FINAL REPORT

TITLE: Effect of Cover Cropping Systems on Dryland Soybean Plant-vigor, Growth, and Yield MSPB Project (21-2022)

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Background and Objectives

In 2020, an estimated 2 million acres of soybeans were planted in Mississippi. There is approximately 56% of the acres planted in a dryland environment on any given year. Therefore, dryland soybeans can make-up more than half of the soybean acres in Mississippi. Inconsistent yields are a problem for many dryland soybean producers due to the lack of rainfall at critical times during the growing season. In many areas, irrigation is not an option and different production practices need to be evaluated to determine if more consistent yields can be achieved each year. There is an increased awareness of soil as a living ecosystem that we rely on to grow quality crops for food, feed, and non-food uses. It has been suggested that cover crops offer many benefits for crop productivity, soil health, and environmental sustainability such as improved water infiltration and soil moisture retention, promotion of soil micro-organism diversity, improved nutrient availability, and enhanced soil organic matter (SOM), which can improve soybean yields. In 2017, cover crops were planted on 15.4 million acres across the U.S. and slightly more than 139,000 acres in Mississippi. This is a 111% increase for planting cover crops in Mississippi from 2012 to 2017. Fertilizer requirements may differ with a cropping systems approach compared to conventional production practices because cover crops scavenge leftover nutrients from the previous crop and release them as they decay. In addition to cover crops, the use of poultry litter as a nutrient source could be used to enhance SOM, increase water holding capacity, improve nutrient retention, and provide micro-nutrients that may be needed to improve soil health. The practice of no-tillage could also be utilized to improve water infiltration and retention of soil moisture, help build SOM by leaving plant residue on the soil surface, and to reduce fuel / labor requirements. No-tillage accounted for 96.5 million acres across the U.S. in 2012, which is nearly 35% of all cropland planted. In Mississippi, no-tillage accounts for a slightly more than 620,000 acres, which is only 16.8% of all cropland. Another strategic factor in this system is cover crop termination and soybean planting date. Earlier plantings tend to have higher yields in Mississippi due to growth and development before drought conditions occur. Therefore, this study is focused on identifying production practices that will provide more consistent soybean yields while improving "soil health" with the following objectives: 1). Evaluate the effects of cover cropping systems on dryland soybean plant-vigor, growth, and yield; 2). Evaluate the effects of cover cropping systems on "soil health"; and 3). Determine the economic benefit of cover crops, source of fertilizer, and planting date on soybean production.

Report of Progress/Activity

Objective 1: Evaluate the effects of cover cropping systems on dryland soybean plant-vigor, growth, and yield. Cover crop (cc) treatments consisting of (1) native vegetation (NV), switched to bare soil last 2 years due to encroachment of adjacent cc, (2) NRCS recommended mix of cereal rye + mustard (CRm), (3) cereal rye (CR), (4) wheat (WH), and (5) vetch (VE) was planted in the fall following soybean. The cc treatments were terminated either *early* (referred to as early system with soybean planted end of April) or *late* (referred to late system with soybean planted end of May). Early planting was accomplished on dates appropriate to the maturity group/variety utilized, while late planting was delayed approximately one month to provide a longer growing period for cc treatments compared to the earlier planting system. Samples (m²) of cc treatments were collected from each plot in the spring prior to termination to determine biomass. Soybeans were no-till planted into cc residue (~ 2 weeks after cc termination) for both early and late systems. In addition to cc crop species and cc growth periods, subplots of three fertilizer regimes were evaluated, namely (1) no fertilizer check, (2) standard inorganic (ING) commercial fertilizer where rate is based on MSU soil test recommendations, and (3) poultry litter that coincides with the ING fertilizer rate based on nutrient analysis of the litter. Physiological parameters of soybeans, such as stand counts and leaf area index, were measured. Yield data were collected at the end of season and cc treatments were reestablished in the fall. Conventional early season insect and weed management was practiced attributing any observed differences in soybean vigor, growth, and yield to treatment effects.

Crop properties

Temperature and rainfall distribution for all years were recorded (Table 1). A summary of statistical analyses for crop-related variables are described in the analysis of variance (ANOVA), shown in Table 2 (early planting) and Table 3 (late planting).

The variation in annual soybean yield across all treatments for early planting and late planting are illustrated in Figure 1. For the early planting (Fig. 1a), yield in 2019 was greater than all other years, where 2018 and 2022 yields were the lowest among the five years (note: 3500 kg/ha equals 52 bu/Ac). With respect to late planted soybean yields (Fig. 1b), the 2021 yield was greater than all other years and as with the early planting, 2018 and 2022 yields were the lowest among the years. The general pattern of these results may be attributable in large part to rainfall patters, where the sum of May, June and July rainfall amounts were notably higher for 2019 and 2021. Annual yields as effected by fertilizers are illustrated in Figure 2. For the early planting (Fig. 2a), soybean yield with both ING and poultry litter was greater compared to the no fertilizer check. Notably, yields where nutrients were applied via poultry litter were approximately 6-10 bushels/Ac higher than where ING fertilizer was used for the years from 2019 to 2022 (Fig. 2a.). For late planted soybeans (Fig. 2b), soybean yield with either fertilizer source was also greater compared to the no fertilizer check. However, the positive effect of poultry litter observed in the early planting system did not appear until 2021 and 2022. Previous research in Mississippi has shown that poultry litter can improve the plant-available water holding capacity of soil through its effect on soil structure and can lead to improvements in 'carryover' nutrients from previous applications. Soybean yields as they relate to the interaction between cc species are presented in Figure 3 (see also Tables 2 and 3). For the early planting system, there was only a difference in soybean yield with poultry litter and cc combinations of

NV, CRm, and VE compared to ING (Fig. 3a). However, yield with both sources of fertilizer was greater than the no fertilizer for CR and WH. In the late planting system, there was no difference between combinations of cc and fertilizer source (p = 0.885) (Fig. 3b). However, soybean yield was greater with both fertilizer sources compared to the no fertilizer check.

Annual soybean leaf area index (LAI) as effected by fertilizer treatment for early and late planted soybeans is illustrated in Figure 4. An LAI measurement is the ratio of one-sided leaf area per unit ground area and therefore quantifies the amount of leaf material in a canopy. Since this variable represents the space available for light interception it is related to net photosynthetic rate and transpiration, which can affect yield making it an indicator of crop development. For early planted soybean (Fig. 4a) fertilizer applications resulted in higher LAI values as compared to the no fertilizer check. For the years beginning with 2020, poultry litter applications resulted in higher LAI than ING fertilizer. LAI for late planted soybeans followed a similar pattern, with the poultry litter effect appearing in 2021 and 2022 (Fig. 4b). LAI measurements reflect the same trend as soybean yield across years (see Fig. 2a and b).

The soybean chlorophyll index for each year as effected by fertilizer source is shown in Figure 5. The chlorophyll index contains green and red spectral bands and is used to calculate the total chlorophyll content in plants. It is reported as being well-correlated with the crop nutrient status, the harvest index, and yield, which is a makes it a useful indicator of crop status. In this study, chlorophyll index values were only higher with fertilizer treatments compared to the no fertilizer check in 2 of 5 years in the early and 3 of 5 years in the late system. Therefore, it did not prove to be a strong indicator of crop status in this study.

The soybean leaf tissue concentration of phosphorous (P) and potassium (K) are shown in Figures 6-8. For early and late planted soybeans, the P tissue content as effected by fertilizer applications followed the same trends as with LAI, where applied nutrients resulted in higher values (Fig. 6 a and b). Values of tissue K content trended slightly higher with poultry litter compared to ING and no fertilizer check (Fig. 7 and b). A cc-fertilizer effect was observed, but it was only with WH in the early planting system where P tissue content with ING fertilizer was the only fertilizer source that was different compared to no fertilizer.

Cover Crop properties

In general, more cover crop biomass delivers more benefits such as nutrient retention, weed suppression, and soil structure/water availability. Annual cover crop biomass, which was collected prior to termination, is shown in Tables 4 and 5. Although there were significant differences in the biomass among the cc treatments, clear patterns were not readily observed due to a high degree of variability in the data (note values of standard deviation).

Soil variables properties

The statistical analysis summaries and soil test nutrient-based properties are shown in Tables 6-9. These samples were used to devise fertilizer application rates. With respect to the treatment, they can be useful in assessing carryover nutrient contents. The effects observed (see Tables 6 and 7) were due primarily to differences where nutrients were applied (poultry litter and ING fertilizer) as compared to the no fertilizer check (see Tables 8 and 9).

Objective 1 Graphics and Tables

Table 1: Monthly average temperature and precipitation data over the experimental period, from 2018-2022.

	Temperature (°C)							Precipitation (mm)					
Month	2018	2019	2020	2021	2022	2018	2019	2020	2021	2022			
January	3.5	6.3	8.7	6.5	4.7	59.9	189.2	188.0	53.3	96.5			
February	11.6	11.0	8.7	5.0	7.6	325.4	405.9	260.1	148.6	170.7			
March	12.1	11.0	16.0	14.7	12.8	86.1	89.2	176.5	164.6	141.5			
April	14.0	17.4	15.6	15.9	16.5	181.9	239.0	144.8	104.1	164.3			
May	24.0	23.5	20.6	20.2	22.7	125.5	138.2	109.7	162.1	131.6			
June	26.3	23.1	25.1	24.9	26.5	165.1	146.3	124.0	207.0	43.7			
July	27.0	27.1	27.7	26.4	28.1	73.2	234.4	189.2	210.6	86.1			
August	26.5	27.0	26.3	26.5	26.5	115.8	190.0	150.6	205.5	147.6			
September	25.3	27.2	22.9	23.2	23.3	127.0	1.5	106.4	74.7	39.1			
October	19.2	17.9	17.8	19.1	16.4	58.4	175.8	114.3	61.2	85.1			
November	8.7	9.0	13.0	10.2	11.9	137.7	69.3	35.6	4.3	80.5			
December	8.1	9.1	7.1	13.4	7.8	159.0	127.8	123.4	96.0	164.6			

Table 2: Analysis of variance (ANOVA), probability values (p-values) for the treatment effects and interactions between cover crop, fertilizer source treatment, and year for the growth, physiological parameters, and tissue nutrient content of soybean in early planting conditions.

