

# **Oregon Beef Council** Report

# **2020 Edition**



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# Oregon Beef Council Report



Beef Cattle Sciences

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Report



BEEF192

**Beef Cattle Sciences** 

# Use of In Virto-In Vivo Hybrid Approach to Study the Nutrigenomic Effect of Fatty Acids on Cattle <sup>1</sup>

Massimo Bionaz<sup>2</sup> and Sebastiano Busato<sup>3</sup>

## Synopsis

The use of multiple fatty acids, specifically palmitic acid, stearic acid, and lauric acid, is more effective in activating the nuclear receptor Peroxisome Proliferator-Activated Receptor in cow than the use of a single fatty acid.

## Summary

Dairy cows undergo a significant degree of metabolic stress in the transition between pregnancy and parturition. The resulting negative energy balance causes a breakdown of body fat, which enters the bloodstream as nonesterified fatty acids (NEFA). It is unclear if NEFA have nutrigenomic properties, but it is difficult to assess this in vivo. In order to overcome such challenge, in the present work we developed a in vivo-in vitro hybrid method. We hypothesized that high circulating NEFA in the peripartum can be used as a substrate for the activation of the Peroxisome Proliferator-Activated Receptor (PPAR), a transcription factor with known nutrigenomic effects. We collected blood from 3 jersey cows at three different points (-40d, -10d and +10d relative to parturition) and used the serum from those to treat mammary (MAC-T) and liver (BFH-12) cells to assess PPAR activation. The results

showed a great response in both cell lines to increasing concentration of circulating NEFA in the blood, demonstrating the causal link between the two in the transition period. Furthermore, we demonstrated that circulating NEFA preferentially activate PPARδ, followed by PPAR $\gamma$  and PPAR $\alpha$  (but not in BFH-12). By comparison, we tested the effect of palmitic acid, one of the most common fatty acids, showing that palmitic acid alone activated PPAR $\delta$  and PPAR $\alpha$ , but not PPAR $\gamma$ . This suggests that additional fatty acids found in the NEFA pool are responsible for the activation of PPAR in the transition period. Our results substantiate the importance of acquiring further insight on the interaction between NEFA and PPAR, and highlight the viability of the in vivoin vitro hybrid system developed and reinforce the importance of nutrigenomics as a potential tool for farmers.

#### Introduction

Nutrigenomics: a revolutionary approach to improve efficiency and well-being in cattle (Bionaz et al., 2015). Nutrigenomics is a scientific branch of nutrition that studies how nutrient compounds contained in feedstuff can modify the biology of the organism by interacting with the animal's genetic code (Bionaz et al., 2015). Because the potential of

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nutrigenomic studies is enormous: once deciphered the effect on the genome of a compound present in the feed, such effect can be leveraged to fine-tune the biology of an organism by increasing (or decreasing) the amount of that specific compound in the diet. The discovery of the nutrigenomic effect of feedstuff compounds can allow to have a powerful and economically viable means to improve health, efficiency of production and/or quality of the cattle products. This poses a question: which dietary compound has known nutrigenomics properties that can be used in cattle?

#### Fatty acids have nutrigenomics properties

It is becoming evident that fatty acids (the main components of the fat in the diet) can affect the expression of genes. They can do this by binding and activating proteins (i.e., transcription factors) in cells that turn on or off specific genes. It is known that certain fatty acids, such as palmitic acid (one of the most common fatty acid presents in animal products) have a positive effect on the overall physiology of the cattle when supplemented to cows (Loften et al., 2014). Besides the overall physiological improvement, this fatty acid also increases the production of milk fat (Bionaz et al., 2015). Data generated by our and other research teams indicated that the positive effect of palmitic acid in cattle is due to a nutrigenomic effect by activating a transcription factor called peroxisome proliferator-activated receptor (PPAR) (Bionaz et al., 2013). Why should we care about PPAR?

## Role of PPAR in cattle and differences between species

Since its discovery in 1992, PPAR has been studied extensively in several species. Through subsequent molecular studies, three distinct PPAR have been identified: PPAR $\alpha$ , PPAR $\beta/\delta$ , and PPAR $\gamma$ . Individual PPAR have been studied extensively and have been shown to play different role in essential biological processes (Bionaz et al., 2013, Bionaz et al., 2015). PPAR $\alpha$  is a master regulator of fat metabolism in the liver, PPAR $\beta/\delta$  plays a central role in muscle function and tissue repair,

and PPAR $\gamma$  is essential for the biology of the fat tissue. All of them play key roles in the immune system and inflammation (Mandard and Patsouris, 2013). All those functions are crucial in cattle, especially early post-partum where the metabolism of fat increase dramatically, the immune system is impaired, and a lot of tissue repair has to happen (Bionaz et al., 2013, Loor et al., 2013, Bionaz et al., 2015). The biological importance of the three PPAR justifies the need for accurate information on their activation and function. While for some species we possess abundant information for the three PPAR, ruminant PPAR studies are still rather primitive. Furthermore, several key differences have been identified between bovine PPAR and other species; for example, the involvement of PPAR in controlling milk fat production appears to be unique of ruminants (Osorio et al., 2016). Due to the different importance of each of the three PPAR, how can we activate those individually using dietary fatty acids?

#### Activation of PPAR by individual fatty acid.

Data on activation of each PPAR by dietary fatty acids have been generated in mouse and human, but not in cattle. Unfortunately, data generated in mouse or human are not relevant for bovine (Bionaz et al., 2013). Prior data clearly indicated that PPAR in mouse responds strongly to fatty acids enriched in oil (Moya-Camarena et al., 1999), while bovine PPAR is more sensitive to solid fat (Bionaz et al., 2013, Bionaz et al., 2015, Vargas-Bello-Perez et al., 2019), including the fat commercially available as supplement for cattle. Among these, palmitic acid appears to be a potent activator of PPAR in cattle, while it is a weak activator of PPAR in mouse (Bionaz et al., 2013). For the above reasons it is essential to investigate activation of each individual PPAR by dietary fatty acids in bovine. How can we study the activation of each PPAR by dietary fatty acids?

## Circulating fatty acids activate PPAR in bovine cells.

In a prior work founded by the Oregon Beef Council, we demonstrated that circulating fatty acids (a.k.a., non-esterified fatty acid of NEFA) activate PPAR in a dose-response fashion (Busato and Bionaz, 2020). The same study revealed that NEFA activate PPAR $\beta/\delta$  and PPAR $\gamma$  but not PPAR $\alpha$ . Furthermore, the data indicated that the addition of palmitic acid into the serum of dairy cows increase further the activation of PPAR, particularly PPARB/8 and PPAR $\alpha$ but not PPAR $\gamma$ . All of the above support a strong role of fatty acids in activating PPAR and the possibility of a nutrigenomic intervention to modulate those nuclear receptors. However, it remains to be determined which fatty acid is the strongest activator of PPAR, what the optimal dose of each fatty acid to activate PPAR, and the determination of the mixture of fatty acids that maximize PPAR activation in dairy cows. Thus, the objectives of the present study was to determine the activation of PPAR in bovine by the most important fatty acids present in the diet of dairy cows and 2) investigate the mixture of fatty acids that maximize PPAR activation in bovine.

### **Materials and Methods**

## Culture, Transfection and Measurement of PPAR activation in immortalized bovine cells

Mammary alveolar cells transformed (MACT) already present in our laboratory and immortalized bovine hepatic cell line (BFH-12) obtained from another laboratory (Gleich et al., 2016) were used. Culture medium was changed every 48 h and cells were subcultured to 70 to 80% confluence (approximately every 3 to 4 d). For the experiments, approximately 30,000 or 3,000 cells/well were plated in 96-well or 384well plate, respectively. Twenty-four hours later the cells were co-transfected with a PPAR Response Element associated with luciferase and a renilla plasmid at 50:1 ratio of luciferase/renilla plasmid. Treatments were applied 16 or 24 h post-transfection. Luciferase and renilla activity were measured via luminometer. For the experiments, also a HP 300e Digital Dispenser was used.

### Treatments

#### Dose-response of single fatty acids

In order to assess the activation of PPAR by various fatty acids we used purified fatty acids available commercially, those included palmitic acid, stearic acid, palmitoleic acid, octanoic acid, decanoic acid, dodecanoic acid, myristic acid, linoleic acid, linolenic acid, and phytanic acid. Cells previously transfected with the plasmids as above described, were treated with increased dose of each fatty acid using a HP D300e Digital Dispenser. Luminescence as index of PPAR activation was measured 24h later. The cells were treated with the fatty acids in an artificial media used to cultivate and growth cells.

#### Response to mixture of fatty acids

Based on the results from the doseresponse to single fatty acids, cells were treated with a mixture of 2 fatty acids at the dose that reached the higher activation of PPAR or using a mixture of 3 fatty acids but using 1/3 of the dose that maximize PPAR activation (all fatty acids were suspended in DMSO, which is toxic above a certain level). PPAR activation was assessed via luminometer 24h later. *Statistical Analysis* 

Data were analyzed using a generalized linear model (proc GLM, SAS 9.4) with LSD contrasts. Linear, quadratic and cubic effect of the dose response was assessed using GLM. Significance was declared with P<0.05.

## Results

### Dose-response activation of PPAR reveals lauric acid to be a potent PPAR activator

The results of the dose-response of the various fatty acids is reported in Figure 1. Results clearly indicated that most of the fatty acids known to be present in the diet of dairy cows activate PPAR in vitro, with higher activation detected by palmitic, stearic, and lauric acid. Most fatty acids have a quadratic effect, with a peak activation around 150-200 µM, similar to the level of NEFA in blood of cows in mid-lactation (Cozzi et al., 2011). The largest activation was observed with lauric acid; however, with dose  $>500 \mu$ M that is very high. Palmitic acid and stearic acid had the highest activation with doses around 150-200 µM, which support prior data in bovine using gene expression techniques (Bionaz et al., 2013). Other fatty acids, such as oleic acid and

palmitoleic acid surprisingly and contrary to prior data (Kadegowda et al., 2009, Bionaz et al., 2013, Vargas-Bello-Perez et al., 2019) did activate PPAR with dose around 100  $\mu$ M. Other fatty acids, including linoleic acid did not activate PPAR.

# Mixture of fatty acids is more effective in activating PPAR compared to single fatty acids.

Although theoretically PPAR bind fatty acids individually, it has been reported that certain types of PPAR bind more than one fatty acid simultaneously (Bionaz et al., 2013, Bionaz et al., 2015). More importantly, circulating fatty acids are composed of a mixture of fatty acids. Therefore, based on results of Figure 1, we combined 2 or 3 fatty acids with the dose showing to maximally activate PPAR. Results clearly indicated that combination of 3 fatty acids is more effective than single or 2 mixture of 2 fatty acids to activate PPAR (Figure 2). More importantly, the dose of 3 fatty acids was 1/3 of the dose used for the mixture of 2 fatty acids. Interestingly, the activation of PPAR with the mixture of fatty acids was up to 14-fold compared to non-treated cells while the maximal activation of PPAR by using a single fatty acid was 6-fold (Figure 1).

### Conclusions

Our data clearly indicated that each fatty acid has an optimal dose to activate PPAR. In addition, our experiment demonstrated that a mixture of specific fatty acids even with a 1/3 the dose that maximize activation of PPAR when used as single fatty acids is more effective in activating PPAR than the use of each fatty acid or the mixture of 2 fatty acids.

Our data clearly indicated that we need to discover the right mixture of fatty acids and the proper dose of it in order to be able to activate PPAR in vivo. The produced data are providing fundamental data to move toward in vivo application of nutrigenomics. However, *in vivo* dosing is not easy, especially considering the complexity of the utilization of dietary fatty acids in ruminants, as recently reviewed (Bionaz et al., 2020). Thus, next step would be to perform an in vivo experiment using the mixture we detected to be most effective. This is our current experiment that was again funded by the Oregon Beef Council.

## Acknowledgments

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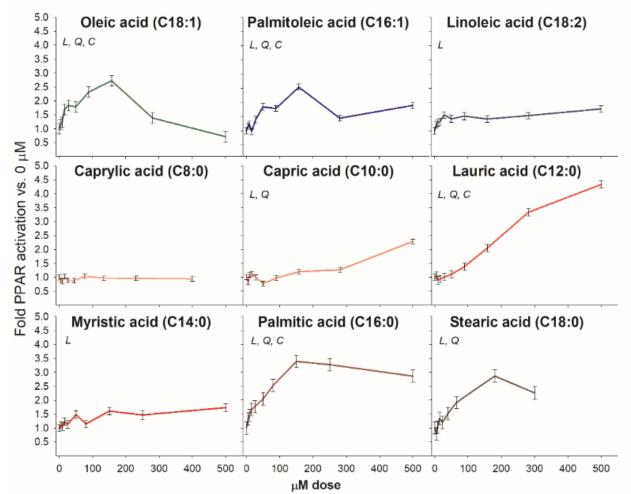
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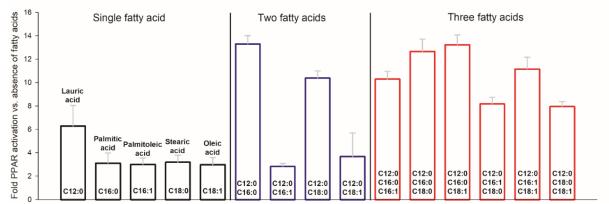
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**Figure 1**. Bovine cells were treated with an increase concentration of several fatty acids in media and activation of the PPAR was measured. Letter in the graph denote significant (P<0.05) linear (L), quadratic (Q), or cubic (C) statistical effect.



**Figure 2**. Hepatic bovine cells were treated with single or combination of fatty acids and activation of PPAR measured. Presented are the combinations of each fatty acid with lauric acid (C12:0).



Report



BEEF193

**Beef Cattle Sciences** 

## Endometrial Inflammatory Cytokine Expression in Postpartum Beef Heifers Follow Platelet Rich Plasma Treatment <sup>1</sup>

Victor O. Perez<sup>2</sup> and Michelle A. Kutzler<sup>3</sup>

### **Synopsis**

Intrauterine treatment with platelet rich plasma after calving does not alter endometrial inflammatory cytokine expression.

### Summary

Current treatments for reproductive problems in beef cattle involve hormones and antibiotics which are expensive and often ineffective. Our objective was to test the response of endometrial pro-inflammatory cytokine (IL-6, IL-8,  $TNF\alpha$ ) expression in postpartum beef heifers to intrauterine platelet rich plasma (PRP), platelet poor plasma (PPP), and saline. Twelve Angus-crossbred heifers calved under supervision. Nine heifers calved normally (eutocia) and three heifers needed assistance (dystocia). The eutocia heifers were divided equally into groups: PRP, PPP, and saline-treated eutocia (EUT). The dystocia heifers were also administered intrauterine saline. Endometrial cells were collected at 2and 4-weeks post-calving using a doubleguarded endometrial swab past through the cervix. Intrauterine treatment was administered once after samples were collected 2-weeks postcalving. Total RNA was isolated from endometrial cells using a standard phenol-

chloroform protocol. Complementary DNA (cDNA) was prepared from the total RNA using a commercial kit. Previously validated forward and reverse primer sequences for IL-6, IL-8, and TNF $\alpha$  as well as the housekeeping gene ( $\beta$ actin) were used in the current study for realtime polymerase chain reactions to determine relative gene expression. Data were analyzed using the 2- $\Delta\Delta$ CT method followed by paired Student's t tests to compare the relative expression of each group to saline-treated eutocia controls. There was no significant effect of treatment (PRP, PPP) on expression of IL-8, IL-6, or TNFα (p>0.05). However, TNFα expression was significantly reduced in the saline-treated dystocia heifers compared to the saline-treated eutocia heifers. In addition to the small sample size, another limitation in this study was that none of the heifers treated had clinical evidence of endometritis. Follow-up research investigating cytokine gene expression in heifers with endometritis and in heifers that experienced dystocia that are treated with platelet rich plasma is needed.

### Introduction

Calving difficulty in first-calf heifers is an important economic issue in the beef industry not only because of the risks to the calf, but also

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because of the effects of impaired fertility following delivery on the mother. While efforts are made to minimize factors that contribute to calving difficulty (e.g. using expected progeny differences (EPDs) for lower birth weight or improved calving ease), the overall prevalence of beef heifers needing assistance is still 10-20%. Failure to conceive at second mating is the most common reason for heifer attrition. About 4% of heifers are culled at second mating after being diagnosed non-pregnant and about 2.3% are carried over as non-pregnant 3-year-old heifers.

During calving, the uterus is exposed to bacterial contamination, which can cause inflammation of the uterine lining (referred to as "endometritis"). If calving is prolonged, the severity of bacterial contamination and endometritis increases. In healthy cattle following a normal calving, bacterial contamination and endometritis are spontaneously cleared within two weeks, which can be confirmed using bacterial culturing endometrial cytology methods (e.g. reduction in the number of neutrophils). However, about 40% of beef cattle fail to spontaneously clear bacterial infections and/or have prolonged inflammatory conditions that persist more than 50 days postpartum, which severely affects fertility. It is important to note that these animals do not show any external evidence that there is a problem (referred to as "subclinical"). Ricci and coworkers (2015) reported that only 13% of beef heifers with subclinical endometritis were pregnant within 130 days postpartum. In addition, Sheldon and colleagues (2016) reported that subclinical endometritis reduces pregnancy rates in beef heifers by 16%. Despite this, research in this area has been very limited.

In cattle, many therapeutic agents and procedures have been used to treat endometritis, including systemic or intrauterine administration of antibiotics, or administration of PGF2 $\alpha$ (Lutalyse®) or its analogue (Estrumate®). The efficacy of most of these treatments is low, while the costs (labor and drugs) are high. It would be highly desirable for beef producers to have a specific treatment aimed at reducing uterine inflammation that would not result in meat residues.

Platelet rich plasma (PRP) is an emerging therapeutic application in tissue regeneration because of its enrichment with growth factors and anti-inflammatory properties. Platelet rich plasma is known to accelerate the healing process in human medicine and has been used in facial surgery, muscle and tendon repair, and reversal of skin ulcers. In veterinary medicine, it has been mainly used for promoting equine tendon repair, but there are some reports of its use in intestinal wound healing in pigs and in skin wound healing in dogs. In dairy cattle, PRP has been used to treat mastitis, repeat breeders, and to increase embryo production in embryo transfer programs. Previous Oregon Beef Council funded research demonstrated that administration of intrauterine PRP to normal calving heifers with no overt evidence of endometritis significantly decreased the number of inflammatory cells (specifically macrophages) present on endometrial cytology compared to control heifers treated with intrauterine platelet-poor plasma (PPP) or saline groups (Puttman & Kutzler, 2018). In an in vitro study, PRP downregulated pro-inflammatory genes (e.g. interleukin-8) in cultured bovine endometrial cells (Ghasemi et al 2012). Further research needs to be done to study the in vivo effect of PRP on endometrial pro-inflammatory genes.

The objective of the proposed research was to investigate the in vivo effect of postpartum PRP treatment on bovine endometrial proinflammatory cytokine gene expression. We hypothesized that intrauterine treatment with PRP will decrease endometrial interleukin-6 (IL-6), interleukin-8 (IL-8), and tumor necrosis factor-alpha (TNF $\alpha$ ) gene expression in postpartum beef heifers.

### Materials and Methods Animals, Sample Collection, and

**Treatment Protocol:** The platelet rich plasma was prepared by collecting blood from a donor Jersey in good health and free of contagious

diseases (e.g. Johne's disease, bovine leukemia virus), using the double centrifugation method used by Lange-Consiglio and colleagues (2014). Individual centrifuge tubes of PRP and platelet poor plasma (PPP) were frozen at  $-80^{\circ}$ C, thawed at 37°C, and then combined. The PRP and PPP were subjected to aerobic and anaerobic bacteriological examination to verify their sterility. The platelet concentration of PRP was determined through use of a hemocytometer and the concentration was determined to be  $1 \times 10^{9}$  platelets/ml. Finally, the PRP and PPP were aliquoted in 10 ml ready-to-use doses and kept frozen at  $-20^{\circ}$ C until use.

