

**Results and discussion**

We will present the results for and discuss the soil fertility gradient that we found across farms, the impact of soil quality on cover crops, resulting cover crop quality and differences we found between treatments across farms, N mineralization rates over time, and resulting corn production. We have only had a few weeks to conduct an initial analysis of the complete dataset. We will continue to analyze the data in new ways as we produce presentations and publications from this study.

**Soil Fertility Gradient:** At the beginning of the project, we collected soil samples and analyzed them for soil fertility properties including soil texture, micro and macro nutrients, and different fractions of soil organic matter. We found a gradient of soil fertility levels, driven by different soil metrics, indicating that we achieved our goal of conducting this cover crop experiment across a range of baseline soil fertility. Tables 1-5 show all baseline soil data across the seven fields that completed the study (F1, F2, F5, F6, F7). Here we have included the mean, standard error, and range for each soil property.

**Table 1:** Baseline means, standard error (SE), and range for bulk density and soil texture for soils collected across all farms in the study.

	<b>Bulk Density</b>	<b>% Sand</b>	<b>% Clay</b>	<b>% Silt</b>
<b>Mean</b>	1.56	57.47	21.00	21.52
<b>SE</b>	0.014	2.751	1.830	1.208
<b>Range</b>	0.37	56.03	38.92	27.15

**Table 2:** Baseline means, standard error (SE), and range for pH, organic matter (%), phosphorus (P), potassium (K), magnesium (Mg), calcium (Ca), cation exchange capacity (CEC), N and C for soils collected across all farms in the study.

	<b>pH</b>	<b>Organic Matter (%)</b>	<b>P (ppm)</b>	<b>K (ppm)</b>	<b>Mg (ppm)</b>	<b>Ca (ppm)</b>	<b>CE C</b>	<b>%N</b>	<b>%C</b>
<b>Mean</b>	6.66	2.61	70.79	111.54	175.00	1344.64	9.73	0.14	1.52
<b>SE</b>	0.09	0.143	7.06	7.85	7.95	59.26	0.51	0.008	0.079
<b>Range</b>	2.00	3.30	165.00	197.00	190.00	1350.00	11.8	0.18	1.73

**Table 3:** Baseline means, standard error (SE), and range for the amount, N, and C of free particulate organic matter (FPOM) for soils collected across all farms in the study.

	<b>g FPOM/kg soil</b>	<b>kg FPOM/ha</b>	<b>% N</b>	<b>% C</b>	<b>mg N FPOM/kg soil</b>	<b>mg C FPOM/kg soil</b>
<b>Mean</b>	2.48	7653.72	1.50	27.28	37.36	669.29
<b>SE</b>	0.139	419.512	0.028	0.391	2.246	36.250
<b>Range</b>	3.75	11135.78	0.86	12.14	53.97	817.43

**Table 4:** Baseline means, standard error (SE), and range for the amount, N, and C of occluded particulate organic matter (OPOM) for soils collected across all farms in the study.

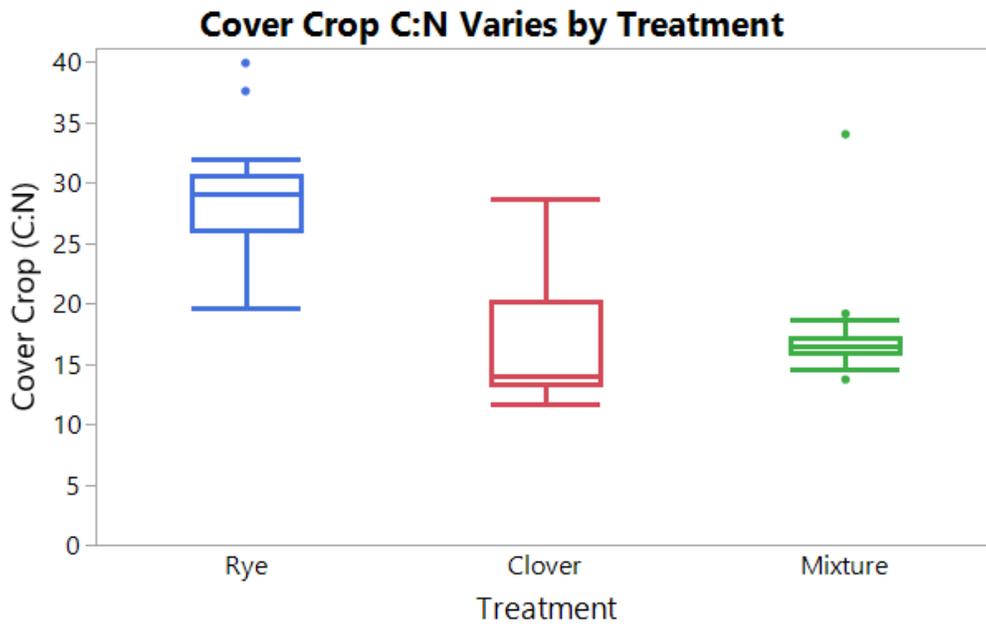
	<b>g OPOM/kg soil</b>	<b>kg OPOM/ha</b>	<b>%N</b>	<b>%C</b>	<b>mg N OPOM/kg soil</b>	<b>mg C OPOM/kg soil</b>
<b>Mean</b>	3.87	11674.39	1.67	25.32	64.11	968.51
<b>SE</b>	0.19	514.05	0.03	0.40	3.23	44.29
<b>Range</b>	5.70	16280.00	1.09	12.33	104.74	1310.01