		Plant														
Factors	Yield	stand/ac	PHT	LAI	CI	N	P	K	Ca	Mg	В	Zn	Mn	Cu	Fe	S
CC	0.449	0.091	0.156	0.013	0.438	0.031	0.172	0.544	0.007	0.001	0.204	0.406	< 0.001	0.318	0.489	0.781
Fert	< 0.001	0.430	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.374	0.010	0.003	< 0.001	< 0.001	0.172	0.335
Year	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.003	< 0.001	< 0.001	< 0.001	0.228	< 0.001	0.006	0.152
CC*Fert	0.003	0.771	0.020	0.108	0.022	0.204	0.325	0.907	0.104	0.822	0.992	0.622	0.538	0.038	0.693	0.636
CC*Year	0.729	0.076	0.429	0.215	0.048	0.482	0.016	0.016	0.847	0.959	0.651	0.004	0.691	0.005	0.434	0.320
Fert*Year	< 0.001	0.002	0.025	< 0.001	< 0.001	< 0.001	< 0.001	0.003	< 0.001	< 0.001	0.003	< 0.001	0.031	< 0.001	0.545	0.090
CC*Fert*Year	0.999	0.778	0.854	0.741	0.948	0.527	0.968	0.999	0.684	0.981	0.997	0.919	0.968	0.092	0.723	0.417

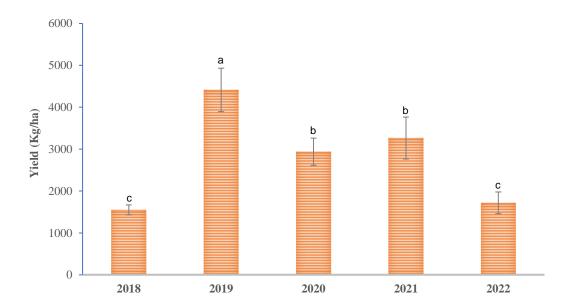
Note: CC, Cover Crop; Fert, Fertilizer; PHT, Plant Height; LAI, Leaf Area Index; CI, Chlorophyll Index; N, Nitrogen; P, Phosphorous; K, Potassium; Ca, Calcium; Mg, Magnesium; B, Boron; Zn, Zinc; Mn, Manganese; Cu, Copper; Fe, Iron; S-Sulfur. Bold values are significant at $\alpha = 0.05$; n=4.

Table 3: Analysis of variance (ANOVA), probability values (p-values) for the treatment effects and interactions between cover crop, fertilizer source treatment, and year for the growth, physiological parameters and tissue nutrient content of soybean in late planting conditions.

		Plant														
Factors	Yield	stand/ac	PHT	LAI	CI	N	P	K	Ca	Mg	В	Zn	Mn	Cu	Fe	S
CC	0.221	0.866	0.008	0.003	0.693	0.041	0.557	0.011	0.040	0.010	0.845	0.745	0.002	0.432	0.436	0.446
Fert	< 0.001	0.126	< 0.001	< 0.001	0.0014	< 0.001	< 0.001	< 0.001	< 0.001	0.044	0.007	0.883	< 0.001	0.085	0.496	0.003
Year	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.036	< 0.001	< 0.001	< 0.001	0.014	0.083	< 0.001	0.014
CC*Fert	0.885	0.788	0.117	0.406	0.436	0.336	0.647	0.409	0.761	0.373	0.586	0.012	< 0.001	0.380	0.622	0.053
CC*Year	0.986	0.420	< 0.001	0.049	0.197	0.347	0.453	0.492	0.299	0.133	0.057	0.939	0.024	0.525	0.941	0.838
Fert*Year	< 0.001	0.025	< 0.001	< 0.001	0.044	< 0.001	0.004	< 0.001	0.091	0.003	0.195	< 0.001	< 0.001	0.286	0.377	0.028
CC*Fert*Year	0.999	0.896	0.204	0.551	0.739	0.951	0.945	0.970	0.726	0.776	0.982	0.887	0.078	0.516	0.919	0.437

Note: CC, Cover Crop; Fert, Fertilizer; PHT, Plant Height; LAI, Leaf Area Index; CI, Chlorophyll Index; N, Nitrogen; P, Phosphorous; K, Potassium; Ca, Calcium; Mg, Magnesium; B, Boron; Zn, Zinc; Mn, Manganese; Cu, Copper; Fe, Iron; S-Sulfur. Bold values are significant at α = 0.05; n=4.

a.



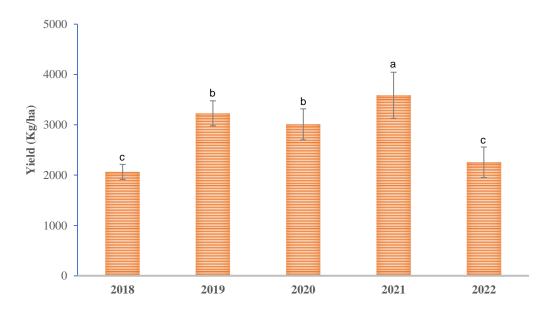
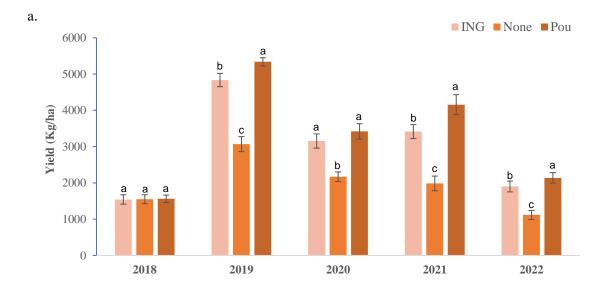


Figure 1: Average soybean yield among the years under different cover crop and fertilizer source treatments in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p<0.05.



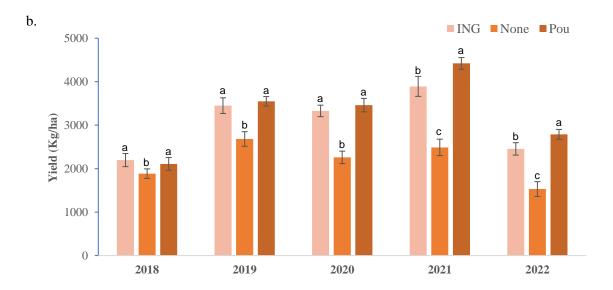
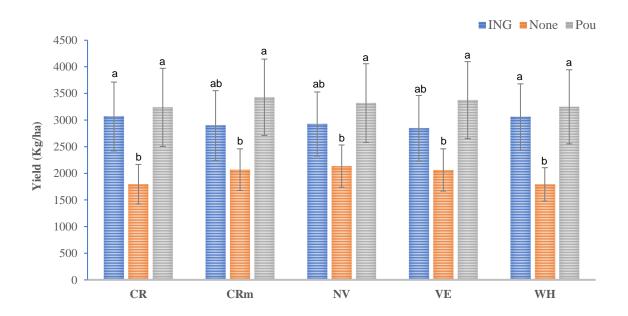


Figure 2: Effect of different fertilizer source treatment on soybean yield within the year in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p<0.05.

a.



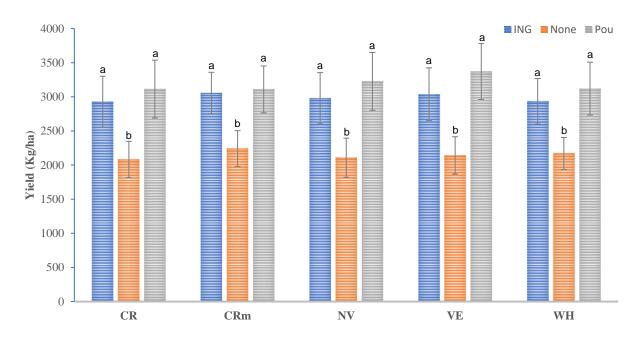
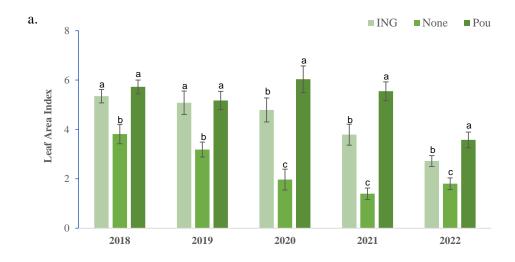


Figure 3: Soybean yield variation as affected by interaction of different cover crop and fertilizer source treatments in early (a) and late (b) planting conditions during the crop season from 2018-2022. Treatments with different letters are statistically significant at p<0.05. CR, Cereal Rye; CRM, Cereal Rye + Mustard; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING- Inorganic fertilizer; Pou- Poultry litter.



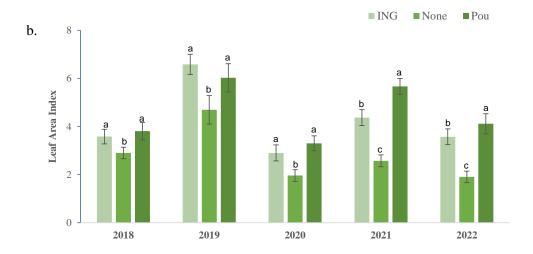
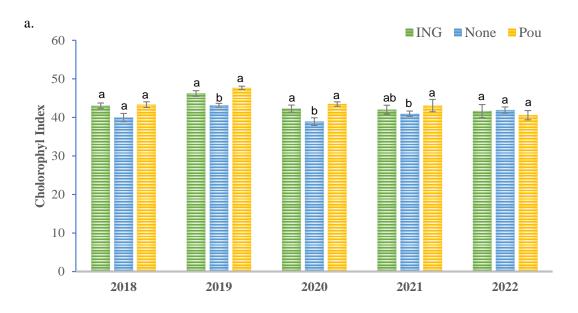


Figure 4: Soybean Leaf Area Index under different fertilizer source treatment during the cropping season within the year in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p<0.05.



