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Article

Kernza in Wyoming: Perennial Grains for Vulnerable Lands

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Abstract

Kernza®, a perennial grain crop created from intermediate wheatgrass (*Thinopyrum intermedium*), has the potential to mitigate soil degradation in semiarid croplands of the Northern High Plains. From 2021 to 2023, Kernza was grown for the first time in Wyoming and compared at the field scale to winter wheat–fallow and Conservation Reserve Program (CRP) systems on a working farm. We measured grain and forage yields, root biomass, and soil health and microbiology in bulk and rhizosphere soils. The first growing season was dry, and Kernza produced substantial forage (2995 kg ha^{-1}) but insufficient grain for harvest. In the second year, Kernza produced 286 kg ha^{-1} of grain, compared to 2172 kg ha^{-1} for wheat. After two years, Kernza and wheat differed in rhizosphere—but not bulk—soil properties; Kernza rhizosphere organic matter, enzyme activities, and microbial communities were more similar to the rhizosphere of intermediate wheatgrass from CRP than to that of winter wheat. Kernza also produced nearly three times more root biomass and rhizosphere organic matter than winter wheat. Although Kernza remains a low-yielding crop in development, potential soil health benefits, a high market value, and the flexibility to harvest grain or forage may make it a viable option for this region.

Keywords: perennial grains; dryland agriculture; soil health; soil microbiology; rhizosphere

1. Introduction

Most crops grown worldwide are annuals, and annual systems are generally more vulnerable to climatic stress, use water and fertilizers less efficiently, and support less biodiversity than perennial systems [1–3]. Historically, farmers have planted perennial crops as biofuels, forages, and some fruits and vegetables, but until recently, no perennial grain crops existed. Moreover, around one-third of native perennial prairie worldwide has been converted to annual grain crops such as corn (*Zea mais*, L.) or wheat (*Triticum aestivum*, L.) [4,5], resulting in soil degradation in regions such as the US Great Plains [3,6]. For example, winter wheat–fallow cropping systems in the Wyoming High Plains have lost 33–63% of their native soil organic carbon (SOC), contributing to diminished crop yields [7,8]. This two-year rotation includes 14 months of fallow to store soil moisture for the wheat crop, which makes the soil vulnerable to erosion and disrupts microorganisms that rely on living plant roots [9]. However, converting these wheat–fallow systems to perennial grasses and forbs can restore up to 90% of lost SOC within 15 years of transition [8].

To address this land degradation, researchers are now breeding perennial grain crops that can mimic the ecosystem services of native prairie. One outcome is Kernza, a cultivar of intermediate wheatgrass (IWG; *Thinopyrum intermedium* (Host) Barkworth and D.R. Dewey)

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that can be harvested for both grain and forage for 3–5 years after planting [10]. Kernza's extensive root system can remediate land degradation by stabilizing soil, accessing deeper water and nutrients [3,11], and increasing SOC at depth [12,13]. Kernza also supports soil microbial communities more similar to native prairie than to annual wheat, with higher abundances of some symbiotic microorganisms [14,15]. These microorganisms mainly interact with plants in the rhizosphere, the soil zone surrounding roots, and are integral to plant health [16]. For example, arbuscular mycorrhizal fungi (AMF) improve plant access to water and nutrients as well as resistance to stressors such as drought [17,18].

Kernza is still an experimental crop and has only been grown commercially since 2015. Grain yields are typically less than a quarter of that of wheat, though Kernza grain prices are often 10–20 times higher [19]. Farmers often plant Kernza in marginal or degraded areas, valuing it for weed suppression, soil improvement, and the flexibility to harvest forage and/or grain [20]. This flexibility, along with a high water-use efficiency, makes Kernza a good candidate for semiarid landscapes [1,21,22]. However, few studies have grown Kernza in regions as dry as southeastern Wyoming, where mean precipitation (419 mm) is around one-half of that of Kernza breeding sites in Kansas. Drought and hailstorms are common in this region, creating a need for farmers to prioritize risk mitigation and to convert cropland to rangeland or Conservation Reserve Program (CRP) [6,23]. Therefore, Kernza may offer another alternative for farmers transitioning away from wheat–fallow. There is a knowledge gap regarding Kernza's performance in semiarid on-farm conditions, particularly with respect to soil health indicators and microbial dynamics.

This study compared Kernza, winter wheat–fallow, and CRP fields on a working farm in southeastern Wyoming by collecting root biomass, forage and grain yields, and bulk and rhizosphere soils. To evaluate soil health, we analyzed organic matter pools, structure, microbial biomass, enzyme activities, and bacterial and AMF communities. Overall, we aim to present a preliminary trial of Kernza in a previously untested environment and to compare soil health and microbial communities across these three systems. We hypothesized that Kernza would promote greater root biomass, soil organic matter pools, and microbial activity than winter wheat and would support a microbial community more similar to a perennial reference system (CRP) than to annual wheat.

2. Materials and Methods

2.1. Site Description and Experimental Design

The study was conducted from 2021 to 2023 on a working farm near Chugwater, Wyoming (41.787461 N, −104.756330 W, 1640 m elevation). Soils are mixed, mesic Ustic Torriorthents and Aridic Calciustolls [24] with a loam or silt loam texture and 15.1% clay, 1.0% SOC, 2.6% calcium carbonate, and a pH of 7.57 on average (Table 1). The climate is semiarid, with a mean annual precipitation of 419 mm, most occurring between April and July (Figure 1). Mean monthly temperatures range from −1.9 °C in January to 21.4 °C in July [25].