Commercial crossbred Angus heifers (n=12) from the Oregon State University (OSU) Soap Creek Ranch were used for this study. Heifers were closely monitored for signs of calving at the OSU Hogg Animal Metabolism Building under supervision. Nine heifers calved normally without any assistance (eutocia) and three heifers had difficulty calving and needed to have their calves pulled (dystocia). Heifers and their calves were transported to the OSU Soap Creek Beef Ranch within 48 hours of calving. Two weeks after calving, eutocia heifers were randomly divided into three equal groups: saline-treated heifer eutocia (SHE; n=3), PPPtreated (n=3), and PRP-treated (n=3). All dystocia heifers were treated with saline (SHD; n=3)

To obtain endometrial cells, a doubleguarded endometrial swab was passed through the cervix. Briefly, heifers were separated from their calves and restrained in a squeeze chute. Feces was manually removed from the rectum to facilitate manipulation of the swab through the cervix. The perineal area was thoroughly cleaned with a disinfectant soap and water to remove any feces or other debris on the vulva or within the vestibule. Then an endometrial swab was passed through the vulvar lips and vagina to the external cervical os. Using the left hand to maneuver the cervix through the rectal wall, the endometrial swab was threaded through the cervical lumen into the uterine body with the right hand. Once inside the uterus, the inner guard was advanced past the outer guard and the

swab was advanced past the inner guard. The swab was rolled on the lining of the endometrium several times to facilitate removal of endometrial cells. The swab was then withdrawn into the inner guard, the inner guard was withdrawn into the outer guard, and the double-guarded swab was removed from the uterine, cervix, vagina, and vulva. The endometrial swab was then cut with scissors to a length of 2 cm and put into RNAse-free microcentrifuge tubes containing 1 mL of TRIzol<sup>TM</sup> reagent (cat# 15596026, Invitrogen<sup>TM</sup>, Carlsbad, CA). The tube was then flash frozen in liquid nitrogen and transported from the farm back to the laboratory. Tubes were stored at -80°C until total RNA was isolated.

Following endometrial sample collection, SHE and SHD heifers received an intrauterine infusion of 10 mL of 0.9% sterile saline, while the PRP and PPP groups received equal volume of platelet rich plasma or platelet poor plasma, respectively. Two weeks later (4 weeks after calving), endometrial samples were collected in the same way as previously described.

**RNA Isolation and Generation of Complementary DNA:** Tubes containing the swabs were thawed on ice and vortexed briefly. Swabs were then removed with forceps and total RNA was isolated using a standard chloroform and ethanol extraction. The total RNA concentration was determined using a bioanalyzer (NanoDrop One, catalog #A38189, ThermoFisher Scientific, Carlsbad, CA).

Complementary DNA was then made from total RNA samples using a commercial cDNA kit (SuperScript <sup>TM</sup> First-Strand Synthesis System for RT-PCR, #11904018, Invitrogen<sup>TM</sup>, Carlsbad, CA) following the manufacturer's instructions. Complementary DNA samples were stored at -20°C until used for real-time polymerase chain reaction (RTPCR).

**Primers and Real-Time Polymerase Chain Reaction:** Forward and reverse primer sequences for IL-6, IL-8, and TNF $\alpha$  as well as the housekeeping gene ( $\beta$ -actin) had been previously validated for use in bovine endometrial gene expression studies (Ghasemi et al 2012). Commercial primers (25 nmole DNA Oligo standard desalting, Integrated DNA Technologies, Coralville, IA) were prepared from these sequences. Each primer was then reconstituted with 250  $\mu$ L of buffer, vortexed briefly, and then diluted in a new microcentrifuge tube to a concentration of 500 nM in 100  $\mu$ L.

The primers were added RTPCR kit components (Fast SYBR <sup>TM</sup> Green Master Mix, #4385612, Applied Biosystems, ThermoFisher Scientific, Carlsbad, CA) to create a mastermix, which was briefly vortexed and centrifuged. For each RTPCR reaction, 45  $\mu$ L of master mix and 5  $\mu$ L of each cDNA sample was added to the respective wells of the 96-well plate and mixed by pipetting. The RTPCR plate was then placed it into an RTPCR thermocycler (7500 Fast Real-Time PCR System, 4351106, ThermoFisher Scientific, Carlsbad, CA) with manufacturer's recommended cycling conditions.

**Data Analysis:** Data were analyzed using the  $2-\Delta\Delta CT$  method followed by paired Student's t tests to compare the relative expression of each group to saline-treated eutocia controls.

### Results

There was no significant effect of either treatment (PRP, PPP) on expression of IL-8 (p=0.482 and p=0.149, respectively; Figure 1), IL-6 (p=0.086 and p=0.355, respectively; Figure 2), or TNF $\alpha$  (p=0.147 and p=0.195, respectively; Figure 3) gene expression. In addition, there was no significant effect of dystocia (SHD) on expression of IL-8 or IL-6 (p=0.316 and p=0.059, respectively; Figures 1 and 2). However, TNF $\alpha$  gene expression was significantly reduced in heifers with dystocia treated with saline compared to heifers with eutocia treated with saline (p<0.000; Figure 3).

### Limitations

A limitation of the current study was the small sample size (three heifers per group). Although not significant, there was a trend for a reduction in endometrial IL-6 gene expression following intrauterine PRP treatment. In addition to the small sample size, another limitation in this study was that none of the heifers treated had clinical evidence of endometritis. However, heifers that had experienced dystocia and were treated the same as controls had a trend towards a reduction in IL-6 and a significant reduction in TNF- $\alpha$  gene expression. Additional research is needed to evaluate changes in endometrial gene expression in normal calving and difficult calving heifers to determine if these changes could contribute to reduced pregnancy rates.

## Acknowledgments

This research study was financially supported by the Oregon Beef Council.

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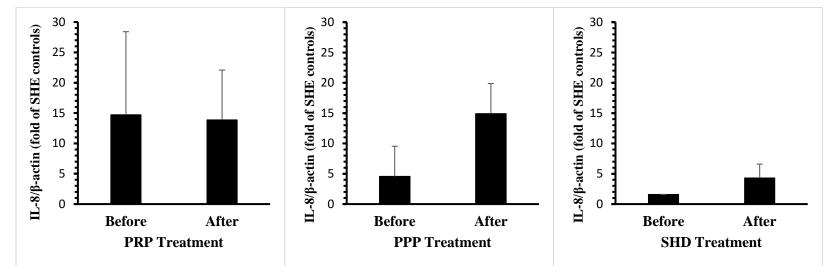
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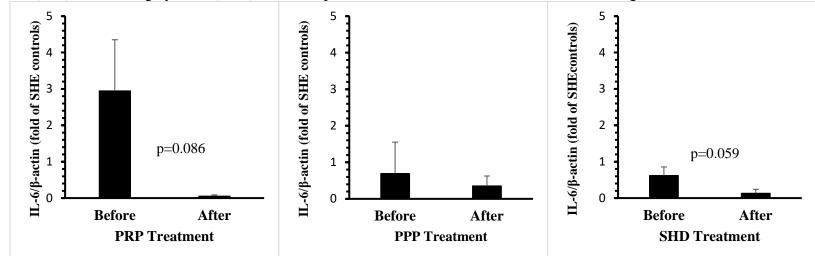
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Endometrial Inflammatory Cytokine Expression in Postpartum Beef Heifers Follow Platelet Rich Plasma Treatment

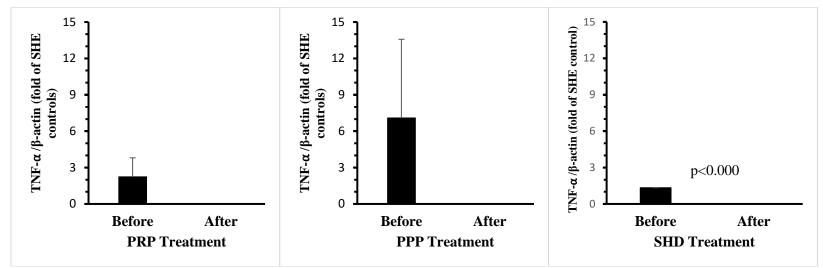


**Figure 1.** There was no significant difference in IL-8 gene expression in either treatment group (platelet rich plasma (PRP) or platelet poor plasma (PPP) or following dystocia (SHD) when compared to intrauterine saline-treated normal calving heifers.



**Figure 2.** There was no significant difference in IL-6 gene expression in either treatment group (platelet rich plasma (PRP) or platelet poor plasma (PPP) or following dystocia (SHD) when compared to intrauterine saline-treated normal calving heifers.

Endometrial Inflammatory Cytokine Expression in Postpartum Beef Heifers Follow Platelet Rich Plasma Treatment



**Figure 3.** There was no significant difference in TNF- $\alpha$  gene expression in either treatment group (platelet rich plasma (PRP) or platelet poor plasma (PPP) when compared to intrauterine saline-treated normal calving heifers. However, there was a significant reduction in the saline-treated heifers that had dystocia.



## Report



**Beef Cattle Sciences** 

## Progress Reports – Animal Sciences<sup>1</sup>

## Monitoring Cattle Behavior to Identify Cattle Disturbance Remotely

Contact Person: Sergio Arispe, Associate Professor, Oregon State University Extension Service-Malheur County, Ontario, OR 97914 Email: Sergio.arispe@oregonstate.edu

**Project Objectives:** The research *goal* is to classify and monitor cattle behavior to identify when calves are removed from their mothers during a simulated theft. The hypothesis is that machine learning will detect a change in maternal behavior after the calf is removed.

**Project Start Date**: The project begins spring 2021 after calving. The GPS cow collars will be purchased using the limited funding amount.

#### Project Completion Date: Fall 2021

**Project Status and Preliminary Findings:** Introduction: Cattle theft—*rustling*—is as common today across the vast landscapes of central and eastern Oregon as it was in the late 1800s. Local cattle producers believe that thieves, or rustlers, have an intimate knowledge of the terrain, but more importantly, are familiar with the cow-calf producers' routine schedules, which are closely associated with the culture and heritage of rural communities. They suspect that *rustlers* heist unbranded, newborn calves when ranchers least suspect it—during Sunday church or during a community event. Historically, the owners would rely on either local law enforcement or the brute force of a rifle to deter theft. Today, there is arguable a more powerful, non-lethal weapon—*artificial intelligence*—that could give cattle producers an edge.

In Malheur County, the problem is widespread. To combat the problem, the Malheur County Sheriff Department increases aircraft surveillance when beef cows are calving each spring. Around April 1<sup>st</sup>, cow-calf operators move cows away from the ranch headquarters to more distant rangelands where cows and calves have access to fresh grass. Cows that have not calved by that time will give birth on the new pasture.

Ranchers typically wait until the end of the calving season to brand calves with their unique brand—indicating ownership. It is in this window of opportunity where cattle rustlers come in and heist the calves. Today, a 550-pound weaned calf costs nearly \$800. However, in a 2015 economy, that same weaned calf would sell for nearly \$1,600. Multiply that average by 70 calves heisted in two years and you will have a sense of how much money Malheur County cow-calf producer has lost in the past two

<sup>1.</sup> This document is part of the Oregon State University – 2020 Oregon Beef Council Report. Please visit the Beef Cattle Sciences website at <u>http://blogs.oregonstate.edu/beefcattle/research-reports/</u>

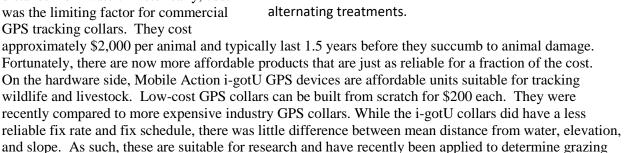
years—\$84,000. The cow-calf operator, like other cow-calf operators who fall victim to rustling, is searching for ways to mitigate losses so that his family operation can remain economically viable. Recent advances in hardware and software may be able to help cattle producers.

Every day, computers can process data more efficiently and more purposefully. Machine learning (ML) is a field of data analysis whereby a human automates analytical models to identify patterns. It is a branch of artificial intelligence centered on the notion that minimum human intervention is necessary for a system to learn from patterns within data. For example, you may have previously seen an alert on your smart phone indicating that the traffic situation has changed and you should budget in additional time or change your route. Another common instance is a smartphone alert indicating that you based on your location, you will be late to a meeting if you fail to move quicker. Artificial intelligence,

made from ML, provides direct feedback based on large data sets.

Global positional system (GPS) devices can be research instruments as they communicate with a network of satellites to fix the device location. Researchers have successfully used GPS device technology to monitor grazing distribution and activity (Anderson et al., 2012). The location can be combined with a digital elevation model to obtain additional data. such as elevation use, slope, and

distance from water. Historically, cost was the limiting factor for commercial GPS tracking collars. They cost

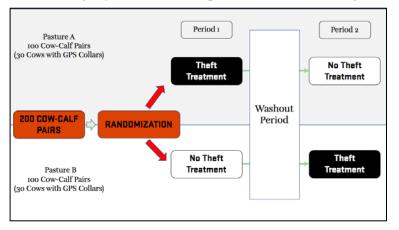


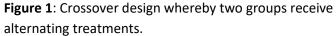
distribution (Knight et al., 2018).

On the software side, interdisciplinary research efforts have developed innovative algorithms to predict cattle behavior. For example, a group in Australia trained ML algorithms to classify cattle behavior from collar, halter, and ear tag accelerometer sensors (Rahman et al., 2018). Accelerometers measure the change in gravitational acceleration on or near the head and can indicate livestock behavior. Different magnitudes produced by the accelerometer correspond to different behaviors. In this case, the authors used ML to estimate the distribution of grazing, resting, walking, standing, ruminating, or other actions. It demonstrates that ML can be applied to assess cattle behavior.

Methods & Materials: To test the hypothesis that maternal behavior will change after a simulated theft, the researcher will use an industry cost match a cow-calf operator to purchase 60 Mobile Action i-gotU GPS collars. To test the hypothesis, the researcher will use a crossover design whereby two groups receive similar treatments-control (no simulated theft of calves) and a simulated theft of calves. A Malheur County cow-calf operator has also committed 200 beef cows, and their calves (cowcalf pairs), to the proposed experiment.

Two hundred cow-calf pairs will be randomly divided into two groups of 100 cow-calf pairs (Figure 1). On the initial day (day 0), the researcher and cow-calf operator will deploy 60 GPS collars on 30 random cows in each group. The GPS device within the collar will collect a waypoint every two





seconds. Cows will be given 48 hrs to adjust to the GPS collars. On day 2, we will simulate a theft treatment in Pasture A. The cow-calf operator will drive a trailer into the pasture, load calves into a trailer, and then drive the trailer across the fence line at the nearest gate. The cow-calf operator will wait for 15 minutes before returning the calves to the pasture with their mothers. There will be a 24-hour washout period whereby cattle in Pasture A adjust to the treatment disturbance. On the fourth day, cow-calf pairs in Pasture B will receive the same simulated theft treatment as Pasture A cow-calf pairs. GPS collars will be removed 24 hrs after the second simulated theft, and the data will be processed.

The GPS waypoint data that is acquired every two seconds will be downloaded and stored in a spreadsheet. The researcher will use the Python computing environment and the scikit.learn machine learning library. Supervised machine learning will be used in this experiment. Supervised learning is one type of machine learning that trains the machine using example input and output data. The input data in this case will be GPS position and characteristics (e.g., lateral cow speed). The output data will be a "Yes" if the calf was taken from the cow and a "No" if the calf was not taken. In this way, the algorithm will learn which patterns in the GPS data indicate that the calf was taken. This type of classification scheme is a called a binary classification. The event (i.e., theft) either happened or it did not. The researcher will use a subset of each group of cows to train the machine and then use cross-validation methods with the remaining cow-calf pairs to determine, quantitatively, how accurate this approach is. During this process different supervised learning algorithms (e.g., support vector machines and decision trees) will be tested to determine if one method out-performs the others.

During the data process and classification, the two second data will be down-sampled to 5s, 10s, 30s, and 60s to identify the sensitivity of the classification scheme to the sample rate. The goal of this is to determine the minimum amount of GPS time series data needed to accurately classify the cow behavior. This is an important parameter to know because the less frequent GPS data is collected, the longer the GPS sensor can collect data due to the finite battery life within the collars. The data sample rate also has implications for real-time data transfer and monitoring via cellular or satellite modems that could be installed in a future generation of the collar.

**Projected Impact:** The proposed interdisciplinary research has potential impacts that are scientific and economic. Potential scientific impacts are that our research will provide foundational work using ML to identify theft from cattle behavior. If our hypothesis is correct, we can expand different theft scenarios to characterize and compare cattle behavior. For example, does maternal behavior from a simulated theft from a cornstalk pasture differ from a simulated theft on the sagebrush steppe. Additionally, does maternal behavior differ when cattle are collected with four wheelers compared to horseback? Potential impacts will also be economic. The mere site of 60 cows with GPS collars may deter cattle rustlers from approaching the herd. Tangible potential impacts could also be developed with real-time GPS monitoring and combined with messages sent to the producer when abnormal behavior is encountered. Once this technology develops, cow-calf operators would theoretically receive updates on their movements and location. Those data have the potential to allow cow-calf operators to develop their pasture to get a more even grazing distribution. In the distant future, potential impacts could also be economic by deterring livestock wildlife interaction (e.g., cattle and wolves).

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## Report



**Beef Cattle Sciences** 

## Progress Reports – Animal Sciences<sup>1</sup>

## **Evaluating Methods to Reduce Calf Stress during Processing in Unweaned Bulls**

Contact Person: Sergio Arispe, Associate Professor, Oregon State University Extension Service-Malheur County, Ontario, OR 97914 Email: Sergio.arispe@oregonstate.edu

**Project Objectives:** The *research objective* is to test the hypothesis that managing cow-calf pairs during branding and processing will reduce the levels of the stress hormone, *cortisol*, which is acutely released at that time. We anticipate that our results will promote a *positive image* within the beef industry—in Oregon and across the west—and have implications directly related to positive *economic returns*.

Project Start Date: The project begins winter 2021 after calving.

Project Completion Date: Fall 2021.

**Project Status and Preliminary Findings:** Introduction: Across the western U.S., cattle processing is a standard procedure performed by cow-calf operators. It is a stressful event that occurs within the first three months of a calf's life when they are earmarked, branded, vaccinated, dehorned, and when bull calves are castrated. Producers use this time to both provide a necessary form of identification and boost overall herd health. When taking into consideration the sheer number of beef cattle that populate the rugged terrains and vast expanses of western rangelands, it is not uncommon for the general public to come across ranchers processing their calves. Because it is commonly in public view, traditional practices associated with calf processing are subjected to increased scrutiny that have the potential to either enhance or damage cattle producers' image through the lens of urban America.

Oregon cow-calf operators skillfully implement practices supporting sustainable operations by implementing practices that are *economically viable*, *socially diligent*, and *environmentally responsible*. Incidentally, many of these practices are consistent with best management practices highlighted by the National Beef Quality Assurance (NBQA) Program, which provides science-based and common sense curriculum to ensure producers are implementing practices that result in a wholesome and quality beef product. Today, the NBQA Program is implemented at the state level, which allows BQA trainers to use science and local understanding to identify best management practices that are consistent with the broader NBQA context. Unfortunately, there is limited science-based information characterizing the best management practice during calf branding and processing. The most common scenario across the western

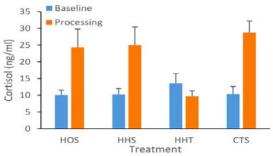
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U.S. is to use horseback to rope and secure calves prior to branding and processing—either with or without the cow. Interestingly, these effects on calf stress have not been formally reported within the scientific literature, which is to say that producer intuition has gone largely unvalidated.

The discipline of *animal welfare* spans negative/bad welfare to positive/good welfare, is tightly associated with health & performance, and is linked with a society's values and moral interpretation (Ohl et al., 2012). At branding and processing, cattle handling practices have the potential to either enhance or inhibit the overall performance of a calf, depending on the level of stress they experience. Previous work highlights castration and branding as acutely stressful times in a calf's life (Schwartzkopf-Genswein et al.,

1997; Tucker et al. 2014), which has implications on economic returns. Unfortunately, little is known about the science of traditional management techniques that cow-calf operators implement branding and processing calves as cow-calf pairs.

The proposed study improves on the limitations of a pilot study we conducted on a southeastern Oregon cow-calf operation in 2018. At that time, we studied four commonly used methods to process calves and highlighted that processing calves as a cow-calf pair reduced level of stress (Fig. 1). Baseline cortisol was obtained a week before processing and the four different processing treatments included—Heel Only Separating Pairs (HOS); Head & Heel Separating Pairs (HHS), Head & Heel Together

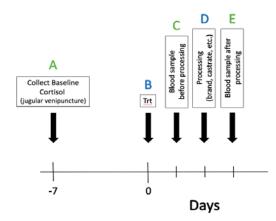


**Figure 1:** Cortisol levels before (baseline) and after branding and processing. Four treatments included: Heel Only Separate (HOS); Head & Heel Separate (HHS), Head & Heel Together (HHT); and Calf Table Separate (CTS).

(HHT); and Calf Table Separating Pairs (CTS). Figure 1 illustrates cortisol levels exhibited by unweaned beef calves before and after branding and processing. Immediately following processing, both the HOS and HHS expressed nearly identical levels of cortisol release—around 24 ng/ml. However, the CTS treatment yielded the greatest levels of cortisol release at 32 ng/ml, whereas the HHT treatment yielded the lowest levels of cortisol release at 9 ng/ml. These preliminary data illustrate that keeping cow-calf pairs together reduces calf stress at branding and processing.

