with more perennial grass cover can be expected to start the growing season with a lower NDVI due to the delayed onset of growth relative to invasive annual grasses; they might further be expected to remain green longer into the summer. This study found that both sprayed and non-sprayed plots peaked in greenness around the same time in late June, with sprayed plots having a slower and lower decline in NDVI later into the season.

Cover of litter, which was mostly from annual grasses that were growing prior to treatment, also increased in the herbicide sprayed plots (Chapter 2). This litter may have contributed to the lower NDVI early in the season along with the perennial grasses that were small and slow to begin growth. Winter annual grasses can accumulate large quantities of litter, or thatch, on the soil surface as plants senesce yearly and decompose slowly (Clark et al., 2019; Evans & Young 1970). Herbicides removed annual grasses from the research plots, but the

remaining litter from previous years' growth was still present and especially noticeable early in the season.

The ability to detect NDVI differences between herbicide-treated areas and non-treated areas can give land managers a better understanding of where herbicides are effective at reducing *ventenata*. Multi-temporal UAV data can be helpful at detecting and capturing *ventenata* senescence. At my research site, there were differences in NDVI, with the sprayed plots having a higher value as the growing season progressed and staying green longer. The change between treated and non-treated areas was particularly apparent between the last week of June and the first week of July. This seemed to be the critical time at which annual grasses dried out, but perennial grasses retained their greenness longer into the season. Herbicide treated plots had more perennial grass cover (Chapter 2), which is likely why there was a higher NDVI compared to the non-sprayed plots during the second half of the time series.

Phenological differences can facilitate remote detection of nonnative invasive annual grasses (Bradley, 2014). Studies have used satellite remote sensing-derived maps of heavily invaded areas to create invasion risk models. For example, Bradley & Mustard (2006) used a phenology-based classification of heavy infestations of cheatgrass to correlate invasions with landscape scale features, including disturbances such as roads and powerlines as well as topography. However, the spatial and temporal resolution needed to identify phenological events at finer spatial scales prevent many satellite-based remote sensing approaches (Klosterman et al., 2018). Difficulties separating two species with similar phenologies, such as cheatgrass and medusahead, can present challenges in capturing differences (Bateman et al., 2020). This is also likely a difficulty for *ventenata* when using satellite imagery. Satellite platforms such as Landsat

30 m resolution data are ideal for time series analysis of invasive annual grasses, both for the spatial resolution and the length of the imagery archive. However, a challenge with using Landsat imagery is the 16-day return interval, since it may miss the green up of nonnative grasses which is a brief period during the early season. This is underscored in this study where there was a big shift in NDVI in a 7-day period between June 30 and July 7. This is something UAVs can overcome. UAV-derived imagery has a fine spatial resolution leading to more detailed and accurate identification and mapping (Wood et al., 2022; Weisberg et al., 2021; Yao et al., 2019).

Change detection is an important application in remote sensing, and for UAVs an advantage is fine spatial resolution which leads to the ability to detect small changes (Yao et al., 2019). Additionally, UAVs can capture more frequent imagery than satellite platforms, allowing for more timely management. Multi-date imagery can also help with detecting change in phenology which can be variable year to year for annual grasses (Bradley and Mustard, 2005; Bradley et al., 2018). *Ventenata* may be like other invasive annual grasses, but there is potential to differentiate it from them by using time series imagery and linking subtle phenological differences (Weisberg et al., 2021). In a study in Nevada, multi-temporal UAV imagery was used to distinguish multiple co-occurring species of invasive annual grasses from native vegetation, based on phenological differences in the timing of plant life phases over the course of a growing season (Weisberg et al., 2021). By sampling eight different dates of UAV imagery, a species-specific classification was best optimized using three flight dates: 19 May, 1 June, and 20 July (Weisberg et al., 2021). In a study in Montana, multi-date UAV flights were used to measure phenological variability within vegetation types to improve classification detail and accuracy

(Wood et al., 2022). They found an increase between a single flight and multiple flights during the growing season, and their approach demonstrated improved classification accuracy by utilizing multi-flight UAV classification approaches (Wood et al., 2019).

Species distribution models aim to map the areas likely to be suitable to a species (Guisan & Thuiller, 2005). Remote sensing can also help by identifying areas that are most vulnerable to invasion and prioritizing where to focus control efforts. For example, researchers from the University of Montana's Spatial Analysis Lab used satellite imagery from PlanetScope and Sentinel-2 to create a *ventenata* presence probability map for the Crow Reservation (Spatial Analysis Lab, 2021). My study complements their work by providing multi-date UAV imagery and has the potential to link the data to satellite imagery, which can further our understanding of *ventenata* in this part of Montana. Future work should include more UAV analyses that include classification efforts to improve detection of *ventenata* and based on training data with GPS locations. For example, high-resolution imagery from a UAV was used to create reference data for satellite-based image classification for the invasive grass buffelgrass (*Cenchrus ciliaris* L.) (Elkind et al., 2019). This should be done for *ventenata* in the future.

Managers need the ability to identify *ventenata* across entire landscapes, time and cost-effective approaches to evaluate risk of *ventenata*, and maps to develop targeted control efforts so they can work towards effective and efficient management approaches. A *ventenata*-specific map can give managers the flexibility to prioritize and direct management needs when attempting to control the spread of *ventenata* into non-invaded areas. NDVI is only one way to assess *ventenata* on the Crow Reservation, and more in-depth analyses of phenological differences will be needed to better understand *ventenata* and other annual grasses such as

Japanese brome (*Bromus japonicus* Thunb.) that can co-occur. Additional research is needed in this area and should focus on these phenological analyses that can link UAV data with satellite data to better understand the landscape. There is potential to dive further into remote sensing analyses for ventenata, such as classification of species, estimating cover of ventenata, and improving presence probability mapping by linking to UAV-derived data.

