Evaluation of diagnostic tools and fertilizer sources for phosphorus and sulfur management in winter wheat

by

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Abstract

Three studies were conducted to assess winter wheat (*Tritticum aestivum*) response to phosphorus (P) and sulfur (S) and evaluate soil test methods and tissue tests as diagnostic tools. The first study compared different commercially available soil test P methods in Kansas soils. This study was performed on 24 locations in 2019 and 2020. It was designed to include various soils across the wheat-growing regions of Kansas, and locations were selected based on initial soil test P concentrations. Fertilizer treatments included pre-plant broadcast P fertilizer at rates of 0, 45, 90, and 135 kg of P_2O_5 ha^{-1}. Soil samples were collected at the 0-15 cm depth and analyzed using six soil test P methods. Results from this study found that most soil test P (STP) methods correlate well with the Mehlich-3 (M3) method. The highest correlation was with the Bray-1 (B1), while the lowest was with the H3A. The B1 and H3A method correlations to the M3 method were affected by high soil pH and calcium carbonate. Critical STP levels for winter wheat differed between STP methods. The lowest critical level for wheat grain yield was 12.2 mg of P kg^{-1} for the Olsen test, while the highest was 36.7 mg of P kg^{-1} with the Mehlich-3 ICP test. The second study evaluated soil test S methods and wheat response to sulfate and elemental S fertilizer sources. Sulfur application rates included 0, 11, and 45 kg of S ha^{-1}, and all plots received a balanced blanket application of N and P fertilizer. Profile soil samples (0-30 and 30-60 cm) were taken before fertilization and tested with four different S soil test methods. Plant tissue samples were collected at the Feekes 6 growth stage, and flag leaf samples were collected at the Feekes 10.5 growth stage. Results from this study indicate that the four sulfur soil test methods had a wide range of correlations indicating that different methods could be extracting S from different sulfur pools of S in the soil. When comparing these methods, the calcium phosphate and ammonium acetate methods correlated the best between all method comparisons

to one another. When analyzing tissue S concentration, S rates had the biggest impact while the source of S had limited response at any location. The third study evaluated the effects of blending P fertilizer with winter wheat seed prior to drilling. Treatments included a factorial combination of two P fertilizer sources, three P rates, and four storage times of the seed-fertilizer blend prior to drilling. Four locations were established in 2019 and 2020. Fertilizer sources included diammonium phosphate (DAP) and MESZ; rates included 34, 67, and 135 kg of P_2O_5 ha^{-1}; and blend storage times of 0, 7, 22, and 34 days. Results from this study found that the blend storage time and fertilizer source had limited impacts on winter wheat NDVI, P uptake, biomass, and grain yield responses in the field. P fertilizer rate had the biggest impact on NDVI, P uptake, biomass and grain yield responses and was the biggest response in this study which was likely due to locations being lower in P compared to general guidelines.

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Chapter 1 - Literature Review and Thesis Organization

Phosphorus is an essential element in the winter wheat life cycle and is commonly applied to all crops used in food production. My study focuses on how soil phosphorus and phosphorus fertilizer affect plant growth as well as grain yield. Knowing how this nutrient is applied in an agricultural setting will help to identify the current research needs for winter wheat. This crop is grown extensively in Kansas and makes up a large portion of the revenue made by producers in the area. Years of research has shown that P fertility is one of the important factors affecting grain yield (Kastens, 2000). Every crop grown and removed from a location means elements such as phosphorus are removed from the soil. Removal can occur through the removal of biomass, grain, and fiber, which all contain different concentrations of phosphorus. Commercial fertilizers are commonly used to replenish phosphorus to maintain crop productivity.

However, many factors need to be addressed to ensure an economic return is achieved when a crop such as winter wheat is produced. Over-fertilization can mean a producer spent too much money on fertilizer and can generate environmental risks, while under-fertilization can mean a yield was not economical due to a nutrient-limited crop. Phosphorus deficiencies in winter wheat can be easily prevented through the use of modern scientific techniques to analyze the soil, especially when they are paired with application techniques used by producers today. This review will focus on how phosphorus affects wheat growth and will describe how source, placement, rate, and critical soil test values affect winter wheat's overall economic productivity.

Crop Response to Phosphorus

To understand how phosphorus affects the growth of winter wheat, we first need to review the wheat plant life cycle. Winter wheat is planted in the fall at an early enough time to allow the crown, stems, and leaves to develop properly before winter arrives. This stage is known as the tillering stage. Tillering is when a grass plant, or wheat in this case, grows multiple stems from a centralized budding location which is located at the base of the wheat plant. Tillers play a crucial role in producing grain, and sufficient numbers of productive tillers are needed to achieve good crop yields (Xiu-Xiu Chen, 2019). Once tillering is completed, wheat can survive through the winter with limited plant growth until temperatures increase in the spring. When spring temperatures increase, tillers will grow in size and eventually produce spikes that will flower. If these spikes are fertilized properly, they will produce grain that can mature and be harvested and removed from the field.

Phosphorus's Effects on Wheat Growth

Phosphorus is an important nutrient during fall tillering. Critical levels of phosphorus are needed to ensure productive tillers for the winter wheat crop. A productive tiller or shoot is one that produces a viable spike for grain production. Research has shown that the dry weight of tillers increases as the phosphorus concentration of the tiller increases in a linear relationship (Xiu-Xiu Chen, 2019). This increase in biomass may allow tillers to survive winter and remain productive in the spring. Tillers having a concentration of 2.7 g/kg phosphorus resulted in 64% of the tillers being productive (Xiu-Xiu Chen, 2019). The productive tillers result in spikes that produce grain that is harvestable. A study in China showed 50% of grain yield could be explained by the number of spikes (Xiu-Xiu Chen, 2019). This also confirms that productive spikes are necessary for good grain yields. Spikes can have different numbers of seeds, which can affect crop yield. However, the number of seeds in a spike accounts for only between 13% and 3% of variability measured in grain yield and is likely not a major factor contributing to yield differences (Xiu-Xiu Chen, 2019).

Phosphorus Fertilizer Sources and Applications

Many phosphorus fertilizer sources are available, but the source does not seem to influence positive crop responses in wheat. Most, if not all, fertilizers have been labeled according to the percentages of each of the major nutrients that are found in the fertilizer. An example would be 18-46-0; this fertilizer would contain 18% Nitrogen, 46% P2O5, and 0% K2O. When Gokmen and Sencar (1988) compared DAP 18-46-0 (Diammonium Phosphate) to TSP 0-45-0 (Triple Super Phosphate), they found no differences in the wheat growth responses (Gokmen and Sencar, 1998). In their study, the nitrogen was balanced to ensure any nitrogen response would be accounted for. Applying a fertilizer containing phosphorus to a wheat crop often occurs in many different manners.

Two main placement techniques are used when phosphorus fertilizer is applied to wheat. These are broadcasting and banding (Larson and Herron, 2000). When broadcasting is used, fertilizer is spread evenly across the soil surface. Broadcasting fertilizer is a more common practice in wheat production compared to other methods of fertilizer applications (Gokmen and Sencar, 1998). This is likely due to the ease of application for the producer. Timing the application may not be as critical when broadcasting is used, which likely is why producers prefer this application technique. With banding, fertilizer is injected into the soil, and it forms a concentrated band at or below the soil surface. The width between bands can change, but the concept is still the same.

Banding is more effective than broadcasting and generally increases productivity on soils with high fixation and low levels of phosphorus (Gokmen and Sencar, 1998). In high calcareous soils, lime has been known to bind with phosphorus causing fixation (Price, 2006). In these high calcareous soils, banding phosphorus has been known to increase phosphorus's availability for plant growth compared to a broadcast application (Larson and Herron, 2000). Banding with or around the seed area is the most efficient use of phosphorus fertilizer (Vitosh, 1998). This is because the band has less surface area in contact with the soil and results in less phosphorus fixing to lime compared to broadcasting, which allows more fertilizer contact with the soil.

Banding phosphorus fertilizer is generally completed in two ways, banding with the seed or banding to the side or below the seed (Gokmen and Sencar, 1998). Banding with the seed is said to efficient because phosphorus is in close proximity to the germinating wheat seed but comes with some risk (Vitosh, 1998). Applications exceeding 30 kg $ha^{-1}$of N plus K_2O per acre with the seed are known to cause germination injury (Vitosh, 1998). Because many commercial phosphorus fertilizers contain low concentrations of these elements, banding them with the wheat seed is possible. Researchers from Turkey reported that fertilizers placed in contact with the seed at the time of planting prevent or delay germination and drastically reduce emergence (Gokmen and Sencar, 1998). The extent of the injury to the wheat crop will likely be linked with how much nitrogen and K_2O were contained in the nutrient that was banded with the seed at planting time.

Another method is banding to the side or below the seed. Banding below the seed has been found to prevent fertilizer injury to germinating seedlings and may result in higher yields (Gokmen and Sencar, 1998). Banding to the side of the seed provides a way to effectively apply phosphorus fertilizer without the potential negatives of placing fertilizer in contact with the wheat seed. When Gokmen and Sencar (1998), banded fertilizer below the seed, it resulted in a higher number of spikes compared to banding to the side or with the seed. Both broadcast application and banding applications have advantages and disadvantages. However, banding phosphorus fertilizers away from the seed leads to greater crop safety.

Phosphorus Rates, Economics, and Critical Soil Test Levels

Optimal phosphorus rates are highly dependent upon application methods and economics (Larson and L. Herron, 2000). The application method affects how much phosphorus fertilizer is needed to achieve a yield response in wheat. Researchers have found that seed-banded phosphorus can be three to four times more effective than broadcasting (Peterson, 1981). This can greatly affect how much fertilizer is needed. However, if rates are too low, phosphorus fertilizer may not be enough to replace what the crop is removing in the long term. To determine the correct fertilizer rate, soil testing should be performed to determine how much fertilizer is needed for an economic yield response (Kastens, 2000).

Knowing the critical soil test level of phosphorus is a way of knowing when a wheat crop will respond and will not respond to phosphorus fertilizer. A critical soil test value of 46 mg/kg was found to be profit-maximizing when using a Bray 1 soil test was used (Kastens, 2000, p. 9). However, the critical soil test value was found to be 19 mg/kg when a Bray 1 soil test for winter wheat was used, which was higher than for both corn and soybeans (Sucunza and Boem, 2018, p.91). In Oklahoma, a critical level for winter wheat was found to be 25 ppm using a Mehlich 3 soil test (Arnall, 2010). These different critical levels for phosphorus mean finding a critical phosphorus level could be dependent on many factors that could include soil characteristics, environmental, and many other factors. Because phosphates exist in plant available and non-available, the availability of phosphorus is not easy to predict (Ciampitti et al., 2011). Without being able to predict the availability of phosphorus due to these soil interactions, identifying a critical level could depend on the soil type.

Soil test methods also affect the critical level of phosphorus as each can produce different results (Mallarino and Blackmer, 1992). Figure 1-1 displays corn response to phosphorus characterized by three different soil test methods in Iowa: Bray 1, Mehlich 3, and Olsen. These results show that commonly used soil tests have different crop responses at similar soil test phosphorus values for phosphorus in crop production (Mallarino and Blackmer, 1992). The results from a plot study showed a strong correlation between an Olsen soil test and a Mehlich 3 (Sedlar, 2018). While these two soil tests showed different critical levels, they correlated well enough to cross-reference each other when determining a critical phosphorus soil test level.

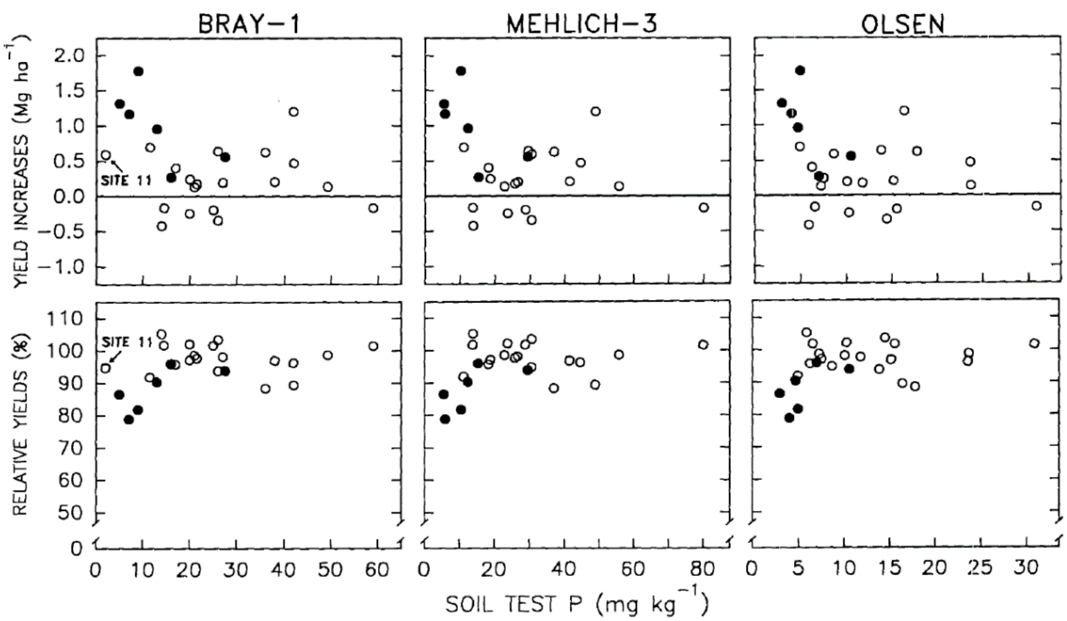

Figure 1-1. Phosphorus extracted from three soil test extractants compared to relative yield and yield increases (Mallarino and Blackmer, 1992).

Conclusion

In conclusion, winter wheat responds to phosphorus both in the soil and as fertilizer in many different ways. The life cycle of winter wheat depends on a proper nutritional balance. Without proper phosphorus fertilization, growth stages like tillering and fertility of spikes could be hampered. To protect against deficiencies, many different strategies, such as fertilizer placement and different soil test methods, are used to produce economic wheat yields. However, what is not truly known is exactly how much more efficient the banding application is compared to broadcast applications. One source showed banding was three to four more times efficient, but with soil properties affecting the availability of phosphorus, this result could depend on the growing environment. Further research should also be done to compare different soil test methods. Although many methods appear to correlate with each other, how these tests respond across different environments and soils is not fully understood.

Thesis Organization

This thesis is divided into five chapters. Following this introductory chapter, there are three chapters, each of them consisting of one research project. Titles of chapter 2, 3, and 4 are: "Determining critical concentrations for six soil test phosphorus methods for winter wheat", "Evaluation of soil test methods and plant analysis for sulfur in winter wheat," and "Blending phosphorus fertilizer with wheat seed: response to source, rate, and time". The fifth chapter is the overall conclusion. Each chapter contains an abstract, introduction, materials and methods, results and discussion, conclusions, and references.

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Chapter 2 - Determining critical concentrations for six soil test phosphorus methods for winter wheat

Abstract

Winter wheat (*Tritticum aestivum*) response to phosphorus (P) fertilization and new soil test phosphorus (STP) methods have not been recently evaluated in Kansas. The objective of this study was to evaluate six commercially available STP methods in different soil types. Phosphorus response trials were established at 24 Kansas locations in 2019 and 2020. Fertilizer treatments included four P rates using mono ammonium phosphate (MAP) (11-52-0) applied at 0, 45, 90, and 135 kg P_2O_5 ha^{-1}. All P fertilizer treatments plus 50 kg N ha^{-1} as urea were broadcast in the fall before sowing, followed by an additional 62 kg N ha^{-1} topdressed at Feekes 5 in the spring. The N applied with MAP was balanced, so all treatments received a total of 112 kg N ha^{-1}. A randomized complete block design with four replications was used in this study. Soil samples were collected at the 0-15 cm depth by rep at each site and analyzed for six different STP methods prior to fall fertilization. At Feekes 6, wheat biomass was collected and analyzed for P concentrations, and normalized difference vegetation index (NDVI) values were also collected. Grain yield was measured at harvest. Soil P extraction with the Bray-1 and H3A methods was significantly decreased at pH > 7.45, affecting correlation with other methods and wheat response predictive performance under high pH conditions. Critical STP values based on relative yield using the Mehlich-3 colorimetric method was 25.5 mg P kg^{-1} using the linear plateau model. When using other STP methods, the critical value varied from 12.2 to 36.7 mg P kg^{-1} for the Olsen and Mehlich-3 ICP methods, respectively. In general, all soil test methods did correlate well with crop response. The Bray-1 and H3A had similar critical values when locations with high pH were excluded. Early-season NDVI was related to both early season P

uptake as well as grain yield and was a valid way to determine an early-season critical STP level. Overall, in normal soils, knowing how a specific soil test P method correlates to crop yield is more important than picking a specific STP P method.

Abbreviations: N, nitrogen; P, phosphorus; K, potassium; S, sulfur; STP, soil test phosphorus (STP); normalized difference vegetation index, NDVI.

Introduction

Winter wheat is often considered to be one of the more responsive crops to phosphorus (P) fertilization. One of the best ways to identify responsive soils to P fertilizer is the use of soil testing. Soil testing helps identify fields or areas of a field that are likely to see a response to the addition of P fertilizer. Phosphorus fertilizer application is strongly based on the potential for an economic return on investment both in the short term as well as long term. However, this analysis can be challenging and is strongly dependent upon which soil test method is being used (Mallarino, 1995). This subject has become increasingly dynamic with the introduction of new soil test phosphorus methods that are now widely available for use in production agriculture (Dari, 2018). Identifying how each method correlates with crop response across a broad range of soils requires an ongoing field correlation-calibration process (Tang, 2009).

Kansas has a wide range of soil characteristics and properties that can affect how different soil test methods perform. In the west, soils can have pH values well over 7.5 and some even above 8.0, and in the southeast, soils that have a pH below 5 are not uncommon. Even within a single field, large swings in soil pH can be seen across a production field along with different soil properties, which could affect how an individual soil test would perform (Usowicz, 2017). This becomes a problem when using site-specific management, such as grid soil sampling. This management strategy purposefully takes soil samples from multiple regions or points in a field to identify areas of a field where P fertilizer inputs will yield a return on investment (Wollenhaupt, 1994). Using this sampling scheme results in the segregation of soil properties within a field. Samples taken within one field can express a wide range of variability that could impact how a soil test performs in the lab. Composite soil sampling usually does not result in such large ranges of soil properties because samples get mixed together and diluted out prior to soil testing in the lab.

Soil testing for crop response is not the only reason to use tests to assess soil phosphorus. Sharpley (1996) identified a positive relationship between soil test P levels and the amount of dissolved P in runoff water. This furthers the need to supply enough P fertilizer to attain an economic crop response while not over-applying P that could cause environmental concern.

This study's objectives were to i.) compare various soil test P (STP) methods and assess the correlation to winter wheat crop response, and ii.) identify critical STP levels and methods that correlate well with crop response under Kansas soil conditions.

Materials and methods

Treatments, Experiment Design, and Implementation

Twenty-four P fertilization trials were conducted in 2019 and 2020 in Kansas (Table 2-1). The study locations were on farmer's fields (18 locations) and university experiment fields (6 locations). Locations on farmer's fields were sowed by the farmer with all other field operations completed with research equipment, including N and P fertilizer applications. Research spraying equipment included a John Deere Gator with a 6-meter boom sprayer for wheat weed herbicides at Feekes 6 and a 3.6-meter boom CO_2 sprayer for fungicides at heading. For harvest, a 1.4-meter Wintersteiger combine was used. Either conventional tillage or no-till was used, with conventional tillage typically including 3 to 5 passes using a field cultivator prior to sowing, depending on the location. Crop rotations varied depending on the region and each farming operation. The most common rotations included wheat sowed directly after harvesting soybeans or wheat after summer fallowing.

The individual plot size was 13.7 m in length and 1.8 m in width, except location 21, with a length of 13.7 m and a width of 3.0 m. This change in plot area was used so the site could be planted to corn the next year. Row spacing depended on equipment but was either 25 cm or 19 cm (Table 2-1). Wheat varieties were selected by regional performance and those normally used by the farmer.

The experimental design was a randomized complete block design with four replications. Fertilizer treatments consisted of four rates of phosphorus (P) fertilizer (0, 45, 90, and 135 kg of P_2O_5 ha^{-1}) using mono-ammonium phosphate (MAP) (11-52-0). A non-limiting rate of N was applied for all the plots and adjusted to balance the N applied with the P fertilizer. Nitrogen from all fertilizer sources was balanced to a rate of 112 kg of N ha^{-1} of N for each treatment using Urea (46-0-0), except for locations 2, 6, 7, and 24, which received a total of 157 kg of N ha^{-1}. This increase in N was due to the producer applying pre-plant N using UAN or Urea prior to location establishment. Approximately half of the N fertilizer was broadcast-applied in the fall within a week before sowing and combined with the P fertilizer treatments. The remaining N fertilizer using urea was broadcast by hand as a topdress in the spring at the Feekes 5 growth stage.

Soil Sampling and Analysis

Composite soil samples of 10-12 cores (1.9 cm diameter) were collected at the 0-15 cm depth by block prior to sowing and fertilizer application. Soil samples were then dried at 41^{o} C for five to ten days. After samples were dried, the soil was ground to pass through a 2 mm sieve.

Soil samples were analyzed for pH 1:1 (soil:water) (Watson and Brown, 1998), soil carbonate content by the manometer method (Micah, 1963), organic matter by loss on ignition (Combs and Nathan, 1998), and potassium, calcium, magnesium, and sodium by the ammonium acetate extraction (Warncke and Brown, 1998). Cation exchange capacity was determined by summation (Chapman, 1965). Soil test P extraction methods included Mehlich-3 colorimetric (Frank et al., 1998), Bray 1 (Frank et al., 1998), Olsen (Knudsen, 1980), Haney H3A (Haney, 2017), and Resin (Hedley et al., 1982 with modifications of Condron et al., 1985). The Mehlich-3 extraction method was also analyzed by the ICP-OES (Inductively Coupled Plasma Optical Emission Spectrometry).