The site had been managed primarily with a wheat–fallow rotation for the past century, consisting of 10 months of winter wheat followed by 14 months of fallow, with non-inversion tillage for weed control. The study included three fields representing distinct treatments: a wheat–fallow field (6.8 ha, 850 m × 80 m), an adjacent Kernza field of equal size, and a CRP field (16.2 ha, 780 m × 208 m) located 680 m southwest. Each field was treated as an independent experimental unit. All fields had similar soil texture, pH, and EC (Table 1). Only one field was used for each farming system, due to logistical constraints. Eight plots (3 m × 3 m) were randomly spaced along the first 300 m of each field. These plots served as spatial replicates, resulting in pseudoreplication, which is a common experimental design used in large-scale agronomic trials [26,27].

Table 1. Soil pH, electrical conductivity (EC), and texture in bulk soil in the three study fields. Data is presented as mean ± standard deviation. There were no significant differences between fields ($p > 0.05$ by ANOVA).

	Wheat	Kernza	CRP
pH	7.68 ± 0.11	7.72 ± 0.21	7.32 ± 0.24
EC (µS cm^{-1})	14.5 ± 2.94	24.8 ± 9.09	18.0 ± 9.5
Clay (%)	13.0 ± 0.3	15.5 ± 2.0	16.9 ± 3.9
Silt (%)	49.0 ± 5.1	50.6 ± 2.4	50.8 ± 3.2
Sand (%)	38.1 ± 5.3	33.9 ± 3.7	32.3 ± 7.1

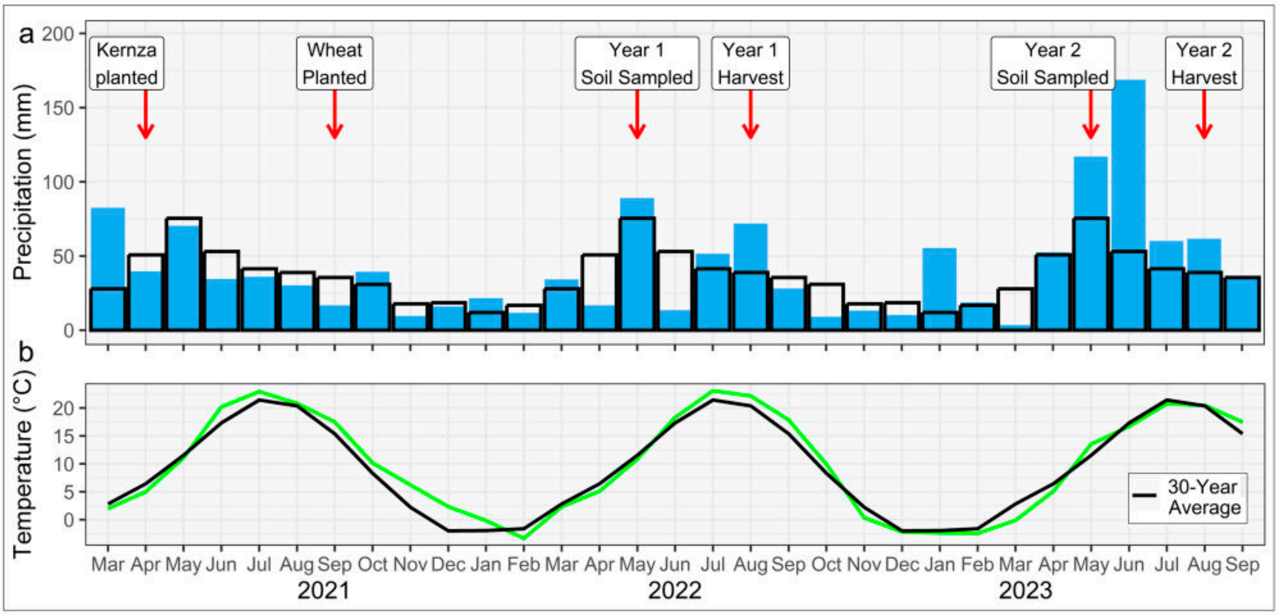

Figure 1. Precipitation (**a**) and temperature (**b**) at the study site compared to the 30-year averages (in black). Arrows indicate the timing of major events in the study.

In the wheat–fallow field, wheat was planted mid-September 2021 following fallow and harvested late July 2022 (variety Westbred 2262, 78 kg ha^{-1} seeding rate, 28.9 cm row spacing) (Figure 1). After harvest, the field returned to fallow, with light disk tillage for weed control. Both the CRP and Kernza fields were managed as wheat–fallow prior to conversion. Kernza was planted in April 2021 (variety TLI-C5, 13.5 kg ha^{-1}, 24.4 cm row spacing), following 10 months of fallow. The field was sprayed with 2,4-dichlorophenoxyacetic acid (2,4-D) (0.804 L ha^{-1}) in July 2021 and mowed in August 2021 for weed control. The CRP field was established in 2012 by planting a mixture of wheatgrasses and alfalfa (34% pubescent wheatgrass (*Thinopyrum intermedium* sp. *barbulatum*), 30% IWG, 16% slender wheatgrass (*Elymus trachycaulus* (Link) Gould ex Shinners), 14% western wheatgrass (*Pascopyrum smithii* (Rydb.) A. Löve), and 6% alfalfa (*Medicago sativa* L.) by seed weight). None of the study fields were fertilized or irrigated, as is typical in this system.

Due to farmer retirement and the fact that the wheat–fallow field was in the fallow phase in year 2, no wheat was grown at the study farm in 2023. As a result, year 2 wheat biomass and rhizosphere soil were collected from a nearby farm (41.885861 N, −104.747139 W, 1590 m elevation) with comparable soil type and field management history (a long-term wheat–fallow rotation with light tillage for weed control and no fertilization).

