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## ASSESSING THE IMPACT OF SUPPLEMENTS, FOOD AVERSIONS, AND SILICA

## ON MEDUSAHEAD USE BY SHEEP

by

Tyler Hamilton

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

of

## MASTER OF SCIENCE

in

Range Science

Approved:

Juan J. Villalba Major Professor Elizabeth Burritt Committee Member

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UTAH STATE UNIVERSITY Logan, Utah

2014

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## ABSTRACT

## Assessing the Impact of Supplements, Food Aversions, and Silica on Medusahead Use by

Sheep

by

Tyler Hamilton, Master of Science

Utah State University, 2014

Major Professor: Dr. Juan J. Villalba Department: Wildland Resources

Medusahead now covers about 1 million acres of Oregon and California and is spreading across 10 Western states containing between 30 million and 76 million acres of infested public and private land at an estimated rate of 12% per year. The main issue with medusahead is that once it invades an area, it creates thick patches of thatch that crowd out other forage, producing areas that are primarily medusahead only. In Nevada, medusahead is infesting tens of thousands of acres across northern reaches of the state to the point that has been considered Nevada's "No. 1 weed of concern." From 1962 to 2004, medusahead spread from 18 to 31 of Oregon's 36 counties. In Idaho, rangelands infested by medusahead more than doubled between 1957 and 1992. Livestock grazing losses can reach \$20 an acre each year. Grazing capacity of land infested with medusahead can be reduced by up to 80%. In addition, medusahead invasion can exacerbate the decline of sensitive species such as sage grouse as it replaces plant communities that provide critical habitat for the bird.

The objectives of this study were to determine in sheep: a) the impact of the nutritional context on medusahead intake and preference during different phenological stages of the weed, b) whether medusahead conditions a food aversion, and c) if silica is involved in the process which constrains medusahead intake during grazing. Groups of lambs (n=8) were individually penned and randomly assigned to 4 supplementation treatments: Beet pulp:barley (70:30) (HE), alfalfa:soybean meal (60:40; HP), a choice of HE and HP (CHOICE), or no supplement (Control). After supplementation all animals had *ad libitum* access to medusahead in early reproductive-Trial 1, late reproductive-Trial 2, and thatch-Trial 3 phenological stages. On the last day of each trial, lambs had choices between medusahead and tall fescue hay. Lambs in HE consumed more medusahead than lambs in Control (P < 0.10; Trials 1 and 2). Lambs in HE, HP, and CHOICE showed greater intake of and preference for medusahead thatch than Control lambs during Trial 3 (P < 0.05). Lambs in HE, HP, and CHOICE at more feed and had greater ADGs than Control lambs (P < 0.05). In Trial 4, three groups of lambs (n=10) were fed beet pulp and then received intraruminal infusions of: 1) tall fescue hay (4 g/kg BW), 2) lithium chloride (LiCl; 150 mg/kg BW), and 3) medusahead (4 g/kg BW). Medusahead infusions did not reduce intake of beet pulp relative to infusions of tall fescue hay (P > 0.10). whereas infusions of LiCl conditioned a food aversion (P < 0.05). In Trial 5, three groups of lambs (n=10) were fed:1) alfalfa, 2) alfalfa-2.5% silica, and 3) alfalfa-4.5% silica. Lambs in group 2 had the lowest intake of alfalfa (P < 0.05). This research suggests that supplements can increase preference for and intake of medusahead by sheep. These results also suggest that food aversions do not explain the low palatability of the weed, which may be better understood by the high concentrations of silica in its tissues.

(73 pages)

## **PUBLIC ABSTRACT**

Assessing the Impact of Supplements, Food Aversions, and Silica on Medusahead Use by Sheep

by

## **Tyler Hamilton**

Ranchers, farmers, land managers, and resource management agencies have been plagued with various types of invasive weeds for decades, ultimately affecting millions of acres of viable wildlife habitat, grazing, and recreational opportunities. Invasive weeds often have nutritional contexts that negatively affect animals in some way, increasing the incentive to avoid consuming it by various animal species and domestic livestock. With the current amount of land infested with this weed, and the limited knowledge of the reason for avoidance, the underlying causes of livestock avoidance require in-depth and fundamental understanding.

In my MS program, I sought to explore the relationship of plant and herbivore interactions in regards to grazing, animal diet selection through the use of supplementation, along with understanding the chemical compounds that cause avoidance of medusahead by domestic animals. This research will help us better understand the reasoning behind grazing avoidance and preference, and provide further understanding and management approaches using livestock as an alternative management tool in hopes of creating sustainable ecosystems with complementary benefits for soils, plants, animals, and land managers.

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I would like to thank my committee members for their willingness to help and dedication to the assistance of my degrees completion. Thank you to Beth Burrit for your knowledge, wisdom, for accepting the many editing challenges, insight, and expertise offered, for the thought provoking discussions, and for providing guidance and clarity when needed. Thank you to Dr. Ralph Whitesides for allowing me to be influenced by your love of agriculture, and specifically the study of weeds. Your exceptional knowledge, experience, character, passion for your work, and your influence on the people you associate with is contagious, and has always inspired me to follow my passions and to strive to be of service to others. I would like to express a public thank you to Marsha Bailey for her continued support during my degree completion. Her knowledge, guidance, and willingness to help whenever needed has been a huge factor in allowing me to complete this degree.

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Tyler Hamilton

For my family

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#### **INTRODUCTION**

Medusahead (*Taeniatherum caput-medusae* (L.) Nevski) is an annual grass native to the Mediterranean region, which has invaded millions of acres in the Pacific Northwest, California, Utah and Nevada (Zimmerman *et al.*, 2002). It impacts whole ecosystems, reducing habitat, grazing and biodiversity, commercial and recreational value.

Chemical control of the weed can be effective but it has potential negative side effects such as undesirable impacts on soils and native plants. Moreover, chemical control is often temporary in nature (Davies and Johnson, 2008) and the need for repeated applications makes it unaffordable for many ranchers. Grazing may represent a sustainable, efficient, and low-cost alternative to chemical control. Instead of destroying the plant with costly chemicals, animals could use the carbohydrates, protein and minerals in the plant for their own nutrition while reducing medusahead's abundance and competitiveness. In addition, grazing influences the N cycle by changing litter quality, thereby affecting conditions for N mineralization, and by adding readily available N to upper levels of the soil in urine and feces (Hobbs, 1996). Unfortunately, ungulates typically avoid grazing medusahead (Murphy and Turner, 1959; Bovey et al., 1961), particularly after the weed produces the head or awns, when it becomes too dry and prickly (Coatney, 2008). Efforts have been directed at forcing animals to consume medusahead (i.e., at high animal densities) despite its low palatability in an attempt to understand grazing as a viable alternative for its control. For instance, during spring and in the early boot stage, cattle and sheep eat medusahead at high animal densities (Davy et

*al.*, 2009). However, this approach affects animal welfare and productivity and it may be unrealistic for managing large medusahead infestations (DiTomaso *et al.*, 2008). Moreover, once grazing pressure is relaxed animals will continue avoiding the weed, and probably even more than before because animals' negative experiences with food strengthen food avoidances (Provenza, 1995, 1996).

New paradigms on foraging behavior, such as the importance of positive experiences with the biochemical context (provided by the plant community or supplements) on preference for target feeds may help create innovative management approaches that enhance voluntary intake of medusahead by herbivores. Preference for a particular food depends not only on its intrinsic (e.g., nutritional, toxicological) properties, but also on the nutritional context where that target food is ingested, also referred to as the induction effect, which consists of an increased intake of an unpalatable food when it is associated with the ingestion of a preferred food in a sequence familiar to the animal (Flaherty, 1996; Freidin et al., 2011, 2012). It was hypothesized that preference for medusahead by herbivores is influenced by their experiences with the nutrients consumed in association with the weed. Thus, it was predicted that animals experiencing medusahead in the "right" nutritional context (i.e., with appropriate supplementation) would increase their intake of and preference for the weed relative to animals experiencing medusahead in the "wrong" nutritional context (i.e., when animals are forced to consume the weed without the appropriate supplementation) The biochemical context in which unpalatable foods are consumed is critical for enhancing their use and preference. Thus, the specific array of foods encountered and the sequence of encounters could turn out to be crucial in determining an animal's food preferences

(Villalba *et al.*, 2006; Bergvall and Balogh, 2009). These contextual effects may occur due to the digestive interaction among feeds, induction, or the interplay between these mechanisms (Provenza *et al.*, 2003; Freidin *et al.*, 2011). Studies have shown that supplementation programs (i.e., use of molasses) without understanding the reason animals avoid unpalatable foods like medusahead are not successful. It is not simply the presence or amount of supplemental nutrients, but the *type* and/or proportion of supplemental nutrients offered that influences the use of unpalatable weed species by livestock. Consumption of single foods is not only determined by their intrinsic chemical properties but also by the interaction with other foods in the diet (Flaherty, 1996; Provenza *et al.*, 2003).

#### Silica and Its Effect on Intake

It has been claimed that grazing animals reject medusahead at any stage of growth, possibly due to its low feed value to livestock (Murphy and Turner, 1959; Bovey *et al.*, 1961). Particular attention has been given to the antinutritional effects of the high silica (Si) content of the plant (Swenson *et al.*, 1964). The presence of Si in plant tissue is thought to be a defense mechanism developed by the plant to defend against herbivory (McNaughton *et al.*, 1985). However, Si may not be the main constraint on utilization of medusahead, since a variety of other plants contain high Si concentrations (e.g., rice straw) and animals do not show comparable degrees of avoidance for these examples (Van Soest, 2006). In addition to Si, it was hypothesized that other unaccounted factors are involved in the rejection or avoidance of the weed. It is possible that medusahead contains other secondary compounds that constrain its utilization and cause negative post ingestive feedback inducing a food aversion, which would in part explain the low palatability of the weed to herbivores from such compounds. If true, these compounds at specific concentrations may cause a food aversion, which will limit utilization of the weed (Provenza, 1996). Limited information on the secondary chemistry of the plant is available, and very little is known about the plant chemistry in general.

#### **Physical Properties of the Plant**

A common theory is that livestock do not like medusahead, particularly after it produces the head or awns, especially when it becomes too dry and prickly (Coatney, 2008). Earlier studies reported that as plants matured, sheep selectively avoided medusahead (Lusk *et al.*, 1961). With multiple possible combining factors and the alarming rate of infestation that is occurring across the landscape, identifying and isolating these factors is a key subject of investigation to determine potential mediations of these problems.

#### **Hypothesis and Objectives**

The objectives of this study were to determine in sheep: a) the impact of the nutritional context on medusahead intake and preference during different phenological stages of the weed, b) whether medusahead conditions a food aversion, and c.) if silica is involved in the process which constrains medusahead intake during grazing. I hypothesized that due to the weeds nutritional value, a supplementation program could be used to mitigate the effects of the weeds low nutritional value by using either 1) a high protein based supplement, 2) high energy supplement, or 3) a choice (preference test) between the two. By conducting an aversion trial to ascertain if medusahead is in fact

aversive, and a silica response trial to determine if silica is responsible for a possible aversion, we would be better able to understand its chemical constraints and effects of supplemental preference following herbivory.

## **Expected Benefits**

Completion of this research will enable us to better understand the dynamics of medusahead avoidance by livestock, and gain better understanding of the solution or mitigation of these dynamics to better formulate viable management solutions of the weed using livestock as holistic and sustainable methods of control.

## MATERIALS AND METHODS

The study was conducted at the Green Canyon Ecology Center in Logan, UT according to procedures approved by the Utah State University Institutional Animal Care and Use Committee (Approval # 1551). Lambs in all trials were dewormed against internal and external parasites (Ivermectin; 0.2 mg/kg of BW) and vaccinated against clostridial diseases (*Clostridium perfringens* types C & D and tetanus toxoid; 2 ml/lamb).

