Soil organic matter and microbial community responses to semiarid croplands and grasslands management

Rajan Ghimire[a,b,*], Vesh R. Thapa[b], Amanda Cano[c,d], Veronica Acosta-Martinez[c]

[a] New Mexico State University, Agricultural Science Center, Clovis, NM, USA
[b] New Mexico State University, Department of Plant and Environmental Sciences, Las Cruces, NM, USA
[c] USDA Agricultural Research Service, Lubbock, TX, USA
[d] Texas Tech University, Lubbock, TX, USA

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ABSTRACT

Livestock integration in cropping systems and conversion of croplands into grazing lands has been increasingly considered to improve agricultural sustainability, yet their roles in soil health and resilience are not clear due to the complex interactions of soil, climate, and agricultural systems. A study was conducted to evaluate the effects of cropland and grassland management systems on soil organic carbon (SOC) and total nitrogen (N) across the soil profile (0–20, 20–40, 40–60, and 60–80 cm) and microbial community size, structure, and activity in the soil surface (0–20 cm) as indicators of soil health. Cropland systems compared included conventional-tilled winter-grazed cropland (CTGC) and no-tilled and strip-tilled croplands (NTC and STC) without livestock grazing. Grassland systems included grazed grassland (GGL) and ungrazed grassland (UGL). Grassland soils accumulated 18% greater SOC and 13% greater total N than cropland soils in the 0–80 cm profile. Microbial community size (sum of ester-linked fatty acid methyl esters [El-FAME]) in the surface 0–20 cm was 90% greater, and enzyme activities were 131–155% greater in the grasslands than in the croplands. Within grasslands, cattle (*Bos taurus*) grazing increased microbial community size by approximately 42%, which was mainly due to greater fatty acid methyl esters (FAME) markers for gram-positive bacteria (51%) and Actinobacteria (73%). Grazed cropland had 95% more β-glucosaminidase activity than ungrazed croplands. This study suggests light grazing and grassland restoration has potential to improve soil health and resilience through an increase in SOC and microbial community responses related to nutrient cycling.

1. Introduction

Grasslands constitute the largest land cover in the subhumid to semiarid regions of the Great Plains of the US. Grassland soils supported the proliferation of a diverse microbial community and accumulated SOC, a proxy of the soil organic matter (SOM), from thousands of years of plant production and belowground residue decomposition (DeLuca and Zabinski, 2011). Cultivation in grasslands in the past century has significantly altered microbial community size and structure, and thereby deteriorated soil health (Mackelprang et al., 2018). Similarly, the SOC stocks in the croplands of the Great Plains has depleted by 28–59% threatening the agricultural sustainability in the region (Paustian et al., 1997). Ogallala Aquifer, one of the largest aquifers in the world that supplies irrigation water to eight Great Plains states, is the main source of irrigated crop production in the Great Plains. Declining irrigation water level in the Ogallala Aquifer is expected to

change vast area of irrigated-lands into drylands further depleting soil health and decreasing agricultural productivity (Cano et al., 2018). The increased challenge in crop production associated with soil health deterioration highlights the need for quantifying the potential of grassland and cropland soils to sequester C, store nutrients, and support growth of diverse microbial community to develop a sustainable agricultural system.

Integrated crop-livestock system and conversion of croplands into grazing lands have been increasingly considered in the past few decades to improve soil tilth, fertility, and increase C sequestration (Russelle et al., 2007). Integrated crop-livestock system promotes the proliferation of microbial communities that drive SOC and nutrient cycling (Acosta-Martinez et al., 2010), water infiltration, soil aggregation, and structural stability (Franzluebbers, 2007; Liebig et al., 2011). Grazing may also contribute to higher crop productivity when it is a part of a land use rotation by enhancing mineralization of nutrients such as N, P,

and S from organic resources (Giller et al., 1997). However, soil microbial communities in croplands and grasslands vary with cropping practices, grazing intensity, duration, and frequency, and they likely influence SOC and nutrient cycling (Bronson et al., 2004; Ghimire et al., 2014; Schuman et al., 1999).

Adopting conservation systems that reduce soil disturbance and increase residue cover on the soil surface could be another strategy to improve soil health and resilience through increased SOC storage and nutrient cycling (King and Hofmockel, 2017). Adopting conservation systems has reduced the fallow frequency and significantly minimized soil erosion in the crop-fallow system typically practiced in the Great Plains region, and stored more water in the soil profile than conventional systems (Baumhardt and Jones, 2002; Baumhardt et al., 2015). With these changes, conservation systems also shifted microbial communities toward higher soil fungal proportions and increased soil enzyme activities, which could help in the development of healthy and resilient agroecosystems (Acosta-Martinez et al., 2007).

Increasing SOC storage and associated improvements in soil health of agricultural fields is critical not only for maintaining agronomic production but also for harnessing environmental benefits through soil C sequestration and greenhouse gas mitigation. Improvements in soil health is more critical in low-fertility soils and hot, dry, semiarid environment of the Southern Great Plains than more productive humid environments because SOC, nutrients, and enzyme activities in low-productivity agroecosystems have not been very responsive to changes in soil management (Acosta-Martinez et al., 2011; Hansen et al., 2012). Soils in the Southern Great Plains typically have low fertility with < 1.5% in SOC content (Bronson et al., 2004; Ghimire et al., 2019). In addition, plant biomass production and soil C inputs are limited by low precipitation and high temperature in a semiarid southern Great Plains (Zhou et al., 2009), which ultimately slows down the process of SOC accumulation and negatively affects soil health.