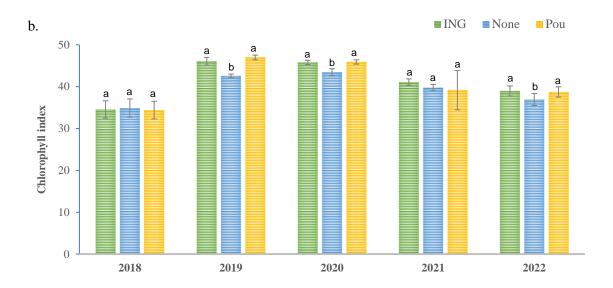
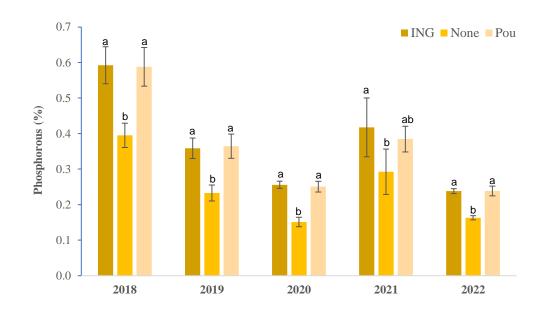


Figure 5: Soybean chlorophyll index under different fertilizer source treatment during the cropping season within the year in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p < 0.05.

a.



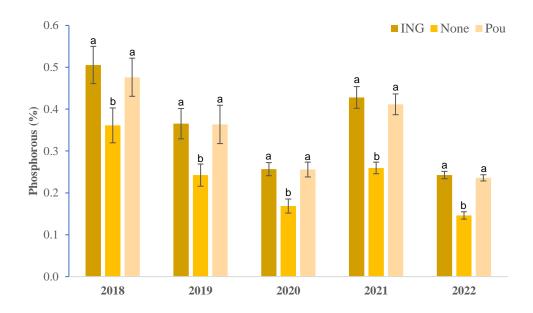


Figure 6: Soybean leaf tissue phosphorus content within the year differences under different fertilizer source treatment in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p<0.05.

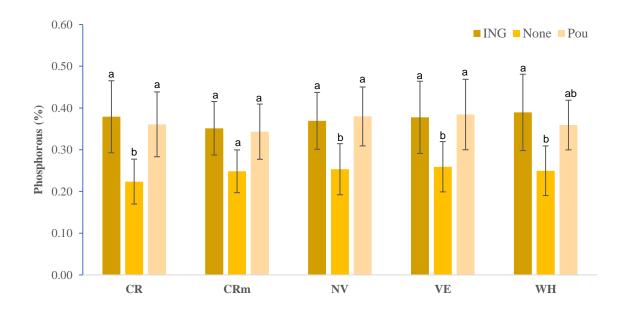
a.





Figure 7: Soybean leaf tissue potassium content within the year differences under different fertilizer source treatment in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p<0.05.

a.



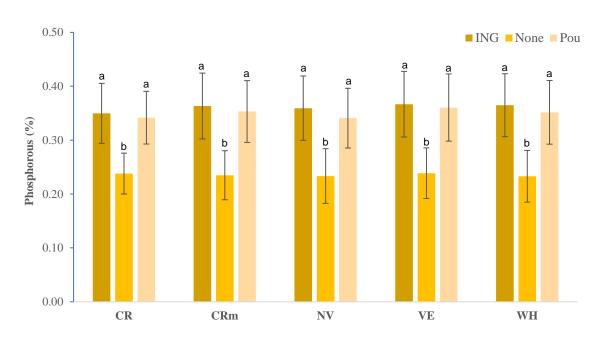


Figure 8: Soybean leaf tissue Phosphorous content for the interaction of different cover crop and fertilizer source treatments in early (a) and late planting (b) conditions. Treatments with different letters are statistically significant at p<0.05.

Table 4: Cover crop biomass (dry weight) in year, 2019, 2020, 2021 and 2022 under combination of cover crop and fertilizer source treatment in early planting conditions.

	Cover crop biomass (Kg/ha)							
Cover crops	2019	2020	2021	2022				
CR	3303.7 ± 543.52 a	1755.8 ± 469.82	835.5 ± 307.7 b	1456.7 ± 410.8 b				
CRm	3673.2 ± 490.07 a	1882.5 ± 447.38	1878.8 ± 781.72 ab	$1443.3 \pm 287.78 \text{ b}$				
NV	1276.2 ± 189.86 c	1435 ± 332.41	*	*				
VE	1987.8 ± 357.77 bc	1320.8 ± 278.69	$1145.2 \pm 441.05 \text{ b}$	$995 \pm 127 \text{ b}$				
WH	2950.5 ± 423.01 ab	1919.2 ± 302.61	2865.3 ± 637.58 ab	2596.7 ± 344.27 a				
P value	< 0.001	0.197	< 0.001	< 0.001				

Note: within a column, means and standard errors followed by the different letter are statistically significant (p<0.05). CR, Cereal Rye; CRM, Cereal Rye + Mustard; NV, Native Vegetation; VE, Vetch; WH, Wheat. "*", data not available.

Table 5: Cover crop biomass (dry weight) in year, 2019, 2020, 2021 and 2022 under combination of cover crop and fertilizer source treatment in late planting conditions.

	Cover crop biomass (Kg/ha)							
Cover crops	2019	2020	2021	2022				
CR	4221.7 ± 417.5 a	3466.7 ± 643.2 ab	2003.8 ± 482.7 b	2313.3 ± 602.1 b				
CRm	4222.5 ± 457.6 a	4187.9 ± 794.5 a	$3696.2 \pm 1116.1 \text{ b}$	$2575 \pm 768.4 \text{ b}$				
NV	$2678.3 \pm 478.1 \text{ b}$	$2736.8 \pm 480.2 \text{ b}$	*	*				
VE	$2934.2 \pm 631.8 \text{ b}$	$2707.2 \pm 596.2 \text{ b}$	3911.5 ± 985.7 ab	$2310 \pm 566.6 \text{ b}$				
WH	5154.2 ± 605.7 a	3533.6 ± 564.9 ab	5901.5 ± 1010.5 a	$3888.3 \pm 907.6 a$				
P value	< 0.001	0.027	< 0.001	0.03				

Note: within a column, means and standard errors followed by the different letter are statistically significant (p<0.05). CR, Cereal Rye; CRM, Cereal Rye + Mustard; NV, Native Vegetation; VE, Vetch; WH, Wheat. "*", data not available.

Table 6: Analysis of variance (ANOVA), probability values (p-values) for the treatment effects and interactions between cover crop, fertilizer source treatment, and year for the soil chemical properties in early planting conditions.

Factors	pН	P	K	Ca	Mg	Na	Zn	В	Mn	Fe	S
CC	0.251	< 0.001	0.011	0.032	< 0.001	0.620	0.827	0.252	0.004	0.033	0.055
Fert	< 0.001	< 0.001	< 0.001	0.473	< 0.001	0.025	< 0.001	0.301	0.792	0.039	0.001
Year	0.008	0.002	< 0.001	0.170	0.527	0.0011	< 0.001	< 0.001	0.548	0.0013	< 0.001
CC*Fert	0.437	0.491	0.062	0.007	< 0.001	0.336	0.120	0.099	0.995	0.289	0.589
CC*Year	0.987	0.321	0.941	0.994	0.995	0.496	0.818	0.534	0.033	< 0.001	0.778
Fert*Year	0.788	< 0.001	< 0.001	0.989	0.002	0.573	< 0.001	0.734	0.970	0.002	0.141
CC*Fert*Year	0.951	0.406	0.986	0.997	0.826	0.250	0.247	0.579	1.000	0.900	0.969

Note: P, Phosphorous; K, Potassium; Ca, Calcium; Mg, Magnesium; Na, Sodium; Zn, Zinc; B, Boron; Mn, Manganese; Fe, Iron; S, Sulfur. Bold values are significant at $\alpha = 0.05$.

Table 7: Analysis of variance (ANOVA), probability values (p-values) for the treatment effects and interactions between cover crop, fertilizer source treatment, and year for the soil chemical properties in late planting conditions.

Factors	pН	P	K	Ca	Mg	Na	Zn	В	Mn	Fe	S
CC	0.680	0.292	0.057	0.025	0.022	0.030	0.814	0.219	0.026	0.805	0.205
Fert	0.024	< 0.001	< 0.001	0.002	< 0.001	0.071	< 0.001	0.441	0.761	0.003	0.002
Year	0.001	< 0.001	0.002	< 0.001	0.027	0.029	0.001	< 0.001	0.795	< 0.001	< 0.001
CC*Fert	0.001	0.110	0.865	0.031	0.814	0.970	0.230	0.737	0.910	0.772	0.452
CC*Year	0.970	0.668	0.998	1.000	1.000	0.012	0.778	0.912	0.015	0.544	0.606
Fert*Year	0.944	0.001	0.001	0.987	0.205	0.902	< 0.001	0.818	0.997	0.002	0.202
CC*Fert*Year	0.972	0.900	1.000	0.980	0.999	0.998	0.443	0.940	1.000	0.998	0.219

Note: P, Phosphorous; K, Potassium; Ca, Calcium; Mg, Magnesium; Na, Sodium; Zn, Zinc; B, Boron; Mn, Manganese; Fe, Iron; S, Sulfur. Bold values are significant at $\alpha = 0.05$.

Table 8: Effect of different fertilizer source treatments on soil chemical properties in soybean early planting conditions under no-tillage in 2019, 2020, 2021 and 2022.