All lambs were individually penned outdoors, under a protective roof in individual, adjacent pens measuring  $1.5 \times 2.5$  m. Throughout the study, lambs had free access to water and trace mineral salt blocks. Lambs were weighed prior to, and after each trial. Average daily gains (ADG) were estimated in trials that lasted 10 or more days.

Trials using medusahead in vegetative and early reproductive stage were conducted from May 29 to August 31, 2012. Trial 3, using medusahead thatch, was conducted from May 14 to May 24, 2013.

## Medusahead and Fescue Hay

Naturally established stands of medusahead (*Taeniatherum caput-medusae* (L.) Nevski) were fenced off with poly mesh electric fence on private land in Mantua, located in the north east part of Box Elder County in Utah (<u>41°29'51"N 111°56'32"W</u>). Medusahead was harvested using a lawnmower (particle size 5 cm in length) in the early reproductive stage (from emergence of awns to early emergence of seedhead) during feeding Trial 1 (May 29 to June 8, 2012), and during the late reproductive stage (mature) for feeding Trial 2 (June 20 to June 29, 2012). Thatch was collected from the same area using a rake during early spring of 2013 (May 3, 7 and 8) to conduct Trial 3 (May 14 to May 24, 2013).

Medusahead for Trials 1 and 2 was harvested fresh daily from fenced areas, and transported to the Green Canyon Ecology Center, and fed to lambs upon arrival. Proper transport of medusahead between sites, and disposal of unwanted feed was done to minimize the risk of spread or contamination of areas clean from medusahead. Endophyte-free tall fescue (E-; *Lolium arundinaceum* [Schreb.] Darbysh) hay was harvested and baled in spring of 2012.

The experimental hay was passed through a hydraulic bale grinder (Gehl commercial forage grinder) and cut into lengths of 1-4 mm particle size. The ground material was bagged in 20 kg moisture resistant bags and stored in a shaded building.

## **Animals and Pre-Conditioning Period**

Thirty-four commercial crossbred lambs (2-3 months of age) with an average initial body weight (BW) of  $35 \pm 0.5$  kg were blocked by BW and randomly assigned to one of four treatment groups. Treatment groups were: 1) Control; lambs received no supplement; 2) High Energy (HE); lambs received in a plastic feeder a supplement with a high energy/protein ratio (table 1) containing 70% beet pulp and 30% ground barley (1-2 mm particle size); 3) High Protein (HP); lambs received in a plastic feeder a supplement with a high protein/energy ratio (table 1) containing 60% ground alfalfa hay and 40% soybean meal (1-2 mm particle size); and 4) CHOICE; lambs received a simultaneous offer of HE and HP supplements presented in separate plastic feeders. For 5 days before the onset of Trial 1, lambs in Groups 2-4 were fed 500 g of their respective supplements

at 0800 in order to familiarize the animals to the supplements and experimental protocol. All lambs had alfalfa pellets in *ad libitum* amounts throughout the adaptation period.

#### **Trial 1-Medusahead in Early Reproductive Stage**

Each day from 0830 to 0850 lambs in Groups 2-4 were offered their respective supplements in *ad libitum* amounts. Supplements were then removed and intake was calculated by subtracting the amount refused to the amount offered. All animals then received freshly harvested and chopped medusahead in *ad libitum* amounts in their wooden feeders from 0900 to1400. Throughout the trial lambs were offered between 200-500g of medusahead (as-fed basis), and additional amounts of medusahead were added to the feeders when the amounts remaining were below 20% of the amounts offered initially. Medusahead intake for each animal was estimated as described before.

Following removal of medusahead, all groups received *ad libitum* amounts of endophyte-free tall fescue hay from 1400 to1600. Tall fescue intake for each animal was estimated as described before and no other food was offered until the next day at 0830. The trial was conducted from May 29 to June 7, 2012.

On June 8, after collecting supplement refusals, from 0900 to 1400 all lambs received a choice of tall fescue hay and medusahead. After this period, tall fescue hay and medusahead refusals were collected and intake was estimated as described before. No other food was offered. On the following day, all animals were weighed and then offered alfalfa pellets in *ad libitum* amounts until Trial 2 started.

## **Trial 2-Medusahead in Late Reproductive Stage**

The day before Trial 2 started, alfalfa pellets were removed at 1600. The protocol for this study was as described for Trial 1. Medusahead (late reproductive stage) was offered from June 20 to June 28, 2012. A preference test was conducted on June 29, 2012.

## **Trial 3-Medusahead – Thatch**

A new group of 34 commercial crossbred lambs (2-3 months of age) with an average initial body weight (BW) of  $27 \pm 1.1$  kg were used for this trial. The protocol for this study was as described for Trial 1. Medusahead thatch was offered from May 14 to May 23, 2013. A preference test was conducted on May 24, 2013.

#### **Trial 4- Is Medusahead Aversive to Sheep?**

A new group of 30 commercial crossbred lambs (2-3 months of age) with an average initial BW of  $32 \pm 0.7$  kg were used for this trial. Lambs were blocked by BW and then randomly assigned to three treatment groups: 1-Negative Control; 2-Positive Control; 3-Medusahead. All lambs were fed *ad libitum* amounts of a basal diet of alfalfa pellets.

*Familiarization with Flavored Beet Pulp.* For five consecutive days and at 0830, each lamb received 200 g of beet pulp (1-2 mm particle size) containing 3% of coconut flavor (Lucta, S.A., Montornés del Vallés, Spain). A flavor was added to beet pulp in order to enhance the likelihood of animals forming strong associations between the taste of beet pulp and the postingestive treatments provided during the trial (Provenza, 1996).

Administration of Treatments. On July 22, 2012 alfalfa pellets were collected at 1700 and no other food was offered until the next day. On July 23, 2012 and at 0800 all lambs were offered 300 g of coconut-flavored beet pulp. At 0850 refusals were collected and weighed. Intake was estimated as described for Trial 1. After refusal collection, lambs in Group 1-Negative Control were given 4 g/kg BW of endophyte-free tall fescue hay in a volume of 2 L tap water by oral gavage. Lambs in Group 2-Positive Control were given an emetic toxin; LiCl at 150 mg/kg BW via oral gavage in a volume of 2 L tap water. Lambs in Group 3-Medusahead were given 4 g of medusahead/kg BW by oral gavage in 2 L tap water. The medusahead infused was in the early reproductive stage and the dose selected corresponded to the highest average amount of medusahead (DM basis) consumed during a 5- hr period by lambs during Trial 1, when medusahead was offered to lambs in the same phenology. Prior to the trial, medusahead was collected from the described location, stored in a freezer at -20°C and then freeze dried. Both medusahead and tall fescue hay were ground using a Wiley Mill (1-mm mesh). Doses of medusahead and tall fescue were mixed with water inside a plastic bottle immediately before gavaging.

At 1700, all lambs were offered alfalfa pellets in *ad libitum* amounts until 1900. Refusals were collected and intake was estimated. No other food was offered until the next day.

*Testing*. At 0800 all lambs were offered 400 g of coconut-flavored beet pulp for 15 minutes. Refusals were collected and weighed as described for the previous section. At 1700, all lambs were offered alfalfa pellets in *ad libitum* amounts until 1900, when

refusals were collected and no other food was offered until the next day. Testing was conducted during 2 consecutive days (July 24 and 25, 2012).

#### Trial 5- Is Silica Involved in the Low Use of Medusahead by Sheep?

Thirty lambs from Trials 1 and 2 were (with an average initial BW of  $38 \pm 0.7$  kg) were randomly assigned to 3 treatment groups. Each day at 0830 lambs received in *ad libitum* amounts: 1-Control: Ground alfalfa pellets (1-2 mm particle size); 2-Silica-2.5%: Ground alfalfa pellets containing 2.5% Silica; 3-Silica-4.5%: Ground alfalfa pellets containing 4.5% Silica. At 1400 refusals were collected and weighed. Following removal of feeds, all groups received *ad libitum* amounts of ground endophyte-free tall fescue hay (1-2 mm particle size) from 1400 to1600. Tall fescue intake for each animal was recorded as described before and no other food was offered until the next day at 0830. Lambs were given their respective treatment diets (Control; Silica-2.5%; Silica-4.5%) for 5 days (August 14 to August 18, 2012) to familiarize them with the feeding protocol. Then, they were fed *ad libitum* amounts of alfalfa pellets until August 21, when refusals were collected at 1600 in order to fast animals overnight before the ensuing trial.

The trial was conducted from August 22 to August 31, 2012. For 5 days (from August 22 to August 26, 2012) all animals were fed ground alfalfa pellets from 0830 to 1400 and then ground fescue hay from 1400 to 1600 in order to determine intake of alfalfa hay and tall fescue hay during a baseline period without silica. Lambs were then fed their respective treatment diets (Control; Silica-2.5%; Silica-4.5%) and tall fescue hay during the same time periods from August 27 to August 31, 2012.

## **Chemical Analyses**

Every day that medusahead was fed, representative samples of the weed were placed in plastic bags and transported to a freezer where they were kept at -20 °C and subsequently freeze dried. Dried medusahead, alfalfa pellets and tall fescue hay, and beet pulp samples were ground through a Wiley mill with a 1-mm screen, and analyzed for dry matter (Method 930.15 AOAC, 2000), neutral detergent fiber (NDF), acid detergent fiber (ADF) (Van Soest *et al.*, 1991), and nitrogen (N) (Method 990.03 AOAC, 2000). Medusahead and tall fescue hay were also analyzed for acid insoluble ash (AIA) (Van Keulen and Young, 1977) as an estimate of silica content.

Representative samples of the feeds were placed in paper bags and dried in a forced-air oven at 60  $^{\circ}$ C for 48 hr to estimate dry matter content, in order to express intake values on a dry matter basis.

#### **Statistical Analyses**

Analyses were computed using a mixed model (MIXED procedure; SAS Inst., Inc. Cary, NC; Version 9.1 for Windows). The variance-covariance structure used were those (autoregressive order-1, compound symmetry, variance components), which yielded the lowest Bayesian information criterion. The model diagnostics included testing for a normal distribution of the error residuals and homogeneity of variance. Means were analyzed using pairwise differences (DIFF) of least squares means (LSMEANS).

Food intake, expressed as grams of feed consumed/kg of metabolic body weight  $(BW^{0.75})$ , was analyzed as a mixed model with repeated measures (day) and lambs (random factor) nested within treatment group (fixed factor).

During preference tests, food intake and preference [(intake of a plant species/total intake) x 100] were analyzed as a mixed model with lambs (random factor) nested within treatment group (between-subject factor). Plant species (medusahead, tall fescue hay) was the within-animal factor in the analyses.

Separate analyses were conducted for the CHOICE treatment (Trials 1 to 3) to estimate lamb preference [(intake of a supplement/total supplement intake) x 100] for each supplement. In this case, animal and supplement (HE or HP) were the whole-plot factors and day was the repeated measure.

Average daily gains (ADG) were analyzed as a mixed model with lambs (random factor) nested within group (fixed factor).

#### RESULTS

## **Nutritional Analyses**

Nutritional analyses for the feeds used in the study are reported in table 1. Medusahead quality declined with maturity, particularly CP, although AIA values were the lowest for thatch. Tall fescue hay had greater concentration of CP and lower concentration of ADF, ADF, and AIA than medusahead at any phonological stage tested.

