# UNIVERSITY OF CALIFORNIA, SANTA CRUZ

# EFFECTS ON SOIL CARBON AND NITROGEN CYCLING ASSOCIATED WITH GRAZING BY DIFFERENT SPECIES IN A KANSAS TALLGRASS PRAIRIE

A Senior Thesis submitted in partial satisfaction of the requirements for the degree of

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by

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ABSTRACT: The world's soils are an enormously important sink of atmospheric CO<sub>2</sub>, sequestering significantly more carbon. Consequently, alterations to soil carbon cycling can have huge implications concerning climate change. In cases of heavy grazing, intensity-reduction or exclusion has been shown to increase primary productivity and sequester carbon, as opposed to release. However, these studies focus on sheep or cattle-grazed land. It has been almost 30 years since the near-extinction of the Plains bison in Midwest America and their numbers are returning. Aboveground comparisons have been made between cattle and bison-grazed lands, but not much has been researched belowground. This experiment conducted on Konza Prairie Biological Station (KPBS), a tallgrass prairie in Manhattan, Kansas, analyzes root and soil properties, using microbial biomass carbon (MBC), total nitrogen (TN) and total carbon (TC) as proxies for carbon and nitrogen cycling. MBC content was over 60% greater, while TC and TN were 40% and 35% greater respectively in the bison-grazed site compared to cattle-grazed. These differences are quite significant and could have long lasting effects on soil nutrient cycling in middle North America.

KEYWORDS: Konza Prairie Biological Station, Soil carbon/nitrogen cycling, Grazing, Climate change

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## Introduction

There is more carbon stored in the soils of the world than there is in the atmosphere; so ecosystem-level fluctuations in carbon cycling and sequestration can have profound effects on global warming and climate change (Davidson and Janssens, 2006). This study uses soil microbial biomass carbon (MBC) measurements as an indication of soil carbon cycling to compare bison (*Bos bison*) and cattle (*Bos taurus*) grazed lands with an ungrazed control. MBC can be useful as such an indicator because it has numerous direct and indirect effects on soil carbon pools and their turnover, in addition to the biomass being a carbon pool itself. For example, microbial communities dominated by fungi exhibit increased turnover rates and soil microbes are known to enhance aggregation, which can physically entrap soil organic matter (Six et al., 2006).

The reintroduction of the Plains bison to Midwest America has been hailed as a great conservation success, however little is known about the grazing effects of bison in comparison to cattle grazing. The majority of research concerning the ecological effects of the two ungulate species focuses mainly on how differences in behavior and resource selection affect plant community composition and diversity. Previous studies have suggested that grassland grazed by bison has been shown to have a more rapid increase in annual and perennial forb cover as well as a trend towards greater overall species richness. However, despite observations of slight alterations to some vegetation components, the plant communities did not vary significantly between the bison and cattle pastures. Instead, variations in how the herbivores are managed have been hypothesized as the major contributors to differences between pastures grazed by the two species (Towne, Hartnett, & Cochran, 2005). This study will see how these differences affect soil properties and if they translate into variations in soil carbon and nitrogen pools.

## Methods

Study Site



Figure 1. Ariel photograph of Konza Prairie Biological Station depicting the sampling locations in each of the three treatment sites as indicated by the red circles (Konza Prairie Biological Station, 2015).

Samples were collected on Konza Prairie Biological Station (KPBS), a 3,487-hectare native tallgrass prairie located in the Flint hills of Northeastern Kansas (39°05' N, 96°35' W) owned by The Nature Conservancy and Kansas State University. Three sites were chosen on KPBS with care taken to ensure the same soil type, Benfield-Florence complex, and microclimate conditions such as slope and accessibility by grazers. Figure 1 displays the three sampling sites, with one site harboring the natural grazers of the area, the Plains bison (N1B), another being home to cattle (C1A), and the final site being an ungrazed control (1D). All three sites are burned annually in the spring, with the most recent controlled burns taking place in the end of March and the beginning of April, 2015.

#### Cattle and Bison Management

The goal when managing the stocking rate at KPBS is to remove 25% of the available forage annually, which works out to approximately eight grazable acres per cow/calf pair and 11 grazable acres per bison. A notable difference between the two plots is that the bison graze year-round, while the cattle have a grazing season of May through October. However, this is a normal practice in cattle-grazed systems.

#### *Experimental design*

There is inevitably going to be variability in such a complex, natural ecosystem, so in an attempt to compensate and observe statistically significant effects of the treatments a mass bulking approach was utilized.

Samples were taken at each site during a three-day period at the end of August 2015. In order to isolate the effect of the treatment, grazing, care was taken to randomize the sampling design by throwing a ball in a random direction when setting up the transects. Two 45 meter transects were created for each treatment. Evenly spaced along each transect (every 9 meters), five 1-square meter quadrats that had been divided into 16 equal (.0625m<sup>2</sup>) squares were placed. Then, a random number generator was used to pick five of the .0625m<sup>2</sup> squares, in which 1-inch diameter soil cores were taken to a depth of 10 centimeters. Additionally, a 2-inch diameter soil core was taken at each quadrat location and wrapped in plastic wrap for determination of bulk density and gravimetric water content. The 1-inch cores taken within each quadrat were homogenized and the roots picked for determination of root biomass, C%, and N%. All samples were refrigerated at 4 degrees Celsius until analyses could be completed.

