



# Enhancement of photosynthesis and growth of an aridland perennial in response to soil nitrogen pulses generated by mule deer

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## Abstract

The altitudinal range of the aridland herbaceous perennial *Cryptantha flava* overlaps with the wintering range of mule deer (*Odocoileus hemionus*) in the Colorado Plateau region of the western United States. Deer do not feed on *C. flava*, but do influence its microhabitat via excretion of nitrogenous compounds. We were interested in determining the importance of Mule deer in redistributing N across the landscape as well as determining if *C. flava* was capable of exploiting the ephemeral pulse of this redistributed N. Based on pellet group counts, we found that mule deer deposited on average  $16 \text{ g N m}^{-2}$  per year to our study site in northeastern Utah, but these deposits were highly variable spatially. The distribution of pellet groups varied from 3 to 49 per  $100 \text{ m}^2$ , resulting in 6% of individual plants in the population occurring within 20 cm of a pellet group. We applied a one-time treatment of fresh fecal pellets (0.53 g N per pellet group) and experimental urine (6.4 g urea N) to small pre-reproductive plants to examine plant response to N pulses. Within 1 week of application, leaf N concentrations, instantaneous photosynthetic rates, carboxylation efficiencies, and  $\text{CO}_2$  saturated photosynthetic rates were significantly higher for plants receiving simulated urine. Increased photosynthetic rates were maintained for at least 3 weeks post treatment. Over the next 2 years, urine-treated plants had higher growth rates, greater overall size, higher flowering rates, and produced more seed per plant than those not receiving artificial urine. We conclude that *C. flava* is capable of rapid root uptake and incorporation of nitrogen into photosynthetic structures in response to nitrogen pulses. Mule deer activity has direct population level consequences on this species by increasing spatial nutrient heterogeneity, and the growth and reproductive rate of individuals.

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**Keywords:** Colorado plateau; *Cryptantha flava*; Desert; *Odocoileus hemionus*; Pellet groups; Urine pulse

## 1. Introduction

Low rainfall and limited soil nutrients result in low primary productivity in aridland systems (Smith et al., 1997). Low primary productivity further reduces soil nutrient levels by reducing the amount of organic matter available for recycling.

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In addition, low rainfall slows the breakdown of organic matter, further reducing the rate of recycling and nutrient uptake from soils. Both water and nitrogen additions have been found to increase plant productivity in aridland systems (Gutierrez and Whitford, 1987; Gutierrez et al., 1988). Nitrogen addition alone may increase perennial plant growth by more than 100% (Ettershank et al., 1978). These results suggest that many aridland plants may be water regulated, but nitrogen limited.

Associated with low soil resource levels in aridland ecosystems is a high degree of spatial and temporal heterogeneity. Spatially, soil nutrients may vary at scales comparable to the sizes of individual plants (Burke, 1989). In many aridland systems, the highest nutrient concentrations are in the upper 20 cm of the soil (Evans and Ehleringer, 1994). Local processes such as earthworm casts (Zaller and Arnone, 1999), nutrient islands associated with perennial shrubs (Shmida and Whitaker, 1981; Jackson and Caldwell, 1993; Shumway, 2000) or large mammal deaths and/or excretion (Afzal and Adams, 1988) can increase the spatial heterogeneity of soil nutrients. Seasonally, variability in precipitation and snowmelt cause pulses of nutrients through increases in mineralization rates (Burke, 1989; Gallardo and Schlesinger, 1992).

Nutrient pulses can be an important component of the annual nutrient supply for plants (Campbell and Grime, 1989). Therefore, plants in habitats with high temporal and spatial heterogeneity in soil nutrient availability are hypothesized to have evolved mechanisms to exploit these pulses (Robinson, 1994). For example, plants may increase fine root growth (Jackson and Caldwell, 1989; Caldwell et al., 1991; Larigauderie and Richards, 1994), and/or they may increase root uptake kinetics to exploit microsite nutrient enrichment (Jackson et al., 1990; Caldwell et al., 1992). Most ecological work on plant response to nutrient patches and pulses has concentrated on either root-level responses or community-level changes, while few studies have incorporated measurements of leaf-level physiology (Day and Detling, 1990a,b; Jaramillo and Detling, 1992a,b).

Large mammals may play a significant role in communities by changing the dynamics and spatial heterogeneity of nitrogen cycling. Mammal herds have been shown to change species diversity (Steinauer and Collins, 1995), community structure (Pastor et al., 1993) and productivity (McNaughton et al., 1988; Day and Detling, 1990a). These effects often occur through species-specific responses to mammal activity (Day and Detling, 1990a,b). Most of the research on mammal impacts on plant communities have been conducted in grassland, savanna, or forest systems. However, there are a number of large mammals inhabiting arid ecosystems. In the southwestern United States, large mammals include pronghorn antelope (*Antilocapra americana*), elk (*Cervus canadensis*) and mule deer (*Odocoileus hemionus*). The effects of large mammals on soil nutrient heterogeneity and particularly plant response to mammal-generated nutrient patches are less well characterized in these arid systems.