Fertilizer		P	K	Ca	Mg	Na	Zn	В	Mn	Fe	S
source	pН	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)
					Year_201	9					
ING	$5.75 \pm 0.2 \text{ a}$	14.23 ± 2.3 a	$148.33 \pm 8.6 a$	1356.35 ± 133.8 a	112.13 ± 10.3 a	*	$1.13 \pm 0.1 \text{ b}$	$1.43 \pm 0.2 \text{ a}$	$307.45 \pm 43.1 \text{ a}$	$82.95 \pm 4.2 \text{ a}$	$24.43 \pm 2.1 \text{ a}$
None	$5.88 \pm 0.2 \ a$	$10.43 \pm 1.8 \text{ b}$	152.75 ± 13.3 a	1305.68 ± 156.5 a	$104.33 \pm 14.9 a$	*	$1.4 \pm 0.2 \text{ b}$	$1.31 \pm 0.2 a$	$315.5 \pm 44 a$	$84.58 \pm 4.8 a$	$22.35 \pm 1.9 a$
Pou	$5.79 \pm 0.2 a$	$13.73 \pm 2.8 \text{ ab}$	$162.48 \pm 12.9 a$	$1299.65 \pm 101.8 a$	$111.4 \pm 9.2 a$	*	$1.83 \pm 0.3 \text{ a}$	$1.42 \pm 0.2 a$	$313.95 \pm 43.4 a$	$84.9 \pm 5.3 \text{ a}$	$23.9 \pm 1.8 \text{ a}$
					Year_202	0					
ING	$5.74 \pm 0.1 \text{ b}$	$7.39 \pm 1 \text{ a}$	$106.23 \pm 6.3 \text{ b}$	1121.28 ± 88.7 a	$92.8 \pm 6 \text{ a}$	$11.47 \pm 0.7 \text{ b}$	$0.64 \pm 0.1 \text{ b}$	$2.75 \pm 0.5 \text{ a}$	$278.69 \pm 40.6 a$	$83.13 \pm 5 \text{ a}$	*
None	$5.92 \pm 0.1 a$	$3.31 \pm 0.5 b$	$112.93 \pm 8.5 \text{ b}$	1106.11 ± 73.9 a	$90.96 \pm 8.2 a$	$10.98 \pm 0.6 \mathrm{b}$	$0.64 \pm 0.1 \text{ b}$	2.52 ± 0 a	$287.46 \pm 47.2 a$	$80.38 \pm 5 \text{ a}$	*
Pou	$5.87 \pm 0.1 \text{ ab}$	$7.86 \pm 2.2 a$	$127.02 \pm 9.5 a$	$1105.08 \pm 80.1 a$	$100.64 \pm 7 \text{ a}$	$12.67 \pm 0.5 a$	$1.23 \pm 0.2 a$	$2.54 \pm 0.1 a$	$280.1 \pm 42.2 \text{ a}$	$82.15 \pm 5.1 a$	*
					Year_202	1					
ING	$5.77 \pm 0.2 \text{ b}$	13.92 ± 3.3 a	124.64 ± 8.2 b	1246.45 ± 102.5 a	$101.84 \pm 8.8 \text{ b}$	$40.19 \pm 9.4 a$	$0.73 \pm 0.1 \text{ b}$	2.7 ± 0 a	365.91 ± 47.4 a	105.66 ± 6.3 a	18.06 ± 1.8 a
None	$6.02 \pm 0.1 a$	$2.05 \pm 0.5 b$	$129.08 \pm 10.7 \text{ b}$	$1203.72 \pm 89.5 a$	$101.09 \pm 9.3 b$	$35.96 \pm 9.4 a$	$0.61 \pm 0.1 \text{ b}$	$2.7 \pm 0 a$	$356.04 \pm 45.9 a$	$106.25 \pm 8.6 a$	$17.64 \pm 1.5 a$
Pou	$5.99 \pm 0.1 a$	$14.96 \pm 3.5 a$	$170.12 \pm 12.9 a$	1212.29 ± 81.9 a	$120.12 \pm 10 \text{ a}$	$43.29 \pm 8.8 \text{ a}$	$2.34 \pm 0.3 a$	$2.68 \pm 0 a$	$360.07 \pm 49.5 a$	101.34 ± 7.6 a	$17.87 \pm 1.7 a$
					Year_202	2					
ING	$5.56 \pm 0.1 \text{ b}$	16.55 ± 2.9 a	$92.63 \pm 7.5 \text{ b}$	1113.51 ± 97.3 a	$95.15 \pm 7.5 \text{ b}$	$10.86 \pm 0.6 \mathrm{b}$	$1.58 \pm 0.4 \text{ b}$	$2.63 \pm 0.1 \text{ a}$	305.25 ± 41.1 a	115.25 ± 4.8 a	$16.06 \pm 1.2 a$
None	$5.86 \pm 0.1 a$	$1.5 \pm 0.2 \text{ b}$	$102.8 \pm 10.8 \text{ b}$	1114.26 ± 87.7 a	$96.39 \pm 7.7 \text{ b}$	$10.31 \pm 0.4 \text{ b}$	$0.43 \pm 0.1 \text{ c}$	$2.61 \pm 0 a$	$324.5 \pm 48.4 \text{ a}$	$102.75 \pm 5.5 \text{ b}$	$11.93 \pm 1 \text{ b}$
Pou	$5.8 \pm 0.1 \text{ a}$	$11.84 \pm 4.8 \text{ a}$	$142.95 \pm 16.8 a$	1111.6 ± 78 a	$115.81 \pm 7.5 \text{ a}$	$13.37 \pm 0.8 \text{ a}$	$2.8 \pm 0.5 a$	$2.67 \pm 0 \text{ a}$	$320.93 \pm 45.1 a$	109.11 ± 5.4 ab	$15.09 \pm 1.3 a$

Note: within a column, means and standard errors followed by the different letter are statistically significant (p<0.05). P, Phosphorous; K, Potassium; Ca, Calcium; Mg, Phosphorous; Ca

Magnesium; Na, Sodium; Zn, Zinc; B, Boron; Mn, Manganese; Fe, Iron; S, Sulfur. "*", data not available.

Table 9: Effect of different fertilizer source treatments on soil chemical properties in soybean late planting conditions under no-tillage in 2019, 2020, 2021 and 2022.

Fertilizer		P	K	Ca	Mg	Na	Zn	В	Mn	Fe	S
source	pН	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)
					Year_2	019					
ING	$5.89 \pm 0.2 \text{ a}$	$17.73 \pm 2.2 \text{ a}$	126.53 ± 11.9 a	$1375.38 \pm 147 a$	$94.13 \pm 10.5 a$	*	$1.35 \pm 0.2 \text{ b}$	$1.46 \pm 0.2 \text{ a}$	$337.8 \pm 39.5 a$	$92.48 \pm 4.2 \text{ a}$	$26.8 \pm 4.6 a$
None	$6 \pm 0.2 \text{ a}$	$11.82 \pm 1.2 \text{ b}$	$129.42 \pm 9.4 a$	1399.71 ± 120.7 a	$90 \pm 6.7 \text{ a}$	*	$1.29 \pm 0.2 b$	$1.44 \pm 0.2 \text{ a}$	$339.97 \pm 37.7 a$	$90.95 \pm 3.6 a$	$22.61 \pm 1.9 a$
Pou	$5.89 \pm 0.2 a$	$15.68 \pm 1.8 a$	$133.3 \pm 14.8 \text{ a}$	1272.58 ± 110.5 a	$96.58 \pm 12.8 \text{ a}$	*	$1.75 \pm 0.2 \text{ a}$	$1.43 \pm 0.2 \text{ a}$	$334.7 \pm 37.2 \text{ a}$	$94.48 \pm 4.3 \text{ a}$	$25.18 \pm 2.7 \text{ a}$
					Year_2	020					
ING	$5.91 \pm 0.1 a$	$8.87 \pm 1.7 \text{ a}$	$96.54 \pm 6.5 \text{ b}$	1176.29 ± 79.3 a	$80.38 \pm 6.7 \text{ a}$	$10.92 \pm 0.4 \text{ b}$	$0.83 \pm 0.1 \text{ b}$	$2.6 \pm 0.1 \text{ a}$	$306.23 \pm 33.3 \text{ a}$	$94.02 \pm 5.3 \text{ a}$	*
None	$6 \pm 0.1 \text{ a}$	$3.49 \pm 0.5 \text{ b}$	104.09 ± 10.3 ab	1200.03 ± 82.5 a	$81.72 \pm 6.9 \text{ a}$	$10.54 \pm 0.4 \text{ b}$	$0.76 \pm 0.1 \text{ b}$	$2.59 \pm 0.1 a$	$309.3 \pm 34.4 a$	$87.4 \pm 4.6 a$	*
Pou	$5.89 \pm 0.1 a$	$8.23 \pm 1.5 \text{ a}$	116.95 ± 11.3 a	1110.85 ± 77.3 a	$92.39 \pm 9.8 a$	12.44 ± 0.6 a	$1.36 \pm 0.2 a$	$2.61 \pm 0 a$	$301.98 \pm 33 a$	$94 \pm 5.6 a$	*
					Year_2	021					
ING	$6.01 \pm 0.1 \text{ a}$	$13.52 \pm 3.4 \text{ a}$	107.49 ± 9 b	1244.23 ± 127.6 a	$87.36 \pm 9.2 \text{ b}$	$33.25 \pm 10 \text{ a}$	$0.71 \pm 0.1 \text{ b}$	2.72 ± 0 a	304.6 ± 39.6 a	$98.44 \pm 6.8 \text{ a}$	19.88 ± 1.3 a
None	$6.13 \pm 0.2 a$	$2.56 \pm 0.6 \ b$	$110.02 \pm 7.1 \text{ b}$	$1289.88 \pm 91.4 a$	$87.29 \pm 5.5 \text{ b}$	$34.79 \pm 9.4 a$	$0.74 \pm 0.2 \text{ b}$	$2.72 \pm 0 a$	$307.05 \pm 46.6 a$	$101.98 \pm 7.5 a$	$19.41 \pm 1.6 a$
Pou	$6.06 \pm 0.1 \text{ a}$	$12.76 \pm 2.5 \text{ a}$	$145.9 \pm 12.7 \text{ a}$	1199.13 ± 94.3 a	$107.97 \pm 9 a$	$35.82 \pm 10.6 a$	$2.34 \pm 0.3 \text{ a}$	$2.84 \pm 0.2 a$	$308.94 \pm 34.3 \text{ a}$	$98.79 \pm 5.1 \text{ a}$	$18.89 \pm 1.7 \text{ a}$
					Year_2	022					
ING	$5.74 \pm 0.1 \text{ b}$	$13.71 \pm 2.2 \text{ a}$	$79.91 \pm 7.5 \text{ b}$	1177.29 ± 104.4 ab	$81.31 \pm 8.8 \text{ b}$	$11.17 \pm 0.5 \text{ b}$	$1.64 \pm 0.5 \text{ b}$	$2.59 \pm 0 \text{ b}$	330.91 ± 37.4 a	$122.89 \pm 7.8 \text{ a}$	$15.14 \pm 0.9 a$
None	$5.95 \pm 0.1 a$	$3.08 \pm 3.5 c$	$82.31 \pm 10.9 \text{ b}$	1248.72 ± 85.5 a	$85.26 \pm 7.2 \text{ b}$	$10.66 \pm 0.3 \text{ b}$	$0.5 \pm 0.1 c$	2.61 ± 0 ab	$341.47 \pm 35.6 a$	$105.22 \pm 6.2 \text{ b}$	$11.31 \pm 0.6 \text{ b}$
Pou	$5.74 \pm 0.1 \text{ b}$	$8.59 \pm 2.3 \text{ b}$	119.6 ± 14.1 a	1095.8 ± 80.6 b	$104.99 \pm 9.2 a$	13.42 ± 0.7 a	2.52 ± 0.3 a	$2.66 \pm 0 \text{ a}$	325.31 ± 38.3 a	$120.04 \pm 7.9 a$	14.43 ± 1.1 a