**Table 5:** Means, standard error (SE), and range for soil inorganic nitrogen in Kg N/ha and in concentrations from extractions (EXT) and ammonium from incubations (INC) for soils collected across all farms in the study at the time of cover crop planting.

	<b>Kg NO3-N/ha (EXT)</b>	<b>ug NO3-N/g soil (EXT)</b>	<b>Kg NH4-N/ha (EXT)</b>	<b>ug NH4-N/g soil (EXT)</b>	<b>Kg NH4-N/ha (INC)</b>	<b>ug NH4-N/g soil (INC)</b>
<b>Mean</b>	165.29	55.03	27.05	8.74	240.35	79.57
<b>SE</b>	17.08	5.82	1.60	0.52	15.10	5.37
<b>Range</b>	389.17	134.40	40.66	13.10	413.61	162.32

**Cover Crop Litter Quality:** Across farms (F1, F2, F7A, and F7B), we found that cover crop biomass quality varied between treatments. In a mixed effects model with treatment as a fixed effect and block nested in field as a random effect, we found that rye had the highest mean C:N of 28.7, which is a typical value for grasses, and both treatments with clover had significantly lower C:N ratios of 16.8 for clover grown alone and 17.5 for both species combined in mixture ( $p < 0.0001$ ) (**Figure 1**). Rye on average made up 43% of total biomass in the mixture treatments, with a range from 14% to 68%. Adding rye to the mixture treatment did not significantly increase C:N compared to clover grown alone. This indicates that the mixture provided the same quality cover crop litter inputs as the clover biomass.

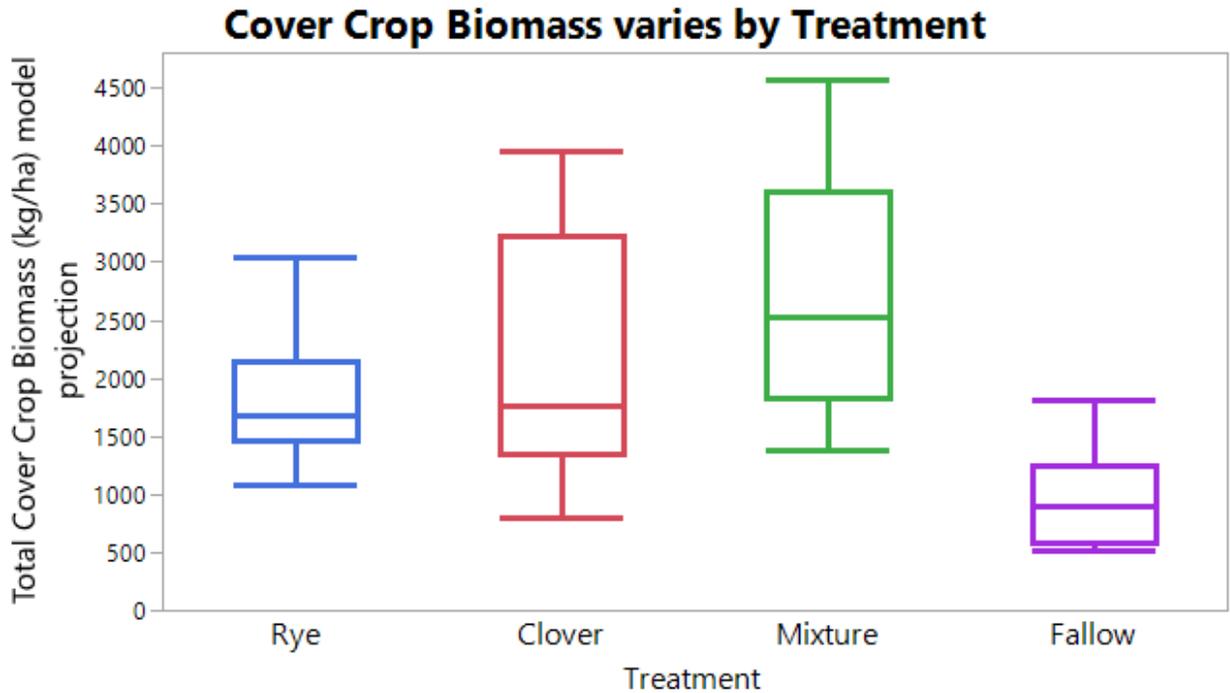
**Figure 1:** Box plot of cover crop carbon to nitrogen ratio (C:N) by cover crop treatment across the four fields where we collected cover crop biomass (F1, F2, F7A, and F7B). Rye’s C:N was statistically higher than clover’s or mixture’s C:N ( $p < 0.0001$ ).