The main objective of this study was to compare soil health and resilience of grasslands and croplands in semiarid Southern Great Plains by evaluating the profile SOC and N distribution and the surface soil microbial community size, structure, and activity. Specifically, we measured SOC, total N, potentially mineralizable carbon (PMC), potentially mineralizable nitrogen (PMN), microbial community size (sum of bacteria, fungi, and protozoa), structure (e.g., fungal to bacterial ratio via FAME profiling), and enzyme activities associated with SOM dynamics and nutrient cycling in grasslands and croplands.

2. Materials and methods

2.1. Study site and treatments

Soil samples for this study were collected from research plots at the New Mexico State University Agricultural Science Center (ASC), Clovis, NM, USA (34°35′ N, 103°12′ W, elevation 1348 m) and adjacent farmers' fields. The study area has a semiarid climate with average maximum and minimum temperatures of 22.1 °C and 4.28 °C, respectively. Average annual rainfall is 466 mm, 70% of which occurs between May and September. This area experiences high inter-annual variability in precipitation and frequent short-term drought periods within a crop growing season. All the study plots (ASC and on-farm) were mapped as Olton clay loam (fine, mixed, superactive, thermic aridic paleustolls) under the USDA soil classification system (Soil Survey Staff, 2017). Soil texture analysis of the surface 0–20 cm depth had sand, silt, and clay contents of 47–48%, 17–20%, and 33–35%, respectively.

Five agricultural management systems were evaluated including three croplands and two grasslands varying in management practices. The cropland systems included conventional-tilled winter grazed cropland (CTGC), and no-tilled and strip-tilled croplands (NTC and STC) without livestock grazing while grassland systems included grazed grassland (GGL) and ungrazed grassland (UGL) (Table 1). The NTC and

STC were part of a randomized experiment and had 24-m by 30-m plots replicated three times, while CTGC, GGL, and UGL were spatially randomized three 10-m by 10-m plots within a large field (> 10 ha) adjacent to the experimental station plots (Table 1). The CTGC followed conventional management of wheat *(Triticum aestivum* L.)-sorghum [*Sorghum bicolor* (L.) Moench]-fallow with low-intensity grazing of sorghum stubble for three months during the winter. The typical conventional system in the area uses moldboard plow, disk, and DMI ripper (Case IH, LLC, Racine, WI) to 0–20 cm depth for tillage and incorporates crop residues into soils, leaving < 15% of the soil surface covered by residues. The NTC and STC were also under conventional wheat-sorghum-fallow rotation until 2013 but had no cattle grazing. No-tillage and strip tillage corn (*Zea mays* L.)-sorghum rotations were started in 2013. The NTC involved no tillage whereas STC used one pass of a strip tiller (Twin Diamond Industries, LLC, Minden, NE) followed by planting with a no-till drill (John Deere, Moline, IL) in either system. After the adoption of conservation tillage systems, NTC and STC maintained 42 and 74% crop residue on the soil surface, respectively. The GGL and UGL were under native grasses, predominantly bluegrama [*Bouteloua gracilis* (Willd. ex Kunth) Lag. ex Griffiths] (~80%), crested wheatgrass (*Agropyron cristatum* L.), buffalo grass [*Buchloe dactyloides* (Nutt.) J.T. Columbus], and plains lovegrass (*Eragrostis intermedia* Hitchc.). The GGL was never broken for cultivation but was grazed at low intensity (0.3–0.4 beef cattle ha^{-1}) throughout the year whereas UGL was converted to native grasses in 1964, and the grasses were never harvested or grazed.

All the cropland plots had sorghum in 2017 when soil samples were collected for this study. Sorghum was planted in May at ~69,000 seeds per hectare and 76 cm row spacing. All plots received 25 kg N ha^{-1} at planting, and no P and K fertilizers were applied. Weeds, pests, and diseases in all plots were controlled as needed by using chemical measures as recommended by New Mexico State University pest management guidelines. Grassland plots were never sprayed or fertilized.

2.2. Soil sampling and laboratory analysis

Soil samples were collected in summer 2017. Baseline SOC data for each site were unavailable. Therefore, undisturbed grassland was considered as a reference site to compare changes in SOC, N, and soil microbial community size and functions under various agricultural management systems considering that SOC level remains at a steady state under undisturbed grasslands (Ogle et al., 2005). Within each plot, two 4.5 cm diameter cores were collected from 0 to 80 cm depths using a tractor-mounted Giddings hydraulic probe (Giddings Machine Company Inc., Windsor, CO) with plastic liner tubes. In the fields under sorghum cropping, one core was taken between crop rows and one core in a crop row to capture the variability within a cultivated field. Soil samples collected from each site were transported to the laboratory and divided into 0–20 cm, 20–40 cm, 40–60 cm, and 60–80 cm depth increments. Soil samples within each 20 cm depth increment were weighed, and 20 g sub-samples were taken for soil moisture estimation. Studies have demonstrated greater differences in SOC and N stock and microbial community size across management and land use within the soil surface than at lower soil depths (e.g., Eilers et al., 2012; Fierer et al., 2003). Thus, subsamples from 0 to 20 cm depth were stored in a −20 °C freezer for soil microbial community analysis and 50 g sub-samples were stored in a 4 °C refrigerator for PMC and PMN estimation. The rest of the samples were air dried and ground to pass through a 2-mm sieve for analysis of soil pH, electrical conductivity (EC), SOC, and total N.

Soil pH and EC were measured in the air-dried soils (1:5 soil-water ratio) by using a pH electrode and conductivity meter standardized against known buffer solutions. The SOC and total N contents were determined by a dry combustion method (LECO Corporation, St. Joseph, MI) in samples ground to pass through a < 0.5 mm sieve. Soil inorganic C was removed by treating samples with a 6 M HCl solution.

Table 1
Crops, tillage, and livestock management under diverse cropland and grassland systems.