Note: within a column, means and standard errors followed by the different letter are statistically significant (p<0.05). P, Phosphorous; K, Potassium; Ca, Calcium; Mg, Magnesium; Na, Sodium; Zn, Zinc; B, Boron; Mn, Manganese; Fe, Iron; S, Sulfur. "*", data not available.

Objective 2. Evaluate the effects of cover cropping systems on "soil health" (physical characteristics, microorganisms, soil moisture, etc.). Soil microbial communities are important indicators for understanding the effects of soil management practices and soil quality. To study the influence of different cover crop and fertilizer source treatment on soil microbial diversity and community composition, soil samples were collected after 20 days of cc termination in early and late planting systems, in 2019 and 2022. Soil genomic DNA was extracted from the collected soil samples and amplicon sequencing of bacterial and fungal genes were carried out using the 16SrRNA and ITS2 fragments, respectively. Extracted DNA was sent to Novogene (https://en.novogene.com/; Sacramento, CA, USA) and sequenced on Illumina HiSeq platform (250-bp paired-end reads). Sequence processing was done using the QIIME 2 (version 2022.11) (Quantitative Insights Into Microbial Ecology) software. Statistical analysis was performed using the MicrobiomeAnalyst (https://www.microbiomeanalyst.ca/) pipeline.

To determine the effect of treatments on microbial (bacterial and fungal) diversity, we analyzed and compared the microbial alpha and beta diversity indices across the years, 2019 and 2022. In the early planting system, significant differences were observed in both richness (Chao1) (p=0.001) and diversity (Shannon Diversity index) (p=0.02) between 2019 and 2022. Specifically, 2022 had higher richness and diversity than 2019 (Fig. 1a and b). Similar results were observed with the late system (Fig. 2a and b). We next studied microbial community structure in response to cc and fertilizer source treatment using Bray-Curtis dissimilarity metric, as this is a quantitative measure of the abundance of organisms presented in studied microbial populations. Bacterial community structure was different (p=0.001) and showed a distinct cluster for both years with early and late system conditions (Fig. 3a and b).

Given that the bacterial species diversity differed across the years, similar alpha and beta diversity measures were used to assess fungal species diversity. Fungal richness and diversity were different (p<0.001) between years and higher in 2022 with both early and late systems (Fig. 4a and b). Bray-Curtis analysis with PERMANOVA revealed that treatments shaped the fungal community composition. For each year, there was a distinct clustering of samples in both early and late planting conditions (Fig. 5a and b). These differences in the soil microbial communities suggest the influence of cc and fertilizer source treatments over the years. In addition, soil microbial communities in agricultural soils are temporally variable, spatially patterned and often correlated to soil moisture and temperature.

Further, with differences in bacterial and fungal diversity and community structure, we next studied the potential differences in microbial diversity within the year under cc and fertilizer source treatments. In early planting, bacterial species richness and diversity differed significantly under cc treatments in both years. Bacterial richness with CR was greater than WH in 2019 and CR and VE was greater than WH in 2022. In addition, NV was not different than CR or VE in both years. However, bacterial species richness and diversity increased for all cc treatments from 2019 to 2022. When comparing fertilizer sources within the year, bacterial species richness and

diversity was higher in poultry litter compared to ING fertilizer in 2019 and 2022 (Table 1). In late planting, similar differences were observed with cc treatments. However, no differences between fertilizer source were observed for either year (Table 2). The beta diversity of bacterial communities was significantly (p=0.041) shaped by fertilizer source treatment in early planting conditions in 2019 (Fig. 2). Notably, no differences in beta diversity of bacterial communities were observed in cc soil samples between fertilizer sources in 2019 or 2022. However, there was an increase in richness and diversity between years (over time), which was also observed with the early planting system.

The richness of soil fungal communities was affected by cc treatments in early and late planting system conditions (Table 3). In 2019 and 2022, WH exhibited the highest fungal richness compared to other cc treatments and different compared to NV with the early planting system. In late planting, WH was also highest, but only in 2022 (Table 4). The was no difference in fungal richness and diversity between fertilizer sources in either year. However, there was an increase in richness and diversity from 2019 to 2022 with cc and fertilizer source treatments. Suggesting an improvement in health overtime. In addition, fungal community structures were affected by cc treatments in early and late planting condition in both years (Fig. 7 a, b and 8a). In 2022, fungal communities were affected by fertilizer source treatments, only in early planting conditions (Fig. 8b).

Having shown that microbial communities are influenced by cc and fertilizer source treatments, to explain the phylum-level microbial community profiling, a combination of pattern correlation and heat map analyses was carried out to evaluate correlations between highly abundant bacterial or fungal phyla and treatments. In 2019, in the bacterial community profile, the phylum *Cyanobacteria* showed strong positive correlation with the (0.7; p<0.001) WH and *Bacteroidetes* showed positive correlation (0.40) with poultry litter treatment in early planting system conditions (Fig. 9a and b). However, with late planting there was not a strong correlation between bacterial phyla and treatments. These bacterial phyla play multiple roles in nutrient cycles (N-fixation) (Priya *et al.*, 2015) and organic matter degradation in soil (Fierer *et al.*, 2007), respectively, thus helps in soil productivity. Additionally, *Cyanobacteria* acts as a C source provider in the dryland soil. In 2022, we observed strong positive correlation of phyla *Spirochaetes* (0.8; p<0.001) and *Bacteroidetes* (0.78; p<0.001) with WH and *Crenarchaeota* (0.55; p=0.006) with poultry litter treatment (Fig. 15 a and b) in early planting conditions.

For the fungi, a combination of pattern correlation and heat map analysis showed that there was a positive correlation with fungal phyla *Mucoromycota* (0.52; p=0.01) and *Mortierellomycota* (0.48; p=0.018). This result is complimented by the heatmap, showing highest presence of these phyla with WH and VE in early planting system conditions (Fig. 11a). The Phylum *Ascomycota* showed positive correlation (0.44; p=0.03) and higher abundance in poultry litter treatment (Fig. 11b) in 2022. *Ascomycota* is dominant in dry land soils (Maestre et al., 2015) and these results suggest that fungal communities are strongly affected by cc and fertilizer source treatments.

We then studied the abundance of bacteria and fungi in the samples at the genus level, it revealed the presence of nitrogen fixing bacterial genera in both early and late planting conditions. We observed both nonsymbiotic and symbiotic nitrogen fixing bacteria such as, Rhizobium, Bradyrhizobium, Mesorhizobium, Azospirillum, Nitrospira (Wagner, 2011) in the study. In 2019, abundance of genera Bradyrhizobium and Nitrospira were higher with CR and WH, respectively compared to other cover crops in late planting conditions (Fig. 12a and b). The members of Azospirillum species are associated with cereal grasses (Steenhoudt and Vanderleyden, 2000) and higher abundance was observed with WH compared to other cc treatments (Fig 12c). In 2022, poultry litter treatment showed a higher abundance of Nitrospira compared to ING fertilizer in early planting system conditions (Fig. 17e). The members of Rhizobium associated with leguminous plants to ensure the nodule formation (Zahran, 1999). We observed an increased abundance of Mesorhizobium with VE, Rhizobium and Bradyrhizobium with WH, followed by VE (Fig 12d, f, g, and h) in early and late planting system conditions. We also noticed that the NV treatment possessed a lower abundance of nitrogen-fixing bacteria compared to cc treatments. These networks of nitrogen-fixing genera promote soybean growth and development, thus helps in crop productivity.

For the fungi, in 2019, the genera *Rhizopus* and *Aspergillus* were found to be significantly different in relative abundance with late planting system contitions, where there was higher abundance with WH compared to other cover crops (Fig. 13a and b). Regarding fertilizer source treatments, *Trichoderma* showed a higher relative abundance with ING fertilizer compared to poultry litter (Fig. 13c). Members in the genera, *Rhizopus*, *Aspergillus* and *Trichoderma* are plant growth promoting fungi that produce plant growth hormones. These play role in mineralization of major and minor nutrients required to reinforce plant growth and productivity (Karunarathna et al., 2021).