## **Trial 1-Medusahead in Early Reproductive Stage**

Lambs in HE consumed more medusahead than lambs in Control (June 2, 5, and 6; Group x Day; P < 0.10; figure 1A). Lambs in CHOICE ingested the greatest amount of supplement (Group; P < 0.0001; figure 2A). No differences among groups were detected in intake of tall fescue hay (Group; P = 0.50; Group x Day; P = 0.99; figure 2A).

Lambs in CHOICE selected similar amounts of high-energy and high-protein supplements (16.0 vs.16.8± 1.6 g/Kg<sup>0.75</sup>, respectively; Supplement; P = 0.72; Supplement x Day; P = 0.58). In addition, lambs in CHOICE ingested the greatest amount of total feed (medusahead+supplement+hay), whereas lambs in Control ingested the least amount (Group; P < 0.0001; figure 2A). The pattern of ADG followed the same pattern, with the greatest ADG for CHOICE and the least ADG for the Control group (Group; P = 0.0002; table 2).

When offered a choice between medusahead and tall fescue hay, lambs in HE tended to eat more medusahead than the other groups (Group; P = 0.13; figure 3A). Preference for medusahead for lambs in HE, HP, CHOICE, and Control was 10.6; 5.1; 6.5; and 5.6± 2.3%, respectively (Group; P = 0.32).

#### **Trial 2-Medusahead in Late Reproductive Stage**

Averaged across days, lambs in HE consumed more medusahead than lambs in the Control treatment (2.2 vs.  $1.2 \pm 0.3$  g/Kg<sup>0.75</sup>; Group; P = 0.04; Group x Day; P = 0.82; figure 1B). Lambs in CHOICE ingested the greatest amount of supplement (Group; P < 0.0001; figure 2B). Lambs in Control (23-24 and 26-28 June) and HP (24 and 28 June) ate more tall fescue hay than lambs in HE, whereas lambs in CHOICE ate more tall fescue hay than lambs in HE and HP (June 21) and in Control (22 June) (Group; P = 0.38; Group x Day; P = 0.007).

Lambs in CHOICE ingested greater amounts of the high-energy than of the highprotein supplement (18.3 vs.  $15.2 \pm 1.0 \text{ g/Kg}^{0.75}$ , respectively; Supplement; P = 0.04; Supplement x Day; P = 0.09). Lambs in CHOICE ingested the greatest amount of total feed, whereas lambs in Control ingested the least amount (Group; P < 0.0001; figure 2B). Consistent with this, lambs in the Control group lost weight (negative ADG), in contrast to the rest of the groups which showed positive values (Group; P = 0.006; table 2).

When offered a choice between medusahead and tall fescue hay, lambs in HE and CHOICE tended to display a greater intake of medusahead than lambs in Control (Group; P = 0.18; figure 3B). Preference for medusahead for lambs in HE, HP, CHOICE, and Control was 7.4; 4.0; 5.4; and  $1.2\pm 1.8\%$ , respectively (Group; P = 0.12). Preference for medusahead in the Control treatment was not different from 0 (P = 0.49).

## **Trial 3-Medusahead – Thatch**

No differences in thatch intake were detected among groups of lambs (Group; P = 0.94; Group x Day; P = 0.29; figure 1C). Lambs in CHOICE ingested the greatest

amount of supplement (Group; P < 0.008; figure 2C). No differences among groups were detected in intake of tall fescue hay (Group; P = 0.36; Group x Day; P = 0.42; figure 2C). Lambs in CHOICE selected similar amounts of high-energy and high-protein supplements (15.4 vs.  $16.9 \pm 2.1 \text{ g/Kg}^{0.75}$ ; Supplement; P = 0.62; Supplement x Day; P = 0.67). Lambs in CHOICE ingested the greatest amount of total feed, whereas lambs in Control ingested the least amount (Group; P < 0.0001; figure 2C). Consistent with this, lambs in the Control group lost weight (negative ADG), in contrast to the rest of the groups, which showed positive values (Group; P = 0.03; table 2).

When offered a choice between medusahead thatch and tall fescue hay, lambs in HE, HP and CHOICE showed greater intake of medusahead thatch than Control lambs (Group; P = 0.002; figure 3C). Preference for medusahead for lambs in HE, HP, CHOICE (9.4; 8.4; and 15.6± 2.0%, respectively) was also greater than for lambs in Control (3.7± 2.0%; Group; P = 0.002).

Item			%DM	
nem	CP <sup>6</sup>	NDF <sup>7</sup>	ADF <sup>8</sup>	AIA <sup>9</sup>
HE <sup>1</sup>	11.3±0.4	31.7±0.5	19.8±0.6	$ND^{10}$
$HP^{2}$	28.8±0.5	31.9±0.6	25.1±0.4	ND
Medusahead – Trial 1 <sup>3</sup>	6.9±0.1	44.9±0.3	65.2±0.5	10.2±0.2
Medusahead – Trial 2 <sup>4</sup>	5.4±0.1	46.8±0.5	65.8±0.5	9.4±0.2
Medusahead – Trial 3 <sup>5</sup>	3.7±0.1	55.0±0.7	70.0±0.1	6.6±0.4
Tall Fescue Hay – Trials 1-3, and 5	14.9±0.2	32.6±0.4	45.7±0.2	4.9±0.2
Beet Pulp – Trial 4	9.8±0.1	26.2±0.3	38.1±0.4	ND
Alfalfa Hay Trials 1-5	15.6±0.6	30.1±0.8	41.0±0.4	ND

**Table 1.** Nutritional composition of supplements, medusahead, endophyte-free tall fescue, beet pulp, and alfalfa hay during the study.

<sup>1</sup>High-energy supplement: 70% Beet Pulp, 30% Barley

<sup>2</sup>High-protein supplement: 60% Alfalfa, 40% Soybean Meal

<sup>3</sup>Medusahead was fed in the early reproductive stage (May 29 to June 8, 2012)

<sup>4</sup>Medusahead was fed in the late reproductive stage (June 20 to June 29, 2012)

<sup>5</sup>Medusahead thatch collected during May 3, 7 and 8, 2013

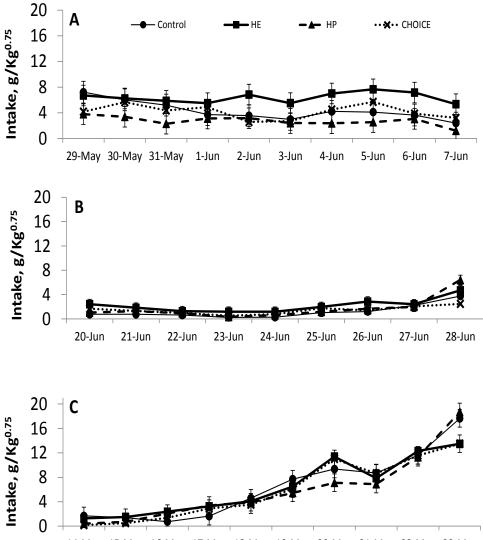
<sup>6</sup>Crude protein

<sup>7</sup>Neutral detergent fiber

<sup>8</sup>Acid detergent fiber

<sup>9</sup>Acid insoluble ash

<sup>10</sup>Not determined



14-May 15-May 16-May 17-May 18-May 19-May 20-May 21-May 22-May 23-May

Figure 1. Intake of medusahead by four groups of lambs during three trials when medusahead was fed in three different phenological stages. Before ingesting medusahead lambs were supplemented with: 1) a high-energy supplement (HE): beet pulp:barley (70:30), 2) a high-protein supplement (HP): alfalfa:soybean meal (60:40), 3) a choice of HE and HP (CHOICE), or no supplement (Control). All animals then had *ad libitum* access to fresh-cut medusahead (5 h/d) and subsequently a basal diet of tall fescue hay (2h/d). **A.** Trial 1. Medusahead- early reproductive stage. **B.** Trial 2. Medusahead- late reproductive stage. **C.** Trial 3. Medusahead – thatch. Values are means for 8 lambs; SE are represented by vertical bars.

Figure 1

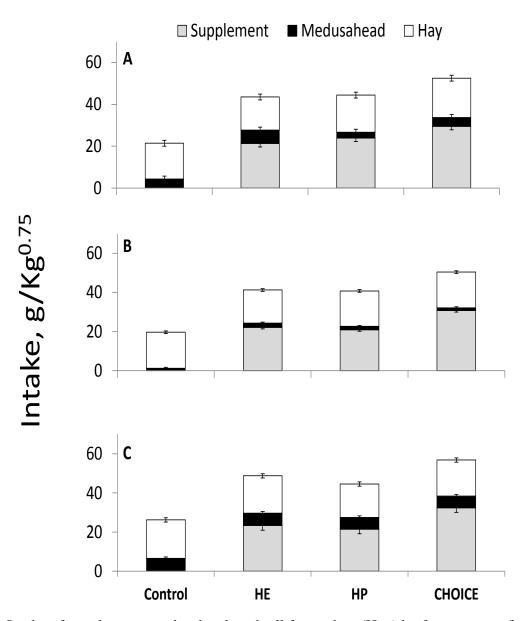


Figure 2. Intake of supplement, medusahead, and tall fescue hay (Hay) by four groups of lambs during three trials when medusahead was fed in three different phenological stages. Before ingesting medusahead lambs were supplemented with: 1) a high-energy supplement (HE): beet pulp:barley (70:30), 2) a high-protein supplement (HP): alfalfa:soybean meal (60:40), 3) a choice of HE and HP (CHOICE), or no supplement (Control). All animals then had *ad libitum* access to fresh-cut medusahead (5 h/d) and subsequently a basal diet of tall fescue hay (Hay; 2h/d). A. Trial 1. Medusahead- early reproductive stage. B. Trial 2. Medusahead- late reproductive stage. C. Trial 3. Medusahead – thatch. Values are means for 8 lambs; SE are represented by vertical bars.

Figure 2

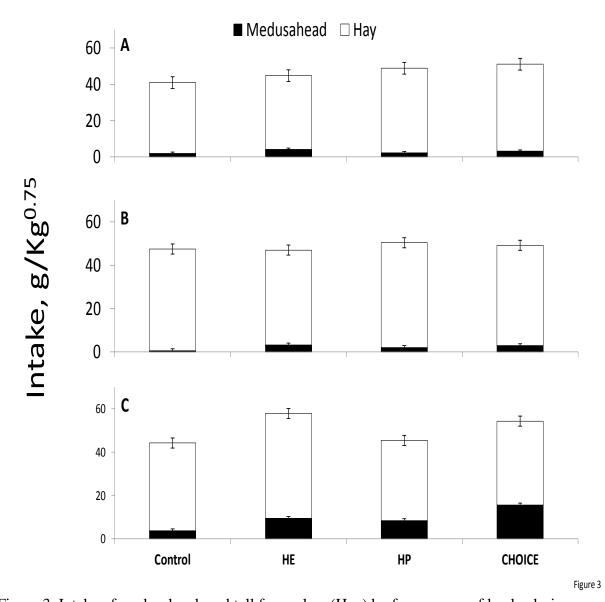


Figure 3. Intake of medusahead, and tall fescue hay (Hay) by four groups of lambs during three preference tests, each conducted after periods of feeding medusahead at three different phenological stages and supplementing lambs with: 1) a high-energy supplement (HE): beet pulp:barley (70:30), 2) a high-protein supplement (HP): alfalfa:soybean meal (60:40), 3) a choice of HE and HP (CHOICE), or no supplement (Control). **A.** Trial 1. Medusahead – early reproductive stage. **B.** Trial 2. Medusahead – late reproductive stage. **C.** Trial 3. Medusahead – thatch. Values are means for 8 lambs; SE are represented by vertical bars.

