Rangeland Ecology & Management 68 (2015) 285–289

Contents lists available at ScienceDirect

Rangeland Ecology & Management

journal homepage: http://www.elsevier.com/locate/rama

Improving Cattle Nutrition on the Great Plains with Shrubs and Fecal Seeding of Fourwing Saltbush☆

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ARTICLE INFO

Key words:
dormant-season
grazing
protein intake
winter
winterfat

ABSTRACT

Two in vitro trials were conducted for estimates of dietary percentage of fourwing saltbush (*Atriplex canescens*; FS) or winterfat (*Krascheninnikovia lanata*; WF) for improved dietary digestibility when cattle graze mature cool-season grass. Three in vitro trials were conducted to estimate percentage of FS and WF seeds that could survive passage through the bovine gastro-intestinal tract (GIT) with potential for fecal seeding. Mixtures of FS and mature smooth bromegrass (*Bromus inermis*; SB) or WF and SB had greater apparent digestibility than SB alone ($P < 0.0001$). There were positive linear relationships ($r^2 \geq 0.93$) between the amount of each shrub in digested mixtures and digestibility. Similar relationships were observed with mixtures of FS and mature Altai wildrye *(Leymus angustus)*. Germination of Dakota FS seeds in the first trial, incubated for 24 or 48 h, was 55% and 47%, respectively, with no difference in germination of seeds for the 24- and 48-h incubations ($P = 0.26$), but more seeds germinated if incubated versus not ($P \leq 0.002$). Germination of Utah FS seeds, which were incubated for 24 or 48 h with high-, medium-, or low-quality forage, averaged 9% and 8%, respectively. Length of incubation, forage quality, and their interaction did not influence germination ($P \geq 0.45$). Germination of nonincubated Utah FS seeds was 21% and greater than for incubated seeds ($P = 0.004$). Average germination of WF seeds was 0.6% and 0.1% for 24- and 48-h incubations, respectively, with incubation length, forage quality, and their interaction not significant ($P \geq 0.31$). Nonincubated WF seeds had greater germination (42%) than incubated seeds ($P < 0.0001$). Results from the third trial were confirmatory for Dakota FS seed. FS and WF can improve diet quality of grazing cattle in late summer through winter, and some FS seeds have potential for fecal seeding.

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Introduction

Increasing the ability of noncultivatable semiarid lands to support ruminant livestock production is important partly because grain may eventually become less affordable for ruminant meat production and use of grain and oilseed by-products may shift toward production of milk from ruminants or meat from monogastrics, which can utilize these feedstuffs more efficiently (Oltjen and Beckett, 1996; Flachowsky, 2002; Hoffman and Baker, 2011). When grassland herbaceous plants are mature and have lower nutritional quality, ruminant livestock will graze significant amounts of some shrubs if available (Cook and Harris, 1968; Grings et al., 2001). Therefore, increasing the diversity of grassland vegetation with cold-tolerant, palatable, and often native shrubs can improve forage quality for these livestock (and wild ruminants) by providing protein, calcium, phosphorus, and other nutrients in late summer, fall, and winter (Smoliak and Bezeau, 1967; Cook and Harris, 1968; Schellenberg and Banerjee, 2002). For example, if sufficient amounts are available, the native and palatable shrubs fourwing saltbush *(Atriplex canescens)* and winterfat *(Krascheninnikovia lanata)* can provide forage with higher protein concentration than mature grass (Smoliak and Bezeau, 1967; Romo, 2004; Derner and Hart, 2005) and can improve protein intake substantially when the basal diet of grazing livestock is mature grass. Shoop et al. (1985) determined that when fourwing saltbush was abundant on shortgrass range in northern Colorado, it was a major constituent of cattle diets in winter (an average of 32% between late November and March and peaking at 55% in March) and also a major constituent of cattle diets in August (42%). Additionally, grassland with significant amounts of evergreen shrubs may sequester more soil carbon (Johnson et al., 1999; Svejcar et al., 2008).

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http://dx.doi.org/10.1016/j.rama.2015.03.003
1550-7424/Published by Elsevier Inc. On behalf of Society for Range Management.