2.2. Soil and Plant Sampling

Plant and soil samples were collected from each plot (24 plots total) once per year. Aboveground plant biomass was sampled in 2022 and 2023 at plant maturity, 1–2 weeks before harvest (July for wheat and CRP; August for Kernza) (Figure 1). Plants were cut at ground level using hand clippers in three 60 cm^2 quadrats per plot in CRP or three 1 m row sections per plot in Kernza and wheat. Row spacing was recorded to convert biomass to area-based units (kg ha^{-1}). Aboveground plant biomass was dried, weighed, and threshed using a laboratory thresher (Haldrup LT-21, HALDRUP GmbH, Ilshofen, Germany), and hand-cleaned to determine yield. Kernza grain yield was corrected (reduced) by 20% to account for hull mass, which is typically removed during industrial processing (personal communication, Hana Fancher, The Land Institute). Root biomass was sampled in June 2023, when winter wheat was transitioning to heading (stage 10.1) and root biomass was expected to be greatest [28]. All roots were excavated from a 1 m^2 area in plots 1, 4, and 8 to a depth of 60 cm. This maximum depth was decided based on the fact that no roots were observed below 60 cm. Roots were washed, separated from the roots of weeds and any aboveground biomass, air-dried, and weighed.

Bulk soil samples were collected from each plot in late May 2022 and 2023, around peak plant growth (the wheat boot stage, stage 10). Each sample consisted of eight 0–15 cm cores composited together. Separate cores were simultaneously collected for soil bulk density (0–15 cm, 2.03 cm diameter, five cores per plot). Rhizosphere soil was sampled 1–2 weeks later (early June) from the same plots. To collect rhizosphere soil, roots were carefully excavated from beneath 3–5 plants per plot (up to 30 cm depth) and shaken to remove loose soil. The remaining soil adhering to roots was brushed and shaken into sterilized containers, composited per plot, and transferred to sterile Whirl-Pak® bags (Merck KGaA, Darmstadt, Germany). In the CRP field, rhizosphere soil was collected from wild-type IWG plants only.

Soil samples were kept cool and sieved in the lab to 8 mm within 24 h for field-moist soil analyses (gravimetric moisture and dissolved organic C and N (DOC and DON)). All soil was later sieved to 2 mm to remove remaining gravel. At the same time, subsamples were sieved to 2 mm and frozen at −20 °C for phospholipid fatty acid (PLFA) analysis, enzyme assays, and DNA extraction. The remaining soil was air-dried for analysis of pH, electrical conductivity (EC), soil protein, permanganate oxidizable C (POXC), aggregate stability, total C and N, inorganic C, and texture.

2.3. Soil Analyses

Soil structure (bulk density, aggregate stability, and texture) was analyzed on bulk soil samples only. Bulk density was analyzed by drying and weighing cores of a known volume. Aggregate stability was analyzed by wet sieving to 250 µm with a Yoder-style apparatus for 5 min, then drying and weighing intact aggregates [29]. Texture was analyzed by hydrometer on three plots per field only [30]. Gravimetric moisture was analyzed by oven-drying at 105 °C for 24 h. Soil pH and EC were analyzed by electrodes (Thermo Scientific Orion Start A215, Thermo Fisher Scientific Inc., Waltham, MA, USA) in a 1:2 (*w*:*v*) soil:deionized water mixture.

For organic matter dynamics, we analyzed SOC, total N, DOC, DON, POXC, and soil protein. DOC and DON were extracted with 0.5 M potassium sulfate in a 1:5 (*w*:*v*) soil:solution mixture, horizontally shaken for 30 min at 200 rpm, filtered through Q5 filter paper (Fisher Scientific, Waltham, MA, USA), stored at −20 °C, and quantified using combustion catalytic oxidation/non-dispersive infrared detection on a Shimadzu TOC-VCSH (Shimadzu Corporation, Kyoto, Japan) [31]. Soil protein was extracted using the autoclaved citrate-extractable protein method and quantified on a BioTek Synergy

HTX multimode reader (BioTek, Winooski, VT, USA) at 562 nm [32]. POXC was analyzed by reaction with 0.2 M potassium permanganate and quantified on the same reader at 550 nm [33]. Total C and N were analyzed by dry combustion on a LECO C/N Analyzer 928 Series (LECO Corporation, St. Joseph, MI, USA). Inorganic C was analyzed by reaction with 6 M HCl and 3% $FeCl_2$, with CO_2 quantified on an LI-COR LI-820 CO_2 Gas Analyzer (LI-COR, Lincoln, NE, USA) [34]. SOC was calculated as the difference between total C and inorganic C. SOC, total N, DOC, and DON were only analyzed in year 2.

We measured microbial biomass using PLFAs [35] and microbial activity using potential extracellular enzyme activities [36]. PLFAs were extracted from 1 g of freeze-dried soils using a modified Bligh–Dyer method as in the NEON protocol [37] and analyzed on an Agilent 6890 gas chromatograph (Agilent Technologies, Santa Clara, CA, USA). Peaks were identified using the MIDI Sherlock software v4.5, with quantification based on the internal standard 19:0. Total microbial biomass was calculated as the sum of all PLFA biomarkers, according to Joergensen [35]. Extracellular enzyme activities were analyzed by reaction with the appropriate 4-methylumbelliferone (MUB) or 7-amido-4-methylcoumarin (AMC)-labeled substrate and fluorescence measurement on a Synergy HTX multimode reader at 360 nm excitation and 450 nm emission, as in Custer et al. [38]. Fluorescence conversions were based on 10 μM MUB and AMC standards. Eight enzymes representing C, N, P, and S cycling were assayed: cellobiohydrolase (CBH) and β-glucosidase (BG) target cellulose, β-xylosidase (BX) targets hemicellulose, α-glucosidase (AG) targets starch, N-acetyl glucosamine (NAG) targets chitin, leucine aminopeptidase (LAP) targets polypeptides, phosphatase (PHOS) targets phosphate esters, and arylsulfatase (SUL) targets ester sulfates.

