Variation in Response to N and S Among Spring Wheat Genotypes Grown on Irrigated and Non-irrigated Soils

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Research Questions

1) Study the effect of sulfur rate on spring wheat grain yield and protein concentration and quality.

2) Determine whether spring wheat varieties differ in the potential response to sulfur fertilizer.

3) Evaluate if plant tissue analysis (flag leaf samples collected at anthesis) can indicate the responsiveness of spring wheat varieties to N or S

4) Determine the economic optimum nitrogen rate for spring wheat grown under irrigation.

Results

Wheat Sulfur Studies – Table 1 and 2 summarize planting data and initial soil test information for the sulfur trials.

Statistical significance, by location, for spring wheat grain yield, grain protein concentration, and the total amount of protein produced per acre is summarized in Table 3. Main effect averages are given in Table 4 summarized by location and for the 3 location average for the 2016 growing season. As expected, yield differed consistently among the varieties at all locations. RB07 produced the greatest yield across sites followed in order by Mayville, Faller, Select, Glenn, and lastly, Vantage. Grain protein concentration was greatest for Vantage and Glenn while Faller producing the least. Total protein produced per acre was greatest for the top yielding variety. As in previous years, Staples had the highest average grain protein compared to the other two locations. It is unclear why Staples has had a higher potential protein concentration. However, the inverse relationship between protein and yield may explain the high protein concentration as wheat grain yield was substantially lower at Staples than the other two locations.

There was no detectable increase in yield from sulfur at the Crookston and Fergus Falls locations (Table 4). In 2014 the 7.5 lb S rate produced a significant yield increase at Staples and there was no further increase to the 15 lb rate. In 2015 grain yield was again increased at Staples but only for the 15 lb S rate. The actual yield increase was less at 4 bu/ac in 2015 than occurred in 2014 (2014 and 2015 data are included in previous reports and not shown in the current report). There was a yield decrease due to the application of the 15 lb S rate at Staples in 2016. The reason for the decrease was not clear. In 2016, grain yield was increased when 15 lbs of S was applied at Fergus Falls. This response to S at Fergus Falls is the first that has been found outside of the Staples location.

Grain protein content was increased by sulfur at Staples. At Staples, the 7.5 and 15 lb S rate produced an increase of 0.2-0.3% protein. Average protein levels were above 14% so the increase in grain protein concentration would not have resulted in a discount. Grain protein produced on a per acre basis was only impacted by sulfur at Staples due to impacts of S on both grain yield and protein concentration. Grain protein concentration and total protein produced per acre was not increased or decreased at Crookston or Fergus Falls.

Asparagine data was collected on samples taken from the 2014 sulfur studies (Table 5). Asparagine is important as it is an indicator of the production of acrylamide. Acrylamide can be produced during baking or frying and can have negative health impacts on humans. Asparagine content in the wheat grain varied among varieties across locations. Glenn typically had the lowest asparagine content followed by Vantage and Faller. Past research on hard red winter wheat has shown that sulfur can reduce the amount of asparagine in the grain. For hard red spring wheat, application of sulfur decreased asparagine content at Staples and Kimball. In both cases the 7.5 lb S rate produced the greatest decrease in asparagine content. It was interesting that the content of asparagine was higher at Staples than the other locations. The higher asparagine content at Staples could have been a result of higher protein content at Staples compared to the other locations. Samples were saved from the 2015 site to test for asparagine content if funds allow.

Wheat grain yield and grain protein concentration data were summarized across years for individual environments of Crookston, Fergus Falls and Kimball, and Staples (Table 6). These environments were chosen as they represent, respectively, an increase in the likelihood of response to S fertilizer. Overall, grain protein and yield were only increased at Staples. The increase was a result of the 7.5 lb S per acre application rate. There was no further increase, on average, for the 15 lb S rate versus the 7.5 lb rate. On average, grain yield was increased by 4 bu/ac with an average increase of 0.5% in grain protein concentration at Staples. The lack of an increase in grain yield and protein concentration at the other two environments indicates little potential of an economic benefit of S for wheat based on the current structure for marketing spring wheat. It should be noted that the amount of S applied in the irrigation water was measured at Staples but the totals were negligible over the years studied. In other studies the amount of S supplied can be substantial thus the chance for a response to fertilizer S may be greater on sandy irrigated soils but the need for S may be low due to S supplied through irrigation water.