Trial	Weight <sup>1</sup>	ADG <sup>2</sup>	SEM <sup>3</sup>	<b>P</b> <sup>4</sup>
1. Medusahead – Early Reproductive Stage			0.05	0.0002
Control	35.4	$+0.11^{a}$		
HE	35.0	$+0.25^{b}$		
HP	34.8	$+0.24^{b}$		
CHOICE	35.4	+0.49 <sup>c</sup>		
2. Medusahead – Late reproductive stage			0.05	0.006
Control	36.6	-0.07 <sup>a</sup>		
HE	37.7	+0.12 <sup>b</sup>		
HP	37.4	+0.23 <sup>b</sup>		
CHOICE	40.8	+0.10 <sup>b</sup>		
3. Medusahead– Thatch			0.08	0.03
Control	26.5	- 0.009 <sup>a</sup>		
HE	27.4	+0.25 <sup>b</sup>		
HP	27.5	$+0.25^{b}$		
CHOICE	27.0	$+0.32^{b}$		
5. Silica			0.04	0.84
Control	37.6	+0.14		
2.5%	38.1	+0.14		
4.5%	38.4	+0.11		

**Table 2.** Weight of lambs in Trials 1, 2, 3, and 5.

<sup>2</sup>Average daily gains (ADG; kg). Within each trial, means with different characters differ; P < 0.10.

<sup>3</sup>Standard error of the mean for ADG.

<sup>4</sup>Treatment effect

## **Trial 4- Is Medusahead Aversive to Sheep?**

No differences in intake of coconut-flavored beet pulp among groups of lambs were detected before infusions (July 23). However, on the 2 days after the infusions, lambs in the Positive Control (LiCl infusions) ingested the least amounts of coconutflavored beet pulp. No differences in ingestion of beet pulp were detected throughout the trial between the groups that received intraruminal infusions of medusahead or tall fescue hay (Negative Control) (Group; P = 0.0008; Group x Day; P = 0.03; figure 4). Likewise, lambs in the Positive Control ingested the least amounts of alfalfa pellets and no differences were detected between the groups that received intraruminal infusions of medusahead or tall fescue hay (Negative Control) (10.5 vs. 15.4 and  $15.5 \pm 0.9 \text{ g/Kg}^{0.75}$ , respectively; Group; P = 0.001).

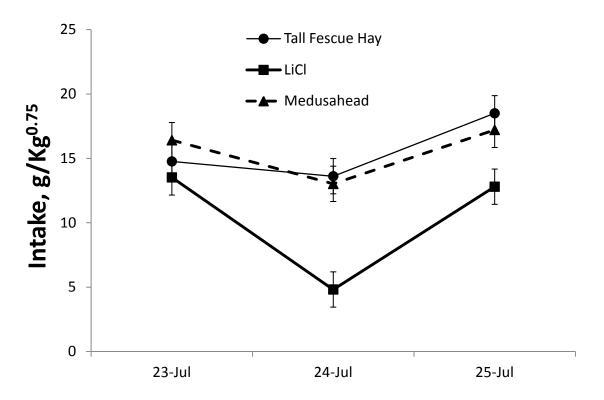


Figure 4. Intake of flavored beet pulp by lambs that received intraruminal infusions of: 1) tall fescue hay (Hay; Negative Control), 2) LiCl (Positive Control), and 3) medusahead. Values are means for 10 lambs; SE are represented by vertical bars.

#### Trial 5- Is Silica Involved in the Low Use of Medusahead by Sheep?

No differences in intake of ground alfalfa (Group; P = 0.90; Group x Day; P = 0.84) or tall fescue hay (Group; P = 0.68; Group x Day; P = 0.34) were detected between groups before lambs were introduced to their respective treatment diets containing silica (August 22-26; figure 5A,B).

When silica was mixed with alfalfa, lambs in the group Silica-2.5% displayed the lowest values of intake (P < 0.05). Lambs in the group Silica-4.5% showed lower intake values than lambs fed ground alfalfa (Control) only during the first day of the trial (Group; P = 0.006; Group x Day; P = 0.04; figure 5C). Lambs in Silica-4.5% ate more tall fescue hay than Control lambs (P < 0.05), and lambs in Silica-2.5% ate more tall fescue hay than Control lambs during days 1-2 and 4-5 of the trial (Group; P = 0.01; Group x Day; P = 0.008; figure 5D).

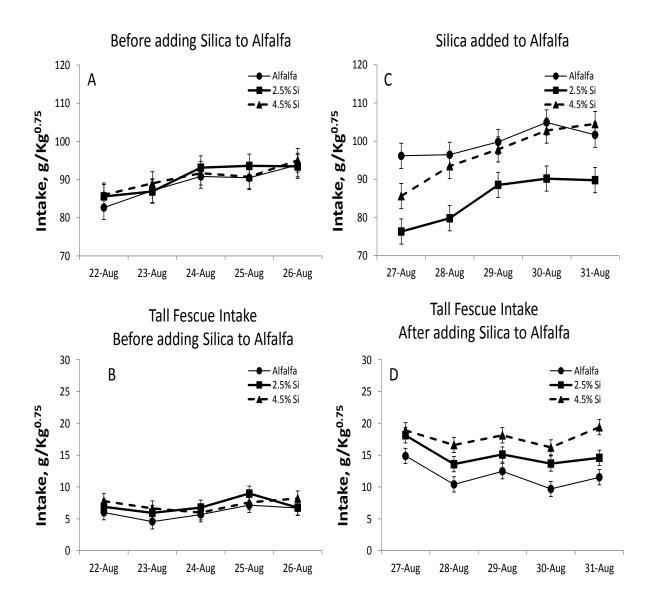


Figure 5. Intake values by three groups of lambs fed: 1) alfalfa, 2) alfalfa containing 2.5% silica (2.5% Si), and alfalfa containing 4.5% silica (4.5% Si). After consuming these feeds, lambs were offered ground tall fescue hay. **A.** Intake of alfalfa hay before adding silica to the alfalfa diets. **B.** Intake of tall fescue hay before adding silica to the alfalfa diets. **C.** Intake of alfalfa hay when silica was added to the alfalfa diets. **D.** Intake of tall fescue hay when silica was added to the alfalfa diets. SE are represented by vertical bars.

#### DISCUSSION

I evaluated the effect of different supplements on intake of and preference for medusahead by sheep and determined whether this weed causes a food aversion and if silica has a negative impact on food intake. I found that sheep are constrained in their ability to ingest medusahead and that high-energy supplements have a positive effect on both medusahead intake and preference. My results did not support the hypothesis suggesting that medusahead causes a food aversion. Finally, silica seems to be involved in constraining food intake by sheep.

#### Nutritional Context and Preference for Medusahead

Consumption of single food is not only determined by its intrinsic chemical properties but also by the interaction with other foods (Flaherty, 1996; Provenza *et al.*, 2003). The biochemical context in which unpalatable foods are consumed is critical for enhancing their use and preference. Thus, the specific array of foods encountered and the sequence of encounters could turn out to be crucial in determining an animal's preference for target feeds (Villalba *et al.*, 2006; Bergvall and Balogh, 2009). These contextual effects may occur due to the digestive interaction among feeds, induction, or the interplay between these mechanisms (Provenza *et al.*, 2003; Freidin *et al.*, 2011). Interactions among feeds (i.e., associative effects) are commonly observed in ruminant nutrition studies and can lead to positive associative effects such as increases in diet digestibility (Van Soest, 1994). Sheep and goats eating mixed diets on rangeland display daily intakes much greater (2X) than reference intake values obtained with animals fed single forages of similar nutritive value in confinement (Agreil and Meuret, 2004).

Intake induction or facilitation results when animals repeatedly ingest a less preferred food in association with a highly preferred food. The intake induction effect consists of increased consumption of the low-valued meal relative to controls where animals do not have access to the preferred food (e.g., Flaherty and Grigson, 1988; Freidin *et al.*, 2011, 2012). This induction may be a consequence of animals partially attributing the postingestive effects of the preferred food to the low-palatable food because of the close temporal proximity between both ingestive events (Yearsley *et al.*, 2006). Results from this study are consistent with an induction effect; lambs offered a high-energy supplement (Trials 1-3), a high-protein supplement or a choice of supplements (Trial 3) displayed greater preference for medusahead than nonsupplemented lambs (Control). Lambs in these treatments increased their preference for medusahead, even when the alternative food in the choice test (tall fescue hay) was of greater nutritional quality (table 1).

Previous studies have enticed livestock to consume medusahead through the use of attractants (Doran, 2008; Davy *et al.*, 2009). However, liquid molasses or salt applied directly on medusahead during the late, dry season did not affect use by cattle or sheep (Davy *et al.*, 2009). In another study, molasses applied at different concentrations to medusahead was found to be an ineffective control method (Doran, 2008). Bovey *et al.* (1961) used molasses, beet pulp, and molasses plus beet pulp to enhance medusahead consumption by sheep. Even when sheep were hungry, they rejected the weed which made the authors conclude ".... to date, there appears to be no satisfactory method whereby medusahead can be utilized by animals." It is likely that in these studies the sequence of presentation of palatable and unpalatable feeds (i.e., molasses applied directly to medusahead) led to a "simultaneous negative contrast" as opposed to an induction effect. A simultaneous negative contrast occurs when as a result of comparisons made among foods of different quality, animals show an exaggerated decrease in the intake of the lower quality options (Flaherty, 1996).

Despite the induction effect found in the present study, ingestion of medusahead was low, particularly when medusahead was in the early (Trial 1) and late (Trial 2) reproductive stage. Lambs in the Control treatment ingested strikingly low amounts of medusahead, and in one preference test (Trial 2), intake values by Control lambs were not different from 0. This suggests that during targeted grazing treatments, when stocking densities are high and animals graze without supplements, intake of medusahead may be nil and the use reported in some studies may be just due to trampling effects. Our study, to our knowledge is the first to report actual intake values of medusahead during specific time intervals and across different phenological stages.

Supplementation in the present study had a significant impact on the response of lambs' BW. In contrast, lambs in the Control treatment displayed the lowest ADG (Trial 1), or lost weight (Trials 2-3). Thus, forcing Control animals to consume medusahead had a negative impact on the animals' productivity. Moreover, even when Control lambs were forced to ingest medusahead they never ate more of the weed than supplemented animals. On the contrary, in some instances (e.g., Trials 1 and 2) supplemented animals ate greater amounts of medusahead.

During the late reproductive stage (Trial 2), CP concentration in medusahead decreased whereas fiber concentration increased in the weed relative to the previous feeding period (Trial 1). Lambs responded to this decline in quality by decreasing their intake of medusahead. Nevertheless, concentration of CP was much lower and concentration of fiber much greater for medusahead thatch. Yet, intake and conditioned preferences were the greatest for thatch. Acid insoluble ash was ~30% lower for thatch than for the other phenological stages of medusahead tested. Thus, we hypothesize that this lower concentration of Si explains, at least in part, the greater utilization and induction effects observed for medusahead thatch.

The greater impact of high-energy supplements on medusahead intake and preference by lambs, added to the greater preference for the high-energy supplement displayed by lambs offered a choice of supplements (CHOICE, Trial 2), suggests that medusahead consumption promotes a constraint on the energy metabolism of the ruminant. Silica taken up by forage plants reduces microbial accessibility to cell walls (Smith and Nelson, 1975; Harbers *et al.*, 1981), reducing apparent forage digestibility (Van Soest and Jones, 1968). It has been suggested that *in vitro* dry matter digestibility of forages decreases in 4 units for each unit of Si present in the forage (Mayland and Shewmaker, 2001). Collectively, the greater intake values of medusahead thatch (lower concentration of Si) – despite the low concentrations of CP and high concentrations of fiber in this stage – and positive effects of high-energy supplements on medusahead intake and preference suggest lambs were more constrained in their need for energy rather than protein while consuming the weed. Silica or other antinutritional factors that restrict energy availability to herbivores may be involved in the process.