2.4. DNA Extraction, Sequencing, and Processing

Amplicon sequencing was used to characterize bacterial and AMF community composition in rhizosphere soil only. DNA was extracted from 0.25 g of soil using a Qiagen DNeasy PowerSoil Pro Kit (Qiagen, Hilden, Germany) and stored at −20 °C until library prep. DNA concentration and quality were assessed on a Synergy HTX multi-mode reader. Library preparation and PCR were conducted with a one-step library protocol with 9 μL of PCR master mix (3 μL 5X KAPA HiFi Buffer, 0.45 μL 10 M dNTPs, 0.3 μL Kapa HiFi HotStart DNA Polymerase, and 5.25 μL HPLC H_2O) combined with 2 μL of 0.75 μM paired primers and 2 μL of normalized (10 ng μL^{-1}) DNA. Primers contained sample-specific barcodes. PCR amplification conditions for bacteria were: 95 °C for 3 min, 35 cycles of 98 °C for 30 s, 35 cycles of 62 °C for 30 s, 35 cycles of 72 °C for 30 s, 72 °C for 5 min, and a hold at 4 °C. For AMF, the PCR conditions were: 95 °C for 10 min, 35 cycles of 95 °C for 30 s, 35 cycles of 55 °C for 30 s, 35 cycles of 72 °C for 60 s, 72 °C for 9 min, and a hold at 4 °C. Following amplification, PCR products were purified using modified manual AxyPrep MagBead PCR Clean-Up (Corning Incorporated, Corning, NY, USA), and molar concentrations were checked on a BioTek Synergy HTX multimode reader using a BioTek Take3 Micro-Volume Plate (BioTek, Winooski, VT, USA).

Samples were sequenced at the University of Wyoming Genome Technologies Lab on an Illumina NextSeq 2000 (Illumina, San Diego, CA, USA) using paired-end 2 × 300 bp chemistry. Bacteria were assessed by amplifying the V4 region of the 16S rRNA gene using primers 515F [39] and 806R [40]. AMF were assessed by amplifying a region of the 18S rRNA gene using primers AMV4.5NF and AMDGR [41]. Initial sequence processing was performed using Vsearch v2.27.0 [42]. Raw reads were trimmed of primers, merged, and filtered to remove sequences with >1 expected error. Reads were clustered into exact sequence variants (ESVs) at 100% similarity. Contaminants were identified as taxa with higher mean abundance in blanks than in samples and were removed. Taxonomy was assigned using the RDP 16S database (v18) for bacteria [43] and the MaarjAM 2019 database for AMF [44].

2.5. Statistical Analyses

Data were analyzed in the program R (version 4.3.1). All data and analysis scripts are available at https://github.com/hrodgers5/Kernza_Wyoming (accessed on 1 November 2025), and amplicon sequence data have been deposited in the National Center for Biotechnology Information Sequence Read Archive under BioProject ID PRJNA1173200. Differences in soil properties and plant biomass between the three study fields were evaluated using spatial linear models fit by generalized least squares (GLS) in the nmle package, with field as a fixed effect. Because treatments were applied at the field level without true replication, spatial autocorrelation among plots was accounted for using an exponential correlation structure based on plot geographical coordinates. This approach accounts for non-independence among plots due to spatial proximity. Each year was analyzed separately due to significant year × field interactions for most variables. Pairwise comparisons among fields were conducted using estimated marginal means with Tukey-adjusted pairwise comparisons in the emmeans package. Model residuals were tested for normality using the Shapiro–Wilk test, with Box–Cox power transformations to achieve normality when necessary. p-values were considered statistically significant at $p < 0.05$.

Multivariate differences in enzyme activities were assessed using permutational multivariate ANOVA (PERMANOVA) based on Bray–Curtis dissimilarity matrices in the vegan package [45]. Post hoc pairwise comparisons between fields were conducted with the pairwiseAdonis package [46]. Principal component analysis (PCA) was used to visualize these differences. For amplicon sequence data, alpha diversity (ESV richness and Shannon index) was analyzed after rarefaction using the phyloseq package [47]. Differences in alpha diversity between fields were analyzed using spatial linear models, as described above. Beta diversity was analyzed using PERMANOVA on Bray–Curtis distance matrices and visualized using non-metric multidimensional scaling (NMDS) in the *vegan* package.

3. Results

3.1. Grain and Biomass Yield

The year 1 growing season (August 2021–July 2022) was dry (351 mm of precipitation, 84% of the 35-year average), and crops experienced hail damage in July (Figure 1). Kernza and winter wheat produced marginal grain yields but substantial aboveground biomass (Figure 2a,b). In contrast, the year 2 growing season (August 2022–July 2023) was wet (610 mm, 145% of the long-term average), and Kernza produced 13% of the grain and 59% of the aboveground biomass of wheat, but 280% of the root biomass of wheat (Figure 2a–c). Representative Kernza, wheat, and CRP IWG plants and their entire root systems are shown in Figure 3.

3.2. Bulk Soil Properties

Most bulk soil health indicators did not differ between fields, with no differences between Kernza and wheat–fallow (Table 2). Kernza differed from CRP by having lower bulk density in year 1 and higher DOC in year 2. Enzyme activity differed between all fields in both years, with the highest overall activity in CRP (PERMANOVA $p < 0.01$) (Figure 4a,b). Soil moisture was highest in fallow and lowest in winter wheat (Table S1).