Plant Biomass, NDVI, and Grain Yield

Early-season biomass samples and normalized difference vegetative index (NDVI) were collected at the early jointing (Feekes 6) growth stage. Biomass samples were collected from 2 rows at a length of 76 cm from the back 3 meters of each plot. This sample area was excluded from future measurements. Samples were oven-dried at 60^{o} C for five to seven days and then weighed for dry biomass. Dry samples were ground to pass through a 2 mm screen using a Wiley benchtop cutting mill (Thomas Scientific, Swedesboro NJ). Samples were then digested using the sulfuric acid/peroxide digest method (Matsunaga & Shiozaki, 1987). Phosphorus concentration was determined by ICP–OES. Phosphorus nutrient concentration and biomass were used to calculate total plant P uptake.

The NDVI measurements were taken at the Feekes 6 growth stage using an active sensor RapidSCAN CS-45 handheld crop sensor (Holland Scientific, Lincoln NE). This sensor uses a wavelength of 670 nm, red-edge light at 730 nm, and near inferred light at 780 nm. For this study, the red and near inferred wavelengths were used to calculate the NDVI for each treatment.

The sensor was held at the height of 1.2 meters above the canopy, and the reading was collected for the entire length of each plot. At the end of each plot, the average measurement was recorded.

Grain yield was determined by harvesting the center 1.4 meters of each plot using a plot combine with a platform header. Subsamples were taken from each plot and analyzed for moisture and test weight using a Dickey-John GAC-2500-AGRI grain analysis computer.

Statistical Analysis

For regression analysis, the geom smooth function was used from ggplot in R version 4.0.3 (R Core Team, 2020). Stat poly eq was used to extract the formula of the regression. Regressions were all compared to the Mehlich-3 method because it is currently the standard phosphorus soil test method at Kansas State University and seen as a control for all other methods. For linear mixed-effects models the lme4 package was used in R version 4.0.3 (R Core Team, 2020). Blocks were included as a random effect in the model**.** For analysis across locations, blocks and locations were included as random effects in the model. Statistical significance was set at alpha = F 0.05. Relative values for NDVI and yield were defined as the response of the only N applied treatment divided by the highest response treatment containing both N and P. Critical soil test P values were determined for each extractant using the bivariate Cate-Nelson analysis (Cate and Nelson, 1971) and the segmented regression linear-plateau model (nls package) R version 4.0.3 (R Core Team, 2020).

Results and discussion

Phosphorus Soil Test Methods

Soil test P levels varied from 2 to 48 mg of P kg^{-1} across the 24 locations using a Mehlich-3 colorimetric (M3-COL) method (Table 2-2). A range of soil pH’s (5.4-8.3) and calcium carbonates (0-11.4 mg kg^{-1}) was observed at some locations in our study. Other parameters such as soil organic matter, cations, and CEC also varied between site locations.

The M3-COL method correlated well with the Bray-1 (B1) method and had the highest R^2 at 0.97 when samples with pH>7.45 were excluded (Figure 2-1A). Culman (2020) found a lower R^2 of 0.85 when comparing these two methods for soils in Ohio; however, soils with a higher pH were not excluded from the analysis. The intercept of 1.69 and a slope of 0.758 indicate a close relation for these two methods; however, the interpretation is not exactly the same based on our set of soils (Figure 2-1 A). The linear regression parameters presented here excluded soils with a pH > 7.45 (Figures 2-2 A & B). Although these two methods had the strongest correlation among all soil tests compared to the M3-COL, the presence of calcium carbonates decreased the amount of P extracted with the B1 method. This same trend has been documented in other studies that contained calcareous soils (Mallarino and Blackmer 1992). In general, the M3-COL method extracted greater amounts of P than the B1 method when soils exceeded a pH of 7.45 (Figure 2-1 A); previous studies found similar trends (Culman, 2020). Using the Mehlich-3 extraction with the measure of P using ICP (M3-ICP) the M3-ICP and B1 methods showed a similar slope (0.765) and an R^2 of 0.91 (Table 2-4). The biggest difference when comparing the M3-COL or the M3-ICP method to the B1 method was the y-intercept. This change was 6.5 mg of P kg^{-1} higher with the M3-ICP method (Table 2-4, Figure 2-1 A).

The M3-COL and Olsen methods correlated well in a linear regression with an R^2 of 0.89 and slope of 0.395 (Figure 2-1 B). Like previous methods, high pH soils were excluded from the analysis to ensure the same soil points are being used in each analysis. However, soils with high pH and carbonates showed little effect on the relationship of Olsen and M3-COL, indicating a better correlation between these two methods for a range of soil pH's. The Olsen method is known to extract smaller amounts of P than other methods, particularly when compared to the M3-COL method (Mallarino, 1995). Similar trends were found when comparing Olsen to the M3-ICP method (slope of 0.378), but a lower R^2 of 0.77 and slightly lower intercept (Table 2-4).

The H3A method resulted in the lowest R^2 value compared to the M3-COL method with a value of 0.74 (Figure 2-1 C). Even though the resulting R^2 is the lowest, previous studies indicated that these two methods generally correlated well (Rutter, 2020). Similar to previous studies, the H3A method extracted slightly smaller amounts of P than the M3-COL (Rutter, 2020). When soil pH and carbonates increased, the H3A method did not follow the same trend as the M3-COL method and extracted significantly less P. (Figures 2-2 C & D). These results build upon previous literature that shows the H3A extraction method is affected by high soil pH (Rogers, 2019). The relation of the H3A method to the M3-ICP had a slightly lower R^2 of 0.65 with a slope of 0.648, which is also lower than the comparison of H3A with M3-COL (Table 2-4, Figure 2-1 C).

The Resin STP extraction method correlated well to the M3-COL method, resulting in an R^2 of 0.87, a slope of 0.524, and an intercept of 3.09 (Figure 2-1 D). While correlating well, the Resin method generally extracted lower amounts of P than the M3-COL method (Figure 2-1 D). High pH soils were excluded from the regression analysis as indicated by red-colored dots in Figure 2-1; however, the effect of high pH was negligible when compared to B1 or H3A but was excluded for consistency. Comparing Resin to the M3-ICP method, we see slopes and R^2 values that are nearly identical to those for the relationship between Resin and M3-COL methods (Table 2-4). The main difference was the y-intercept which was approximately 4.6 mg of P kg^{-1} lower when comparing the M3-COL to Resin relation.

The M3-COL and M3-ICP methods showed a good overall correlation with a slope close to one (0.931) and R^2 of 0.91 (Figure 2-1 E). However, the intercept of the regression comparing these two methods was 8.94 mg of P kg^{-1} (Figure 2-1 E). This result is important because of the potential implications in the interpretation of critical STP values and fertilizer recommendations. The M3-ICP method is extracting around 9 mg of P kg^{-1} more than the M3-COL. This result is also supported when comparing the other soil test methods to the M3-ICP as the slopes were mostly unchanged, but the intercepts were different when compared to those for regressions with M3-COL. The commercial use of M3-ICP is increasing in popularity due to the potential for high throughput analysis in the laboratory when combining a multi-element extraction (Mehlich-3) and analysis with the ICP. However, the potential implication of using the ICP determination versus the more traditional colorimetric method on established STP critical values and current guidelines is crucial.

Early season NDVI and P Uptake

The values of NDVI collected at the Feekes 6 growth stage showed a statistically significant treatment effect at 14 locations (Table 2-3). The locations with statistically significant NDVI response to P fertilizer had soil test values ranging from 8.5 to 26.3 mg of P kg^{-1} using the M3-COL method (Table 2-2). Unresponsive locations had soil test values ranging from 10.8 to 48.3 mg of P kg^{-1} (Table 2-2). This was comparable to trends found for plant biomass and P uptake at the same growth stage.

Across all locations, there was a significant increase in NDVI values up to 90 kg of P_2O_5 ha^{-1} (Figure 2-3 A). However, the majority of NDVI response was seen at the 45 and 90 kg of P_2O_5 ha^{-1} rates. Increases of 0.08 and 0.05 in NDVI value were seen between these fertilizer rates, respectively, compared to the no P fertilizer treatment (Figure 2-3 A). Across locations, P fertilizer statistically increased NDVI values at Feekes 6 by 29 percent. Evaluating wheat response to P fertilizer with NDVI is not common in the literature, and NDVI is more commonly used to identify nitrogen response. However, NDVI is directly correlated to the amount of biomass (Moges, 2004), and this data supports that winter wheat shows positive biomass increases to P fertilizer under lower STP conditions. The increase in wheat biomass is likely due to increased tillering in the late fall and spring, reflecting a higher NDVI value at the Feekes 6 growth stage. The increase in early biomass accumulation also resulted in greater total P uptake (Figure 2-3 B). Increased tillering would lead to more red light being absorbed by the canopy and more NIR light reflected back to the sensor. This relationship may result in difficulty defining whether the wheat response is due to nitrogen or P response in a production wheat crop that has had both nitrogen and P fertilizers applied.

Early P uptake was measured at the Feekes 6 growth stage, and the responsive locations had an M3-COL test ranging from 8.5 to 20.1 mg of P kg^{-1} (Table 2-3). Across all locations, there was a significant increase in early season P uptake up to the highest P fertilizer rate of 135 kg ha^{-1} P_2O_5 (Figure 2-3 B). The lowest uptake occurred when no P fertilizer was applied and resulted in 2.5 kg of P ha^{-1} uptake (Figure 2-3 B). Overall, the largest response occurred between the 0, 45, and 90 kg of P_2O_5 ha^{-1} rates with an increase of 1.2 kg of P ha^{-1} with each fertilizer rate increase (Figure 2-3 B). The addition of P fertilizer increased P uptake at the Feekes 6 growth stage by 133 percent compared to the control treatment with no P fertilizer applied.

Calcareous STP Method Distribution Compared to Relative Yield

Now that the data has been analyzed to determine how P fertilizer affects the productivity of winter wheat, it can now be determined how this productivity is related to the soil test methods discussed earlier in the chapter. As found earlier, pH played a role in determining how each of these methods compared to the M3-COL method. Identifying how pH affects a test method's correlation to crop productivity is important and needs to be done before a full-scale analysis can be done to address the critical values of each test method.

Figure 2-4 shows the relative yield compared to all five soil test P methods and is color-coded to pH to help identify sites with soil pH's greater than 7.45. The M3-COL method generally did not seem to be affected by high pH sites falling in line with the general trend (Figure 2-4 A). However, the B1 method's correlation to crop yield response was affected by high pH soils, with these sites falling to the left of the trend seen in the data (Figure 2-4 B). The Olsen method correlated well with crop yield response, with all points following the same trend (Figure 2-4 C). The H3A method showed more variability when comparing it to crop yield response, and all points seemed to follow the same trend. However, these soil points were low testing which placed them close to the y axis making it hard to tell if they fall in line with general crop response trends (Figure 2-4 D). Knowing that the H3A follows the same trend as the B1 in high pH soils makes it hard to determine if these sites are following the same trend or not. The second to last soil test P method is resin, similar to M3 and Olsen, it too followed the same trend when compared to crop yield response (Figure 2-4 E). The last method analyzed was the M3-ICP method. With this method, higher pH soils followed general trends of crop response much like the M3-COL method (Figure 2-4 F). Overall, due to crop response not being accurately represented in high pH soils, sites having a pH greater than 7.45 were omitted from further analysis determining critical soil test P levels with the B1 and H3A methods.

Grain Yield and Critical Soil Test Phosphorus Values

From the harvested locations, 14 showed statistically significant yield differences with P fertilizer treatments at the 0.05 probability level (Table 2-3). Responsive locations had STP

(M3-COL) values ranging from 8.5 to 41.0 mg of P kg^{-1} (Table 2-2). Averaged across locations, there was a significant increase in grain yield up to the 90 kg of P_2O_5 ha^{-1} rate that resulted in a 673 kg ha^{-1} yield increase compared to the control treatment with no P fertilizer (Figure 2-3 C). There was no significant difference between the 90 and 135 kg of P_2O_5 ha^{-1} rates of P fertilizer (Figure 2-3 C). Across locations, grain yield showed a statistically significant increase from 2953 kg ha^{-1} without P fertilizer to 3626 kg ha^{-1} resulting in a 23 percent yield increase (Figure 2-3 C).

Grain yield correlated well with values of NDVI collected at early season (Feekes 6 growth stage) (Figure 2-5 A). A linear regression resulted in an R^2 value of 0.69 and a p-value of <0.001 (Figure 2-5 A). Previous studies have shown that NDVI has a relationship with the final yield of winter wheat (Moges, 2004). The values of NDVI also showed a strong correlation with total P uptake at the Feekes 6 growth stage, with an R2 of 0.56 and a p-value of <0.001 (Figure 2-5 B). This strong relation of NDVI with P uptake and grain yield in response to P fertilizer may indicate a valid approach to using relative NDVI values to estimate critical soil test P levels. Early-season NDVI measurements used to estimate critical STP values could be useful as a quick and less costly P response assessment method, provided that a co-founding response to other nutrients such as N is removed.

Critical soil test phosphorus (STP) levels for the different extraction methods using the linear plateau (LP) and cate nelson (CT) statistical methods using early-season NDVI values to characterize wheat response are shown in Table 2-5. Overall the resulting critical values were larger with the LP model than with the CN model in our dataset and with the range of STP values across locations. For the M3-COL method, the LP and CN models resulted in critical values that differed by 7.3 mg of P kg^{-1}, with the LP method having a higher level (Table 2-5).

Current soil test interpretation guidelines with the M3-COL method suggest a critical value of 20 mg kg^{-1} for all crops in Kansas (Leikam, 2003). For the B1 method, the LP and CN models resulted in an R^2 of 0.74 and 0.77, respectively, and critical values of 21.2 and 12.9 mg of P kg$^{-1,}$ respectively (Table 2-5). It is important to note that soils with a pH greater than 7.45 were removed for the determination of critical levels using the B1 soil test method. For the Olsen method, the LP and CN models resulted in an R^2 of 0.50 and 0.90, respectively, and critical values of 15.1 and 8.9 mg of P kg$^{-1,}$ respectively (Table 2-5).

For the H3A method, the LP and CN models resulted in an R^2 of 0.67 and 1.0, and critical values of 25.5 and 11.7 mg of P kg^{-1}, respectively (Table 2-5). Due to challenges with this soil test P method under high soil pH conditions, all soils above 7.45 were removed before determining the critical levels. For the Resin method, the LP and CN models resulted in an R^2 of 0.59 and 0.82, respectively, and critical values of 19.0 and 10.4 mg of P kg$^{-1,}$ respectively (Table 2-5).

The M3-ICP method resulted in an R^2 of 0.72 and 0.89 for the LP and CN statistical models, respectively (Table 2-5). The critical values established by the LP and CN models were 34.9, and 25.6 mg of P kg^{-1}. Results from our study indicated that even though the same extractant is used, the use of ICP for P determination resulted in larger STP critical values, and these methods cannot be used interchangeably for fertilizer recommendations.

The estimation of STP critical levels using relative yield as crop response is shown in Table 2-5. For the M3-COL method, the estimated STP critical values were 25.5 and 17.9 with the LP and CT models, respectively; these values were similar to those found using NDVI (Table 2-5). DeLong (2006) found similar results for critical STP values that suggest the critical value using the M3-COL for winter wheat to be around 25 mg P kg^{-1}. For the B1 method, values were also similar to those estimated using early-season NDVI. Similar values were found by Sucunza and Gutierrez (2018) when using the B1 method for winter wheat, with a critical level of 19 mg of P kg^{-1} at the 90% sufficiency level for their study. For the Olsen method, critical STP levels were 12.2 and 9.5 mg kg^{-1} using the LP and VN statistical models, respectively. Khan (2018) found critical values of 14 and 16 mg of P kg^{-1} using the Olsen method for winter wheat with a sampling depth of 0-20 cm. Tang (2009) found that critical levels for winter wheat using the Olsen method, with values ranging from 12.5, 16.4, and 17.3 mg of P kg^{-1} using a sampling depth of 20 cm. Overall the critical levels found in our study are in agreement with previous research for winter wheat.

Critical values for the H3A extraction were 26.4 and 14.5 using the LP and CN models (Table 2-5). Field calibration of the H3A method is scarce and, to our knowledge, non-existent for winter wheat. The Resin method resulted in critical levels 19.6 and 11.9 mg kg-1 using the LP and CN models (Table 2-5). In general, relative yield followed similar trends to that of NDVI collected at the Feekes 6 growth stage. Overall the models using NDVI resulted in a higher R^2 in our dataset.

Conclusions

In general, the B1, Olsen, H3A, Resin, and M3-ICP methods correlated well with the M3-COL extraction method. The H3A method had the weakest fit with the M3-COL method which indicates that it could be extracting P in the soil from different pools. Soil test method relationships with the M3-COL were lost when soil carbonates increased and affected the B1 and H3A methods which extracted significantly less P in these soils. The poor predictive performance of B1 and H3A when calcareous soils were included required the removal of locations with calcareous soils to allow fitting our statistical models. The Olsen and Resin methods did not show a negative effect from high pH and carbonates. In general, winter wheat was highly responsive to P fertilization in Kansas soils when soil test P values were low. Predicting how this crop responds to moderate soil test values is heavily dependent upon which soil test method is being used. In general, the M3-ICP method had the highest soil test P critical value, followed by the M3-COL, H3A, B1, Resin, and Olsen soil test methods in decreasing order. When using the B1 and H3A methods, correlation to crop response was affected by pH and soil carbonates, and these methods should be avoided for fields with these characteristics. However, the methods evaluated in this study can be used successfully to predict potential wheat response to P fertilizer in Kansas soils that exhibit normal properties such as neutral pH or low calcium carbonates.

Results of this study show that using the appropriate soil test method is a vital tool to estimate agronomic wheat response to fertilizer. Knowing how these test methods are affected by variations in soil types, particularly changes in pH and carbonate concentrations, is essential when selecting a soil test method. This could be increasingly important in the case of a site-specific management setting in production agriculture. Sampling methods in these systems are designed to sample areas of the field that are similar or specific points of the field. This sampling scheme could result in large changes in soil characteristics between samples that would usually go unnoticed when whole field composite samples are collected and mixed. Equally as important is knowing how the selected soil test compares to crop response. Each method needs to be calibrated in order to have the best performance in a production setting. Results from this study provide estimated STP critical values for various methods and soils in the wheat-producing regions of Kansas.

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Tables and Figures

Table 2-1. Study locations, soil classification, and tillage system information. Wheat variety, planting dates, and row spacing.

Location	Year	County	Soil Series	Sub Group†	Variety	Tillage System‡	Planting Date	Row Spacing
								cm
1	2019	Barton	New Cambria	C. Haplustolls	Larry	CT	10/3/2018	25
2	2019	Barton	Harney	Ty. Argiustolls	Larry	CT	10/3/2018	25
3	2019	Franklin	Woodson	A. Argiaquolls	Everest	NT	11/21/2018	19
4	2019	Gove	Ulysses	To. Haplustolls	Oakley CL	CT	9/29/2018	25
5	2019	Gove	Ulysses	To. Haplustolls	Oakley CL	CT	9/29/2018	25
6	2019	Gove	Ulysses	To. Haplustolls	Oakley CL	CT	9/27/2018	25
7	2019	Gove	Buffalo Park	A. Haplustepts	Oakley CL	CT	9/27/2018	25
8	2019	Riley	Smolan	P. Argiustolls	Everest	NT	11/14/2019	19
9	2019	Reno	Ost	U. Argiustolls	Everest	CT	10/31/2018	19
10	2020	Ellsworth	Hord	C. Haplustolls	Everest	NT	10/13/2019	25
11	2020	Ellsworth	Hord	C. Haplustolls	Everest	NT	10/13/2019	25
12	2020	Gove	Keith	A. Argiustolls	Avery	CT	9/22/2019	25
13	2020	Gove	Ulysses	To. Haplustolls	TAM 114	CT	9/24/2019	25
14	2020	Gove	Ulysses	To. Haplustolls	TAM 114	CT	10/3/2019	25
15	2020	Gove	Ulysses	To. Haplustolls	TAM 114	CT	9/28/2019	25
16	2020	Marion	Ladysmith	P. Ud. Argiustolls	WB4303	NT	9/25/2019	25
17	2020	McPherson	Goessel	U. Haplusterts	CP7909	NT	9/30/2019	25
18	2020	McPherson	Goessel	U. Haplusterts	CP7909	NT	9/30/2019	25
19	2020	Riley	Ivan and Kennebec	C. Hapludolls	Larry	NT	10/25/2019	19
20	2020	Riley	Wymore	Aq. Argiudolls	Tatankta	NT	10/25/2019	19
21	2020	Reno	Ninnescah	F. Endoaquolls	Everest	CT	11/1/2019	19
22	2020	Sheridan	Keith	A. Argiustolls	TAM 114	CT	10/1/2019	25
23	2020	Sheridan	Keith	A. Argiustolls	TAM 114	CT	10/1/2019	25
24	2020	Smith	Harney	T. Argiustolls	Tatankta	NT	10/14/2019	19

† C, Cumulic; Ty, Typic; A, Abruptic; To, Torriorthentic; P, Pachic; U, Udic; A, Aridic; Ud, Udertic; Aq, Aquertic; F, Fluvaquentic;
‡CT, Conventional Tillage; NT, No Till.

Table 2-2. Soil test information from samples collected before wheat planting and fertilizer application at 24 locations in Kansas where the response to phosphorus fertilizer was evaluated in 2019-2020. Samples collected at the 0-15 cm depth.