Conclusions:

- Our results demonstrate that cover crop and poultry litter amendment treatments have a positive influence on the richness, diversity, and composition of the soil microbial community. As, microbial richness and diversity were found to be higher in 2022 than 2019.
- In both the years, cover crop and poultry litter treatments resulted in the highest change in bacterial richness and diversity in early planting conditions. However, only the cover crop treatment promoted the change in fungal community richness.
- Fertilizer treatments recorded significant impact on the bacterial community structure in early planting. In 2022, both cover crops and fertilizer source influenced the fungal community structure in early planting conditions.

- In 2019 and 2022, cover crops, wheat and poultry litter promoted a greater abundance of beneficial bacterial phyla in early and planting conditions.
- Vetch and wheat cover crops exhibited the highest abundance of N- fixing bacterial genera, while native vegetation showed the lowest abundance.
- The use of cover crop and poultry litter amendment promoted a beneficial soil microbiome, which is essential for maintaining soil health and encourages soybean growth and productivity. In addition, this study provides evidence that the soil microbial communities differ temporally and spatially in the given environment.

Objective 2 Graphics and Tables

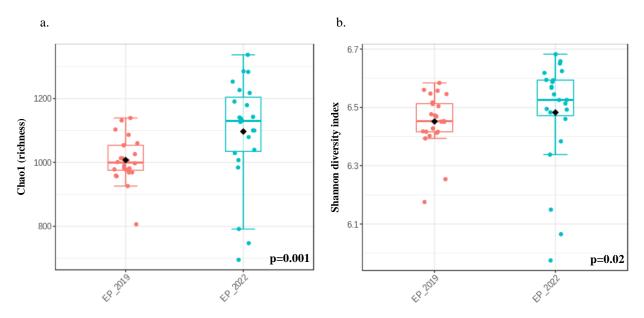


Figure 1: Alpha diversity indices, Chao1(a) and Shannon diversity index (b) of bacterial communities in soils sampled in the year 2019 and 2022 under different cover crop and fertilizer source treatments in early planting (EP) conditions.

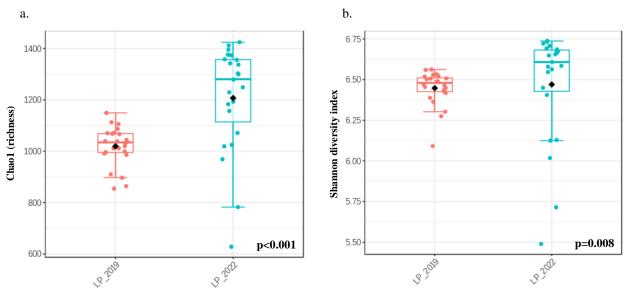


Figure 2: Alpha diversity indices, Chao1(a) and Shannon diversity index (b) of bacterial communities in soils sampled in the year 2019 and 2022 under different cover crop and fertilizer source treatments in late planting (LP) conditions.

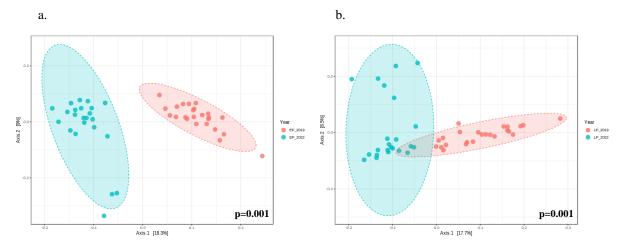


Figure 3: Bacterial beta diversity across the years (2019 and 2022) under no tillage, different cover crop and fertilizer source treatments in early (a) and late (b) planting conditions. Beta diversity was derived using Principal coordinate analysis (PCoA)-Bray Curtis dissimilarity matrix. EP, Early Planting; LP, Late Planting.

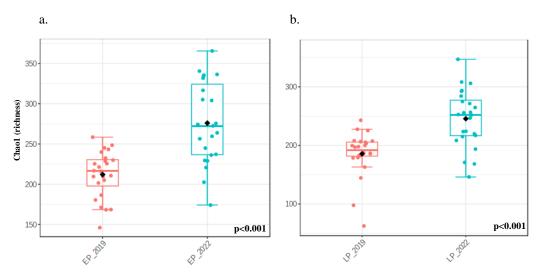


Figure 4: Alpha diversity index, Chao1(richness) of fungal communities in soils sampled in the year 2019 and 2022 under different cover crop and fertilizer source treatments in early (a) and late (b) planting conditions.

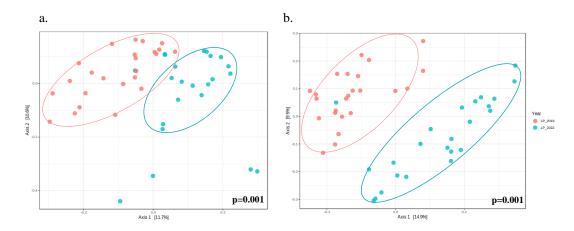


Figure 5: Fungal beta diversity across the years (2019 and 2022) under no tillage, different cover crop and fertilizer source treatments in early (a) and late (b) planting conditions. Beta diversity was derived using Principal coordinate analysis (PCoA)-Bray Curtis dissimilarity matrix. EP, Early Planting; LP, Late Planting.

Table 1: Soil bacterial species richness and diversity in early planting conditions under different cover crop and fertilizer source treatments in 2019 and 2022.

	Year-2019		Year-20	022
	Early planting		Early pla	nting
Cover crops	Richness	Diversity	Richness	Diversity
-	(ChaO1)	(Shannon)	(ChaO1)	(Shannon)
CR	1057.67 ± 29.79 a	6.53 ± 0.03 a	1204.18 ± 43.98 a	6.6 ± 0.04 a
NV	1053.42 ± 21.05 a	6.53 ± 0.03 a	1275.71 ± 23.26 a	6.61 ± 0.02 a
VE	997.76 ± 17.87 ab	6.47 ± 0.02 ab	1261.88 ± 52.38 a	6.65 ± 0.03 a
WH	$966.33 \pm 23.79 \text{ b}$	$6.41 \pm 0.02 \text{ b}$	$970.21 \pm 87.89 \text{ b}$	$6.32 \pm 0.1 \text{ b}$
p-value	0.032	0.008	< 0.05	< 0.05
Fertilizer source				
ING	1005.93 ± 4.48 b	$4.73 \pm 0.02 \text{ b}$	1155.39 ± 48.4	6.56 ± 0.05
Pou	$1031.65 \pm 2.7 \text{ a}$	$4.8 \pm 0.01 \ a$	1216.03 ± 52.04	6.56 ± 0.05
p-value	0.033	0.004	ns	ns

Note: within a column, means and standard errors followed by the different letter are statistically significant. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING- Inorganic fertilizer; Pou- Poultry litter.

Table 2: Soil bacterial species richness and diversity in late planting conditions under different cover crop and fertilizer source treatments in year, 2019.

	Year-2019	
	Late planting	
Cover crops	Richness	Diversity
•	(ChaO1)	(Shannon)
CR	1061.01 ± 14.2 a	6.5 ± 0
NV	1022.92 ± 13.2 a	6.49 ± 0
VE	$1024.94 \pm 20.5 a$	6.43 ± 0.1
WH	$928 \pm 22.7 \text{ b}$	6.39 ± 0
p-value	0.001	ns
Fertilizer source		
ING	1001.2 ± 17.73	6.46 ± 0.02
Pou	1017.24 ± 20.33	6.45 ± 0.04
p-value	ns	ns

Note: within a column, means and standard errors followed by the different letter are statistically significant. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING- Inorganic fertilizer; Pou- Poultry litter.

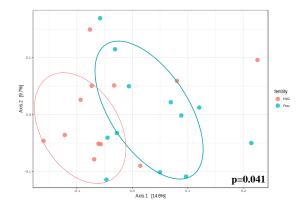


Figure 6: Principal coordinate analysis (PCoA) plot of Bray Curtis distances of soil bacterial community structures under different fertilizer source treatments in early planting conditions, in year 2019. ING, Inorganic fertilizer; Pou, poultry litter.

Table 3: Soil fungal species richness and diversity in early planting conditions under different cover crop and fertilizer source treatments in 2019 and 2022.

	Year-2019	Year-2022
	Early planting	Early planting
Cover crops	Richness	Richness
	(ChaO1)	(ChaO1)
CR	211.92 ± 8.94 ab	318.39 ± 22.16 ab
NV	$183.66 \pm 13.09 \text{ b}$	241.05 ± 17.63 c
VE	215.47 ± 9.39 ab	257.38 ± 9.09 bc
WH	$236.3 \pm 7.71 a$	357.15 ± 16.92 a
p-value	0.012	< 0.001
Fertilizer source		
ING	220.53 ± 7.34	303.57 ± 18.64
Pou	203.14 ± 9.32	278.95 ± 16.59
p-value	ns	ns

Note: within a column, means and standard errors followed by the different letter are statistically significant. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING- Inorganic fertilizer; Pou- Poultry litter.

Table 4: Soil fungal species richness and diversity in late planting conditions under different cover crop and fertilizer source treatments in 2022.

	Year-2022		
	Late planting		
Cover crops	Richness	Diversity	
	(ChaO1)	(Shannon)	
CR	276.64 ± 16.67 ab	$3.61 \pm 0.11 \text{ b}$	
NV	$222.54 \pm 13.85 \text{ b}$	$3.33 \pm 0.16 \mathrm{b}$	
VE	$281.93 \pm 10.25 \text{ a}$	$3.76 \pm 0.16 \text{ ab}$	
WH	$326.85 \pm 15.2 \text{ a}$	4.17 ± 0.09 a	
p-value	< 0.001	0.02	
Fertilizer source			
ING	277.97 ± 13.71	3.74 ± 0.12	
Pou	276 ± 15.62	3.69 ± 0.14	
p-value	ns	ns	

Note: within a column, means and standard errors followed by the different letter are statistically significant. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING- Inorganic fertilizer; Pou- Poultry litter.