Mule deer, *O. hemionus*, have a wide range in western North America, occurring west of a line from southwestern Saskatchewan through central North and South Dakota, Nebraska, Kansas, and western Texas (Anderson, 1984). Mule deer are browsing ruminants, ranging from 43 to 150 kg in adult size. Mule deer prefer highly digestible, succulent forage, and diets consist of approximately 50% woody and 50% herbaceous forage (Short, 1981). Food intake averages about 22 g kg<sup>-1</sup> body weight per day, although this varies seasonally with the availability of high quality forage (Anderson, 1984). The behavioral patterns of *O. hemionus* increase its potential impact on the nutrient cycling and patchiness of local habitats. A social system of maternally related females serves as a resource defense unit (Kucera, 1978; Geist, 1981). These groups confine daily movements to discrete home ranges, and migrate between the same winter and summer home range over multiple years. Fidelity to the same home range results in concentrated deposition of urine and feces over time.

*Cryptantha flava* (A. Nels.) Payson (Boraginaceae) is a widespread, C<sub>3</sub> herbaceous perennial that occurs throughout the Colorado Plateau region of Utah, Colorado and Arizona. The species can be

found on low nitrogen, sandy soils, over a range of different precipitation regimes. This broad distribution in a spatially and temporally heterogeneous environment make it an ideal study system with which to examine plant response to pulses in resource availability. Previous work has shown that *C. flava* responds dramatically to environmental heterogeneity. Gebauer and Ehleringer (2000) showed that *C. flava* was capable of rapid uptake of moisture and N pulses from upper soil layers after watering. Casper (1996) demonstrated that large and small individuals respond differently to rainfall, resulting in the reversal of population size hierarchies after dry years. Since reproduction is positively related to plant size, changes in size hierarchies could potentially affect population genetic structure.

*C. flava*'s growth habit consists of a woody caudex supporting rosettes of nearly vertical leaves. In northeastern Utah, where our study site is located, seed germination and aboveground vegetative growth begins in late March, flowering occurs in mid-May and seeds are set by mid-July. Most vegetative rosettes senesce during the hot, dry summers, followed by active vegetative growth and some seedling germination in the fall.

This study addresses the photosynthetic, individual growth, and population-level response of *C. flava* to nutrient pulses generated by a sympatric, overwintering herd of *O. hemionus*. We hypothesized that due to the natural spatial and temporal heterogeneity of its habitat (Bilbrough and Caldwell, 1997; Forseth et al., 2001) *C. flava* would be able to exploit ephemeral pulses of soil N through root uptake. However, we did not know how the uptake of pulsed N might be utilized by the plant, or whether the amount of N deposited by mule deer would be enough to affect population level processes. Accordingly, we asked the following questions: How much N from mule deer is deposited in this system? What is the spatial pattern of N deposition from mule deer in relation to plant distribution? Would plants in close proximity to deer pellet groups be capable of taking up N from the excretion patch and would there be a differential response from the added urine or fecal pellets? Would plants incorporate pulsed N into current photosynthetic structures, or

build new leaves? Would pulsed N uptake increase flowering and/or seed set? What is the time scale of flowering response to N uptake, e.g. does it occur in the same year as the N pulse, the next year, or future years? Finally, is the combination of the amount and pattern of N deposition by mule deer and individual plant response to N patches capable of affecting population level processes in *C. flava*?

## 2. Materials and methods

### 2.1. Study system

This research was conducted on land managed by the US Bureau of Land Management (1730 m elevation, 40°30'N, 109°22'30"E). Vegetation is dominated by the shrubs *Artemisia tridentata* Nutt. and *Chrysothamnus nauseosus* (Pallas) Britt. and the small tree *Juniperus osteosperma* (Torr.) Little. The study area is characterized by substantial environmental variation, both seasonally and spatially. Annual precipitation averages 215 mm and is highly variable (coefficient of variation of monthly rainfall ranges from 50 to 125% of the mean). The presence of rock outcrops, soil depth differences, shrubs, and variable drainage patterns generate a considerable amount of spatial heterogeneity in water availability and solar radiation (Forseth et al., 2001).

Mule deer contribute to the variability in the spatial landscape by depositing N in excretion patches and by creating compressed trails, but do not feed on *C. flava*. Deer typically urinate at the same time as defecating, so we were able to use pellet groups as indicators of urine and feces deposition (Wallmo, 1981). This site acts primarily as winter range for Mule deer, typically spend 6 months of the year in our lower elevations to escape the cold climate and find adequate forage (Richens, 1967). Deer densities range from 31 to 52 km<sup>-2</sup>, but is highly variable due to forage quality and quantity (Richens, 1967).

## 2.2. Mule deer N redistribution

To document the spatial characteristics of mule deer excretions relative to the population of *C. flava*, we established sixteen contiguous 10 × 10 m plots. In the spring of 1998, we mapped all plants and all pellet groups deposited over the 1997–1998 winter. Current year pellet groups could be distinguished from the previous year's pellet groups because the latter are bleached white and appear desiccated, while the former are darkly colored and appear moist. We classified pellet groups within 20 cm of a *C. flava* individual as being potentially exploitable patches.