**Cover Crop Biomass Variability Between Treatments:**

Using the pictures and plant heights we took across all farms, combined with the cover crop biomass data we collected on four farms, we were able to estimate cover crop biomass for the farms on which we couldn’t sample due to the pandemic. Using these data for all farms in a mixed effects model with treatment as a fixed effect and block nested in field as a random effect, we found that the mixture produced higher biomass (mean = 2731 kg/ha) than the rye (mean = 1818 kg/ha) or clover (mean = 2151 kg/ha) treatments ( $p < 0.0001$ ). All cover crop treatments produced more aboveground biomass than the weedy fallow control (mean = 956 kg/ha). These results support our hypothesis that the mixture would produce higher overall biomass inputs than either species grown alone. While this supports our hypothesis, we did not find a different C:N between mixture and clover.

**Figure 2:** Boxplots of cover crop biomass (estimated from plant coverage and height) across all farms.



As visualized in the boxplots in **Figure 2**, we found high variability in cover crop biomass across farms indicating that factors other than cover crop treatment impact cover crop outcomes. The next section explores soil properties that were most highly correlated with cover crop outcomes.

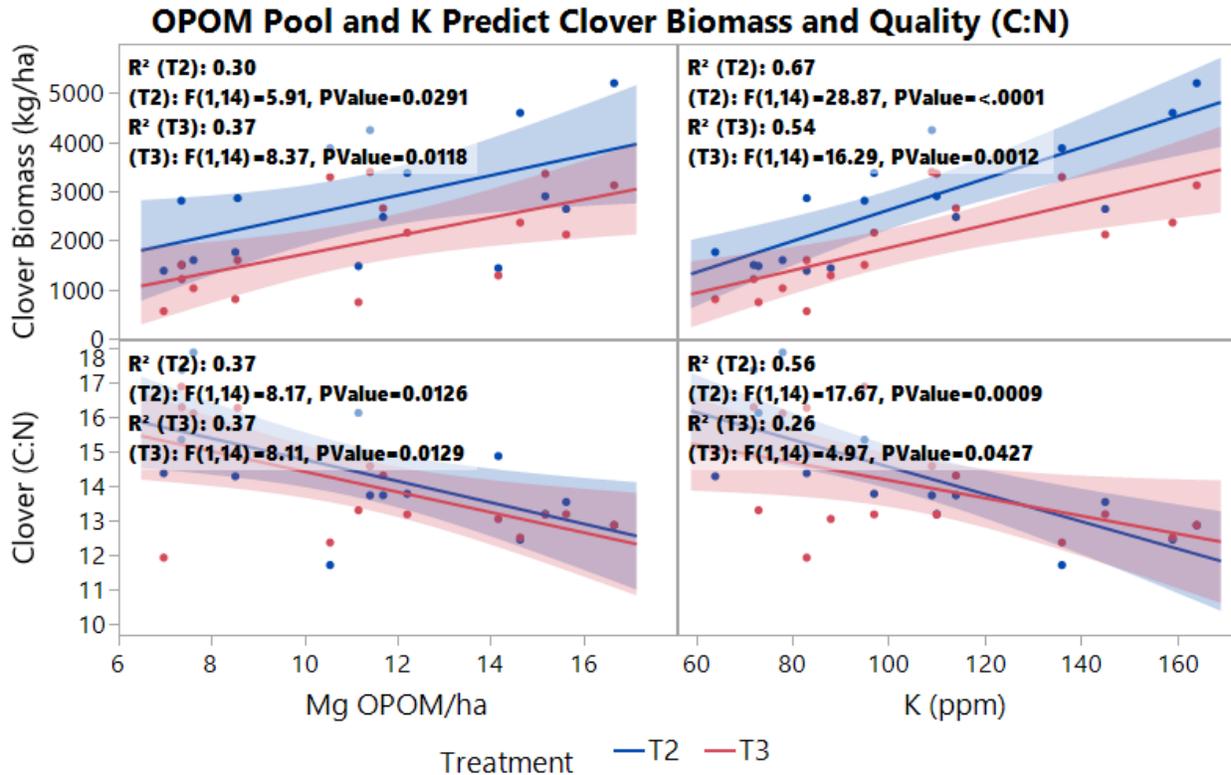
### ***Soil properties affect cover crops:***