Palatable and nutritious shrub species such as *Atriplex* and *Krascheninnikovia* are found in many arid and semiarid regions of the world, including western North America, southern Europe, northern Africa, Turkey, and the Middle East (e.g., Mediterranean saltbush, *Atriplex halimus*), central and eastern Europe, Eurasia and China (e.g., Pamirian winterfat, *Krascheninnikovia ceratoides*), and Australia (e.g., bladder saltbush, *Atriplex vesicaria*), and may be less abundant in some of these regions because of excessive browsing by livestock in the past. Semiarid grasslands where fourwing saltbush and winterfat can grow in North America include large areas of southern Alberta and Saskatchewan, western North and South Dakota, and eastern portions of Montana and Wyoming (Stubbendieck et al., 1997). However, there is little information available relative to degree of improvement in dry matter (DM) digestibility when various amounts of these cold-tolerant and nutritious shrubs are combined with mature cool-season grass, and consequently it is difficult to estimate how many shrubs would be necessary to improve the diet quality of grazing cattle during the dormant season for cool-season grass.

For improved seeding success of winterfat, relatively inexpensive broadcasting seeding in the fall is recommended (Schellenberg, 2004); however, drilling the seed is the suggested method for fourwing saltbush (Thompson et al., 2006). Drilling seed is an expensive approach for establishing new plants in grassland, yet most research has focused on drilling seed (Malik and Waddington, 1990), with less research relative to potentially less costly methods such as feeding seed to cattle and establishing new plants from seed in dung (fecal seeding, Ocumpaugh et al., 1996). Moreover, drilling seed is difficult on rough lands, and using fecal seeding to establish desirable plants may be an effective but underutilized approach if done thoughtfully (Herrick et al., 1996; Whitacre and Call, 2006). However, many seeds cannot be used for fecal seeding because the seed coats of these species do not protect the seed as it passes through the gastro-intestinal (GI) tract, and diet quality, digesta passage rate, and rumen microbial activity, which are positively related, can affect seed survival during passage through the GI tract (Ocumpaugh and Swakon, 1993; Traveset, 1998). Therefore, this study used two in vitro trials to provide information on the amount of improvement in DM digestibility when various percentages of fourwing saltbush or winterfat were mixed with mature, low-quality cool-season grass. Three in vitro assessments were also conducted to determine the percentage of fourwing saltbush and winterfat seeds that might survive passage through the bovine GI tract such that they might have potential for establishment via carefully managed fecal seeding and if diet quality and length of time in the digestive environment could influence seed survival.

Materials and Methods

Forage Digestibility Evaluation

Two two-phase in vitro DM digestibility trials (modified slightly from Tilley and Terry, 1963) were conducted in 100-mL Nalgene centrifuge tubes at 39°C with forage mixtures digested in 10 mL of rumen fluid and 40 mL of buffer solution for 48 h (phase 1) and then for 48 h with a HCl acid–pepsin solution (phase 2). Each tube contained 0.5 g of forage substrate in it, but the composition of the forage substrate varied with the experimental treatments: 95%, 90%, 80%, 70%, 60%, and 50% mature smooth bromegrass (*Bromus inermis*; 5.5% CP) combined with 5%, 10%, 20%, 30%, 40%, and 50% fourwing saltbush or winterfat (14.4% and 15.0% CP, respectively), respectively (first trial), or 70%, 60%, and 50% mature Altai wildrye (*Leymus angustus*; 3.6% CP) combined with 30%, 40%, or 50% fourwing saltbush (second trial). Treatments of 100% mature smooth bromegrass or Altai wildrye were also included in the trials, and each treatment had three replicates. The number of treatment levels evaluated in the second trial was reduced after consideration of results from the first. The smooth bromegrass, fourwing saltbush, and winterfat were collected in mid-August, and the Altai wildrye was collected in mid-November. Only living leaves and fine stem material close to leaves were collected from fourwing saltbush and winterfat plants and used in the trials. All forage materials were collected at the Northern Great Plains Research Laboratory in Mandan, North Dakota (46°46′12″N, 100°54′57″W), and all four species were dried in a forced-air oven at 43°C and then ground in a Wiley mill to pass through a 1-mm screen. Rumen fluid was obtained from two cows on a medium-quality hay diet and strained through eight layers of cheese cloth. Equal proportions from each cow were mixed together and added to the prewarmed buffer solution and tubes. All tubes were purged with CO_2 after rumen fluid was added and then were capped with a rubber stopper, which had a small vent hole in it. Composition of buffer solution was distilled water with 0.68 mM $CaCl_2$, 2 mM $MgSO_4$, 73 mM KH_2PO_4, 142 mM Na_2CO_3, 8.6 mM $NaCl_2$, 7.9 mM Na_2SO_4, and 8.3 mM NH_2CONH_2. During the first phase of the trials, the tubes were swirled at the beginning and at 2, 4, 20, and 28 h after the trial began. The acid-pepsin solution was composed of 8.25 mL of concentrated hydrochloric acid in 1000 mL of distilled water plus 0.2% w/v pepsin. During the second phase of the trials, the tubes were swirled at the beginning and at 2, 5, 24, and 36 h after the trial began. At the end of the second phase the undigested matter in the tubes was filtered through filter paper, and the dry weight of the undigested matter was determined. Three blank tubes (no forage substrate) were used in the trial to determine the weight of nondigested DM that was added via the rumen inoculum.