The current study will implement a uniform calf crops and characterize cortisol release before, during, and after branding and processing.

**Methods & Materials:**The proposed experimental site is on the Marchek Ranch near Harper, OR (43°86' N, 117°6' W). It will consist of 50 crossbred, commercial (Angus x Hereford x Charolais), cowcalf pairs consisting of bull calves between 2-3 months old. Calves will be selected for uniformity and randomly assigned to the following treatments—Head & Heel Together (HHT) and Head & Heel Separating Pairs (HHS). Figure 2 illustrates an example of a blood extraction timeline to measure plasma cortisol concentrations. Baseline cortisol will be collected seven days before implementing the treatments after running calves through a chute system. Blood will then be collected before processing, immediately after processing, and then after 30 and 60 minutes.



**Figure 2:** Timeline of blood collection to analyze cortisol: A) 7 days before implementing treatment; B) Treatment (Trt) Separate or maintain cow-calf pairs; C) Sample blood before processing; D) Processing procedure; E) Blood sample after processing immediately, after 30 mins, and after 60 mins.

will be followed to collect blood within this treatment.

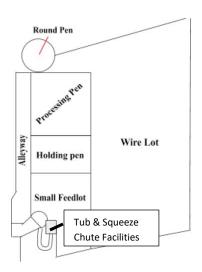
The Marchek Ranch has the infrastructure to conduct the applied research project. It has six fenced areas, including an alleyway leading to a round tub and squeeze chute (Figure 3). Cattle will be held in the Wire Lot area prior to data collection with ample access to feed and water. All designated cow-calf pairs will remain in the pasture during collection of baseline data. Designated handlers will capture each calf utilizing the heel and drag method, and the calf will then be restrained by placing a rope over the front and back feet and holding in place. One blood sample per calf will be collected via jugular venipuncture. This method was selected as it presents the least risk of injury to calves and handlers, and the animals will remain in a familiar environment.

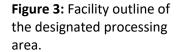
Each blood sample will be transferred to a centrifuge within 30 min after collection at 2,500 x g for 30 min. Plasma will then be separated and frozen at  $-20^{\circ}$ C prior to sending for lab analysis.

On the day of processing, all 50 bull calves will be randomly separated into two groups of 25 and assigned to either the HHT or HHS treatment. The first

set of 25 calves will consist of the HHS group and be restrained utilizing the head and heel roping technique. Processing procedures will include branding, castration, earmarking, subcutaneous injections of Multimin and Vision 8 Somnus, applied in doses of 3 ml and 2 ml respectively, application of Enforce 3 and Once PMH intranasally at 2 ml each, and 36 mg of Ralgro injected under the skin of the ear. Immediately following processing, another blood sample will be collected. The HHS treatment group will then be moved to the middle holding pen in order to keep the two treatment groups separate. Calves will then be roped to secure blood samples 30 mins and 60 mins after initial processing. The second set of 25 calves will consist of the HHT treatment and also be restrained utilizing the head and heel roping technique. Amount of time taken to restrain each calf will be recorded. The same protocol

Coordination will be arranged at least one month prior to conducting the experiment. The Primary Investigator will work closely with Kerry and Audrey Marchek on phone calls to familiarize ropers with the experimental protocol. For consistency purposes, each handler will be given a designated task to perform throughout the duration of both treatments.





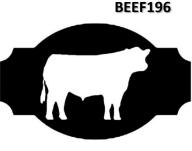
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## Report



**Beef Cattle Sciences** 

## **Progress Reports – Animal Sciences <sup>1</sup>**

## Feeding Spent Hemp Biomass to Lambs as a Model for Cattle

**Contact Person:** Serkan Ates, Assistant Professor, Oregon State University, Corvallis, OR 97331 **Email:** Serkan.ates@oregonstate.edu

**Project Status and Preliminary Findings:** Synopsis: Lamb feed intake and liveweight gains were similar to control diet when they were fed diets containing low (10%) spent hemp biomass. While the feed intake of the lambs that were fed diets containing high spent hemp biomass (20%) was lower than those were fed control diet, their liveweight gains did not differ indicating a greater feed conversion efficiency.

Summary: The objective of this study was to determine the effect on health and meat quality and THC and CBD residuals in finishing lambs fed spent hemp biomass in place of alfalfa as the roughage source. We investigated the effects of feeding level and withdrawal period of spent hemp biomass in place of alfalfa in feedlot-finishing lamb diets. Specifically, we assessed THC and CBD residuals in meat cuts, along with evaluating growth performance, carcass traits, and meat quality. Lambs are chosen as proxy to cattle in this study to reduce the cost of the experiment. Over an 8 week-trial period followed by 3 week transition, our result indicated that lambs the lambs that consumed high (20%) spent hemp biomass (HH groups) in their diets had a lower feed intake, while those receiving low (10%) spent hemp biomass (LH groups) had similar feed intake to control diet that contained no spent hemp biomass. While the liveweight gains of the lambs were similar in period 1, the lambs that were fed high spent hemp biomass (20%) containing diet in period 1 had a greater feed intake and liveweight gain when they were fed control diet in period 2. Carcass characteristics and meat quality related parameters are currently being investigated.

Introduction: The 2018 Farm Bill removed hemp (*Cannabis sativa*) from the Controlled Substances Act, classifying it as an agricultural product. Thirty-eight states in the U.S. are in the process of implementing a program for regulating industrial hemp, allowing its cultivation. This has led to a flourishing industry. Oregon is among the leading producers of hemp to produce cannabidiol (CBD), a process that generates a highly-nutritive spent byproduct that could be fed to livestock. Byproducts of hemp are not yet FDA approved for use in animals for food production. Studies conducted in Europe have revealed that hempseed cake is safe for inclusion in livestock feed (EFSA, 2011). However, there are no studies that have investigated feeding spent hemp biomass (i.e., hemp biomass after CBD has been extracted) to livestock, specifically to ruminant animals. The most critical aspect of feeding any hemp byproducts is the potential for cannabinoid residuals in milk or meat, particularly the psychoactive tetrahydrocannabinol (THC). Data from Europe have indicated the presence of unacceptable levels of

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THC in animals fed fresh hemp, but acceptable levels when using spent byproducts such as hempseed cake. Therefore, there is a critical need to assess the levels of THC residuals in milk and meat to obtain FDA approval to feed hemp byproducts to livestock.

Our long-term goal is to implement the safe use of hemp byproducts in livestock diets and take full advantage of their nutritional and potential medicinal properties to improve animal health and the quality of animal products. The objective of the present proposal is to assess the presence of THC and CBD residuals in meat of ruminant animals fed diets that contain spent hemp biomass. These data are essential for obtaining FDA approval for use of hemp byproducts in ruminant diets. This proposal is predicated upon the fact that hemp is a rapidly growing industry that can provide economically viable and potentially health-improving byproducts to feed ruminants. However, data to obtain FDA approval for hemp use are not available in the U.S. Therefore, there is a critical need to assess the presence of cannabinoid residuals in milk and meat in ruminants fed hemp byproducts and their effect on quality of the animal products and animal health and performance. Our rationale is that data on THC and CBD residuals in animal byproducts and safety are essential for use as livestock feed and this research is needed for the process of obtaining FDA approval for the feeding of hemp byproducts to ruminants. The expanding hemp industry is of very high interest to Oregon and other U.S. livestock producers due to the possibility of obtaining cost-effective, high-quality feed supplements with potential health benefits for animals.

Materials and Methods: A pen feeding trial was conducted using 35 weaned Polypay lambs (44.7kg and 5-6 month-old) for a period of 8 weeks following a 3-week adaptation period. Lambs were individually housed in pens equipped with feeders and water buckets where they were fed corn-barley-soy based finishing diets with 70/30 concentrate/roughage ratio (as fed). Lambs were stratified by weight and assigned randomly to 5 treatments with 7 lambs per treatment. The experiment consisted of two fourweek periods from August 7 to October 1. Spent hemp biomass replaced alfalfa in diets as follows: control diet; 70% concentrate-20% alfalfa (+10% grass hay) with no hemp inclusion (control; CON); 2) low hemp diet in period 1: 70% concentrate plus 10% alfalfa and 10% spent hemp biomass (+10% grass hay) during only period 1 (28 d) and 20% alfalfa in period 2 (+10% grass hay) (LH-1); 3) low spent hemp biomass in both periods: 70% concentrate plus 10% alfalfa and 10% spent hemp biomass (+10 grass hay) during period 1 (28 d) and period 2 (28 d) (LH-2), 4) high spent hemp biomass in period 1: 70% concentrate plus 20% spent hemp biomass (+10 grass hay) during only period 1 (28 d) and 20% alfalfa (+10 grass hay) in period 2 (HH-1); and 5) high spent hemp biomass in both periods: 70% concentrate plus 20% spent hemp biomass (+10 grass hay) during period 1 (28 d) and period 2 (28 d) (HH-2). All diet ingredients were ground with a hammer mill and mixed to prevent feed selection. However, chopped grass hay (10% as fed) was offered to lambs separately to avoid possible acidosis problems. The diets were balanced for energy and protein and formulated to contain 12.1 g kg<sup>-1</sup> CP (DM basis). Each animal was offered a trace mineral-salt mixture.

Lamb LWG and dry matter intake (DMI) were monitored. Lamb weights were recorded at the beginning of the trial and at 28-day intervals thereafter, following a 12 h fast. Feed samples and unconsumed feed from each group were taken three times each week for DM and nutrient intake determination. All lambs were fed twice daily and the unconsumed feed was collected and weighed prior to morning feeding to determine feed consumption per lamb. The unconsumed feed represented 15-20% of feed offered to lambs.

At the end of the feeding experiment, all lambs were slaughtered at the OSU Clark Meat Lab to study the carcass characteristics and meat quality. Carcass characteristics (hot and cold carcass weight, dressing percentage, offal parts, and dissected components of the left carcass) of the animals were determined. Meat quality parameters (tenderness, cooking loss, pH, color, shelf life etc.) and fatty acid profile are currently being determined.

Results: Our preliminary results indicated that the lambs receiving high (20%) spent hemp biomass (HH groups) in the diet had a significant reduction of feed intake, while those receiving low (10%) spent hemp biomass (LH groups) had similar feed intake to control diet that contained no spent hemp biomass (Figure 2). However, the decrease in feed intake did not affect the average daily gain. The animals receiving rations containing high (20%) spent hemp only in Period 1 (HH1 group) had a large increase in their feed intake in Period 2 when they were fed control diet, resulting in the group with the highest feed intake during this period. The animals receiving low (10%) spent hemp biomass during both periods (group LH2) had a greater feed intake compared to control or the LH1 group in Period 2. The average daily gain tended to be higher in animals that received spent hemp biomass in Period 1 but received the control diet in Period 2. In animals receiving high (20%) spent hemp biomass during only Period 1 (HH1), feeding spent hemp biomass tended to make the animals more efficient in Period 2 when they had control diets. No other differences in performance were observed. Besides not being detrimental to the animals, feeding spent hemp biomass resulted in increased feed intake in the long run, as demonstrated by the feed intake being larger compared to control group in animals receiving 10% spent hemp biomass to feed ruminants, but also are indicative of an effect of the spent hemp biomass on the appetite of the animals. The higher feed intake induced by the spent hemp biomass could be due to an effect on the rumen or could be a systemic effect.

Conclusions: Our results indicated that spent hemp biomass can successfully be included in ruminant diets without causing any detrimental effect to animal performance and health. However, it is of note that high terpene content of spent hemp biomass should be eliminated before feeding to the livestock to improve the 'palatability' of the feed. The meat quality related parameters will be presented in the final report.

Acknowledgements: This research study was financially supported by the Oregon Beef Council.

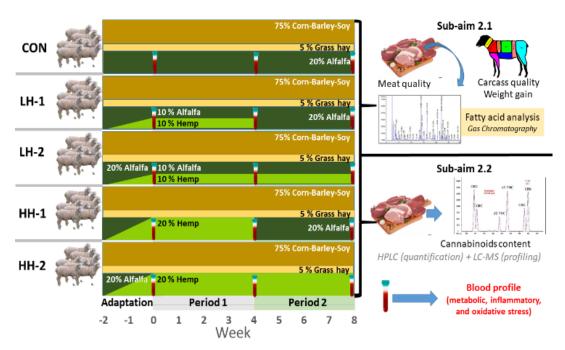


Figure 1. Overview of the experimental design and measurements

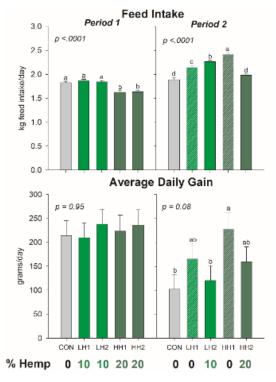
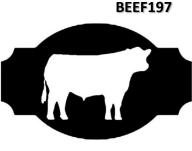


Figure 2. Feed intake (kg) and average daily gain (grams/day) from the lamb experiment. CON = lambs receiving no hemp; LH1 = lambs receiving 10% spent hemp biomass during Period 1 only; LH2 = lambs receiving 10% spent hemp biomass during Periods 1 and 2; HH1 = lambs receiving 20% spent hemp biomass during Period 1 only; HH2 = lambs receiving 10% spent hemp biomass during Periods 1 and 2. See Figure 1 for details. The overall effect is indicated by the p-values reported in the graph (significant when equal or less than 0.05 and a tendency when P equal or less than 0.10) and different letters denote statistical difference between groups.



## Report



**Beef Cattle Sciences** 

## Progress Reports – Animal Sciences<sup>1</sup>

## Self-Regenerating Annual Clovers in Western Oregon Forage Systems

**Contact Person:** Serkan Ates, Assistant Professor, Oregon State University, Corvallis, OR 97331 **Email:** Serkan.ates@oregonstate.edu

**Project Status and Preliminary Findings:** Synopsis: Self-regenerating annual legumes in particular, balansa clover increased the legume content of the pastures by 25.3% in early spring, providing an excellent quality of forage for grazing or silage. The higher legume content in spring led to greater heifer liveweight gains.

Summary: The objective of this experiment was to evaluate the effects of self-regenerating annual legumes in pasture and animal production in dairy production systems in western Oregon. The perennial pastures containing either only perennial species or mix of perennial and self-regenerating annual legumes were sown on October 2019. Following the harvest of the forages for silage in early spring, pastures were rotationally grazed from May 28<sup>th</sup> to October 29<sup>th</sup>. Our first year results indicated that balansa clover has a high potential to increase legume content of establishing pastures in early spring with its rapid growth rates. Higher legume content of pastures also led to greater heifer liveweight gains by 110 g/head/day during spring period. The persistence of balansa clover and subterranean clover will be monitored in the second year of the experiment. Pastures will be grazed from early April to November in 2021 and heifer liveweight chances will be recorded.

Introduction: Pastures that contain annual legumes are more productive in spring and persistent in summer dry areas owing to lower temperature requirements and drought avoidance strategy of annual legumes. These legumes avoid dry conditions by dying in early summer after seed setting and have an added advantage of growing earlier in spring so providing high quality feed in early lactation. For example, when drilled into grass-dominated pastures, subterranean clover increased pasture yield as much as 40% in New Zealand (Ates *et al.*, 2010). Balansa clover, not as widely tested as subterranean clover, can outperform subterranean clover in heavy clay soils of western Oregon due to its high tolerance of poor drainage. Potential of balansa clover to support high animal production was highlighted in a sheep grazing study at OSU in spring 2018. Lambs that grazed tall fescue-balansa clover or birdsfoot trefoil. The greater liveweight gain obtained from balansa clover was mainly due to higher clover content of tall fescue-balansa clover pastures (over 35% vs. <20%) in late spring period (Gultekin et al., 2020).

Self-regenerating annual clovers need to produce adequate amount of seeds to persist in a permanent pasture. The production and persistence of annual clovers are mainly dependent on rainfall, grazing management and flowering time (maturity) of the varieties. Early flowering cultivars exploit winter

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rainfall and ensure high quality forage and seed production early in spring. Later flowering annual clover cultivars always produce greater herbage production, if soil moisture is present. Current subterranean clover seeds available in the market are predominantly earliest flowering varieties (suitable for areas with <15 inch rainfall). However, the amount and seasonal distribution of rainfall in Western Oregon would permit successfully growing mid-late flowering subterranean clover varieties and therefore producing greater amount of high quality forage. Within annual clover pastures, sowing cultivars together in mixtures that differ in flowering time may be useful in exploiting and coping with variable spring rainfall. Therefore, in this study, we have been investigating the total and seasonal pasture productivity, nutritive quality and persistence of annual legumes that belong to different groups of maturity in irrigated and rainfed pastures. The overall objective is to explore the potential of annual legumes in permanent pastures and develop sustainable management practices for higher productivity and persistence. Specific objectives of the study are:

- Determine the forage and livestock production from pastures containing annual legumes with different maturities.
- Assess the production and persistence of self-regenerating annual legume varieties and perennial pasture species in irrigated and rainfed conditions.

Materials and Methods: Site, pasture establishment and measurements: The study has been carried at the Oregon State University Dairy Farm in Corvallis, Oregon (44° 34' N, 123° 18' W 78 m. a.sl.). Multispecies pasture mixtures either containing the combination of perennial species and self-regenerating annual legumes or only perennial pastures species (perennial ryegrass, orchard grass, chicory and white clover) were established in a 2.1-ha plot on October 11 in 2019. Prior to establishment, pasture paddocks were divided into three blocks to serve as replicates for the experiment. Each block was divided into 3 subplots, which were randomly allocated to a combination of (1) perennial species + self-regenerating annual clover mixtures (2) or only perennial pastures without any annual clovers, giving a total 6 plots. Pastures with annual clovers were further divided into three subplot and planted with either early maturing annual clovers, later maturing annual clovers or mix of early-late maturing annual clovers.

Grazing trial: Prior to start of the grazing, pastures were harvested for silage in May. Then they were grazed with dairy heifers from May 28<sup>th</sup> to October 29<sup>th</sup>. Dairy heifers were offered a dietary treatment of (1) perennial pastures overdrilled with annual clovers or (2) perennial pastures only (with no annual clovers). Twenty-four multiparous Jersey heifers were blocked for age and liveweight and randomly assigned to the two dietary treatments. Pastures overdrilled with three groups of annual legumes (early, mid and mix) were grazed commonly, as one pasture at the same time. Each group of 3-4 heifers were randomly assigned to one of 6, 0.35-ha pasture paddocks where they rotationally grazed within the same pasture at the stocking rate of 8.6-11.4 heifers/ha. Rotation length was adjusted based on the seasonal pasture growth and leaf number counts of the dominant grasses. Each treatment had a core group of 3 heifers (testers) with spare heifers (regulators) to be used in a put-and-take grazing system to match feed demand with changing supply. Heifers were offered an estimated pasture allocation of 8 kg of DM/heifer per day above a post-grazing pasture residual of 1200-1400 kg of DM/ha. Plastic water troughs were moved into the new grazed area to allow ad libitum access to water as heifers were offered new pastures.

Pasture dry matter production and botanical composition: Dry matter production (kg/ha) and herbage growth rates (kg/ha per day) of each pasture combination were measured at each grazing cycle (or cutting for silage) before the animals turned onto pasture plots during active growth in spring, summer and autumn. For the grazed pastures, herbage growth were measured inside 1-m<sup>2</sup> grazing exclosure cages. Herbage growth was measured from a 0.25 m<sup>2</sup> quadrat by cutting to a stubble height of approximately 5 cm. Exclosure cages were placed over a new representative area pre–trimmed to 3 cm stubble height at the start of each new growth period. After cutting, cages were relocated to new pre–trimmed sites in each pasture treatment. All herbage from the quadrat cuts will be dried in an oven (65 °C) until constant weight. Quadrat cuts will be sub–sampled for sorting into botanical fractions (grass, legume, herb, weed and dead material) before they are dried.

Liveweight gains of heifers: Liveweight gain of the heifers were determined prior to and following each grazing period (28-36 day intervals). All animals were weighed "empty" after a 12-h withdrawal from feed.

Results: Although in spring (April and May), pastures with annual clovers provided 249-256 kg DM/ha greater forage production, the total annual forage production of pastures did not differ. Overall pastures sown with and without annual clovers had similar total annual forage production (11.3 vs. 11.4 t DM/ha). In April 2020, pastures containing self-regenerating annual legumes had 31.3% legume content. In contrast, perennial pastures planted without any annual legumes contained only 5.0% legume. However, perennial pastures were not sown with annual legumes had greater legume and chicory contents during summer. In September 2020, both pasture types had similar legume content (31.0-33.4%). It is possible that faster growth rates of annual legumes, in particular balansa clover in early spring might have suppressed the growth of white clover and chicory, reducing their contents in the pastures during summer. Together with the senescence of annual legumes, this caused a reduced legume content in the pastures sown with annual legumes. However, it appears that white clover recovered this early suppression later in the season once the pressure of annual legumes disappeared.