#### Root Analysis

The roots picked from the homogenized 1-inch soil cores were used to compare average root density, then were ground in a ball mill grinder and weighed for carbon and nitrogen elemental composition. Carbon and nitrogen elemental composition was determined by Dumas combustion using a Carlo Erba 1108 elemental analyzer coupled to a ThermoFinnigan Delta Plus XP isotope ratio mass spectrometer. Analyses were run at the University of California, Santa Cruz Stable Isotope Laboratory.

#### Soil Analysis

Substrate-induced respiration (SIR) measurements were taken using the protocol outlined in Anderson & Domsch (1978) to get an indication of microbial biomass carbon (MBC). MBC was estimated from the substrate-induced respiration measurements, while carbon availability was determined by the ratio of basal respiration to substrate-induced respiration. (Samples had to be transported back to the University of California, Santa Cruz before SIR could be completed, so the results may be slightly skewed). The remainder of the soil from the homogenized 1-inch cores was oven-dried at 105 degrees Celsius for 72-hours. The oven-dried soil was then ground, weighed, and analyzed for carbon and nitrogen elemental composition using the same method outlined above.

#### Statistical Analysis

One-way ANOVA was used to compare the effects of the grazing treatments using the variables in question. JMP Pro 12 was used for all statistical analyses and the significance level was set at P < 0.05.

# Results

Bulk Density





\*Site N1B was very rocky making it extremely difficult to take intact cores.

The bulk density was slightly greater in the site hosting cattle, compared to the bison and

ungrazed sites, however there is no statistically significant difference between them (Figure 2).



Figure 3. Mean total soil carbon values for the three treatment sites ( $\mu$ g C g<sup>-1</sup> of soil). (1D - n=9, C1A - n=7, N1B - n=9)

There is a trend of increasing TC associated with grazing as well as the bisongrazed plot having a 40% increase in TC when compared to cattle-grazed. C1A and N1B are not statistically different (Figure 3).



Figure 4. Mean total soil nitrogen values ( $\mu$ g N g<sup>-1</sup> soil) for the three treatment sites. (1D – n=9, C1A – n=7, N1B – n=9)

Similar to TC, there is a trend of increasing TN associated with grazing in addition to the bison-grazed plot having a 35% increase in TN when compared to cattle-grazed. However, C1A and N1B are statistically different (Figure 4).



Figure 5. Mean carbon:nitrogen ratio of roots across the three sites. (1D - n=10, C1A - n=9, N1B - n=9)

C:N analysis of roots showed the ungrazed site, 1D, having a significantly greater ratio than the two grazed sites. There was no significant difference in the C/N content of roots from either of the grazed sites, but N1B did have a slightly lower mean value (figure 5).



Figure 6. Mean carbon:nitrogen ratio for soil across the three treatment sites. (1D - n=9, C1A - n=7, N1B - n=9)

The C:N of the soil was similar between the three sites with values between 11 and 12, but there was still a statistically significant difference between them. 1D had the greatest C:N with a mean of 11.98, followed by N1B and C1A with 11.58 and 11.12, respectively (Figure 6). *Microbial Biomass* 



Figure 7. Mean microbial biomass carbon in  $\mu$ g carbon/g soil for the three treatment sites. (1D, C1A, N1B – n=10)

The ungrazed control had the lowest mean soil microbial biomass carbon followed by the site with cattle having a slight increase, and finally the bison grazed site with the greatest (Figure 7). Microbial biomass carbon is approximately 60% greater at the bison grazed site when compared to the cattle grazed site. All means are statistically significant.





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Figure 8. Mean root density in kilograms, determined by dry weight, per cubic meter of soil. (1D - n=9, C1A - n=7, N1B - n=9)
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The density of roots in sites 1D and C1A was not significantly different, but N1B had a value about half that of the other two sites (Figure 8).

## Discussion

#### Bulk Density

From the data in this experiment, there does not seem to be an effect of grazing on the bulk density of the soil, as there is no significant difference between the values for the three sites. However, sampling for bulk density was very difficult at N1B because there were many rocks throughout the site, which made taking intact 2-inch cores challenging. Studies on the movement patterns of the two grazers propose cattle would create the most compaction because they tend to congregate in specific areas and avoid roaming, however this seems to have had little effect in this case (Kohl et al., 2013). Through observation N1B seemed to be the most dense when sampling, although there were a fair amount of rocks, and 1D was definitely the least. The ungrazed control had very dense, tall grasses resulting in an array of small grass roots that create an abundance of micro-pores, and would logically have the lowest bulk density, so further sampling should be carried out before a definitive conclusion can be made.