Location	SC†	pH	OM	P	K	Ca	Mg	Na	CEC
	mg kg^{-1}		g kg^{-1}	- - - - - - - - - - - - - - - - - mg kg^{-1} - - - - - - - - - - - - - - - -					(meq/100g)
1	2.4	6.5	22.5	48	347	1861	337	2.5	13.0
2	11.4	7.7	24.8	18	249	4912	85	4.9	25.9
3	3.0	6.2	35.0	9	127	2425	307	20.0	15.1
4	0.0	6.9	25.3	17	629	2829	318	4.4	18.4
5	1.6	6.7	26.0	21	604	2655	327	3.9	17.5
6	10.5	7.6	20.3	18	595	5166	206	4.6	29.1
7	2.7	6.4	25.3	20	528	2263	311	2.2	15.2
8	3.1	5.7	31.3	16	283	2367	427	15.8	16.1
9	6.1	7.7	20.3	29	191	4060	49	6.6	21.2
10	1.1	6.2	22.8	9	304	1586	239	16.3	10.7
11	0.7	5.6	22.5	26	270	1321	234	12.3	9.3
12	0.5	7.2	23.0	20	712	3042	547	17.3	21.6
13	2.5	8.3	23.3	18	702	5336	239	15.8	30.5
14	0.1	7.3	25.3	14	613	3100	401	13.5	20.4
15	2.1	7.6	23.5	16	770	3849	364	13.3	24.3
16	0.1	6.5	25.3	14	227	3404	475	43.8	21.7
17	0.2	5.4	27.3	13	153	1762	315	17.3	11.9
18	0.0	5.6	29.8	12	154	2212	381	25.3	14.7
19	0.0	6.2	28.8	11	176	2077	292	24.0	13.3
20	0.0	6.6	28.3	9	294	2500	395	15.3	16.6
21	0.0	6.7	18.0	2	78	749	187	177.5	6.3
22	0.5	6.8	24.0	20	632	2503	394	14.3	17.4
23	0.1	6.8	24.3	29	626	2838	395	14.0	19.1
24	0.2	5.6	28.3	41	427	1883	339	16.3	13.4

† SC, Soil Carbonates; OM, Organic Matter; P, Mehlich-3 colorimetric phosphorus; K, Potassium; Ca, Calcium; Mg, Magnesium; Na, Sodium; CEC, Cation Exchange Capacity

Table 2-3. Significance of F values for fixed effect of phosphorus fertilizer application rates on NDVI, biomass, and phosphorus uptake at the Feekes 6 growth stage and grain yield at harvest.

Location	Sampling at Feekes 6			Yield
	NDVI	Biomass	Phosphorus uptake	
	- - - - - - - - - - - - - - - - - - - p > F	- - - - - - - -	- - - - - - - -	- - - - - - - -
1	0.843	0.507	0.351	0.540
2	<0.001	<0.001	0.005	<0.001
3	0.010	0.021	--†	--
4	<0.001	0.013	--	--
5	0.014	0.076	--	--
6	<0.001	<0.001	<0.001	<0.001
7	<0.001	0.011	<0.001	0.01
8	<0.001	<0.001	<0.001	0.008
9	0.721	0.605	0.849	0.809
10	<0.001	0.009	0.004	<0.001
11	0.014	0.024	0.020	0.049
12	0.017	0.373	0.035	0.039
13	0.072	0.095	0.056	0.814
14	--	--	--	0.019
15	0.211	0.186	<0.001	0.025
16	0.005	0.046	0.007	0.260
17	<0.001	<0.001	<0.001	<0.001
18	<0.001	<0.001	<0.001	0.002
19	0.094	0.046	0.106	0.148
20	<0.001	<0.001	<0.001	<0.001
21	--	--	--	0.327
22	0.251	0.683	0.084	0.668
23	0.922	0.816	0.198	0.049
24	--	--	--	0.006
Across Locations	<0.001	<0.001	<0.001	<0.001

†Samples were not collected

Table 2-4. Linear regression coefficients for the Mehlich-3 test with ICP compared to Bray-1, Olsen, H3A, and Resin methods.

Test Method	Intercept	Slope	R^2	p > F
Bray-1	-4.81	0.765	0.91	<0.001
Olsen	0.815	0.378	0.77	<0.001
H3A	-3.33	0.648	0.65	<0.001
Resin	-1.48	0.529	0.86	<0.001

Table 2-5. Statistical models (linear plateau and Cate-Nelson) used to identify critical soil test P levels for six different soil test phosphorus methods. Analysis completed using relative NDVI values at Feekes 6 and relative grain yield as wheat response variable. †

	Linear Plateau			Cate-Nelson		
STP Method	Critical STP§	Equation	R^2	Critical STP	P > F	R^2
	mg kg^{-1}			mg kg^{-1}		
- - - Relative NDVI at Jointing - - -						
Mehlich 3 Col	26.1	0.96 + 0.020 * (x - 26.1) * (x <= 26.1)	0.77	18.8	<0.001	1.00
Bray 1	21.2	0.96 + 0.026 * (x - 21.2) * (x <= 21.2)	0.74	12.9	0.007	0.77
Olsen	15.1	0.96 + 0.034 * (x - 15.1) * (x <= 15.1)	0.50	8.9	<0.001	0.90
H3A	25.5	0.97 + 0.016 * (x - 25.5) * (x <= 25.5)	0.67	11.7	<0.001	1.00
Resin	19.0	0.97 + 0.026 * (x - 19.0) * (x <= 19.0)	0.59	10.4	<0.001	0.82
Mehlich 3 ICP	34.9	0.97 + 0.019 * (x - 34.9) * (x <= 34.9)	0.72	25.6	<0.001	0.89
- - - Relative yield- - -						
Mehlich 3 Col	25.5	0.93 + 0.016 * (x - 25.5) * (x <= 25.5)	0.51	17.9	<0.001	0.90
Bray 1	26.8	0.95 + 0.014 * (x - 26.8) * (x <= 26.8)	0.56	16.3	0.001	0.87
Olsen	12.2	0.91 + 0.040 * (x - 12.2) * (x <= 12.2)	0.51	9.5	<0.001	0.90
H3A	26.4	0.95 + 0.012 * (x - 26.4) * (x <= 26.4)	0.55	14.5	0.001	0.88
Resin	19.6	0.95 + 0.018 * (x - 19.6) * (x <= 19.6)	0.33	11.9	<0.001	0.80
Mehlich 3 ICP	36.7	0.95 + 0.013 * (x - 36.7) * (x <= 36.7)	0.41	26.0	<0.001	0.80

† Locations 2, 6, 9, and 13 were removed from all Bray-1 and H3A analysis due to high pH and calcium carbonate content, affecting correlation.

§ The critical STP value indicated by the model as the point at which responses to P fertilizer stopped.

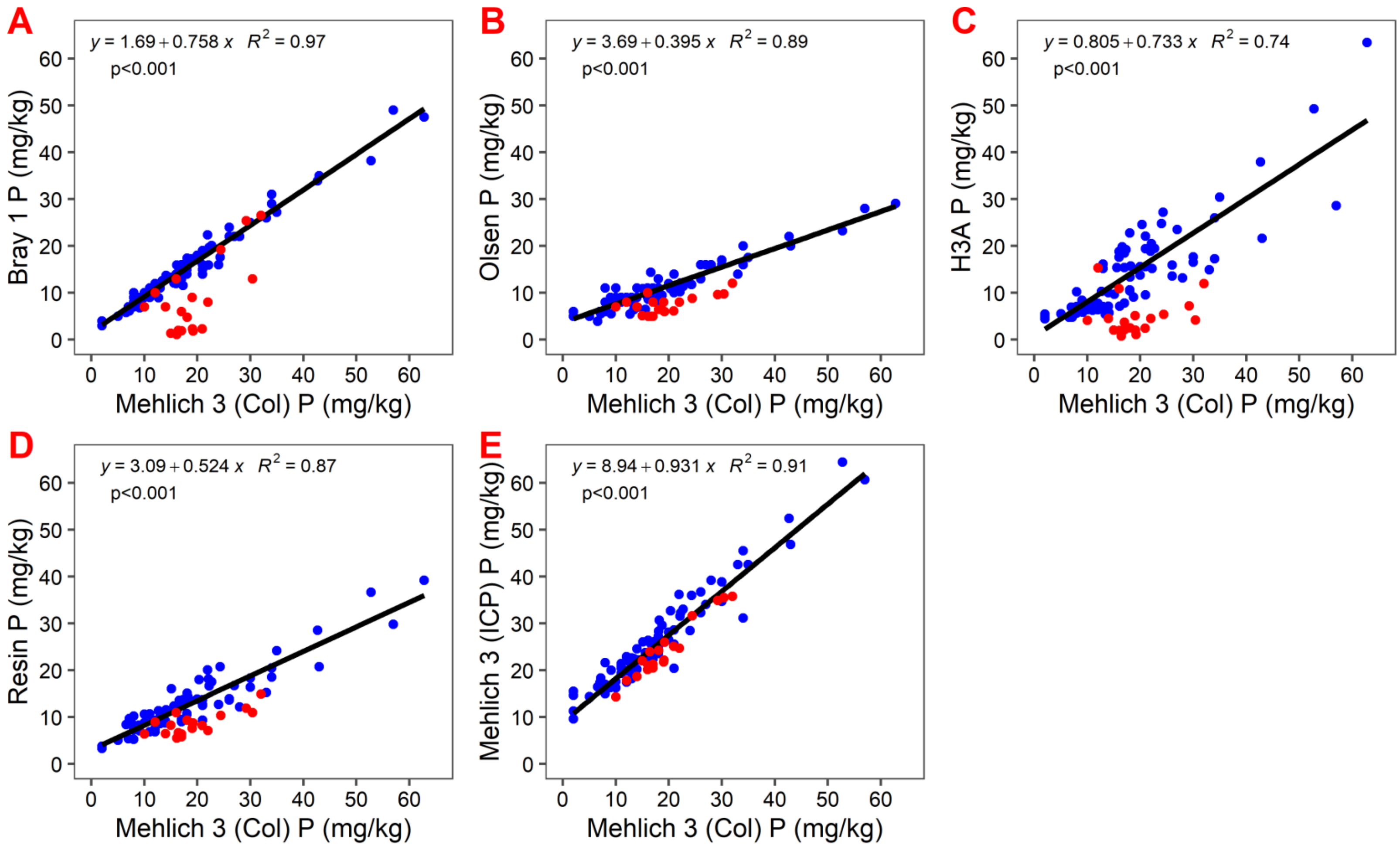

Figure 2-1. Correlation between the Mehlich-3 colorimetric extraction method and Bray-1 (A), Olsen (B), H3A (C), Resin (D), and Mehlich-3 ICP (E). Samples with a pH above 7.45 were removed for the regression analysis (Figure 2-2).

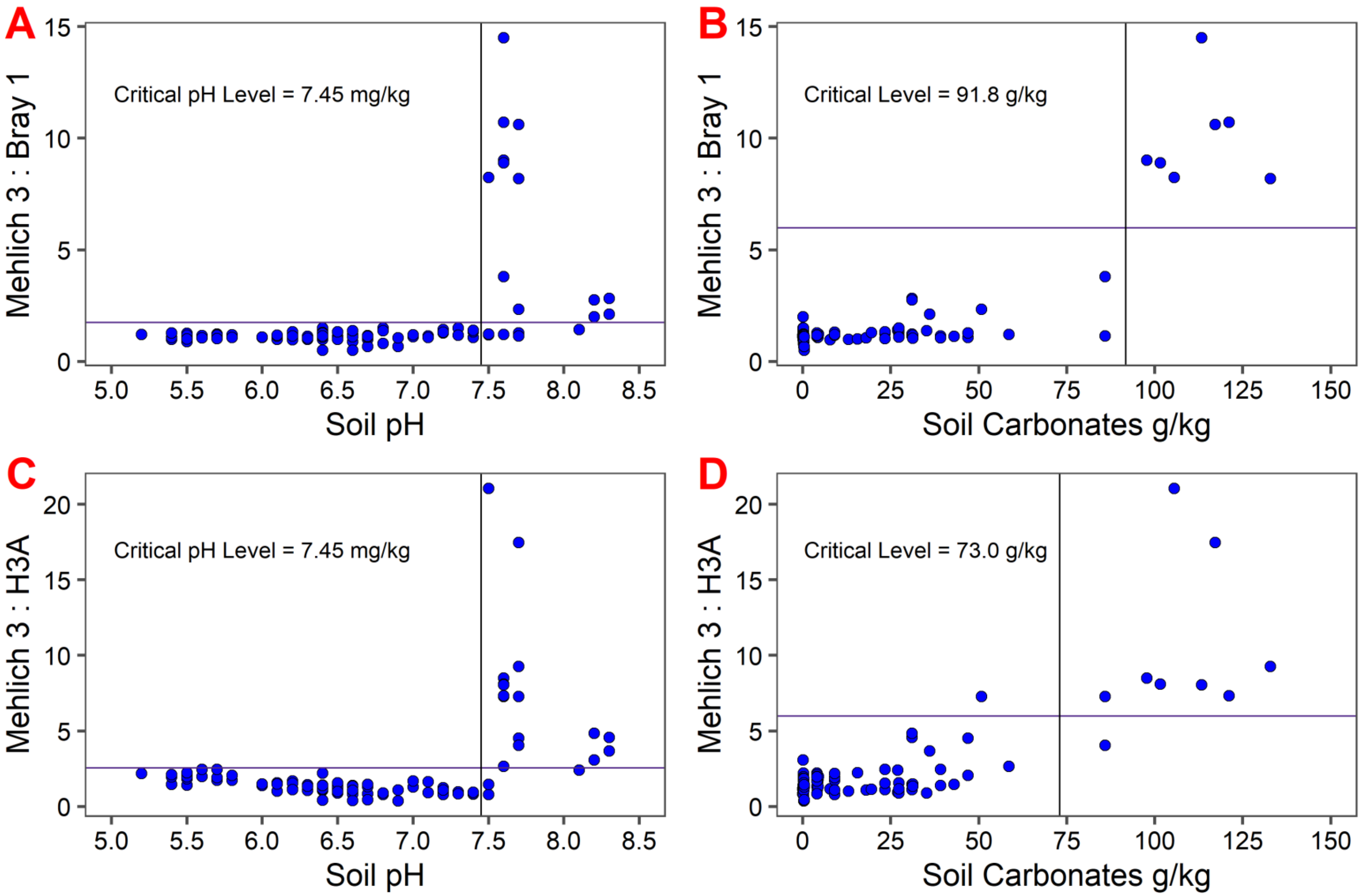

Figure 2-2. Relationship and critical break points between soil pH and the ratios of Mehlich-3 to Bray-1 (A) and Mehlich-3 to H3A (C). Soil carbonates and the ratios of Mehlich-3 to Bray-1 (B) and Mehlich-3 to H3A (D).

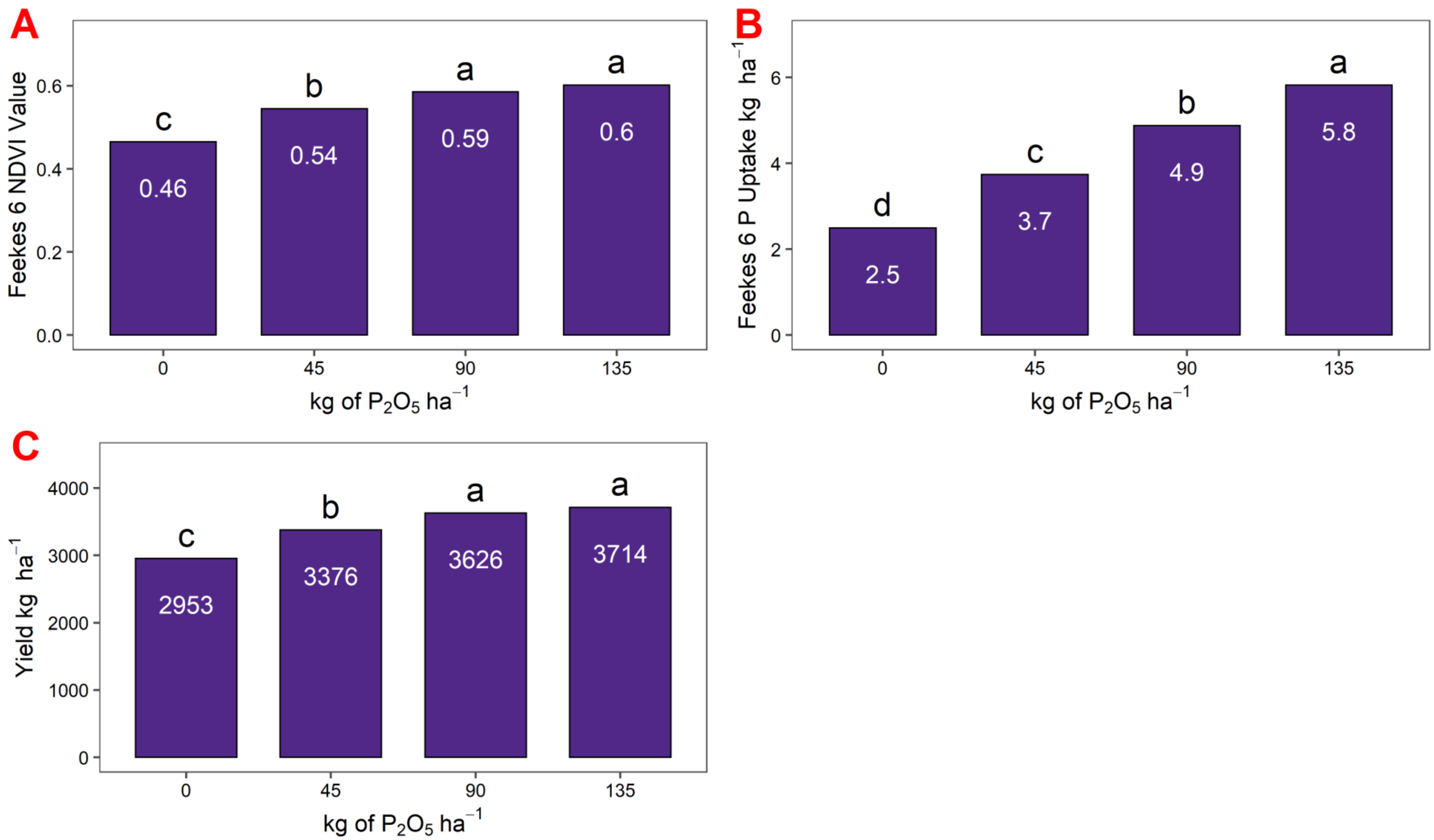

Figure 2-3. Average wheat response to P fertilizer rates across locations during the early vegetative growth stage. Feekes 6 NDVI (A), Feekes 6 P uptake (B) and grain yield (C). Location and blocks were set as random effects in the model and P fertilizer rates as a fixed effect. All P treatments were balanced to 112 kg of N. Means with different letters are statistically different at the 0.05 probability level.

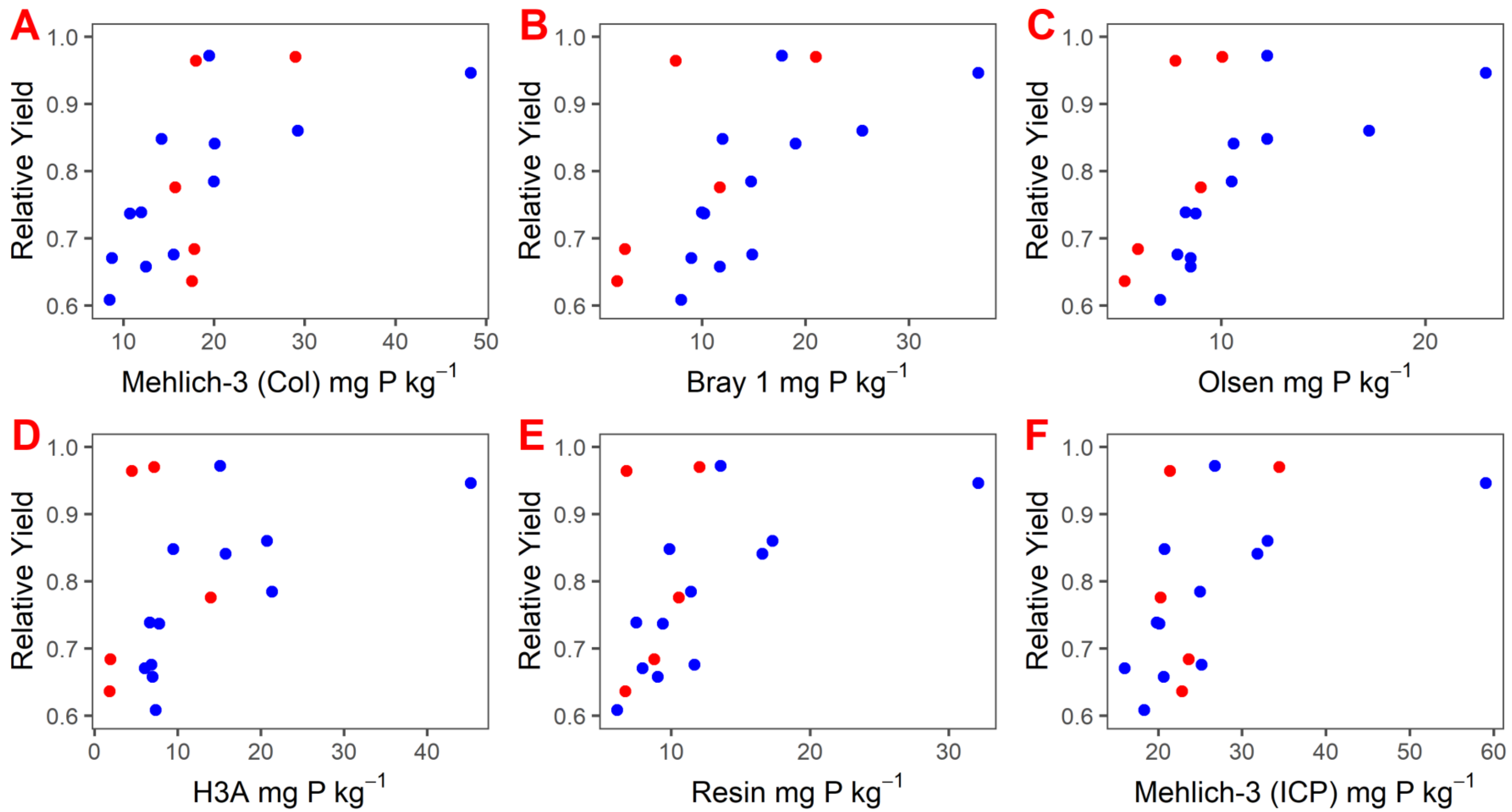

Figure 2-4. Effects of soil pH on correlation with relative yield by soil test phosphorus methods. Soils with a pH >7.45 are colored red. Mehlich-3 Col (A), Bray-1 (B), Olsen (C), H3A (D), Resin (E) and Mehlich-3 ICP.