#### **Medusahead and Food Aversions**

Food aversions in ruminants are triggered by stimulation of the emetic system of the brain stem (Provenza *et al.*, 1994). Upon ingesting a food containing a toxin, afferent impulses to the central nervous system promote malaise, which causes animals to decrease intake of the food associated with the induction of such malaise (Provenza, 1996). The typical food aversion learning paradigm (i.e., Positive Control in the present study) involves offering sheep a specific food for 15-20 min, and immediately after its ingestion, gavaging animals with the toxicant LiCl. This procedure reduces intake of that specific food by sheep on subsequent days (Provenza *et al.*, 1992). Consistent with this, lambs in the Positive Control treatment developed a food aversion to the food (coconutflavored beet pulp) ingested prior to LiCl infusions. However, lambs infused with medusahead or tall fescue hay (Negative Control) did not display such response. Since doses utilized in the study were in line with the amounts of medusahead ingested by lambs I conclude that it is unlikely that the low palatability of medusahead is caused by a conditioned food aversion.

#### Silica and Food Intake

Medusahead contains silicified cell walls with Si forming over 70% of the ash or about 11% of the dry matter of the plant (Bovey *et al.*, 1961). This concentration of Si could not only reduce palatability due to nutrient dilution, but also because Si interferes with mineral metabolism (Van Soest, 1994), and inhibits cell wall degradation and apparent digestibility of herbage to about the same extent as the depression accountable to lignin (Van Soest and Jones, 1968). Silica in grass is composed of three different fractions: opaline silica bodies, silica associated with cell walls, and "free" silica (Blackman, 1968; Blackman and Bailey, 1971). It has been shown in digestibility studies that the proportion of silica loss to the rumen fluid from grass hay incubated in nylon bags ranges from 22 to 39% (Blackman and Bailey, 1971). Considering that silica content in medusahead is about 11% of the dry matter of the plant (Bovey *et al.*, 1961) and that 22 to 39% solubilizes in the rumen, we added soluble silica to alfalfa hay in concentrations of 2.5 and 4.5%.

Lambs fed Si-2.5% displayed the lowest levels of alfalfa hay intake, suggesting that silica constraints food intake in sheep. However, lambs fed silica at 4.5% ate less than lambs fed ground alfalfa (Control) only during the first day of the trial. Thus, a greater concentration of silica did not lead to a proportional reduction in food intake. It is likely that at concentrations of 4.5% silica, lambs attempted to compensate for the dilution of nutrients in alfalfa. Consistent with this, lambs in this treatment ingested the greatest amounts of tall fescue hay. This dilution effect, compounded with the high-quality of alfalfa, and low cell wall binding characteristic of silica, likely attenuated the negative impact of silica on alfalfa intake.

#### CONCLUSIONS

My research suggests that supplementation with a high-energy supplement is a viable option to enhance intake of and preference for medusahead. Even when such enhancement was modest, animals displayed much greater gains of BW than non-supplemented (i.e., forced to ingest medusahead) animals. Intake values of thatch were the greatest, and supplementation led to significant increases in thatch preference. Thus, supplementing thatch during early spring may lead to significant thatch reductions in the plant community which in turn may reduce the likelihood of medusahead establishment and create better environmental conditions for the establishment of perennials in the plant community and/or introduced grasses and legumes during restoration efforts or to prepare for chemical application of herbicides. Finally, these results suggest that food aversions do not explain the low palatability of the weed, which may be better understood by the high concentrations of silica in plant tissues, compounded with the low nutritional quality of the weed. The possibility of post ingested chemical change and rumenal effects of medusahead may be of further interest in understanding intake constraints.

#### REFERENCES

- Agreil C. and M. Meuret. 2004. An improved method for quantifying intake rate and ingestive behaviour of ruminants in diverse and variable habitats using direct observation. *Small Ruminant Res.* 54:99–113.
- A.O.A.C. 2000. Official Methods of Analysis. 17th ed. Assoc. Offic. Anal. Chem., Gaithersburg, MD.
- Bergvall, U. A. and A. C. C. Balogh. 2009.Consummatory simultaneous positive and negative contrast in fallow deer: implications for selectivity. *Mammalian Biol. Zeitschrift fur Saugetierkunde* 74: 236–239.
- Blackman, E. 1968. The pattern and sequence of opaline silica deposition in rye (*Secate Cereale* L.). *Annals of Botany* 32:207–218.
- Blackman, E. and C. Bailey. 1971. Dissolution of silica from dried grass in nylon bags placed in the rumen of a cow. *Canadian J. Anim Sci.* 51:327–332.
- Bovey, R.W., D. LeTourneau, and L.C. Erickson. 1961. The chemical composition of medusahead and downy brome. *Weeds* 9:307–311.
- Coatney, K. 2008. Researchers look at new ways to control medusahead. *California Farm Bureau Federation presentation*.
- Davies, K. W. and D. D. Johnson. 2008. Managing medusahead in the intermountain west is at a critical threshold. *Rangelands* 30:13–15
- Davy, J., Laca, E., and Forero, L. 2009. Medusahead: What is being tested to reduce it? Northern California Ranch Update. University of California Cooperative Extension. Vol. 3. 4 pp.

DiTomaso, J. M., Kyser, G. B., George, M. R., Doran, M. P., and Laca, E. L. 2008. Control of medusahead (*Taeniatherum caput-medusae*) using timely sheep grazing. Invasive *Plant Sci Manage*. 1:241–247.

Doran, M. P. 2008. Luring sheep with molasses. Western SARE Professional Development Program Report. Available at: www.sare.org/content/download/1627/11466/FW06\_304\_txt.pdf. Accessed 26 January 2014.

- Flaherty C. F. 1996. Incentive Relativity. Cambridge University Press, Cambridge.
- Flaherty, C. F. and Grigson, P. S. 1988. From contrast to reinforcement: role of response contingency in anticipatory contrast. J. Experimental Psychology: Animal Behavioral Processes14:165–176.
- Freidin E., F. Catanese, N. Didone, and R.A. Distel. 2011. Mechanisms of intake induction of a low-nutritious food in sheep (*Ovis aries*). *Behavioural Processes* 87:246–252.
- Freidin E., F. Catanese, M. I. Cuello, and R. A. Distel. 2012. Induction of low-nutritious food intake by subsequent nutrient supplementation in sheep (*Ovis aries*). *Animal* 6:1307–1315.
- Harbers, L. H., D. J. Raiten, and G. M. Paulsen.1981. The role of plant epidermal silica as a structural inhibitor of rumen microbial digestion in steers. Nutr. Rep. Int. 24:1057–1066.
- Hobbs, N. T. 1996. Modification of ecosystems by ungulates. *J. Wildlife Manage*. 60:695–713.

- Mayland, H. F. and G. E. Shewmaker. 2001. Animal health problems caused by silicon and other mineral imbalances. *J. Range Manage*. 54:441–446.
- McNaughton, S. J., J. L. Tarrants, M. M. Mcnaughton, and R. H. Davis. 1985. Silica as a defense against herbivore and a growth promoter in African grasses. *Ecology* 66:528–535.
- Murphy, A. H. and Turner, D. 1959. A study on the germination of medusahead seed. *The Bulletin.* California State Department of Agriculture. 48:6–10.
- Provenza, F. D., J. A. Pfister, and C. D. Cheney. 1992. Mechanisms of learning in diet selection with reference to phytotoxicosis in herbivores. *J. Range Manage*. 45:36–45.
- Provenza, F. D., L. Ortega-Reyes, C. B. Scott, J. J. Lynch, and E. A. Burritt. 1994.Antiemetic drugs attenuate food aversions in sheep. *J. Anim Sci.* 72:1989–1994.
- Provenza, F. D. 1995. Postingestive feedback as an elementary determinant of food preference and intake in ruminants. *J. Range Manage*. 48: 2–17.
- Provenza, F. D. 1996. Acquired aversions as the basis for varied diets of ruminants foraging on rangelands. J. Anim Sci. 74:2010–2020.
- Provenza, F. D., J. J. Villalba, L. E. Dziba, S. B. Atwood, and R. E. Banner. 2003. Linking herbivore experience, varied diets, and plant biochemical diversity. *Small Ruminant Res.* 49:257–274.
- Smith, G. S. and A. B. Nelson. 1975. Effects of sodium silicate added to rumen cultures on forage digestion, with interactions of glucose, urea and minerals. *J. Animal Sci.* 41:891–899.

- Swenson, C. F., D. LeTourneau, and L. C. Erickson. 1964. Silica in medusahead. *Weeds* 12:16–18.
- Van Keulen, J. and Young, B. A. 1977. Evaluation of acid-insoluble ash as a natural marker in ruminant digestibility studies. *J. Anim Sci.* 44:282–287.
- Van Soest, P. J. and L. H. P. Jones. 1968. Effect of silica in forages upon digestibility. J. Dairy Sci. 51:1644–1648.
- Van Soest, P. J., J. B. Robertson, and B.A. Lewis. 1991. Methods for Dietary Fiber, Neutral Detergent Fiber, and Nonstarch Polysaccharides in Relation to Animal Nutrition. J. of Dairy Sci. 74:3583–3597.
- Van Soest, P. J. 1994. Nutritional Ecology of the Ruminant. 2<sup>nd</sup> Ed. Cornell University, Ithaca, NY.
- Van Soest, P. J. 2006. Rice straw, the role of silica and treatments to improve quality. J. Anim. Feed Sci. Technol. 130:137–171.
- Villalba J. J., F. D. Provenza, and R. Shaw. 2006. Initial conditions and temporal delays influence preference for foods high in tannins and for foraging locations with and without foods high in tannins by sheep. *App. Animal Behav. Sci.* 97:190–205.
- Yearsley J. M., J. J. Villalba, I. J. Gordon, I. Kyriazakis, J. R. Speakman, B. J. Tolkamp, A. W. Illius, and A. Duncan. 2006. A theory of associating food types with their post-ingestive consequences. *Am. Naturalist* 167:705–716.
- Zimmerman, J., W. S. Johnson, and M. E. Eiswerth. 2002. Medusahead: Economic Impact and Control in Nevada. University of Nevada Extension Service. Reno, Nevada. Fact Sheet FS-02-37.

