

Soil carbon and nitrogen after eight years of rotational grazing in the Nebraska Sandhills meadows

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ABSTRACT

Grassland provides many ecosystem services; therefore, sustainable management practices of grassland are crucial for maintaining and enhancing its ecosystem health and resilience. Rotational grazing at a high stocking density (a.k.a., ultrahigh stocking density) is purported to sequester greater amounts of carbon (C) in grassland soils than rotational grazing at low stocking densities. This study was conducted in the Nebraska Sandhills meadows for eight years to evaluate how rotational grazing with different stocking densities can affect soil C and total nitrogen (TN) in bulk soils, soil organic matter fractions, and sequestration rate. The grazing management included a high stocking density with one grazing cycle (MOB), a 4-pasture rotation system with one grazing cycle at a low stocking density (4PR1), and no grazing (CNT). Results showed that soil C and N contents were higher in the total particulate organic matter fraction than in the soil mineral-associated organic fraction at the 0–10 cm depth and visa versa at the 10–20 cm depth. Grazing management did not affect C and N contents in the bulk soils (SOC averaged 28.19 g C kg⁻¹ and TN averaged 2.67 g N kg⁻¹ at the 0–10 cm depth). Both MOB and 4PR1 grazing treatments recorded similar C and N contents in all SOM fractions. However, the MOB grazing treatment increased C and N contents in the macro-aggregate occluded particulate and dissolved organic matter fractions compared to the CNT. The 4PR1 grazing treatment increased soil C and N contents in the dissolved organic matter and mineral-associated organic matter fractions and increased SOC sequestration rate compared to the CNT. Rotational grazing at a low stocking density for one cycle appears to enhance long-term SOC accumulation in these Sandhills meadows soil.

1. Introduction

Grasslands make up approximately half of the land on earth (Wang and Fang, 2009), emphasizing its importance as a potential carbon (C) sink. Further, healthy grasslands provide benefits such as enhancing environmental quality and improving vegetation productivity and, thus, food security (Teague et al., 2013). For example, the Nebraska Sandhills, one of the most intact semiarid rangelands in the Great Plains of North America characterized by a mixed-grass ecoregion, provides a diverse range of habitats for wildlife and regulating services (Stephenson et al., 2019). Sustainable management of rangelands is essential for maintaining and, more importantly, enhancing its ecosystem health and resilience (Wagner et al., 2021; Guretzky et al., 2020; Booker et al., 2013).

In rangelands, grass (aboveground and belowground) affects soil

organic matter (SOM) formation, the important storage for soil C and nitrogen (N) (Chen et al., 2015; Fisher et al., 1994). Soil organic matter is an important indicator of soil health in rangelands, examples being its ability to provide soil nutrients for vegetation, enhancing aggregate stability, and increasing soil water retention (Cotrufo and Lavelle, 2022; Lal, 2020; Yu et al., 2020). Stabilization of the SOM either through chemical stabilization (adsorption of SOM onto mineral surfaces) or through physical stabilization (occlusion of SOM within the aggregate) facilitates the longer soil C residence time, thus having a significant impact on soil C sequestration (Cotrufo et al., 2019; Blanco-Canqui and Lal, 2004; Torn et al., 1997).

Management practices such as grazing can affect SOM as clarified in our conceptual diagram (Fig. 1, Jing et al., 2023; Zhang et al., 2022; Liu et al., 2015). The enhancement of SOM is important in most grazing ecosystems, including Nebraska Sandhills rangelands. One of the

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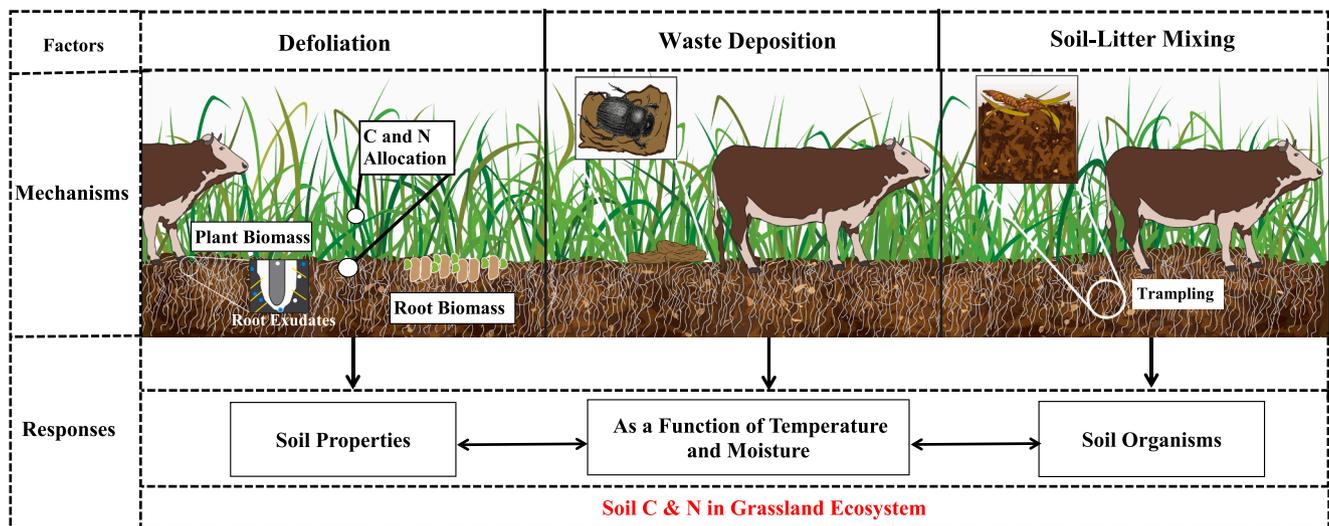


Fig. 1. Conceptual diagram illustrating the potential pathways by which grazing can affect soil organic matter, ultimately affecting soil carbon and nitrogen levels (Drawing illustration by Lana Johnson).

important mechanisms by which grazing management can influence SOM is plant defoliation (Fig. 1). Defoliation intensity can affect C and N allocation within plant-soil system, thus impacting SOM formation and plant growth (Ma et al., 2021). High defoliation frequency can reduce a large amount of photosynthetic tissue, leading to plants allocating more C aboveground to build new tissue; which can affect root biomass and exudates (Bicharanloo et al., 2022). According to Zhang et al., (2021a), defoliation timing can also affect forage production and subsequent SOM inputs, with defoliation during peak growth increasing plant production compared to early defoliation. Defoliation can also affect litter decomposition (Fig. 1), because it removes part of the plant, which increases the penetration of solar radiation into the soil, thus, increasing soil temperature and promoting microbial activity (Butenschoen et al., 2011). Defoliation can also influence species diversity, thus affecting quantity and quality of litter being produced and root production (Li et al., 2022). In addition, in a grazed ecosystem, deposition of animal excreta adds C and N to the soil (Evans et al., 2019, Fig. 1). Both urine and dung contain significant amounts of C and N, which can be incorporated in the soil through soil fauna (Zhu et al., 2021; Lovell and Jarvis, 1996). Whitehead (1995) suggested that the high percentage of N consumed by grazing animals (approximately 75–90 %) is returned to the soil in the form of urine and dung. Grazing management can also affect SOM through its effect on soil-litter mixing (Fig. 1) through hoof trampling, which affects microbial activity and soil properties (Wei et al., 2021; Ludvíková et al., 2014). Soil microbial processes are the primary processes responsible for the C and N transformations (Teutscheroová et al., 2021).

Various grazing management methods are practiced on semiarid rangelands, with rotational grazing being promoted as a sustainable rangeland grazing approach for its ability in enhancing soil, forage and livestock productivity (Wagner et al., 2021; Enri et al., 2017; Teague et al., 2013). Under rotational grazing, a grazing unit is divided into multiple paddocks with livestock rotated through the paddocks (Mosier et al., 2021). A number of studies reported that rotational grazing management, such as stocking density (relationship between number of animals and the specific unit of land being grazed at a point in time) and rest period, can affect grazing distribution, animal wastes, and soil productivity (James et al., 2017; Peterson et al., 2013; Barnes et al., 2008).

In this study, we conducted a field experiment for 8-yr with three grazing treatments varying in grazing densities (high and low) and number of grazing cycles (one and two) per year. We initiated grazing during the vegetative stages of grasses to be more susceptible to

trampling. Data published from this grazing research documented that trampling was higher on the high stocking density grazing pastures (also known as MOB grazing) than the low stocking density grazing pastures, possibly because the animals on the MOB pastures graze in a smaller area for a short period (Andrade et al., 2022; Guretzky et al., 2020). While low stocking densities and long grazing periods result in selective grazing, the short grazing periods and ultrahigh stocking densities (MOB grazing) result in extremely high grazing pressure (i.e., animal demand per unit forage), thus reducing the grazing animal's ability to selectively graze available vegetation and what is not grazed is trampled into the soil (Bork et al., 2021; Harmel et al., 2021; Roberts and Johnson, 2021). Reed et al. (2019) has also suggested that MOB grazing in the US Midwest region increased the trampling of vegetation compared to a low stocking rotational grazing, potentially increasing the SOM content.

Rotational grazing at an ultrahigh stocking density such as MOB grazing has gained much attention from both the rancher and scientific communities. However, due to the lack of consistent results, recommendations for implementation of MOB grazing must be given with caution. Several researchers reported that ultrahigh stocking density and short grazing periods had positive effects on soil, such as increasing soil microbes biodiversity (Teutscheroová et al., 2021) when compared to continuous grazing, increasing N stock (Contosta et al., 2021) when compared to no grazing, and increasing C storage (de Otálora et al., 2021) when compared to rotational grazing with a shorter rest period and a longer grazing period. On the other hand, other researchers reported that ultrahigh stocking density and long grazing periods negatively affected soil productivity compared to no grazing (Roberts and Johnson, 2021) and compared to continuous grazing (Dowhower et al., 2020).