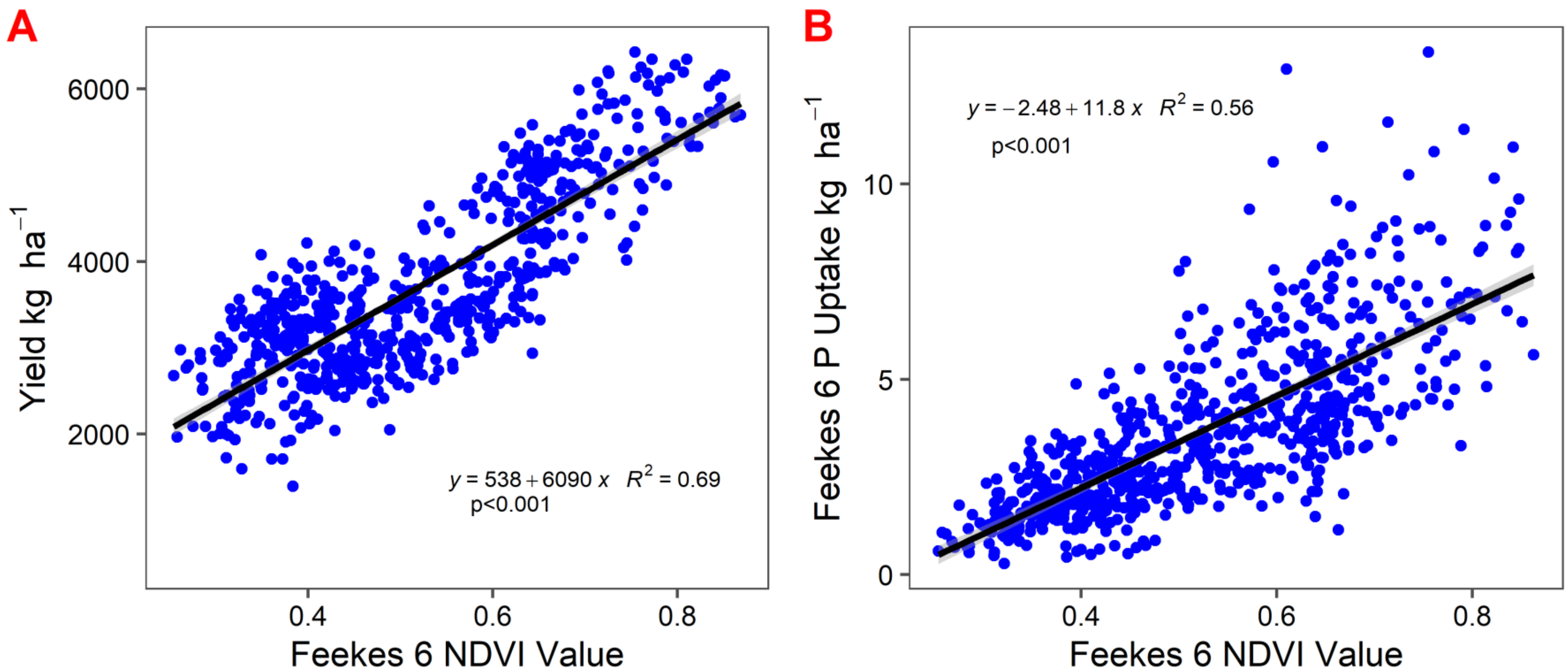

Figure 2-5. Relationship between Feekes 6 NDVI and grain yield (A) and the relationship between Feekes 6 NDVI and Feekes 6 P uptake (B). A linear regression was applied to both comparisons.

Chapter 3 - Evaluation of soil test methods and plant tissue analysis to assess sulfur nutritional status in winter wheat

Abstract

Identifying how winter wheat will respond to sulfur (S) fertilization through the use of soil test S (STS) methods has been a challenge across Kansas soils. The objective of this study was to evaluate diagnostic tools used to manage S and included soil test S extraction methods and plant S nutritional status using different S fertilizer sources and rates. Sulfur response trials were established at 24 Kansas locations during two years (2019 and 2020). Fertilizer rate treatments included a control with 112 kg of nitrogen (N) ha^{-1} and 45 kg of P_2O_5 (P) ha^{-1} using urea and mono ammonium phosphate. Sulfur treatments included three rates, 0, 11, and 45 kg of S ha^{-1} applied using ammonium sulfate AMS (21-0-0-24S), and two additional S fertilizer sources applied at 11 kg of S ha^{-1} using Micro-Essentials SZ “MESZ” (12-40-0-10S-1Zn) and elemental sulfur (0-0-0-90S), for a total of five treatments. Nitrogen was balanced accordingly for all treatments. All P and S and 50 kg of N ha^{-1} of N were broadcast in the fall, followed by a 62 kg N ha^{-1} topdress application at Feekes 5. A randomized complete block design was used for the experiment with four replications. Soil samples were taken by replication at depths of 0-15, and 0-60 cm and analyzed using four different STS methods. At Feekes 6 (jointing) normalized difference vegetation index (NDVI) and biomass samples were taken. At heading, flag leaf tissue samples were taken as well as biomass samples at Feekes 11.2 (soft dough); grain yield was also measured at harvest. Both biomass and grain samples were analyzed for S and N concentrations. Results indicate STS methods varied in their correlation with one another. The most closely correlated methods were calcium phosphate extraction and the ammonium acetate extraction, which resulted in an R^2 of 0.96. Although yield and Feekes 6 NDVI showed little

response to S fertilizer treatments, the majority of tissue samples showed an increase in S concentration as rates of S increased as well as a decrease in S concentration when elemental S was used as the source. The median internal efficiency was found to be 425 kg grain/ kg S uptake. Overall, diagnostics tools that are used to evaluate S response in wheat varied greatly with one another making it difficult to clearly identify similar responses between diagnostics tools.

Abbreviations: N, nitrogen; S, sulfur; STS, soil test sulfur; normalized difference vegetation index, NDVI.

Introduction

Sulfur (S) deficiency in winter wheat has become more common in recent years. In the past, crops located in coal-burning regions received sufficient S directly from the atmosphere (Wainwright, 1984). However, as the Clean Air Act was implemented, much of this sulfur has been removed from the atmosphere for environmental reasons (Popp, 2003). This means the amount of S accumulated from the atmosphere that had previously been available to plants has been decreasing in the past decades. Even though this may seem like a small input compared to the demand of today's crops, it is important to note that a typical crop does not need a substantial amount of S compared to other macronutrients (Hagin, 1982). Slight reductions in sulfur deposits from the atmosphere can play a big role in the supply of S throughout the growing season. In addition to this, the typical fertilizer used in production agriculture has changed. Concentrations of S in commercial fertilizers have decreased significantly in recent years with improvements in fertilizer processing (Scherer, 2001). This reduction in sulfur in typical fertilizers containing other macronutrients has also led to a decrease in sulfur inputs into crop production systems.

Identifying the need for additional S as fertilizer has typically been done by using profile soil sampling, much like what is used for nitrogen. Both nitrogen and S have similar cycles in the soil, and soil test recommendation methods are similar. Applying the correct amount of this nutrient to obtain an economic return is the main driver of S fertilization in crops like winter wheat. Identifying how efficiently the crop utilizes this nutrient and how effectively the crop can uptake it from the soil is key to understanding how to manage this nutrient. There are different soil test S (STS) methods being used to identify S-deficient soils in Kansas. This makes it a challenge when determining where a response could be seen. This is especially apparent if an STS method has not been well correlated with crop response.

How well a crop responds to sulfur depends largely on which form of sulfur is in the soil. S in the sulfate form is readily available to the plant, while other sources such as S in organic matter need to be mineralized to become plant available (Scherer, 2009). This same trend is seen in many of the fertilizers that are typically applied to a growing crop. Although many fertilizers contain S in the sulfate form, others contain S in the elemental form, which requires oxidation before becoming plant available (Wainwright, 1984). This process is dependent upon soil moisture as well as temperature because sulfur oxidation is largely performed by microorganisms (Wainwright, 1984). With the transition to no-till, soil temperatures are typically lower due to previous crop residue providing a temperature buffer in the spring (Ruiz Diaz, 2018). In addition to this, wheat is grown during the cooler and drier part of the year in Kansas. How much elemental sulfur is converted to the plant-available sulfate form is difficult to determine. The objective of this study was to evaluate soil test extraction methods for S as well as plant S nutritional status using different S fertilizer sources and rates and determine how responses relate to crop response.

Materials and methods

Treatments, Experiment Design, and Implementation

A total of 24 locations were established in 2019 and 2020 (Table 3-1). These locations were located on farmer's fields (18 locations) and university experiment fields (6 locations). Locations on farmer's fields were sowed by the farmer, with all other field operations completed with research equipment. Field operations on university experiment fields were completed with university equipment. Research spraying equipment included a John Deere Gator with a 6-meter boom sprayer for wheat weed herbicides at Feekes 6 and a 3.6-meter boom CO2 sprayer for fungicides at heading. For harvest, a 1.4-meter Wintersteiger combine was used. Either conventional tillage or no-till was used for all locations. Conventional tillage typically included 3 to 5 passes of tillage using a field cultivator prior to sowing. Common crop rotations were used at each location and varied depending on the region and each farming operation. The individual plot size was 13.7 m in length and 1.8 m in width, except location 21, with a length of 13.7 m and a width of 3.0 m. This change in plot area was used so the site could be planted to corn the next year. Row spacing depended on equipment but was either 25 cm or 19 cm (Table 2-1). Wheat varieties were selected by regional performance or by the farmer based on characteristics of production systems and soils (Table 3-1).

The experimental design used was a randomized complete block design with four replications. Five fertilizer treatments consisted of three rates of sulfur fertilizer (0, 11, and 45 kg S ha^{-1}) using ammonium sulfate (AMS) (21-0-0-24S), and two treatments consisting of MESZ and elemental S fertilizer; both applied at 11 kg S ha^{-1}. All S fertilizer treatments were applied before sowing. All treatments received a uniform application of 45 kg of P_2O_5 ha^{-1} using mono ammonium phosphate (11-52-0) before sowing. Nitrogen from all fertilizer sources were balanced to a rate of 112 kg of N ha^{-1} of N for each treatment using Urea (46-0-0), except for locations 2, 6, 7, and 24, which received a total of 157 kg of N ha^{-1}. This increase in N was due to the producer applying pre-plant N using UAN or Urea prior to location establishment. Roughly half of the N fertilizer was broadcast-applied in the fall within a week before sowing (in combination with the S treatments and P fertilizer). The remaining N fertilizer was broadcast as a top-dress in the spring at Feekes 5.

Soil Sampling and Analysis

Composite soil samples of 10-12 cores were collected by block prior to sowing and fertilizer application at the 0-15 and 0-60 cm depths. Soil samples were dried at 41^{o} C (5-10 days). After samples were dried, the soil was ground to pass through a 2 mm sieve. Soil samples from the 0-15 cm depth were analyzed for pH 1:1 (soil:water) (Watson and Brown, 1998) and organic matter percent by loss on ignition (Combs and Nathan, 1998). Samples collected from the 0-60 cm depth were analyzed for nitrate and ammonium using 1N KCl (Gelderman and Beegle, 1998) and for chloride using calcium nitrate (Gelderman et al., 1998). All samples were extracted for S using four methods: calcium phosphate, resin, Mehlich 3, and ammonium acetate. Sulfur was determined with the ICP–OES (Inductively Coupled Plasma Optical Emission Spectrometry) for all extraction methods. Due to dry and hard soils, locations 11, 17, and 18 were not sampled at the 0-60 cm depth.

Early Season NDVI and Plant Sampling

Normalized difference vegetative index (NDVI) measurements were taken at the Feekes 6 (jointing) growth stage using a Holland CS-45 (Holland Scientific, Lincoln NE). The sensor senses red light at a wavelength of 670 nm, red-edge light at 730 nm, and near inferred light at 780 nm. For this study, the red and near inferred wavelengths were used to calculate the NDVI for each treatment. The sensor was held at the height of 1.2 meters above the crop canopy, and the reading was collected for the entire length of each plot. At the end of each plot, the average measurement was recorded. NDVI values for locations 14, 21, and 24 were not collected due to inclement weather at the time of sampling.

Biomass samples were collected at the early vegetative (Feekes 6) stage. Samples were collected from 2 rows at a length of 76 cm. All biomass samples were collected from the back 3 meters of each plot, which was excluded from future measurements. Samples were oven-dried at 60^{o} C for 5-7 days and weighed to measure dry biomass. Dry samples were ground through pass through a 2 mm screen, and then digested for sulfur (S) using a nitric perchloric digest (Gieseking et al., 1935). Sulfur concentration was determined by ICP–OES (Inductively Coupled Plasma Optical Emission Spectrometry). Sulfur uptake was calculated by multiplying plant tissue concentration by biomass weight.

Late Season Plant Sampling and Yield

Flag leaf tissue samples were collected between the Feekes 10.1 and Feekes 10.5 growth stages. For this sampling timing, 50 flag leaves were collected from each plot and then oven-dried at 60^{o} C for 3-5 days. Dry samples were then ground to pass through a 2 mm screen. Samples were then digested for sulfur (S) using a nitric perchloric digest (Gieseking et al., 1935). Plant biomass samples were collected at the late reproductive (Feekes 11.2 -Soft dough) stage. This stage is considered the point of maximum nutrient uptake in wheat. The sample was taken from wheat rows that had not already been sampled previously at the Feekes 6 stage. Sampling protocols were the same as described for early season biomass sampling, except for chopping and weighing wet samples and then collecting a sub-sample for moisture determination. The subsamples were oven-dried at 60 ^{o}C for 5-7 days to estimate moisture. The original whole plant biomass was corrected for moisture and expressed on a dry weight basis. Subsamples were ground to pass through a 2 mm screen and digested for sulfur (S) using a nitric perchloric digest (Gieseking et al., 1935). Sulfur concentration was determined by ICP–OES (Inductively Coupled Plasma Optical Emission Spectrometry). Biomass samples were also analyzed for nitrogen (N) using the dry combustion method (LECO) (Matsunaga & Shiozaki, 1987).

Grain yield was determined by harvesting the center 1.4 meters of each plot using a plot combine with a platform header. Locations 3, 4, and 5 were not harvested due to hail damage. Subsamples were taken from each plot and tested for moisture and test weight. Grain subsamples were then digested for sulfur (S) using a nitric perchloric digest (Gieseking et al., 1935) and sulfur concentration determined by ICP–OES. Results were used to determine S removal in the harvest grain.

Nutrient Balance Calculations

Sulfur use efficiency was evaluated with the partial nutrient balance (PNB), agronomic efficiency (AE), apparent recovery efficiency (AR), and internal efficiency (IE). The PNB is typically used as a long-term indicator of fertilizer usage trends (Fixen, 2015). The equation uses the nutrient content of the harvested portion of the crop divided by the amount of fertilizer applied. The PNB was estimated for S as well as N to evaluate the effect of S fertilizer treatments on N PNB. Agronomic efficiency was calculated by taking the yield with S fertilizer application minus the yield without S fertilizer and divided by the amount of S applied with fertilizer (Fixen, 2015). The apparent recovery efficiency by difference was calculated by using the total plant S uptake (total biomass) (Fixen, 2015). This indicator would show how much S the crop accumulated compared to how much was applied as fertilizer while taking into account the amount of S accumulated in the no S treatment. Internal efficiency was estimated by dividing grain yield by whole plant S uptake at Feekes 11.2.

Statistical Analysis

For regression analysis, the geom smooth function was used from ggplot in R version 4.0.3 (R Core Team, 2020). Stat poly eq was used to extract the formula of the regression. Regressions were all compared to the calcium phosphate method as it is currently the standard for use method at Kansas State University and seen as a control for all the methods. Statistical analysis was completed using linear mixed-effects models (lme4 package) in R version 4.0.3 (R Core Team, 2020); blocks were included as random factors in the model**.** For analysis across locations, block and locations were considered as random effects in the model. Statistical significance was set at alpha = F 0.05.

Results and discussion

Sulfur Soil Test Methods Comparison

Soil test S levels from samples collected at the 0-60 cm varied from 1.3 to 152 mg of S kg^{-1} using a calcium phosphate extraction (Table 3-2). When comparing the relationship of the CP method to other methods, the highest R^2 (0.96) was with the Ammonium Acetate (AA) extraction (Table 3-3 and Figure 3-1). The lowest R^2 was with the Mehlich-3 (M3) method with an R^2 of 0.31 (Table 3-3 and Figure 3-1). The Resin (R) method had an R^2 of 0.76 when compared with the CP method. The M3 and R extraction methods showed the lowest R^2 and large variability in values (Table 3-3). The relationships between the other three methods are shown in Table 3-3. When comparing the R and AA method, an R^2 of 0.75 was calculated, which was similar to the R and CP methods (Table 3-3). The relationship between the M3 to AA showed a lower R^2 of 0.32 with a general trend but scattered points (Table 3-3).

Overall, the CP and AA methods showed the best correlation compared to the other STS methods. The M3 method, in general, seemed to extract more sulfur than other methods when looking at the dense points that run along the y axis. The R method generally correlated well with other methods but also had significant variability. One reason these methods have variability when comparing test methods to one another could be due to different methods extracting S from different pools of S in the soil, such as organic or inorganic S.

Early season tissue S and NDVI

Out of the 21 locations where NDVI was measured at jointing, two showed significant treatment effects at the $\alpha = 0.05$ statistical level, and analysis across locations showed no statistically significant response to S fertilizer treatments (Table 3-4 and Figure 3-2). The locations that were responsive had a starting sulfur level between 2.1 and 2.6 mg of S kg^{-1} (Table 3-2). Furthermore, NDVI values in wheat are generally driven primarily by biomass. Sulfur concentration in the tissue was generally decreased with greater biomass accumulation (Figure 3-5 C). Therefore, production factors affecting wheat biomass at jointing would affect S tissue concentration and should be considered for the interpretation of S nutritional status with the tissue test.

Out of the 18 locations where sulfur concentration was measured in the tissue at jointing, 13 showed a significant response to S fertilizer applications (Table 3-4), and analysis across locations also showed a significant response (Figure 3-2 B). The locations that were responsive had a starting sulfur level between 1.2 and 12.3 mg of S kg^{-1} (Table 3-2). Across locations, the highest tissue S concentration was attained with the 45 kg S ha^{-1} rate. Evaluation of S fertilizer sources showed that the MESZ treatment had the same S tissue concentration as the ammonium sulfate treatment; however, the elemental sulfur was significantly less and similar to the no-S fertilizer control (Figure 3-2). This could be due to low S oxidation during the winter months and limited S availability from elemental S at the time of jointing (Feekes 6) in the spring (Wainwright, 1984).

Late-season flag leaf tissue S, uptake and yield

The most consistent wheat response to S fertilizer treatments was flag leaf tissue S concentration. Of the 20 locations analyzed, 15 showed a significant response (Table 3-4). The locations that were responsive had a starting sulfur level between 1.2 and 12.3 mg of S kg^{-1} (Table 3-2). In the evaluation of S rates across locations, significant increases in flag leaf S were observed at each S fertilizer rate (Figure 3-2 C). The control treatment had the lowest S concentration of 2.43 g of S kg^{-1}, and the highest rate of 45 kg ha^{-1} S fertilizer rate had the highest flag leaf S concentration of 3.0 g kg^{-1} (Figure 3-2 C). When comparing S fertilizer sources, elemental S had the lowest flag leaf S concentration of 2.47 g kg^{-1} with no statistical difference from the non-fertilized control. In comparison, the MESZ source was similar to the ammonium sulfate source (Figure 3-2 C). Overall, the addition of S fertilizer increased flag leaf S concentration by 23 percent compared to the control (Figure 3-2 C). Flag leaf S concentration was related to early-season tissue S concentration (Figure 3-5 A). However, a low R^2 value indicates a weak relationship, and early-season tissue S may not be a good predictor of late-season S nutritional status in wheat.

There were 20 locations sampled for S uptake at the Feekes 11.2 growth stage. Out of these locations, 8 locations showed response due to S fertilizer treatments (Table 3-4). The locations that showed an S uptake response to S fertilizer had S soil levels between 1.2 and 7.0 mg of S kg^{-1} (Table 3-2). Analysis of sulfur fertilizer rates across locations showed a significant increase in S uptake at each S fertilizer rate (Figure 3-3 A). The control treatment had the lowest

S uptake of 7.3 kg of S ha^{-1}, and the highest rate of 45 kg ha^{-1} S fertilizer rate had the highest S uptake of 9.9 kg of S ha^{-1} (Figure 3-3 A). When comparing S fertilizer sources, there were no significant differences between any of the three S sources applied at 11 kg ha^{-1} (Figure 3-3 A). Overall, the addition of S as ammonium sulfate increased S uptake by 36 percent compared to the control (Figure 3-3 A).

Of the harvested locations, three showed a significant effect of the S fertilizer treatments (Table 3-3 and 3-4). The responsive locations to S fertilizer had soil S levels ranging from 2.1 to 3.0 mg of S kg^{-1} (Table 3-2). Across all locations, neither S rate nor S source had a significant impact on yield (Table 3-4 and Figure 3-3 B). Sulfur removal with harvested grain showed similar trends as yield (Table 3-3 and Figure 3-3 C). Across locations, the rate of S fertilizer had no effect on grain S removal, but the source of S fertilizer did have an effect (Table 3-3 and Figure 3-3 C). The source with the lowest S removal was elemental S at 4.3 kg of S ha^{-1}, and the highest S removal with the grain was seen with the MESZ fertilizer source at 4.7 kg of S ha^{-1} (Figure 3-3 C).

Sulfur nutritional status in the plant

Results across locations indicate that partial nutrient balance (PNB) for S was lowest at the 45 kg S ha^{-1} rate with differences between sources of S fertilizer being seen between MESZ and elemental S with elemental S having a lower PNB (Figure 3-4 A). The rate and source of S fertilizer had no effect on the PNB of N (Figure 3-4 B). Results across locations showed no difference in agronomic efficiency between the rates and sources of S fertilizer treatments (Figure 3-4 C). This lack of difference is likely due to larger variations in the data as these graphs do have varying results between sources and rates of S. Across all treatments, the apparent recovery of S rate using AMS was the highest with the 11 kg S ha^{-1} and the lowest with the 45 kg S ha^{-1} rate (Figure 3-4 D). When comparing sources of S, there was no significant differences between sources however a decreasing trend was seen from AMS to MESZ and finally elemental S. Results from our study showed a value of approximately 425 kg grain/ kg S uptake for the median of the data set.