Data from the trials were analyzed as a completely randomized design using the general linear model (GLM) procedure (SAS Institute Inc., Cary, NC). Amount of shrub was the fixed effect. Differences were considered significant at $P \leq 0.05$.

Seed Tolerance Evaluation

The three in vitro evaluations of seed tolerance for the simulated environment of the bovine GI tract were done according to the modification of Ocumpaugh and Swakon (1993) to the two-stage in vitro dry matter digestion procedure (Tilley and Terry, 1963), which was to shorten the acid-pepsin phase to 7 h. Seeds were tested for germination after 24 or 48 h of total incubation time (at 39°C) because most viable seeds are excreted during the first or second day after ingestion (Ortman et al., 1998; Gokbulak and Call, 2004). Seeds were also tested for germination with no incubation in a simulated bovine GI environment. The "wings" were mechanically removed from the fourwing saltbush seeds, as is commonly done to improve their use in mechanical seeding. During the first 17 h of the 24-h incubation, the seeds were in an environment that simulated rumen conditions (phase 1 with rumen fluid and buffer solution as described earlier). For the final 7 h they were in an environment simulating the bovine gastric stomach and small intestine (phase 2 with acid-pepsin solution as described earlier). For the 48-h incubations, the final 7 h were the same as for the 24-h incubation, but the simulated rumen digestion (phase 1) lasted 41 h. For the first trial, 100 seeds of wild-type fourwing saltbush that were produced in North Dakota at the U.S. Department of Agriculture, Natural Resource Conservation Service's Bismarck Plant Materials Center (PMC) were placed in each centrifuge tube along with 500 mg of medium-quality ground forage that was ground through a 1-mm screen in a Wiley mill. Three replicate tubes were used for each incubation time.