The occluded particulate organic matter (OPOM) fraction was a predictor of quality (i.e., C:N) and quantity for crimson clover when grown alone and in mixture across farms (F1, F2, F7A, and F7B). OPOM is a fraction of soil organic matter that is physically protected inside soil aggregates and changes slowly with a turnover time of decades. We found that the OPOM pool (Mg OPOM/ha) was significantly correlated with clover biomass (kg/ha) and quality (C:N). As the OPOM pool increased, we found a decrease in C:N (the proportional amount of N increased). Since legumes fix nitrogen, legumes growing in higher quality soils with larger intermediate soil carbon pools, may be fixing more nitrogen than legumes in poorer quality soils. This corresponds with higher total N in clover biomass and higher overall biomass production. We found a similar trend with potassium (K), a critical nutrient for legumes (**Figure 3**).

While OPOM pools and K significantly improved clover outcomes across farms, we did not see the same strong positive trends with mg N in OPOM/kg soil, which was not a predictor of clover biomass or quality. This observation is reasonable given that the legume, clover, fixes its own N and is thus not dependent on soil N pools for biomass production. We also expect legume biomass to be lower in the mixture in soils with more POM N because it is less competitive with grasses that do need soil N. Soil carbon storage, an indicator of soil quality, and nutrients such as K are more important metrics for predicting legume success. Farmers with

higher OPOM and K pools may benefit more from planting clover as a cover crop than farmers with lower quality soils in the early years of cover cropping.

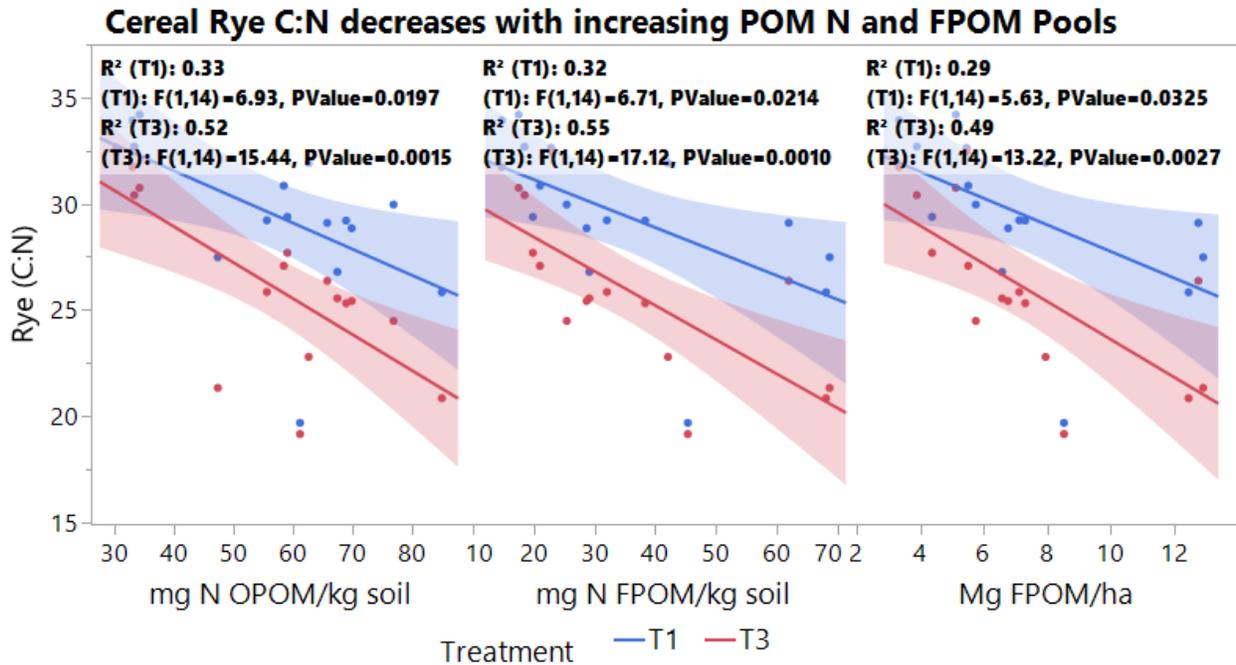
**Figure 3:** Linear models of the OPOM pool (Mg OPOM/ha) plotted against clover biomass (kg/ha) and clover quality (C:N) across farms. Models split by treatment into clover grown alone (T2) and clover grown in mixture with rye (T3).