We estimated N input from mule deer to the system by adding estimated urine and fecal N inputs. To calculate urine-derived N input, we multiplied the average number of pellet groups by the mean content of N in a mule deer urination (6.4 g of urea N, based on minimum levels in ungulates on low protein diets during winter months Moen and DelGiudice, 1997). We estimated fecal-derived N by collecting 20 pellet groups, drying them at 80 °C to a constant weight, and then weighing them to the nearest 0.01 g. Pellets were then ground with a mortar and pestle to a fine powder and analyzed for % N by weight using a Perkin–Elmer 2400 Series CHN analyzer (Norwalk, CT). We determined total N per pellet group by multiplying the average number of pellets in a group by the mean content (by weight) of N in a pellet.

## 2.3. N pulse response

In 1998 completely randomized experimental design was used, with 200 small (< 6 rosettes), pre-reproductive plants being chosen from the population and assigned to one of five treatments, three with fertilization and two different controls. In addition, plants were chosen to minimize any microhabitat effects by choosing similar slope and aspects as well as in open habitats by being greater than 1 m from the canopies of any associated shrub species. The groups were: (1) fecal pellet treatment (F), where current year's fecal pellet groups (collected in early June) were collected and placed in a circle less than 10 cm

from base of experimental plants; (2) urine treatment (U), where one liter of experimental urine was poured in a circle into the soil around the base of a plant; (3) urine and fecal pellet treatment (UF), where both pellet groups and simulated urine were applied to the base of plants; (4) control (C), where no manipulation was done to plants; (5) deionized water control (DI), where 1 l of DI water was poured in a circle immediately around the base of the plant (a control for any water addition effects). This treatment design would allow which constituent would be responsible for any effects on our response variables, or if any synergistic effects are present with the addition of both urine and fecal N.

The simulated urine followed the recipe of Stillwell (1983). The main constituent was urea, totaling 6.4 g of nitrogen in 1 l DiH<sub>2</sub>O reflecting minimum urea nitrogen levels found in ungulates on low protein diets during winter months (Moen and DelGiudice, 1997). The volume of urine per urination event varies widely (0.5–2 l per event). We chose 1 l as an estimate of the median volume per urination event (G. DelGiudice, Minnesota Department of Natural Resources, personal communication). Treatments were applied between June 3 and 4, 1998.

Instantaneous photosynthetic rates and leaf N concentrations were measured on random subsamples of the plants 1 week prior to treatment and 1 week after treatment. Photosynthetic and N concentrations were not necessarily measured on the same leaves, however, due to the small nature of most plants, this could have occurred although was not noted. In addition, we measured instantaneous photosynthetic rates once a week for 2 more weeks. We measured instantaneous photosynthetic rates at saturating light levels and ambient CO<sub>2</sub> concentrations using a Li-Cor 6200 portable photosynthesis system (LiCor Inc., Lincoln, NE). Photosynthetic measurements were restricted to the hours of 09:00–14:00 h MST to ensure saturating light levels and to restrict the range of ambient temperature and vapor pressure deficit. One week after treatment application, we also measured the response of photosynthesis to intercellular CO<sub>2</sub> concentration ( $c_i$ ) using a Li-Cor 6400 portable photosynthetic system (LiCor Inc.). This

instrument was only available for a limited time, restricting its use to the photosynthetic response to  $c_i$  measurements (instrument courtesy of D.A. Wait, Southwest Missouri State University). Response curves using the Li-Cor 6400 are preferred over the Li-Cor 6200 due to the rigorous control of chamber conditions. Photosynthetic response to  $c_i$  was measured at a light level of  $2000 \mu\text{mol m}^{-2} \text{s}^{-1}$ , a leaf temperature of  $32^\circ\text{C}$ , and a leaf-to-air vapor pressure deficit of  $3.5 \text{ kPa Pa}^{-1}$ . Steady-state photosynthetic rates were measured initially at an ambient  $\text{CO}_2$  concentration of  $370 \mu\text{mol mol}^{-1}$ , then  $\text{CO}_2$  concentration was changed to  $350 \mu\text{mol mol}^{-1}$  where photosynthesis was recorded again, followed by successive measurements at  $\text{CO}_2$  concentrations of 250, 150, 50, 350, 500, 800, 1000 and  $1200 \mu\text{mol mol}^{-1}$ . Lower  $\text{CO}_2$  concentrations were measured first to prevent stomatal closure effects.

Fully expanded leaves from the study plants were collected 1 week prior to, and 1 week after treatment for elemental analysis. Leaves were dried at  $80^\circ\text{C}$  for a minimum of 24 h, ground to a fine powder using a mortar and pestle with liquid  $\text{N}_2$  and analyzed using a Perkin–Elmer 6400 series CHN analyzer (Perkin–Elmer, Shelton, CT). To test for N uptake, pre- and post treatment leaves were measured for isotopic composition of  $^{15}\text{N}/^{14}\text{N}$  ( $\delta^{15}\text{N}$ ) to compare with the  $\delta^{15}\text{N}$  signature of non-treated plants using a continuous flow isotopic ratio mass spectrometer (SIRFER, University of Utah).