APPENDIX

### APPENDIX

# **SAS Outputs**

### **Trial 1 2012**

## Total Medusahead intake

Type 3 Tests of Fixed Effects									
Effect	Num DF	Num DF Den DF F Value Pr > I							
Group	3	28	1.28	0.2999					
Day	9	252	8.86	<.0001					
Group*Day	27	252	1.34	0.1293					

# **Total Supplement Intake**

Type 3 Tests of Fixed Effects							
Effect	Num DF	Den DF	F Value	Pr > F			
Group	2	21	4.82	0.0189			
Day	9	189	11.40	<.0001			
Group*Day	18	189	2.59	0.0007			

Least Squares Means								
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t	
Group	Choice		32.4096	2.1712	21	14.93	<.0001	
Group	HE		23.1959	2.1664	21	10.71	<.0001	
Group	HP		26.1216	2.1712	21	12.03	<.0001	
Group*Day	Choice	1	35.9853	2.7429	189	13.12	<.0001	
Group*Day	Choice	2	33.8764	2.7429	189	12.35	<.0001	
Group*Day	Choice	3	40.6741	2.7429	189	14.83	<.0001	

			Least Squ	ares Means			
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t
Group*Day	Choice	4	31.7049	2.7429	189	11.56	<.0001
Group*Day	Choice	5	28.7492	2.7429	189	10.48	<.0001
Group*Day	Choice	6	20.7703	2.7429	189	7.57	<.0001
Group*Day	Choice	7	31.1160	2.7429	189	11.34	<.0001
Group*Day	Choice	8	34.0608	2.7429	189	12.42	<.0001
Group*Day	Choice	9	33.0304	2.7429	189	12.04	<.0001
Group*Day	Choice	10	34.1286	2.7429	189	12.44	<.0001
Group*Day	HE	1	22.3151	2.7391	189	8.15	<.0001
Group*Day	HE	2	21.1282	2.7391	189	7.71	<.0001
Group*Day	HE	3	27.7755	2.7391	189	10.14	<.0001
Group*Day	HE	4	23.8495	2.7391	189	8.71	<.0001
Group*Day	HE	5	21.8649	2.7391	189	7.98	<.0001
Group*Day	HE	6	17.2214	2.7391	189	6.29	<.0001
Group*Day	HE	7	22.6432	2.7391	189	8.27	<.0001
Group*Day	HE	8	25.3924	2.7391	189	9.27	<.0001
Group*Day	HE	9	24.4636	2.7391	189	8.93	<.0001
Group*Day	HE	10	25.3058	2.7391	189	9.24	<.0001
Group*Day	HP	1	23.5869	2.7429	189	8.60	<.0001
Group*Day	HP	2	24.0297	2.7429	189	8.76	<.0001
Group*Day	HP	3	29.2739	2.7429	189	10.67	<.0001
Group*Day	HP	4	27.3257	2.7429	189	9.96	<.0001
Group*Day	HP	5	23.3902	2.7429	189	8.53	<.0001
Group*Day	HP	6	20.3033	2.7429	189	7.40	<.0001
Group*Day	HP	7	35.4129	2.7429	189	12.91	<.0001
Group*Day	HP	8	26.0520	2.7429	189	9.50	<.0001
Group*Day	HP	9	25.4728	2.7429	189	9.29	<.0001
Group*Day	HP	10	26.3687	2.7429	189	9.61	<.0001

# Total Tall Fescue Hay Intake

Type 3 Tests of Fixed Effects									
Effect	Num DF	Num DF Den DF F Value Pr > I							
Group	3 28 0.80 0.5								
Day	9 252 39.31 <.000								
Group*Day	27	252	0.45	0.9926					

# **Group Choice Supplement Preference**

Type 3 Tests of Fixed Effects									
Effect	Num DF	Num DF Den DF F Value Pr >							
Food	1	14	0.13	0.7202					
Day	9 126 3.93 0.00								
Food*Day	9	126	0.84	0.5786					

	Least Squares Means								
Effect	Food	Day	Estimate	Standard Error	DF	t Value	$\Pr >  t $		
Food	HE		15.9821	1.5853	14	10.08	<.0001		
Food	HPro		16.8096	1.5853	14	10.60	<.0001		
Day		1	18.1837	1.6611	126	10.95	<.0001		
Day		2	17.1292	1.6611	126	10.31	<.0001		
Day		3	20.5281	1.6611	126	12.36	<.0001		
Day		4	16.0435	1.6611	126	9.66	<.0001		
Day		5	14.5656	1.6611	126	8.77	<.0001		
Day		6	10.5761	1.6611	126	6.37	<.0001		
Day		7	15.7490	1.6611	126	9.48	<.0001		
Day		8	17.2214	1.6611	126	10.37	<.0001		
Day		9	16.7062	1.6611	126	10.06	<.0001		
Day		10	17.2553	1.6611	126	10.39	<.0001		
Food*Day	HE	1	19.2790	2.3597	126	8.17	<.0001		

			Least Sq	uares Means			
Effect	Food	Day	Estimate	Standard Error	DF	t Value	Pr >  t
Food*Day	HE	2	15.7953	2.3597	126	6.69	<.0001
Food*Day	HE	3	20.0592	2.3597	126	8.50	<.0001
Food*Day	HE	4	14.6945	2.3597	126	6.23	<.0001
Food*Day	HE	5	14.0149	2.3597	126	5.94	<.0001
Food*Day	HE	6	10.8618	2.3597	126	4.60	<.0001
Food*Day	HE	7	14.1873	2.3597	126	6.01	<.0001
Food*Day	HE	8	17.5335	2.3597	126	7.43	<.0001
Food*Day	HE	9	18.2229	2.3597	126	7.72	<.0001
Food*Day	HE	10	15.1722	2.3597	126	6.43	<.0001
Food*Day	HPro	1	17.0883	2.3597	126	7.24	<.0001
Food*Day	HPro	2	18.4631	2.3597	126	7.82	<.0001
Food*Day	HPro	3	20.9969	2.3597	126	8.90	<.0001
Food*Day	HPro	4	17.3924	2.3597	126	7.37	<.0001
Food*Day	HPro	5	15.1164	2.3597	126	6.41	<.0001
Food*Day	HPro	6	10.2905	2.3597	126	4.36	<.0001
Food*Day	HPro	7	17.3107	2.3597	126	7.34	<.0001
Food*Day	HPro	8	16.9094	2.3597	126	7.17	<.0001
Food*Day	HPro	9	15.1895	2.3597	126	6.44	<.0001
Food*Day	HPro	10	19.3384	2.3597	126	8.20	<.0001

Total Intake (Medusahead + Tall fescue Hay + Supplement)

Type 3 Tests of Fixed Effects									
Effect	Num DF	Num DF Den DF F Value Pr >							
Group	3	28	17.63	<.0001					
Day	9 252 11.44 <.00								
Group*Day	27	252	2.09	0.0019					

			Least Squa	ares Means			
Effect	Group	Day	Estimate	Standard Error	DF	t Value	$\Pr >  t $
Group	Choice		52.7478	3.2469	28	16.25	<.0001
Group	Control		21.9433	3.2241	28	6.81	<.0001
Group	HE		42.2807	3.2241	28	13.11	<.0001
Group	HP		44.8663	3.2469	28	13.82	<.0001
Group*Day	Choice	1	49.1762	3.7528	252	13.10	<.0001
Group*Day	Choice	2	56.1528	3.7528	252	14.96	<.0001
Group*Day	Choice	3	58.8177	3.7528	252	15.67	<.0001
Group*Day	Choice	4	52.5866	3.7528	252	14.01	<.0001
Group*Day	Choice	5	46.6011	3.7528	252	12.42	<.0001
Group*Day	Choice	6	41.8239	3.7528	252	11.14	<.0001
Group*Day	Choice	7	53.5967	3.7528	252	14.28	<.0001
Group*Day	Choice	8	57.8208	3.7528	252	15.41	<.0001
Group*Day	Choice	9	55.3044	3.7528	252	14.74	<.0001
Group*Day	Choice	10	55.5981	3.7528	252	14.82	<.0001
Group*Day	Control	1	18.0069	3.7331	252	4.82	<.0001
Group*Day	Control	2	22.8546	3.7331	252	6.12	<.0001
Group*Day	Control	3	20.3058	3.7331	252	5.44	<.0001
Group*Day	Control	4	20.0197	3.7331	252	5.36	<.0001
Group*Day	Control	5	20.6359	3.7331	252	5.53	<.0001
Group*Day	Control	6	22.7460	3.7331	252	6.09	<.0001
Group*Day	Control	7	24.6136	3.7331	252	6.59	<.0001
Group*Day	Control	8	24.1373	3.7331	252	6.47	<.0001
Group*Day	Control	9	23.8649	3.7331	252	6.39	<.0001
Group*Day	Control	10	22.2485	3.7331	252	5.96	<.0001
Group*Day	HE	1	35.4397	3.7331	252	9.49	<.0001
Group*Day	HE	2	39.5064	3.7331	252	10.58	<.0001

			Least Squa	ares Means			
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t
Group*Day	HE	3	42.9690	3.7331	252	11.51	<.0001
Group*Day	HE	4	41.1499	3.7331	252	11.02	<.0001
Group*Day	HE	5	41.3757	3.7331	252	11.08	<.0001
Group*Day	HE	6	38.2324	3.7331	252	10.24	<.0001
Group*Day	HE	7	44.3649	3.7331	252	11.88	<.0001
Group*Day	HE	8	47.4745	3.7331	252	12.72	<.0001
Group*Day	HE	9	47.0145	3.7331	252	12.59	<.0001
Group*Day	HE	10	45.2801	3.7331	252	12.13	<.0001
Group*Day	HP	1	37.3652	3.7528	252	9.96	<.0001
Group*Day	HP	2	41.8948	3.7528	252	11.16	<.0001
Group*Day	HP	3	44.6510	3.7528	252	11.90	<.0001
Group*Day	HP	4	45.3091	3.7528	252	12.07	<.0001
Group*Day	HP	5	42.5771	3.7528	252	11.35	<.0001
Group*Day	HP	6	40.6816	3.7528	252	10.84	<.0001
Group*Day	HP	7	53.8242	3.7528	252	14.34	<.0001
Group*Day	HP	8	47.1754	3.7528	252	12.57	<.0001
Group*Day	HP	9	48.4832	3.7528	252	12.92	<.0001
Group*Day	HP	10	46.7010	3.7528	252	12.44	<.0001

### **Average Weight Gains**

	Type 3 Tests of Fixed Effects								
Effect	Effect Num DF Den DF F Value $Pr > F$								
Group	Group 3 28 9.46 0.0002								

	Least Squares Means									
Effect	Group	Estimate	Standard Error	DF	t Value	Pr >  t				
Group	Choice	0.4864	0.05059	28	9.61	<.0001				
Group	Control	0.1136	0.05059	28	2.25	0.0328				
Group	HE	0.2511	0.05059	28	4.96	<.0001				
Group	HP	0.2375	0.05059	28	4.69	<.0001				

### **Choice Between Medusahead and Tall Fescue Hay**

Type 3 Tests of Fixed Effects									
Effect	Num DF Den DF F Value Pr > 1								
Group	3	28	1.79	0.1711					
Food	1	28	505.17	<.0001					
Group*Food	3	28	1.89	0.1543					

### **Preference for Medusahead**

Type 3 Tests of Fixed Effects								
Effect	fect Num DF Den DF F Value $Pr > F$							
Group	Group 3 28 1.22 0.3215							

### **Trial 2 2012**

Type 3 Tests of Fixed Effects									
Effect	Num DF Den DF F Value Pr >								
Group	3	28	0.70	0.5579					
Day	8	224	12.04	<.0001					
Group*Day	24	224	0.88	0.6285					

### **Total Medusahead Intake**

# **Total Supplement Intake**

Type 3 Tests of Fixed Effects									
Effect	ct Num DF Den DF F Value $Pr > F$								
Group	3	28	296.41	<.0001					
Day	8	224	6.29	<.0001					
Group*Day	24	224	2.99	<.0001					

	Least Squares Means									
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t			
Group	Choice		30.6929	0.7519	28	40.82	<.0001			
Group	Control		0.03481	0.7514	28	0.05	0.9634			
Group	HE		22.1064	0.7514	28	29.42	<.0001			
Group	HP		20.9338	0.7519	28	27.84	<.0001			
Group*Day	Choice	1	28.0337	1.0696	224	26.21	<.0001			
Group*Day	Choice	2	27.5946	1.0696	224	25.80	<.0001			
Group*Day	Choice	3	30.5708	1.0696	224	28.58	<.0001			
Group*Day	Choice	4	28.6467	1.0696	224	26.78	<.0001			
Group*Day	Choice	5	34.1615	1.0696	224	31.94	<.0001			