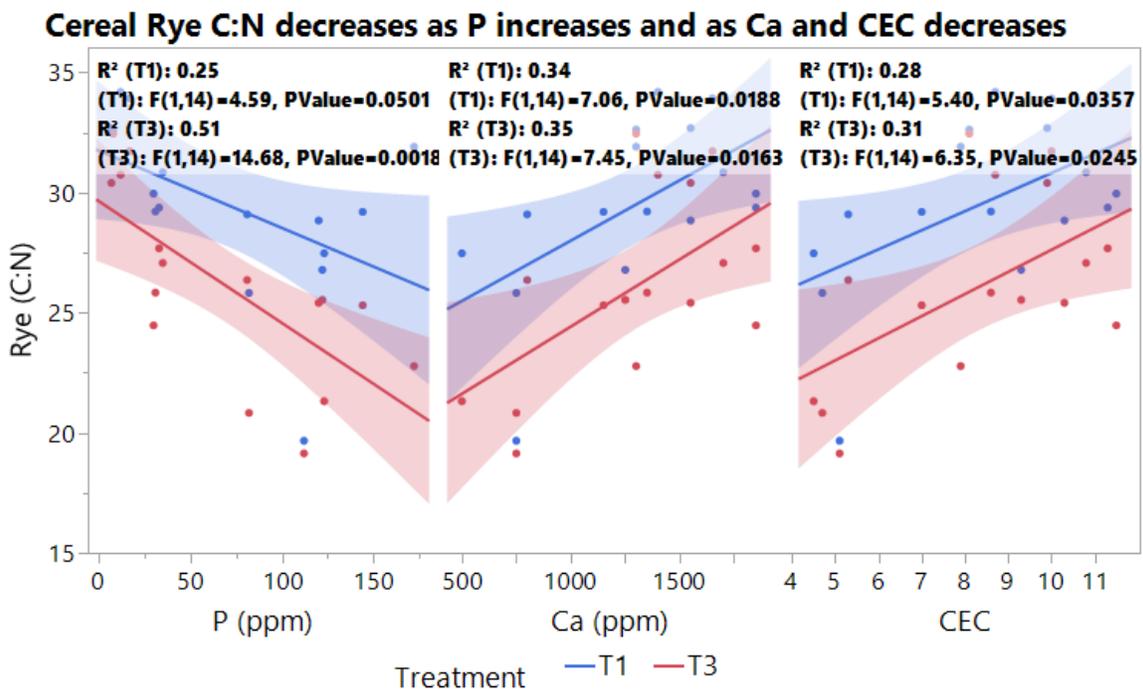
We found that a range of soil properties impacted cereal rye quality across farms (F1, F2, F7A, and F7B). First, we found that the concentrations of OPOM and FPOM N were significantly and negatively correlated with the C:N ratio of cereal rye when grown alone and in mixture, with a stronger relationship in the mixture. We found a similar negative relationship between the FPOM pool and C:N (**Figure 4**).

POM was not the only soil property that helped explain cereal rye quality. We also found significant and positive relationships between calcium (Ca) and cation exchange capacity (CEC) and cereal rye C:N and a negative relationship between P and C:N (**Figure 5**), indicating that as P increases, N concentration also increases in rye biomass. Since P is a limiting nutrient, rye may be able to absorb more N from the soil when P is not limiting. This relationship was stronger in the mixture than in the monoculture, which also suggests that higher P levels may be allowing the rye to compete more against the clover. We did not find any effect of these nutrients on total rye biomass, though, indicating that these soil properties influenced the quality, rather than the quantity, of the cereal rye biomass. Cover crop quality matters to future cash crop growth in addition to just cover crop biomass, so it is an important metric for farmers to consider when managing their cover crops.

**Figure 4:** Linear models of particulate organic matter (POM) plotted against cereal rye's carbon to nitrogen ratio (C:N) across farms. Models are separated between rye grown alone (T1) and rye grown in mixture with clover (T3).