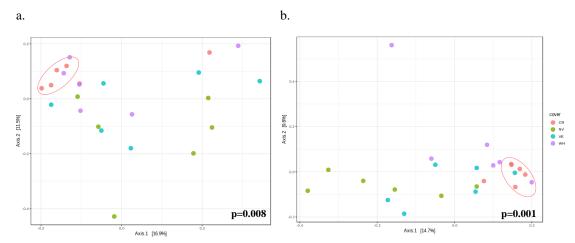


Figure 7: Principal coordinate analysis (PCoA) plots of Bray Curtis distances of soil fungal community structures under different cover crop treatments in early (a) and late (b) planting conditions, in year 2019. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat.

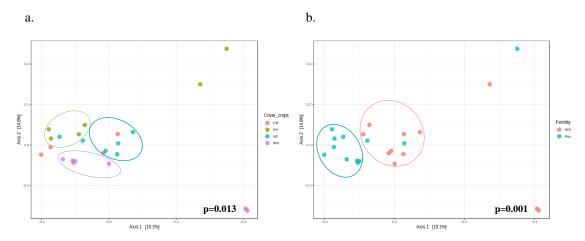


Figure 8: Principal coordinate analysis (PCoA) plots of Bray Curtis distances of soil fungal community structures under different cover crop (a) and fertilizer source (b) treatments in early planting conditions, in year 2022. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING, Inorganic fertilizer; Pou, Poultry litter.

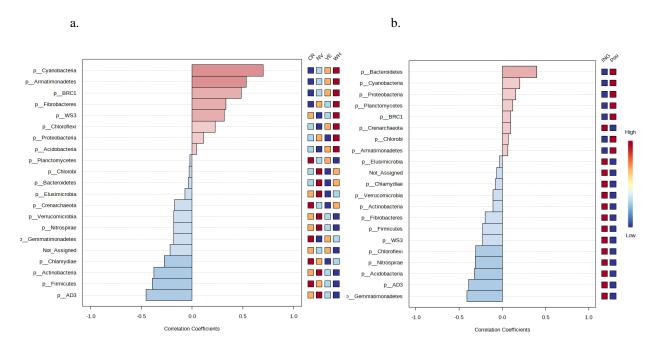


Figure 9: Pattern correlation and heat map analysis of bacterial phyla under different cover crop (a) and fertilizer source (b) treatments in 2019 in early planting conditions. Bars indicate the correlation coefficients of the different phyla which are ranked by correlation and to the right, heatmap showing levels of abundance (red-higher; blue-lower).

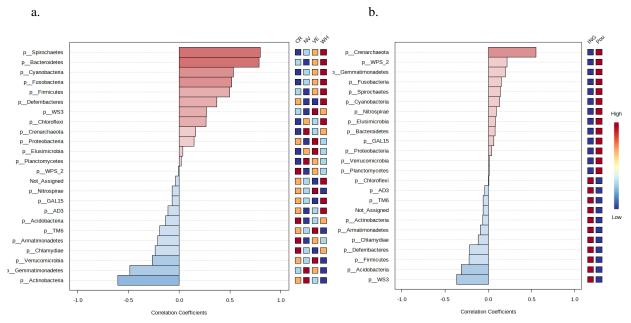


Figure 10: Pattern correlation and heat map analysis of bacterial phyla under different cover crop (a) and fertilizer source (b) treatments in 2022 in early planting conditions. Bars indicate the correlation coefficients of the different phyla which are ranked by correlation and to the right, heatmap showing levels of abundance (red-higher; blue-lower).

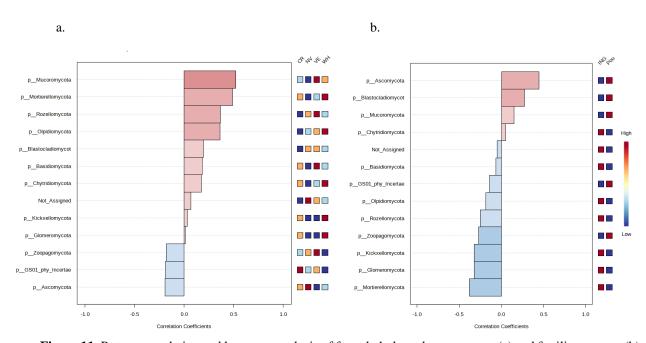


Figure 11: Pattern correlation and heat map analysis of fungal phyla under cover crop (a) and fertilizer source (b) treatment in early planting in the year,2022. Bars indicate the correlation coefficients of the different phyla which are ranked by correlation and to the right, heatmap showing levels of abundance (red-higher; blue-lower). CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING, Inorganic fertilizer; Pou, Poultry litter.

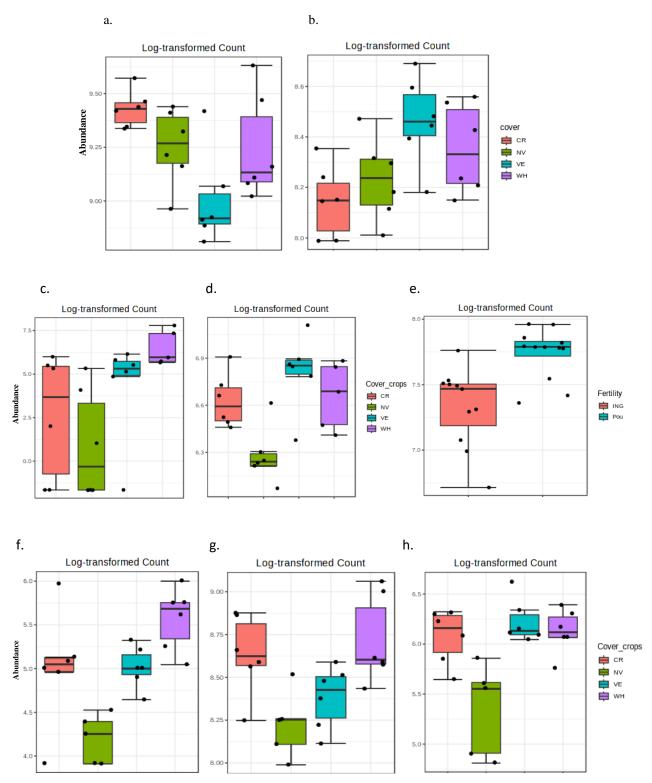


Figure 12: Nitrogen fixing bacteria abundance under different cover crops and fertilizer source treatments across the years. Abundance of genera *Bradyrhizobium* (a) and *Nitrospira* (b) in late planting in year, 2019; Abundance of genera *Azospirulum* (c), *Mesorhizobium* (d), *Nitrospira* (e) in early and *Rhizobium* (f), *Bradyrhizobium* (g) and *Mesorhizobium* (h) in late planting conditions in the year, 2022. Significance at p<0.05. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING, Inorganic fertilizer; Pou, Poultry litter.

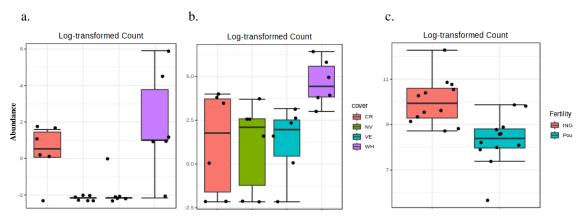


Figure 13: Plant growth promoting fungal genera under different cover crop and fertilizer source treatments in late planting conditions. Abundance of genera *Rhizopus* (a) and *Aspergillus* (b) in year 2019, p<0.05; abundance of genus *Trichoderma* (c) in year 2022, p<0.001. CR, Cereal Rye; NV, Native Vegetation; VE, Vetch; WH, Wheat; ING, Inorganic fertilizer; Pou, Poultry litter.

Objective 3. Determine the economic benefit of cover crops, source of fertilizer, and planting date on soybean production. A partial budget analysis was used to determine the net economic benefit of adopting cc and fertilizer practices. In a partial budget analysis, only the changes from varying practices are compared to a baseline control. Changes can be classified as positive (additional revenue; reduced expenses) or negative (reduced revenue; additional expenses) (Olson, 2010). This analysis focuses solely on the costs and benefits of adopting cc establishment and using poultry litter instead of ING fertilizer. In this study, additional expenses include cc establishment and fertilizer source. The assumed per acre cost of cc seed is based on prices collected from local vendors (Table 1). An additional expense of \$18.25 per acre is assumed to account for the additional machinery cost of drilling in the cc obtained from the 2023 Mississippi State University Soybean Planning Budget (Gregory, 2023). ING fertilizer cost is calculated using the average price from 2019-2023 of N, P, and K published in the Mississippi State University Soybean Planning Budgets (Gregory, 2019, 2020, 2021, 2022, 2023). Poultry litter cost was provided in private conversations with a large poultry company and farms located throughout the Mid-South. This company provided survey data on litter prices and hauling costs in central Mississippi. Based on this information, litter cost is assumed to be \$28.00 per ton plus \$21.00 per ton to haul 100 miles. Assuming an application rate of 2 tons per acre, the total litter cost is \$98 per acre. Net returns are calculated for each combination of cc species and fertilizer source for early and late system establishment dates. Net returns are calculated based on three different price scenarios. These prices represent the minimum (\$8.36/bu), average (\$10.72/bu), and maximum (\$14.50/bu) Mississippi season average price from 2007-2022 reported by the National Agricultural Statistics Service.