Demographic data were taken on experimental plants several times during the growing season in 1998. We measured the number of rosettes and the length and width of the largest leaf on the plant. None of these demographic measurements changed significantly during the 1998 season (data not shown). In 1999 we measured the same suite of traits in addition to the number of flowering stalks. Flowering stalks were collected upon senescence and examined for seed production. Fifteen random flowers from each flowering individual were collected and all mature seeds (nutlets) from these flowers were counted.

Statistical analyses of instantaneous photosynthetic rates, leaf N concentration and number of rosettes used a repeated measures analysis of

variance for data collected prior to and 1 week post treatment application. In addition, rosette number was also analyzed using a repeated measures approach for data collected in 1998 and 1999. The results of these analyses led us to believe that the primary response of both physiology and growth came from the urine constituent. In all cases, the urine and urine + feces treatment showed significant increases in all three parameters measured;  $A_{\text{max}}$ , % leaf N, and number of rosettes (Fig. 1). Based on these three results, we grouped all subsequent analyses into two treatments, those that received urine and those that did not. To control for making excessive Type I errors, we chose Scheffe's multiple comparison procedure to control experimentwise error rate at 0.05. This procedure is extremely conservative, recommended for posteriori contrasts and controls for all comparisons, regardless if they are made (Jones, 1984; Snedecor and Cochran, 1989). Photosynthetic ( $A$ ) response to intercellular  $\text{CO}_2$  ( $c_i$ ) was analyzed using a nonlinear mixed model approach where each plant was assigned a unique number and analyzed across time (Peek et al., 2002). Individual curves were fit to a Mitscherlich model in the form:

$$A = A_{\text{max}}[1 - e^{-\text{CE}(c_i - \text{Comp})}] \quad (1)$$

where three parameters were estimated and analyzed statistically; maximum photosynthetic rates at saturating  $\text{CO}_2$  ( $A_{\text{max}}$ ), carboxylation efficiency (CE, initial slope), and  $\text{CO}_2$  compensation point (Comp, x-intercept). Analyses of  $\delta^{15}\text{N}$  concentrations and demographic data were completed using a one-way ANOVA model. Flowering response was calculated as the percent of the total number of individuals in a treatment category that flowered. Seed set data is expressed as the percentage of total flowers with two mature seeds per treatment category. Both flowering and seed set were analyzed using a  $\chi^2$  contingency table to determine significant differences between treatments.

### 3. Results

Mule deer deposited on average 24.3 pellet groups per plot in our study site, or approximately

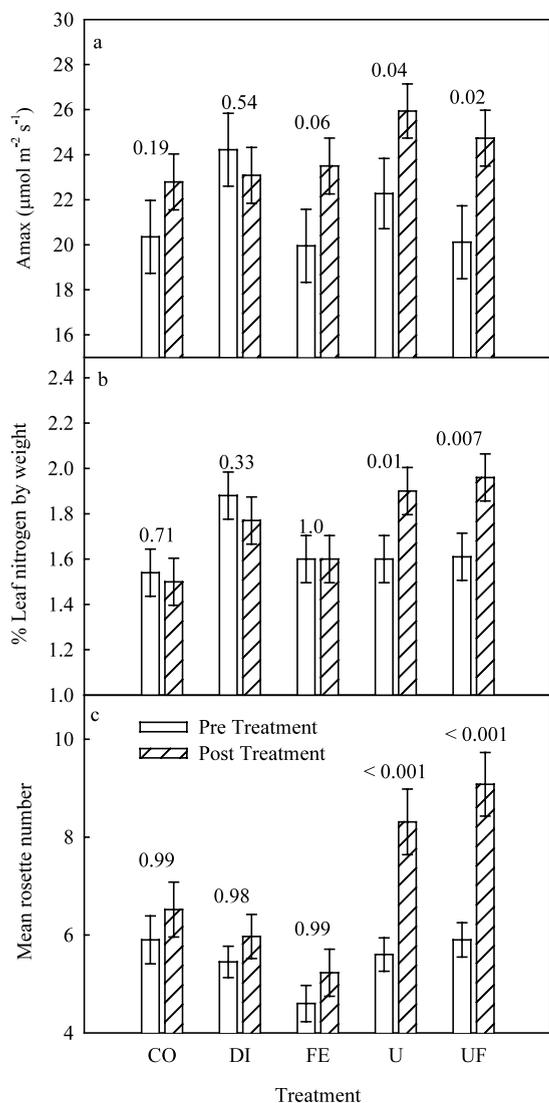


Fig. 1. Repeated measures analysis of (a) instantaneous photosynthetic rates ( $n = 15$  per treatment), (b) leaf nitrogen concentrations ( $n = 15$  per treatment) 1 week prior to and 1 week post treatment application, and (c) the number of rosettes per plant ( $n = 40$  per treatment) 1 year after treatment application. Treatments correspond to control (CO), deionized water (DI), fecal pellets (FE), urea derived N (U) and fecal pellets + urea N (UF). Means  $\pm$  1 S.E. are presented with numbers above bars representing probabilities of significant differences between time periods from the repeated measures ANOVA.