The positive effect of the higher legume content of the pastures containing annual legumes were reflected from the heifer liveweight gains. Heifer grazing pastures with annual legumes gained 100 g head/d more than those grazing pastures with only perennial pasture species (420 g/head/d vs. 530 g/head/d) during May-June period. See Figure 1.

Conclusion: In conclusion, balansa clover has a great potential in increasing the legume contents and pasture quality in early spring with its rapid growth rates due to its lower temperature requirement than perennial legumes. Although the pasture containing high balansa clover content was not grazed (but ensiled) in early spring, it is highly probably that the higher legume content by 25.3% would have led to greater animal performance as well in early spring. The importance of high legumes for animal performance was still apparent in May-June period when heifers gained 110 g/head/d more liveweight on pastures containing self-regenerating annual legumes.

Acknowledgments: This research study was financially supported by the Oregon Beef Council Literature Cited: Ates, S., Tongel, M. O., and Moot, D. J. (2010). Annual herbage production increased 40% when subterranean clover was over-drilled into grass-dominant dryland pastures.



Pastures with (31.3% clover) and without<br/>annual legumes (5%) on 10 April 2020.Heifers grazing pastures on 7 June.

Figure 1. Pasture photos

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## Report



**Beef Cattle Sciences** 

## **Progress Reports – Animal Sciences <sup>1</sup>**

## In Vivo-In Vitro Dose-Effect Response of Bovine Liver to Rumen-Protected Fatty Acids: Implementation of a Nutrigenomic Approach in Dairy Cows

**Contact Person:** Massimo Bionaz, Oregon State University, Corvallis, OR 97331 **Email:** Massimo.bionaz@oregonstate.edu

**Project Objectives:** The objective of the present proposal is to determine the dose of dietary rumenprotected fatty acid mixture with the maximal activation of PPAR.

Project Start Date: September 2019

Project Completion Date: December 2020

**Project Status and Preliminary Findings:** We improved the experimental design to best accomplish the objective (**Figure 1**). The modifications included the addition of a treatment group by feeding cows a commercially available fatty acid mixture to use as a positive control. Based on IACUC recommendations, we decided to use more cows (30 cows) and have 5 groups. According to IACUC biopsies in cows can be repeated only each 20 days. Thus, we had to change the feeding time to 20 days .We have also run additional *in vitro* experiments to test the mixture of fatty acids in cells cultivated in blood serum, so to better mimic the *in vivo* situation. Data will be presented in the final report. We originally planned to perform the experiment in spring 2020; however, the lockdown of COVID precluded us to start the experiment. For this reason we are now organizing the experiment to start in mid-November.

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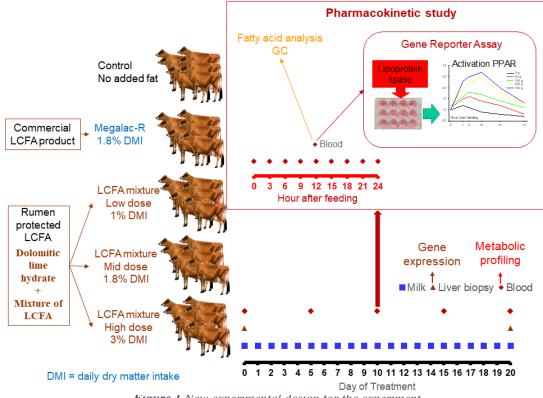


Figure 1 New experimental design for the experiment



## Report



**Beef Cattle Sciences** 

## Progress Reports – Animal Sciences<sup>1</sup>

## Using GPS-Activated Shock Collars to Prevent Cattle Grazing of Burned Rangeland

**Contact Person:** Juliana Ranches, Assistant Professor, Eastern Oregon Agricultural Research Center, Burns, OR 97720 **Email:** Juliana.ranches@oregonstate.edu

**Project Objectives:** The main objective of this study was to determine the efficacy of GPS-activated shock collars (virtual fence) for excluding cattle from burned sagebrush steppe. A second objective of this study was to evaluate cattle behavior, when using GPS-activated shock collars.

Project Start Date: May 2020

Project Completion Date: May 2021

**Project Status:** Both field portions of the study (grazing and behavior) were completed over summer 2020. Data from both portions of the study are currently being summarized and analyzed.

#### **Preliminary results:**

**Grazing:** Preliminary results are very encouraging, with virtual fencing effectively keeping cattle out of the burned areas compared to cattle without virtual fencing. More importantly, forage utilization of the burned areas protected from cattle grazing by the virtual fence technology was negligible to none compared with heavy utilization (almost 70%) without the virtual fence. This preliminary data shows that virtual fence technology holds tremendous potential as a land and livestock management tool. **Fence-line contrast description:** Fence-line contrast photo of the burned area after grazing. Control treatment (cattle not restricted access) on left and virtual fence treatment (cattle restricted access) on right.

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BEEF200

## **Report** Developing Conservation Measures to Restore and Rehabilitate Rangelands on Degraded Sage-Grouse Habitat in Southeastern Oregon <sup>1</sup>

Sergio Arispe<sup>2</sup>, Kirk Davies<sup>3</sup>, and Dustin Johnson<sup>4</sup>

## Synopsis

Implementation and evaluation of broad management tools to improve degraded sagebrush rangelands within greater sage-grouse habitat.

### Summary

The research objective was to evaluate the effectiveness of a variety of sagebrush and inasive annual grass (IAG) treatments, which will be followed up with native or introduced perennial grass seedings. During fall of 2016 and 2017, five treatments were implemented on four cow-calf operations in southeastern Oregon. Treatments included a modified rangeland drill (MD), spring 2016 disking (D), fall 2016 prescribed burn with imazapic and glyphosate herbicide (B+IG), fall 2017 prescribed burn (B), fall 2016 imazapic and glyphosate application (IG), and a control plot. Each of the whole plots, save the control, were seeded with native and introduced perennial bunchgrasses while herbaceous cover and density were assessed 1.5 years after seeding. The B+IG treatment appeared to be most effective at establishing perennial bunchgrasses and mitigating annual grass cover and density. The IG treatment also seemed to suppress annual grass density and cover but was not as effective at perennial bunchgrass establishment. The B and D treatments appeared to increase total

herbaceous density with annual grasses contributing to the greatest proportion of the density. In short, mechanical action may further degrade these sagebrush steppe plant communities while IG or B+IG promote opportunities to establish deep rooted perennial bunchgrasses.

#### Introduction

Greater sage-grouse (sage-grouse) are a sagebrush obligate that has experienced a decline in population across 11 western states since the 1960s. In March 2010, the United States Fish & Wildlife Service (USFWS) identified sage-grouse as a candidate species citing primary range-wide threats such as habitat loss, fragmentation, degradation, and inadequate regulatory mechanisms. Despite the federal decision not to list sage-grouse in 2015, a future listing could negatively impact rangelandbased cattle operations that contribute to Oregon's leading agricultural commodity.

In anticipation of a final listing by the USFWS, six Soil & Water Conservation Districts (SWCDs) assembled Candidate Conservation Agreements with Assurances (CCAAs) allowing landowners to voluntarily work with them to develop a site specific plan (SSP) whereby private landowners commit to address threats to sage-grouse on enrolled lands by implementing Conservation measures (CMs) actions that improve or maintain sage-grouse habitat on lands to be enrolled (Harney Soil and Water

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<sup>3.</sup> Lead Rangeland Research Scientist, US Dept. of Ag – Ag Research Service (USDA-ARS), Burns, OR 97720.

<sup>4.</sup> Rangeland Outreach Specialist, Oregon State University, Eastern Oregon Ag Research Center, Burns, OR 97720.

Conservation District, 2014). Private landowners entering into CCAAs will receive assurances against additional regulatory requirements if sage-grouse are listed under the Endangered Species Act—as long as they follow through with their commitments in the SSP. The CCAAs have the potential to provide assurances to private landowners on approximately 3.5 million acres of sage-grouse habitat across eight Oregon counties.

One of the primary threats to sage-grouse is invasive annual grass (IAG) invasion and dominance of sagebrush rangelands, which can devastate and alter a functional sagebrush steppe ecosystem with widespread negative consequences. Currently, there are at least 70 million acres of IAGs in the Intermountain West. These grasses are highly competitive and can exclude preferred plant species leading to decreases in biodiversity and forage production. They can also germinate in the fall and establish root growth at low winter temperatures giving them a competitive advantage over the seedlings of desirable perennial grasses. During spring, they develop an extensive root system that depletes soil moisture. In degraded low elevation sagebrush rangelands where IAGs are the primary sagebrush understory, cheatgrass and medusahead create fine fuels on the landscape that increase the frequency of fire to the detriment of native vegetation. We developed the current proposal with rangeland-based beef cattle producers in Harney and Malheur counties because producers in both counties have experienced wildfires that have burned over two million acres in the last seven years. These wildfires disturbed rangelands, diminishing wildlife habitat, and limited cattle production.

Our experiment evaluates the effectiveness of a variety of sagebrush and IAG treatments, which will be followed up with native or introduced seedings of perennial gasses. Our hypothesis is that fire, herbicide, a fire-herbicide combination, and mechanical action treatments will promote reduce IAG cover and density, where introduced seeding promote perennial bunchgrass compared to a native seeding. The objective of the current study is to examine the potential to develop new CMs for CCAA participants.

#### **Materials and Methods**

#### Study Area

There are four study sites in southeastern Oregon with three study sites located in Malheur County and one study site in Harney County.

Within Malheur County, one experimental site is located 3 miles east of Crowley, OR and 60 miles southeast of Burns, OR. The two additional sites are located 100 miles southeast and 72 miles northeast of Burns, OR, respectively. The experimental site in Harney County is located 145 miles south of Burns, OR. Study site elevations range from 2,700 to 4400 feet above sea level. Long-term (1981-2010) average annual precipitation was between 10.3 inches and 11.5 inches (PRISM Climate Group, 2014). The rangeland ecological sites for our study was predominately Loamy 10-12 PZ (Natural Resource Conservation Service 2015). Vegetation on this rangeland ecological site was traditionally Wyoming big sagebrush, bluebunch wheatgrass, and Thurber's needlegrass. Currently, all four study sites are classified as *Ecological State C* sites within low elevation sagebrush rangelands according to the state-and-transition models within the Harney and Malheur County CCAAs.

#### Treatments/Design

The experimental design used to compare treatments was a randomized complete block design with four sites (blocks). Mechanical, chemical, and prescribed burn treatments before seeding perennial bunchgrasses included, 1) 2016 fall prescribed burn plus 8 oz/acre imazapic with 12 oz/acre glyphosate (B+IG), 2) fall 8 oz/acre imazapic with 12 oz/acre glyphosate (IG), 3) Spring 2017 disking (D), 4) 2017 fall burn (B), 5) 2017 modified rangeland drill (MD), and 6) control (C; Not Seeded). The treatments were applied to 30 x 11 m plots that included a 5-m buffer between the different treatments. In 2016, invasive annual grasses were controlled using prescribed burning, which occurred in mid-October. The relative humidity and wind speed ranged between 25-55% and 0-10 miles  $\cdot$  hr<sup>-1</sup>, respectively. Furthermore, air temperature fluctuated between 50-61°F. The burns removed the invasive annual grass litter and leaves were burned off the sagebrush. An IG herbicide treatment was applied the week after the prescribed burn using a pressurized backpack sprayer with EUS 02 nozzles at 35 psi releasing 20 gallons  $\cdot$  acre<sup>-1</sup> of spray solution using a metronome. The prescribed burn treatment consisted of using strip-head fires ignited with drip torches. In fall 2017, five treatment plots at the four experimental sites were split lengthwise and followed up with native or introduced seedings of perennial grasses. Native species included bluebunch wheatgrass, and bottlebrush squirreltail. Introduced species included desert and Siberian

wheatgrass. All plots were drill-seeded in October 2017 with 12 pounds of perennial grass pure live seed per acre.

Vegetative cover and density were measured and compared across all plots. Data are presented as averages with their respective variation.

#### Results

Cover

Cover appears to vary by treatment whereas it is not clear the effect of seeding on vegetative response. Annual grass cover responded similarly to treatments in both years (Fig. 1A). The B+IG and IG treatments reduced annual grass cover, whereas the disking and burn treatments seem to increase the response. Total perennial herbaceous cover exhibits a similar trend across years (Fig. 1B). The MD and D treatments exhibited the least cover while other treatments appear similar to the control. Total herbaceous cover generally highlights that the B treatment produced the highest percent cover, whereas D produced higher cover during 2019 and a lesser response in 2020 (Fig. 1C). Bare ground was greatly influenced by B+IG across both years (Fig. 1D). Rock cover varied across treatments with no clear trend between years (Fig. 1E). Finally, litter displayed similar responses across years. The B+IG treatment exhibited the lowest litter cover (Fig. 1F).

#### Density/Diversity

Large perennial bunchgrass density appears to be greatest in the B+IG treatment and this was consistent across year (Fig. 2A). Annual grass suppression appears to be greatest with both the B+IG and IG treatments (Fig. 2B). In 2020, annual grass tends to increase in the D treatment and the B treatment with native seeding. Annual forb density was clearly attenuated by the B+IG and IG treatments for both years. It also seems that annual forb density increases for both D and B treatments from 2019 to 2020 (Fig. 2C). Total herbaceous density reflects similar trends with the B+IG and IG treatments suppressing density (Fig. 2D). Total perennial herbaceous density highlights that B+IG promotes the greatest response (Fig. 2E).

#### Conclusions

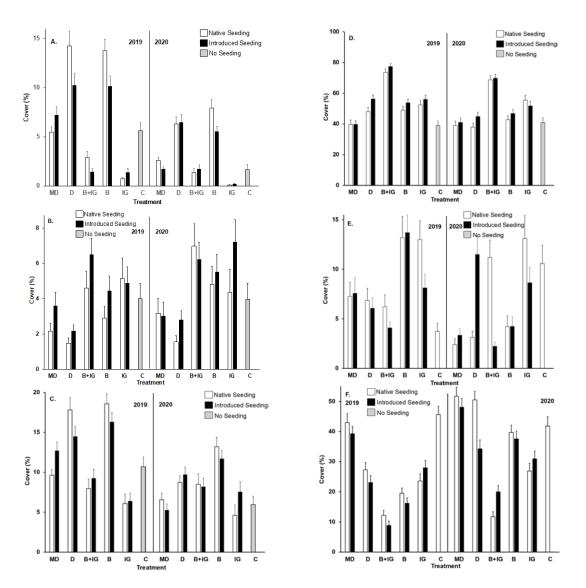
In conclusion, results from this experiment suggest that large perennial bunchgrasses are able to establish after treated with B+IG. The burn destroys the sagebrush canopy and thus reduces sage-grouse habitat. The IG treatment is just as effective at reducing annual grass density. Unfortunately, it does not promote perennial bunchgrass density. Disturbing the plant community with either mechanical action—MD or D—or a prescribed burn did not appear to influence annual grass density in 2019. However, the D and B treatments exhibited augmented responses in 2020 highlighting that disturbance may promote annual grass density over time. These data suggest that restoring and rehabilitating a degraded sagebrush where invasive annual grasses are the predominant plant functional group in the interspace is a long-term process that may be enhanced with either B+IG or IG treatments.

#### Acknowledgments

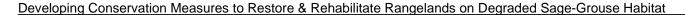
This research study was financially supported by the Oregon Beef Council and cow-calf operators in Malheur and Harney counties, including the Stacey Davies, Bob Skinner, Steve Russell, and Bill Romans. Furthermore, the work would not have been possible without funding for student technical support. Finally, the work would not have been possible without the help from rangeland technicians out of the Eastern Oregon Agricultural Research Center-Burns.

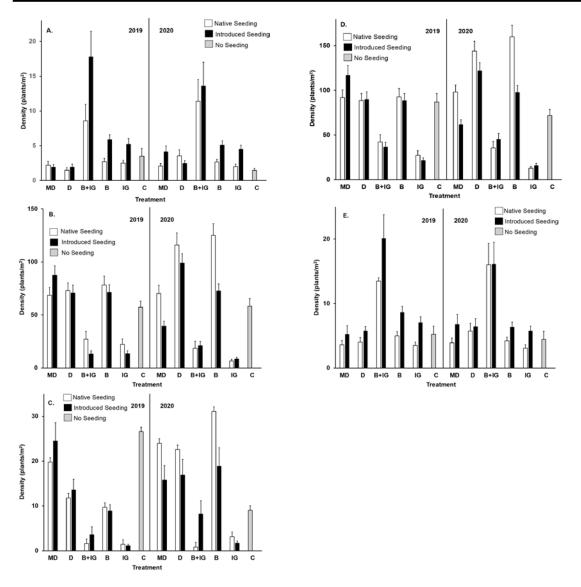
#### Literature Cited

Harney Soil and Water District. 2014. Greater sage-grouse programmatic candidate conservation agreement with assurances for private landowners in Harney County.



**Figure 1.** Cover % relative to **A**, Annual grass, **B**, total perennial herbaceous, **C**, total herbaceous, **D**, bare ground, **E**, rock, **F**, litter (mean  $\pm$  SE). Treatments are MD = modified rangeland drill, D = disking, B+IG = fall 2016 burn + imazapic + glyphosate, IG = 2016 fall imazapic + glyphosate. Seeding treatments included desert & Siberian wheatgrass with forage kochia (introduced) and bluebunch wheatgrass, bottlebrush squirreltail, and Wyoming big sagebrush (native). The control (C) was not seeded.





**Figure 2.** Density with **A**, large perennial bunchgrass, **B**, annual grass, **C**, annual forbs, **D**, total herbaceous, **E**, total perennial herbaceous (mean  $\pm$  SE). Treatments are MD = modified rangeland drill, D = disking, B+IG = fall 2016 burn + imazapic + glyphosate, IG = 2016 fall imazapic + glyphosate. Seeding treatments included desert & Siberian wheatgrass with forage kochia (introduced) and bluebunch wheatgrass, bottlebrush squirreltail, and Wyoming big sagebrush (native). The control (C) was not seeded.

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Report



BEEF201

**Beef Cattle Sciences** 

# Grazing Season of Use Effects on Greater Sage-Grouse Habitat <sup>1</sup>

Vanessa Schroeder<sup>2</sup>, Dustin Johnson<sup>3</sup>, David Bohnert<sup>3</sup>, Jonathon Dinkins<sup>4</sup>, Chad Boyd<sup>5</sup>, Travis Miller<sup>6</sup>, and Kirk Davies<sup>5</sup>

#### Synopsis

Moderate levels of rotational and dormant season grazing did not lead to meaningful short-term differences in major vegetation or wildlife habitat variables. These results support our initial hypotheses that short-term moderate rotational grazing would not drastically change plant communities or wildlife habitat, however this experimental design with controls and pre-treatment data fills a considerable knowledge gap in the literature that provides critical information to support decision making surrounding grazing management.

#### Summary

Currently there is no published literature from a rigorous comparison of direct or indirect effects of contemporary, extensively used grazing practices on sage-grouse habitat. We tested the effects of timing of grazing on rangeland health metrics important to nesting grouse, applying three different grazing regimes (no grazing, rotational spring-defer grazing and dormant grazing) at moderate intensity at three different sites located at the Northern Great Basin Experimental Range (NGBER) in southeast Oregon.

After three years of treatment we did not find any meaningful differences in major vegetation or wildlife habitat variables associated with moderate levels of rotational and dormant season grazing. While we did observe a decline in herbaceous cover (Figure 2), a metric thought to be important to nesting grouse, it was not in conjunction with a reduction in visible obstruction at heights pertinent to nesting sage-grouse. Our replicated, controlled experiment with pre-treatment data, is a critical supplement to observational studies that comprise the majority of the existing literature, as our design allows us to attribute observed changes directly to the grazing treatment application, something that is not possible with observational landscape scale studies lacking controls or pre-treatment data.

#### Introduction

The sagebrush ecosystem is currently under threat from landscape scale processes including invasion by annual grasses, an unprecedented rise in wildfire, and encroachment by conifers (Connelly et al., 2004). Greater sage-grouse (*Centrocercus urophasianus*; hereafter sage-grouse) have experienced significant population declines during the past several decades. Sage-grouse currently occupy approximately 56% of their historic (pre-1800) distribution. The primary threats to sage-

<sup>1.</sup> This document is part of the Oregon State University – 2020 Oregon Beef Council Report. Please visit the Beef Cattle Sciences website at <a href="http://blogs.oregonstate.edu/beefcattle/research-reports/">http://blogs.oregonstate.edu/beefcattle/research-reports/</a>

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grouse are habitat based with significant loss and fragmentation of habitat contributing to population declines (Connelly et al., 2004). Sage-grouse are dependent upon the sagebrush ecosystem, requiring adequate levels of sagebrush and herbaceous cover for nesting sites (Connelly et al., 2000; Knick et al., 2003).