Conclusions

Results from our study showed that the four soil test sulfur methods compared had limited correlation with one another besides the calcium phosphate and ammonium acetate methods. These two could be similar due to these methods extracting S from similar pools of this nutrient in the soil while the other methods could be extracting S from different or more pools of S in the soil. Further research on soil test sulfur methods will need to be done to identify how each method represents crop response in the field.

In early growth, early season NDVI values were driven primarily by wheat biomass, and therefore unless severe S deficiency symptoms are present, it is unlikely that NDVI alone will be a good indicator of S nutritional status. Early-season tissue S levels showed a better relationship with S fertilizer application; however, late-season flag leaf tissue S concentration was the most responsive to changes in S supply from fertilizer application. The poor performance of early-season diagnostic indicators makes the determination of S nutritional status challenging for conditions in our study with no severe S deficiencies.

Grain yield response to S fertilization was limited, but response variables including tissue S concentration at the Feekes 6 stage, flag leaf S concentration, and total S plant uptake indicated statistically significant differences between S fertilizer treatments. Results from our study showed differences in the short-term sulfur availability from different fertilizer sources, with generally lower S supply from elemental S as indicated by tissue test at the Feekes 6 stage and flag leaf S concentration. The process of S oxidation relies on microbial activity, and therefore temperature and moisture availability are critical driving factors for S oxidation and plant availability. Fall planting dates and S fertilizer application (late September to early November) correspond to the beginning of lower precipitation and soil temperature period in the wheat-growing region (Wainwright, 1984). Results from our study suggest that under average winter wheat growing conditions, a preferred S fertilizer source should contain the readily plant available sulfate form. Results also indicate that predicting S response in Kansas is still difficult and needs more research to accurately predict responsive locations.

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Tables and Figures

Table 3-1. Study location, soil classification, and tillage system information. Wheat variety and sowing dates.

			Soil classification				
Location	Year	County	Series	Subgroup†	Variety	Tillage System‡	Sowing Date
1	2019	Barton	New Cambria	C. Haplustolls	Larry	CT	10/3/2018
2	2019	Barton	Harney	Ty. Argiustolls	Larry	CT	10/3/2018
3	2019	Franklin	Woodson	A. Argiaquolls	Everest	NT	11/21/2018
4	2019	Gove	Ulysses	To. Haplustolls	Oakley CL	CT	9/29/2018
5	2019	Gove	Ulysses	To. Haplustolls	Oakley CL	CT	9/29/2018
6	2019	Gove	Ulysses	To. Haplustolls	Oakley CL	CT	9/27/2018
7	2019	Gove	Buffalo Park	A. Haplustepts	Oakley CL	CT	9/27/2018
8	2019	Riley	Smolan	P. Argiustolls	Everest	NT	11/14/2019
9	2019	Reno	Ost	U. Argiustolls	Everest	CT	10/31/2018
10	2020	Ellsworth	Hord	C. Haplustolls	Everest	NT	10/13/2019
11	2020	Ellsworth	Hord	C. Haplustolls	Everest	NT	10/13/2019
12	2020	Gove	Keith	A. Argiustolls	Avery	CT	9/22/2019
13	2020	Gove	Ulysses	To. Haplustolls	TAM 114	CT	9/24/2019
14	2020	Gove	Ulysses	To. Haplustolls	TAM 114	CT	10/3/2019
15	2020	Gove	Ulysses	To. Haplustolls	TAM 114	CT	9/28/2019
16	2020	Marion	Ladysmith	P. Ud. Argiustolls	WB4303	NT	9/25/2019
17	2020	McPherson	Goessel	U. Haplusterts	CP7909	NT	9/30/2019
18	2020	McPherson	Goessel	U. Haplusterts	CP7909	NT	9/30/2019
19	2020	Riley	Ivan and Kennebec	C. Hapludolls	Larry	NT	10/25/2019
20	2020	Riley	Wymore	Aq. Argiudolls	Tatankta	NT	10/25/2019
21	2020	Reno	Ninnescah	F. Endoaquolls	Everest	CT	11/1/2019
22	2020	Sheridan	Keith	A. Argiustolls	TAM 114	CT	10/1/2019
23	2020	Sheridan	Keith	A. Argiustolls	TAM 114	CT	10/1/2019
24	2020	Smith	Harney	T. Argiustolls	Tatankta	NT	10/14/2019

† C, Cumulic; Ty, Typic; A, Abruptic; To, Torriorthentic; P, Pachic; U, Udic; A, Aridic; Ud, Udertic; Aq, Aquertic; F, Fluvaquentic;
‡CT, Conventional Tillage; NT, No Till.

Table 3-2. Soil test information from samples collected before wheat sowing and fertilizer application.

	0-15 cm depth						0-60 cm depth			
Location	pH	OM	P	K	S	CEC	NO3‡	NH4	S	Cl
		g kg^{-1}	-mg kg^{-1}-			(meq/100g)	- mg kg^{-1} -			
1	6.5	23	48	347	3.4	13.0	5.7	5.5	23.1	3.9
2	7.7	25	18	249	4.4	25.9	22.4	5.5	4.5	2.5
3	6.2	35	9	127	2.7	15.1	1.5	5.2	4.2	5.4
4	6.9	25	17	629	3.6	18.4	10.4	3.3	4.8	6.1
5	6.7	26	21	604	3.2	17.5	8.3	3.6	5.7	6.4
6	7.6	20	18	595	2.6	29.1	8.6	3.4	5.6	5.2
7	6.4	25	20	528	2.5	15.2	5.0	4.5	7.0	3.5
8	5.7	31	16	283	2.2	16.1	1.6	6.4	3.2	4.2
9	7.7	20	29	191	2.0	21.2	7.5	4.9	3.2	3.6
10	6.2	23	9	304	0.7	10.7	1.2	4.1	1.2	2.5
11	5.6	23	26	270	1.1	9.3	2.9	3.5	1.5	2.4
12	7.2	23	20	712	2.4	21.6	3.7	2.7	2.5	3.4
13	8.3	23	18	702	2.4	30.5	5.1	2.7	2.1	3.0
14	7.3	25	14	613	2.0	20.4	4.1	2.2	2.8	3.4
15	7.6	24	16	770	1.8	24.3	6.5	2.2	2.5	4.5
16	6.5	25	14	227	1.4	21.7	2.2	4.2	2.5	2.0
17	5.4	27	13	153	1.8	11.9	3.3	4.0	12.3	1.5
18	5.6	30	12	154	1.0	14.7	3.1	4.7	2.1	1.9
19	6.2	29	11	176	1.4	13.3	1.7	2.9	1.6	1.3
20	6.6	28	9	294	1.8	16.6	0.6	3.7	3.0	1.3
21	6.7	18	2	78	12.8	6.3	1.3	2.2	151.9	64.2
22	6.8	24	20	632	2.3	17.4	5.7	2.1	3.5	4.1
23	6.8	24	29	626	2.6	19.1	5.1	2.1	2.6	3.8
24	5.6	28	41	427	1.5	13.4	1.7	3.0	1.9	2.3

† OM, Organic Matter; P, Mehlich-3 Phosphorus; K, Potassium; CEC, Cation Exchange Capacity

‡ NO3, Nitrate; NH4, Ammonium; S, Sulfur; Cl, Chloride;

Table 3-3. Comparison of sulfur soil test methods through linear regression.

Sulfur extraction methods		Intercept	slope	R^2	p > F
X	Y				
Calcium Phosphate	Resin	1.77	1.380	0.76	<0.001
Calcium Phosphate	Mehlich-3	55.30	0.897	0.31	<0.001
Calcium Phosphate	Ammonium Acetate	2.20	1.120	0.96	<0.001
Resin	Calcium Phosphate	0.97	0.549	0.76	<0.001
Resin	Mehlich-3	56.40	0.479	0.22	<0.001
Resin	Ammonium Acetate	3.19	0.625	0.75	<0.001
Mehlich-3	Calcium Phosphate	-13.70	0.348	0.31	<0.001
Mehlich-3	Resin	-16.40	0.469	0.22	<0.001
Mehlich-3	Ammonium Acetate	-14.10	0.406	0.32	<0.001
Ammonium Acetate	Calcium Phosphate	-1.56	0.854	0.96	<0.001
Ammonium Acetate	Resin	-0.55	1.200	0.75	<0.001
Ammonium Acetate	Mehlich-3	53.60	0.795	0.32	<0.001

Table 3-4. Significance of F values for fixed effects of sulfur fertilizer on NDVI, biomass, and sulfur tissue concentration collected at the Feekes 6 growth stage. Flag leaf S tissue concentration, uptake at the Feekes 11.2, and grain yield.

	Feekes 6			Flag Leaf	Feekes 11.2	Grain Harvest	
Location	NDVI	Biomass	S† Con.‡	S Con.	S Uptake	Yield	S Removal
	- - - - - -	- - - - - - - - - - - - - - - - - -	- - - - - -	p > F	- - - - - - - - - - - - - - - - - -	- - - - - - - - - - - - - - - - -	- - - - - -
1	0.737	0.632	0.226	0.653	0.283	0.558	0.710
2	0.247	0.308	0.028	0.202	0.719	0.679	0.460
3	0.100	0.069	§	§	§	§	§
4	0.146	0.279	§	§	§	§	§
5	0.517	0.944	§	§	§	§	§
6	0.722	0.262	0.514	<0.001	0.669	0.560	0.670
7	0.092	0.669	0.003	<0.001	0.010	0.776	0.271
8	0.131	0.553	0.588	0.724	0.513	0.129	0.013
9	0.172	0.575	0.067	0.406	0.181	0.304	0.247
10	0.256	0.793	<0.001	<0.001	<0.001	0.134	0.079
11	0.189	0.610	0.006	<0.001	0.006	0.039	<0.001
12	0.628	0.916	0.006	<0.001	0.342	0.210	0.403
13	0.039	0.050	0.008	<0.001	0.037	0.343	0.121
14	§	§	§	0.012	0.070	0.979	0.976
15	0.930	0.236	0.005	<0.001	0.585	0.473	0.566
16	0.647	0.413	0.443	<0.001	0.001	0.026	0.206
17	0.057	0.303	0.035	0.044	0.094	0.094	0.127
18	0.516	0.424	0.049	0.004	0.083	0.461	0.396
19	0.099	0.541	0.003	<0.001	0.021	0.073	0.722
20	0.202	0.539	<0.001	0.152	0.314	0.050	0.395
21	§	§	§	§	§	0.483	0.549
22	0.255	0.609	<0.001	0.015	0.111	0.795	0.897
23	0.031	0.320	<0.001	0.015	0.708	0.617	0.754
24	§	§	§	<0.001	0.034	0.052	0.197
Across Locations	0.392	0.944	<0.001	<0.001	<0.001	0.126	0.031

§ Samples were not collected for these locations.

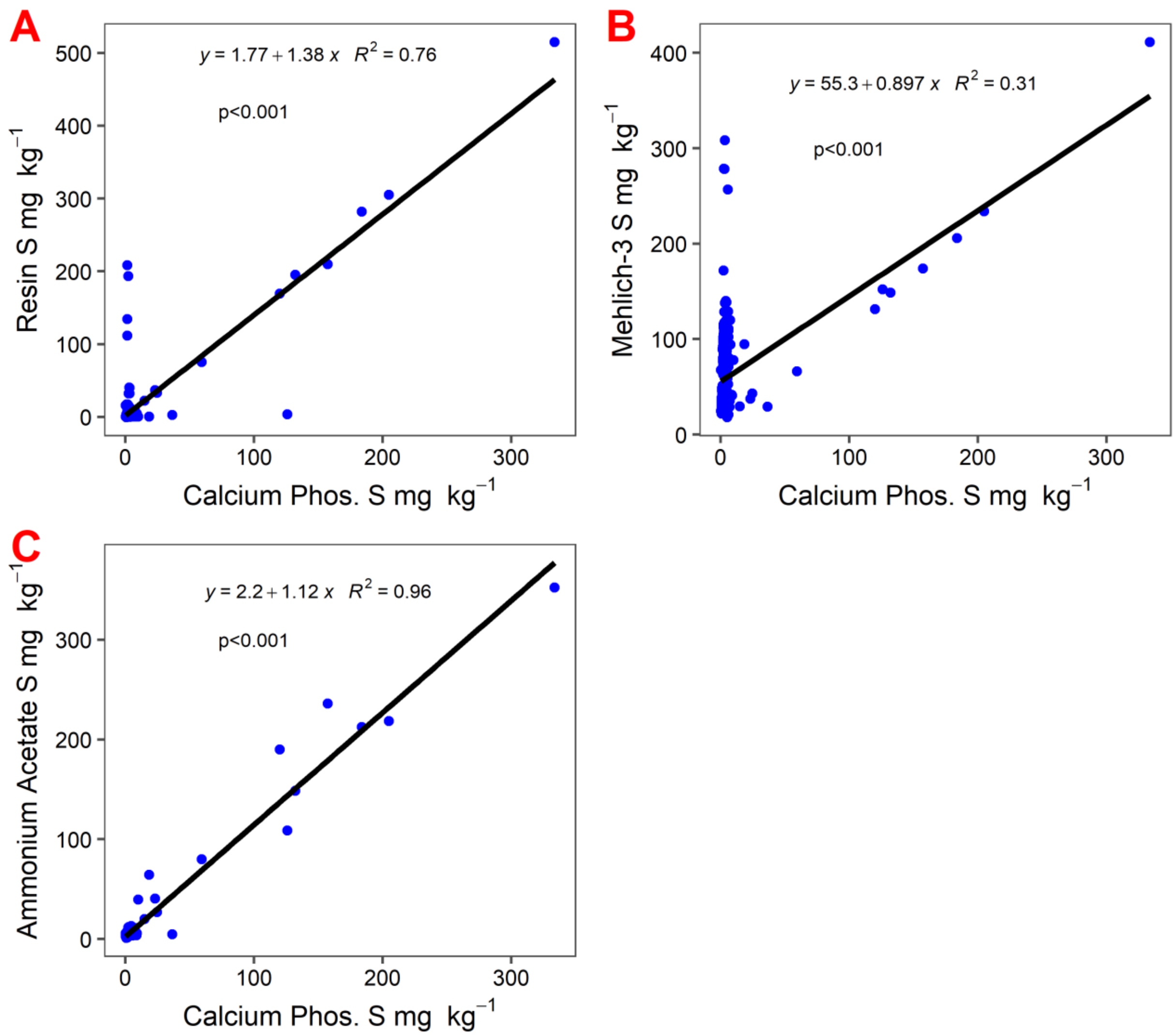

Figure 3-1. Relationships between the calcium phosphate sulfur extraction method, and (A) resin extraction method, (B) Mehlich- 3 extraction method, and (C) Ammonium acetate extraction method. Sulfur was determined with the ICP–OES (Inductively Coupled Plasma Optical Emission Spectrometry).

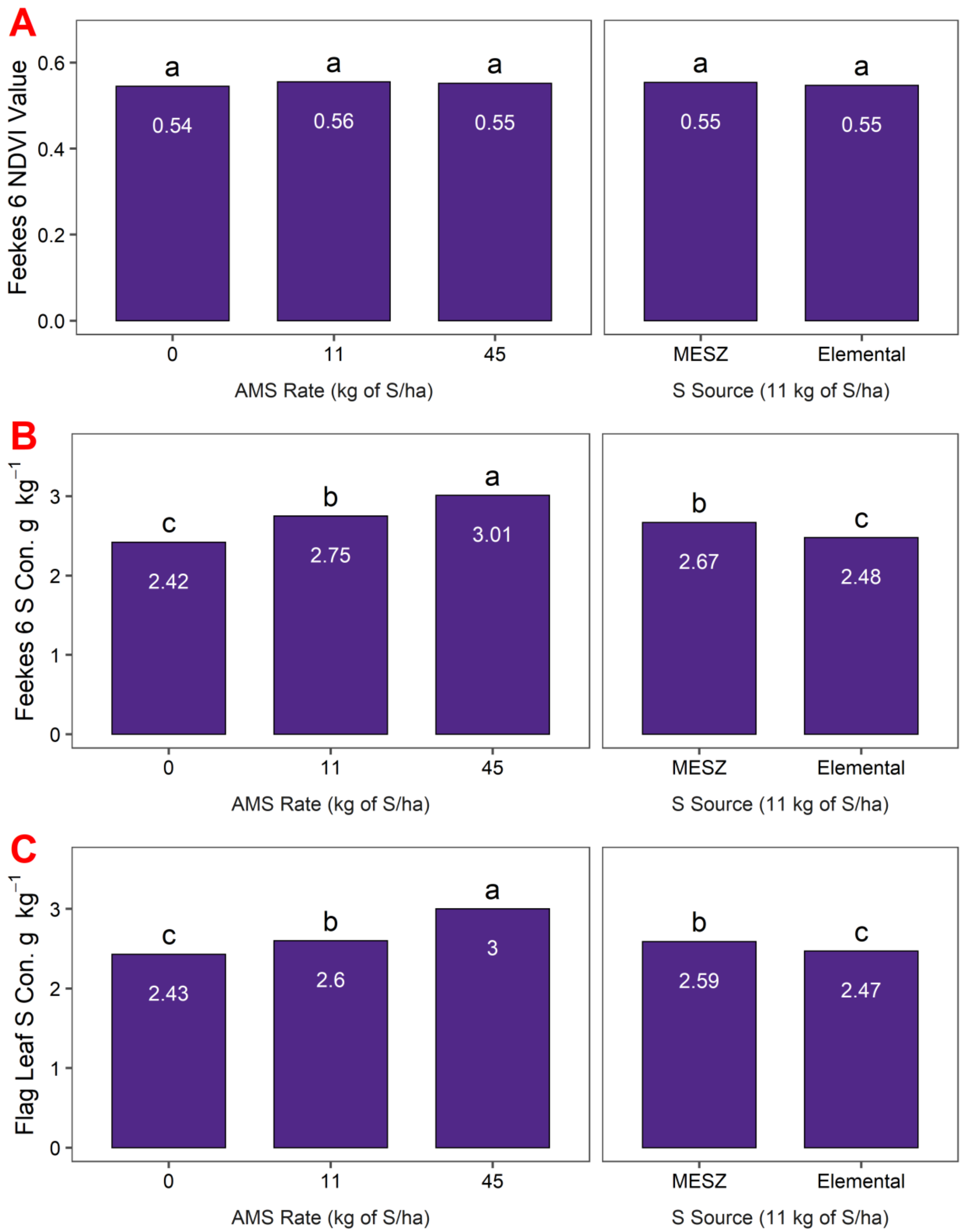

Figure 3-2. Wheat response to S fertilizer rate and source treatments, MESZ and elemental S was applied at 11 kg S ha^{-1}. Response parameters included (A) NDVI values and (B) tissue S concentration collected at the Feekes 6 stage, and (C) S concentration in the flag leaf.

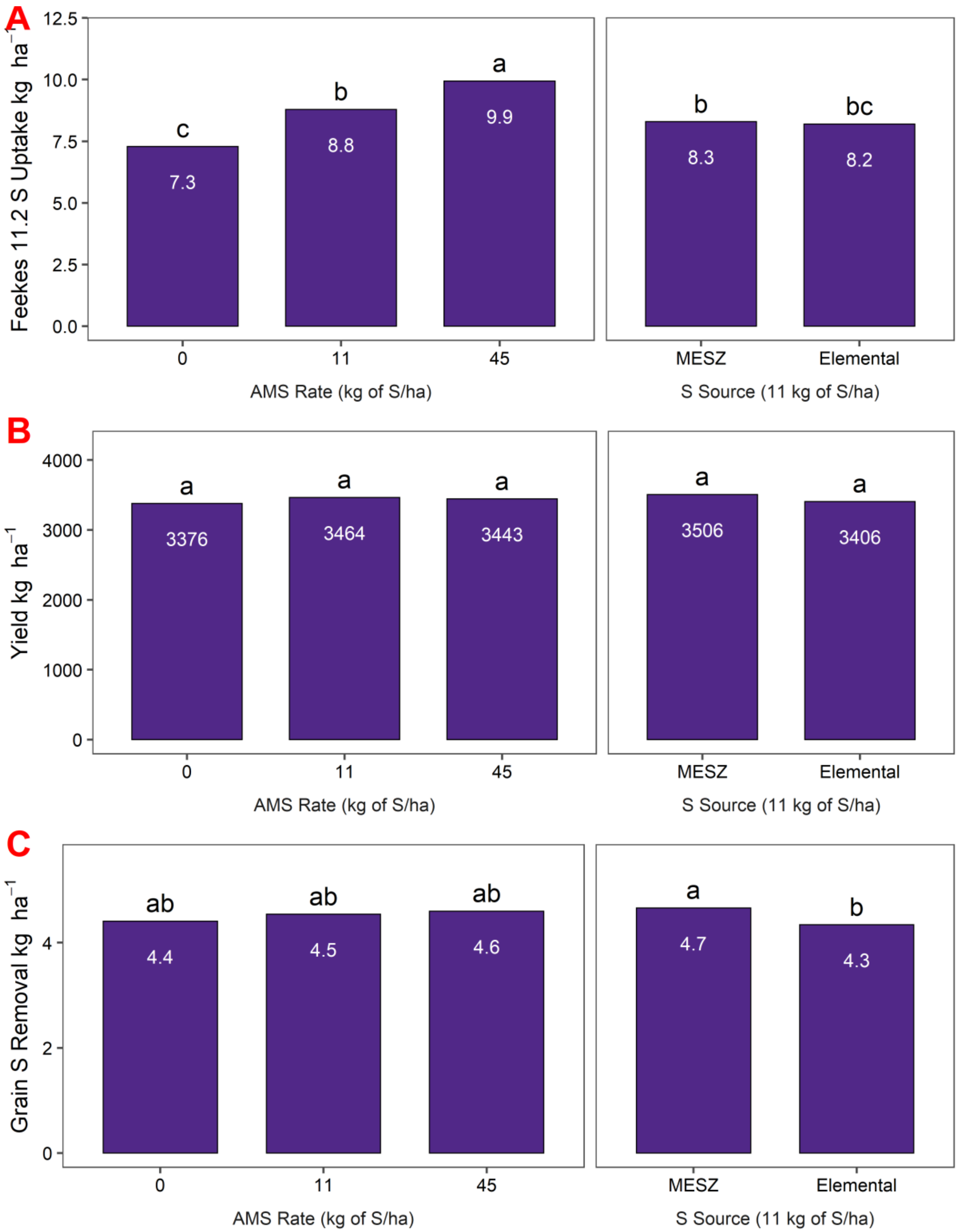

Figure 3-3. Wheat response to S fertilizer rate and source treatments, MESZ and elemental S was applied at 10 kg S ha-1. Late- season response parameters included (A) total S plant uptake at the Feekes 11.2 stage, (B) grain yield, and (C) S removal with the grain.