## Root Density

Comparison of root density between the plots showed no significant difference between C1A and 1D, but the mean root density in N1B was approximately one kg/m<sup>3</sup> less than the other two sites. The low value for N1B may be a consequence of rocks in the soil simply displacing roots within the cores while sampling. It is surprising that C1A and 1D have such similar root densities because of the extremely dense, tall grasses in site 1D. However, previous studies have shown an increase in belowground biomass and carbon allocation associated with grazing (Wang et al. 2015, Hafner et al 2012). Future studies could look closer into the differences in root densities and organic matter decomposition to determine the influence of rhizosphere processes on carbon cycling in these systems. Rhizosphere priming, or altered SOM decomposition in the root zone, can have dramatic effects on soil carbon and therefore when combined with this data can provide more insight into the cycling of these systems (Cheng, 2009).

#### Elemental Analysis

The C:N ratio of the roots show an effect of grazing, with the value for 1D being much higher than those of C1A and N1B, but there was no significant difference between the two grazed sites. This can be attributed to the differences in plant community composition between the grazed and ungrazed sites. For example, it is evident through observation, but studies have also shown dramatic increases in species richness and aboveground diversity associated with grazing (Boughton, et al. 2016). Additionally, it is known that graminoids have a higher C:N than forbs and other herbaceous plants (Towne, Hartnett, & Cochran, 2005). Therefore as a result of predominately graminoid species growing in the ungrazed plot, the root C:N is much higher than those of the grazed plots which have a variety of forbs and herbaceous plant species growing on them.

The values for total nitrogen (TN) and total carbon (TC) in the soils reflect the results of the microbial biomass carbon analysis and therefore can give insight into soil carbon and nitrogen pools in these two systems. The bison-grazed plot had a 35% and 45% increase in TN and TC respectively when compared to the cattle-grazed. Plant community composition has cascading effects on microbial processes and nutrient cycling which can lead to significant effects on soil carbon and nitrogen content (Hooper & Vitousek, 1998). Therefore, the increase in TC and TN in the soils of N1B could be attributed to the known preferences in resource selection between the two ungulates and the resulting differences in plant community composition (Towne, Hartnett, & Cochran, 2005).

The C:N of the soil for the three sites, despite being statistically different, were very similar with all three values falling between 11.12-11.98. However, globally the majority of soil C:N ratios fall within this range, so even a slight variation is a notable difference (Batjes, 1996). Also, feedback from the stable isotope laboratory indicated that high levels of organic matter in the soil resulted in incomplete combustion and chromatography. Therefore, decreasing the sample size for EA-iRMS analysis would provide more accurate results if the analysis were to be repeated.

#### Microbial Biomass

The trend of increasing microbial biomass carbon associated with grazing is supported by studies that found more carbon being allocated belowground when aboveground biomass is being continually removed (Hafner et al, 2012). With more carbon being sent belowground, there is an increased food source for the microbial population as a result of increased root turnover, presence of more root exudates, or both. Similarly, a previous study by Aldezabal et al. (2015) found that an increase in root density and exudation in the top 10 centimeters of the soil as a result of grazing led to increased microbial activity. Likewise, an important factor is the quality (C:N primarily) of the food source for the microbial population. Microbes need a food source with a C:N of approximately 24:1, with anything above that slowing down decomposition. Therefore, root turnover and exudation, being one of the only fresh organic matter inputs below the surface, can greatly influence microbial activity. With 1D having such a high root C:N ratio (mean = 77.2) compared to C1A (52.1) and N1B (44.6), it is not surprising the MBC was lower.

The results for microbial biomass carbon seem higher than anticipated for a healthy prairie soil, but this could be explained by the amount of time between sampling and analysis. There was a three week period before substrate induced resperation was completed when the samples were transported from Kansas to the University of California, Santa Cruz. Even though the soil was refrigerated or kept in a cooler with ice packs at all times to maintain a temperature of 4 degrees celsius, metabolic activity over time would utilize some of the available carbon to grow the microbial population.

# Conclusions

The positive effects of grazing on carbon cycling and the associated implications for carbon sequestration and global warming have been researched in different systems around the globe (Hafner et al., 2012; Wang, 2015, Han et al., 2016), but only for cattle grazed lands and traditionally through grazing exclusion. This study supports those findings and suggests that in a tallgrass prairie, the effect may be even greater when bison as opposed to cattle graze the land. Elevated levels of microbial biomass carbon, total nitrogen, and total carbon equate to increased carbon and nitrogen cycling as well as greater pools sequestered in the soil.

Further research should be done to quantify the direct effects on carbon and nitrogen cycling in lands grazed by various ungulates, as the implications in the United States alone could be dramatic. Bison have largely recovered since the 1980's when over thirty million were killed, and currently graze a large portion of the American Midwest. Their reintroduction and role as grazers could have lasting effects on the battle against global warming and should not be dismissed (Craine, et al. 2015).

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