Landscape scale threats impacting the sagebrush ecosystem, coupled with documented population declines have resulted in multiple petitions for the United States Fish and Wildlife Service (USFWS) to list the sage-grouse as threatened or endangered under the Endangered Species Act. The USFWS ruled on the petition during the fall of 2015, reaching a decision of not warranted, primarily due to the large number of conservation efforts arising throughout the west to bolster and conserve habitat for the species. Leading up to the 2015 decision, the BLM amended Resource Management Plans in ways that could potentially require changes in current grazing on some BLM allotments in an effort to preserve adequate nesting cover for sage-grouse in remaining intact habitat. In addition, while agencies are increasingly directing more research dollars towards restoration of degraded rangelands in an attempt to stem the tide of habitat loss and fragmentation, sustaining existing habitat through effective management is of equal or greater importance. In the face of multifaceted, landscape scale threats to the sagebrush ecosystem, much of the existing habitat, particularly in Oregon, remains relatively intact. Thus, research that improves our understanding of management strategies that contribute to maintaining the productivity and resiliency of intact sagebrush rangeland will be vital for sustaining both healthy wildlife populations, as well as the economic viability of ranching in a significant portion of Oregon.

Grazing of cattle in the sagebrush ecosystem can affect sagebrush and bunchgrass cover and density, thereby impacting available cover for nesting grouse. The intensity and timing of grazing determines the nature of this impact (Beck & Mitchell, 2000). While there is literature available assessing the impact of grazing on grouse habitat, most studies examine the impact of growing season grazing on the plant community (Beck & Mitchell, 2000) or examine the interactive effects of grazing timing and intensity with fire (Davies et al., 2016). Currently there is no published literature from a rigorous comparison of direct or indirect effects of current, extensively used grazing practices on sagegrouse habitat. It is imperative to better understand how different grazing seasons of use affect various aspects of grouse habitat including plant community composition, recruitment and structure, in order to help determine what land managers need to consider when implementing management in the sagebrush ecosystem.

The purpose of this study was to use cattle to test the effects of timing of grazing on rangeland health. Specifically, we evaluated three different grazing regimes (no grazing, rotational spring-defer grazing and dormant grazing) at moderate intensity at three different sites located at the Northern Great Basin Experimental Range (NGBER) in southeast Oregon, to evaluate the impact on rangeland health metrics important to nesting grouse.

#### **Materials and Methods**

Study Area

We conducted our study at the Northern Great Basin Experimental Range (NGBER; lat 43°29'N, long 119°43'W) 50 – 60 km west of Burns, Oregon, from 2017 – 2019. Elevation of the study area ranged from 1,300 – 1,500 meters. The study area normally experiences wet, cool winters followed by hot, dry summers with a long-term (1938 – 2020) crop year (September 1 – August 30th) average precipitation of 278 mm (standard deviation = 82.8 mm) NOAA station (RILEY 10 WSW, OR US). Crop year precipitation amounts during the study were 113%, 74%, 124% and 79% of the long-term average for 2017, 2018, 2019 and 2020, respectively.

Composition of plant communities comprising the study area were representative of those in the northern Great Basin (Davies et al., 2006; Davies & Bates, 2010), and exhibited minimal invasion by exotic annual grasses. Wyoming big sagebrush (Artemisia tridentata ssp. wyomingensis [Beetle and A. Young] S. L. Welsh) and green rabbitbrush (Chrysothamnus viscidiflorus [Hook.] Nutt.) were the dominant shrubs. Dominant perennial bunchgrasses varied by study site (block) but included bluebunch wheatgrass (Pseudoroegneria spicata [Pursh] A. Löve), Thurber's needlegrass (Achnatherum thurberianum [Piper] Barkworth), bottlebrush squirreltail (Elymus elymoides [Raf.] Swezey), Idaho fescue (Festuca idahoensis Elmer), prairie junegrass (Koeleria macrantha [Ledeb.] J. A. Schultes), and needle and thread (Hesperostipa comata [Trin. and Rupr.] Barkworth) as common species.

#### Treatments/Design

We used a randomized complete block design with three blocks to determine the effects of three grazing treatments on sagebrush-obligate songbird nest success and predator community (Figure 1). Grazing treatments consisted of dormant season grazing, rotational grazing and a control experiencing no grazing during the study. Treatments were assigned randomly to one of the three pastures (5.69 - 7.41 ha) in each block. Pasture size varied among, but not within blocks. Pastures within blocks experienced the similar historical grazing regimes, had similar plant communities, soils, aspect and gradients (Schroeder unpublished data). The dormant season grazing treatment occurred during fall or winter from 2017 - 2020, after the native bunchgrasses entered dormancy. The rotational spring-deferred grazing treatment consisted of alternating between deferment of grazing until after seed-set of bunchgrasses (~mid-July 2017 & 2019) and growing season grazing during May (2018 & 2020) (Figure 1).

Herbage production varied among blocks with differences in site potential and within blocks from one year to the next. In practice, using a consistent stocking rate throughout pastures and years would have resulted in variable grazing intensities due to unequal herbage production. Therefore, we varied stocking rates by herbaceous forage amount in order to consistently achieve moderate grazing utilization (Figure 1). We applied moderate grazing by first estimating available forage prior to turning cattle into pastures by clipping herbaceous material in a sample of the pasture  $(1m^2 \text{ per acre})$  to determine available forage, thus the number of cattle and grazing duration. We visually checked each pasture during cattle grazing every few days to prevent over or under-utilization. We assessed the post-treatment level of grazing utilization by using protocols employed by the local Bureau of Land Management (BLM), the landscape appearance method (e.g., Coulloudon et al., 1999). This technique visually estimates forage utilization based on the general appearance of the rangeland. Utilization levels are determined by comparing observations with written descriptions of six different grazing utilization classes from no use to severe use (BLM personal communication): no use (0-5%), slight (6-20%), light (21-40%), moderate (41-60%), heavy (61-80%) or severe (81-100%). After grazing treatments were completed and cattle removed, the same natural resource professional trained in landscape appearance assessed the pastures for grazing

utilization at 15 evenly spaced grid locations throughout the pasture, and averaged for an overall grazing utilization score.

Vegetation cover, density and visual obscurity was measured during the first year pre-treatment in 2017 and the three years post-treatment during the summers of 2018, 2019 and 2020 along twelve 50 meter transects evenly spaced throughout the study pastures to determine the effect of grazing treatments on herbaceous and woody vegetation. Herbaceous canopy cover was measured by using the line-point-intercept method (Herrick et al. 2005). We recorded vegetation data every 1m by dropping a pin at each 1 m mark and recording every plant type intercepted, to species level for grasses and shrubs, and to functional group for forbs (invasive annual forb, native annual forb, invasive perennial forb or native perennial forb). Density of perennial herbaceous species were determined by counting all plants rooted within fifteen 0.25m<sup>2</sup> quadrats placed every three meters along each transect. We measured plot level visible obstruction by using a modified Robel pole method (Robel et al., 1970). Measurements were taken every 10 meters from two directions for a total of 10 readings per transect. Visibility measurements were taken where an observer stands 5m from a banded decimeter pole, viewing the pole from a height of 1m and records the estimated portion of the decimeter band that was obscured to the nearest 5%. To quantify shrub canopy cover by species, we used the line-intercept technique (Canfield 1941) and we measured shrub density by counting all individuals rooted inside  $2 \times$ 50m belt transects.

#### Results

Density of native perennial forbs, deep rooted perennial bunchgrasses, shallow rooted perennial bunchgrasses, sagebrush and rabbitbrush did not vary by year, treatment site or treatment × year (P> 0.1) (Figure 2). Cover of perennial bunchgrasses was 5.23 and 5.10% lower in 2018 and 2020 compared to pre-treatment data in 2017 and trended 3.00% lower in 2019 compared to pre-treatment data (P < 0.01, P <0.02, P=0.14) (Figure 2). Cover of invasive annual forbs did not differ by year, treatment site or treatment  $\times$  year (P>0.1), but native annual forb cover was on average 1.2% higher overall in all dormant season treatment sites regardless of year (P<0.01), and declined 0.73% in 2018 and 0.87% in 2020 relative to pre-treatment year, regardless of treatment. In 2019 dormant season pastures saw a 1.13% decline in native annual forb cover (P <

0.05). Visual obstruction differed by year (P<0.01), but not for treatment  $\times$  year (P>0.1) (Figure 3).

#### Conclusions

After three years of treatment we did not find any meaningful differences in major vegetation or wildlife habitat variables associated with moderate levels of rotational and dormant season grazing (Figure 2 & 3). We detected a grazing treatment effect where cattle grazed perennial bunchgrasses and reduced bunchgrass cover a consistent amount between dormant and spring/defer treatments (Figure 2A). However, visual obstruction of vegetation, thought to potentially be an important metric for wildlife, did not vary across grazing treatments from ground level to 0.5m above the soil surface, or from 0.5m to 1m heights (Figure 3). These results are in line with our expectations that moderate rotational or dormant season use should not drastically affect plant communities, particularly in the short term. A reduction in the density of deep rooted perennial bunchgrasses would be alarming after just a few years of grazing. Often community level changes take years to manifest. Thus it is imperative to continue longer term treatment application and monitoring to confidently assess the effects of grazing on the sagebrush plant community. We found no evidence that grazing negatively affected metrics deemed important by management agencies for sage-grouse habitat (RMPA table 2-2), including sagebrush cover and density. While we did observe a decline in herbaceous cover (Figure 2), a metric thought to be important to nesting grouse, it was not in conjunction with a reduction in visible obstruction at heights pertinent to nesting sagegrouse. Our replicated, controlled experiment with pre-treatment data, allows us to attribute observed changes directly to the grazing treatment application, something that is not possible with observational landscape scale studies lacking controls or pretreatment data.

This report summarizes the vegetation components of a larger study that is also examining the direct effects of grazing and cattle presence on sagebrush-obligate songbirds. The vegetative component of this research is vital to assessing the direct effects of cattle-grazing on the plant community, and quantifying wildlife habitat. However, wildlife habitat incorporates more than just the plant community, as songbirds fill critical role in a complex ecosystem and food web, influenced by the plant community, insect

availability, weather, and predator-prey dynamics. Cattle likely play a larger role in the ecosystem than simply potentially influencing vegetation and related habitat characteristics. Preliminary results from the songbird portion of this study indicate that weather, not grazing, is the primary driver of nest success for sagebrush-obligate songbirds (Schroeder, thesis in prep). Furthermore, there is some evidence to suggest that grazing increases nest success for one species of songbird, the sagebrush sparrow, and has no obvious effect on the Brewer's sparrow (Schroeder, thesis in prep). This could be due to the role cattle play in modifying songbird predator-prey dynamics. Although the role cattle play in influencing predator-prey dynamics is understudied and not well understood, preliminary evidence from our research suggests grazing might reduce badger, rodent and avian predator activity compared to nongrazed pastures (Schroeder, thesis in prep). The research presented here provides evidence that moderate levels of rotational and dormant season grazing have neutral to positive short-term effects on sensitive sagebrush-obligate songbird reproductive success.

#### Acknowledgments

This research was funded in part by the Oregon Beef Council, US Fish and Wildlife Service and Bureau of Land Management. We would like to thank Lynn Carlon and Skip Nyman for their extensive help with cattle management and treatment application, as well as the countless hours of data collection by our tireless seasonal field crews.

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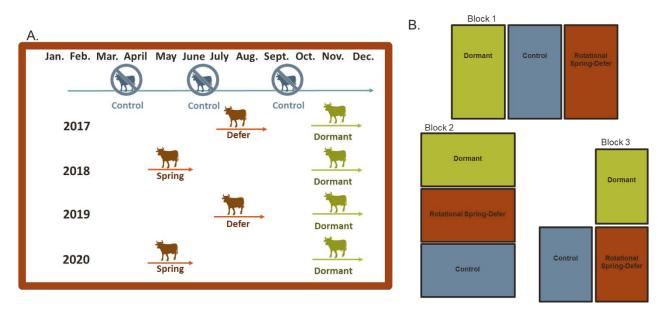
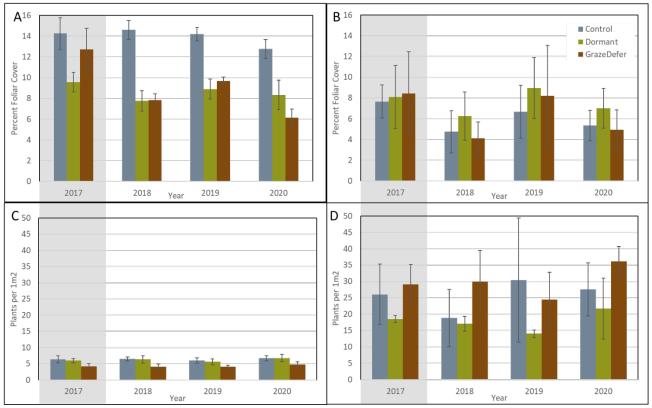
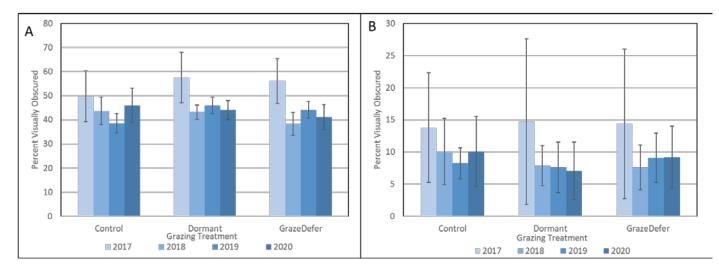


Figure 1. A) Timeline of grazing treatment application and songbird nest success and predator data collection. OBC funds supported the project for 2019/2020. Grazing treatments represented by cow icons and include dormant season (green), rotational spring graze-defer (red-orange) and control (no-grazing, blue). B) Study site layout of randomized experimental block design, consisting of 3 blocks, each with 1 pasture of each treatment. Study conducted at the Northern Great Basin Experimental Range (NGBER) in SE Oregon from 2017 – 2020. Treatments were randomly assigned. Pasture size varied among, but not within, blocks (5.69 – 7.41 HA).



**Figure 2.** Perennial bunchgrass cover (A) and density (C) and native perennial forb cover (B) and density (D) for 4 years of data collection (2017-2020). 2017 represents the pre-treatment year: cattle grazing had not yet been applied (indicated by grey box). There were no differences between the control (blue), dormant season grazed (green) or rotational spring-defer grazed (burnt-orange) for density or cover of perennial forbs or perennial bunchgrass density. We did detect a treatment effect where the cattle grazed the dormant and spring/defer pastures similarly (A).

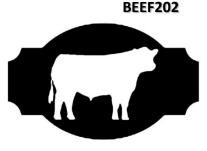


**Figure 3.** The percent visually obscured through time (2017-2020) and across moderate intensity grazing treatments varying in season of use (control experiencing no grazing, dormant season grazing and a rotational spring graze/defer graze treatment) in study pastures located at the NGBER in SE Oregon for a visual band from the soil surface to 0.5m above the ground, viewed from 5m away at a height of 1m, and B) from 0.5m to 1m above the ground. 2017 represents the pre-treatment year: cattle grazing had not yet been applied. Treatments varied significantly across years, but not across grazing treatments.

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Report



**Beef Cattle Sciences** 

### Progress Reports–Rangeland Ecology & Management<sup>1</sup>

#### Fine Fuels Management to Improve Wyoming Big Sagebrush Plant Communities Using Dormant Season Grazing

**Contact Person:** Sergio Arispe, Associate Professor, Oregon State University Extension Service-Malheur County, Ontario, OR 97914 **Email:** Sergio.arispe@oregonstate.edu

**Project Objectives:** The goal of the proposed research is to use an integrated ecological approach to promote rangelands that are resilient to disturbance (specifically fire) and resistant to invasive annual grasses within Wyoming big sagebrush plant communities.

Project Start Date: The funding cycle began July 1, 2019.

**Project Completion Date**: While the current USDA-NIFA CARE funding will end in 2022, the PI is authorized to carry out the research through 2028 and plans to seek funding for summer technical assistants.

#### **Project Status and Preliminary Findings:**

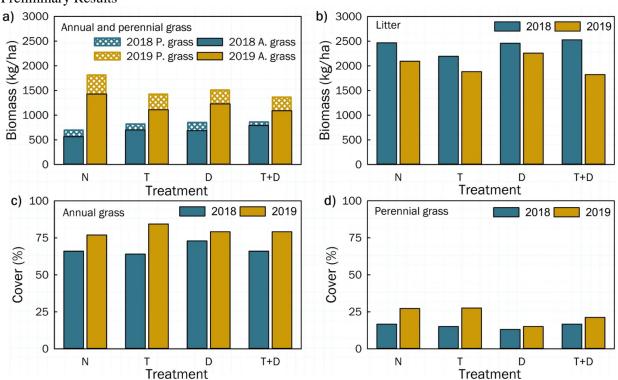
In the fall of 2016, two public land permittees and the Vale District Bureau of Land Management (BLM) Supervisory Rangeland Management Specialist approached the Oregon State University (OSU) Extension Service to implement an experiment to mitigate mega-wildfires in the region. Nearly two years later, the Vale District BLM, OSU Extension Service, and permittees partnered for a landscape-scale dormant season grazing project on three pastures within the Three Fingers Allotment.

Study pastures—McIntyre, South Camp Kettle, and Saddle Butte—are located within the Three Fingers Allotment near Jordan Valley, Oregon (43°19'N, 117°6'W). The allotment is managed by the Vale District BLM with an elevation of approximately 3,800 ft. Annual precipitation ranges between 8"- 12" with the majority falling as rain or snow during the October to March period with an area average annual maximum and minimum temperatures between 40 and 70 F, respectively. Due to repeated wildfires within the pastures, the plant community is dominated by medusahead and cheatgrass; few perennial bunchgrasses and shrubs are present. Historically, livestock grazing on the study pastures has been light to moderate. They are on a rest rotation system so the pastures are not grazed during the same window in consecutive years. During 2020, monitoring took place from June 20-June 30. Researchers took data to quantify cover, gap, herbaceous biomass, density, and height.

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Cover was collected using the line-point intercept method; a pin was dropped every meter along three, 50 m transects (total points = 150 per treatment plot) and all species and ground cover that the pin hit were recorded. Due to the low density of shrubs, shrubs were counted and measured within three, 2 X 50 m belt transects located along each transect. Of those rooted within the belt transect, shrub canopy height, greatest width, and greatest perpendicular width to the first width was recorded and used to estimate canopy cover and biomass using allometric relationships. To determine the extent and distribution of fuels, foliar canopy gaps (including annuals and perennials) greater than 20-cm were measured along each of the three transects.

Herbaceous biomass was collected using a 0.2 m2 quadrat; samples were clipped to  $\sim 1$  cm above ground level and sorted as either perennial bunchgrass, annual grass, forbs, or litter. Biomass was collected every 10 m along each of the three transects (total biomass = 15 samples per treatment plot). Density of 8 life-form categories (perennial tall bunchgrass, perennial short grass, perennial forb, annual forb, exotic annual grass, sagebrush, antelope bitterbrush, and other shrubs) were collected using a 0.2 m2 quadrat and were recorded every 5 m along three, 50 m transects (total quadrats = 30 quadrats per treatment plot). Average vegetation height of grasses and forbs were also recorded using the density quadrats to describe the overall height structure on a treatment plot. Data from summer 2020 are being compiled during the fall 2020.

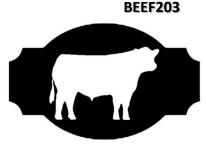


**Figure 1.** Biomass and cover estimates for grazing treatments: no-graze control (**N**), traditional grazing (**T**), dormant season grazing (**D**), traditional + dormant season grazing (**T**+**D**). Annual and perennial grass biomass production (**a**), litter biomass (**b**), annual grass cover (**c**), and perennial grass cover (**d**).

**Preliminary Results** 



Report



**Beef Cattle Sciences** 

## Progress Reports–Rangeland Ecology & Management<sup>1</sup>

#### **Interspace/Undercanopy Foraging Patterns of Horses in Sagebrush Habitats: Implications for Sage-Grouse**

**Contact Person:** David W. Bohnert, 67826-A Hwy 205, Burns, OR 97720, phone: 541-573-8910 **Email:** dave.bohnert@oregonstate.edu

**Project Objectives:** We are using a case study approach to determine the impacts of season-long (8 months/year) horse grazing on 1) sage-grouse nesting habitat structure and composition and 2) behavioral interactions between nesting sage-grouse and grazing horses within active nesting habitat located near a water source.