The effect of rotational grazing at a high stocking density on grass production compared to continuous grazing was not consistent, with an increase in the grass production after twenty years of rotational grazing as reported by Teague and Dowhower (2022) or no effect on the grass production after five years of rotational grazing as reported by Augustine et al. (2020). These contradictory results show that the mechanisms behind the effects of MOB grazing on soil productivity are not fully understood yet.

While the response of above ground vegetation production, species diversity and livestock performance to grazing management is well understood by ranchers (Lindsey, 2016; Stephenson et al., 2015; Schacht et al., 2010), what is happening belowground related to soil C and N storage based on various grazing management approaches is not yet well understood. The objectives of this study were to evaluate how rotational

Table 1

Grazing information for Nebraska Sandhills meadows from 2010 and 2017. Stocking density and stocking rate varied slightly each year because of differences in yearling cattle weight. 4PR1, the 4-pasture rotation system with one grazing cycle; 4PR2, the 4-pasture rotation system with two grazing cycles; MOB, the ultrahigh stocking density system with one grazing cycle; and CNT, no grazing.

Management	4PR1	4PR2	MOB	CNT
No. of pastures	4	4	120	1
Hectare per pasture	0.435	0.642	0.058	0.435
Grazing cycles	1	2	1	
Grazing start date	Mid-June	Late-May	Mid-June	
Grazing duration (d)	60	80	60	
Grazing period (d)/pasture	15	10	0.5	
No. of animals	9	10	36	
Stocking density (kg live weight ha ⁻¹)	7,138	5,374	214,138	

grazing at a range of stocking density can affect C and N in bulk soils and SOM fractions in the Sandhills subirrigated meadows of Nebraska. We hypothesized that MOB grazing with an ultrahigh stocking density would result in higher soil C and N levels relative to rotational grazing treatments at low stocking densities due to the increased vegetation trampling (Andrade et al., 2022; Guretzky et al., 2020).

2. Materials and methods

2.1. Study site

This study began in 2010 and continued to 2018 at the northeastern Sandhills meadows of the University of Nebraska-Lincoln (42°13'N; 99°38'W). Meadows are seasonally wet in early to late spring due to the rising water table. The climate is semiarid in this region. Weather data from 2010 to 2018 at the experimental site was published in Andrade et al. (2022). The mean annual precipitation was 612 mm and the mean temperature was 17.5 °C. The highest annual precipitation was observed in 2018 (840 mm) and the lowest was recorded in 2012 (280 mm) (Andrade et al., 2022). Soils on the meadows are sandy to a fine sandy loam texture. Soil property measurements in 2018 were conducted (pH = 6.4, electrical conductivity (EC) = 0.26 mmho cm⁻¹, nitrate-nitrogen (NO₃-N) = 20 mg kg⁻¹, phosphorus (P) = 35 mg kg⁻¹, potassium (K) = 103 mg kg⁻¹, calcium (Ca) = 1 853 g kg⁻¹, and magnesium (Mg) = 166 mg kg⁻¹).

Vegetation was dominated by non-native cool season grasses (*Phalaris arundinacea* L., *Bromus inermis* Leyss., *Poa pratensis* L., *Elymus repens* (L.) Gould), *Phleum pratense* L.), rushes (*Eleocharis* and *Juncus* spp.) and sedges (*Carex* spp.). Several forbs and native warm season grass species were present and contributed about 15 to 18 % of the annual production (Shine et al., 2022; Wagner et al., 2021). Annual litter accumulation and vegetation production in this site averaged approximately 2 194 kg ha⁻¹ and 5 100 kg ha⁻¹, respectively (Andrade et al., 2022; Guretzky et al., 2020). Prior to the start of the grazing study, this meadow site had been

hayed during mid-summer for the past 40 years.

2.2. Grazing management

This study quantified the effect of grazing management for eight years (2010–2017) on soil C and N levels at the Nebraska Sandhills meadows. Detailed information about the grazing management can be found in Table 1. This study had four grazing treatments: a 120-pasture, ultrahigh stocking density system with one grazing cycle (MOB), a 4-pasture rotation system with one grazing cycle (4PR1), a 4-pasture rotation system with two grazing cycles (4PR2), and no grazing (CNT) as a control (Table 1). Each grazing treatment had two replicates and the experimental design was a randomized complete block. The rotational pastures were divided into smaller paddocks and had different stocking densities depending on the treatment. Over the years, grazed treatment stocking rates averaged 7.9 AUM ha⁻¹ with a range from 7.4 AUM ha⁻¹ to 8.4 AUM ha⁻¹.

For the MOB grazing treatment, cattle were grazed at ultra-high stocking densities (214 138 kg live weight ha⁻¹) and were moved to a new paddock two times per day. These cattle grazed each paddock once each season. The MOB grazing treatment had 120 paddocks (0.058 ha each) grazed by 36 yearling steers over a 60-day period. Each year, the grazing for the MOB treatment started in mid-June (peak cool-season grass biomass, Stephenson et al., 2019). Stem and upright plants dominate the vegetation in this period, thus trampling will be optimal.

For the 4PR1 grazing treatment, cattle were grazed at low stocking densities (7 138 kg live weight ha⁻¹) and were moved to a new paddock after 15 days. These cattle grazed each paddock once each season. The 4PR1 treatment included four 0.435-ha paddocks grazed by nine steers for 15 days each, resulting in a 60-day grazing season. The grazing start date for the 4PR1 treatment was mid-June, which is similar to the grazing start date as the MOB treatment, so the effects of these treatments on soil and plant productivity will be comparable.

For the 4PR2 grazing treatment, cattle were grazed at low stocking

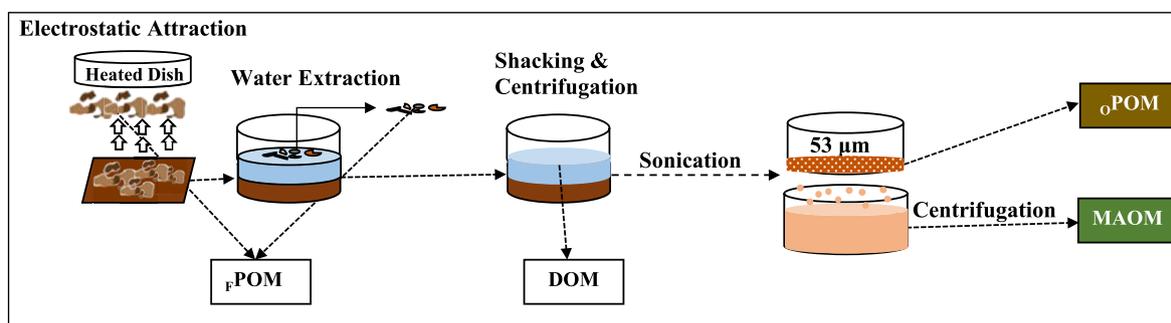


Fig. 2. Procedure for fractionating soil to free particulate organic matter (fPOM), dissolved organic matter (DOM), macro-aggregate occluded particulate organic matter (oPOM), and mineral-associated organic matter (MAOM), adapted from Jilling et al. (2020). First, fPOM was separated by electrostatic attraction and water extraction. Second, the remaining portion of the soil sample and the solution were then shaken, centrifuged and the supernatant was considered as DOM and the pellet remaining was subjected to an ultrasonication and wet sieved using a 53- μ m sieve to separate oPOM from MAOM.

densities (5 374 kg live weight ha⁻¹) and were moved to a new paddock after 10 days. These cattle grazed each paddock twice each season. The 4PR2 treatment had four 0.642-ha pastures grazed by 10 steers for 10 days in each for the two cycles, resulting in an 80-day grazing season. The grazing start date for the 4PR2 treatment was late May, 20 days earlier than for the MOB and 4PR1 treatments. This grazing treatment is commonly used in this region to improve the use of relatively high-quality plant forage (Andrade et al., 2022). The three grazed treatments ended on the same date in August. Water was moved with the cattle as they were moved from pasture to pasture in all grazing treatments. Lastly, the CNT treatment had no cattle present throughout the grazing season.

2.3. Soil sampling

Soil sampling was conducted in April 2010 (prior to the establishment of treatments) and April 2018 (end of the grazing). Samples were collected from two depths (0–10 cm and 10–20 cm) using a handheld soil probe, 1.5 cm in diameter, after all the residues from the soil surface were removed. Soil samples were collected from the 4PR1, 4PR2, MOB, and CNT. The number of samples taken per treatment from each treatment in each year was equivalent to the pasture sizes, with a goal of representing the total variation within the pasture. Fig. S1 showed the number of soil samples taken from each grazing treatment. For each replicate, soil number collected in 4PR1 and 4PR2 was 24 each, in MOB was 60, and in CNT was 6 (Fig. S1). Soil samples were combined for 4PR1, 4PR2, and MOB to create six replicates from each treatment (Fig. S1). All soil samples were refrigerated until laboratory analysis was conducted.