Treatment[a]	Cropping system/plants	Crop and grazing management	Years
CTGC	Wheat-sorghum- fallow 3-year rotation	Mostly tilled with a moldboard plow, disk, and DMI ripper; conventional fertility management (25 kg N ha^{-1}, and no P and K); low-intensity winter grazing with beef cattle (0.3–0.4 steer ha^{-1}); and chemical weed and pest management as needed.	> 20
NTC	Corn-sorghum 2-year rotation	No tillage, seeds, and fertilizers drilled using a no-till drill (John Deere, Moline, IL), fertilizer applied at 25 kg N ha^{-1}, no P and K, and chemical weed and pest management as needed.	4
STC	Corn-sorghum 2-year rotation	Tilled with strip-tiller (Twin Diamond Industries, LLC, Minden, NE) that created about 15 cm wide 10 cm deep tilled zone, conventional fertility management (25 kg N ha^{-1}, and no P and K), no grazing, and chemical weed and pest management as needed.	4
GGL	Native grasses	Predominantly bluegrama (80%), low-intensity grazing (0.3–0.4 steer ha^{-1}) throughout the year, no tillage, fertilizer application, nor weed and pest management.	> 50
UGL	Native grasses	Predominantly bluegrama (80%) and no grazing, harvesting, tillage, fertilizer application, nor weed and pest management.	> 50

[a] CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, and UGL - ungrazed grassland.

Approximately 20 g subsample collected from each field was dried at 105 °C for 24 h to estimate soil water content. Soil bulk density was estimated as a ratio of dry soil mass to soil volume within each depth increment (Blake and Hartge, 1986). Soil bulk density estimated for each depth was used for converting SOC and N concentrations to SOC and N contents. The SOC and N stocks in the soil profile were calculated by summing SOC content in individual soil depths as described in Ghimire et al. (2015). Soil PMC content was estimated by aerobic incubation of 20 g soil samples in 1 L mason jars for two weeks (Zibilske, 1994). Soil PMN content was determined as the sum of nitrate (NO_3^-) and ammonium (NH_4^+) nitrogen in an automated flow injection N analyzer (Timberline Instruments, LLC) on incubated soil samples without subtracting an unincubated control.

Soil microbial community structure was characterized via ester-linked fatty acid methyl ester (EL-FAME) analysis in field-moist soil samples (3 g, < 2 mm) as described by Schutter and Dick (2000). Samples were reconstituted in 100 μL of hexane containing methyl nonadecanoate (19:0) at a concentration of 150 nmol g^{-1} soil as an internal standard before being vortexed and transferred to 250 μL glass inserts in 2 mL GC vials. The biomarker FAMEs were analyzed using an Agilent 6890 N gas chromatograph equipped with a 25 m × 0.20 mm × 0.33 μm (5% phenyl)-methylpolysiloxane Agilent HP-5 fused silica capillary column (Agilent, Santa Clara, CA) and flame ionization detector (Hewlett Packard, Palo Alto, CA) with ultra-high-purity nitrogen as the carrier gas. Peak identification and area calculation were performed using the Phospholipid Fatty Acid calibration method from MIDI (Microbial ID, Inc., Newark, DE). The FAMEs were identified by the number of C atoms, a colon, the number of double bonds, and the position of the first double bond from the methyl (ω) end of the molecule. Particular notations are defined as cis isomers (c), cyclopropane group (cy), methyl group (Me), and iso- (i) and anteiso- (a) branched FAMEs. Selected FAMEs were used as microbial markers according to previous research (Zelles, 1999). Bacterial markers included gram-positive bacteria (i15:0, a15:0, i17:0, a17:0), gram-negative bacteria (cy17:0, cy19:0), and Actinobacteria (10Me16:0, 10Me17:0, 10Me18:0); fungal markers included saprophytic fungi (18:1ω9c, 18:2ω6c) and arbuscular mycorrhizal fungi (AMF) (16:1ω5c); and protozoa markers included 20:4ω6c. Absolute amounts of FAMEs (nmol g^{-1} soil) were subsequently converted to mol% and used to evaluate the relative abundance and community structure. The bacterial sum was calculated using the gram-positive, gram-negative, and Actinobacteria markers, and the fungal sum was calculated using saprophytic and AMF fungal markers listed above. The fungal to bacterial ratio was calculated by dividing total fungi by total bacteria. Soil microbial community size was estimated as a sum of all the biomarker fatty acids associated with bacteria, fungi, and protozoa.

Soil enzyme activities were monitored as soil microbial function indicators for SOC and nutrient cycling. Alkaline phosphatase and β-glucosaminidase activities were determined by incubating 0.5 g (< 2 mm air-dried) soil samples at optimal buffer pH for 1 h at 37 °C as described in Tabatabai (1994) and Parham and Deng (2000),

respectively. The results are expressed in mg of p-nitrophenol (PNP) released per kg of soil (dry soil basis) per hour.

2.3. Statistical analyses

All data were tested for normality of residuals and homogeneity of variance in statistical analysis system (SAS v.9.4, SAS Institute, Cary, NC), and passed both tests. The variation in treatment responses was evaluated using a MIXED procedure of SAS, which considered treatment as a fixed effect and replication as a random effect. Data for each soil depth as well as 0–80 cm profile were analyzed separately. F-tests that utilized Type III sums of squares were used to test the significance of fixed effects (treatments). Treatment means were separated using the Fisher's protected LSD at α = 0.05 unless otherwise stated using a PDIFF option of the LSMEANS statement in the SAS. Since the study was conducted on experimental station plots and nearby on-farm locations, it did not have a traditional RCBD design. Therefore, dummy variables were created for spatially randomized treatments (on-farm plots) to conform with the randomized design of the experimental station plots. In addition, data on the microbial community structure, SOC and N components, and soil enzyme activity were reanalyzed using a multi-variate method (principal component analysis, PCA) in a Minitab (V.17.0, Minitab Inc., State College, PA) to compare the relative difference between croplands and grasslands. For this, FAME data for the individual microbial group were normalized to the sum of microbial FAMEs to calculate the relative abundance of each microbial group, and shifts in a microbial community structure and functions were compared. The first two principal components are graphed to summarize the results.