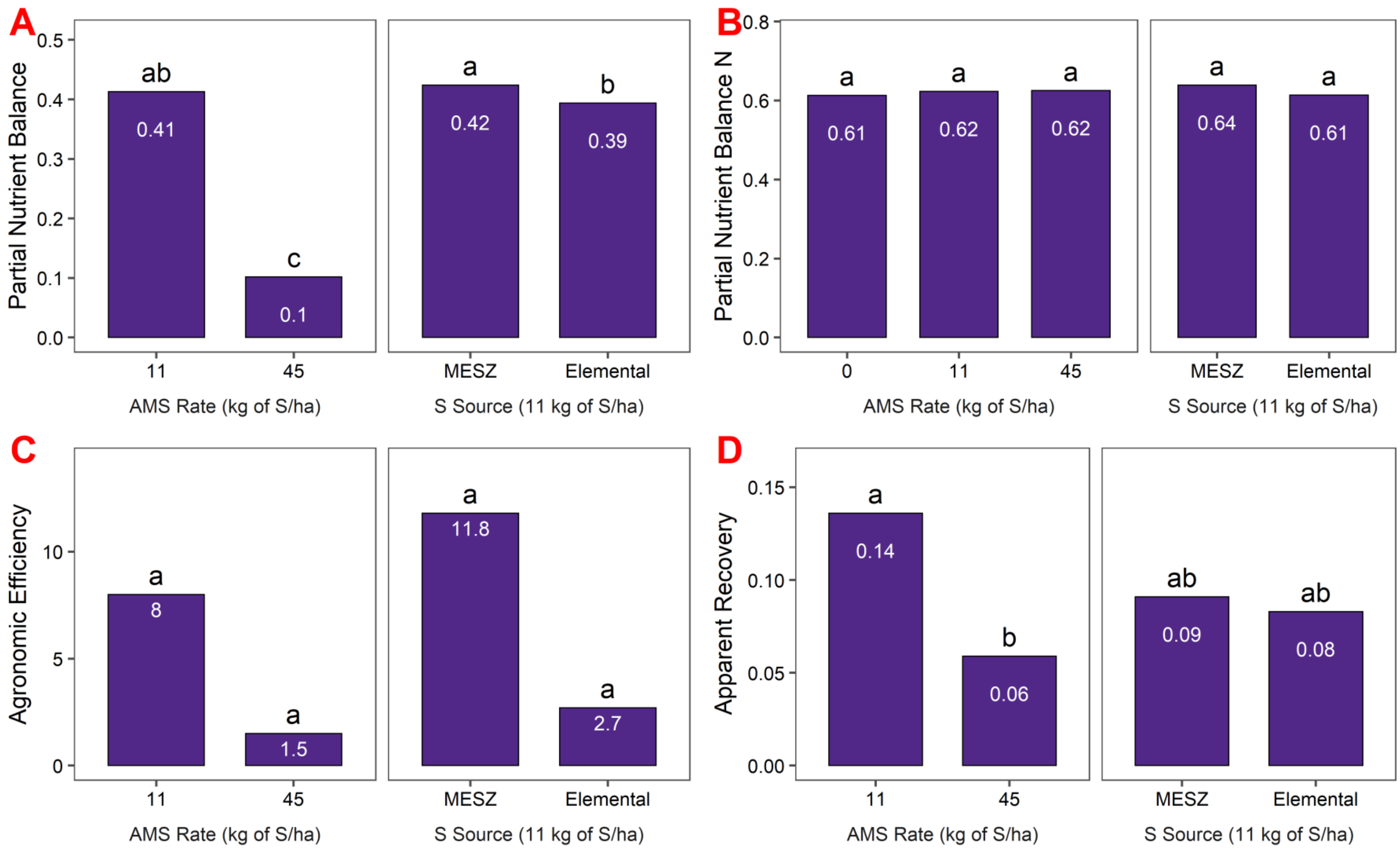

Figure 3-4. Nutrient use efficiency parameters, (A) S partial nutrient balance, (B) N nutrient balance to S application, (C) agronomic efficiency, and (C) apparent recovery efficiency.

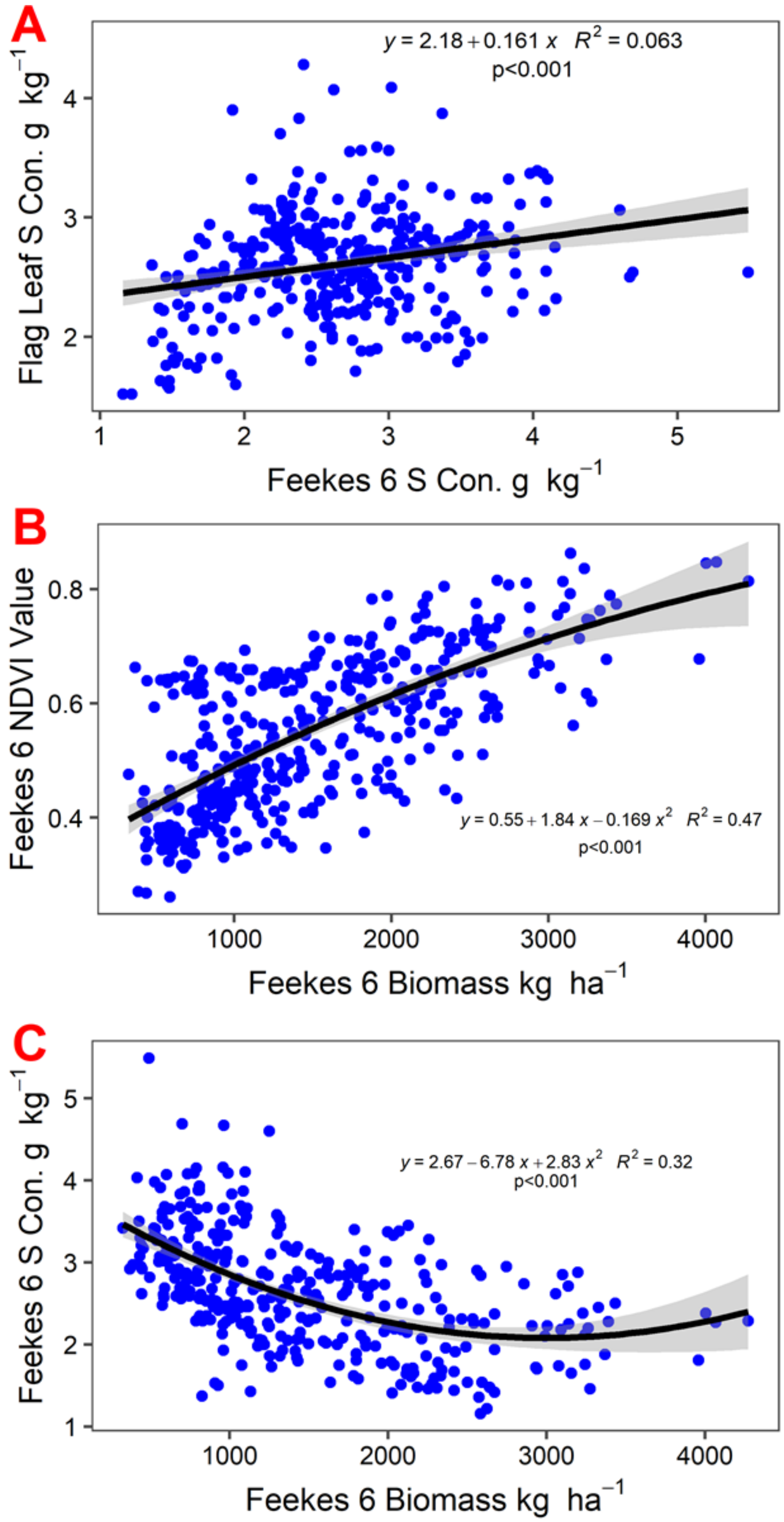

Figure 3-5. Relationships between (A) tissue S concentration in the flag leaf and samples at Feekes 6, (B) biomass and NDVI at the Feekes 6 stage, and (C) biomass and S concentration at the Feekes 6 stage.

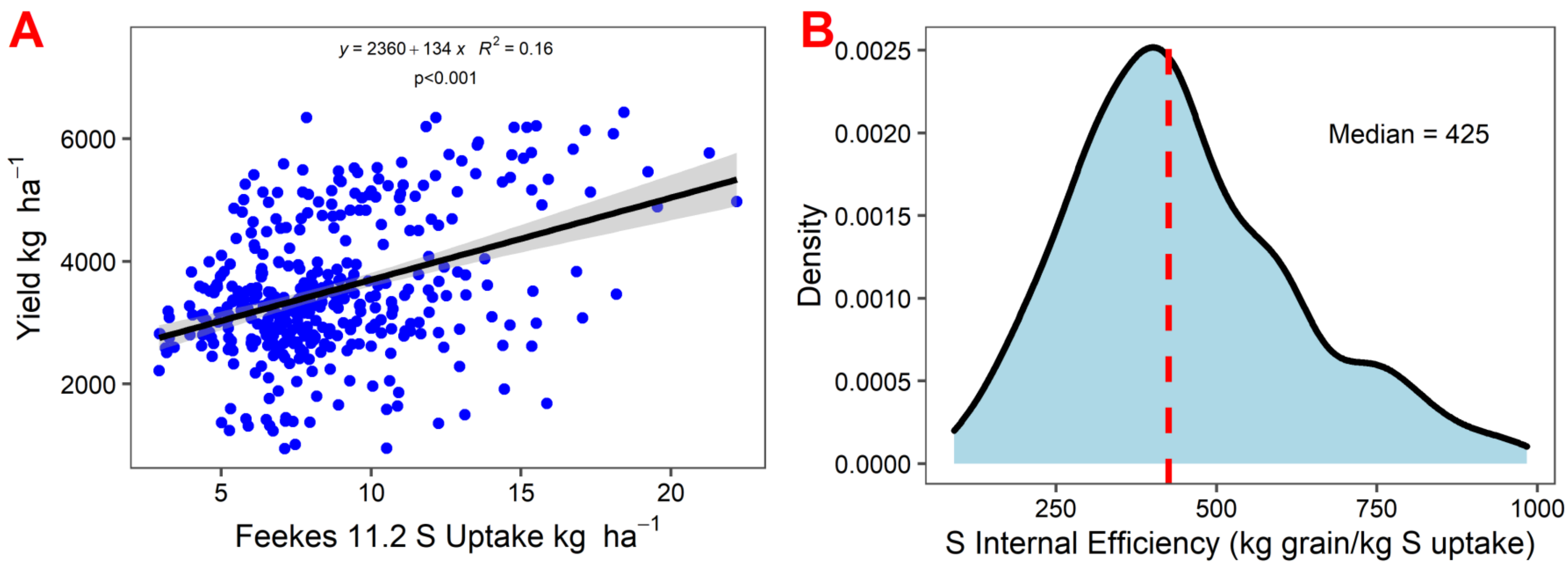

Figure 3-6. Relationship between wheat S uptake at the Feekes 11.2 stage and grain yield (A). Density plot of S internal efficiency and median value (B).

Chapter 4 - Blending phosphorus fertilizer with wheat seed: response to source, rate, and time

Abstract

Blending dry phosphorus (P) fertilizer with winter wheat seed can provide a starter fertilizer benefit to the crop. This study was designed to evaluate the effects of dry phosphorus sources, rates, and storage times of fertilizer-seed blends, effects on early growth, and overall productivity and yield of the crop. Four winter wheat experiments were conducted in the 2019-2020 wheat growing season in Northeast Kansas. The previous crop was soybeans for three locations and corn for one location. The winter wheat was no-till drilled at 79-101 kg ha^{-1} and blended with either diammonium phosphate “DAP” (18-46-0) or Micro-Essentials SZ “MESZ” (12-40-0-10S-1Zn) at rates of 0, 34, 67, and 135 kg P_2O_5 ha^{-1}. Storage times of seed-fertilizer blends were 0, 7, 22, and 34 days. The winter wheat was drilled in October and November and top-dressed with 112 kg of Nitrogen (N) ha^{-1} using urea ammonium nitrate (UAN) 28% at Feekes 5 in the spring. Normalized difference vegetation index measurements (NDVI) and biomass measurements were taken at Feekes 6. Significant increases in Feekes 6 NDVI were observed when overall P_2O_5 rates were increased with both P sources; however, minimal differences were observed when seed-fertilizer blend storage times were compared aside from a slight decrease for the longest storage period. Feekes 6 biomass and P uptake were significantly increased with increasing rates of fertilizer but were not affected by seed-fertilizer blend storage time or source. Biomass and whole plant P uptake at Feekes 11.2, and grain yield followed the same trends. Both were increases as fertilizer rates increased with minimal effects occurring with increasing storage time or fertilizer source.

Abbreviations: N, nitrogen; P, phosphorus; normalized difference vegetation index, NDVI.

Introduction

Applications of P fertilizer for wheat are typically made before or around drilling to ensure the crop is not P deficient in lower testing P soils. Many methods are used to apply P fertilizer, with standard methods being broadcast, banding, or in-furrow applications. All methods are potentially effective and are typically dependent upon P source, application equipment, and producer preference. Some producers who lack the equipment on their drills to apply a starter fertilizer in-furrow opt to blend the fertilizer with the wheat seed before drilling. The drill measures the P fertilizer and wheat seed at the same time, depositing both the seed and fertilizer into the furrow.

How this method affects the germination of the wheat crop is partly unknown but is a question of concern. It is already known that increases in salt concentration in-furrow or the soil, in general, can lead to decreases in germination (Pandya and Mer, 2005). This negative effect is amplified when fertilizers are concentrated in a specific area, such as an in-furrow fertilizer application. Fertilizer then dissolves into a small area, which increases the salt concentration of the soil moisture in the area around the seed (Rader, 1942). This higher salt concentration leads to a lower osmotic pressure, which can affect seed germination. However, what is unknown is how the direct contact of fertilizer with the wheat seed prior to drilling affects the performance of winter wheat throughout the growing season. In addition, when a producer blends wheat seed with fertilizer, it is typically not drilled immediately, and this blend may sit for some time. Typically, it has been advised not to leave seed-fertilizer blends to sit for more than a few days. However, weather can play a large role in the progression of wheat drilling in the fall. Unfavorable weather conditions can result in this seed-fertilizer blend being left sit for extended periods of time. With little to no knowledge on this topic available, the question arises if the seed is still viable and productive.

Newer equipment that has onboard fertilizer units that separate the fertilizer from wheat seed has eliminated the concern of extended blend storage times. However, as stated earlier, producers that are new to this application technique still rely upon blending wheat seed with P fertilizer. Even producers with separate seed-fertilizer units are still concerned with the amount of P fertilizer they can put in-furrow without seeing crop injury. Understanding interactions between fertilizer rates, sources, and blend times are essential for all producers who seek to apply banded fertilizers in wheat production and look to maximize the use of planting equipment for higher rates of fertilizer application. This study's objectives were to i.) identify how storage time of fertilizer-seed blend, fertilizer rate, and source with wheat affect the overall production of the crop, and ii.) provide recommendations based on results to help guide producers that are implementing this management strategy.

Materials and methods

Four locations were established in 2019 and 2020, under no-till and after soybean at locations 1, 3, and 4, while conventional tillage after corn was used at location 2 (Table 4-1). Soil test phosphorus (P) levels ranged from 11-20 mg of P kg^{-1} using the Mehlich-3 test. The individual plot sizes were 9.1 – 12.2 m in length and 1.8 m in width. The row spacing at sowing was 19 cm for all locations. The selection of wheat varieties was based on regional performance.

Wheat seed and P fertilizer were blended thoroughly and stored in plastic containers until sowing. Three storage periods were evaluated (0, 7, 22, and 34 days). Two P fertilizer sources were blended with the wheat seed: diammonium phosphate (DAP) (18-46-0) and Micro-Essentials SZ (MESZ) (12-40-0-10S-1Zn). Phosphorus fertilizer rates were 34, 67, and 135 kg of P_2O_5 ha^{-1}. All combinations of storage time (4), fertilizer source (2) and P fertilizer rate (3) were included in the study in addition to a control with no P fertilizer application to assess wheat response to P application. Wheat/fertilizer blends were sowed with a Great Plains drill with a SRES cone attached (Seed Research Equipment Solutions, Hutchison KS). A cone planter was used because various sizes of seed-fertilizer blends were needed to be planted in a specific area. Although the amount of wheat seed did not change, the amount of fertilizer blended with it did change. The cone planter allowed this difference by dropping the wheat/fertilizer blend into a rotating cone that made one revolution per plot length to distribute this blend for even planting. Nitrogen (N) was applied at 112 kg N ha^{-1} using urea ammonium nitrate (UAN, 28% N) in the spring at green-up using streamer bars to dribble the liquid fertilizer in 12.7 cm spacing. Nitrogen was applied uniformly across the study, and therefore, N applied with P fertilizer at sowing in the fall contributed with additional N depending on the fertilizer application rate (range from 0 to 53 kg N ha^{-1} for the highest rate). The MESZ fertilizer source also included sulfur and zinc with rates varying from 0 to 34 kg of S ha^{-1} and 0 and 3.4 kg of Zn ha^{-1}. Annual and historical weather data were collected from automated weather stations located < 1 km from the field locations.

Composite soil samples of 10-12 cores were collected at a depth of 0-15 cm from each study area before sowing wheat. Soil samples were dried at 41^{o} C for 5-10 days and ground to pass through a 2 mm sieve. Soil tests included pH 1:1 (soil:water) (Watson and Brown, 1998), organic matter by loss on ignition (Combs and Nathan, 1998), and phosphorus using the Mehlich-3 extraction (Frank et al., 1998).

Normalized difference vegetative index (NDVI) measurements were taken at the Feekes 6 (jointing) stage using an active sensor RapidSCAN CS-45 handheld crop sensor (Holland

Scientific). The sensor senses red light at a wavelength of 670 nm, red-edge light at 730 nm, and near inferred light at 780 nm. For this study, the red and near inferred wavelengths were used to calculate the NDVI for each treatment. The sensor was held at the height of 1.2 meters above the crop canopy, and readings were collected from the entire length of each plot. At the end of each plot, the average measurement was recorded.

Wheat biomass samples were collected at early vegetative (Feekes 6) and late reproductive (Feekes 11.2, Soft Dough) stages. Samples were collected from 2 rows at a length of 76 cm (0.29 m^2). All biomass samples were collected from the back 3 meters of each plot, which was excluded from future measurements. The samples collected at Feekes 11.2 were taken from wheat rows that had not been sampled previously. Biomass samples were oven-dried at 60^o C for 5-7 days and weighed to estimate dry biomass. Biomass sampling at Feekes 11.2 followed similar procedures. However, from the total biomass collected, a sub-sample was oven-dried at 60^o C for 5-7 days to estimate moisture, which was used to convert the fresh mass of the whole sample to dry biomass. Dry samples were ground to pass through a 2 mm screen and then digested for total P using a sulfuric acid/ peroxide digest (Matsunaga & Shiozaki, 1987). Phosphorus concentration was determined by inductively coupled plasma optical emission spectrometry (ICP-OES). Phosphorus concentration and biomass were multiplied to estimate plant P uptake. Grain yield was determined by harvesting the center 1.4 meters of each plot using a plot combine with a platform header. Subsamples were taken from each plot and tested for moisture and test weight.

The experimental design was a randomized complete block design with four replications. Statistical analysis was completed using a linear mixed effect model (lme4 package) in R version 4.0.3 (R Core Team, 2020), with blocks included as random effects in the model. For analysis across locations, locations and blocks were included as random effects in the model. Significant treatment differences were established at alpha = 0.05.

Results and discussion

Precipitation patterns in the first year were generally higher than average, but the second year followed average precipitation trends (Figure 4-1). It is important to note the spikes in precipitation before drilling in the fall of 2018 (Figures 4-1 A & B). These conditions led to the high moisture content in the soils during the seeding time frame. In the fall of 2019, this large spike in precipitation did not occur (Figures 4-1 C & D). However, when drilling, the drill was set to a normal depth that penetrated into moist soil on the 2019-2020 wheat crop.

Dry conditions and coarse-textured soils are considered as conditions leading to a higher risk for seedling damage from fertilizer application in-furrow. Soil texture was similar at all locations; however, lower soil organic matter content was observed at locations 2 and 4 compared to locations 1 and 3 (Table 4-1). Soil pH values were near 7.0, except for location 1 with a pH of 5.8. Soil pH values below 5.5 combined with soluble aluminum can cause aluminum toxicity in plant roots (Shroyer, 2013), and the application of P fertilizer in-furrow can reduce the incidence of aluminum toxicity due to P binding with available aluminum in the soil (Hagin, 1982). Soil test P at or below 20 ppm at all locations were below the critical value, and response to P fertilizer would be expected (Leikam, 2003).

Feekes 6 NDVI, Biomass, and P Uptake

Early season parameters at Feekes 6 (NDVI, biomass, and P uptake) showed a significant response to P fertilizer rates (Table 4-2). Values of NDVI showed a significant interaction of P fertilizer source by blend storage time for the analysis across locations. A decrease in NDVI values at the early-season growth stage was observed with a longer storage time of the fertilizer-seed blend, but the decrease was less pronounced for the DAP fertilizer source (Figure 4-4). The MESZ fertilizer source has a slightly higher NDVI compared to DAP at the 0 day blend period and ended with a lower NDVI compared to DAP and the 34 day blend period. The NDVI could have been slightly higher with the MESZ fertilizer source compared to the DAP source because of the S that is in the MESZ fertilizer source. There was a brief time in the spring where a slight difference could be visually seen between DAP and MESZ, with MESZ being greener which confirms the higher NDVI as stated before. There is not enough data to describe why the NDVI decreased below DAP and maybe more due to normal variation. A producer is unlikely to leave a seed-fertilizer blended for this long, which makes this interaction less important in typical producer practices.