The core ring (1.5-cm in diameter) method was used to determine the bulk density (Bd) from all plots in 2010 and 2018. Soil cores were divided by depth at 0- to 10-cm and 10- to 20-cm depths Soil Bd was calculated using the following equation:

$$Bd(\text{g cm}^{-3}) = \text{mass of dry soil} / \text{soil volume} \quad (1)$$

Soil moisture content for all treatments at the time of Bd sampling in both years was measured and found similar for all treatments in each year. Before conducting the following soil analysis, soil samples were tested for the presence of carbonates by penetrating with 10 % hydrochloric acid. The test confirmed no inorganic C in the soil.

2.4. Soil measurements

2.4.1. Soil carbon and nitrogen in bulk soils

Soil organic C and TN in bulk soils were determined for all grazing treatments in 2010 and 2018. Soil samples at each depth were air-dried, and milled into a powder (~0.149 mm). Soil organic C and TN concentrations were determined using a 2000 Organic Elemental Analyzer

(CE Elantech Inc). Soil organic C or TN stocks (C stock and N stock) for 2010 and 2018 were calculated for all grazing treatments using the following equations (Shi and Han, 2014):

$$C_{\text{stock}} = \text{SOC} \times \text{Bd} \times \text{H} \times 10 \quad (2)$$

$$N_{\text{stock}} = \text{TN} \times \text{Bd} \times \text{H} \times 10 \quad (3)$$

where H is the soil depth (m), and 10 is a factor to adjust units.

Because soil Bd may change over time, which can result in an inaccurate estimate of C and N stocks, the C and N stock values in 2018 were multiplied by the change in Bd (Bd in 2010/Bd in 2018) to adjust for Bd change.

Soil C and N sequestration rates were calculated using the following equations (Zhang et al., 2021b):

$$\text{Soil C sequestration rate} = (C_{\text{stock 2018}} - C_{\text{stock 2010}}) / t \quad (4)$$

$$\text{Soil N sequestration rate} = (N_{\text{stock 2018}} - N_{\text{stock 2010}}) / t \quad (5)$$

where t is the duration of this experiment (years).

2.4.2. Soil carbon and nitrogen in soil organic matter fractions

Soil organic matter fractionation was conducted for all grazing treatments in 2010 for the 0–10 cm depth and in 2018 for both the 0–10 cm and 10–20 cm depths. Soil organic matter was fractionated (Fig. 2) using a particle size-based technique that combines electrostatic attraction, sonication, and centrifugation as described in Anuo et al. (2023), Greenberg et al. (2019), and Jilling et al. (2020). These fractions included particulate organic matter (which consists of free particulate organic matter (fPOM) and macro-aggregate occluded particulate organic matter (oPOM)), mineral-associated organic matter (MAOM), and dissolved organic matter (DOM). Free particulate organic matter was first separated from soil by electrostatic attraction, and then the soil was mixed gently with water in a centrifuge bottle to keep aggregate disruption at a minimum. The suspension was then allowed to settle for five minutes. The fPOM floating on the surface of the suspension was removed by pipetting the supernatant over a 53 μ m sieve (Kaiser et al., 2010), then combined with the electrostatically isolated fPOM (Fig. 2). The remaining portion of the soil sample and the supernatant that passed through the 53 μ m sieve was then shaken for 12 h at 10,000 x g for 30 min and centrifuged, and the final supernatant was filtered using a membrane filter (0.45 μ m). The filtrate was used for quantifying DOM.

The pellet remaining in the centrifuge bottles was transferred to glass beakers and subjected to a low energy ultrasonication at 60 J cm⁻³ to disperse macro-aggregate occluded organic fraction, then the sample was wet sieved using a 53- μ m sieve. Sample material above the sieve represented oPOM while the material below the sieve represented MAOM (Fig. 2). Samples of free and macro-aggregate occluded particulate, and mineral-associated organic fractions were freeze-dried and

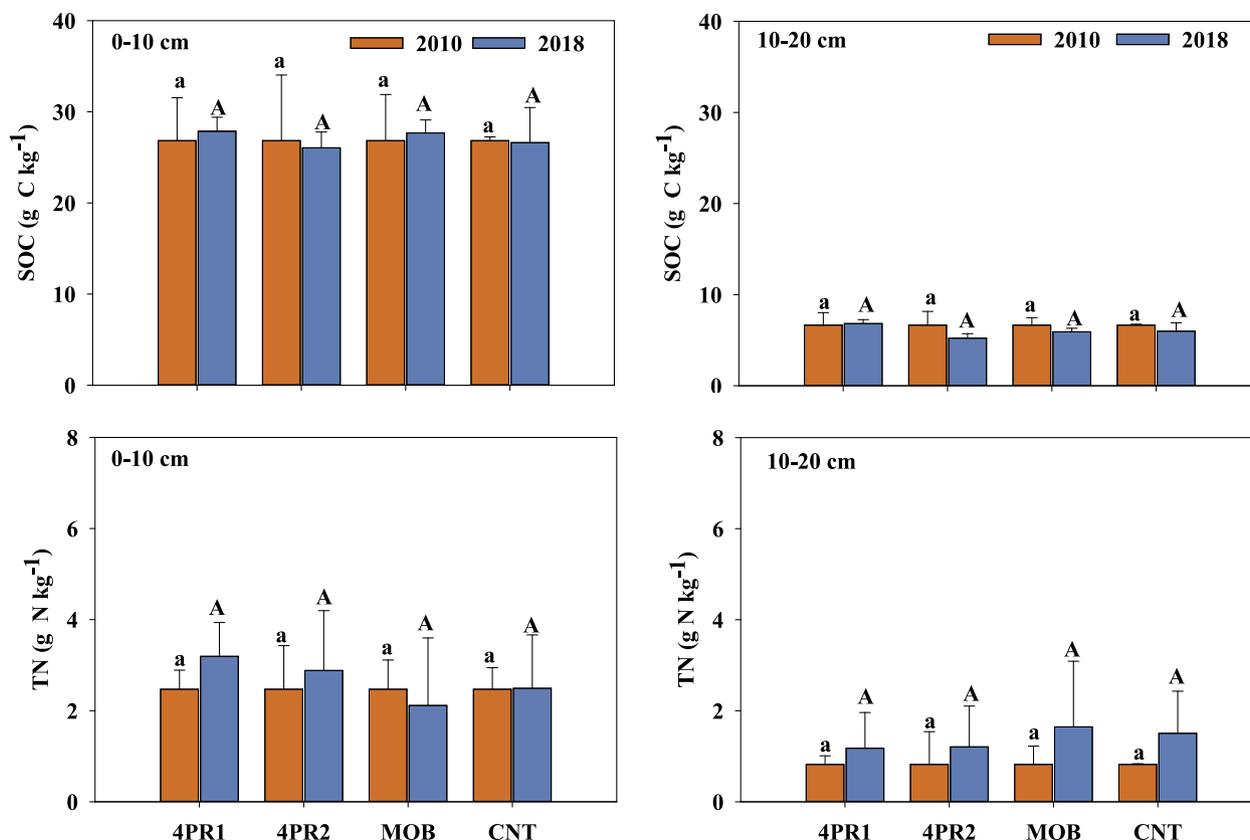


Fig. 3. Soil organic C (SOC) and total N (TN) under the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) in 2010 and 2018 at the 0–10 cm and 10–20 cm depths at Barta Brothers Ranch, Nebraska Sandhills meadows. Vertical bars indicate standard errors of the means. Different lower case letters indicate a significant difference among treatments in 2010, and different capital letters indicate a significant difference among treatments in 2018 at the 0.05 probability level.

weighed. These samples were subsequently milled into powder, and C and TN concentrations were determined using above Analyzer. Samples for water extractable OM were measured for dissolved organic C and N using a Total Organic Carbon Analyzer (OI Analytical Aurora, Baltimore, MD). Carbon and TN associated with SOM of different fractions were calculated by multiplying C or TN concentrations (g kg^{-1} of soil in fraction size) by the percent weight of the fraction. The C and TN contents in the SOM fractions will be referred to with $-C$ and $-N$ at the end of these fractions, in the form of ${}_F\text{POM-C}$, ${}_F\text{POM-N}$, ${}_O\text{POM-C}$, ${}_O\text{POM-N}$, MAOM-C , MAN-N , DOM-C , and DOM-N .

2.5. Statistical analysis

Analysis of variance was conducted on soil C and TN data using the MIXED model in SAS 9.3. For each depth, treatments were considered as a fixed factor and block as a random factor. For treatment comparisons, an LSD Fisher test was used at a 0.05 probability. We conducted principal component analysis to evaluate how grazing management affected each soil parameter. To define the relationships among soil C and N parameters, correlation analysis was conducted.

3. Results

3.1. Carbon and nitrogen concentrations in bulk soils as affected by grazing over eight years

The means for SOC and TN for bulk soils under all grazing managements in 2010 and 2018 at the 0–10 cm and 10–20 cm depths are shown in Fig. 3. The SOC and TN for bulk soils were not significantly different among treatments in either year and at either depth (p values

> 0.05 for 0–10 cm and 10–20 cm depths). In 2010, the means for SOC and TN were $26.44 \text{ g C kg}^{-1}$ and 2.56 g N kg^{-1} , respectively, at the 0–10 cm depth and 6.57 g C kg^{-1} and 0.86 g N kg^{-1} , respectively, at the 10–20 cm depth (Fig. 3). In 2018, SOC averaged 28.19 and 5.99 g C kg^{-1} at the 0–10 cm and 10–20 cm, respectively, and TN averaged 2.67 and 1.38 g N kg^{-1} at the 0–10 cm and 10–20 cm, respectively. Soil organic C was not significantly affected by grazing management at either depth in 2018 ($p = 0.3791$ for 0–10 cm and $p = 0.4520$ for 10–20 cm). Similarly, grazing management did not affect TN in 2018 at the surface depth ($p = 0.1164$), and at the subsurface depth ($p = 0.6804$) (Fig. 3). Soil organic C and TN contents over time (from 2010 to 2018) did not significantly change among treatments or between depths (Fig. 3 and Table 2).