**Project Start Date**: May of 2018 **Project Completion Date**: May 2022

**Project Status and Preliminary Findings:** An approximately 1,100 acre pasture has been fenced and excluded from grazing by livestock. In addition, due to infrastructure challenges we modified the experimental design. This will result in a longer study but will generate comparable data. Briefly, instead of having 2 separate pastures we will use the same overall acreage in a single pasture with 3 yr of preliminary sage-grouse nesting habitat structure and composition data collected prior to horse grazing. We will then graze horses for at least 2 years and collect comparable data in response to horse grazing. **Vegetation Sampling:** All vegetation measurements will take place in June of each year of the study. Pre-treatment measurements began in 2018. The north and south halves of the pasture were split into three north/south bands that represent increasing distance from water (Figure 1).

**Sage-Grouse:** Preliminary sage-grouse nesting data has been collected in the study area for almost 10 years. We captured additional grouse the spring of 2018 (Figure 2), 2019, and 2020 and placed additional sage-grouse tracking collars on them. This practice will continue for the duration of the study. **Horse Grazing:** We anticipate beginning horse grazing in 2021 depending on the quality of preliminary data collected. We currently plan on using approximately 1 horse/100 acres from April through November. This stocking rate will be based on horse density in the nearest HMA (South Steens). Horses will be unmanaged during the grazing period to replicate feral horse grazing. A perennial drainage on the east end of the plots will provide water for horses.

**Expected outcomes/products:** This research will result in first-of-its-kind data that can be used to characterize the magnitude and nature of the effects of horse grazing on nesting habitat attributes important to sage-grouse and, potentially, the influence of horse grazing on sage-grouse nesting behavior and nest success. These outcomes would be the basis for two peer reviewed journal publications.

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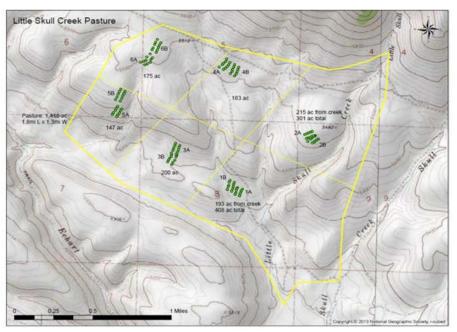


Figure 1. Study Site.

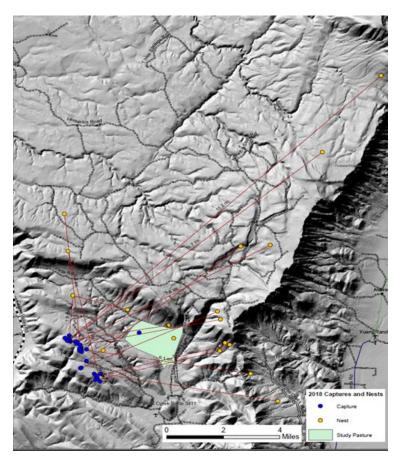
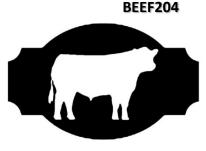


Figure 2. Sage-grouse capture and nesting sites – 2018.



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**Beef Cattle Sciences** 

## Progress Reports–Rangeland Ecology & Management<sup>1</sup>

## Influence of Juniper Removal in Aspen Stands on Greater Steen's Mountain Wildlife

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dustin.johnson@oregonstate.edu

Collaborators: Jonathan Dinkins, Vanessa Schroeder, Samantha Wolfe, Dustin Johnson and Holly Higgins

#### **Project Objectives:**

- 1. Evaluate differential avian predator densities across a juniper cover gradient in association with aspen, riparian, and sagebrush vegetation
- 2. Compare abundance of sensitive wildlife species, specifically mule deer and songbirds, before and after juniper removals associated with aspen, riparian, and sagebrush vegetation

Project Start Date: 2019 (OBC funding for 2020)

Project Completion Date: May 2025

**Project Status:** Increasing juniper encroachment is threatening rangeland ecosystems and associated sensitive wildlife species. Management agencies have implemented and continue to propose conservation actions to mitigate negative effects of juniper encroachment in critical plant communities. Limited information currently exists on the effects of juniper removal in sensitive habitats, such as aspen stands and riparian areas. Understanding how and when juniper encroachment negatively affects sensitive wildlife species, such as songbirds and mule deer, is essential to ensure long-term effective restoration success on multi-use landscapes. We have conducted on the ground surveys and deployed automated audio recording units to collect data on occupancy, species richness, and abundance of sensitive songbirds, raptors, and other avian species. We also deployed game cameras to assess changes in intensity of habitat use of mule deer before and after juniper removals associated with aspen, riparian, and sagebrush vegetation. Juniper treatments were completed in two of the treatment locations (Figures 1–2).

**Preliminary findings:** We conducted 61 songbird surveys, deployed 58 cameras and 29 automated recording units in 2020, capturing 43 songbird species and 7 avian predator species during visual surveys,

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and we successfully captured mule deer, elk, pronghorn, coyote, birds, badgers, rodents and rabbits on game cameras spread across juniper and aspen study sites (Figures 3–4). We are still collecting game cameras and audio units from the field and starting to process automated audio recording unit and game camera data.

Anticipated outcomes: Our research will help inform management decisions on the effectiveness of juniper removal as a conservation strategy for multiple wildlife species. This study will provide crucial information on abundance of numerous avian species and mule deer relative to aspen, juniper, and sagebrush ecosystems in the greater Steens Mountain area. Understanding the relationship between junipers and key sage-grouse avian predators will help land managers make better informed decisions when managing for multi-species and multi-use landscapes. This will also allow for the application of lessons learned regarding bird abundance and diversity and mule deer use to future management across the landscape of the greater Steens area. The results from the study will allow for improved management of game and non-game wildlife on public lands, thereby, increasing wildlife viewing and hunting opportunities. This research will also help improve restoration success on public and private lands. We anticipate several peer reviewed publications resulting from this study:

- 1. Differential raptor and raven abundances along a juniper gradient
- 2. Effects of juniper removal on mule deer abundance and use in aspen and riparian habitats
- 3. Influence of juniper removal on songbird abundance and diversity in the greater Steens Mountain area
- 4. Multi-species juniper management (Extension publication)

Additionally, information garnered from this research will be integrated into OSU's and the EOARC's current outreach/extension programs related to sustainable land management.



Figure 1. Photo of treatment site before juniper removal.



Figure 2. Photo of treatment site after juniper cutting.

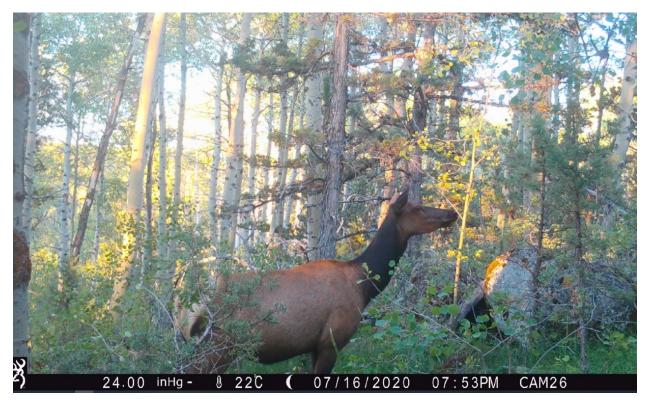


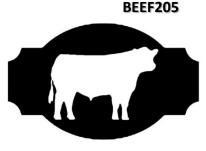
Figure 3. Photo of elk captured in a juniper encroached aspen study survey site.



Figure 4. Photo of a mule deer captured on a study game camera in an aspen survey location.



Report



**Beef Cattle Sciences** 

## Progress Reports–Rangeland Ecology & Management<sup>1</sup>

#### Influence of Ravens on Baker, Oregon Sage-Grouse Population: Assessment of Raven Removal for the Benefit of Sage-Grouse

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lindsey.perry@oregonstate.edu

**Project Objectives:** Our research will help inform management decisions on the effectiveness of raven removal as a conservation strategy for sage-grouse. This research will also assess habitat quality (specifically related to annual grass and fire) for sage-grouse as interactive effects with ravens—a potentially overabundant predator. Sufficient data on sage-grouse and ravens was gathered this past year to be included in analyses related to our objectives. Specific objectives

- 1) Evaluate interactive effects of ravens (presence and/or abundance) with anthropogenic subsidies, annual grass, and fire on sage-grouse.
- 2) Evaluate differences in sage-grouse habitat use, nest success, and chick survival in areas with proportionally more annual grass and/or burned area.
- 3) Assess benefits of raven removal on nest success, chick survival, and habitat use of sage-grouse.
- 4) Identify habitat characteristics associated with habitat use, abundance, and nest success of ravens, including anthropogenic subsidies, annual grass, and fire.
- 5) Evaluate efficacy of raven removal on radio-marked ravens.
- 6) Compare long-term sage-grouse lek trends in the Baker sage-grouse population before and after raven removal to reference areas without raven removal.

**Project Date:** This report details fall trapping from 2018 through the end of the breeding season 2020.

**Project Status and Preliminary Findings:** Introduction: Greater sage-grouse (*Centrocercus urophasianus*: hereafter "sage-grouse") distribution and abundance in western North America has declined over the last century (Connelly et al. 2011), which has prompted multiple petitions to the U.S. Fish and Wildlife Service to list sage-grouse as Threatened or Endangered. Many factors have been attributed to this decline including habitat fragmentation, habitat loss, and predation (Connelly et al. 2011). Several studies suggest that quantity and condition of breeding habitat (micro and landscape scale habitat) dictate the productivity of sage-grouse (Connelly et al. 2011). Herbaceous cover is important to conceal sage-grouse nests from predators (Coates and Delehanty 2010), and microhabitat characteristics

<sup>1.</sup> This document is part of the Oregon State University – 2020 Oregon Beef Council Report. Please visit the Beef Cattle Sciences website at <u>http://blogs.oregonstate.edu/beefcattle/research-reports/</u>

such as sagebrush (*Artemisia* spp.) cover and grass height can influence predation rates at sage-grouse nests (Coates and Delehanty 2010). Landscape factors, such as juniper encroachment, annual grasses, and fire, also have negative consequences on sage-grouse population growth. Understanding mechanisms influencing sage-grouse habitat use and demographic rates related to habitat quantity and quality, including interactions among habitat and predators, is essential to ensure long-term effective restoration success.

Even in excellent sage-grouse habitat, most sage-grouse nests are lost to predators (red fox [*Vulpes vulpes*], badgers [*Taxidea taxus*], coyotes [*Canis latrans*], black-billed magpies [*Pica hudsonia*; hereafter "magpies"], and common ravens [*Corvus corax*; hereafter "ravens"]). Breeding success and population growth of ground-nesting birds can be suppressed by generalist predators, such as ravens (Coates and Delehanty 2010, Dinkins et al. 2016). Raven presence has been negatively associated with sage-grouse nest success and lek trends. Loss and degradation of concealment cover (e.g., sagebrush cover and grass height) combined with increasing raven abundance interactively reduce sage-grouse nest success. For example, sage-grouse nests with greater sagebrush cover were less likely to be depredated by a raven (Coates and Delehanty 2010).

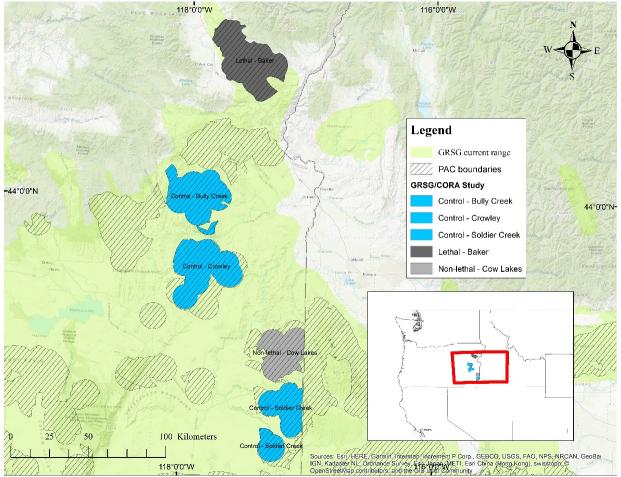
Ravens can reach high densities in landscapes with human-subsidized resources by utilizing human-provided food resources (road-kill, dead livestock, and garbage), perch structures (buildings, power lines, oil and gas wells, etc.), and overwintering shelter (industrial facilities). Sources of perch and nesting structure attract ravens and may increase their foraging ability. In addition, ravens have greater use of areas where intact sagebrush habitat adjoins disturbed habitat. Sage-grouse minimize the risk of predation indirectly by avoiding risky habitat and directly by avoiding avian predators (magpies, *Buteo* hawks, ravens, golden eagles, and northern harriers). Combined effects of avoidance of suitable sagebrush habitat with high raven abundance, raven presence negatively influencing sage-grouse nest success, and increasing raven abundance in sagebrush habitats may have considerable implications for sage-grouse population growth in the future. These findings suggest increases in raven abundance along with habitat degradation—in the form of anthropogenic features, juniper encroachment, annual grass invasion, and fire—may interactively reduce nest success and use of functional habitat available to sage-grouse.

Unlike other population limiting factors (e.g., weather and drought), predation can realistically be reduced by wildlife management agencies. For example, lethal raven removal by Wildlife Services has been demonstrated as a potential tool to reduce negative impacts of raven depredation on sage-grouse nests (Dinkins et al. 2016). However, it may be difficult to implement raven removal for the benefit of sage-grouse, and long-term solutions to reduce raven impacts on sage-grouse are necessary—such as reducing food subsidies and overwinter shelter for ravens while improving sage-grouse habitat. Sagegrouse populations in severe decline may benefit from raven removal followed by identification of longterm management actions to keep raven abundance lower. The Baker, Oregon sage-grouse population has been in severe decline with approximately 350 birds remaining as of spring 2016, and Oregon Department of Fish and Wildlife has quantified extremely high densities of ravens throughout this sage-grouse population. We have initiated a study to evaluate the influence of ravens and the potential benefit of Wildlife Services lethally removing ravens for the benefit of sage-grouse in the Baker, Oregon sagegrouse population. Generally, this project will be focused on identifying habitat characteristics associated with high densities of ravens, raven habitat use (movement, foraging habitat, and nest-sites), and raven nest success. Simultaneously, we will compare sage-grouse habitat use, nest success, chick survival, and population growth among years before and after raven removal. Focus of secondary objectives will generally evaluate the influence of annual grass and fire on ravens and sage-grouse in the Baker sagegrouse population and the Bully Creek, Cow Lakes, Crowley, and Soldier Creek sage-grouse Core Areas in Oregon.

#### Materials and Methods: Study area

The originally proposed project included the Baker, Bully Creek, and Crowley Sage-Grouse Priority Areas for Conservation (PACs); ODFW Grand Ronde and Malheur Watersheds, and the Beulah, Catherine Creek, Keating, Lookout Mountain, Malheur River, and Owyhee Wildlife Management Units (WMUs), on a mixture of public lands administered by the BLM, and private lands. These were the areas within the project for 2017. However, we were able to increase the number of PACs in the study from three to five for 2018 and 2019. The addition included the Cow Lakes and Soldier Creek PACs, which occur in the ODFW Malheur Watershed within the Owyhee and Whitehorse WMUs.

Prior to the start of the 2020 season, we opted to reduce data collection in the Crowley PAC due to reductions in personnel and funding. The Burns Paiute Tribe and a local landowner have elected to continue raven surveys at 19 random points on the west side of the PAC to be use in our long-term comparisons of raven density and lek counts.



**Figure 1.** Boundaries for our five study PACs. PACs designated with blue are reference study PACs; whereas, ravens will be manipulated with lethal or non-lethal management techniques in study PACs designated with grey starting in 2021.

#### Study design

Our study has been stratified by a study area planned to have lethal raven removal (nest destruction and/or adult removal) implemented by ODFW and Wildlife Services (raven-removal study area), a nonlethal raven management area (removal of roadkill/bone pits, perch deterrents, etc) and two study areas without raven management in eastern Oregon. The Baker sage-grouse population now has four years of data collection without raven removal (2017–2020) that will be compared to four year where ODFW and Wildlife Services will implement raven removal (2021–2024). The Cow Lakes PAC now has three years of data collection without raven management (2018–2020) and will be targeted for nonlethal raven management (2021–2024). The Bully Creek and Soldier Creek PACs will not have any type of raven management and will be monitored across the duration of the study. We will compare the relative change in sage-grouse seasonal habitat use, nest success, and chick survival before and after management

of ravens. In addition, we will evaluate raven habitat use, abundance, and nest success before and after management actions.

Sage-grouse monitoring: We will maintain a sample of approximately 80 radio-marked sagegrouse females each year of our study. Captures will occur at night using spotlights and hoop-nets during the spring near lek locations and in the fall around roosting sage-grouse locations. We have and continue to deploy 20 VHF-collars and/or GPS units in each study area. We have and continue monitor VHFcollared sage-grouse females with ground tracking using radio telemetry receivers and Yagi antennas during April–August and aerial surveys during the remainder of the year. Locations have and continue to be recorded bi-weekly via ground tracking and monthly via aerial surveys. Female sage-grouse marked with GPS units have and continue to provide 2–5 locations/day depending on unit type (store-on-board or PTT) and season (breeding or non-breeding).

Female survival have and continue to be recorded with the aid of telemetry signal (mortality switch). Mortality sites have and continue to be visited as soon as possible to assess sage-grouse carcasses and potentially identify cause of death (e.g., disease, fence or power line strike, predator, etc.). Nest locations have and continue to be visually documented while ground tracking. We have and continue to assess nest fate as successful or unsuccessful after a hen has left her nest. A successful nests are defined as having evidence that at least 1 egg hatched as determined by shell membrane condition. We have and continue to assess brood survival bi-weekly by either visually detecting chicks or observing hen behavior that indicates the presence of chicks (e.g., hesitation to flush, feigning injury, or clucking). Brood failure has been and continues to be determined as 3 consecutive visits without detecting chicks and counting chicks at night 35-days after estimated hatch date.

Raven monitoring: To quantify the relative abundance of ravens, we have and continue to conduct 10-minute point count surveys at random locations and 100–200 m away from sage-grouse locations (non-reproductive female, nest, and brood). Point count surveys at sage-grouse locations have and continue to be conducted within a line-of-sight to the actual sage-grouse location. Survey distance away from sage-grouse locations prevent disturbing sage-grouse females and causing observer instigated predation events. Point count surveys at random locations have and continue to be conducted in all study areas. We have and continue to conduct 2–4 point count surveys per month at each random and sage-grouse location April–August and as time and access allow in other seasons.

We have and continue to fit up to 40 ravens with GPS-collars to evaluate raven habitat use, survival, and locate nests in or around our study areas. Raven nests have and continue to be monitored visually by observers in the field to assess nest success. Clusters of GPS locations have and continue to be used to identify nest locations and areas of concentrated use. Areas of concentrated have and continue to be evaluated as potential areas of subsidized resources.

Micro and macro habitat assessment: To assess habitat quality, we will evaluate both micro and macro habitat variables at sage-grouse (nest and brood) and random locations. We sample vegetation at the microhabitat scale with on the ground plots and digital images throughout the breeding season. In brief, we document vegetation at sage-grouse nest and brood and random locations with Robel pole readings, line-intercept, Daubenmire quadrats, and digital cameras. Robel pole readings be used to assess concealment (line-of-sight visibility). We use line-intercept to measure percent canopy cover and height of shrubs. Daubenmire quadrats have and continue to be used to measure herbaceous vegetation variables (grass height and percent canopy cover of annual and perennial grass, residual grass, forbs, litter, cryptobiotic soil, bare ground, and rock). Whereas, macrohabitat scale vegetation (proportion of tree, shrub, grass, etc.) and habitat features (power lines, roads, buildings, etc.) will be quantified with available GIS layers or manually digitized then associated with sage-grouse and random locations throughout each year.

#### Data analysis: Raven abundance

We plan to quantify abundance of ravens with N-mixture models implemented in the Unmarked package or similar package in R. The Unmarked package has functions that allow for inclusion of habitat covariates to describe differences in abundance across the landscape while simultaneously using

covariates to describe differences in detection probability. This will allow us to compare the density of ravens before and after raven removal and among study areas.

Comparison of raven and sage-grouse demographic rates, seasonal habitat use, and movement We will assess raven survival and nest success and female sage-grouse adult, nest, and chick survival with models in program MARK or Cox proportional hazards models. Seasonal habitat use and movement of raven and female sage-grouse will be assessed using resource selection functions, generalized linear mixed models, and/or step-selection functions. Micro and macro habitat variables (including proximity and density of trees, burned area, and annual grass) will be used as predictors of raven and female sage-grouse habitat use and movement. In addition to habitat variables, raven abundance will be assessed as interactive effects with habitat variables influencing sage-grouse habitat use, nest success, and brood success. Interactions among raven abundance and habitat variables will evaluate whether sage-grouse survival rates and seasonal habitat use are disproportionally influenced by the combination of pairs of these variables. To evaluate the long-term influence of ravens on sage-grouse populations and benefits of raven removal, sage-grouse lek trends in removal and non-removal study areas will be compared to raven abundance across approximately eight years.