Group*Day C	Choice Choice	6	28.3395	1.0696	224	26.50	<.0001
1 2	Choice						
		7	32.8157	1.0696	224	30.68	<.0001
Group*Day C	Choice	8	30.4910	1.0696	224	28.51	<.0001
Group*Day C	Choice	9	35.5828	1.0696	224	33.27	<.0001
Group*Day C	Control	1	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	2	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	3	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	4	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	5	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	6	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	7	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	8	0.03481	1.0693	224	0.03	0.9741
Group*Day C	Control	9	0.03481	1.0693	224	0.03	0.9741
Group*Day H	HE	1	19.9976	1.0693	224	18.70	<.0001
Group*Day H	HE	2	21.8328	1.0693	224	20.42	<.0001
Group*Day H	HE	3	22.4655	1.0693	224	21.01	<.0001
Group*Day H	HE	4	21.9905	1.0693	224	20.57	<.0001
Group*Day H	HE	5	23.0414	1.0693	224	21.55	<.0001
Group*Day H	HE	6	22.2819	1.0693	224	20.84	<.0001
Group*Day H	HE	7	23.6753	1.0693	224	22.14	<.0001
Group*Day H	HE	8	22.0811	1.0693	224	20.65	<.0001
Group*Day H	HE	9	21.5913	1.0693	224	20.19	<.0001
Group*Day H	HP	1	19.7748	1.0696	224	18.49	<.0001
Group*Day H	HP	2	19.9492	1.0696	224	18.65	<.0001
Group*Day H	HP	3	20.5664	1.0696	224	19.23	<.0001
Group*Day H	HP	4	20.6786	1.0696	224	19.33	<.0001
Group*Day H	HP	5	21.6427	1.0696	224	20.24	<.0001
Group*Day H	HP	6	21.6759	1.0696	224	20.27	<.0001
Group*Day H	HP	7	21.4569	1.0696	224	20.06	<.0001

Group*Day	HP	8	21.0808	1.0696	224	19.71	<.0001
Group*Day	HP	9	21.5787	1.0696	224	20.18	<.0001

# **Total Tall Fescue Hay Intake**

Type 3 Tests of Fixed Effects									
Effect	Num DF Den DF F Value $Pr > F$								
Group	3	28	1.07	0.3767					
Day	8	224	29.42	<.0001					
Group*Day	24	224	1.95	0.0066					

			Least Squa	ares Means			
Effect	Group	Day	Estimate	Standard Error	DF	t Value	$\Pr >  t $
Group	Choice		18.2440	0.6830	28	26.71	<.0001
Group	Control		18.3999	0.6830	28	26.94	<.0001
Group	HE		16.8612	0.6830	28	24.69	<.0001
Group	HP		18.1215	0.6830	28	26.53	<.0001
Group*Day	Choice	1	16.9217	0.8653	224	19.56	<.0001
Group*Day	Choice	2	16.6549	0.8653	224	19.25	<.0001
Group*Day	Choice	3	18.2554	0.8653	224	21.10	<.0001
Group*Day	Choice	4	18.7053	0.8653	224	21.62	<.0001
Group*Day	Choice	5	18.7800	0.8653	224	21.70	<.0001
Group*Day	Choice	6	18.3688	0.8653	224	21.23	<.0001
Group*Day	Choice	7	18.8648	0.8653	224	21.80	<.0001
Group*Day	Choice	8	18.8576	0.8653	224	21.79	<.0001
Group*Day	Choice	9	18.7875	0.8653	224	21.71	<.0001
Group*Day	Control	1	16.0922	0.8653	224	18.60	<.0001
Group*Day	Control	2	14.9774	0.8653	224	17.31	<.0001
Group*Day	Control	3	16.1896	0.8653	224	18.71	<.0001
Group*Day	Control	4	20.5178	0.8653	224	23.71	<.0001

			Least Squa	ares Means			
Effect	Group	Day	Estimate	Standard Error	DF	t Value	$Pr > \left  t \right $
Group*Day	Control	5	19.3976	0.8653	224	22.42	<.0001
Group*Day	Control	6	17.7036	0.8653	224	20.46	<.0001
Group*Day	Control	7	20.4805	0.8653	224	23.67	<.0001
Group*Day	Control	8	20.7803	0.8653	224	24.02	<.0001
Group*Day	Control	9	19.4600	0.8653	224	22.49	<.0001
Group*Day	HE	1	16.1935	0.8653	224	18.71	<.0001
Group*Day	HE	2	14.5979	0.8653	224	16.87	<.0001
Group*Day	HE	3	16.2766	0.8653	224	18.81	<.0001
Group*Day	HE	4	18.2352	0.8653	224	21.07	<.0001
Group*Day	HE	5	16.9483	0.8653	224	19.59	<.0001
Group*Day	HE	6	16.3784	0.8653	224	18.93	<.0001
Group*Day	HE	7	18.3509	0.8653	224	21.21	<.0001
Group*Day	HE	8	17.9255	0.8653	224	20.72	<.0001
Group*Day	HE	9	16.8446	0.8653	224	19.47	<.0001
Group*Day	HP	1	15.3315	0.8653	224	17.72	<.0001
Group*Day	HP	2	14.5060	0.8653	224	16.76	<.0001
Group*Day	HP	3	17.1109	0.8653	224	19.77	<.0001
Group*Day	HP	4	19.7558	0.8653	224	22.83	<.0001
Group*Day	HP	5	19.6188	0.8653	224	22.67	<.0001
Group*Day	HP	6	18.0260	0.8653	224	20.83	<.0001
Group*Day	HP	7	19.7339	0.8653	224	22.81	<.0001
Group*Day	HP	8	19.7492	0.8653	224	22.82	<.0001
Group*Day	HP	9	19.2617	0.8653	224	22.26	<.0001

# **Group Choice Supplement Preference**

Ty	Type 3 Tests of Fixed Effects									
Effect	ffectNum DFDen DFF Value $Pr > F$									
Food	1	14	5.06	0.0410						
Day	8	112	2.18	0.0337						
Food*Day	8	112	1.78	0.0882						

			Least Sq	uares Means			
Effect	Food	Day	Estimate	Standard Error	DF	t Value	Pr >  t
Food	HE		18.2720	0.9591	14	19.05	<.0001
Food	HPro		15.1906	0.9591	14	15.84	<.0001
Food*Day	HE	1	16.1988	1.7168	112	9.44	<.0001
Food*Day	HE	2	17.5324	1.7168	112	10.21	<.0001
Food*Day	HE	3	17.3211	1.7168	112	10.09	<.0001
Food*Day	HE	4	14.5263	1.7168	112	8.46	<.0001
Food*Day	HE	5	19.6796	1.7168	112	11.46	<.0001
Food*Day	HE	6	18.2821	1.7168	112	10.65	<.0001
Food*Day	HE	7	20.4999	1.7168	112	11.94	<.0001
Food*Day	HE	8	20.1091	1.7168	112	11.71	<.0001
Food*Day	HE	9	20.2988	1.7168	112	11.82	<.0001
Food*Day	HPro	1	14.3607	1.7168	112	8.36	<.0001
Food*Day	HPro	2	12.5478	1.7168	112	7.31	<.0001
Food*Day	HPro	3	16.0082	1.7168	112	9.32	<.0001
Food*Day	HPro	4	16.7025	1.7168	112	9.73	<.0001
Food*Day	HPro	5	17.5697	1.7168	112	10.23	<.0001
Food*Day	HPro	6	12.6113	1.7168	112	7.35	<.0001
Food*Day	HPro	7	15.2801	1.7168	112	8.90	<.0001
Food*Day	HPro	8	13.1331	1.7168	112	7.65	<.0001
Food*Day	HPro	9	18.5022	1.7168	112	10.78	<.0001

Type 3 Tests of Fixed Effects							
Effect	Num DF	Den DF	F Value	Pr > F			
Group	3	28	119.02	<.0001			
Day	8	224	24.92	<.0001			
Group*Day	24	224	1.43	0.0951			

# Total Intake (Medusahead + Tall fescue Hay + Supplement)

	Least Squares Means								
Effect Group Day			Estimate	Standard Error	DF	t Value	Pr >  t		
Group	Choice		50.4724	1.1893	28	42.44	<.0001		
Group	Control		19.6164	1.1882	28	16.51	<.0001		
Group	HE		41.4091	1.1882	28	34.85	<.0001		
Group	HP		40.6161	1.1893	28	34.15	<.0001		
Group*Day	Choice	1	46.7199	1.6093	224	29.03	<.0001		
Group*Day	Choice	2	45.6831	1.6093	224	28.39	<.0001		
Group*Day	Choice	3	49.8744	1.6093	224	30.99	<.0001		
Group*Day	Choice	4	47.9882	1.6093	224	29.82	<.0001		
Group*Day	Choice	5	53.8159	1.6093	224	33.44	<.0001		
Group*Day	Choice	6	48.5609	1.6093	224	30.18	<.0001		
Group*Day	Choice	7	53.1767	1.6093	224	33.04	<.0001		
Group*Day	Choice	8	51.4878	1.6093	224	31.99	<.0001		
Group*Day	Choice	9	56.9449	1.6093	224	35.39	<.0001		
Group*Day	Control	1	16.8579	1.6085	224	10.48	<.0001		
Group*Day	Control	2	15.7306	1.6085	224	9.78	<.0001		
Group*Day	Control	3	16.8254	1.6085	224	10.46	<.0001		
Group*Day	Control	4	20.7777	1.6085	224	12.92	<.0001		
Group*Day	Control	5	19.6829	1.6085	224	12.24	<.0001		
Group*Day	Control	6	18.7285	1.6085	224	11.64	<.0001		

			Least Squa	ares Means			
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t
Group*Day	Control	7	21.6981	1.6085	224	13.49	<.0001
Group*Day	Control	8	23.0071	1.6085	224	14.30	<.0001
Group*Day	Control	9	23.2399	1.6085	224	14.45	<.0001
Group*Day	HE	1	38.8626	1.6085	224	24.16	<.0001
Group*Day	HE	2	38.5062	1.6085	224	23.94	<.0001
Group*Day	HE	3	40.2536	1.6085	224	25.03	<.0001
Group*Day	HE	4	41.6503	1.6085	224	25.89	<.0001
Group*Day	HE	5	41.4016	1.6085	224	25.74	<.0001
Group*Day	HE	6	40.8612	1.6085	224	25.40	<.0001
Group*Day	HE	7	45.1179	1.6085	224	28.05	<.0001
Group*Day	HE	8	42.6562	1.6085	224	26.52	<.0001
Group*Day	HE	9	43.3721	1.6085	224	26.96	<.0001
Group*Day	HP	1	35.9902	1.6093	224	22.36	<.0001
Group*Day	HP	2	35.5855	1.6093	224	22.11	<.0001
Group*Day	HP	3	38.4474	1.6093	224	23.89	<.0001
Group*Day	HP	4	40.6096	1.6093	224	25.23	<.0001
Group*Day	HP	5	41.7179	1.6093	224	25.92	<.0001
Group*Day	HP	6	40.7385	1.6093	224	25.31	<.0001
Group*Day	HP	7	42.7094	1.6093	224	26.54	<.0001
Group*Day	HP	8	42.6682	1.6093	224	26.51	<.0001
Group*Day	HP	9	47.0782	1.6093	224	29.25	<.0001

# **Average Weight Gains**

	Type 3 Tests of Fixed Effects								
Effect	Effect Num DF Den DF F Value $Pr > F$								
Group	Group 3 28 5.21 0.0055								

	Least Squares Means								
Effect	Group	Estimate	Standard Error	DF	t Value	$Pr > \left  t \right $			
Group	Choice	0.1023	0.05383	28	1.90	0.0678			
Group	Control	-0.06705	0.05383	28	-1.25	0.2233			
Group	HE	0.1193	0.05383	28	2.22	0.0350			
Group	HP	0.2307	0.05383	28	4.29	0.0002			