3. Results

3.1. Soil bulk density, pH, and electrical conductivity

The average bulk density was 1.14 g cm^{-3}, 1.24 g cm^{-3}, 1.25 g cm^{-3}, and 1.21 g cm^{-3} at 0–20 cm, 20–40 cm, 40–60 cm, and 60–80 cm depth, respectively and not different between treatments within a soil depth (Table 2). Soil pH values were also not different between treatments within a soil profile; however, they tended to increase with depth. Soil EC was significantly different between treatments. Specifically, it was higher in CTGC than all other treatments in 40–60 cm and 60–80 cm depths. Soil EC did not differ among treatments in 0–20 and 20–40 cm depths.

3.2. Soil organic carbon and nitrogen dynamics

The SOC content was significantly different between treatments in 0–20 cm depth (Fig. 1a). The SOC content in grazed grassland, GGL, was statistically similar to ungrazed grassland, UGL, and significantly greater than all cropland systems. The SOC at 0–20 cm depth for the means of grasslands was 37% greater than the means of croplands

Table 2
Depth distribution of soil pH, electrical conductivity (EC), and soil bulk density (BD) under diverse cropland and grassland systems.

Parameters	Soil depth (cm)	Treatments[†]				
		Croplands			Grasslands	
		CTGC	NTC	STC	GGL	UGL
BD (g cm^{-3})	0–20	1.14	1.24	1.19	1.07	1.09
	20–40	1.24	1.33	1.31	1.16	1.19
	40–60	1.19	1.26	1.33	1.22	1.29
	60–80	1.15	1.22	1.29	1.20	1.21
Soil pH (1:5)	0–20	6.7	7.0	6.7	6.6	6.4
	20–40	7.8	7.6	7.9	7.8	7.5
	40–60	7.8	8.3	8.1	8.0	7.9
	60–80	8.2	8.5	8.4	8.4	8.3
EC (dS m^{-1})	0–20	0.13	0.15	0.11	0.09	0.08
	20–40	0.17	0.14	0.13	0.14	0.09
	40–60	0.12a	0.08b	0.09b	0.09b	0.09b
	60–80	0.16a	0.08b	0.09b	0.08b	0.09b

[†] CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, UGL - ungrazed grassland, BD - bulk density, and EC – electrical conductivity. Mean values within a row followed by different lowercase letters indicate a significant difference between treatments within a depth ($P < 0.05$, LSD test), no letters indicate no significant difference.

(18.7 Mg ha^{-1}). However, SOC content in lower soil depths (20–40 cm, 40–60 cm, and 60–80 cm) was not significantly different between cropland and grassland treatments. The SOC stock in the 0–80 cm profile of UGL with 65.7 Mg ha^{-1} was not significantly greater than that of GGL with 65.6 Mg ha^{-1} but was greater than in the croplands whose SOC stocks were in the range of 50.0 Mg ha^{-1} to 60.3 Mg ha^{-1} (Fig. 2). The SOC stock in 0–80 cm for the means of grasslands was 18% greater than the means of croplands (55.7 Mg ha^{-1}).

Management system significantly influenced total N content at the top 0–20 cm and not in the lower depths (Fig. 1b), a response similar to SOC content. At 0–20 cm depth, the total N content was 18% greater in the mean of grasslands than the croplands (1.98 Mg ha^{-1}). Among croplands, CTGC, using conventional tillage and cattle grazing, had significantly more total N content than STC, using strip tillage management and no cattle grazing. The NTC, using no tillage, remained between the other two cropland systems and was not significantly different from either. In 0–80 cm soil profile, UGL had 7.26 Mg N ha^{-1}, which was not significantly greater than total N in GGL (7.09 Mg ha^{-1}) but was significantly greater than the croplands (Fig. 2). Total N stock in 0–80 cm profile in the croplands ranged between 6.07 and 6.53 Mg ha^{-1} and was not significantly different, but it was 13% greater than that in the croplands (6.33 Mg ha^{-1}).

Soil PMC content, a labile SOC fraction that accounted for 0.9% to 2% of SOC within 0–20 cm depth, was also different between cropland and grassland systems (Fig. 3a). It was 540 kg ha^{-1} in GGL, which was significantly greater than that in UGL (344 kg ha^{-1}) as well as in croplands (152–264 kg ha^{-1}). While the mean of grasslands had 107% more PMC than that of croplands, GGL accumulated 57% more PMC than UGL. Soil PMC content was not significantly different among croplands. Soil PMN content, a labile N fraction that composed 0.7% to 2.5% of total N, was significantly greater in the croplands than in the grasslands (Fig. 3a). It was 19.4 kg ha^{-1} and 25.4 kg ha^{-1} in GGL and UGL, respectively, while 41.3–52.3 kg ha^{-1} in croplands. The PMN content was not significantly different between grasslands nor among croplands. Average PMN content was 53% less in grasslands than in croplands.