Remotely sensed classification of invasive plants relies on field data to train and validate resulting maps, however, this study did not collect the required data needed for these additional analyses to occur. Reference data for multiple species are needed. My study did not collect GPS coordinates around each treatment plot or sampling frame from Chapter 2 to link plant data to UAV imagery, and data were only collected from one relatively small site. This study was also limited to a site that was easy to access and did not have as high of ventenata cover as other sites. Two other sites had more cover (Chapter 2) and may be ideal sites to capture and continue future work.

Results from this study indicate that multi-date NDVI may serve as a useful indicator of vegetative change after annual grasses like ventenata are treated with herbicide. This can give land managers a better understanding of where herbicides are effective at reducing ventenata. Differences in NDVI are most notable in late June to early July when annual grasses begin senescing. A shift in NDVI over time was observed, as initially the non-sprayed plots had higher NDVI than sprayed plots. Without monitoring sprayed areas with on-the-ground estimates of vegetation cover, we may not know if an herbicide application was able to control ventenata. UAVs can help monitor large or inaccessible areas on the Crow Reservation to better inform land managers. Remote sensing can be a useful tool for managing invasive annual grasses with

its ability to cover large areas quickly and help distinguish between annual and perennial grasses. As the threat of invasive annual grasses increases in Montana, the importance of remote sensing will increase, making it a popular tool for land managers to use in management and monitoring.

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CHAPTER FOUR

CONCLUSIONS

Summary of Findings and Implications

The goal of this thesis was to test and find adequate control options for ventenata (*Ventenata dubia*) in southeastern Montana on the Crow Reservation and to test whether Unmanned Aerial Vehicles (UAVs) could be used to detect differences in Normalized Difference Vegetation Index (NDVI) of vegetation in response to treatments. Ventenata has been well documented in the Intermountain West and has been expanding into new ecosystems like the sagebrush steppe and Great Plains. Ventenata was listed as a noxious weed in Montana in 2019 and managers have been searching for control strategies. On the Crow Reservation, there is more recent growing concern for ventenata spread. The research summarized in this thesis provides two control options that proved to be effective, and that UAVs may be able to detect changes in NDVI between sprayed and non-sprayed areas to inform effectiveness of herbicides.

The field study described in Chapter 2 provided promising results for reducing ventenata at four sites one growing season post-treatment. My objective was to determine effectiveness of indaziflam (Rejuvra®) and imazapic (Plateau®) applied at two water carrier rates, and the soil amendment (NutraFix™, Edaphix™), applied at a high and low rate, for reducing ventenata on grasslands in southeastern Montana on the Crow Reservation. Based on previous studies (Harvey, 2019; Hart & Mealor, 2021; Sebastian, et al., 2016, 2017), I hypothesized that indaziflam would provide the highest reduction for ventenata, followed by imazapic and the soil amendment. I also hypothesized that the higher rates would reduce ventenata cover more so than

the lower rates. Results showed that imazapic is just as effective as indaziflam after one growing season post-treatment. The soil amendment reduced ventenata to some degree at two of the four sites but was not as effective as the herbicides. High and low application rates did not differ. Litter and bare ground increased in the herbicide plots, but western wheatgrass also increased in those plots suggesting that this desirable perennial grass can recover after herbicide treatment.

The remote sensing field study described in Chapter 3 used a UAV to capture weekly multispectral imagery and provided a time series that began on 24 May and ended 08 August 2022. The objective was to determine differences in NDVI in response to the treatments from the treatment-based field study focused at one of the four sites. I hypothesized that there would be a difference in NDVI between the herbicide sprayed plots and the non-sprayed control and that the relationship would change over the course of the growing season due to the different abundances of and green up times of annual grasses vs perennial grasses. I expected the sprayed plots to have lower cover of annual grasses, mainly ventenata, and higher cover of the dominant perennial grass western wheatgrass, therefore, I expected NDVI to be higher and stay greener longer in these plots compared to the non-sprayed control. Results demonstrated that NDVI was higher in the sprayed plots and remained higher than the non-sprayed after 07 July. This may have been when annual grasses senesced leaving those plots with a lower NDVI. By using UAV imagery post-treatment, managers may be able to capture this phenology difference between invasive annual grasses and native perennial grasses to monitor effectiveness of treatments. Flying a UAV in early July may capture this NDVI shift. This could be particularly helpful over large areas like those encompassed by the reservation.

These two studies provide land managers with tools to reduce and stop the spread of ventenata. There are limited studies on this species and more research is needed, especially in this part of Montana. More effective management tools will help to control ventenata and, in turn, protect our rangelands and our tribal lands.

Future Research

More research is needed on control options for ventenata and longer monitoring beyond one growing season is needed. Furthermore, testing indaziflam, imazapic, and the soil amendment in combination with each other and other herbicides known to control invasive annual grasses may provide more effective options. Findings from the UAV study show that NDVI can shift between sprayed and non-sprayed plots which can suggest when to monitor for effectiveness. More research is needed on linking UAV data to satellite platforms to further monitor NDVI changes and to detect ventenata over the landscape. Research can include ventenata specific maps for targeted management. Combining on the ground methods with remote sensing can lead to better management of ventenata.

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