2430 pellet groups  $\text{ha}^{-1}$  (Table 1). Using an average from Moen and DelGiudice (1997) of 6.4 g of urea N per excretion event, this would

Table 1  
Summary of plant and pellet group density

	Mean # per plot $\pm$ S.D.	Minimum	Maximum
Plants	155.4 $\pm$ 151.1	10	537
Fecal pellet groups	24.3 $\pm$ 14.6	3	49
'Exploitable' groups	8.7 $\pm$ 10.2	0	36
% Plants	5.6 $\pm$ 4.2	0	9.7

Mean number of plants, mean number of pellet groups, mean number of 'exploitable pellet groups' (defined as a pellet group within a circle of 20 cm radius from the outer edge of a plant canopy), and mean percentage of plants within 20 cm of a pellet group in 16 100  $\text{m}^2$  plots.

translate to approximately 15.6 kg N  $\text{ha}^{-1}$  per year of urea-derived N. The mean dry weight per pellet group was  $33.4 \pm 2.3$  g ( $n = 20$ ). With a mean concentration of  $1.6 \pm 0.1\%$  N ( $n = 20$ ), this results in 0.5 g N per pellet group and fecal N of 1.2 kg N  $\text{ha}^{-1}$  per year. Thus, the total was approximately 16.8 kg N  $\text{ha}^{-1}$  per year being redistributed into concentrated patches by mule deer at our study site.

The spatial distribution of pellet groups was highly variable, with a range of 3–49 groups found in a 100  $\text{m}^2$  plot (Table 1). This spatial patchiness has two ramifications. First, the distribution of pellet groups in relation to individuals of *C. flava* resulted in approximately 6% of all plants surveyed occurring within 20 cm of a current year's pellet group (Table 1). Second, the amount of N redistribution can vary by over 16 fold between plots. We found a mean of 2430 pellet groups per hectare, with a minimum number of 300 and maximum of 4900 pellet groups per hectare. Therefore, N redistribution by Mule deer on a per hectare basis can be as little as 2.1 kg N  $\text{ha}^{-1}$  per year or as great as 33.8 kg N  $\text{ha}^{-1}$  per year.

One week following N treatment the simulated urine (U) and urine + feces (UF) treated plants increased photosynthetic rates under ambient temperature, high-light conditions from  $22 \pm 1.6$  and  $19 \pm 1.6$   $\mu\text{mol m}^{-2} \text{s}^{-1}$  to  $26 \pm 1.2$  and  $25 \pm 1.2$   $\mu\text{mol m}^{-2} \text{s}^{-1}$ , respectively ( $F_{4,71} = 7$ , \*,  $P < 0.05$ , Fig. 1a). The increase in instantaneous photosynthetic rates was accompanied by increased leaf N

concentrations in the same treatments ( $F_{4,71} = 10$ ; \*\*,  $P < 0.01$ ; Fig. 1b). Differences among treatments came primarily from the urine treatment, whereas the fecal pellet treatment alone (F) showed an increase after 1 week at the  $P = 0.06$  level in instantaneous photosynthetic rates, but no significant increase in leaf N concentration (Fig. 1b). Repeated measures analysis of growth, number of rosettes, 1 year later also showed a significant increase in those plants receiving the urine ( $F_{4,169} = 6.25$ ; \*\*\*,  $P < 0.001$ ; Fig. 1c). Since we could not have predicted a priori that the response would come only from the added urine, we chose to analyze only those three, two physiological and one growth, variables separately. For all other analyses, we grouped treatments into plant receiving urine and plants not receiving urine. Although, longer-term effects from fecal N released from mineralization could provide a plant with available N (Ruess and McNaughton, 1988; Pastor et al., 1993), we believe this is negligible due to the very low C:N ratio of our soil. Forseth et al. (2001) reported a mean C:N ratio of 70:1, resulting in intense competition for N from microbes (Gallardo and Schlesinger, 1995). For these reasons, and since the N in an excretion event is approximately 95% urea derived N, we grouped treatments into those containing urine and those not containing urine.

The increase in photosynthetic rates was maintained for at least 3 weeks after treatment application, with an overall increase in photosynthesis in both treatments resulting from several precipitation events in June (Fig. 2). When all of the post-treatment  $A_{\max}$  measurements were pooled, the F treatment did not differ significantly from control treatments (C and DI), but was significantly lower than the treatments receiving urine (U and UF).