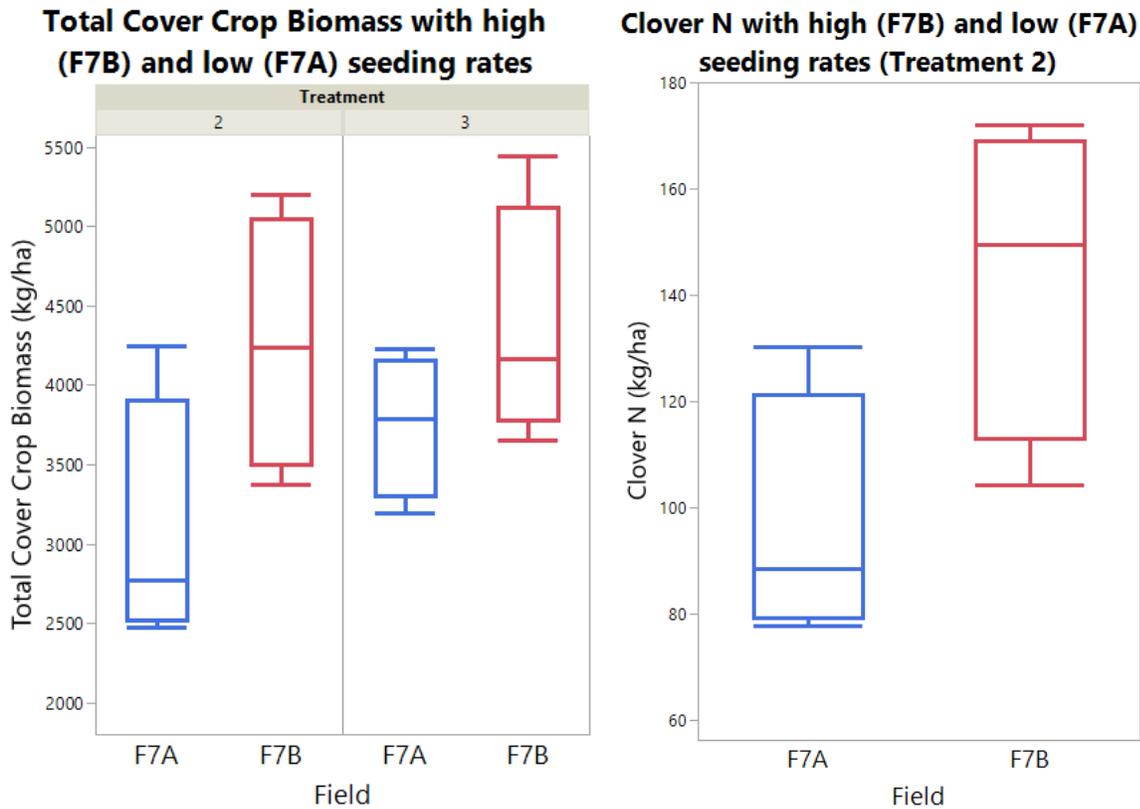
**Figure 5:** Linear models of soil phosphorus (P (ppm)), calcium (Ca (ppm)) and cation exchange capacity (CEC) vs. rye C:N across farms. Models are separated between rye grown alone (T1) and rye grown in mixture with clover (T3).



**Effect of Crimson Clover Seeding Rate on Biomass:**

On farm 7, clover was accidentally planted at 28 lbs/acre in F7B, while F7A, on the same farm, was planted to 16 lbs/acre (the intended rate for the study). This mistake allowed us to compare cover crop biomass outcomes based on seeding rates (**Figure 6**). In the clover monoculture treatment (T2), we found higher (but not significantly,  $p = 0.0799$ ) biomass (kg/ha) and significantly higher total N (kg/ha) ( $p = 0.0463$ ) in the treatment that was planted double (F7B). There was no difference between total cover crop biomass, C, or N (clover + rye) in the mixture treatment, or in clover biomass in the mixture treatment. Competition with rye likely reduced the effect of clover seeding rate on clover biomass in the mixture. Since we found no difference when clover was planted in mixture, farmers may not benefit from increasing seeding rates of legumes in cover crop mixtures. However, increasing seeding rates when legumes are planted alone has the potential to moderately increase total biomass although may not be worth the higher price.

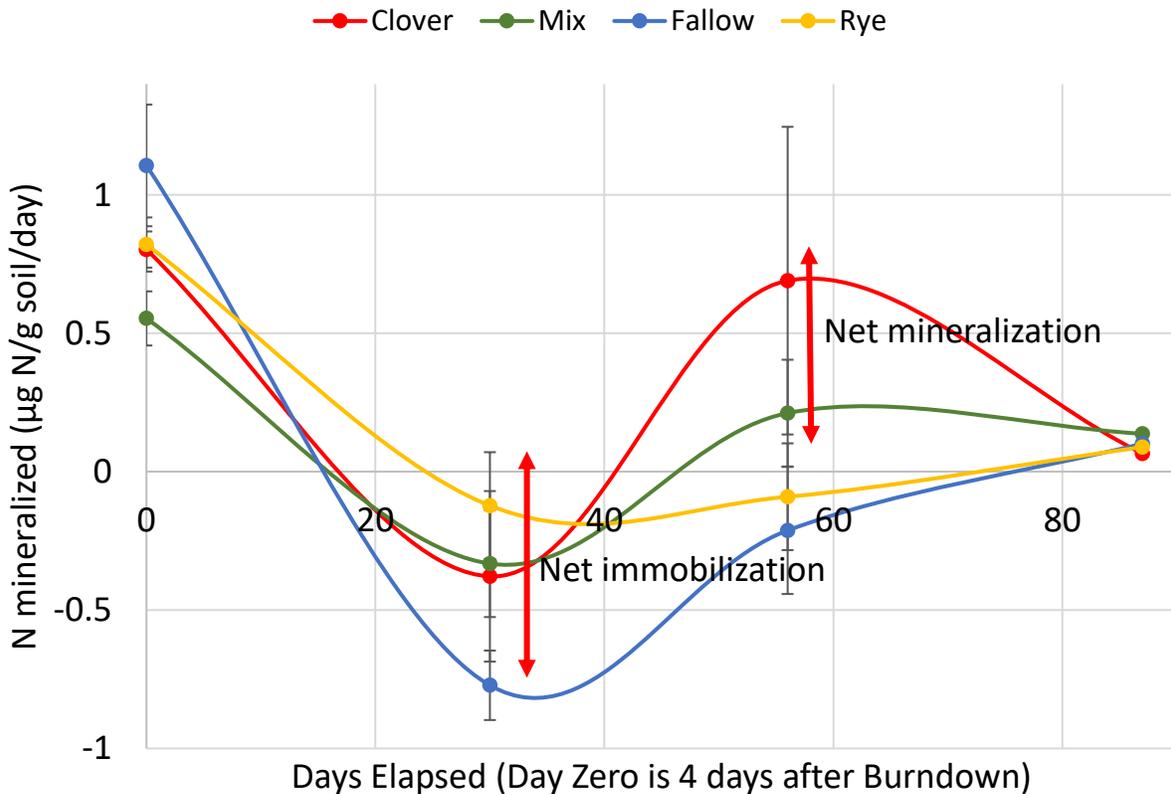
**Figure 6:** Total cover crop biomass and cover crop N in monoculture vs. mixture between fields with high (F7B) and low (F7A) seeding rates.