Grazing treatments did not affect soil bulk density in this study (averaged 0.89 and 1.41 Mg m^{-3} in 2010 and averaged 1.19 and 1.61 g Mg m^{-3} in 2018 at the 0–10 cm and 10–20 cm depths, respectively). Means for C stock and N stock for grazing treatment in 2010 and 2018 at the 0–10 cm and 10–20 cm depths are shown in Fig. 4. Regardless of the grazing treatment, C stock averaged 20.43 and $8.87 \text{ Mg C ha}^{-1}$ in 2010 and averaged 21.44 and $9.38 \text{ Mg C ha}^{-1}$ in 2018 at the 0–10 cm and 10–20 cm, respectively. Nitrogen stock, regardless of grazing treatment, averaged 1.97 and $1.11 \text{ Mg N ha}^{-1}$ in 2010 and averaged 2.95 and $1.49 \text{ Mg N ha}^{-1}$ in 2018 at the 0–10 cm and 10–20 cm, respectively. Carbon and N stocks were not significantly affected by grazing treatment at either depth in both years (p values > 0.05 , Fig. 4). In addition, C stock and N stock for both depths over time (from 2010 to 2018) did not change for each treatment (Fig. 4 and Table 2).

Carbon and N sequestration rate means under grazing management over time at the 0–10 cm and 10–20 cm depths are shown in Fig. 5. Regardless of the grazing management, C sequestration rate averaged 0.57 and $-0.003 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ at the 0–10 cm and 10–20 cm,

Table 2

Changes in soil C and N from 2010 to 2018 at the 0–10 cm depth under the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) at Barta Brothers Ranch, Nebraska Sandhills meadows. The symbols indicate changes from 2010 to 2018 in the C and N concentration of each column, with boxes meaning no change, arrows pointing up meaning increase, and arrows pointing down meaning decrease.

	SOC	TN	C stock	N stock	fPOM-C	fPOM-N	oPOM-C	oPOM-N	MAOM-C	MAOM-N	DOM-C	DOM-N
	<i>4PR1</i>											
2018	↓	↓	↓	↓	↑	↓	↓	↓	↓	↓	↓	↓
2010	↓	↓	↓	↓	↑	↓	↓	↓	↓	↓	↓	↓
	<i>4PR2</i>											
2018	↓	↓	↓	↓	↑	↓	↓	↓	↓	↓	↓	↓
2010	↓	↓	↓	↓	↑	↓	↓	↓	↓	↓	↓	↓
	<i>MOB</i>											
2018	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓
2010	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓
	<i>CNT</i>											
2018	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓
2010	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓	↓

SOC, soil organic C in bulk soils; TN, total N in bulk soils; C_S, C stock; N_S, N stock; fPOM-C, C content in the free particulate organic matter; fPOM-N, N content in the free particulate organic matter; oPOM-C, C content in the macro-aggregate occluded particulate organic matter; oPOM-N, N content in the macro-aggregate occluded particulate organic matter; MAOM-C, C content in the mineral-associated organic matter; MAOM-N, N content in the mineral-associated organic matter; DOM-C, C content in the dissolved organic matter; and DOM-N, N content in the dissolved organic matter.

respectively, and N sequestration rate averaged 0.039 and $-0.015 \text{ Mg N ha}^{-1} \text{ yr}^{-1}$ at the 0–10 cm and 10–20 cm, respectively. At the 0–10 cm depth, grazing management influenced C sequestration rate but did not affect N sequestration (Fig. 5). At the 0–10 cm depth, C sequestration rate for 4PR1 was significantly higher than for CNT but 4PR2 and MOB were intermediate and did not differ from CNT (Fig. 5). At the 10–20 cm depth, grazing management did not influence C and N sequestration rates (Fig. 5).

3.2. Carbon and nitrogen concentrations in soil organic matter fractions as affected by grazing over eight years

Percent of C and N in SOM fractions to SOC and TN in bulk soils, respectively, at the 0–10 cm and 10–20 cm depths in 2010 and 2018 are shown in Fig. 6. Across grazing treatments, the concentrations of C and N in SOM fractions at the 0–10 cm depth in 2010 was the highest in the total particulate organic matter fraction (free and occluded OM), constituting approximately 58 and 51 % of the SOC and TN in bulk soils, respectively (Fig. 6). Similarly, in 2018, the concentrations of C and N in the SOM fractions (across grazing treatments) were the highest in the total particulate organic matter fraction, constituting approximately 65 and 54 % of the SOC and TN, respectively, in the bulk soils at the 0–10 cm (Fig. 6). However, the concentrations of SOC and TN in the SOM fractions at the 10–20 cm depth (regardless of grazing treatment) was the highest in the MAOM fraction, constituting approximately 55 and 92 % of the SOC and TN in the bulk soils, respectively (Fig. 6).

Carbon and TN means in SOM fractions in all grazing treatments in 2010 and 2018 at the 0–10 cm depth are shown in Figs. 7 and 8. At the 0–10 cm depth in 2010, $f\text{POM-C}$ was 5.18 g C kg^{-1} and $f\text{POM-N}$ was 0.48 g N kg^{-1} (Fig. 7). At the same year and depth, $o\text{POM-C}$ was 7.17 g C kg^{-1} and $o\text{POM-N}$ was 0.46 g N kg^{-1} (Fig. 7). MAOM-C was 8.63 g C kg^{-1} , MAOM-N was 0.90 g N kg^{-1} , DOM-C was 0.05 g C kg^{-1} , and DOM-N was $0.005 \text{ g N kg}^{-1}$ (Fig. 8). At the 0–10 cm depth in 2018, $f\text{POM-C}$ was 7.29 g C kg^{-1} , $f\text{POM-N}$ was 0.50 g N kg^{-1} , $o\text{POM-C}$ was 5.60 g C kg^{-1} , $o\text{POM-N}$ was 0.44 g N kg^{-1} , MAOM-C was 7.52 g C kg^{-1} , MAOM-N was 0.80 g N kg^{-1} , DOM-C was $0.023 \text{ g C kg}^{-1}$, and DOM-N was $0.001 \text{ g N kg}^{-1}$ (Figs. 7 and 8).

As shown in Figs. 7 and 8, C and TN means in SOM fractions did not

differ at the 0–10 cm depth in 2010 (p values > 0.05). In 2018, grazing treatment did not influence $f\text{POM}$ ($p = 0.3999$ for $f\text{POM-C}$ and $p = 0.4629$ for $f\text{POM-N}$) at the 0–10 cm depth. For $o\text{POM}$ in 2018; however, $o\text{POM-C}$ ($p = <0.0001$) and $o\text{POM-N}$ ($p = <0.0001$) at the 0–10 cm depth among grazing treatments differed significantly (Fig. 7). At the 0–10 cm depth, $o\text{POM-C}$ did not differ in 2018 between the MOB and 4PR1 treatments and among the 4PR1, 4PR2, and CNT treatments; however, $o\text{POM-C}$ in MOB was approximately 66 % and 70 % higher than 4PR2 and CNT (Fig. 7). A similar trend was also observed in $o\text{POM-N}$ at the 0–10 cm depth in 2018, with MOB having 73 % and 80 % higher $o\text{POM-N}$ compared to 4PR2 and CNT, respectively (Fig. 7).

Grazing treatment also significantly influenced MAOM-C ($p = <0.0001$) and MAOM-N ($p = <0.0001$) concentrations at the 0–10 cm depth in 2018 (Fig. 8). Concentrations of MAOM-C and MAOM-N did not differ among 4PR1, 4PR2, and MOB treatments and for MOB and CNT treatments in 2018; however, 4PR1 and 4PR2 had approximately 62 % and 63 %, respectively, higher MAOM-C than CNT and approximately 69 % and 72 %, respectively, higher MAOM-N than CNT at the 0–10 cm depth (Fig. 8). Grazing treatment also significantly influenced DOM-C ($p = <0.0321$) and DOM-N ($p = <0.0012$) concentrations at the 0–10 cm depth in 2018 (Fig. 8). DOM-C and DOM-N levels did not differ between 4PR1 and MOB treatments in 2018; however, 4PR1 and MOB both had approximately 37 % and 50 %, respectively, higher DOM-C than 4PR2 and CNT at the 0–10 cm depth (Fig. 8). Similarly, both 4PR1 and MOB had approximately 57 % and 113 %, respectively, higher MAOM-N than 4PR2 and CNT at the 0–10 cm depth (Fig. 8).