Preliminary Results: Accomplishments related to raven specific objectives: During the 2018–2020 reporting period, we collected pre-treatment data on 1) raven demographic rates on GPS-marked ravens, 2) conducted raven point count surveys, 3) searched all study areas for raven nests, 4) monitored raven nest success, and 5) banded raven chicks for eventual mark-capture-recapture analyses. We deployed 46 PTT and 2 VHF transmitters on adult ravens, and trapping was conducted year-round (Table 1). Unfortunately, the VHF transmitters did not function. Adult raven capture was attempted in the Baker, Bully Creek, and Cow Lakes PACs. These PACs were prioritized for intensive raven monitoring as lethal and non-lethal manipulation treatment study PACs (Baker and Cow Lakes PACs, respectively) and a reference study PAC (Bully Creek PAC). We will use data from individual ravens to analyze movement, habitat use, and adult survival.

Study PAC	VHF-marked	GPS-marked
Baker	2	30
Bully Creek	0	1
Cow Lakes	0	15
Crowley	0	0
Soldier Creek	0	0
2019 Total	2	46

**Table 1.** Adult raven marking by PAC between August 2018 and August 2020.

Raven nests: During the 2020 breeding season, 51 nests were identified throughout all five study PACs during the months of April, May, and June (Table 2). Previously occupied nest sites were checked for use, and additional nests were located while performing other field work duties. Efforts were focused in the Baker, Bully Creek and Cow Lakes PACs due to time constraints, and all raven nests were documented (Table 2). Nest or fledgling success will be estimated from nest checks, and nest-site selection will be evaluated.

Study PAC	Nests	Chicks	
Baker	23	11	
Bully Creek	10	2	
Cow Lakes	17	0	
Crowley	0	0	
Soldier Creek	1	0	
2019 Total	51	13	

Table 2. Raven nest and banded chick sample sizes by PAC during breeding season 2020.

Chicks: We hand-captured and banded chicks at as many nests as time allowed (Table 2). Banded chicks will be used in mark-capture-recapture survival analyses. All chicks were banded with a metal federal ID band, as well as three color bands representing their capture year and location. Morphometrics were collected and submitted to the Bird Banding Lab. While we place wing tags on ravens caught as adults, we did not place wing tags on chicks in order to minimize potential negative effects on wing development. This also allow for easy identification of banded individuals that were captured as chicks from our study PACs.

Point count surveys: To assess the effect of raven manipulation by management agencies on raven density, point count surveys were conducted during the 2020 field season with 3 surveys between May and July. Random point counts were surveyed throughout sage-grouse PACs and within an 8 km buffer around leks near study PACs where sage-grouse hens were captured or were likely to be captured in subsequent years. Raven point count surveys are described in greater detail in long-term sage-grouse objective section.

Accomplishments related to sage-grouse specific objectives: During the 2018–2020 reporting period, we completed collection of pre-treatment data on 1) sage-grouse demographic rates and habitat use, 2) microhabitat at sage-grouse nest sites, and 3) raven abundance to assess the effect of ravens on sage-grouse. Since fall of 2018, we have deployed 40 VHF and 6 PTT transmitters, and trapping was conducted during fall 2019 and spring 2020 (Table 3). Beginning in fall 2019, all sage-grouse fitted with VHF transmitters also had a Lotek PinPoint-75/120 GPS unit attached to the back of their VHF necklace, which resulted in a combined transmitter weight of <26 g. The Lotek PinPoint-75/120 units are store-on-board GPS technology designed for use on songbirds. We programmed these units to gather 2–3 locations per day. As of August 2020, 30 Lotek units have been deployed in Baker (10), Bully Creek (9), Cow Lakes (3), and Soldier Creek (8). After winter mortalities and a few lost birds, there were 55 sage-grouse at the beginning of the 2020 breeding season (Table 3). All 2020 captures were hindered due to limited personnel from the COVID-19 pandemic. All sage-grouse were monitored via aerial telemetry during winter (November–March) 2019–2020 and ground tracked April–July. Sage-grouse habitat use and survival was monitored for non-reproductive adults, nests, and broods. We located 31 nests and 8 broods (Table 4).

Study Site	August 2018		2019		2020		Total number alive at the start of 2020 breeding season <sup>†</sup>	
	VHF	PTT	VHF*	PTT	VHF*	PTT	VHF*	PTT
Baker	14	0	7	0	1	0	14	0
Bully Creek	9	0	1	0	10	1	15	2
Cow Lakes	0	0	4	4	2	0	2	4
Crowley	0	0	7	2	0	0	4	0
Soldier Creek	0	0	14	0	4	0	10	4

\*includesLotek GPS units deployed with VHF collars

<sup>†</sup>Includes birds captured prior to August 2018

**Table 3.** Total sage-grouse captures August 2018–2020, and number of birds available for ground tracking at the start of the 2020 breeding season.

Study PAC	Nests 2019	Broods 2019	Nests 2020	Broods 2020
Baker	6	2	10	0
Bully Creek	5	3†	10	$4^{\dagger}$
Cow Lakes	8	$1^{\dagger}$	7	unk
Crowley	2	0	2	unk
Soldier Creek	10	2	9	2
Total	31	8	38	6

<sup>†</sup>Includes broods found after nesting season from birds that were in inaccessible areas during poor spring weather.

 Table 4. Sage-grouse nest and brood sample sizes by PAC 2019–2020.

Point count surveys: To assess the effect of raven density on sage-grouse, point count surveys were conducted during the 2019 and 2020 field seasons with 1–2 surveys each month for May–July. Random point counts were surveyed throughout PACs and within an 8 km buffer around leks near study PACs where hens were captured or were likely to be captured in subsequent years. Raven point count surveys are described in greater detail in long-term sage-grouse objective section.

Accomplishments related to long-term sage-grouse objectives: During the 2018–2020 reporting period, we continued data collection on raven abundance throughout five sage-grouse PACs. Raven abundance data will be aligned with sage-grouse lek count data (simultaneously collected by ODFW and partners) to assess the effect of ravens on sage-grouse population trends. Details on raven abundance data collection are below.

Raven point count surveys: Raven point count surveys were conducted during the 2017–2020 field seasons, which has generated sufficient pre-treatment data for the BACI study assessing the benefits of reducing raven abundance for sage-grouse (Table 5). Survey locations were randomly placed, and data was collected 1–2 times each month for May–July. Surveys were completed in Baker, Bully Creek, and Crowley in 2017 and all five PACs in 2018–2020. Point count locations within PACs were conducted at the same spatial positions 2017–2018 in the Baker, Bully Creek, and Crowley PACs. In 2019, some locations in Bully Creek and Crowley were removed from the sample based on access and proximity to sage-grouse capture sites. In Bully Creek, 27 of the original 33 locations were retained for surveys in 2018 and 2019. Of the six locations removed, one was located in unsuitable habitat, while the other five

were inaccessible due to private landowner restrictions. In Crowley, 34 of the original 58 locations were retained. Of the 24 points removed, five locations were removed based on access and time to complete. The remaining 18 locations were removed, because they fell outside of 8 km buffers placed around currently active leks where sage-grouse hens were captured or are likely to be captured in subsequent years. This reduced the survey footprint from 435,000 acres to 314,000 acres. This reduced footprint is much more manageable given logistics and manpower as well as targets density estimates around current sage-grouse use areas. This strategy was also applied to the Cow Lakes and Soldier Creek PACs, resulting in 22 and 27 random points, respectively.

In 2020, adjustments to point counts due to reductions in personnel and funding. Nineteen random points were retained in Crowley but were surveyed by the Burns Paiute Tribe and a local landowner. This will continue for the duration of the study. Additionally, only 17 points on the northeast side of the Owyhee Canyon in the Soldier Creek PAC were retained and surveyed by OSU for the 2020 season. This reduction in point counts will remain through the completion of the study.

Study PAC	2018 Random	2018 GRSG	2019 Random	2019 GRSG	2020 Random	2020 GRSG
Baker	123	10	64	32	86	21
Bully Creek	85	16	71	6	65	18
Cow Lakes	61	9	92	2	57	5
Crowley	106	12	99	5	BPT	2
Soldier Creek	70	28	71	9	42	7
Total	445	75	397	54	250	53

**Table 5.** Total number of completed 10-minute point count surveys during the reporting periods 2018–2020.

Conclusions: Implications of our results will be detailed upon completion of data collection and analysis of data associated with our objectives. This report is associated with year 4 of 8. However, we plan to present and publish research finds upon completion of parts of the project. We anticipate the first set of publication will be out within the next year or two.

Acknowledgments: Our research was funded by the Bureau of Land Management, Oregon Beef Council, Oregon Department of Fish and Wildlife, and US Fish and Wildlife Service. The Burns Paiute Tribe and Oregon Department of Fish and Wildlife provided field equipment and technical support of this study. The USDA/APHIS Wildlife Services National Wildlife Research Center collaborated with raven capture. The Oregon Department of Fish and Wildlife also provided access to sage-grouse lek count data and concurrently collected sage-grouse location data. Our work would not have been possible without the time and effort spent by numerous field personnel, agency staff, and volunteers. In addition, we greatly appreciate the collaboration of over 50 landowners in Baker and Malheur counties; we would not have a large-scale research study without them.

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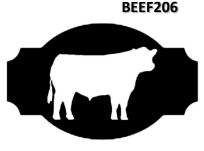
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Report



**Beef Cattle Sciences** 

## Progress Reports–Rangeland Ecology & Management<sup>1</sup>

**Evaluating Rangeland Health, Structure and Function Using Off-The-Shelf Drone Technology to Inform and Enhance Ecosystem Management** 

**Contact Person:** Bryan A. Endress, 205 Badgley Hall, One University Blvd, La Grande, Oregon 97850, phone: 541-962-3115 **Email:** bryan.endress@oregonstate.edu

**Project Objectives:** The goals of this project are to: 1) explore the value and potential of consumer-grade drones as a cost-effective tool to evaluate and monitor critical rangeland resources, and 2) provide guidance and recommendations in their use to livestock producers and land managers. Specifically, the project will evaluate drones use to:

- 1. Measure and analyze rangeland vegetation structure
- 2. Estimate rangeland productivity as it relates to vegetation composition and invasive species (annual grasses).
- 3. Estimate forage utilization and stubble height

Project Start Date: October 2020

**Project Completion Date**: Fall 2023

**Project Status and Preliminary Findings:** Funding will available soon to initiate this research project. In preparation, we have finalized our study sites and have had extensive conversations regarding technological aspects of the project. The drone selected (DJI Phantom 4 Pro + Sentera NDVI Crop Scouting Kit) will allow for both capture of both RGB and NDVI images simultaneously. Drone, software (Pix4D) and computer hardware orders will occur immediately upon funding availability. Over the winter, testing and evaluation will occur will field sampling commencing in the spring of 2021.

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Figure 1: This project will utilize a DJI Phantom 4 Pro drone with a Sentera NDVI single sensor.

Expected outcomes/products: One outcome of this research is the development of new, costeffective tools that can enable livestock producers, land managers and others to more effectively manage land and make resource management decisions based on detailed, high-quality data. The development of protocols and incorporation of drones into to rangeland monitoring efforts represents a new and powerful way to support effective livestock, grazing and ecosystem management plans and activities. The technology has applicability to inform a wide range of management issue including monitoring and mapping of invasive species, tracking forage utilization and stubble height across pastures and allotments, monitoring habitat quality for sage grouse and other wildlife, monitoring rangeland productivity, and evaluating recovery of rangelands following disturbances such as wildfires.



Report



**Beef Cattle Sciences** 

## Progress Reports–Rangeland Ecology & Management<sup>1</sup>

## The Relationship Between Cattle Grazing and the Invasive Annual Grass *Ventenata Dubia* in Oregon

Contact Person: Fara Brummer, Oregon State University Extension-Lake Co, 103 South E St., Lakeview, OR 97630, phone: 541-947-6054 Email: <u>fara.brummer@oregonstate.edu</u> Principal Investigators: Lesley Morris and Fara Brummer

**Project Objectives:** Oregon's working landscapes and wildlife habitat are facing multiple threats to their sustainability. One such threat is the annual grass, ventenata (*Ventenata dubia*), which is a relatively recent invader with very little published research (Wallace et al. 2015). Ventenata has been identified as a problem in several meadow systems of Lake County, Oregon. The increasing abundance of ventenata in these meadows represents a threat to both livestock production and wildlife habitat for Greater sage-grouse. Livestock producers, managers, and researchers in our region all report that cattle will not eat ventenata, but the reasons for low utilization of ventenata unclear. The intersection of cattle and sage-grouse habitat illustrates the need and potential utility to examine the use of targeted grazing as a means for controlling this invasive weed. Based on our previous findings from a pilot study on grazing ventenata (2018-2019), we believe that more research should be done on the differences between early season (May – June) and late season (July- August) grazing on ventenata. We also want to complete another year of forage quality data to bolster the results of our annual forage quality calendar for ventenata.

Project Start Date: Summer 2020 Project Completion Date: October 2021

**Project Status and Preliminary Findings:** We were able to employ an experienced field technician this summer (2020) to clip vententata samples from May 4th to July 4th in a dual meadow system in Lake County that was infested with ventenata. We set up exclosure cages in areas on low, medium, and high ventenata canopy cover and clipped in "paired" areas where cattle were free to graze. All samples will be used for forage quality analysis, including silica measurements.

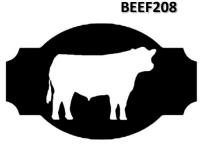
Data analysis of two years of forage quality through the growing seasons and one year examining grazing effect on ventenata cover in low, medium, and high plots after grazing will start in the winter of 2021 with a full report completed by grant completion in fall of 2021. We will also be preparing a manuscript for peer reviewed journal publication during this time frame.

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Report



**Beef Cattle Sciences** 

## Progress Reports–Rangeland Ecology & Management<sup>1</sup>

## A Systems-Based Understanding of Rangeland Watershed-Riparian Systems in Eastern Oregon

**Contact Person:** Carlos Ochoa, 112 Withycombe Hall, Corvallis, OR 97331, phone: 541-737-0933 **Email:** carlos.ochoa@oregonstate.edu

**Project Objectives:** The long-term goal of this project is to improve production and ecological resilience in rangeland watershed-riparian systems of Oregon by providing science-based information to improve upland and riparian areas management. Objectives:

- 1) To characterize biophysical and land use relations influencing water quantity and quality indicators (e.g., stream temperature) in a watershed-riparian rangeland system in eastern Oregon.
- 2) To develop an integrated, systems-based, understanding of ecohydrological relationships and land use information that can be used to develop adaptive management practices, and to inform policy, for achieving or maintaining watershed-riparian system resilience in rangeland ecosystems.
- 3) To collaborate with stakeholders in the co-production of integrated watershed-riparian systems knowledge that will be disseminated through extension and outreach programming.

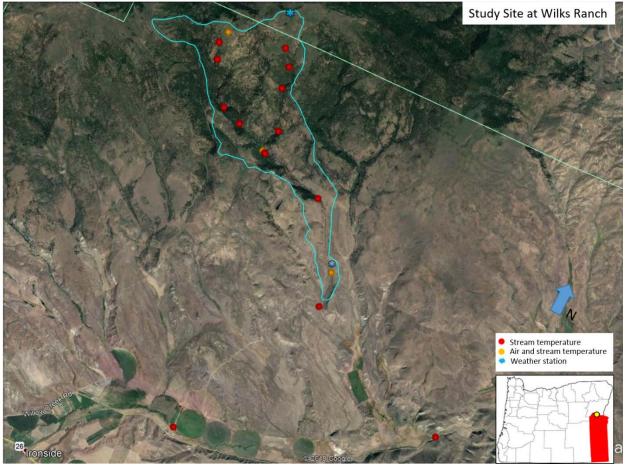
Project Start Date: Spring 2019 (preliminary work started in summer of 2018)

Project Completion Date: Fall 2021

**Project Status and Preliminary Findings:** This long-term project is being established in Malheur County, eastern Oregon. Several ecological and hydrological relationships (e.g., vegetation cover and stream temperature) are evaluated at the Fish Creek watershed-riparian system in Wilks Ranch. This watershed-riparian system offers a great opportunity to understand different land use-environment relationships as it runs through different vegetation types and ecotones. An intensive field monitoring approach is being used to assess ecohydrologic and land use connections at the study site. This field-data collection effort is designed to improve understanding of the effects that critical component interactions (e.g., surface and subsurface water flows) may have on site ecologic functionality and in providing ecosystem services such as forage and water provisioning, habitat, and water quality. The study site was instrumented to monitor multiple hydrologic variables including stream and ambient temperature, soil moisture, streamflow, and weather variables. We installed 17-stream temperature, four air-temperature, and one water-level monitoring stations; from the headwaters to the lower elevation watershed-riparian system. In addition, at upstream and downstream locations we installed two weather stations with satellite-based communication capability for data transfer (Figure 1). We conducted a geologic reconnaissance of the study site, collected

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soil samples at the weather station sites, and in the fall of 2020, we collected water samples for evaluating water quality parameters (e.g., nutrient load) and isotope tracers to detect potential surface-subsurface flow interactions. Also, we established permanent monitoring transects and conducted a vegetation and channel morphology assessment at three different reaches along Fish Creek and Deer Creek.

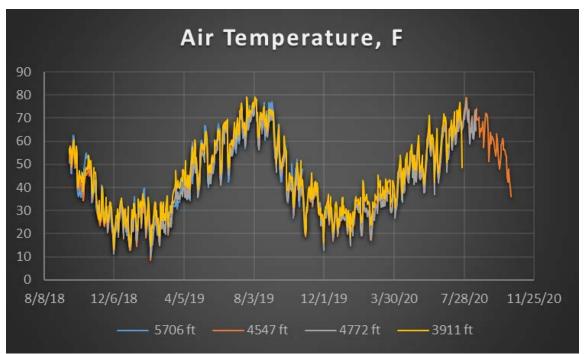


**Figure 1.** Map showing automated field instrumentation at the Fish Creek watershed-riparian system (outlined in blue; 3200 acres) and Willow Creek.

Preliminary results show that a combination of factors including water source (springs), geology, topography, vegetation shade, and channel morphology may contribute to stream temperature along the stream longitudinal gradient. Figure 2 shows water temperature fluctuations in Fish Creek, from August 2018 through September 2020. It can be observed there was a relative constant temperature for the spring water source while temperature in the stream was more variable throughout the year. During the summer, greater stream temperature was observed in lower elevation locations along the stream. A difference of up to 21 degrees Fahrenheit between the spring at 5706 ft and the lower valley stream location at 3911 ft elevation was observed. A difference of up to 12.6 degree Fahrenheit was noted between the stream location at 4970 ft and the lower valley stream location at 3911 ft elevation sensors and the valley at the outlet of the watershed ranged from 0 to 9.6 degree Fahrenheit (Figure 3). Ongoing analysis of the isotopic composition of water at different locations along the stream will provide a better understanding of potential surface-subsurface water flow mixing influencing stream temperature.



**Figure 2.** Daily stream temperature along the longitudinal gradient of Fish Creek, from its spring source at 5706 ft, to its downstream valley at 3911 ft.



**Figure 3.** Daily air temperature along the longitudinal gradient of Fish Creek and Deer Creek watersheds, from its spring source at 5706 ft, to its downstream valley at 3911 ft

Other project photos are below:

Stream temperature sensor and water quality sampling locations.





Schematic representing three transects for measuring stream morphology and riparian-upland vegetation. Also shows a 330-ft reach for measuring riparian vegetation using the 'greenline' method.



Measuring stream morphology and riparian-upland vegetation conditions.



Streamflow measuring device installed at the outlet of Fish Creek.



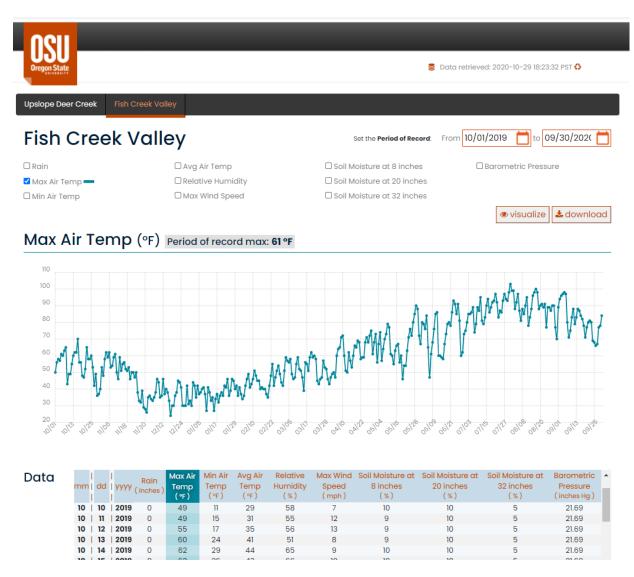
Weather station at the outlet of Deer Creek and Fish Creek watersheds.