### Nitrogen Mineralization Following Cover Crop Treatments:

Our approach to estimating nitrogen mineralization in the field worked well on one of the farms that carefully followed the study design and only fertilized once at corn planting (73 lbs N/acre). This fertilization occurred 13 days before we collected and buried cores to measure N mineralization, and 4 days after cover crops had been burned down. The differences between treatments are not statistically different, however, the fallow treatment had higher N immobilization (mean =  $-0.77 \mu\text{g N/g soil/day}$ ) than the treatments with cover crop inputs in the first month, and the clover treatment had higher N mineralization rates later in the growing season (mean =  $0.69 \mu\text{g N/g soil/day}$ ). Notably, the fallow and rye treatments did not shift from net immobilization (below the 0 line) to net mineralization (above the 0 line) until the end of the measurement period, while the clover and mixture treatments shifted to net mineralization about halfway through the growing season. Based on these results, farmers would benefit most from planting cover crops that include legumes specifically in terms of N availability. Added N from legumes can help drive higher soil mineralization rates through decomposition of organic matter that provides enough N to microbes such that they release excess N back into the soil (mineralization) (**Figure 7**).

**Figure 7: N Mineralized Following Cover Crop Burndown and Corn Planting**

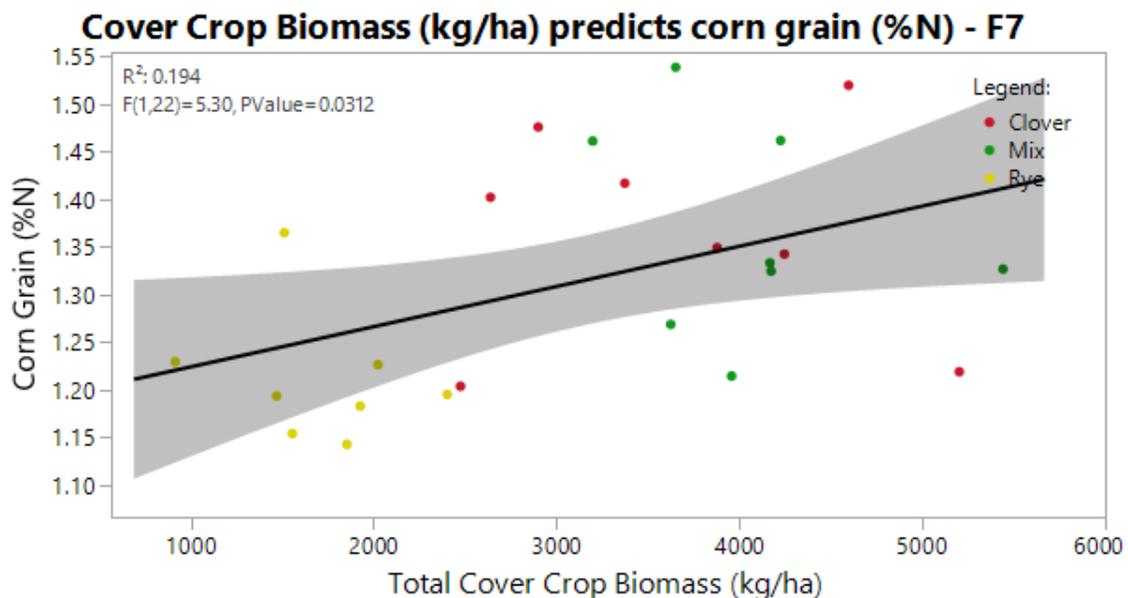


### *Effect of Cover Crops on Corn Yield:*

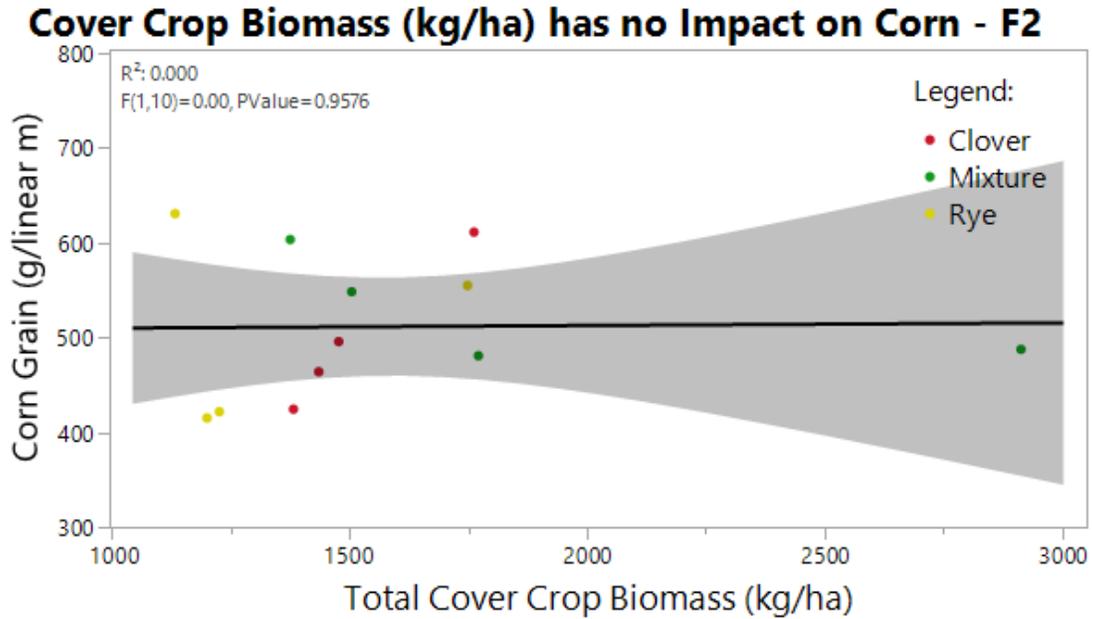
We found variable corn biomass production across farms, each being impacted by soil fertility, weather, and success of growing and terminating cover crops. When corn grain biomass was compared across all farms, there were no differences between cover crop treatments. We did find a difference in corn quality between cover crop treatments across farms. Corn C:N was lower in treatments with clover (the monoculture and the mixture) compared to the rye and fallow treatments ( $p = 0.0012$ ) with the highest % corn N in the mixture treatment (mean = 1.35) followed by the clover treatment (mean 1.34), both significantly higher than the rye treatment (mean = 1.28) ( $p = 0.0033$ ).