Mixed results were seen at each location when comparing blend time intervals with NDVI measurements. Locations 2 and 3 were significant at the 0.05 probability level, but the other two locations were not (Table 4-2). At location 2, there was a slight decrease in NDVI at the longest blend period of 34 days and resulted in a significant decrease of 10% in Feekes 6 NDVI compared to the 0 day blend period (Table 4-4). Location 3 had varying results, with the 0 day blend period being no different than all other treatments except the 22 day blend period, which was significantly different from both the 0 and 34 day blend periods (Table 4-4). Due to these variable results, it's difficult to tell if the trends are substantial or if they are normal variations in the data set. However, across locations, there was a slight decrease in Feekes 6 NDVI, with the longest blend period being statistically less than all the other blend periods with a 4% reduction in NDVI (Figure 4-2 A). As discussed above, this reduction is unlikely to be reproduced in typical production practices especially with the adoption of separate seed and fertilizer compartments farm equipment.

At individual locations, only location 1 showed significant results when comparing P sources (Table 4-2). At location 1, there was a slight decrease in Feekes 6 NDVI with MESZ P fertilizer compared to DAP P fertilizer which resulted in a 2% decrease in Feekes 6 NDVI (Table 4-4). When performing ANOVA across location, a p-value of 0.58 was calculated, indicating there is no significant difference between P sources for NDVI at the Feekes 6 stage (Figure 4-2 A). This slight difference could be due to a sulfur response, but it is difficult to identify this small difference.

The rate of P fertilizer was significant at 3 of the 4 locations for Feekes 6 NDVI. Location 4 was the only location to show no response to P rate and had a p-value of 0.159 (Table 4-2). At responsive locations, Feekes 6 NDVI was significantly increased up to the highest P rate at location 1, lowest rate of 34 kg of P_2O_5 ha^{-1} at location 2, and up to the second-highest P rate at locations 3 (Table 4-4). The addition of P fertilizer increased Feekes 6 NDVI by 36, 12, and 127 percent at locations 1, 2, and 3, respectively. Across locations, Feekes 6 NDVI was significantly increased up to the second-highest P rate and resulted in a 33 percent increase in Feekes 6 NDVI (Figure 4-2 A). NDVI increases with increasing fertilizer rates are likely due to increased plant biomass through increases in leaf area and shoots, which is confirmed by estimates of plant biomass at this stage.

Of the 4 locations, only one location showed significant effects on the time P fertilizer was blended with wheat seed on Feekes 6 biomass (Table 4-2). Results at this location are likely due to field variability as the only significant response was between the 0 and 7 blend periods, with the 7-day blend period being significantly less (Table 4-4). Other than this, there were no significant differences between any of the other blend periods (Table 4-4). Across locations, there was a significant response when comparing storage periods (Table 4-2). The only difference was between the 0 and 34-day duration, which resulted in a 9 percent reduction in Feekes 6 biomass for the longest duration (Figure 4-2 B). Much like the time the blend was stored, the P source did not have any effect on Feekes 6 biomass at each location individually or across locations (Table 4-2, Table 4-4 and Figure 4-2 B). These results indicate there were no effects of source or blend storage time on early season biomass, and in this case, it leaves an abundance of management flexibility in a production agriculture setting.

The rate of P fertilizer was significant at 3 of the 4 locations at the Feekes 6 biomass sampling event (Table 4-2). Location 4 was the only location to show no response to P rate and had a p-value of 0.721 (Table 4-2). At the responsive locations, significant increases in Feekes 6 biomass were observed up to the highest P rate at locations 1 and 2, but location 3 only responded up to the second-highest rate (Table 4-4). The addition of P fertilizer increased Feekes 6 biomass by 154, 75, and 540 percent at locations 1, 2, and 3, respectively (Table 4-4). Across locations, Feekes 6 biomass was significantly increased up to the highest P rate and resulted in a 90 percent increase in Feekes 6 biomass (Figure 4-2 B). Results from the rate of fertilizer fall directly in line with what was seen with NDVI, confirming that NDVI is directly related to the amount of biomass.

Storage time intervals of either P source did not result in significant changes in Feekes 6 P uptake at locations 2, 3, and 4 (Table 4-2). However, location 1 had a significant p-value of 0.016 with a difference between the 0 and 7 day blend periods. This response resulted in a 1.1 kg of P ha^{-1} decrease in P uptake from the 7 to 0 day blend period (Table 4-2, Table 4-4). With this small of a difference and with other blend periods being similar to either one, it is hard to come to a sound conclusion. Across locations, no responses were seen as storage duration increased (Table 4-2, Table 4-4, and Figure 4-2 C). At locations individually and across locations, there were no significant differences between P sources (Table 4-2, Table 4-4, and Figure 4-2 C). These trends are almost identical to what was observed with Feekes 6 biomass.

The rate of P fertilizer significantly affected P uptake at Feekes 6 at all locations (Table 4-2). Locations 1, 2, and 3 responded up to the highest P rate, but location 4 only showed differences between the lowest P rate and the other application rates, including the no P fertilizer treatment (Table 4-4). The addition of P fertilizer increased Feekes 6 P uptake by 195, 230, and 1154 percent at locations 1, 2, and 3, respectively, compared to the control treatment (Table 4-4). Across locations, Feekes 6 P uptake was significantly increased up to the highest P rate and resulted in a 162 percent increase (Figure 4-2 C).

Across early-season responses, the source of P fertilizer had limited effects. A reason for this is likely due to the lack of need for other nutrients that are supplied by the MESZ fertilizer. Because most of the soils would be classified as a type of loam, the amount of S supplied by organic matter likely supplied enough of this nutrient for this yield potential. Although zinc (Zn) was also applied with MESZ, there are limited data supporting winter wheat as a Zn responsive crop in Kansas soils. Storage time did have an effect in some of the results, but results were very scattered, with most responses being similar to the control at responsive locations. Without further research, it’s hard to identify if blend storage duration has an effect at the early growth stage of winter wheat within a typical wheat fertilizer blend time period used by producers. The rate of fertilizer was the largest driving factor of all responses, with all but one location at two sampling times being unresponsive. Positive responses were linked with increased rates of P fertilizer. There are many reasons why this was likely seen. The biggest being the fact that all soil test P levels were at or below the critical level. For this reason, we likely see a starter P response. Probably the most interesting result is that as rates approached and surpassed the generally maximum recommended N application rate in-furrow, no negative responses were identified in the early growth stage. This is especially useful if a producer wants to apply higher rates of P in-furrow with a narrower row spaced crop like winter wheat.

Late Season P Uptake and Yield

Late season response (P uptake and yield) showed a significant response to P fertilizer rates (Table 4-3). Values of P uptake showed a significant interaction of P fertilizer source, P fertilizer rate, and by blend storage time for the analysis across locations (Table 4-3). It is difficult to pick out a clear trend in this interaction (Figure 4-5). The statistical power at this interaction level is greatly reduced compared to looking at single factors, reducing the chance of finding a usable outcome from this interaction. The majority of this interaction is likely due to noise in the data that is causing one of the treatments to be deemed significant from another.

Storage time intervals of P fertilizer did not result in significant changes in Feekes 11.2 P uptake at any of the locations and across all locations (Table 4-3, Table 4-5, and Figure 4-3 A). At locations individually and across locations, there were no significant differences between either P source (Table 4-3, Table 4-5, and Figure 4-3 A). These results follow earlier trends seen in P uptake in the early growth stage.

P-values of locations individually as well as across locations all resulted in values less than 0.05 when analyzing the effects of P fertilizer rate on Feekes 11.2 P uptake (Table 4-3). Locations 3 showed the clearest trend, with the response being seen up to the highest P rate and resulted in a 118 percent increase in Feekes 11.2 P uptake (Table 4-5). Locations 1, 2, and 4 were more variable, with the only significant difference in Feekes 11.2 biomass being between the low P rate of 34 kg of P_2O_5 ha^{-1} and the highest two P rates at location 1 (Table 4-5). Location 2 only showed a difference in Feekes 11.2 P uptake between the control and the highest

P fertilizer rate, while location 4 only showed a difference between the 34 kg of P_2O_5 ha^{-1} fertilizer rate and the highest P fertilizer rate (Table 4-5). Across locations, Feekes 11.2 P uptake increased up to the highest rate of P fertilizer and resulted in a 60 percent increase in Feekes 11.2 P uptake compared to the control treatment (Figure 4-3 A).

Of the 4 locations individually, no locations showed significant effects on yield when comparing the time wheat seed - P fertilizer blend was stored (Table 4-3). Across locations, the time seed was stored with P fertilizer showed significance with a p-value of 0.01 (Table 4-3). However, significant differences were seen only between the 7 and 34 days blend periods, with the 0 day blend period being statistically similar to all other treatments (Figure 4-3 B). At locations individually and across locations, there were no significant differences between P sources (Table 4-3, Table 4-5, and Figure 4-3 B). This means a producer is flexible in their management strategies and can pick which P source they want to use without being worried about the blend duration if a normal time frame is used.

P-values of locations 1, 2, and 3 as well as across locations all resulted in significant values less than the 0.05 probability level when looking at the effects of P fertilizer rate on grain yield (Table 4-3). Location 1 responded up to the highest rate of P fertilizer with an increase of 1487 kg ha^{-1} grain yield compared to the control treatment (Table 4-5). Locations 2 and 3 were responsive up to the second-highest P rate with an increase in grain yield of 785 and 2075 kg ha^{-1}, respectively (Table 4-5). Across locations, grain yield increased up to the highest rate of P fertilizer and resulted in a 1228 kg ha^{-1} increase compared to the control (Figure 4-3 B).

Late season sampling and yield showed trends similar to what the early season showed. There was no effect of seed-fertilizer blend time and no effect from source at any of the locations individually. Although there was a statistical difference across locations when looking at the blend storage duration, the control was no different from any of the blend durations. Much like in early growth, rate played the biggest role, and responses were seen at all locations and sampling timing except one location at grain yield. Again no negative effects were seen late in the season as rates increased, further confirming that these fertilizers did not have an impact even when rates exceeded the recommended level of N in furrow.

Overall Considerations

In general, the rate of P fertilizer was the only factor out of these three to have a consistent effect on winter wheat production when comparing all locations and sampling timings. This is likely due to the individual sites having lower soil test P values than the critical P level (Leikam, 2003). These results provide valuable information that as P rates were increased, there were no negative effects up to the highest rates of P fertilizer, which exceeded the recommended amount (Ruiz Diaz, 2019). For the row spacing used in this study, the maximum recommended amount of N + K_2O is 34 kg ha^{-1} (Ruiz Diaz, 2019). At the highest rates of DAP and MESZ, fertilizer applied 53 and 40 kg of N ha^{-1} respectively. One factor that may have played a role is soil moisture at drilling time. Drier soils will typically result in more salt injury than wet soils (Laboskil, 2015). At all locations, soil moisture was average to above average, which would mitigate any negative effects seen with drier soils. Another reason negative effects may not have occurred is due to the form of N that is present in both P fertilizers. Both contain ammonium, which is considered a stable form of N that does not need to go through processes such as hydrolysis, which urea would need to undergo before becoming a stable plant-available form of N. The time that the seed - P fertilizer blend was stored had minimal impacts on general productivity throughout the season. This result could be due to the wheat seed not being in an actively growing state while blended with the fertilizer. The seed coat and dry conditions are likely barriers that prevented the P fertilizer from affecting the wheat seed. Slight negative effects were seen at the longer storage periods but were not consistent. For future research, increasing the blend period to multiple months may be needed to identify any interactions that may be present. The source of P fertilizer did not have any late-season effects at locations and was likely due to the soil having sufficient amounts of sulfur, which is a component in the MESZ P fertilizer. Early growth measurements of NDVI showed significant differences between sources, with MESZ having a higher value than DAP. This response could be due to the low organic matter at this location, which resulted in less sulfur being mineralized and available to the crop (Ruiz Diaz, 2019).

Conclusions

Overall, the source of P fertilizer had a slight effect at the early stages of winter wheat growth and is likely dependent on whether or not a specific field was deficient in sulfur at this stage. The blend time period did have an effect on specific locations but was limited to the long blend period that was over one month, and its effects were only seen at early growth and in yield. The rate of P fertilizer had effects across most sampling events, locations, and across locations. This response is likely due to the soil test P levels of each site individually. However, the rate of P fertilizer never led to a decrease in production due to salt injury even though the N in fertilizer was greater than recommended standards. Future research in this area could be focused on storage considerations of wheat seed P fertilizer blends and longer blend durations.

Blending P fertilizer with wheat seed can be an effective way to provide a starter P fertilizer effect for a winter wheat crop without needing a separate fertilizer system on drills. This practice is safe for wheat seed with minimal negative effects. It is important to note that keeping the seed-fertilizer blend below the critical humidity of a given P fertilizer is important to prevent caking of the blend in equipment and also avoid any potential negative effects to the seed itself. The typical critical humidity level of a P fertilizer is around 70%; being proactive in storing the seed-fertilizer blends inside during precipitation events and humid periods of the day is critical to prevent any negative effects of this practice. Even though time wheat seed - P fertilizer was stored had little to no effect on wheat productivity, pre-blending weeks in advance of drilling is still not recommended due to storage concerns stated above.

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Tables and Figures

Table 4-1. Field site location, soil classification and tillage system. Wheat variety, planting dates, and soil test information before fertilizer application.

										0-6" samples		
Location	Year	County	Soil Type	Sub Group†	Variety	Soil Texture	Planting Date	Tillage System‡	Row Spacing	pH	P	OM
									cm		ppm	%
1	2019	Riley	Smolan	P. Argiustoll	Everest	Silt Loam	11/19/2019	NT	19	5.8	17	3.2
2	2019	Shawnee	Eudora	F. Hapludoll	Everest	Silt Loam	10/19/2019	CT	19	7.0	18	1.6
3	2020	Riley	Ivan and Kennebec	C. Hapludolls	Larry	Silt Loam	10/29/2020	NT	19	7.4	11	2.9
4	2020	Shawnee	Eudora	F. Hapludoll	Larry	Silt Loam	11/1/2020	NT	19	6.7	20	1.1

† P, Pachic; F, Fluventic; C, Comulic

‡CT, Conventional Tillage; NT, No Till.

Table 4-2. Significance of F values for the fixed effects of fertilizer source, rate, and blend time on early-season wheat response. Values of normalized difference vegetative index (NDVI), biomass, and phosphorus uptake were collected at the Feekes 6 growth stage (jointing).

Source of variation†	Location 1	2	3	4	Across Locations
	- - - - - - - - - -	- - - - - - - - - -	p > F	- - - - - - - - - -	- - - - - - - - - -
			Normalized difference vegetative index		
Phosphorus rate (R)	<0.001	0.040	<0.001	0.167	<0.001
Phosphorus source (S)	0.043	0.410	0.290	0.053	0.580
Blend storage time (T)	0.072	<0.001	0.001	0.460	<0.001
R x S	0.653	0.296	0.077	0.759	0.368
R x T	0.073	0.661	0.012	0.977	0.831
S x T	0.189	0.002	0.450	0.888	0.046
R x S x T	0.189	0.571	0.300	0.093	0.754
			Early biomass		
Phosphorus rate (R)	<0.001	<0.001	<0.001	0.721	<0.001
Phosphorus source (S)	0.152	0.938	0.315	0.127	0.682
Blend storage time (T)	0.011	0.144	0.227	0.283	0.013
R x S	0.456	0.794	0.985	0.744	0.455
R x T	0.903	0.998	0.067	0.590	0.310
S x T	0.225	0.224	0.390	0.230	0.648
R x S x T	0.891	0.802	0.834	0.660	0.766
			Early P uptake		
Phosphorus rate (R)	<0.001	<0.001	<0.001	0.004	<0.001
Phosphorus source (S)	0.199	0.648	0.140	0.104	0.210
Blend storage time (T)	0.016	0.459	0.747	0.372	0.196
R x S	0.423	0.998	0.688	0.863	0.962
R x T	0.153	0.994	0.040	0.533	0.342
S x T	0.200	0.260	0.313	0.190	0.707
R x S x T	0.789	0.820	0.672	0.600	0.734

† Phosphorus rates (R) were 34, 67, and 135 kg P2O5 ha-1; Phosphorus source (S) included Diammonium Phosphate (18-46-0) and Micro-Essentials SZ (12-40-0-10S-1Zn); Blend time (T) of fertilizer-seed were for 0, 7, 22, and 34 days.

Table 4-3. Significance of F values for the fixed effects of fertilizer source, rate, and blend time on late-season wheat response. Total plant P uptake was measure at the Feekes 11.2 growth stage, and grain yield at harvest.

Source of variation†	Location				Across Locations
	1	2	3	4	
	- p > F -				
			Phosphorous uptake		
Phosphorus rate (R)	0.001	0.011	<0.001	0.027	<0.001
Phosphorus source (S)	0.698	0.053	0.888	0.491	0.125
Blend storage time (T)	0.616	0.794	0.573	0.996	0.959
R x S	0.185	0.240	0.360	0.559	0.903
R x T	0.448	0.808	0.399	0.371	0.420
S x T	0.483	0.234	0.555	0.945	0.413
R x S x T	0.117	0.021	0.082	0.641	0.020
			Grain yield		
Phosphorus rate (R)	<0.001	<0.001	<0.001	0.058	<0.001
Phosphorus source (S)	0.101	0.265	0.535	0.800	0.544
Blend storage time (T)	0.143	0.054	0.216	0.517	0.010
R x S	0.212	0.968	0.836	0.322	0.369
R x T	0.793	0.681	0.060	0.563	0.182
S x T	0.108	0.262	0.169	0.711	0.234
R x S x T	0.603	0.999	0.214	0.082	0.608

† Phosphorus rates (R) were 34, 67, and 135 kg P_2O_5 ha^{-1}; Phosphorus source (S) included Diammonium Phosphate (18-46-0) and Micro-Essentials SZ (12-40-0-10S-1Zn); Blend time (T) of fertilizer-seed were for 0, 7, 22, and 34 days.

Table 4-4. Early-season wheat response to the main effect of fertilizer-seed blend storage time, fertilizer rate, and source. Values of normalized difference vegetative index (NDVI), biomass, and phosphorus uptake were collected at the Feekes 6 growth stage.

	Storage time of fertilizer-seed blend (days)				Fertilizer rate (kg of P_2O_5 ha^{-1})				Fertilizer source	
Location	0	7	22	34	0	34	67	135	DAP	MESZ
	-Normalized difference vegetative index-									
1	0.59	0.57	0.58	0.57	0.46 d†	0.54 c	0.58 b	0.62 a	0.59 a	0.57 b
2	0.55 a	0.55 a	0.53 a	0.50 b	0.47 b	0.53 ab	0.53 ab	0.54 a	0.53	0.53
3	0.67 ab	0.69 a	0.70 a	0.66 b	0.31 c	0.64 b	0.70 a	0.71 a	0.68	0.68
4	0.72	0.73	0.71	0.70	0.66	0.71	0.73	0.70	0.70	0.73
	-Biomass (kg ha^{-1})-									
1	3063 a	2578 b	2870 ab	2763 ab	1294 d	2350 c	2827 b	3289 a	2891	2746
2	2282	2093	2177	2000	1363 c	1961 b	2070 b	2383 a	2142	2135
3	1909	2046	2020	1804	321 c	1702 b	2050 a	2083 a	1899	1991
4	1877	1971	1869	1712	2080	1819	1895	1857	1784	1930
	-Phosphorus uptake (kg P ha^{-1})-									
1	6.5 a	5.5 b	5.9 ab	5.8 ab	2.6 d	4.3 c	5.8 b	7.7 a	6.1	5.8
2	8.9	8 .0	8.4	8.1	3.3 c	6.7 b	7.6 b	10.8 a	8.3	8.5
3	5.3	5.6	5.7	5.4	0.6 d	3.9 c	5.6 b	7.0 a	5.3	5.7
4	6.6	7.0	6.9	6.2	6.1 ab	5.8 b	6.8 ab	7.5 a	6.4	7.0

† Means in each row and within each main treatment effect followed by the same letter are not significantly different ($P \leq 0.05$).

Table 4-5. Late-season wheat response to the main effects of fertilizer-seed blend storage time, fertilizer rate, and source. Total P uptake was measured at the Feekes 11.2 growth stage and grain yield at harvest.

	Storage time of fertilizer-seed blend (days)				Fertilizer rate (kg of P_2O_5 ha^{-1})				Fertilizer source	
Location	0	7	22	34	0	34	67	135	DAP	MESZ
	----- Phosphorus uptake (kg P ha^{-1}) -----									
1	10.1	9.8	9.0	9.4	7.5 ab†	7.9 b	10 a	10.7 a	9.4	9.7
2	13.2	13.4	12.9	13.9	8.9 b	13 ab	12.5 ab	14.5 a	12.7	14.0
3	12.7	13.2	13.6	13.6	7.0 d	11.3 c	13.3 b	15.3 a	13.3	13.2
4	14.6	14.5	14.6	14.3	12 ab	13.2 b	14.2 ab	16 a	14.2	14.8
	----- Grain yield (kg ha^{-1}) -----									
1	3125	3154	3115	2989	2079 d	2662 c	3061 b	3564 a	3140	3051
2	3931	3997	3862	3749	3103 c	3728 b	3888 ab	4039 a	3921	3848
3	4600	4639	4701	4516	2665 c	4340 b	4737 a	4766 a	4594	4633
4	4068	4174	3992	3989	3712	3889	4170	4096	4043	4068

† Means in each row and within each main treatment effect followed by the same letter are not significantly different ($P \leq 0.05$).