Populations and vigor of rumen microbial species may affect digestion of seed coats and seed survival in the rumen, and the quality of forage that the microbes digest affects their numbers and vigor, so it was hypothesized that the quality of the forage substrate might affect seed survival. Therefore, for the second trial, 100 seeds of wild-type fourwing saltbush that were collected in Utah or the same number of wild-type winterfat seeds that were collected in New Mexico (both purchased from Wind River Seed Co., Manderson, Wyoming) were placed in each centrifuge tube along with 500 mg of high-, medium-, or low-quality ground forages that were ground as described earlier. Digestibility of the high-, medium-, and low-quality forage substrates was estimated using the formula by Linn and Martin (1989) for total digestible nutrients (TDNs) with TDN $= 0.889 - (\text{ADF\%} \cdot 0.779)$, and the results were 70, 55, and 49, respectively. A third trial using the same high-, medium-, and low-quality forage substrates was conducted with only fourwing saltbush seeds from North Dakota because of unexpected results with the purchased fourwing saltbush seeds from Utah. For the third trial, 100 of the fourwing saltbush seeds from the Bismarck PMC were placed in each centrifuge tube along with 500 mg of one of the same three ground forages used for the second trial. For all three trials, at the end of the second phase, contents of each tube were poured through a small sieve and rinsed with distilled water to remove the acid-pepsin solution. Then the moist contents from each tube were placed on moist germination paper in a germination box, and germination was tested under appropriate environmental conditions for fourwing saltbush and winterfat seeds (16 h of light at 24°C and 8 h of dark at 15°C). Captan fungicide was added to the distilled water used to moisten the germination paper to suppress fungal growth. Germination counts were made for 33 or 34 d. Germination was defined as the radical protruding through the seed coat. Each tube/germination box pair used for the evaluations was considered a statistical replicate of the two time durations of simulated digestion.

Germination data from the first trial were analyzed as a completely randomized design using the GLM procedure (SAS Institute Inc., Cary, NC). Length of time in the simulated GI environment was the fixed effect. Germination data from the second and third trials were analyzed using a factorial design and the MIXED procedure (SAS Institute Inc., Cary, NC) with forage quality of the fermentation substrate and length of time in the simulated GI environment as fixed variables and tube/germination box replicate as the random variable. Means were compared using pairwise differences of the least squares means. Differences were considered significant at $P \leq 0.05$.

Results and Discussion

Forage Digestibility Evaluation

Mixtures of fourwing saltbush and mature smooth bromegrass had greater apparent DM digestibility than bromegrass alone ($P < 0.0001$; Fig. 1), and there was a positive linear relationship between the amount of fourwing saltbush in the mixture and the DM digestibility of the mixture ($r^2 = 0.95$). The highest DM digestibility (55.4%) was observed when equal amounts of fourwing saltbush and smooth bromegrass were combined, and this mixture had 1.45-fold greater digestibility than mature smooth bromegrass alone. Smaller amounts (≤20%) of fourwing saltbush mixed with mature smooth bromegrass would not be enough to raise the overall digestibility of the diet to a sufficient level for mature beef cows in late lactation in late summer or early fall (NRC, 2000). Mixtures of winterfat and mature smooth bromegrass also had greater DM digestibility than bromegrass alone ($P < 0.0001$; Fig. 1), and there was a positive linear relationship between the amount of winterfat in the mixture and the DM digestibility of the mixture ($r^2 = 0.93$). The highest DM digestibility was observed (48%) when equal amounts of winterfat and mature smooth bromegrass were in the mixture, and this mixture had 1.2-fold greater digestibility than the mature smooth bromegrass alone. These results suggest that 40% to 50% of the diet would have to be winterfat in order to meet the nutrient density requirements for a moderate-sized beef cow that is in late lactation in late summer or early fall and grazing mature smooth bromegrass (NRC, 2000).

Mixtures of fourwing saltbush and mature Altai wildrye had greater DM digestibility than Altai wildrye alone ($P < 0.0001$; Fig. 2), and increasing the percentage of fourwing saltbush in the mixture increased the DM digestibility of the mixtures (linear relationship, $P < 0.0001$, $r^2 = 0.99$). The highest DM digestibility (58%) when equal amounts of fourwing saltbush and Altai wildrye were combined had 1.57-fold greater digestibility than mature Altai wildrye alone. These results indicate that a diet of 30% fourwing saltbush may be sufficient for a moderate-sized beef cow in late lactation grazing in late summer or early fall if her basal diet is mature Altai wildrye (NRC, 2000).