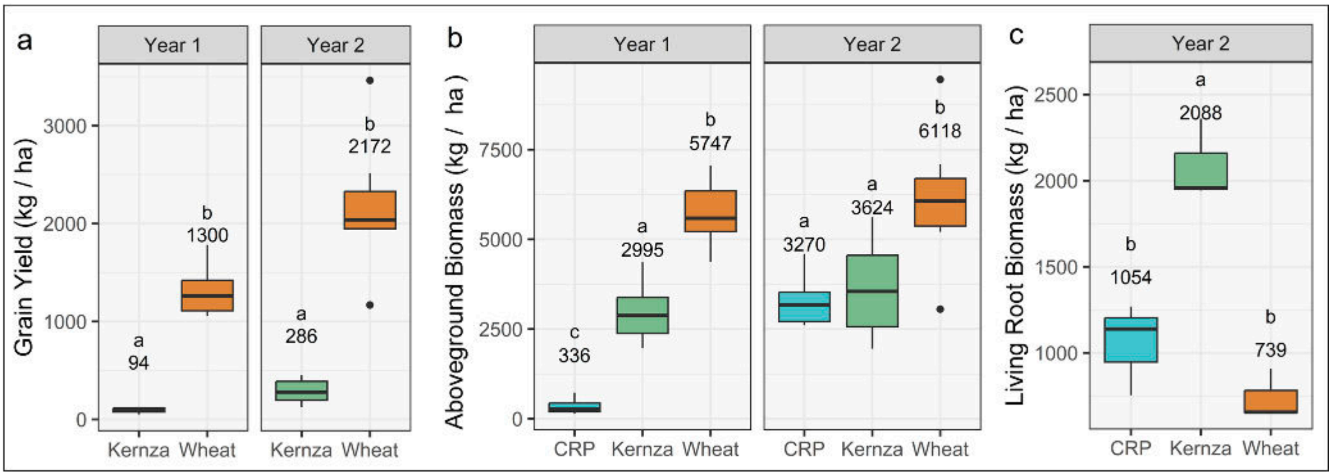

Figure 2. Dry grain yield (**a**), dry aboveground biomass (**b**), and dry root biomass (**c**) from Kernza, winter wheat, and Conservation Reserve Program (CRP) fields. Numbers above boxplots state the mean value, and different letters indicate significant differences between fields ($p < 0.05$ by ANOVA).

Figure 3. Representative plants of winter wheat, Kernza, and wild-type intermediate wheatgrass from the Conservation Reserve Program field (CRP IWG). The entire root system of each plant is shown next to a measuring stick (cm). Root systems were shallow, likely because plants rely more on surface moisture from sporadic rainfall than on soil water stored at depth in this system. Plants were excavated by hand in June of year 2.

Table 2. Soil health indicators in bulk soil from wheat–fallow, Kernza, and Conservation Reserve Program (CRP) fields. Data is presented as mean ± standard deviation. Year 2 data were measured in the fallow year of the wheat–fallow rotation. Different lowercase letters indicate significant differences between fields ($p < 0.05$ by ANOVA). C is carbon, and N is nitrogen.

Bulk Soil				
	Year	**Wheat (yr 1)/ Fallow (yr 2)**	**Kernza**	**CRP**
Soil protein (g kg^{-1})	1	2.51 ± 0.97	2.51 ± 0.57	2.10 ± 0.38
	2	2.55 ± 0.24	2.54 ± 0.31	2.56 ± 0.33
Permanganate oxidizable C (mg kg^{-1})	1	257 ± 55	259 ± 27	265 ± 17
	2	239 ± 41	256 ± 38	280 ± 17

Table 2. *Cont.*

		Bulk Soil		
	Year	**Wheat (yr 1)/ Fallow (yr 2)**	**Kernza**	**CRP**
Water-stable aggregates (%)	1	42.8 ± 8.5	46.6 ± 10.3	49.9 ± 6.3
	2	31.8 ± 14.4	39.9 ± 14.1	43.8 ± 10.1
Bulk density (g cm^{-3})	1	1.39 ± 0.04 ab	1.34 ± 0.05 b	1.44 ± 0.07 a
	2	1.48 ± 0.04	1.46 ± 0.08	1.45 ± 0.08
Microbial biomass (nmol g^{-1})	1	19.9 ± 5.44	19.4 ± 4.43	17.0 ± 5.35
	2	41.3 ± 10.5	43.6 ± 6.7	45.6 ± 11.0
Dissolved organic C (mg kg^{-1})	2	36.3 ± 13.3 ab	39.9 ± 16.5 a	15.3 ± 9.8 b
Dissolved organic N (mg kg^{-1})	2	2.85 ± 0.88 a	2.11 ± 0.60 ab	1.68 ± 0.75 b
Total N (%)	2	0.140 ± 0.007	0.147 ± 0.005	0.138 ± 0.010
Soil organic C (%)	2	0.992 ± 0.08	1.05 ± 0.06	1.01 ± 0.09