3.3. Microbial community size, structure, and activity

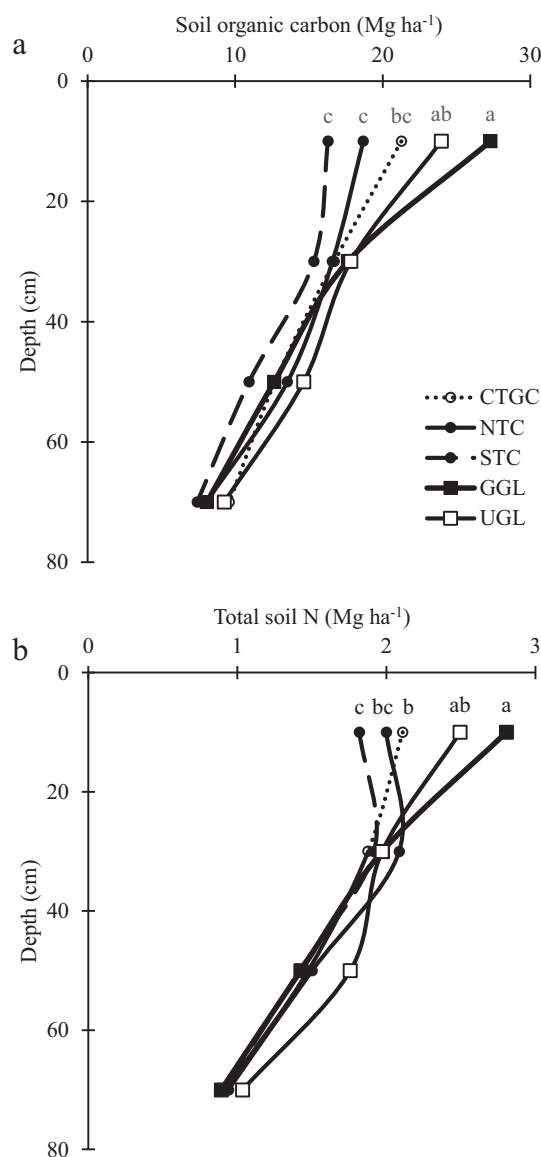Total FAMEs concentration as indicators of soil microbial

Fig. 1. Profile distribution of soil organic carbon (a) and total nitrogen (b) under croplands and grasslands. CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, and UGL - ungrazed grassland. Different letters indicate a significant difference between management systems in a given soil depth ($P \leq 0.05$).

community size (sum of bacteria, fungi, and protozoa FAMEs) was greater in the grasslands (106 and 74.7 nmol g^{-1} soil in GGL and UGL, respectively) than in the croplands (Table 3). The microbial community size in the croplands was not significantly different from each other with values ranging from 43.2 to 53.8 nmol g^{-1} soil. Among markers for different microbial groups, abundance of gram-positive bacteria was significantly greater in GGL (133%) and UGL (54%) than in croplands (average of 14.7 nmol g^{-1} soil). The gram-negative bacteria abundance was not significantly different among all systems compared. Abundance of the Actinobacteria markers was greater in GGL (166%) and UGL (53%) than the average abundance in the croplands (11.1 nmol g^{-1} soil). The abundance of the AMF marker in GGL was greater than in the croplands, but their abundance in UGL was not significantly greater than in CTGC. The GGL, which included cattle grazing, had 51% more gram-positive bacteria and 73% more Actinobacteria, which resulting in 42% greater total microbial community size than UGL, involving no cattle grazing. The FAME marker for protozoa was 155% greater in the grasslands than the croplands but did not differ significantly due to
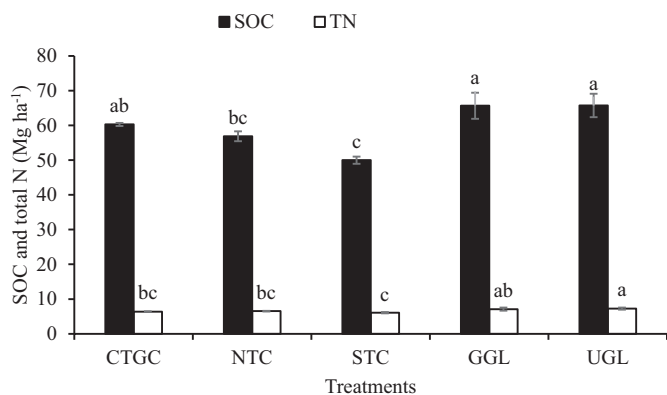
Fig. 2. Soil organic carbon (SOC) and total nitrogen (N) stock in the profile (0–80 cm) under croplands and grasslands. CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, and UGL - ungrazed grassland. Different letters accompanied by bars indicate a significant difference between management systems ($P \leq 0.05$).
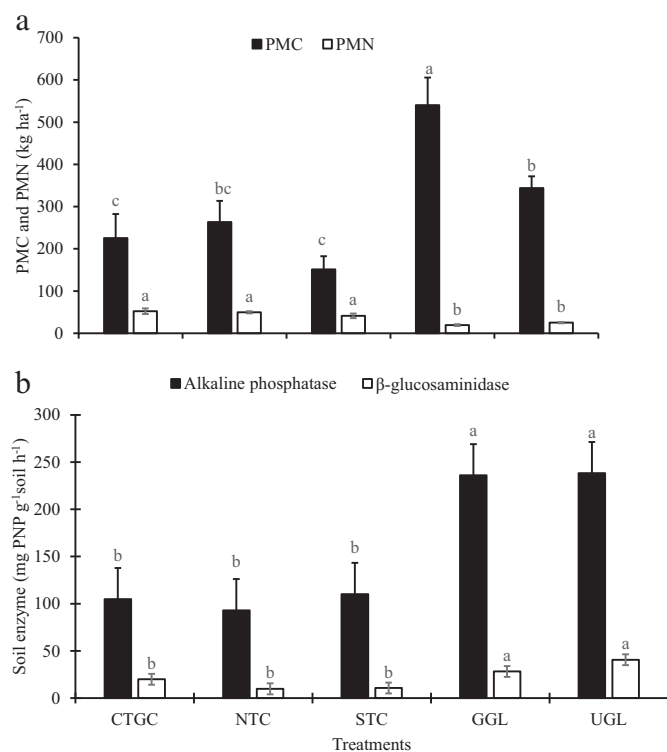

Fig. 3. Potentially mineralizable carbon (PMC) and nitrogen (PMN) (a) and enzymes Alkaline phosphatase and β-glucosaminidase activity (b) in 0–20 cm depth under croplands and grasslands. CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, and UGL - ungrazed grassland. Different letters accompanied by bars indicate a significant difference between management systems ($P \leq 0.05$).

grazing within the grasslands or the croplands. Fungal to bacterial ratio was not significantly different between agricultural systems. Overall, croplands did not differ in the abundance of gram-positive bacteria, Actinobacteria, AMF, and protozoa markers.