# Choice between Medusahead and Tall Fescue Hay

Type 3 Tests of Fixed Effects								
Effect Num DF Den DF F Value $Pr > F$								
Group 3 28 1.77 0.1760								

	Least Squares Means								
Effect	Group	Estimate	Standard Error	DF	t Value	$Pr > \left  t \right $			
Group	CHOICE	2.9207	0.8744	28	3.34	0.0024			
Group	CONTROL	0.5840	0.8744	28	0.67	0.5097			
Group	HiE	3.1544	0.8744	28	3.61	0.0012			
Group	HiPro	2.0693	0.8744	28	2.37	0.0251			

	Least Squares Means								
Effect	Group	Estimate	Standard Error	DF	t Value	$Pr > \left  t \right $			
Group	CHOICE	5.3855	1.7686	28	3.05	0.0050			
Group	CONTROL	1.2332	1.7686	28	0.70	0.4914			
Group	HiE	7.4068	1.7686	28	4.19	0.0003			
Group	HiPro	3.9685	1.7686	28	2.24	0.0329			

#### **Trial 3 2013**

### **Total Medusahead Intake**

Type 3 Tests of Fixed Effects							
Effect	Num DF	Den DF	F Value	Pr > F			
Group	3	28	0.14	0.9358			
Day	9	252	32.94	<.0001			
Group*Day	27	252	1.15	0.2883			

# **Total Supplement Intake**

Type 3 Tests of Fixed Effects							
Effect	Num DF	Den DF	F Value	Pr > F			
Group	2	21	6.08	0.0083			
Day	9	189	11.35	<.0001			
Group*Day	18	189	1.69	0.0437			

	Least Squares Means							
Effect	Group	Estimate	Standard Error	DF	t Value	Pr >  t		
Group	Choice	32.4242	2.3767	21	13.64	<.0001		
Group	HE	23.2933	2.3767	21	9.80	<.0001		
Group	HP	21.4987	2.3767	21	9.05	<.0001		

# **Total Tall Fescue Hay Intake**

Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F						
Group	3	28	1.11	0.3604						
Day	9	252	22.90	<.0001						
Group*Day	27	252	1.04	0.4190						

# **Group Choice Supplement Preference**

Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F						
Food	1	14	0.26	0.6195						
Day	9	126	3.15	0.0018						
Food*Day	9	126	0.75	0.6652						

	Least Squares Means											
Effect	Food	Estimate	Standard Error	DF	t Value	Pr >  t						
Food	Choice HE	15.4456	2.1345	14	7.24	<.0001						
Food	Choice HP	16.9786	2.1345	14	7.95	<.0001						

Ту	Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F							
Group	3	28	20.14	<.0001							
Day	9	252	115.56	<.0001							
Group*Day	27	252	1.19	0.2421							

# Total Intake (Medusahead + Tall fescue Hay + Supplement)

F

	Least Squares Means											
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t					
Group	Choice		57.4700	3.1092	28	18.48	<.0001					
Group	Control		25.2591	3.0975	28	8.15	<.0001					
Group	HE		48.2738	3.0975	28	15.58	<.0001					
Group	HP		44.7368	3.1092	28	14.39	<.0001					

# Average Weight Gains

	Type 3 Tests of Fixed Effects								
Effect	Effect Num DF Den DF F Value $Pr > F$								
Group	3	28	3.42	0.0308					

	Least Squares Means											
Effect	Group	Estimate	Standard Error	DF	t Value	$Pr > \left  t \right $						
Group	CHOICE	0.3169	0.07779	28	4.07	0.0003						
Group	Control	-0.00882	0.07779	28	-0.11	0.9105						
Group	HE	0.2478	0.07779	28	3.19	0.0035						
Group	HP	0.2500	0.07779	28	3.21	0.0033						

Type 3 Tests of Fixed Effects								
Effect Num DF Den DF F Value Pr > F								
Group	3	28	6.13	0.0024				

### **Choice Between Medusahead and Tall Fescue Hay**

	Least Squares Means										
Effect	Group	Estimate	Standard Error	DF	t Value	$\Pr >  t $					
Group	СН	15.5831	1.9770	28	7.88	<.0001					
Group	CTRL	3.6809	1.9770	28	1.86	0.0731					
Group	HE	9.3625	1.9770	28	4.74	<.0001					
Group	HP	8.4154	1.9770	28	4.26	0.0002					

### **Trial 4 Aversion**

# Intake of Coconut Flavored Beet Pulp

Ту	Type 3 Tests of Fixed Effects									
Effect	Num DF	Den DF	F Value	Pr > F						
Group	2	27	9.46	0.0008						
Day	2	54	18.91	<.0001						
Group*Day	4	54	2.86	0.0319						

	Least Squares Means									
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t			
Group	G-Hay		15.6249	0.9712	27	16.09	<.0001			
Group	LiCl		10.3774	0.9700	27	10.70	<.0001			
Group	Medusa		15.5486	0.9712	27	16.01	<.0001			
Day		1	14.8973	0.7895	54	18.87	<.0001			

	Least Squares Means									
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t			
Day		2	10.4827	0.7895	54	13.28	<.0001			
Day		3	16.1709	0.7895	54	20.48	<.0001			
Group*Day	G-Hay	1	14.7614	1.3732	54	10.75	<.0001			
Group*Day	G-Hay	2	13.6135	1.3732	54	9.91	<.0001			
Group*Day	G-Hay	3	18.4999	1.3732	54	13.47	<.0001			
Group*Day	LiCl	1	13.5190	1.3724	54	9.85	<.0001			
Group*Day	LiCl	2	4.8152	1.3724	54	3.51	0.0009			
Group*Day	LiCl	3	12.7982	1.3724	54	9.33	<.0001			
Group*Day	Medusa	1	16.4116	1.3732	54	11.95	<.0001			
Group*Day	Medusa	2	13.0196	1.3732	54	9.48	<.0001			
Group*Day	Medusa	3	17.2146	1.3732	54	12.54	<.0001			

### Total Alfalfa Pellet Intake

Type 3 Tests of Fixed Effects									
Effect	Num DF	Den DF	F Value	Pr > F					
Group	2	27	9.10	0.0010					

	Least Squares Means											
Effect	Group	Estimate	Standard Error	DF	t Value	Pr >  t						
Group	G-Hay	15.5249	0.9481	27	16.37	<.0001						
Group	LiCl	10.5270	0.9481	27	11.10	<.0001						
Group	Medusa	15.4362	0.9481	27	16.28	<.0001						

Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F						
Group	2	27	0.11	0.8992						
Day	4	108	15.20	<.0001						
Group*Day	8	108	0.53	0.8351						

### Alfafla Pellet Intake Prior to adding Silica

# Alfafla Pellet Intake After Adding Silica

Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F						
Group	2	27	6.36	0.0055						
Day	4	108	17.68	<.0001						
Group*Day	8	108	2.10	0.0421						

	Least Squares Means									
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t			
Group	2.5 Silica		85.4350	2.8955	27	29.51	<.0001			
Group	4.5 Silica		95.3075	2.8924	27	32.95	<.0001			
Group	Alfalfa		99.6952	2.8955	27	34.43	<.0001			
Day		1	84.3545	2.1437	108	39.35	<.0001			
Day		2	90.2796	2.1437	108	42.11	<.0001			
Day		3	94.0114	2.1437	108	43.86	<.0001			
Day		4	99.6814	2.1437	108	46.50	<.0001			
Day		5	99.0693	2.1437	108	46.21	<.0001			
Group*Day	2.5 Silica	1	77.8856	3.7325	108	20.87	<.0001			
Group*Day	2.5 Silica	2	81.3037	3.7325	108	21.78	<.0001			

	Least Squares Means									
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t			
Group*Day	2.5 Silica	3	84.7797	3.7325	108	22.71	<.0001			
Group*Day	2.5 Silica	4	91.7653	3.7325	108	24.59	<.0001			
Group*Day	2.5 Silica	5	91.4408	3.7325	108	24.50	<.0001			
Group*Day	4.5 Silica	1	79.1172	3.7301	108	21.21	<.0001			
Group*Day	4.5 Silica	2	93.1892	3.7301	108	24.98	<.0001			
Group*Day	4.5 Silica	3	97.5478	3.7301	108	26.15	<.0001			
Group*Day	4.5 Silica	4	102.48	3.7301	108	27.47	<.0001			
Group*Day	4.5 Silica	5	104.21	3.7301	108	27.94	<.0001			
Group*Day	Alfalfa	1	96.0605	3.7325	108	25.74	<.0001			
Group*Day	Alfalfa	2	96.3460	3.7325	108	25.81	<.0001			
Group*Day	Alfalfa	3	99.7067	3.7325	108	26.71	<.0001			
Group*Day	Alfalfa	4	104.80	3.7325	108	28.08	<.0001			
Group*Day	Alfalfa	5	101.56	3.7325	108	27.21	<.0001			

Tall Fescue Hay Intake Prior to Adding Silica

Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F						
Group	2	27	0.40	0.6761						
Day	4	108	6.17	0.0002						
Group*Day	8	108	1.15	0.3393						

# Tall Fescue Hay Intake After Adding Silica

Type 3 Tests of Fixed Effects										
Effect	Num DF	Den DF	F Value	Pr > F						
Group	2	27	5.12	0.0131						
Day	4	108	6.34	0.0001						
Group*Day	8	108	3.69	0.0008						

Least Squares Means									
EffectGroupDayEstimateStandard ErrorDFt ValuePr							Pr >  t		
Group	2.5 Silica		15.4301	1.0430	27	14.79	<.0001		
Group	4.5 Silica		16.8983	1.0428	27	16.20	<.0001		
Group	Alfalfa		12.2933	1.0430	27	11.79	<.0001		
Day		1	17.1547	0.8401	108	20.42	<.0001		
Day		2	13.2340	0.8401	108	15.75	<.0001		
Day		3	15.3554	0.8401	108	18.28	<.0001		
Day		4	13.3108	0.8401	108	15.84	<.0001		
Day		5	15.3147	0.8401	108	18.23	<.0001		
Group*Day	2.5 Silica	1	18.5718	1.4513	108	12.80	<.0001		
Group*Day	2.5 Silica	2	13.9238	1.4513	108	9.59	<.0001		
Group*Day	2.5 Silica	3	15.5200	1.4513	108	10.69	<.0001		
Group*Day	2.5 Silica	4	14.0871	1.4513	108	9.71	<.0001		
Group*Day	2.5 Silica	5	15.0480	1.4513	108	10.37	<.0001		
Group*Day	4.5 Silica	1	15.1252	1.4511	108	10.42	<.0001		
Group*Day	4.5 Silica	2	15.4694	1.4511	108	10.66	<.0001		
Group*Day	4.5 Silica	3	18.1724	1.4511	108	12.52	<.0001		
Group*Day	4.5 Silica	4	16.2589	1.4511	108	11.20	<.0001		
Group*Day	4.5 Silica	5	19.4657	1.4511	108	13.41	<.0001		
Group*Day	Alfalfa	1	17.7671	1.4513	108	12.24	<.0001		
Group*Day	Alfalfa	2	10.3089	1.4513	108	7.10	<.0001		

Least Squares Means										
Effect	Group	Day	Estimate	Standard Error	DF	t Value	Pr >  t			
Group*Day	Alfalfa	3	12.3739	1.4513	108	8.53	<.0001			
Group*Day	Alfalfa	4	9.5863	1.4513	108	6.61	<.0001			
Group*Day	Alfalfa	5	11.4304	1.4513	108	7.88	<.0001			