Carbon and TN contents in SOM fractions over the 8-yr of the study (from 2010 to 2018) at the 0–10 cm depth significantly changed (Figs. 7 and 8 and Table 2). The $f\text{POM-C}$ for MOB and CNT did not change over time; however, $f\text{POM-C}$ was 42 and 59 % greater in 2018 than in 2010 for 4PR1 and 4PR2, respectively, (Fig. 7 and Table 2). The $o\text{POM-C}$ for 4PR1 and MOB did not change over time; however, $o\text{POM-C}$ decreased significantly by 39 and 40 % from 2010 to 2018 for 4PR2 and CNT, respectively (Fig. 7 and Table 2). Significant change in MAOM-C over the 8-yr of the study was observed for CNT only, with MAOM-C being 41 % lower in 2018 than in 2010 (Fig. 8 and Table 2). Significant change in DOM-C and DOM-N over the 8-yr of the study was observed for 4PR2 and CNT, with DOM-C and DOM-N being lower in 2018 than in 2010

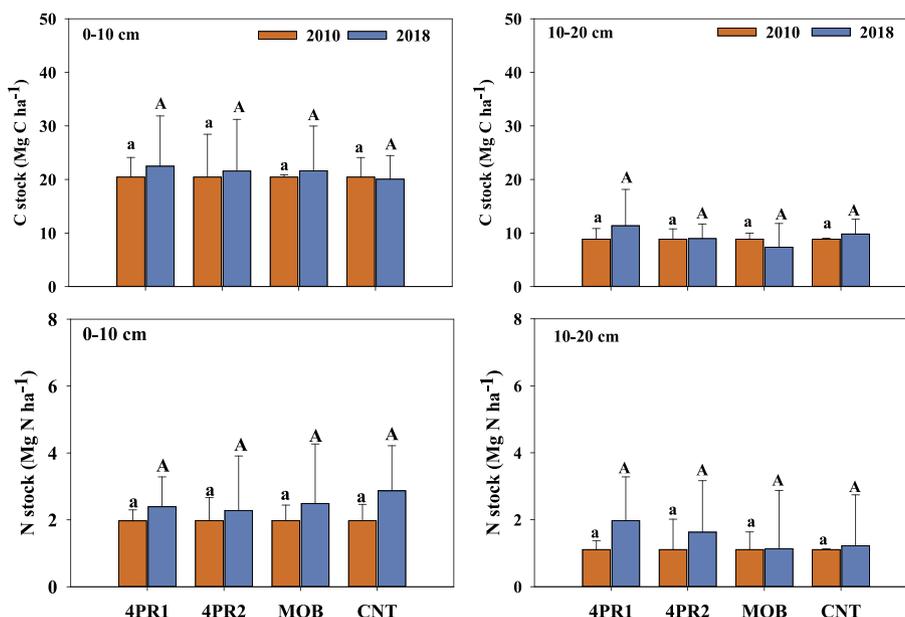


Fig. 4. Carbon stock and N stock for the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) in 2010 and 2018 at the 0–10 cm and 10–20 cm depths at Barta Brothers Ranch, Nebraska Sandhills meadows. Vertical bars indicate standard errors of the means. Different lower case letters indicate a significant difference among treatments in 2010, and different capital letters indicate a significant difference among treatments in 2018 at the 0.05 probability level.

(Fig. 8 and Table 2).

Carbon and TN means in SOM fractions in 2018 at the 10–20 cm depth are shown in Fig. S2. Across grazing treatments in 2018 at the 10–20 cm depth, C and TN content in SOM fractions averaged 1.63 g C kg^{-1} for fPOM-C , 0.01 g N kg^{-1} for fPOM-N , 1.05 g C kg^{-1} for oPOM-C , 0.01 g N kg^{-1} for oPOM-N , 2.82 g C kg^{-1} for MAOM-C , 0.27 g N kg^{-1} for MAOM-N , $0.013 \text{ g C kg}^{-1}$ for DOM-C , and $0.0005 \text{ g N kg}^{-1}$ for DOM-N (Fig. S2). For the 10–20 cm depth in 2018, no significant differences were observed among grazing treatments in C and TN content in SOM fractions (p values > 0.05 , Fig. S2).

3.3. Relationship between soil properties

Fig. 9A shows the principal component analysis of soil properties. Principal component analysis explained 78 % of the changes in soil properties, with PC1 and PC2 accounting for 47.2 % and 30.4 % of the total, respectively. Along the PC1 axis, 4PR1, 4PR2, and MOB are on the right side, and CNT is on the left, potentially representing the degree of soil improvement or degradation due to grazing treatment (Fig. 9A). While the largest contributors to differences among the grazing treatments were MAOM-C, fPOM-C , oPOM-N , and oPOM-C , the lowest contributors were the Bd, C stock and N stock (Fig. 9A). Most of the soil properties were clustered around 4PR1 and 4PR2, meaning these two grazing treatments potentially showed higher values of these properties compared to MOB and CNT. Fig. 9B shows the correlation analysis of soil properties. Correlation analysis showed that oPOM-C was positively ($p = 0.0003$) correlated with oPOM-NDOM-C was positively ($p = 0.0012$) correlated with DOM-N , and MAOM-C was positively ($p = 0.0012$) correlated with MAOM-N (Fig. 9B). The fPOM-N was positively correlated with MAOM-C ($p = 0.0478$), MAOM-N ($p = 0.0348$) and TN ($p = 0.0042$) (Fig. 9B).

4. Discussion

4.1. Carbon and nitrogen in bulk soils after eight consecutive years of grazing

Carbon and N concentrations (i.e. g kg^{-1}) and stocks (i.e. Mg ha^{-1}) in bulk soils in this study decreased with increasing soil depth, a result that is expected and attributed to decreased microbial activity and root growth with increasing soil depth (Eynard et al., 2005). Significant differences among grazing treatments were not observed in C and N concentrations and stocks in bulk soils after eight years of grazing, perhaps because of the non-significant effect of grazing treatments on grass production in this study (annual forage production averaged approximately 5107 kg ha^{-1} over the 8-yr of the study, Andrade et al., 2022). A meta-analysis of 257 published articles showed that soil C is positively correlated to above and belowground plant production (Lu et al., 2011). Briske et al. (2008) reported that plant production was higher under rotational grazing (moderate and high stocking densities) in only three of 23 research studies when compared to continuous grazing. de la Motte et al. (2018) also reported that a high stocking density rotational grazing did not affect aboveground biomass production compared to annual haying in Western Europe.

Our results for C stock are supported by Contosta et al. (2021) who reported that a high stocking density grazing did not affect C stock compared to no grazing for fine sandy loam and sandy loam soils in the US Northeastern states. On the other hand, a high stocking density grazing in several of the US Southern states increased C and N stocks possibly due to increased root and litter decomposition rate compared to annual haying (Conant et al., 2003) or because of the even distribution of organic N inputs from animals to the soil compared to continuous grazing (Mosier et al., 2021). Our expectation was that the longer rest period in MOB grazing would maximize re-growth, thus improving plant

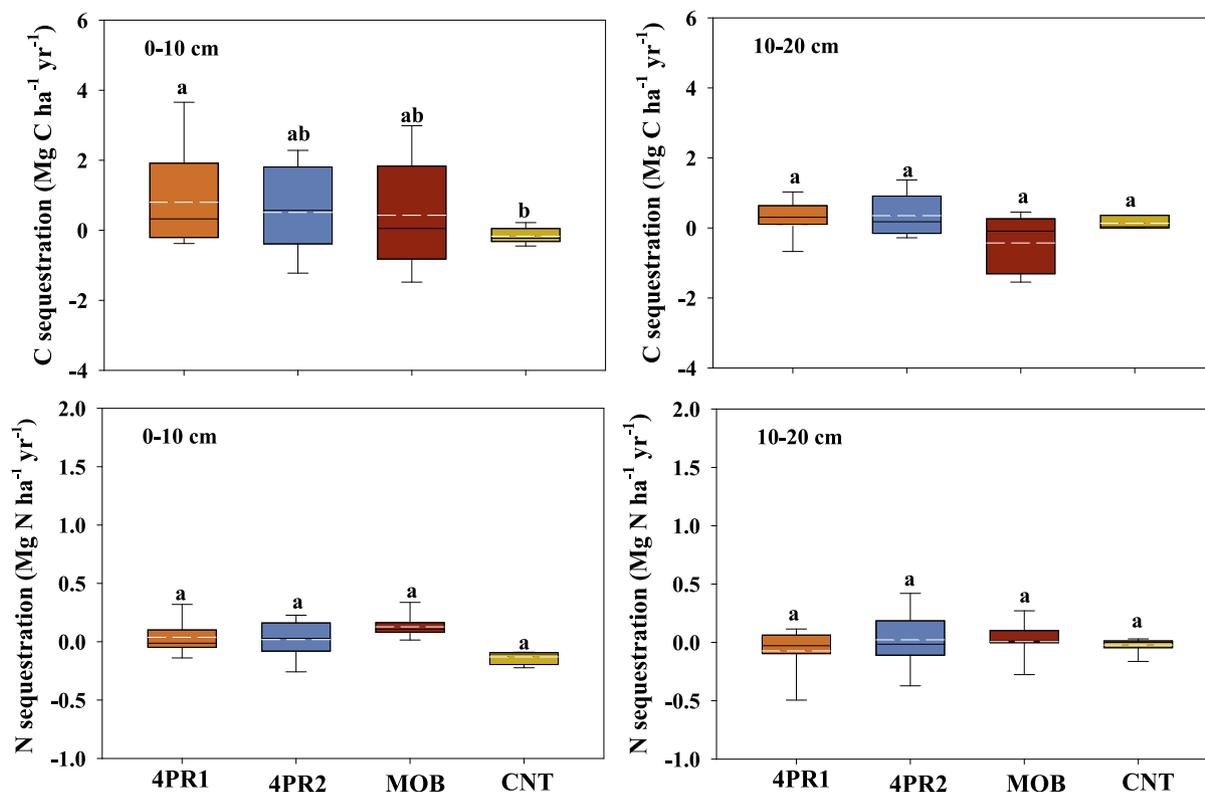


Fig. 5. Carbon and total nitrogen sequestration rates for the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) at the 0–10 cm and 10–20 cm depths at Barta Brothers Ranch, Nebraska Sandhills meadows. Vertical bars indicate standard errors of the means. Different letters indicate a significant difference among treatments at the 0.05 probability level.

productivity (Jordan et al., 2022) and enhancing C and N in bulk soils. This was not confirmed. A longer study covering more than eight years of grazing may lead to changes in C and N contents in bulk soils. In addition, soil type and climate conditions in the current study may be one of the factors affecting the response of C and N levels in the bulk soils to grazing. According to Bonin and Lal (2014), high clay content facilitates the storing of high amounts of C in soil. Our study, however, involved soils with low clay content, a condition that may slow the affects of grazing management on soil C and N concentrations. From 2010 to 2018, both SOC, TN, and C and N stock contents did not significantly change for each treatment in this study. According to Sanderman et al. (2015), more than 15 years of rotational grazing in South Australia is required to obtain the benefits of increasing SOC from the adoption of

rotational grazing.