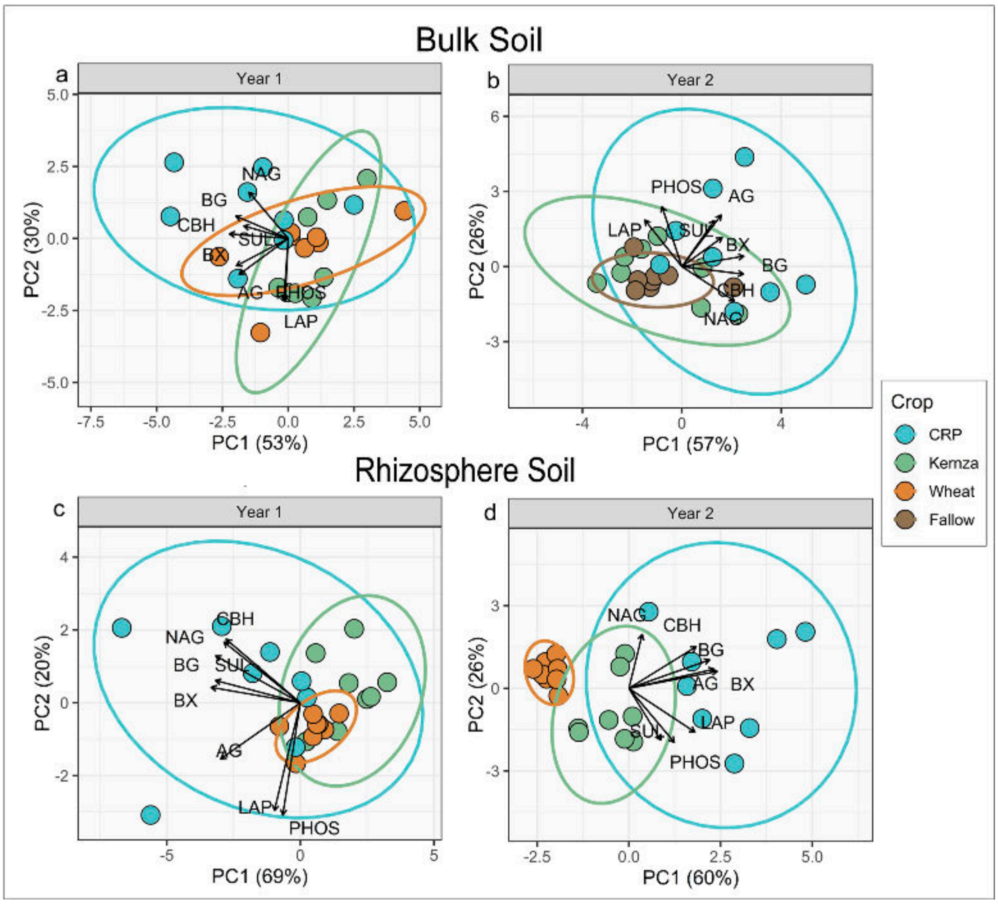

Figure 4. Principal component (PC) analysis of enzyme activities with loadings shown in bulk soil (**a**,**b**) and rhizosphere soil (**c**,**d**) from Kernza, wheat–fallow, and Conservation Reserve Program (CRP) fields. Points closer together represent samples that are more similar. PC1 and PC2 represent the primary patterns of variation in the dataset, and the percentages indicate the proportion of total variation explained by each axis. Rhizosphere soil in the CRP field was taken from wild-type intermediate wheatgrass (IWG) plants. Overall, enzyme activity differed between crops in both bulk soil and rhizosphere soil in both years (PERMANOVA $p < 0.05$). CBH is cellobiohydrolase, NAG is N-acetyl-β-glucosaminidase, BG is β-glucosidase, BX is β-xylosidase, AG is α-glucosidase, LAP is leucine aminopeptidase, PHOS is phosphatase, and SUL is arylsulfatase.

3.3. Rhizosphere Soil Properties

In year 1, the Kernza rhizosphere was more similar to wheat than to CRP IWG (Figure 5a–c). In year 2, the reverse was true: all indicators except DON were higher in Kernza than in wheat and not different from CRP IWG (Figure 5d–j). Enzyme activity differed between all crops (PERMANOVA $p < 0.01$) except between year 1 wheat and Kernza (Figure 4c,d). Enzyme activity was highest in CRP in both years and higher in year 2 in Kernza than in wheat (PERMANOVA $p = 0.002$). Rhizosphere pH and EC did not significantly differ between the three crops (Table S2).