Weather station at Deer Creek headwaters.



Snow depth (cm) on January 4, 2020 at the Deer Creek weather station site.

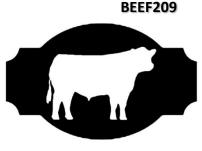


Daily-averaged weather data collected from both weather stations can be accessed at <u>https://ecohydro.live/fish-creek-valley</u> and <u>https://ecohydro.live/upslope-deer-creek/</u>.



**Oregon Beef Council** 

Report



**Beef Cattle Sciences** 

# Progress Reports–Rangeland Ecology & Management<sup>1</sup>

# Irrigation and Seeding Data Effects on Winter Grasses and Forbs Forage Production and Quality in Eastern Oregon

**Contact Person:** Guojie Wang, Eastern Oregon Agricultural Research Center Union, 372 S. 10<sup>th</sup> St., Union, OR 97883, phone: 541-962-3641 **Email:** guojie.wang@oregonstate.edu

**Project Objectives:** The research goal is to search and test the best management practices to produce high quantity and quality forages from winter species in eastern Oregon. The specific objectives are: 1) Evaluate diverse winter forage species to recommend the best choice; 2) Identify the best irrigation management protocol to produce winter forage species; 3) Identify the best seeding time to produce winter forage species; 4) Quantify winter forage species regrowth potential after first cut.

Project Start Date: September 2020

Project Completion Date: August 2021

**Project Status and Preliminary Findings:** We seeded five winter grasses, including winter barley, wheat, rye, triticale, and Italian ryegrass, along with five winter forbs, including hairy vetch, yellow sweet clover, Austrian pea, canola, and radish on September 13 and October 31, 2019. Irrigation in 2020 was carried out according to the experimental design with four treatments: 1) whole season irrigation from May 1 to September 15; 2) late season water shortage irrigation from May 1 to August 1; 3) middle and late season water shortage irrigation from May 1 to June 15; and 4) no irrigation at all. The plots were fertilized, weeded, and monitored after seeding and through the 2020 growing season. We harvested the plots when the corresponding forage species reached their maturity stage and will finish the harvesting this fall of 2020. The forage samples were grinded and sent to the lab for forage quality analysis this fall of 2020. The yield and forage quality data will be presented in the final report next year.

This is an ongoing multi-year project with continuation funding. We collected forage yield data from 2019 and 2020 growing seasons. With one more harvest, the yield data will be comprehensive and finished. We sent forage samples to the lab and will get the results the end of November 2020. So, this report will serve as a progress report for 2020 growing season funding and a partial final report for 2019 growing season funding and preliminary data results.

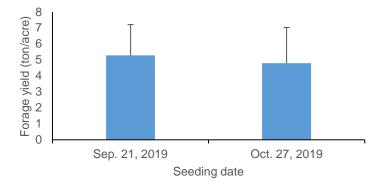
This document is part of the Oregon State University – 2020 Oregon Beef Council Report. Please visit the Beef Cattle Sciences website at <u>http://blogs.oregonstate.edu/beefcattle/research-reports/</u>

Expected outcomes and impacts: This two years study on winter species for forages will result in valuable information about monocultures of winter cereals, biennial legumes, winter legumes, and winter brassicas performances under different irrigation treatments and seeding dates for forage production and quality in eastern Oregon. The production and quality of each winter forage species will be compared with other alternative forage species; the superior number will advocate the best winter forage species as a very good choice under the limited and competing water resources in eastern Oregon. The information will be also used as a guide to select a specific winter forage species, right irrigation practices, and right seeding time that fits a specific farming and ranching situation.

#### Preliminary results from 2019 growing season yield data:

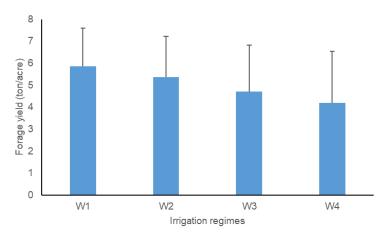
Establishment and winter survival: all five winter grasses established well in 2018 fall seeding based on the seeding density and weed component from the field observations. They were survived the following winter. However, only hairy vetch and winter pea established successfully in 2018 fall seeding and survived the following winter. Brassica and canola did not germinate well, and no competitive stand was established in the spring of 2019. Yellow sweet clover established sporadically.

Seeding date main effects: seeding dates in late fall of 2018 as a main factor did not affect forage yield significantly (Figure 1). However, seeding dates interact with irrigation treatments and forage species affected forage yield (Table 1).



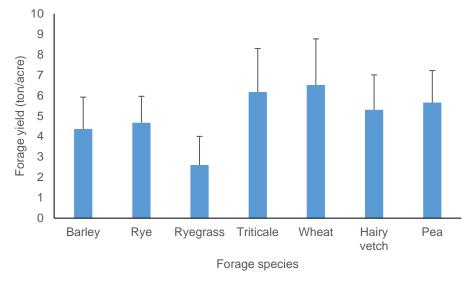
**Figure 1.** Forage yield (ton/acre) in 2019 growing season averaged over seven winter annual grasses and legumes and four irrigation treatments under two seeding dates in 2018 at Eastern Oregon Agricultural Research Center, Union, Oregon.

Irrigation main effects: irrigation treatments in 2018 and 2019 as a main factor did not affect forage yield significantly (Figure 2). However, irrigation treatments interact with seeding dates and forage species affected forage yield (Table 1).



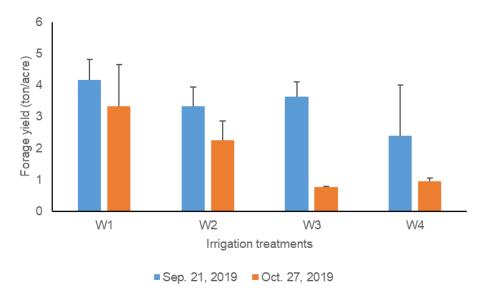
**Figure 2.** Forage yield (ton/acre) in 2019 growing season averaged over seven winter annual grasses and legumes and two seeding dates in 2018 under four irrigation treatments at Eastern Oregon Agricultural Research Center, Union, Oregon. W1: irrigation May 1-September 15; W2: irrigation May 1-August 1; W3: irrigation May 1-June 15; W4: no irrigation.

*Species main effects*: annual ryegrass yield less than the other six species (Figure 3). However, annual ryegrass had the most regrowth under irrigation treatments after the first cut (data not shown).



**Figure 3.** Forage yield (ton/acre) in 2019 growing season averaged over four irrigation treatments and two seeding dates in 2018 under seven winter annual grasses and legumes at Eastern Oregon Agricultural Research Center, Union, Oregon.

Annual ryegrass under less irrigation and late seeding date did not establish well and produce much (Figure 4). The other six winter grasses and legumes produced similar under four irrigation treatments, irrespective to seeding dates (Table 1).



**Figure 4.** Annual ryegrass forage yield (ton/acre) in 2019 growing season under four irrigation treatments and two seeding dates in 2018 at Eastern Oregon Agricultural Research Center, Union, Oregon. W1: irrigation May 1-September 15; W2: irrigation May 1-August 1; W3: irrigation May 1-June 15; W4: no irrigation.

		Seeding on Sep. 21, 2019			S	Seeding on	Oct. 27, 2	2019
	W1†	W2†	W3†	W4†	W1†	W2†	W3†	W4†
Barley	5.47	4.67	5.11	3.89	4.59	4.64	3.27	3.20
Hairy vetch	5.87	5.45	5.39	6.04	5.57	5.35	4.74	3.96
Pea	7.45	6.29	5.31	5.83	4.97	5.53	5.24	4.57
Rye	4.42	4.15	5.01	3.74	6.12	5.72	4.25	3.95
Ryegrass	4.17	3.33	3.63	2.39	3.33	2.26	0.77	0.96
Triticale	7.38	6.21	5.76	4.23	7.93	6.70	5.89	5.29
Wheat	7.66	7.13	6.93	4.91	7.18	7.75	4.84	5.69

<sup>†</sup>W1: irrigation May 1-September 15; W2: irrigation May 1-August 1; W3: irrigation May 1-June 15; W4: no irrigation.

**Table 1.** Seven winter annual grasses and legumes forage yield (ton/acre) in 2019 growing season under four irrigation treatments and two seeding dates in 2018 under at Eastern Oregon Agricultural Research Center, Union, Oregon.

### **REPORT STATUS OF STUDIES FUNDED BY THE OREGON BEEF COUNCIL**

Progress report not required for studies funded prior to 2010-2011 FY and with a full report submitted.

Projects funded in 2007	– 2008 FY		
		Report S	
Abbreviated Project Title	Senior Investigator	Progress	Full
Denveloped Feelom, and Management			
Rangeland Ecology and Management Wolf impact on cattle productivity and behavior	D. E. Johnson		х
Development of digital charting system for range health	D. E. Johnson		X
Livestock, plant community, and sage-grouse food sources	J. Miller		X
Animal Sciences			
Digestibility of cool-season in dairy farms	T. Downing		Х
Female hormones and immune cells in cattle	M. Cannon		Х
Diagnostic test for pregnancy detection in cattle	F. Menino		Х
Assay to assess bovine embryo viability during transfer	F. Menino		Х
Farm-based livestock manure/biogas production	M. Gamroth		X
Glycerol supplementation to cattle	C. Mueller		Х
Copper and Zinc in dairy forage systems	T. Downing		Х
Projects funded in 2008	<u>– 2009 FY</u>		
		Report S	
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Wolf impact on cattle productivity and behavior (cont.)	D. E. Johnson		х
Rangeland vegetation and sediment monitoring	L. Larson	х	X
			~
Animal Sciences			
Late gestation protein supplementation of beef cows	D. Bohnert		Х
Grazing options with Brassicas and Fodder Radishes	C. Engel		Х
Maternal marbling potential and ultrasound technology	C. Mueller		Х
Replacement heifers sired by high or low-marbling bulls	C. Mueller	Х	Х
BVDV and BVDV PI screening to initiate BVDB control	B. Riggs		Х
Selenium supplementation and retention in beef cattle	G. Pirelli	Х	Х
Farm-based livestock manure/biogas production (cont.)	M. Gamroth		Х
Projects funded in 2009	<u>– 2010 FY</u>	Dement C	totus.
Abbrevieted Dreiget Title	Conjer Investigater	Report S	
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Wolf impact on cattle productivity and behavior (cont.)	D E Johnson		Y

D. E. Johnson Wolf impact on cattle productivity and behavior (cont.) Х Х DNA analysis for cattle diet in sagebrush rangelands R. Mata-Gonzales Х Х Behavior and distribution of cattle grazing riparian zones D.E. Johnson Animal Sciences  $PFG2\alpha$  to improve uterine health and reproductive efficiency M. Cannon Х Disposition and reproductive performance of brood cows R. Cooke Х Х Acclimation to handling and heifer development R. Cooke Х Х Х Farm-based livestock manure/biogas production (cont.) M. Gamroth

#### Projects funded in 2010 - 2011 FY

		Report Status	
Abbreviated Project Title	Senior Investigator	Progress	Ful
Rangeland Ecology and Management			
Conflict stressors, spatial behavior and grazing budgets of cattle	D. E. Johnson	Х	Х
Behavior and distribution of cattle grazing riparian zones (cont.)	D. E. Johnson		Х
Grazing and medusahead invasion in sagebrush steppe	D. D. Johnson	Х	Х
Weeds to suppress cheatgrass and medusahead	P. Dysart	Х	Х
Effects of wolves on cattle production systems (cont.)	D. E. Johnson		Х
Quantities diet analysis in cattle using fecal DNA	R. Mata-Gonzales	Х	Х
Animal Sciences			
Protein supplementation to low-quality forage	D. Bohnert	Х	Х
Disposition, acclimation, and steer feedlot performance	R. Cooke	Х	Х
Nutrition during bull development on calf performance	C. Mueller	Х	Х
Extending grazing season with warm season and Brassica forages	S. Filley	Х	Х
Oral Selenium drench at birth to calves	J. Hall	Х	Х

Projects funded in 2011 – 2012 FY
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		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Revegetating sagebrush rangelands Invaded by Medusahead	D. D. Johnson	Х	Х
Potential benefits of Sagebrush consumption by cattle	R. Mata-Gonzales	Х	Х
Effect of wolves on cattle production systems (cont.)	D. E. Johnson		Х
Conflict stressors, spatial behavior and grazing budgets (cont.)	D. E. Johnson	Х	Х
Animal Sciences			
Effects of camelina meal supplementation to beef cattle	R. Cooke	Х	Х
The economics of grassed-based dairying in Oregon	T. Downing	Х	Х
Yeast culture supp. improves feed consumption in cattle	G. Bobe	Х	Х
Western Juniper - Induced Abortions in Beef Cattle	C. Parsons	Х	Х

# Projects funded in 2012 - 2013 FY

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Effect of wolves on cattle production systems (cont.)	D.E. Johnson		Х
Modification of livestock and sage-grouse habitat after juniper control	R. Mata-Gonzales	Х	Х
Prescribed burning and herbicide appl. to revegetate rangelands	D. D. Johnson	Х	Х
Animal Sciences			
Comparison of Ivomec Plus and a generic anthelmintic to beef cattle	R. F. Cooke	Х	Х
Influence of supplement composition on low-quality forages	D. W. Bohnert	Х	Х
Yeast culture supplementation and dairy reproductive performance	G. Bobe	Х	Х
The effect of western juniper on the estrous cycle of beef cattle	C. Parsons	Х	Х

### Projects funded in 2013 - 2014 FY

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Development of forage value index for Ryegrass	T. Downing	Х	Х
Effect of wolves on cattle production systems (cont.)	J. Williams		Х
Jse of herbicide for control of Western Juniper	G. Sbatella		Х
Animal Sciences			
Dxidized lipid metabolites to predict disease in dairy cows	G. Bobe	Х	Х
Cow nutritional status during gestation and offspring performance	R. F. Cooke	Х	Х
Modifying the hormone strategy for superovulating donor cows	F. Menino	Х	Х

#### Projects funded in 2014 - 2015 FY

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Development of forage value index for Ryegrass	T. Downing	Х	Х
Research on stream water temperature and sediment loads	C. Ochoa	Х	Х
Techniques to improve seedling success of forage kochia	D. D. Johnson	Х	Х
Animal Sciences			
Identification of predictive metabolomics markers in dairy cows	G. Bobe	Х	Х
Cow nutritional status during gestation and offspring performance	R. F. Cooke	Х	Х
Modifying the hormone strategy for superovulating donor cows	F. Menino	Х	Х
Energetic output of beef cows based on lactation and calf crop	C. Mueller	Х	
Influence of supplement type and monensin on forage utilization	D. W. Bohnert	Х	Х

#### Projects funded in 2015 - 2016 FY

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Research on stream water temperature and sediment loads	C. Ochoa	Х	Х
Impacts of wolf predation on stress in beef cattle	R. Cooke	Х	Х
Techniques to improve seedling success of forage kochia	D. D. Johnson	Х	Х
Animal Sciences			
Modulation of milk fat synthesis in dairy animals	M. Bionaz	Х	Х
Peripartal vitamin E injections prevent diseases in dairy cows	G. Bobe	Х	
Cow nutritional status during gestation and offspring performance	R. Cooke	Х	Х
Development of enhanced cattle embryo transfer medium	A. Menino	Х	Х
Energetic output of beef cows based on lactation and calf crop	C. Mueller		

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Preventing juniper reestablishment into sagebrush communities	C. Ochoa	X	Х
Research on stream water temperature and sediment loads	C. Ochoa	Х	Х
Greater sage grouse response to landscape level juniper removal	C. Hagen		Х
Greater sage grouse habitat suitability and management in SE Oregon	L. Morris	Х	
Organic fertility effect on alfalfa hay in Central Oregon	M. Bohle	Х	
Annual warm season grasses for forages	G. Wang	X	Х
Animal Sciences			
Peripartal vitamin E injections prevent diseases in dairy cows	G. Bobe	Х	
Feeding immunostimulants to enhance receiving cattle performance	R. Cooke	Х	Х
Development of enhanced cattle embryo transfer medium	A. Menino	Х	Х
n vivo-in vitro hybrid system to perform nutrigenomic studies in cattle	M. Bionaz	X	X
Feeding Se-fertilized hay to reduce parasite load in beef calves	J. Hall	x	X
Evaluation of biological deterrents to manage wolf movements	M. Udel	x	X
L valuation of biological deterrents to manage woll movements	Wi. Odel	~	^

#### Projects funded in 2016 - 2017 FY

# Projects funded in 2017 - 2018 FY

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Preventing juniper reestablishment into sagebrush communities	C. Ochoa	Х	Х
Conservation measures to restore rangeland on sage-grouse habitat	S. Arispe	Х	
How much water do mature and juvenile juniper trees need?	R. Mata-Gonzales	Х	Х
Evaluation of stubble height relationship to riparian health and function	B. Endress	X	Х
Animal Sciences			
Development of enhanced cattle embryo transfer medium	A. Menino		Х
Feeding essential fatty acids to late-gestating cows	R. Cooke	Х	Х
Impacts of estrus expression and intensity on fertility of beef cows	R. Cooke	Х	Х
Increasing milk production in bovine mammary cells	M. Bionaz		Х
Use of platelet rich plasma for endometritis in beef heifers	M. Kutzler	X	Х
Out of Cycle Project			
Identification of cyanobacterium in Lake county	T. Dreher	Х	Х

#### Projects funded in 2018 - 2019 FY

		Report S	Status
Abbreviated Project Title	Senior Investigator	Progress	Full
Rangeland Ecology and Management			
Interspace/Undercanopy foraging by horses in sagebrush habitats	D. Bohnert	Х	
Targeted grazing for control of ventenata dubia in OR meadows	L. Morris	Х	Х
Conservation measures to restore rangeland on sage-grouse habitat	S. Arispe	Х	
Perennial Bunchgrass re-growth under different utilization strategies	D. Johnson	Х	Х
Preventing juniper reestablishment into sagebrush communities	C. Ochoa		Х
Animal Sciences			
Genomic testing for prod.& perf. traits in crossbreed angus cattle	M. Kutzler	Х	Х

#### Projects funded in 2019 - 2020 FY

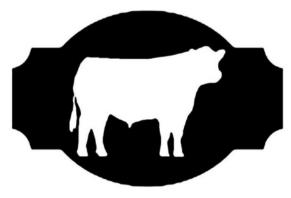
		Report Status	
Abbreviated Project Title	Senior Investigator	Progress	Ful
Rangeland Ecology and Management			
Conservation measures to restore rangeland on sage-grouse habitat	S. Arispe	Х	Х
Fine Fuels Mgt. to improve sagebrush habitat using grazing	S. Arispe	Х	
Influence of Ravens on Sage Grouse in Baker Oregon	J. Dinkins	Х	
Grazing Season of use on Sage-grouse habitat	D. Johnson	Х	Х
Systems-based approach to rangeland riparian systems	C. Ochoa	X	
Animal Sciences			
nvitro/hybrid approach to study nutrigenomic effects of fatty acids	M. Bionaz	Х	Х
Cytokine Expression in Beef Heifers	M. Kutzler	Х	Х
rrigation & Seeding Date effects on Winter forage production systems	G. Wang	Х	
Self-regenerating annual clover in Western Oregon forage Systems	S. Ates	Х	

# Projects funded in 2020 - 2021 FY

Abbreviated Project Title	Senior Investigator	Report Status	
		Progress	Full
Rangeland Ecology and Management			
Fine Fuels Management to Improve Wyoming Big Sagebrush Plant	S. Arispe	Х	
Communities Using Dormant Season Grazing			
Influence of Juniper Removal in Aspen Stands on Greater Steen's	J. Dinkins	Х	
Mountain Wildlife			
Evaluating rangeland health, structure and function, using off the shelf	B. Endress	Х	
drone technology to inform and enhance ecosystem management			
The relationship between Cattle Grazing and the invasive Annual	F. Brummer (L.	Х	
Grass Ventenoto dubio in Oregon	Morris)		
A Systems-based understanding of rangeland watershed-riparian	C. Ochoa	Х	
systems in eastern Oregon			
Irrigation and Seeding Date Effects on Winter Grasses and Forbs	G. Wang	Х	
Forage Production and Quality in Eastern Oregon	-		

S. Arispe	Х
S. Ates	Х
M. Bionaz	Х
S. Arispe	Х
J. Ranches	Х
	S. Ates M. Bionaz S. Arispe





**Beef Cattle Sciences** 