Results from the in vitro digestibility trials indicate that if the basal diet of cattle in late summer is mature perennial cool-season

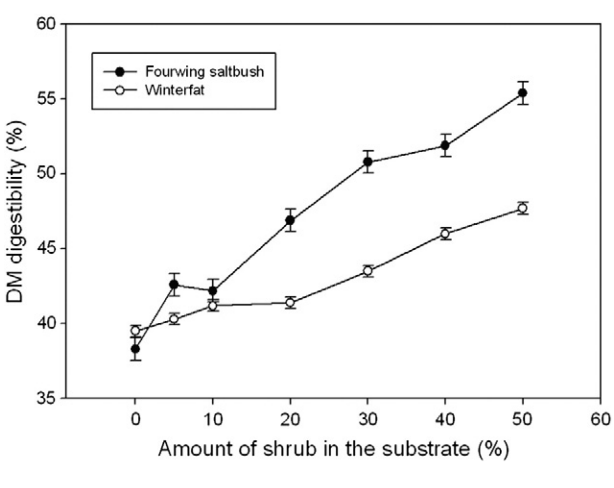

Fig. 1. Apparent dry matter (DM) digestibility of substrates in an in vitro trial with variable amounts of fourwing saltbush or winterfat and mature smooth bromegrass in the digestion substrate (vertical bars represent standard error of mean [SEM]).

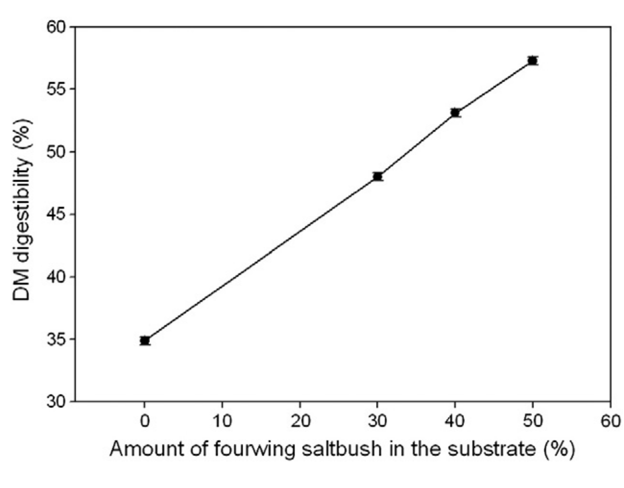

Fig. 2. Apparent dry matter (DM) digestibility of substrates in an in vitro trial with variable amounts of fourwing saltbush and mature Altai wildrye in the digestion substrate (vertical bars represent SEM).

grass such as smooth bromegrass or Altai wildrye, then their diet quality can be improved by addition of fourwing saltbush or winterfat at 30% or more of their diet. This conclusion probably also applies to later in the dormant season because protein concentrations in cold-tolerant fourwing saltbush and winterfat tend to stay relatively high into fall and early winter (Davis, 1979; Otsyina et al., 1982). Rumbaugh et al. (1982) observed that forage and protein yields could be increased in late summer by the addition of fourwing saltbush with the cool-season perennial grass crested wheatgrass *(Agropyron cristatum)*. Otsyina et al. (1982) concluded that fourwing saltbush and winterfat were the most promising shrubs to supplement crested wheatgrass for late fall grazing by sheep with gestating sheep requiring a diet of 56% fourwing saltbush or 69% winterfat if the remainder of their diet was composed of low quality (<2% digestible protein) mature crested wheatgrass.

Nonlactating, pregnant, and mature Hereford cows grazing an ample variety of native grass and shrub species in eastern Montana in November and December did not show a benefit from supplemental protein, and Kartchner (1981) concluded that protein was not a limiting factor for either forage intake or digestibility during this period. The digestibility of dietary dry matter was estimated to be 55% for the nonsupplemented cows, and they gained weight during that period (Kartchner, 1981). These findings along with those of the present study suggest that if mature beef cows in late lactation or that are nonlactating and in early gestation have access to ample amounts of fourwing saltbush and mature cool-season perennial grasses, then their diets may have to be composed of about 30% fourwing saltbush to have adequate protein and dry matter intake. This diet would be less appropriate for young growing beef cattle (NRC, 2000), but better than only mature grass if the cattle were raised for sale to a grass-fed (forage only) beef program. Grings et al. (2001) reported that mature and lactating beef cows grazing eastern Montana grassland, which was similar to the land used for Kartchner's (1981) study, grazed diets composed of as much as 29% and 21% shrubs in September and October, respectively, but did not report dietary composition data for late fall and winter.