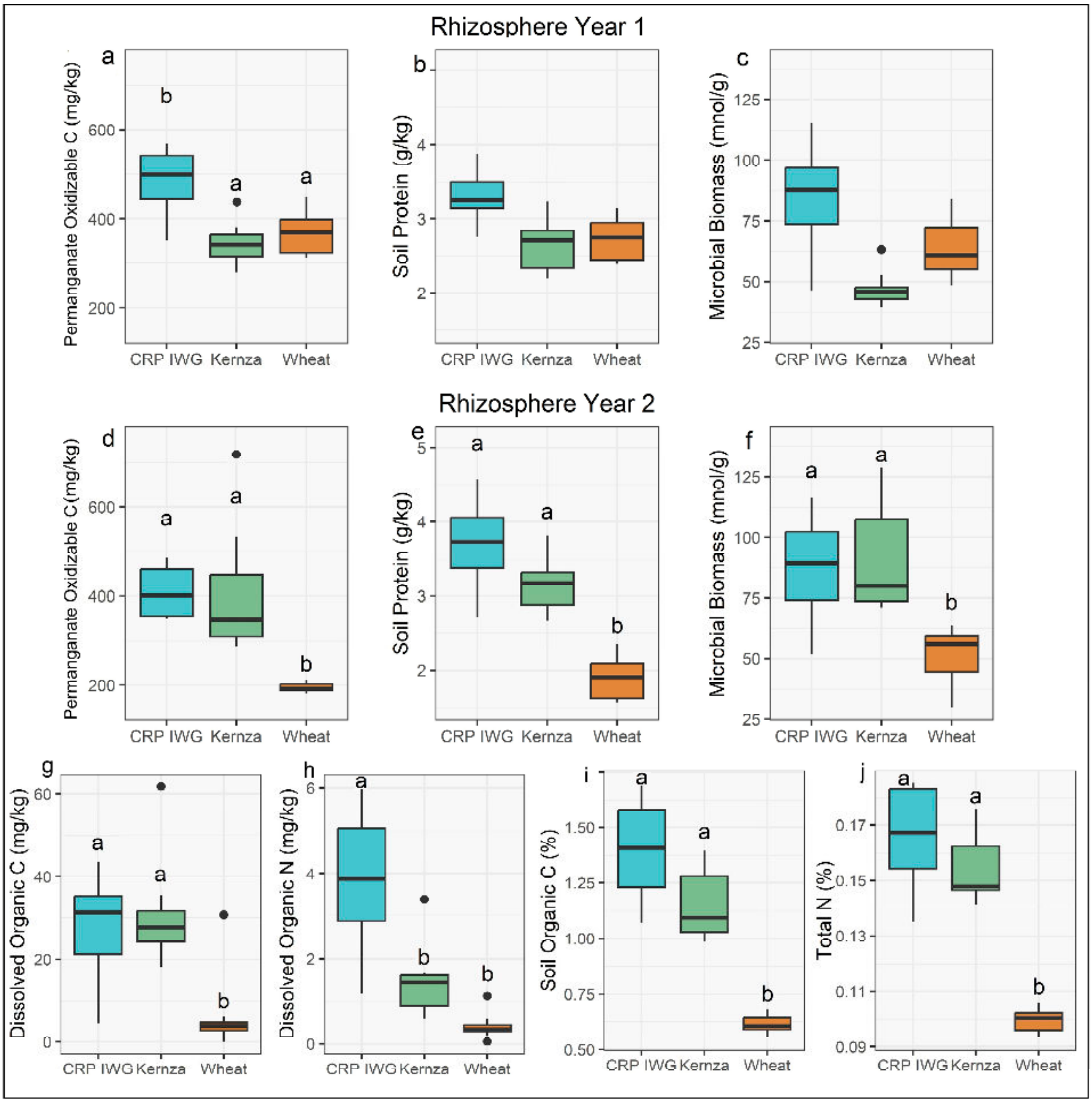

Figure 5. Soil health indicators in rhizosphere soil of winter wheat, Kernza, and wild-type intermediate wheatgrass from the Conservation Reserve Program field (CRP IWG). Permanganate oxidizable C (**a**,**d**), soil protein (**b**,**e**), and microbial biomass (**c**,**f**) were analyzed in both years. Dissolved organic C (**g**), dissolved organic N (**h**), soil organic C (**i**), and total N (**j**) were only analyzed in year 2. Different letters indicate significant differences between fields ($p < 0.05$ by ANOVA). C is carbon, and N is nitrogen.

3.4. Rhizosphere Soil Microbial Community

For bacteria, samples yielded 43,700–122,300 total reads from 19 phyla. A total of 10.9% of ESVs were unassigned at the phylum level and were removed, resulting in 18,769 bacterial ESVs. For AMF, two samples with fewer than 3000 reads were excluded. The remaining samples yielded 3480–69,250 total reads, all in the phylum *Mucoromycota*. In total, 13.6% of ESVs were unassigned at the class level and were removed, resulting in 4479 AMF ESVs. For alpha diversity for bacteria, ESV richness was higher in Kernza than in wheat in year 2, and Shannon diversity was higher in Kernza in both years (Table S3). For AMF, ESV richness was highest in Kernza in both years, and Shannon diversity did not differ between crops. For beta diversity, both AMF and bacterial community composition differed between all three crops in both years (PERMANOVA $p < 0.001$) (Figure 6). Kernza rhizosphere microbial communities were more similar to wheat in year 1 and more similar to CRP IWG in year 2.

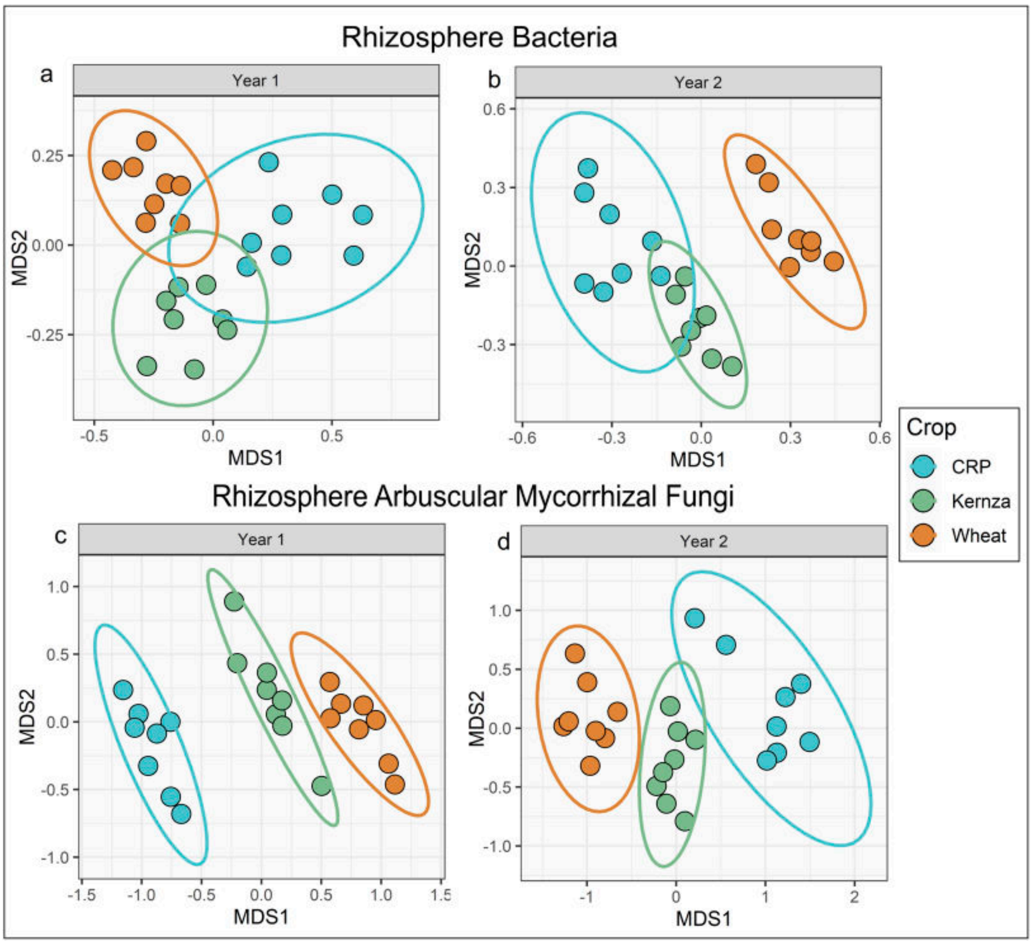

Figure 6. Community composition of bacteria (**a**,**b**) and arbuscular mycorrhizal fungi (**c**,**d**) in rhizosphere soil in Kernza, winter wheat, and Conservation Reserve Program wild-type intermediate wheatgrass (CRP IWG). Community composition differed significantly between crops for both bacteria and arbuscular mycorrhizal fungi in both years (PERMANOVA $p < 0.05$).