Plant uptake of supplied N was indicated by a marked change in leaf  $\delta^{15}\text{N}$  values in the post-treatment urine plants (Fig. 3). Means of  $-2.7$ ,  $-3.0$ , and  $-2.3\text{‰}$  were recorded for pre- and post-no urine treatment and pre-urine treatment, respectively. The pre-urine treatments did not differ significantly ( $F_{1,33} = 2.17$ ,  $P = 0.15$ ), while the post-urine treated value of  $-8.8\text{‰}$  was significantly different from all other treatments ( $F_{1,32} = 288$ ; \*\*\*,  $P < 0.001$ ). In addition, we analyzed the

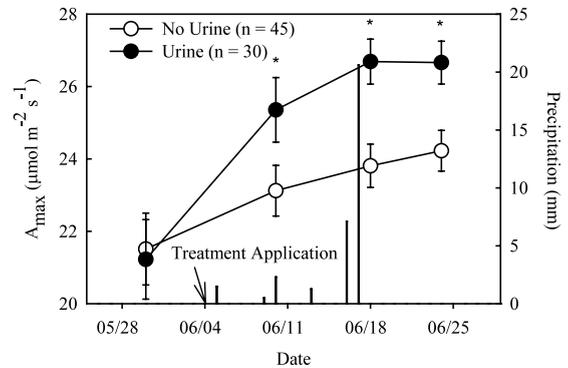


Fig. 2. Mean maximum photosynthetic rates at ambient  $\text{CO}_2$  and saturating light levels (lines and circles, left axis). Means presented are grouped into plants receiving urine (closed symbols) and those receiving no urine (open symbols) for 30 May and 9, 16 and 23 June. Bars are  $\pm 1$  S.E. with sample sizes in parentheses. An asterisk denotes significantly different means at each time period (\*,  $P < 0.05$ ). Also shown are precipitation events during the measurement period (vertical lines, right axis).

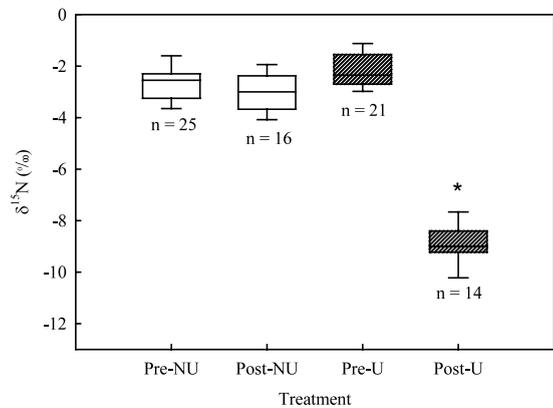


Fig. 3. Leaf  $\delta^{15}\text{N}$  values for plants collected prior to treatment and 1 week after treatment with simulated mule deer urine. Boxes represent 25th and 75th percentiles, bars represent the range and the horizontal line is the mean ( $n = 40$ ) for pre treatment no urine (Pre-NU) and urine (Pre-U) and post treatment no urine (Post-NU) and urine (Post-U) plants. Sample sizes are given below boxes.

five treatments separately prior to the treatment application and found no treatment effects ( $F_{4,30} = 1.58$ ;  $P = 0.2$ ), while the post treatment  $\delta^{15}\text{N}$  values showed a significant treatment effect only for those plants receiving the simulated urine ( $F_{4,29} = 74$ ; \*\*\*,  $P < 0.001$ ).

Photosynthetic responses to intercellular  $\text{CO}_2$  concentration were altered significantly within 1 week of the application of N treatments (Fig. 4). At saturating levels of  $\text{CO}_2$ , the urine-treated plants had a maximum photosynthetic rate 26% higher than that of plants not receiving urine ( $F_{1,12} = 13$ ; \*\*,  $P < 0.01$ ). Carboxylation efficiencies (initial slope of the photosynthesis– $c_i$  curve) of urine-treated plants were also significantly higher than the non-urine treated plants ( $F_{1,12} = 6.3$ ; \*,  $P < 0.05$ ; Fig. 4).

Growth differences among treatments were not detected in 1998, the year of treatment, but were markedly evident in the year following treatment, 1999 (Fig. 1c Fig. 5). The growth of plants from 1998 to 1999 was increased significantly by the addition of urea N (Fig. 5). Plants receiving simulated mule deer urine added on average three more rosettes than those not receiving urine. Average relative rosette growth rate was 0.11 rosettes per rosette per year for plants not receiving urine, compared with 0.37 rosettes per rosette

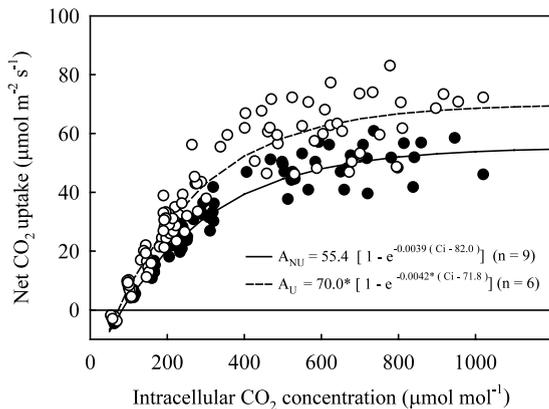


Fig. 4. Photosynthetic response to intracellular  $\text{CO}_2$  concentration for plants receiving urine (open circles) and not receiving urine (closed circles). Curves were fit using a non-linear mixed model, where  $\text{CO}_2$  compensation point, carboxylation efficiency and maximum photosynthesis at saturating  $\text{CO}_2$  levels were estimated from curves from individual plants for each treatment and compared statistically (see text for details). Equations using parameter estimates obtained from individual curves are also shown, an asterisk indicates significantly different parameter estimates, \*,  $P < 0.05$ .