Soil enzyme activities also differed between grassland and cropland systems (Fig. 3b). The mean of alkaline phosphatase activity was 131% higher in the grasslands than in the croplands. However, it was not significantly different between management systems within the croplands, nor between the two grassland systems. The UGL had higher

Table 3
Soil microbial community (mean ± standard error) under diverse cropland and grassland systems.

Treatment	Soil microbial community (nmole g^{-1} soil)[†]									
	Gram-positive bacteria	Gram-negative bacteria	Actinobacteria	AMF	Saprophytic fungi	Protozoa	Bacteria sum	Fungi sum	F:B ratio	Microbial community size
Croplands										
CTGC	17.2 ± 2.22c	1.12 ± 0.23a	13.0 ± 0.70c	3.18 ± 0.45bc	19.0 ± 6.32b	0.44 ± 0.07b	31.2 ± 3.13c	22.2 ± 6.35b	0.69 ± 0.13a	53.8 ± 9.47c
NTC	14.4 ± 0.66c	1.94 ± 0.83a	11.0 ± 0.42c	2.01 ± 0.07c	15.9 ± 0.86b	0.40 ± 0.05b	27.3 ± 0.67c	17.9 ± 0.90b	0.66 ± 0.03a	45.6 ± 1.31c
STC	12.7 ± 1.30c	2.54 ± 0.88a	9.44 ± 0.71c	2.39 ± 0.49c	15.7 ± 2.92b	0.45 ± 0.09b	24.6 ± 2.53c	18.1 ± 3.36b	0.74 ± 0.13a	43.2 ± 5.19c
Grasslands										
GGL	34.4 ± 2.74a	2.71 ± 0.32a	29.6 ± 4.45a	8.00 ± 4.44a	30.1 ± 4.44a	1.00 ± 0.12a	66.7 ± 6.08a	38.1 ± 4.24a	0.58 ± 0.07a	106 ± 8.16a
UGL	22.8 ± 4.24b	2.62 ± 0.53a	17.1 ± 3.95b	5.40 ± 1.53ab	25.6 ± 1.53ab	1.19 ± 0.12a	42.5 ± 7.72b	31.0 ± 2.87a	0.76 ± 0.08a	74.7 ± 10.5b

[†] CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, and UGL - ungrazed grassland. Bacterial markers included gram-positive bacteria (i15:0, a15:0, i17:0, a17:0), gram-negative bacteria (cy17:0, cy19:0), and Actinobacteria (10Me16:0, 10Me17:0, 10Me18:0). Fungal markers included arbuscular mycorrhizal fungi (AMF) (16:1ω5c) and saprophytic fungi (18:1ω9c, 18:2ω6c). Protozoa marker was 20:4ω6c. Mean values within a column followed by different letters indicate a significant difference between management systems ($P \leq 0.05$).
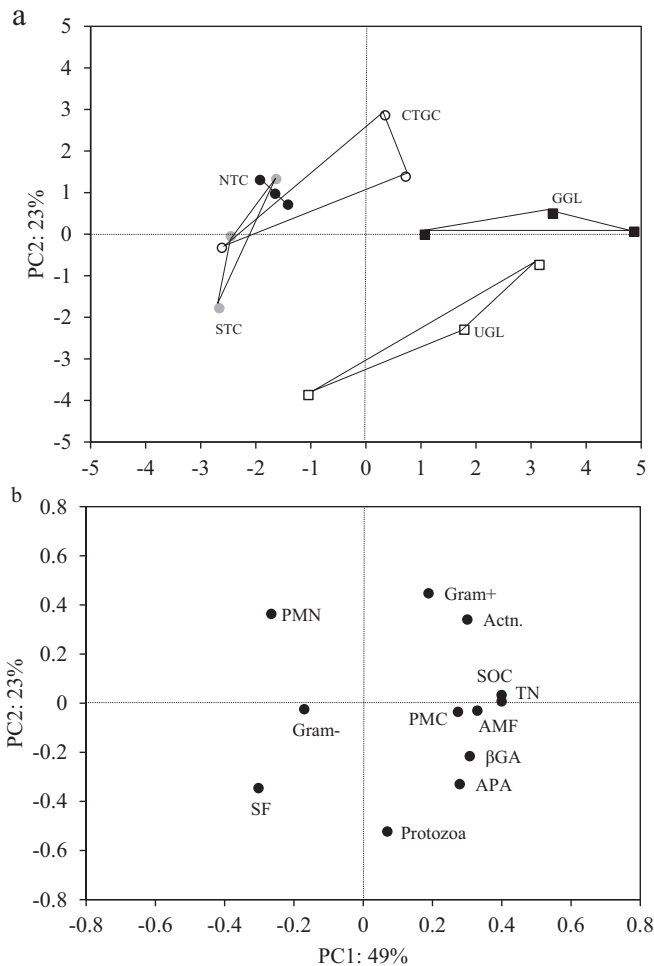
Fig. 4. Principle component analysis of a) microbial community response to cropland and grassland management systems, and b) loading score for different microbial groups. CTGC - conventional-tilled winter grazed cropland, NTC - no-tilled cropland, STC - strip-tilled cropland, GGL- grazed grassland, and UGL - ungrazed grassland. Gram+, gram-positive bacteria; Gram-, gram-negative bacteria; Actn., Actinobacteria; AMF, arbuscular mycorrhizal fungi; SF, saprophytic fungi; SOC, soil organic carbon; TN, total nitrogen; PMN, potentially mineralizable nitrogen; PMC, potentially mineralizable carbon; βGA, β-glucosaminidase; APA, alkaline phosphatase.