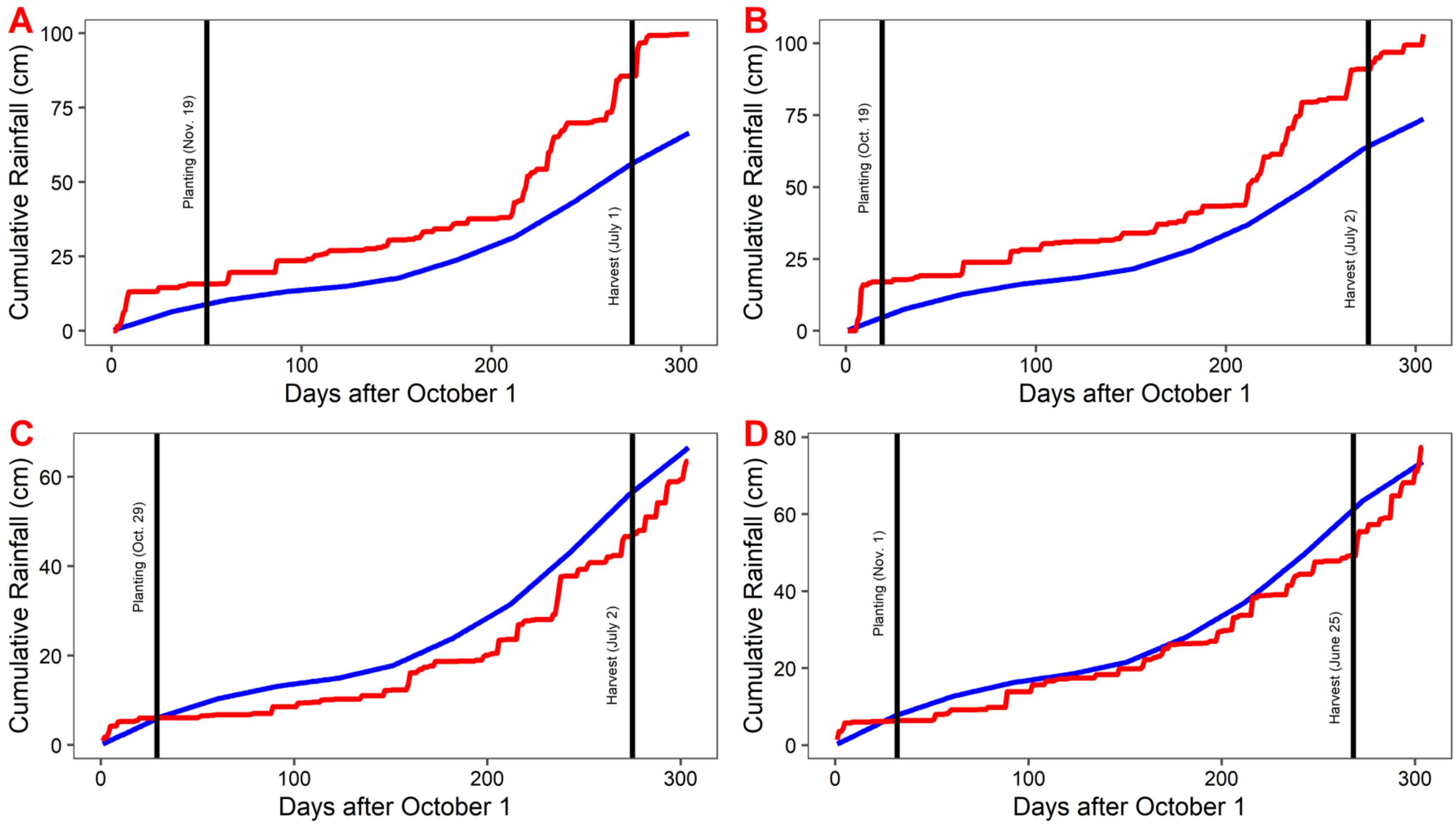

Figure 4-1. Cumulative rainfall by location with planting and harvest dates marked. The blue line represents the historical cumulative precipitation while the red represents the season cumulative precipitation for the wheat growing season. Precipitation data for Riley 2019 (A), Shawnee 2019 (B), Riley 2020 (C) and Shawnee 2020 (D)

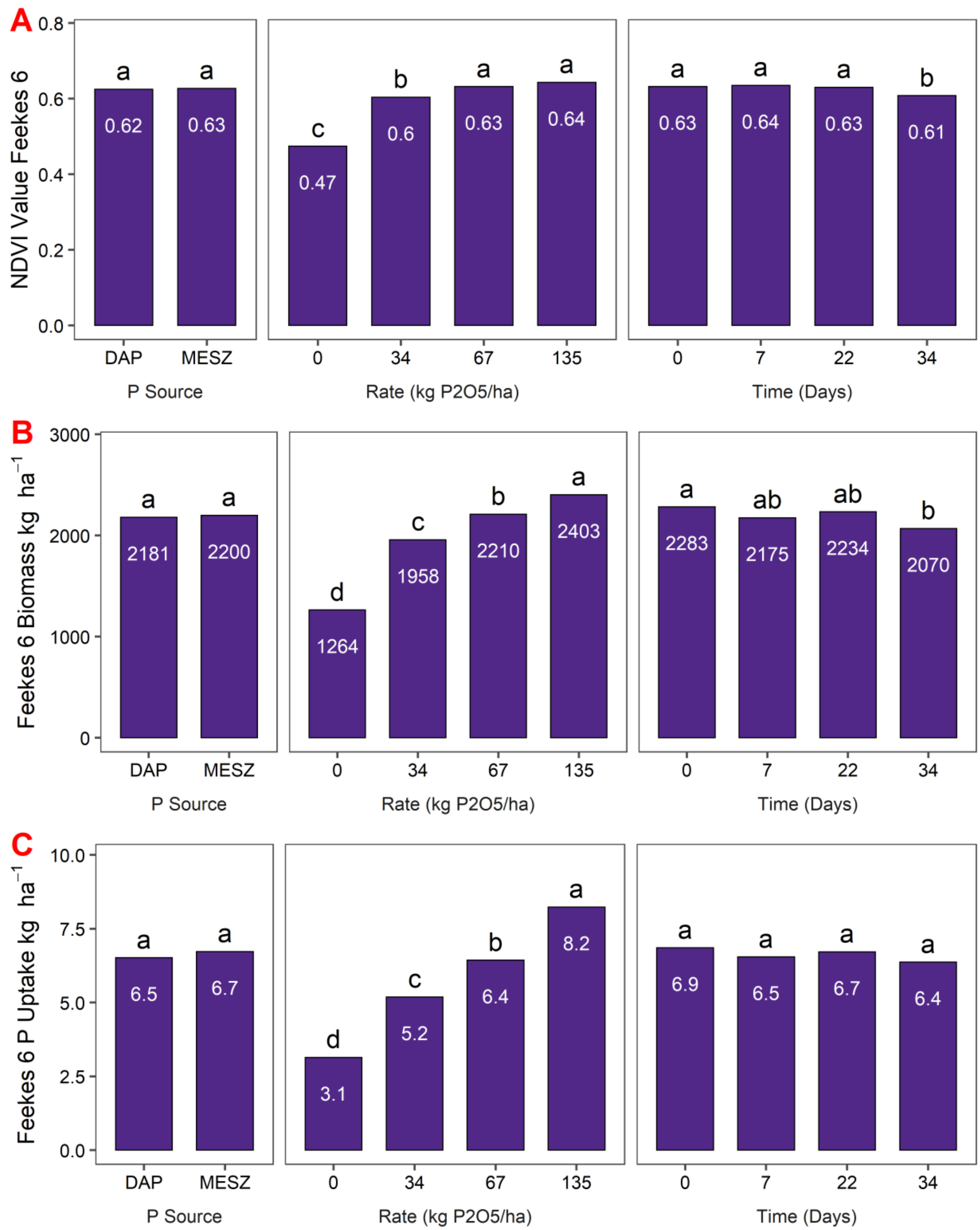

Figure 4-2. Average wheat response to P fertilizer source, rate, and blend duration across locations during the early vegetative growth stage. Feekes 6 NDVI (A), Feekes 6 biomass (B) and Feekes 6 P uptake (C). Location and blocks were set as random effects in the model and P fertilizer source, rates, and blend duration as fixed effect. Means with different letters are statistically different at the 0.05 probability level.

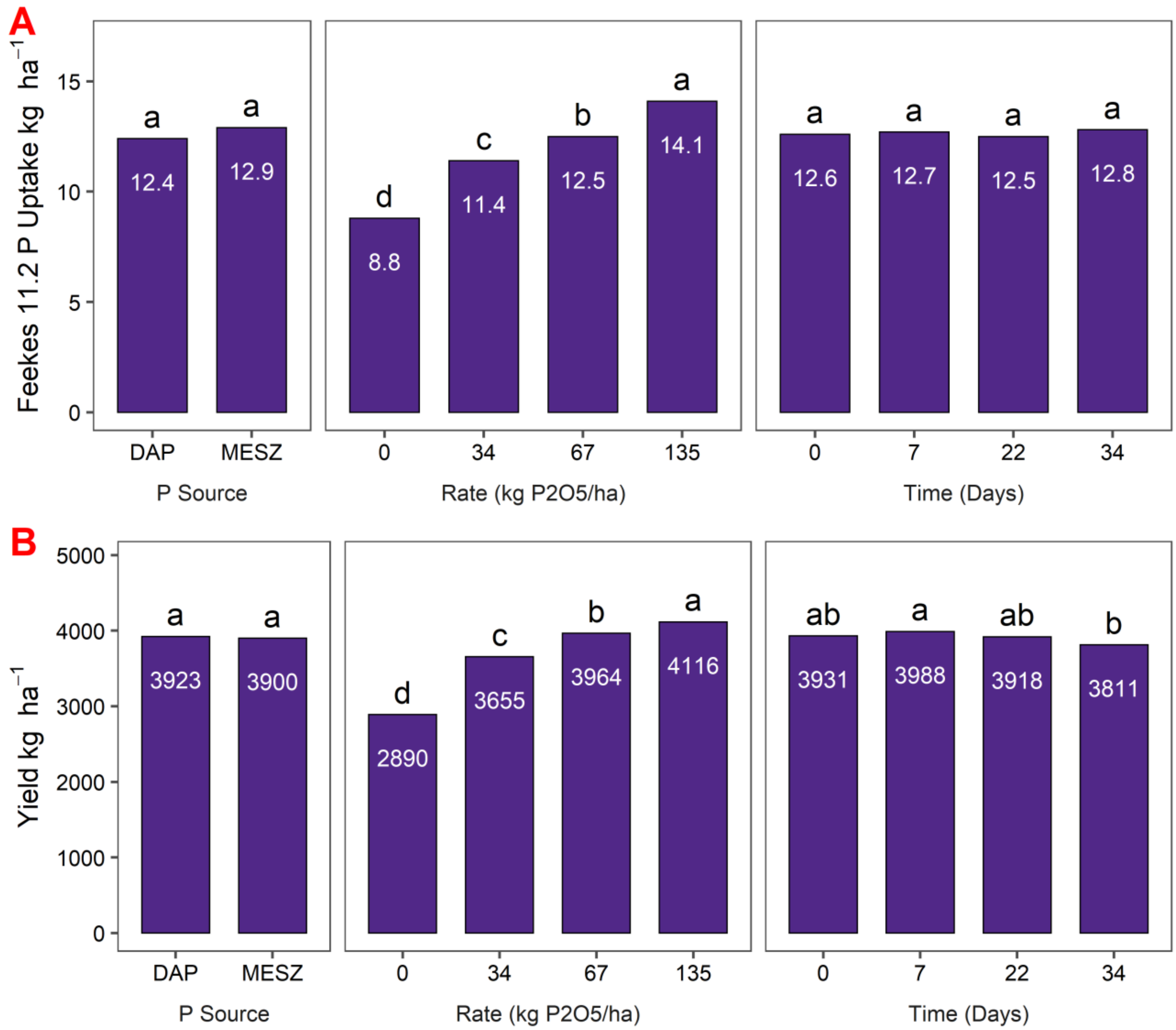

Figure 4-3. Average wheat response to P fertilizer source, rate and blend durations across locations during the late vegetative growth stage and harvest yield. Feekes 11.2 P uptake (A), harvest yield (B). Location and blocks were set as random effects in the model and P fertilizer source, rate and blend duration as fixed effect. Means with different letters are statistically different at the 0.05 probability level.

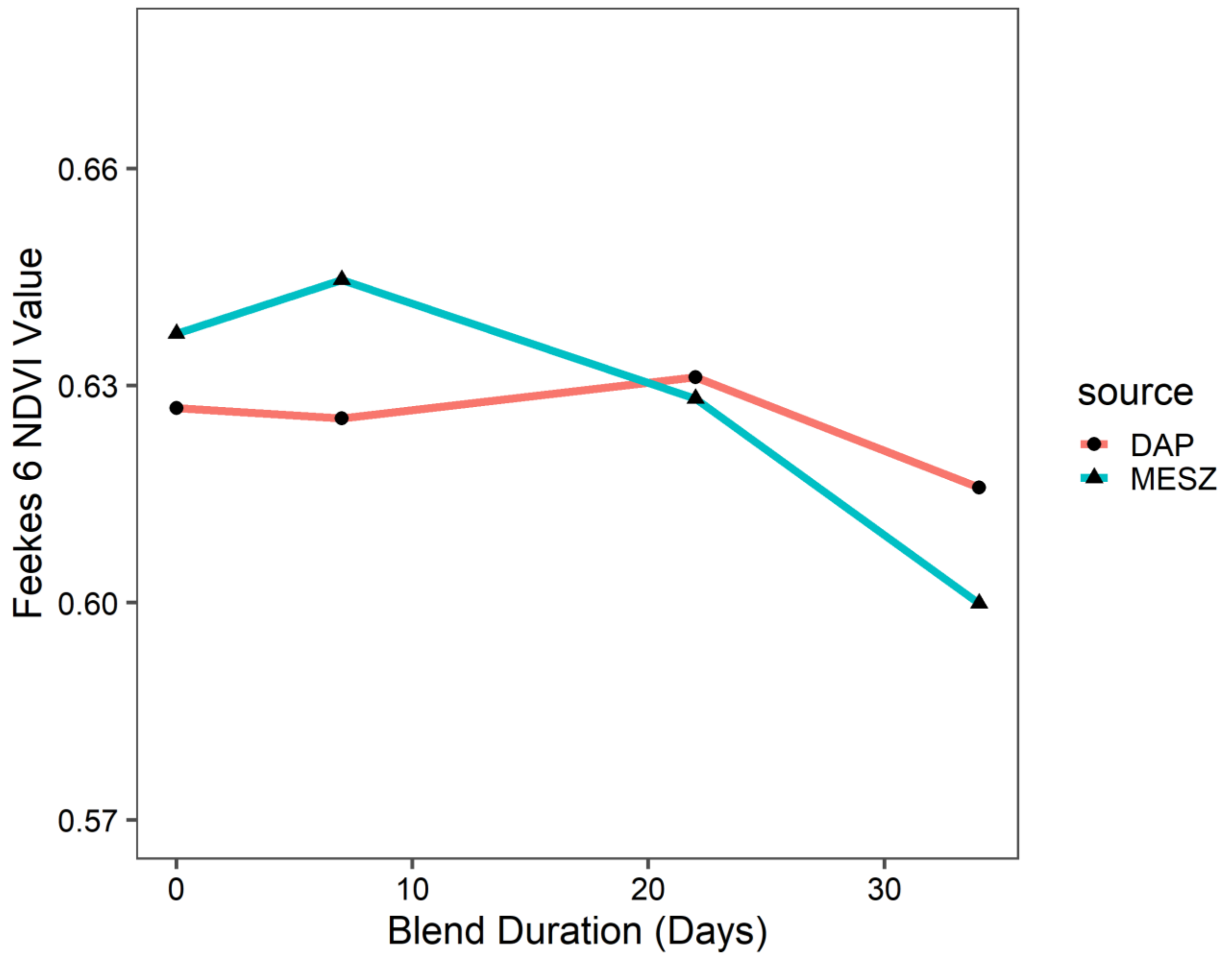

Figure 4-4. Normalized difference vegetative index collected at the Feekes 6 growth stage as affected by the interaction of fertilizer-seed blend storage time and fertilizer source. Location and blocks were set as random effects in the model and fertilizer source and fertilizer blend duration as fixed effect.

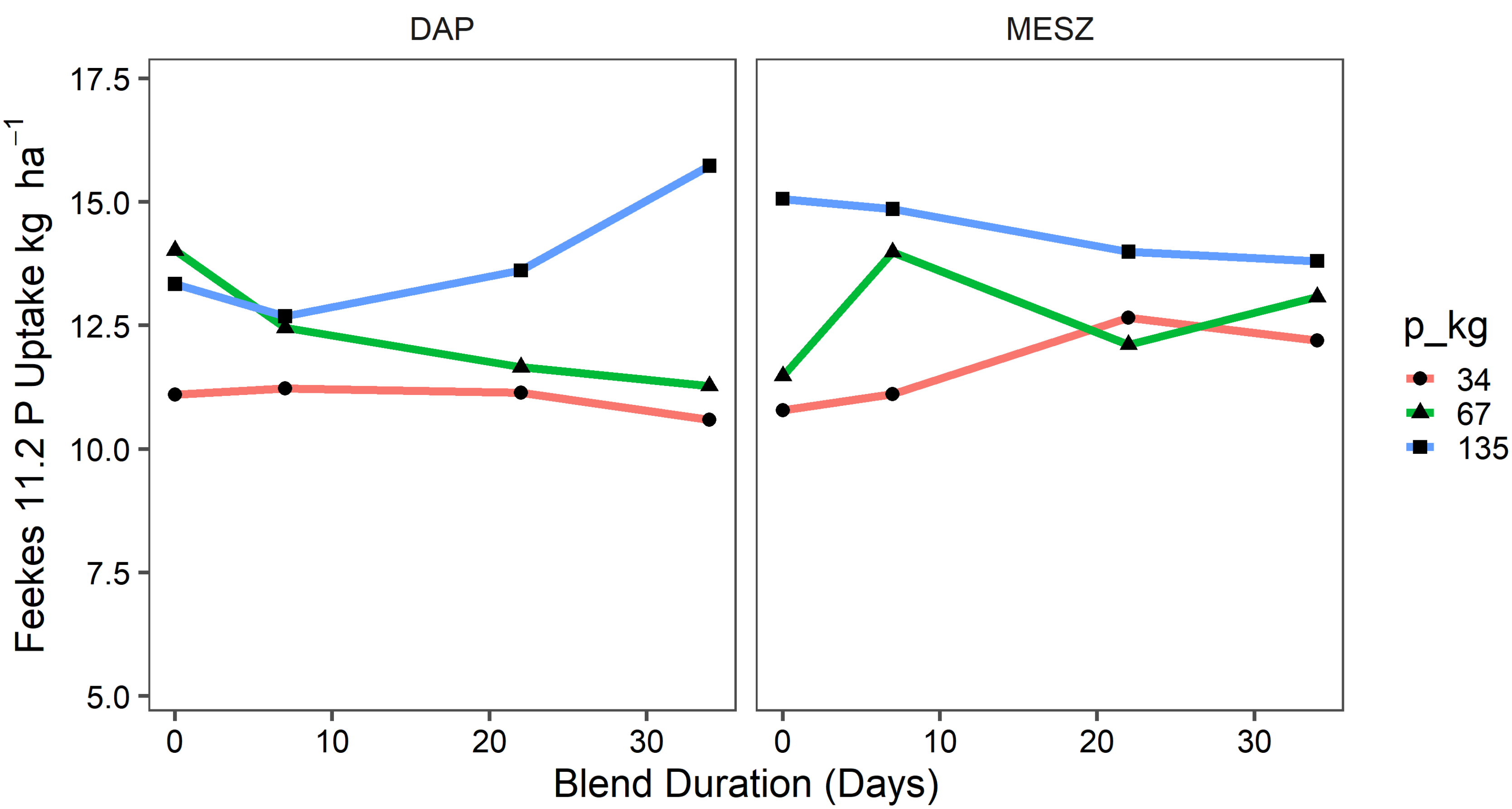

Figure 4-5. Feekes 11.2 P uptake as affected by the interaction of fertilizer source, fertilizer rate and fertilizer-seed blend storage time and fertilizer rate and fertilizer source. Location and blocks were set as random effects in the model and fertilizer source, fertilizer rate and fertilizer blend duration as fixed effect.

Chapter 5 - General Conclusions

Understanding how soil test P methods are affected by soil characteristics in Kansas is critical in identifying profitable winter wheat management practices. Soils that exhibit high pH and high calcium carbonates can significantly affect how a selected soil test P method correlates to winter wheat crop response. Results from our study show that the Mehlich-3 COL, Mehlich-3 ICP, Olsen, and Resin soil test methods correlate well with crop response across various soil types and properties in Kansas. The Bray-1 and H3A correlated well with crop response when soils did not exceed a pH of 7.45. The calcium carbonate concentration of the soil also played a role in crop response correlation. However, within this data set, pH was better able to describe the point at which the Bray 1 and H3A were affected and no longer correlated to other soil test methods. Results showed that the soil test P critical levels for winter wheat should be slightly higher than current guidelines for Kansas.

Sulfur soil test methods vary in their correlation to one another. This makes it a challenge when comparing results between methods. Overall, responses to S fertilization were limited when looking at crop responses like biomass or yield for either rate or source. However, significant effects were found when analyzing tissue samples for S concentration. Increases in S concentration or uptake were generally observed when S fertilizer rates were increased. In addition, small decreases in S concentration or uptake occurred when elemental S was used as an S source compared to other sources. This is likely linked to low S oxidation with elemental S during the winter and early spring months. Sulfur internal use efficiency for wheat was 425 kg of grain/kg S uptake based on our study.

Blending P fertilizer with wheat seed has mixed effects on the productivity of the crop. Across the majority of sampling timings, the source and blend storage duration had limited crop responses. The rate of P fertilizer played a role but was due to lower soil test P levels producing a positive crop response to fertilizer rather than a salt effect on the crop. Results from our study show flexibility in fertilizer application rates up to the highest rate in this study of 135 kg P_2O_5/ ha with negligible adverse effects. Recommendations for this application technique should be based on P fertilizer needs for a responsive wheat crop in low soil test P soil.

The studies included in this thesis comprised multiple soil types and evaluated different diagnostic tools for phosphorus and sulfur. The potential for wheat yield response varies for P and S, and was more common to attain significant yield increases to P management across multiple soils in Kansas. As an immobile nutrient in the soil, P can also benefit from placement near the wheat seed at planting; however, high fertilizer rates and blending options before planting can pose a risk for seedling damage. Results from this study showed that common P fertilizer sources can be applied at high agronomic rates with no measurable damage to wheat growth and yields. Phosphorus fertilizer's overall response for low testing soils was high, and P fertilizers can be applied with the seed to improve wheat response while combining field operations.