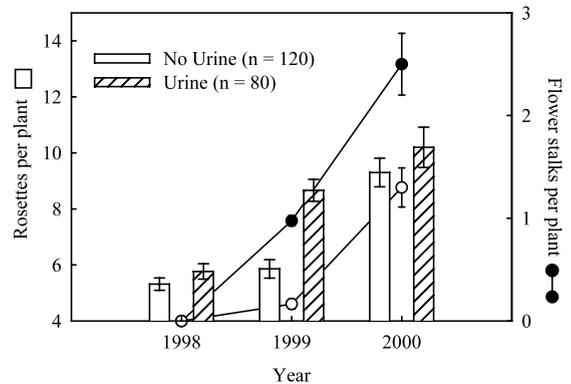


Fig. 5. Mean number of rosettes (vertical bars, left axis) and flowers per individual (lines with circles, right axis) in 1998, 1999 and 2000 for plants receiving urine treatments and those not receiving urine in 1998. Hashed bars and closed symbols correspond to urine treated plants. Error bars represent 1 standard error of the mean.

per year for plants receiving the simulated urine. By 2 years post-treatment (2000) there was no significant difference in rosette numbers among treatments. Plants receiving urine were also more likely to produce more flowering stalks for the 2 years subsequent to urine application, 1999 ( $F_{1,197} = 6.4$ ; \*\*\*,  $P < 0.001$ ) and 2000 ( $F_{1,184} = 6.4$ ; \*\*\*,  $P < 0.001$ ; Fig. 5). In 1999 and 2000, urine plants on average had twice as many flowering stalks per individual than control and feces treated plants.

Forty-three percent of the plants receiving simulated urine in 1998 flowered in 1999, while only 14% of the non-urine treated plants flowered in 1999 (\*\*\*,  $\chi^2_{1,198} < 0.001$ ). In addition to this difference in the production of flower stalks, the number of flowers that produced multiple nutlets was significantly greater (\*\*\*,  $\chi^2_{1,93} < 0.001$ ) in urine-treated plants (38%) than in non-urine treated plants (12%). This translates into an overall greater seed production for urine-treated plants. Of the 15 randomly selected flowers per plant, urine-treated plants produced  $26.4 \pm 0.56$  seeds while non-urine plants produced  $18.4 \pm 0.64$  seeds.

#### 4. Discussion

*C. flava* is capable of rapid root uptake and incorporation into its leaves of urea N supplied in a one-time pulse. These uptake and incorporation responses are translated rapidly into significantly increased photosynthetic rates, that in turn lead to increased growth, flowering, and seed set for at least 2 years following N application. These plant responses, coupled with the herding behavior and repeated occupation of this site by mule deer imply a considerable potential for mule deer to affect local population dynamics of *C. flava*.

Large mammals have significant effects on nutrient cycling in grassland and forest ecosystems (McNaughton et al., 1988; Pastor et al., 1988; Day and Detling, 1990a; Pastor et al., 1993; Steinauer and Collins, 1995). These effects are often mediated through the concentration of nutrients in spatially restricted patches (Jaramillo and Detling, 1992a,b), and variation in plant species response to these nutrients (Day and Detling, 1990a,b). The absolute amount of mule deer-mediated N redistribution will depend on the particular migration patterns, herd size, and residence times in a given area (Hobbs, 1996). However, our results support the contention that large mammal effects may be significant in arid-land shrub systems.

In our study site, the total estimated N input from Mule deer was high (2430 pellet groups per ha) relative to other studies that have shown anywhere from 250 to 1500 pellet groups per hectare (Bennett et al., 1940; Rogers et al., 1958; Collins, 1981). These high values may be due to increased deer density in our site as a result of local habitat loss to a phosphate mine that has reduced winter grazing habitat in the area (Utah Division of Natural Resources, 1994). This, in turn, translates into a large amount of N redistribution within the study site. We calculated deposits of 2–34 kg N ha<sup>-1</sup> per year for mule deer only, while Saunders (1984) estimated additions of N can range from 5 to 10 kg ha<sup>-1</sup> year for sheep in a managed grazing system. Given that our site occurs in low N soils (Forseth et al., 2001), mule deer could have a large impact on local nutrient budgets for the entire community.

For plants other than N-fixers, leaf  $\delta^{15}\text{N}$  values reflect strongly the integrated soil N available to the plant (Handly and Raven, 1992; Hogberg, 1997). Rapid root uptake of N and incorporation into leaf structures was indicated by our measurements of leaf  $\delta^{15}\text{N}$  values, which were not significantly different before treatment, but changed significantly within a week of urine treatment. These results demonstrate that leaf-level responses to N pulses in *C. flava* can occur within days. One week after treatment, plants also showed an increase in total leaf N levels (Fig. 1b) and incorporation of N into photosynthetic machinery, as evidenced by differences in photosynthetic characteristics (Fig. 1a, Figs. 2 and 4). Studies on other aridland plants have shown root-level responses to nutrient pulses can occur on the same time scale, including species common in our site, i.e. *A. tridentata* (Jackson and Caldwell, 1989; Jackson et al., 1990; Gross et al., 1993). Our data extends these studies by showing that the acquisition of pulsed nutrients is rapidly translated into leaf-level photosynthetic responses in *C. flava*.