β-glucosaminidase activity (40.6 mg PNP kg^{-1} soil h^{-1}), which was statistically similar to their activity in GGL (28.2 mg PNP kg^{-1} soil h^{-1}) and significantly higher than the croplands (average: 13.5 mg PNP kg^{-1} soil h^{-1}). The CTGC, which included cattle grazing, had 94% greater β-glucosaminidase activity than STC, which lacked cattle grazing. The NTC had intermediate β-glucosaminidase activity and was not significantly different than CTGC and STC. β-glucosaminidase activity average for the grasslands was 155% greater than for the means of croplands.

3.4. Relationships among soil health indicators: microbial community, soil organic matter components, and enzyme activities

The PCA (principal component analysis) that included gram-positive bacteria, gram-negative bacteria, Actinobacteria, AMF, saprophytic fungi, and protozoa along with soil enzymes and SOC and N components revealed a clear separation of the grasslands from the croplands (Fig. 4a). There was also a clear distinction between the grasslands, but less clear distinction among the management practices within croplands. Cropland that included cattle grazing (CTGC) had the highest

variability in microbial community abundance and functions, while NTC with no soil disturbance had the least variability in microbial composition. Gram-positive bacteria, AMF, Actinobacteria, and protozoa had a positive loading along the PC1 among microbial groups, which was the same as loading for SOC, total N, and PMC contents and soil enzymes (Fig. 4b). Gram-negative bacteria and saprophytic fungi had negative loading along PC1, which was correlated with PMN content. Along the PC2, gram-positive bacteria, Actinobacteria, and PMN content had a positive loading, whereas saprotrophic fungi and soil enzymes had a negative loading and gram-negative bacteria, AMF, and SOC, total N, and PMC contents remained intermediate of the previous two groups.

4. Discussion

4.1. Differences in soil profile carbon and nitrogen

This study comparing different land uses for their potential to improve soil health in the semiarid Southern Great Plains revealed differences between grasslands and croplands in SOM components within the soil profile along with considerable differences in microbial community size and structure in the surface soil. While we found 18% more SOC stock in grasslands than in the croplands at 0–80 cm depth, there was no significant difference in SOC stock (0–80 cm depth) between lightly grazed grassland and ungrazed grassland after > 50 yr of native grass establishment. Perennial grasses typically have deeper and denser root systems than annual agronomic crops such as wheat and sorghum, leading to more SOC storage under grasslands than croplands (Bhandari et al., 2018; Ingram et al., 2008; Schuman et al., 2002). The greater SOC stock in grasslands in this study was consistent with a global meta-analysis result, which revealed an approximately 19% increase in SOC stock when croplands were converted to grassland (Guo and Gifford, 2002). However, the magnitude of difference detected in this study was smaller than the two-fold increase in SOC stock (0–120 cm) under native prairie than under wheat-fallow fields in Wyoming (Hurisso et al., 2013), and 79% more SOC stock (0–60 cm) under native grasses than under wheat-fallow fields in eastern Oregon (Ghimire et al., 2015), likely due to differences in soil types and environmental conditions. The study site has a hot and dry environment in which SOC loss and nutrient mineralization occur faster than in the colder environment.

Grazing can play a significant role in SOC and N accumulation in semiarid agroecosystems where biomass C input is limited by low precipitation. Studies show that light grazing increase SOC storage by stimulating plant biomass production (e.g., Frank et al., 2016). In line with that, we observed significantly greater PMC content in GGL than UGL. Although not statistically significant, the SOC stock in GGL was also greatest among all treatments, and it was greater in CTGC, a winter grazed cropland, than other cropland systems. Significantly greater PMC and marginally greater SOC and total N contents with grazing were possibly a response to the hot, dry environment of the study area. Soils are biologically active and SOC mineralization is rapid in warm soil conditions if soil moisture is not a limiting factor. Comparing grazed and ungrazed sites, Derner et al. (2006) reported 24% greater whole-ecosystem C storage (soil + plant) in grazed grasslands than ungrazed grasslands in a shortgrass community typically present in the semiarid Central and Southern Great Plains region. In croplands, however, such a difference between grazed and ungrazed systems was not observed. Cattles were grazed only for three months once in 3 yr (sorghum stubble) in CTGC, which may not be enough to see the positive effects of livestock in croplands.

Reduced or no tillage is often reported to increase SOC and N compared to conventional tillage when livestock are not a part of dryland agriculture (Ogle et al., 2005). We did not have conventionally tilled plots without cattle grazing to quantify benefits of adopting reduced/no-tillage system on SOC accumulation and nutrient cycling. The NTC and STC plots were also not able to maintain the crop residue

cover throughout the year since crop residues were mostly lost during high-intensity wind in the spring. The CTGC plots were conventionally tilled, but these plots were occasionally low-intensity grazed during winter. Studies show that approximately half of the crop residues in livestock-grazed cropping systems are utilized by livestock during light winter grazing, and the remainder of crop residues, as well as manure, are well mixed into the surface soil by livestock hoof impacts and during spring tillage operations (Franzluebbers, 2007). This ultimately leads to an increase in microbial community size and activity, nutrient cycling, soil aggregation, and soil micro- and macrofauna activity and thereby increases in SOC and N accumulation (Bronson et al., 2004; Ghimire et al., 2014). Although NTC and STC plots used a reduced/no-tillage approach and more intensive cropping since 2013, these plots were never grazed and were under an intensively tilled winter wheat-sorghum-fallow system until then. The winter wheat-sorghum-fallow system produces two crops in 3 yr and leaves 21-month fallow periods in between two crops, leading to continuous depletion in SOC and deterioration of soil health when livestock are not a part of the system. Monitoring of SOC and N in croplands that vary in tillage intensity but with the same cropping practice will reveal the improvements in soil health with conservation tillage. However, numerically higher bacterial and fungal community and enzyme activities in CTGC, which integrated cattle grazing during winter compared to NTC and STC, which adopted more intensive crop rotation and conservation tillage cannot be overlooked.