Seed Tolerance Evaluations

In the first trial, the average germination of fourwing saltbush seeds from North Dakota, which were incubated for 0, 24, or 48 hours in the in vitro simulated GI environment, was 18%, 55%, and 47%, respectively. There was no difference in percent germination of seeds for the 24- or 48-h incubations in the GI environment ($P = 0.25$). However, more seeds germinated after being in the simulated GI environment for 24 or 48 h than for 0 h ($P \leq 0.002$).

For the second trial, average germination of the fourwing saltbush seeds from Utah was 9% and 8% for the 24- and 48-h incubations, respectively, and 21% with no incubation in the simulated GI environment (Fig. 3). Length of incubation, substrate forage quality, and the interaction of these two factors did not influence germination of saltbush seeds ($P \geq 0.45$), but the seeds that were not incubated in the simulated GI environment had greater germination than the seeds that were incubated ($P = 0.004$). Average germination of the winterfat seeds was only 0.6% for the 24-h incubation, 0.1% for the 48-h incubation, and 42% with no incubation in the simulated GI environment. Length of incubation, substrate forage quality, and the interaction of this with incubation time did not influence germination of winterfat seeds ($P \geq 0.31$), but the seeds that were not incubated in the simulated GI environment had greater germination than the seeds that were incubated ($P < 0.0001$). The very low germination of the winterfat seeds that went through the simulated gut environment is not a problem in lieu of the fact that the recommended method for seeding winterfat is to broadcast the seeds. This approach would likely be less expensive and much more effective compared with trying to establish winterfat with bovine fecal seeding. However, the low germination of the fourwing saltbush seeds from Utah when passed through the simulated GI tract was unexpected; therefore, another trial was conducted with North Dakota fourwing saltbush seed to check for response of this seed when it was exposed to high-, medium-, and low-quality fermentation substrates in the simulated bovine GI environment.

In the third trial, average percent germination of the fourwing saltbush seeds from North Dakota was 36% and 33% for the 24- and 48-h incubations, respectively, and 23% with no incubation (Fig. 3). Length of incubation, substrate forage quality, and the interaction of these factors did not influence germination of saltbush seeds ($P \geq 0.30$), but the seeds that were incubated in the simulated GI environment had greater germination than the seeds that were not incubated ($P = 0.03$). The reason(s) for the very different responses in the simulated GI environment by the fourwing saltbush seeds from North Dakota versus Utah were not readily apparent, but the seed coat on the Utah might have been slightly damaged because a little more of the "wing" material was removed from this seed than from the North Dakota seed. This brings up an important point, which is that if the seed is intended for fecal seeding, one should discuss this with the seller of the seed because how a particular species of seed is processed before sale can have a large influence on its suitability for fecal seeding (e.g., scarification may be ideal for normal drill seeding but terrible for fecal seeding).

Results from the seed tolerance evaluations indicate that it may be feasible to use cattle and fecal seeding to establish fourwing saltbush on northern grasslands, and the quality of the forage consumed will probably not influence the viability of the seed defecated. However, the suitability of a particular batch of these seeds should be evaluated with an in vitro test before they are fed to cattle. Barrow and Havstad (1992) placed a small number (800) of intact fourwing saltbush seeds in the rumen of beef steers in gelatin capsules and recovered 7.5% of these seeds within 48 h with 15% of these germinating post recovery. Their low recovery and germination of seeds may have resulted from using fourwing seeds that were less tolerant of the bovine GI environment and/or perhaps due to the fact that they fed seeds with the four "wings" intact on the seeds and also fed their study steers alfalfa hay at only about 2.0% of body weight per d. Consequently, the seeds may have passed out of the rumen slower and may have been ruminated and chewed on more before passing out of the rumen and farther down the GI tract.