4. Discussion

4.1. Rhizosphere and Bulk Soil Health

Because the study relied on pseudoreplication and used a different wheat field in year 2, conclusions about Kernza’s effects on overall soil health are limited. However, we have greater confidence that rhizosphere differences reflect plant-driven effects, given the strong influence of roots on rhizosphere soil [48,49] and the similarity in soil type and management history across fields.

We found few differences in bulk soil properties between Kernza and wheat–fallow fields, likely because soil changes in semiarid environments often require more than three years to become detectable due to slow plant and microbial growth [50–53] (Table 2). In contrast, studies conducted over longer periods or in wetter environments have reported improvements in SOC, microbial biomass, and other soil health indicators following transition from annual cropland to Kernza [11,14,15,54]. Many bulk soil properties also did not differ between CRP and wheat–fallow fields, potentially due to high spatial variability or limited statistical power.

In contrast, rhizosphere soil in year 2 Kernza exhibited higher organic matter, microbial biomass, and enzyme activity than in wheat (Figures 4 and 5). Kernza also produced nearly three times the total root biomass of wheat (Figure 2c), and several studies have linked higher root biomass to improved soil health and SOC accumulation [55–57]. Elevated rhizosphere organic matter likely reflects root exudates, which can contribute to longer-term SOC accumulation [57]. In Kansas, transitioning from winter wheat to Kernza sequestered an estimated 370 g C m^{-2} yr^{-1} over 4.5 years based on eddy covariance measurements [58]. A Michigan-based model predicted that Kernza would allocate 3–7 times more primary production to root exudates (as DOC) than winter wheat, supporting higher soil bacterial biomass and SOC accumulation [59]. Consistent with these predictions, Kernza in our study produced nearly three times more root biomass and five times more DOC per g of rhizosphere soil than wheat.

4.2. Rhizosphere Microbial Community

AMF and bacterial communities in the Kernza rhizosphere were more similar to winter wheat (negative reference) in year 1 but shifted toward CRP IWG (positive reference) in year 2 (Figure 6), possibly in response to higher Kernza rhizosphere organic matter in year 2 (Figure 5). This finding suggests that Kernza may retain some microbial characteristics of wild-type IWG even when managed as a row crop.

Although no studies have directly compared Kernza and wheat rhizosphere microbiomes, findings from bulk soil studies show similar patterns. Research has reported higher microbial biomass and AMF diversity in Kernza than in wheat after 3–5 years [14,60] and has shown that fungal communities under Kernza more closely resemble those under perennial grassland than under wheat [15]. Reduced soil disturbance, recalcitrant root tissue inputs, and year-round roots and ground cover in Kernza fields may contribute to these differences [14,60].

4.3. Agronomic Viability

In year 1, Kernza and wheat both produced marginal grain yields and would likely have been harvested for forage instead of grain (Figure 1a,b). In year 2, Kernza produced 229 kg ha^{-1} (203 lbs $acre^{-1}$) of grain, 27% less than the Great Plains Kernza average that year [61] and only 13.2% of the wheat yield. In contrast to results from the Midwestern US [11], we did not observe yield decline with stand age in the first 2 years, possibly due to the shorter and drier growing season. A concurrent study at the nearby University of Wyoming Sustainable Agriculture Research and Extension Center (SAREC) found that irrigated Kernza produced 737 kg ha^{-1} grain in 2022 and 1242 kg ha^{-1} in 2023 and that irrigation increased Kernza grain yields by 380% and forage by 170% (unpublished data, Hannah Rodgers, 2021–2023). Together, these findings suggest that water limitation suppressed grain yield more than forage yield and that Kernza may be better suited for forage than grain during drought years.

Additionally, an economic analysis building off the current study found that Kernza's higher market value, lower input costs, and forage value could offset low grain yields [62]. The analysis suggested that under some scenarios, Kernza could produce higher economic returns than wheat–fallow in this environment, largely depending on market access and grain price. Overall, Kernza's agronomic performance and soil health effects vary across climatic and soil conditions, warranting further research in new environments.

4.4. Conclusions

This preliminary study provides insights into Kernza's performance in drought-prone environments and highlights directions for future research. Kernza's high root biomass and rhizosphere organic matter suggest the potential for improved soil health in land transitioning from wheat–fallow, though the study was likely not long enough to detect significant changes, and longer-term studies are needed. Kernza also supported a rhizosphere microbial community more similar to CRP IWG than to wheat, indicating that it retains characteristics of wild-type intermediate wheatgrass under row-crop management. Although dryland grain yields were low, they could be partially offset by benefits such as a higher market value, soil health improvements, and the flexibility to harvest either grain or forage during drought. While Kernza remains in the early stages of development, these benefits may make it a viable alternative to annual grains for some farmers in drought-prone environments.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/agronomy16060624/s1. Table S1: Soil moisture in bulk and rhizosphere soils; Table S2: pH and EC in rhizosphere soil; Table S3: Alpha diversity indices for bacteria and arbuscular mycorrhizal fungi in rhizosphere soil.

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Data Availability Statement: All data and scripts used for analysis are available at https://github.com/hrodgers5/Kernza_Wyoming (accessed on 1 November 2025), and genetic sequence data are available through the National Center for Biotechnology Information Sequence Read Archive under BioProject ID PRJNA1173200.

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