4.2. Responses of soil organic matter pools and microbial communities in the soil surface as indicators of soil health

A significant soil health superiority of the grasslands can be attributed to no disturbance for > 50 yr, which was manifested in 37% more SOC, 107% more PMC, and 18% more total N contents than croplands at 0–20 cm. The different SOM dynamics in a grassland compared to the nearby cropland supported shifts in microbial community composition (53–166% greater abundance of different microbial groups) and greater soil biogeochemical potential (131–151% greater enzyme activity). Specifically, community size of Gram positive bacteria, protozoa, and fungi (sum of AMF and saprophytic) were significantly greater in grasslands than in croplands irrespective of cattle grazing suggesting biochemical potential of grasslands to increase SOC storage. These findings may relate to two- to six-times greater biomass C inputs in surface soils of grasslands than in croplands previously found in the semiarid region (Ghimire et al., 2015), which may contribute to higher PMC content, microbial growth and activity, and ultimately increases in SOC storage. In line with our observation, a study in California finds reduction in labile C pools and changes in soil microbial community structure in surface soils with conversion of perennial grassland to annual cropland (DuPont et al., 2010). Beside differences in plant community, biomass C input, and levels of disturbances, croplands received fertilizer N each crop year, which contributed to high N availability and PMN content, and thereby supported the proliferation of selected microbial groups such as gram-negative bacteria.

Light grazing in grasslands could stimulate microbial proliferation, particularly bacterial growth through their effects on biomass growth and addition of labile organic substrates from dung and urine (Bardgett et al., 2001; DuPont et al., 2010). Grazing significantly increased Gram positive bacteria and the size of total bacterial community in grasslands. Evaluation of soil health indicators among croplands suggested that occasional light winter grazing (> 20 yrs) was slightly more effective than conservation tillage with intensive cropping (4 yr) for bacterial and fungal growth and their production of enzymes. Studies have shown high stratification by soil stratum in microbial community responses, and their responses to alternative agricultural management systems are mainly observed in the surface 5 or 10 cm depth, as opposed to lower depths (Eilers et al., 2012; Fierer et al., 2003). The soil samples in this study, however, were taken from the top 20 cm depth,

which may have attenuated the distinction between microbial community responses to the conservation tillage systems (NTC vs. STC) and livestock-integrated system, thus showing only a marginal difference ($p = 0.07$). Our findings comparing STC and NTC indicate that reduced disturbance by strip tillage is leaving enough residues on the soil surface relative to no tillage, resulting in no significant reductions in the labile C pool (PMC). Overall, these systems underwent the same crop rotation (sorghum and corn), which can leave a significant amount of residue serving as the substrate for soil microbial communities and sustaining decomposition processes, leading to soil C sequestration.

Studies often report a positive relationship between fungal proliferation, enzyme activities, and SOC accumulation (Bailey et al., 2002; Kallenbach et al., 2016). Our current study revealed that gram-positive bacteria and Actinobacteria were more favored by grazing than by reduced soil disturbance in grasslands and croplands as indicated by clustering of these microbial groups toward CTGC and GGL. A study in sub-montane ecosystems in UK revealed that microbial biomass, specifically, bacterial growth is maximal at low to intermediate levels of grazing (Bardgett et al., 2001). Unlike gram-positive bacteria and Actinobacteria, proportionate change in the response of gram-negative bacteria was not observed in either system. The gram-negative bacterial mass was negatively correlated to gram-positive bacterial mass in cropland systems, whereas this relationship was not observed in the grasslands, possibly owing to the difference in microbial substrates under grasslands and croplands. Gram-positive bacteria, Actinobacteria, and AMF were closely related to SOC, total N, PMC, and enzyme activities and mostly favored by grassland systems; whereas gram-negative bacteria were more related to PMN content and favored by croplands. Increases in gram-positive relative to gram-negative bacteria are often associated with a diversity of microbial substrates (Carpenter-Boggs et al., 2003; Fierer et al., 2003). Gram-positive bacteria and Actinobacteria are also favored by a high C:N ratio substrate environment, while gram-negative bacteria are favored by more easily decomposable substrates (Fierer et al., 2003). Lack of differences in soil bulk density, texture, and pH among alternative management systems further supported that differences in SOC and N were associated with microbial community responses to organic matter inputs, diversity in cropping, degree of disturbance, and livestock integration in croplands and grasslands.

5. Conclusion

This study comparing two grassland systems and three cropland systems revealed greater SOC and N components, microbial community size, fungal presence, and enzyme activities under grasslands than croplands. Differences in SOC and N accumulation due to crop and grassland management were observed in the surface 0–20 cm depth possibly, likely because of high microbial activity in the surface soil. Conversion of annual cropland to perennial grassland has the potential to improve soil health through its enhancements on soil microbial community responses and SOM components. While light grazing in grassland could increase microbial growth and labile SOM accumulation in grasslands, substantial improvements in cropland soil health due to limited grazing were not observed possibly because of the low frequency of grazing. More comprehensive study in croplands with diverse tillage, livestock-integration, and cropping strategies may reveal the relative potential of alternative systems to improve soil health. This study highlights the SOC dynamics and microbial community responses to grassland restoration and livestock integration, and conservation cropping strategies in semiarid regions.

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