We also analyzed the corn grain production by farm. Except for on farm 1, we generally found positive or neutral relationships between cover crop biomass and chemistry and corn production. On farm 7, across both fields and all treatments, we found that total cover crop biomass (kg/ha) predicted corn grain quality (%N) (**Figure 8**). On farm 2, we found no impact of cover crops on corn production (**Figure 9**). While on farm 7 and farm 2, we did not find a negative effect of cover crops on corn production and quality with some positive outcomes, on farm 1, we found the opposite trend. As cover crop biomass increased, corn biomass decreased (**Figure 10**). In fact, we found that the mixture treatment resulted in significantly lower corn production than the fallow treatment, while the sole clover and rye treatments produced the same corn biomass as the fallow treatment ( $p = 0.03$ ). The unpredictable impact of cover crops on yield is often cited as a reason not to grow cover crops. While we did have one unfavorable outcome on one farm, all other farms saw either positive or neutral impacts of cover crops on corn. Based on this, although there is some risk of damaging corn yield, the overall benefits outweigh the risks.

**Figure 8:** On farm 7, across both fields and all treatments with cover crops, as total cover crop biomass increased, the quality of the corn grain (%N) also increased ( $p = 0.0312$ ,  $R^2 = 0.194$ ).



**Figure 9:** On farm 2 on the field where we collected cover crop biomass, there was no relationship between cover crop biomass and corn grain production ( $p = 0.9576$ ,  $R^2 = 0.000$ ). This also shows that on this farm cover crops did not negatively affect yield.



**Figure 10:** On farm 1, across all treatments with cover crops, we saw that as total cover crop biomass increased corn grain in bu/acre decreased.