The C sequestration rate in our experiment (0.11 to 0.80 Mg C ha⁻¹ yr⁻¹) was within the range of a previous review conducted in temperate grasslands for 17 countries including the United States (Conant et al., 2001). High stocking density rotational grazing in the US Midwestern states sequestered large amounts of soil C as reported by Stanley et al. (2018), however; in this study, MOB grazing did not affect C and N sequestration rates in the experimental period when compared to low stocking density treatments and no grazing. Carbon sequestration rate can be enhanced by minimizing C-loss pathways (Sarkar et al., 2020). The high amount of trampling in the MOB treatment compared with the low stocking density grazing treatments in this study can promote soil-litter mixing (Wei et al., 2021), thus increasing losses of litter C

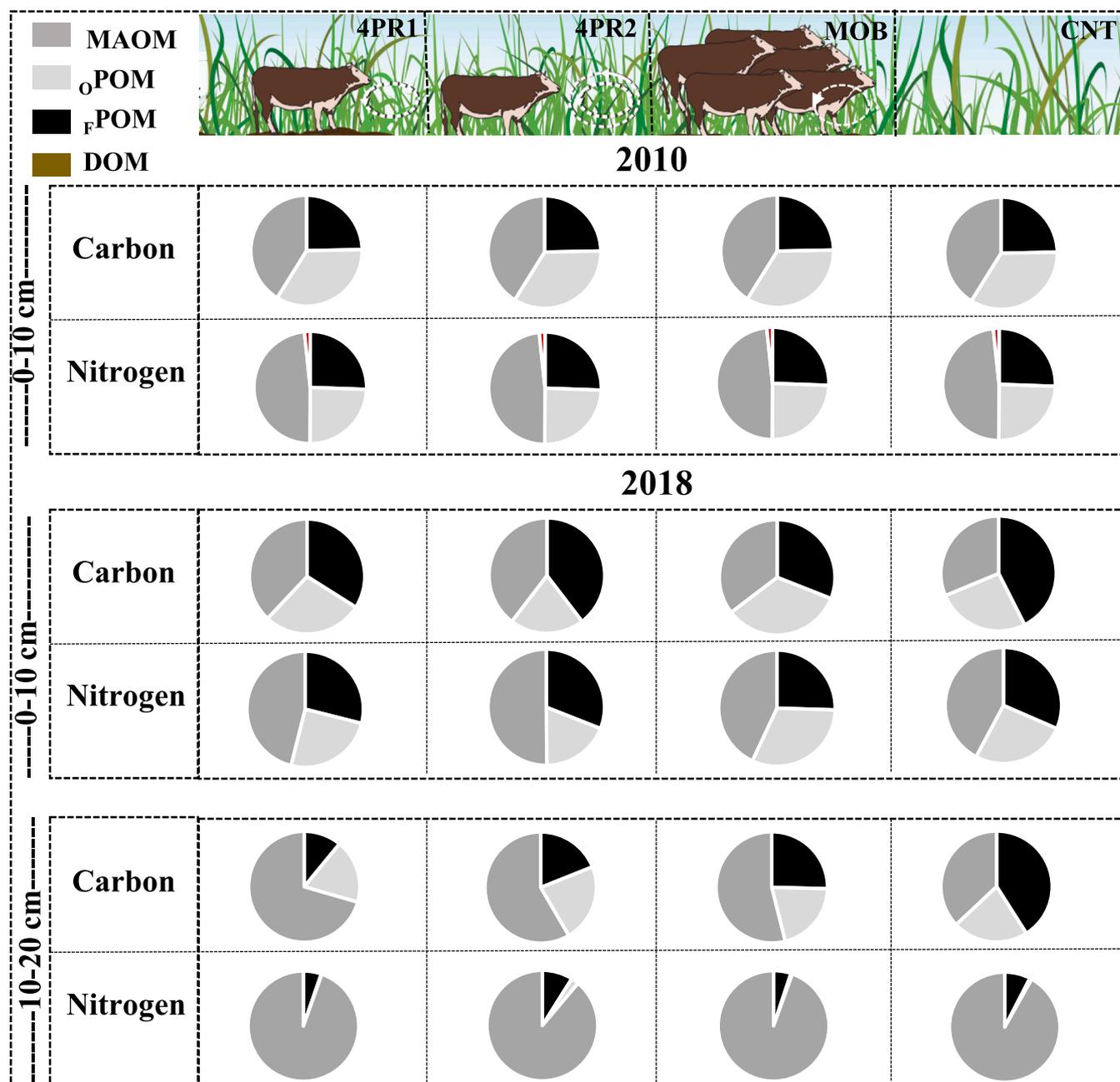


Fig. 6. Proportion of soil organic C (SOC) and total N (TN) found in SOM fractions from the SOC and TN, respectively, at the 0–10 cm and 10–20 cm depths in 2010 and 2018 at Barta Brothers Ranch, Nebraska Sandhills meadows. Treatments included the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT). fPOM, free particulate organic matter; oPOM, macro-aggregate occluded particulate organic matter; DOM, dissolved organic matter; and MAOM, mineral-associated organic matter.

(Hosseiniaghdam et al., 2023). On the other hand, the relatively low amount of trampling in the 4PR1 treatment possibly reduced microbial decomposition (Hewins et al., 2013), and thus resulted in a higher C sequestration rate than the CNT treatment in this study. The 4PR2 treatment had a lower amount of trampling compared to the 4PR1 treatment in this study (Andrade et al., 2022), possibly explaining why this grazing treatment did not significantly increase C sequestration rate compared to the CNT. In contrast to the 4PR1 treatment, grazing in the 4PR2 treatment in this study was initiated when trampling conditions were suboptimal. In addition, our field observations from the experimental site in this study showed that grazing animals in the 4PR2 second cycle tended to prefer new vegetative growth in areas that were grazed in the first cycle, thus avoiding areas that were previously ungrazed and resulting in decreased trampling.

4.2. Carbon and nitrogen in soil organic matter fractions after eight consecutive years of grazing

The finding that the total particulate organic fraction (free and macro-aggregate occluded) was higher than the mineral-associated organic fraction at the 0–10 cm depth in the Nebraska Sandhills meadows was expected because the soil in this study has a low percentage of clay and silt minerals, which is needed to store MAOM fraction (Cotrufo and Lalvallee, 2022). However, the SOM fractionation results at the 10–20 cm depth showed that the soils store more C and N in the mineral-associated organic fraction than in the total particulate organic fraction. Similarly, Cotrufo et al. (2019) found that C storage was more dominant in MAOM than in POM in soils with low C (<12 g kg⁻¹) content in European grasslands, which can be attributed to the fact that MAOM formation can be enhanced by stimulating microbial activity (Wang et al., 2019).

The grazing management strategies investigated in this study did not

affect C and N levels in free particulate organic matter. This fraction can be affected by litter inputs from both aboveground and belowground in the early stages of decomposition (Christensen, 2001). MOB grazing allows a longer rest period due to short-duration grazing compared to the other grazing treatments, thus it was expected to result in higher litter inputs. However, stocking density in this study did not affect annual litter accumulation, which averaged approximately 2 194 kg ha⁻¹ (Guretzky et al., 2020). A previous study at the same site reported that annual forage production was mainly influenced by climate variability rather than grazing intensities (Andrade et al., 2022). This finding was supported by other research in the semi-arid ecosystem, who reported that the effect of climate on vegetation was more pronounced compared to the effect of grazing management (Herrero-Jáuregui and Oesterheld, 2018).