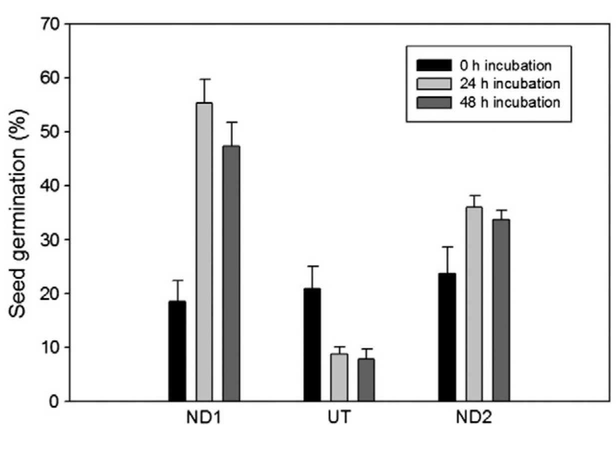

Fig. 3. Seed germination (%) of fourwing saltbush seeds from North Dakota used in an initial trial (ND1), from Utah (UT) or North Dakota and used in a second trial (ND2; vertical bars represent SEM).

Ocumpaugh et al. (1996) compared establishment of switchgrass *(Panicum virgatum)* from seed that was either fed to cattle and deposited in feces or broadcast seeded and found that although germination of seeds that passed through cattle was about 35% lower, establishment of switchgrass plants from seed in cattle feces was equal to or greater than that of the broadcasted seeds. One year after seeding with both approaches, there were similar or more switchgrass plants, respectively, in the fall and spring fecal-seeded plots, and the fecal-seeded plants were larger. Therefore, the second phase of bovine fecal seeding should also be considered important for successfully establishing viable plants on grassland via fecal seeding. The results from Ocumpaugh et al. (1996) also suggested that feces on grassland with less existing vegetation will increase plant establishment from bovine feces, and regular rainfall for many weeks will likely aid plant establishment. There are also factors that can hinder plant establishment, such as location of seeds deep within a large fecal pat and formation of a hard dry crust on the outer surface of a fecal pat, which reduce the ability of germinated seeds to emerge from a fecal pat (Akbar et al., 1995). Therefore, it seems reasonable to assume that fourwing saltbush may be established most effectively with bovine fecal seeding if it is fed to cattle in the spring when they are grazing high-quality forage that leads to faster passage of digesta and seed through the GI tract, thinner fecal droppings, and also a wetter environment that would reduce the rate that a hard, dry crust could form on the feces.

With increasing global demand for food including animal protein and limited availability of arable lands for crop production, arable lands may be increasingly used for production of human foods and for feeds for efficient livestock. The world's remaining grasslands (primarily lands less suitable for grain and oilseed production) will become more important for ruminant meat production, as well as for providing other ecosystem services such as clean water, carbon sequestration, and wildlife habitat. Consequently, increasing the quality and quantity of the forage available on these grasslands is an important endeavor with respect to meat production with ruminants. Estell et al. (2012) have argued that because about 50% of the world's land surface is grazed by livestock and that woody plants are a significant and growing component of the vegetation on much of this grassland, there is a need for approaches to increase shrub use by livestock. This paper argues that some northern grasslands could be improved for late summer, fall, and winter grazing by late-lactating or nonlactating/early-gestational beef cows (or yearling cattle) by the addition of palatable shrubs such as fourwing saltbush and winterfat, and it may be feasible to use cattle to establish this shrub.

Implications

The results of this research indicate that increasing the amount of the palatable, cold-tolerant shrubs fourwing saltbush and winterfat in the diet of cattle grazing mature cool-season grass can increase the DM digestibility of their diet in a linear manner. It also indicates that increasing the amount of cold-tolerant and palatable shrubs such as fourwing saltbush in semi-arid grasslands in western North America may be possible, with a thoughtful and well-informed approach, by feeding its seed to cattle and establishing new plants from fecal pats.

Acknowledgements

The author thanks Clay Erickson, Faye Kroh, Stephanie Schmidt, and Becky Wald (USDA, ARS, Northern Great Plains Research Laboratory, Mandan, ND) for technical assistance with the study and the USDA, NRCS, Plant Materials Center in Bismarck, ND for the ND fourwing saltbush seed used in the study.

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