Plants adapted to chronically low soil N conditions often do not have the capability to respond to increased resource levels (Grime, 1977; Chapin, 1980). It is hypothesized that along a gradient of increasing habitat productivity, plants will show increased capacity for growth rate, high morphological plasticity, and active foraging for nutrients (Grime, 1977; Chapin, 1980). Apparently the tradeoffs inherent in adaptation to low resource environments restrict the ability of these plants to respond to higher resource levels. This was not the case with *C. flava*. Evidently, the spatial and temporal heterogeneity in the sandy, low N habitats (0.5–4 ppm total N; Forseth et al., 2001) where *C. flava* occurs provide enough high N pulses and/or patches to promote a high degree of phenotypic plasticity in *C. flava*. Our research site typically receives approximately 40% of its yearly precipitation as snowfall. Thus, springtime N pulses due to snowmelt are a regular occurrence in this habitat. Additionally, pulses of N may occur due to rainfall-induced increases in mineralization throughout the spring and fall growing seasons.

The rapid uptake and incorporation of the urea N pulse manifested at the leaf level was not evident in growth and flowering responses until the year after treatment. We anticipated this result, based upon the phenology and growth form of this plant. By the time of N application, all of the current growing season's meristems were already determined. Additionally, without the availability of floral meristems (only pre-reproductive plants were chosen) the current year's photosynthate could not be translated into a reproductive response. Jaramillo and Detling (1992a) showed the same delayed growth response to N application in *Agropyron smithii*, but found no evidence of an increase in reproduction. We hypothesize that the increase in N and photosynthetic carbon fixation allowed for greater resource (carbon and N) storage in *C. flava*'s woody caudex, thus enabling plants to grow larger in the following year and increase flowering for the two growing seasons after treatment (Fig. 5). Chapin et al. (1990) point out that storage reserves in perennial plants are important in changing from a vegetative to a reproductive phase. In addition, seasonal storage of carbon tends to be high for plants in arid environments (Bloom et al., 1985).

In addition to the strong flowering response to N treatment, the increased carbon and N storage in urine-treated plants enabled more individual flowers to develop multiple nutlets. Prior work by Casper (1984) on *C. flava* showed that most flowers abort 2–3 nutlets during development, resulting in a vast majority of flowers with only one mature nutlet under normal field conditions. Casper (1994) suggested that abortion would lead to reduced weight of single-seeded calyxes, perhaps in response to strong selection for increased dispersal distance. Our data suggest that maternal resources are also important in multiple ovule development in *C. flava*. Interestingly enough, Casper (1984) was able to show an increase in multiple seeded calyxes in response to supplemental N in greenhouse grown plants, but not for plants in the field.

Despite the strong individual plant response to patches of high N concentration, population level responses may be limited if only a few plants are located near N patches. We combined our data on

distribution of mule deer pellets in relation to plants with individual plant responses to estimate effects of mule deer herds on population seed production. We designated seed production in the absence of mule deer as  $s$ , and seed production in the presence of mule deer as  $s^*$ . Seed production with mule deer will be a function of  $s$  times the proportional effect of plants located near mule deer N excretions and the response of those plants to the N in a mule deer excretion. Thus:

$$s^* = s(1 + 0.06 \times 0.61 \times 3 \times 2 \times 1.5) \quad (2)$$

where 0.06 is the percentage of the plants in the population within 20 cm of a pellet group (Table 1), 0.61 is the percentage of the plant population that were small, pre-reproductive plants in 1998 (Forseth et al., 2001, i.e. the type of plants we used in this study), plants were three times more likely to flower and produce twice the number of flowering stalks, and produce 50% more seeds than controls (see results). If we assign seed production in a given year a value of 1,  $s^*$  would then be 1.33, i.e. seed production is increased for the small pre-reproductive plants in the population by 33%. The effects on the population as a whole were underestimated in this calculation because only small plants were examined. Nevertheless, the increase in flowering and in turn seed production from small plants represents a major contribution to population growth in at least two ways; first, by increasing seed production and second, by decreasing the average age of first flowering. The intrinsic rate of increase of populations ( $\lambda$ ) is highly influenced by age to first reproduction (Caswell, 2001).

There are other potential population and evolutionary phenomena that could result from unpredictable patches of high N in habitats supporting large mule deer herds. Nutrient patches may play a role in the maintenance of genetic diversity within populations (Hartgerink and Bazzaz, 1984) by allowing potentially less fit genotypes the ability to reproduce and set seed. They may also promote phenotypic plasticity within the population because of the fitness benefits of being able to exploit patches and/or pulses of high soil N. Future work will involve examining the entire range of plant sizes and ages in the population to examine these

implications as well as effects of nitrogen pulses on population dynamics.

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