A significant positive correlation between C and N in macro-aggregate particulate organic matter was found in this study, possibly explaining the similar effects of grazing management on these two fractions. Unlike C and N in free particulate organic matter, MOB increased C and N contents in macro-aggregate particulate organic matter and dissolved organic matter compared to low stocking density with two cycles (4PR2) and no grazing. One of the mechanisms by which grazing management can influence soil C and N levels is animal excretion deposition (Fig. 1). Our data from a previous study at the same site showed that MOB grazing paddocks had a higher dung beetle density (Wagner et al., 2021), possibly increasing dung incorporation into the soil and enhancing both oPOM and DOM . In this study, low stocking density with one cycle (4PR1) also increased DOM formation compared to the 4PR2 and CNT treatments. Unlike 4PR2, MOB and 4PR1 treatments began at the same time each year (mid-June) and were grazed for the same period (60 days), perhaps explaining why their effects on the dissolved organic C and N were similar. More research is needed to evaluate how the grazing duration and time of year affects the response of

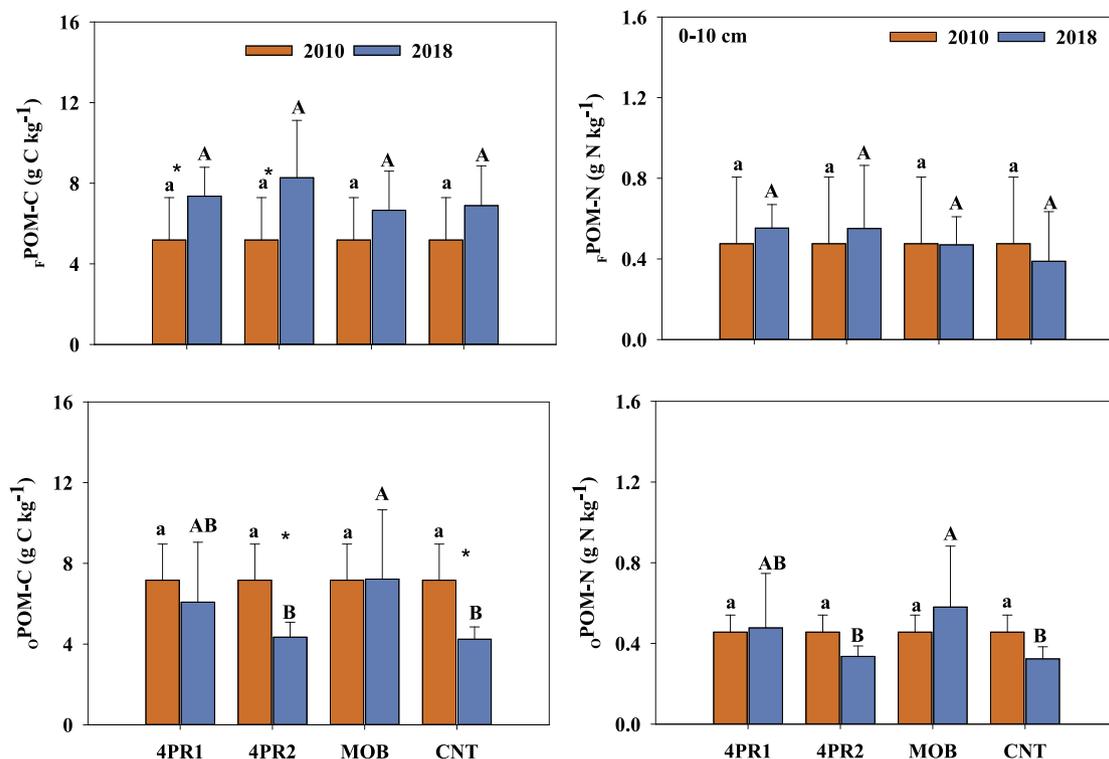


Fig. 7. Carbon and TN contents in SOM fractions in 2010 and 2018 at the 0–10 cm depth. Carbon and TN contents in the free particulate organic matter ($f\text{POM-C}$ and $f\text{POM-N}$) and C and TN contents in the macro-aggregate occluded particulate organic matter (oPOM-C and oPOM-N), under the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) at Barta Brothers Ranch, Nebraska Sandhills meadows. Different lower case letters indicate a significant difference between years in 2010, and different capital letters indicate a significant difference among treatments in 2018 at the 0.05 probability level.

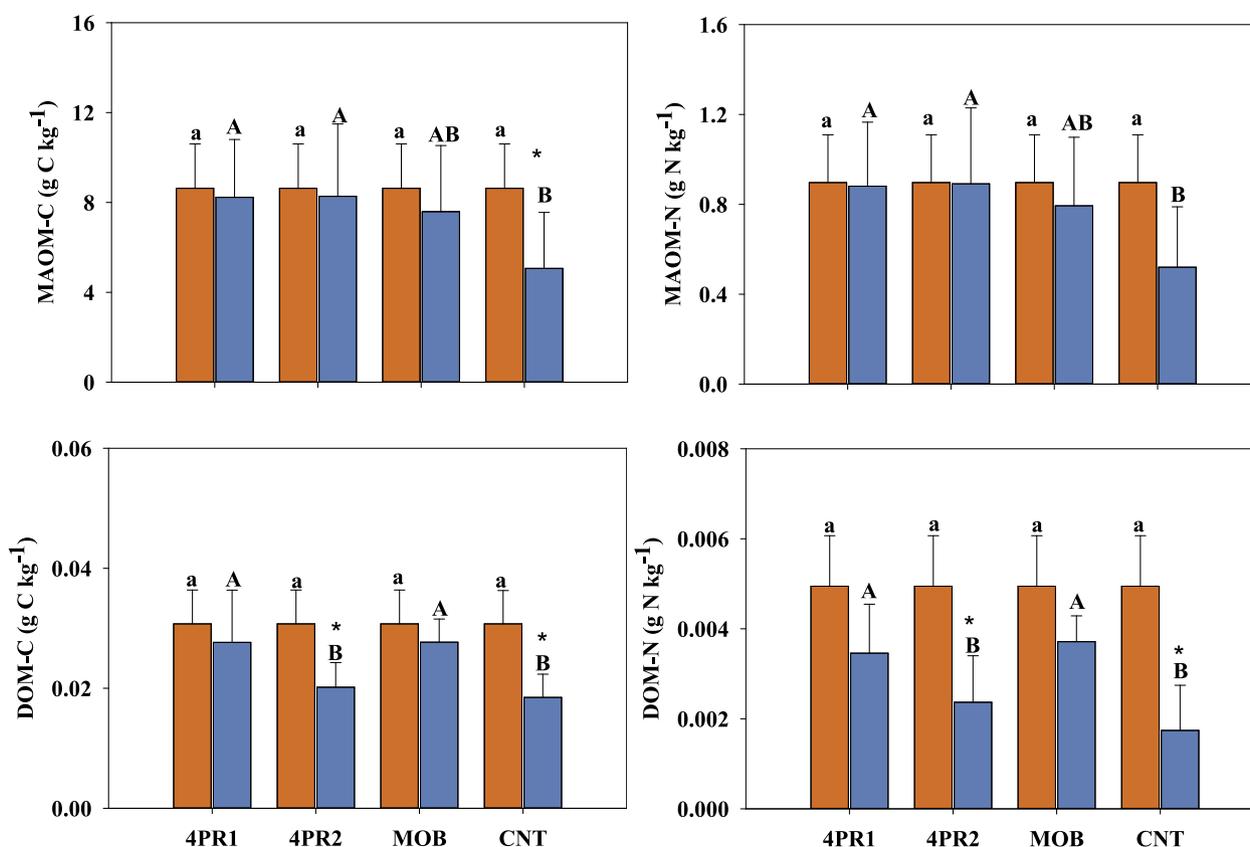


Fig. 8. Carbon and TN contents in SOM fractions in 2010 and 2018 at the 0–10 cm depth. Carbon and N contents in the mineral-associated organic matter (MAOM-C and MAOM-N) and C and TN contents in the dissolved organic matter (DOM-C and DOM-N) under the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) at Barta Brothers Ranch, Nebraska Sandhills meadows. Different lower case letters indicate a significant difference among treatments in 2010, and different capital letters indicate a significant difference among treatments in 2018 at the 0.05 probability level.

DOM formation to different stocking densities.

The increase in oPOM due to MOB grazing and the increase in DOM due to both MOB and 4PR1 grazing practices suggest that these two managements were beneficial in enhancing soil fertility. Particulate and dissolved organic fractions are weakly protected (Abagandura and Kumar, 2021), meaning these fractions are available to microorganisms, so they cycle fast and result in enhancing overall agricultural productivity (Cotrufo and Lavalley, 2022).

Unlike C and N contents in the oPOM and DOM fractions, MOB grazing did not affect C and N contents in the mineral-associated organic fraction compared to the CNT treatment. Grazing management can influence soil C and N levels through trampling (Fig. 1). Increased trampling may disturb soil aggregates (Roberts and Johnson, 2021), thus enhancing the decomposition of aggregate-protected C and N. Mineral-associated organic fraction is always found occluded within the small aggregates (Cotrufo and Lavalley, 2022). Teutschová et al. (2021) reported that intensive short-duration rotational grazing can affect aggregate stability via also its impacts on microbial community structure, affecting soil aggregation and C stabilization within aggregates. Although aggregate stability was not measured in this study, previous research has reported inconsistent effects of rotational grazing on aggregate stability (Dong et al., 2022; Valani et al., 2022; Teague et al., 2011).

However, the mineral-associated organic C and N contents were higher under the 4PR1 and 4PR2 treatments compared to the CNT treatment in our study. Changes in microbial activity can explain how grazing management can affect mineral-associated organic C and N formation due to their effect on its decomposition and stabilization (Li et al., 2021). Grazing management can affect microbial and enzyme

activity as reported by Bai et al. (2013) and Teague and Dowhower (2022). For example, β -glucosidase (an important microbial enzyme involved in the C-cycle) activity was higher under the rotational grazing compared to the continuous grazing in sandy soils (Kotzé et al., 2013). As biological measurements such as microbial community structure and microbial biomass C and N were not made in this study, such measurements would be fundamental in future research to explain the impact of grazing management on C and N dynamics.

The results that rotational grazing at low stocking densities enhanced MAOM fraction suggest that grazing at low stocking densities facilitates a long residence time of C and N, which is very important in preserving SOC in soils with few fine particles. Unlike POM and DOM fractions, MAOM fraction is protected from decomposition through mineral associations and occlusion within small aggregates (Williams et al., 2018), therefore, this fraction can provide valuable information on the effect of soil management on SOC sequestration.

Changes in C and N contents in SOM fraction (POM, DOM, and MAOM) from 2010 to 2018 in this study could be explained by the changes in plant species composition, frequency of occurrence of plants and/or aboveground plant production over time as reported by a previous study (Andrade et al., 2022) conducted at the same experimental site. For example, different plant species composition has different litter quantities and qualities and microbial compositions, thus affecting SOM fractions (Wang et al., 2020). The changes over time in C and N contents in SOM fractions over time may be also due to the changes in microbial decomposition (Campbell et al., 2022).