Net returns by treatment for the early planting system can be found in Table 2. These results have two main takeaways: CC adoption decreases profitability, and fertilizer practices increase profitability, with poultry litter being the preferred choice. CC adoption reduces profitability due to no statistically significant yield benefit but additional incurred expenses. The lowest cost of adoption was with WH at \$50.17 per acre. Both ING fertilizer and poultry litter results show increased profitability, as the yield benefits outweigh the additional expense of fertilization. Across all soybean prices considered, ING fertilizer and poultry litter showed positive net returns. Results also indicate that poultry litter provides higher net returns than ING fertilizer due to a higher yield benefit. Results for the late planting system are reported in Table 3, leading to the same conclusions as those of the early planting system.

While the partial budgeting results show that poultry litter outperforms ING fertilizer in this study, a key concern is an ability to source poultry litter. Poultry litter can be expensive to transport and is located considerably far away. Table 4 and 5 report the maximum sourcing distance (in miles) for poultry litter to be more profitable than ING fertilizer based on litter cost and transportation cost. These results assume an application rate of 2 tons per acre and a soybean price of \$10.72 per bushel. As the cost of litter and the mileage rate increase, the distance from which litter can profitably be sourced decreases. For example, for the early planting system, if litter costs \$28.00 per ton and transportation costs \$0.23 per mile, then a producer could source litter from a maximum distance of 201 miles. If the cost of litter increased to \$32.00 per ton, the sourcing distance would decrease to 192 miles. The sourcing distance decreases for the late planting date due to lower expected yields.

Objective 3 Graphics and Tables

Table 1. Cover Crop Seed and Fertilizer Cost per Acre

Item	Cost/acre
Cereal Rye	\$28.70
Cereal Rye/Mustard	\$82.25
Vetch	\$46.60
Wheat	\$31.92
Inorganic fertilizer (ING)	\$90.44
Poultry Litter	\$98.00

Table 2. Early Planting Net Returns of Cover Crop and Fertilizer Source Treatments by Market Price

Treatmen	nt	Market Price (\$/bu)				
Cover Crop	Fertilizer	\$8.36	\$10.72	\$14.50		
Cereal Rye	ING ¹	-\$14.45	\$20.26	\$75.85		
Cereal Rye/Mustard	ING	-\$68.00	-\$33.29	\$22.30		
Vetch	ING	-\$32.35	\$2.36	\$57.95		
Wheat	ING	-\$17.67	\$17.04	\$72.63		
None	ING	\$32.50	\$67.21	\$122.80		
Cereal Rye	Litter ²	\$23.09	\$70.52	\$146.50		
Cereal Rye/Mustard	Litter	-\$30.46	\$16.97	\$92.95		
Vetch	Litter	\$5.19	\$52.62	\$128.60		
Wheat	Litter	\$19.87	\$67.30	\$143.28		
None	Litter	\$70.04	\$117.47	\$193.45		

¹ Inorganic fertilizer (ING) ² Poultry litter (Litter)

Table 3. Late Planting Net Returns of Treatments by Market Price

Treatmer	<u>ıt</u>	Market Price (\$/bu)			
Cover Crop	Fertilizer	\$8.36	\$10.72	\$14.50	
Cereal Rye	ING ¹	-\$32.88	-\$3.38	\$43.88	
Cereal Rye/Mustard	ING	-\$86.43	-\$56.93	-\$9.67	
Vetch	ING	-\$50.78	-\$21.28	\$25.98	
Wheat	ING	-\$36.10	-\$6.60	\$40.66	
None	ING	\$14.07	\$43.57	\$90.83	
Cereal Rye	Litter ²	-\$15.03	\$21.65	\$80.39	
Cereal Rye/Mustard	Litter	-\$68.58	-\$31.90	\$26.84	
Vetch	Litter	-\$32.93	\$3.75	\$62.49	
Wheat	Litter	-\$18.25	\$18.43	\$77.17	
None	Litter	\$31.92	\$68.60	\$127.34	

¹ Inorganic fertilizer (ING) ² Poultry litter (Litter)

Table 4. Maximum Sourcing Distance (miles) for Poultry Litter to Produce Higher Net Returns than Inorganic Fertilizer – Early Planting

morganic	o i citiiizei	Burry Francing						
			Dollars per Ton					
		\$22	\$24	\$26	\$28	\$30	\$32	\$34
	\$0.11	474	456	438	419	401	383	365
Dollars per Mile	\$0.14	372	358	344	330	315	301	287
	\$0.17	307	295	283	271	260	248	236
	\$0.20	261	251	241	231	221	211	201
	\$0.23	227	218	209	201	192	183	174
	\$0.26	201	193	185	177	170	162	154
	\$0.29	180	173	166	159	152	145	138
	\$0.32	163	157	150	144	138	132	125
	\$0.35	149	143	138	132	126	120	115

Table 5. Maximum Sourcing Distance (miles) for Poultry Litter to Produce Higher Net Returns than Inorganic Fertilizer – Late Planting

		Dollars per Ton						
		\$22	\$24	\$26	\$28	\$30	\$32	\$34
	\$0.11	359	341	323	305	286	268	250
\$0.14 \$0.17 \$0.20 \$0.23 \$0.26 \$0.29 \$0.32 \$0.35	\$0.14	282	268	254	239	225	211	197
	\$0.17	232	221	209	197	185	174	162
	\$0.20	198	188	178	168	158	148	138
	\$0.23	172	163	154	146	137	128	120
	\$0.26	152	144	137	129	121	114	106
	\$0.29	136	129	122	116	109	102	95
	\$0.32	123	117	111	105	98	92	86
	\$0.35	113	107	101	96	90	84	79

Impacts and Benefits to Mississippi Soybean Producers

In 2020, there was an estimated 2 million acres of soybeans were planted in Mississippi. There is approximately 56% of the acres planted to in a dryland environment on any given year. Therefore, dryland soybeans can make-up more than half of the soybean acres in Mississippi. This is over 1 million acres of dryland soybeans in Mississippi that are prone to inconstant yields, due to the lack of adequate moisture during critical stages during the growing season. Dryland soybean yields were more than 15 bu/ac lower than for irrigated soybeans in 2012. Identifying an affordable cover cropping system that will minimize inconsistent dryland soybean yield due to lack of timely rainfall events could provide an additional \$18 million across the state of Mississippi in dryland soybean production based on a 5% increase in yield.

Soil management system certainly have an influence on the soil health by contributing to soil quality. The combination of cover crops and poultry litter amendment in the dryland environment system enriched the bacterial and fungal diversity over the years, as 2022 showed higher microbial richness and abundance than 2019. Also, a no-tillage system creates a conducive environment for soil fungal communities. The adoption of cover crops, such as vetch (legume) and wheat (cereal grass) could enable in enhancing the beneficial microbial communities, which play vital role in organic matter degradation, more carbon, macro and micronutrients cycling involving in soybean plant growth and development. In contrast, native vegetation (no cover crop) treatments shifted towards lower abundance of advantageous microbial communities. These results confirm that the direct linkage between soil management and microbial community structure. In addition, microbial communities are crucial for improving soil properties and structure, thus promoting soil health. These findings could drive decision making and provide direct benefits to soybean growers by identifying the influence of cover crops and poultry litter amendment associated with beneficial microbial communities in the soybean production system. Thus, assist in designing management systems which can support higher yields, while improving the soil quality and health.

End Products-Completed or Forthcoming

Publications:

- ➤ Kodadinne Narayana, N., Kingery, W. L., Shankle, M. W., and Ganapathi Shanmugam, S. (2022). Differential response of soil microbial diversity and community composition influenced by cover crops and fertilizer treatments in a dryland soybean production system. *Agronomy*, 12(3), 618.
- ➤ Pokhrel, S., Kingery, W. L., Cox, M. S., Shankle, M. W., & Shanmugam, S. G. (2021). Impact of cover crops and poultry litter on selected soil properties and yield in dryland soybean production. *Agronomy*, 11(1), 119.

Presentation at conferences:

➤ Poster presentation at Water Resources conference, March 28-30, Starkville, MS. Kovvuri N.R., Feng G., Bi G., Tewolde H., Shankle M. (2023). Cover cropping and poultry litter improve soil physical and hydraulic properties in dryland conditions.

- ➤ Poster presentation at the 4th Annual Mississippi Academy of Sciences (MAS) summer science and engineering symposium, June 8, Starkville, MS. Kodadinne Narayana, N., Kingery, W. L., Shankle, M., and Shanmugam, S. G. (2022). Influence of cover crops and fertilizer treatments on the soil microbial community dynamics in a dryland soybean production system.
- ➤ Poster presentation at the 4th Annual Mississippi Academy of Sciences Summer Science Engineering Research Symposium, June 8, Starkville, MS. Kovvuri, R.N., G. Feng, G. Bi, M. Shankle, and H. Tewolde. (2022). Influence of organic amendments and integration of cover crops on dryland soybean production.
- ➤ Poster presentation at Fall Graduate Research Symposium. Mississippi State University, Oct. 2, Starkville, MS. Kovvuri, R.N., G. Feng, G. Bi, M. Shankle, and H. Tewolde (2022). Effects of integrating cover crop and poultry litter on dryland soybean production and soil hydraulic properties.
- ➤ Poster presentation at the ASA-SSSA-CSSA Annual Meeting, Nov. 5-8 Baltimore, Maryland. Kovvuri, R.N., G. Feng, G. Bi, H. Tewolde, and M. Shankle. (2022). Influence of poultry litter and integration of cover crops on dryland soybean production.
- Results will be written as manuscript/s for publication in peer-reviewed journal and in a MAFES bulletin for use outside of the scientific community (this is the most important priority). The manuscript will describe the long term (5 years) impacts of conservation agriculture practices, such as cover crops and organic amendments (poultry litter) in enhancing the dryland soybean growth, development, and productivity under no-tillage conditions. Further, how these soil management practices improve soil properties and the beneficial soil microbial communities which aids in organic matter decomposition, carbon and nitrogen fixation, thus promotes the soil health.
- ➤ Progress gained by the project will be used to write a project proposal to accomplish the research gap and study further.
- ➤ Results will be presented in local production meetings and regional/national scientific meetings.