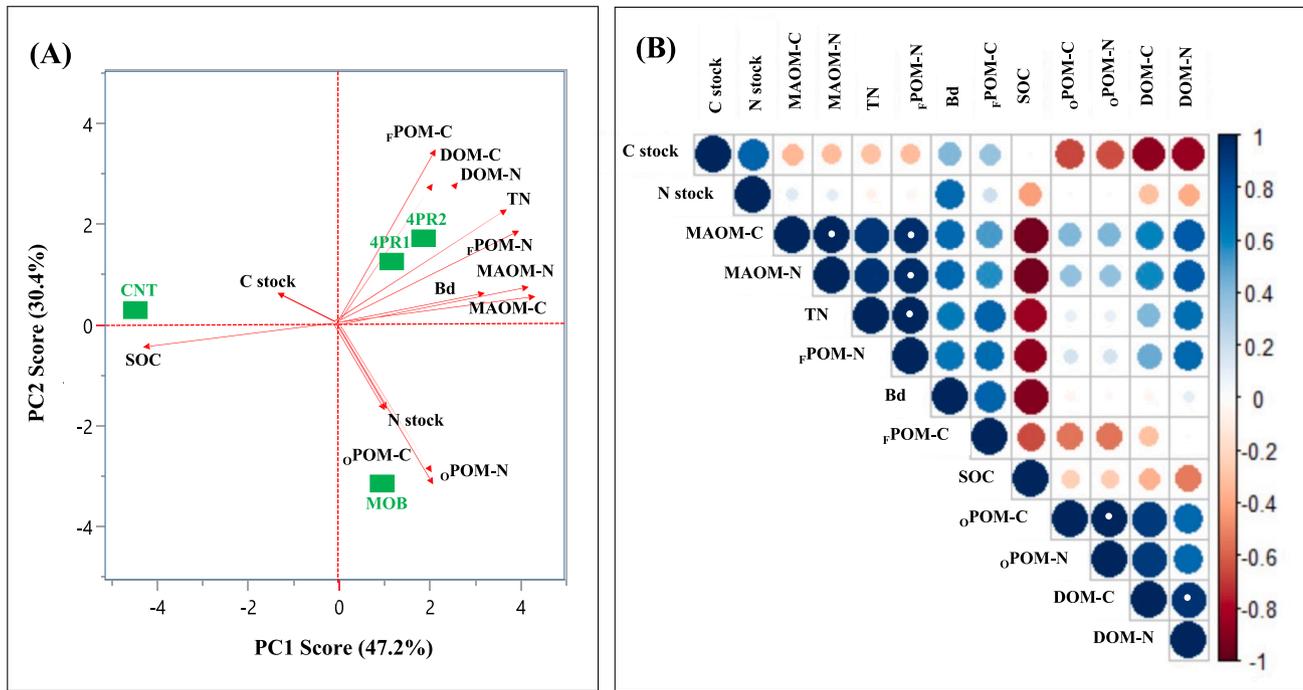


Fig. 9. Principal component analysis of soil properties (A) as impacted by the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) in 2018. The relative contribution of each indicator is reflected in the length and the direction of the arrows. Correlation analysis of soil properties (B) in 2018. The color and size of the circles denote the magnitude and direction of the relationship. White dots within the circles indicate significant correlations. f POM-C, C content in the free particulate organic matter; f POM-N, N content in the free particulate organic matter; o POM-C, C content in the macro-aggregate occluded particulate organic matter; o POM-N, N content in the macro-aggregate occluded particulate organic matter; MAOM-C, C content in the mineral-associated organic matter; MAOM-N, N content in the mineral-associated organic matter; Bd, bulk density; SOC, soil organic C in bulk soils; and TN, total N in bulk soils.

5. Management application

Interest in enhancing and protecting C in the soil has increased worldwide due to the negative impact of GHG emissions. Rotational grazing has been promoted as a sustainable management practice on grasslands, with much research being conducted in the Nebraska

Sandhills region (Andrade et al., 2022; Guretzky et al., 2020; Stephenson et al., 2013; Schacht et al., 2010) and worldwide (de Otálora et al., 2021; Augustine et al., 2020; Billman et al., 2020) evaluating how rotational grazing affects plant production and livestock performance. This research is the first long-term study conducted in the Nebraska Sandhills meadows to evaluate how rotational grazing at conventional

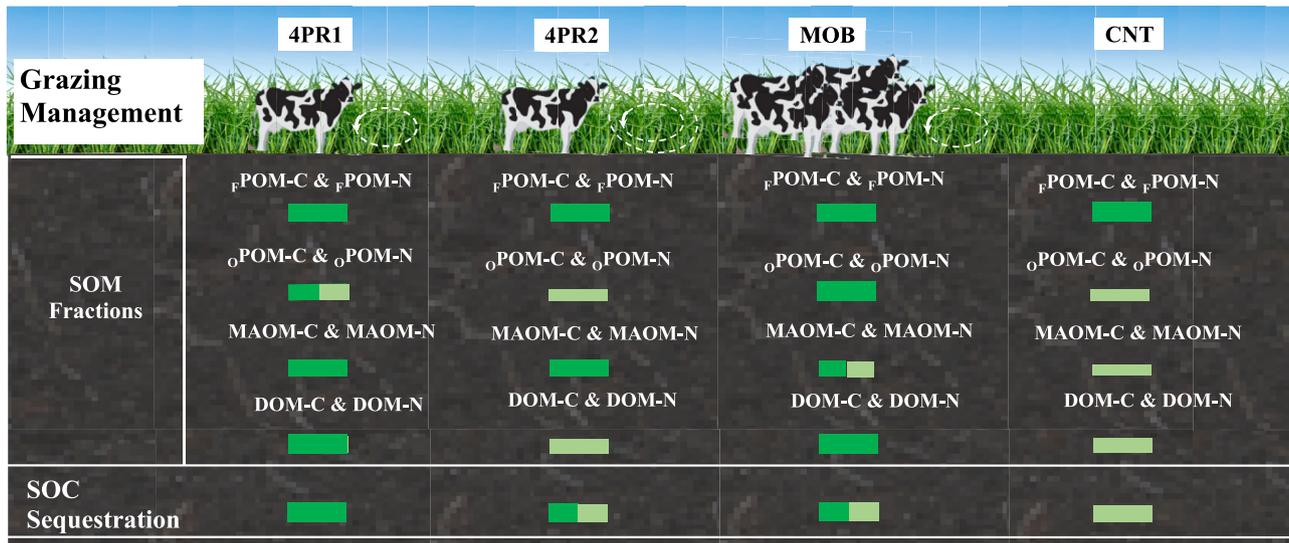


Fig. 10. An overview of the impact of grazing management on soil organic C (SOC) sequestration. Carbon and TN contents in the free particulate organic matter (f POM-C and f POM-N), in the macro-aggregate occluded particulate organic matter (o POM-C and o POM-N), in the mineral-associated organic matter (MAOM-C and MAOM-N), and in the dissolved organic matter (DOM-C and DOM-N) under the 4-pasture rotation system with one grazing cycle (4PR1), the 4-pasture rotation system with two grazing cycles (4PR2), the ultrahigh stocking density system with one grazing cycle (MOB), and no grazing (CNT) at Barta Brothers Ranch, Nebraska Sandhills meadows. Different color indicate a significant difference among treatments. The darker the color, the higher C and N levels in Barta compared to other treatments. A grazing treatment with two different colors indicates that the grazing treatment had similar recorded C and N levels to any treatment with either of the colors.

and extremely high stocking densities can affect C and N levels in bulk soils and SOM fractions. The main findings of this study were summarized in Fig. 10. Regardless of the grazing management, C sequestration rate averaged 0.57 and $-0.003 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ at the 0–10 cm and 10–20 cm, respectively, and N sequestration rate averaged 0.039 and $-0.015 \text{ Mg N ha}^{-1} \text{ yr}^{-1}$ at the 0–10 cm and 10–20 cm, respectively. At the 0–10 cm depth, grazing management influenced C sequestration rate but did not affect N sequestration. At the 0–10 cm depth, C sequestration rate for low stocking density with cycle was significantly higher than for no grazing, but low stocking density with two cycles and high stocking density were intermediate and did not differ from no grazing or the low stocking density with one grazing cycle (Fig. 10). High stocking density grazing increased weakly protected C and N levels compared to CNT (Fig. 10). However, low stocking density grazing with one cycle increased highly protected C and N levels in mineral association compared to no grazing (Fig. 10). This increase in highly protected C and N contents resulted in enhancing SOC sequestration rate under low stocking density grazing with one cycle compared to no grazing (Fig. 10). Results from this study suggest that rotational grazing at a low stocking density for one cycle grazing was beneficial in promoting C; thus, it is a management option that can maintain and enhance long-term SOC accumulation in the Nebraska Sandhills meadows.

CRedit authorship contribution statement

Gandura O. Abagandura: Data curation, Formal analysis, Methodology, Validation, Visualization, Writing – original draft, Conceptualization. **Martha Mamo:** Data curation, Funding acquisition, Investigation, Supervision, Validation, Writing – review & editing, Conceptualization. **Walter H. Schacht:** Conceptualization, Funding acquisition, Writing – review & editing. **Aaron Shropshire:** Data curation, Methodology. **Jerry D. Volesky:** Conceptualization, Funding acquisition, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.geoderma.2024.116776>.

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