Figure 1 summarizes flag leaf sulfur concentration by variety across the six locations conducted between 2014 and 2016. One question we were to address is weather there were differences among the varieties in their distribution of sulfur in the flag leaf tissue following application of sulfur fertilizer. The varieties were selected because they showed some differences in preliminary surveys of variety trials. Flag leaf sulfur concentration was impacted by the application of sulfur across the sites but there was no indication that there was a differing effect by variety (no significant interaction between variety and sulfur rate). The lack of impact of variety on sulfur content could be as a result of the rates being used are too high to achieve differences or the fact that the previous research correlated sulfur content of individual varieties with the average sulfur concentration based on location. Grain S concentration was also measured (Figure 2) with similar results as flag leaf S concentration. Grain sub-samples were collected from 2016 but the samples are still being analyzed in the lab and the data are not available.The question remains as to whether flag leaf S concentration could be used to predict were S deficiencies will occur.

Figure 3 summarizes the relationship between relative yield produced by the control with no S versus the concentration of S in the flag leaf tissue. Relative yield data was generated by dividing the yield of the control for each variety by the average yield of the S fertilized plots at each study location each year. Relative yield data was generated to account for yield variation among the varieties. The dashed line represents the best fit relationship, which indicated that relative yield and flag leaf S were related but the prediction of yield by flag leaf S is poor (R2=0.09). The data for Staples were analyzed separately (not shown) and was found to better correlate but the relationship was linear so a critical level could not be established. Using 95% of maximum yield as a low end of a sufficiency range and the plateau as the upper end, the data indicates that a concentration ranging from 0.20-0.35% S should indicate that sufficient S is present at a location. Taking a sample at anthesis will not provide any useable data for the given growing season and can only be used in subsequent years as an indicator of where S deficiency may occur. There was no indication that the sufficiency level varied by variety thus the data in Figure 3 is shown across varieties.

Nitrogen Fertilization of Irrigated Wheat – Table 8 summarized pertinent soil test data collected prior to the study being established at Staples in 2016.

Data in Table 9 indicated that wheat grain yield, protein concentration, and protein yield were impacted by variety and nitrogen in 2016. There was on significant interaction for protein which indicates some variation in the response to N by variety in 2016. This interaction was not found for grain yield or total protein produced per acre which indicates that the amount of N required to maximize grain yield was the same for all varieties. Current N guidelines are based on yield goal recommendations. The data from 2016 as well as previous years indicates that there is no relationship between grain yield or protein production and the amount of N required. Grain protein concentration may be impacted due to variations in how protein is accumulated by varieties but the data supports the decoupling of yield goals with N guidelines for hard red spring wheat.

The N rate at the maximum return to nitrogen (MRTN) was calculated by year and for the three year average (Table 11). Since the protein concentration tended to be high (above 14%) discounts were not factored in to the calculator. The actual MRTN value varied by year but the suggested N rate for a typical price ratio of 0.05-0.10 ranged from 130 to 155 lbs of N which is less than the MRTN previously calculated for non-irrigated wheat [200-250 lbs of applied N + residual N in the top two feet (data generated though an unpublished study funded by AFREC)]. The residual N in the top 2 feet averaged around 25 lbs of N for Staples which was not substantial. One source that was not accounted for was the amount of N applied in the irrigation water which may account for the difference depending on the number of times irrigated and the N concentration in the irrigation water. Nitrate in the irrigation water also could account for the higher protein concentrations at Staples for both the N study and the S study.

Table 12 summarizes the statistical analysis of grain yield, protein concentration, protein yield (on a per acre basis), and flag leaf N concentration across years. Individual data for the measurements are summarized in Figures 4, 5, 6, and 7, respectively. Nitrogen rate influenced grain yield, protein yield, and flag leaf N but they did not interact. The probability values were close to the accepted level (P<0.10) for the interaction terms for all three variables. The interaction was significant for grain protein concentration. Figure 5 shows the impact of N on grain protein for each of the three varieties. From the data the variety RB07 appears to maximize protein around the 240 lb rate which the other two show increasing protein beyond the 300 lb N rate which was the highest applied in the study. The interaction indicates some evidence that the varieties differ in the accumulation of protein. However, the total protein produced was nearly identical among the varieties in which there was no difference in yield or total protein produced according to the three-year average (Figures 4 and 6). The variety Faller was the only one variety which produced less than 14% grain protein on average with 0 to 60 lbs of N applied.

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Flag leaf nitrogen concentration was measured and the data are reported in Figure 7. There was no indication that the varieties varied in their response to N in the flag leaf tissue. The P value was close to significance but the main effect of variety and nitrogen rate were the only significant factors for the study. The maximum N concentration in the flag leaf tissue was similar for Faller and RB07 and less for Mayville. There was no indication that the accumulation of N following fertilizer application varied by variety. Figure 8 summarizes relative grain yield and protein yield data versus flag leaf N to determine if the concentration of N in the flag leaf could be predictive of the two measurements. Relative data was generated to balance out genetic difference in yield and protein production among the varieties and among the years. In both cases there was a direct relationship between flag leaf N concentration and relative grain yield and protein yield of the three varieties. However, the relationship was linear in both cases making it difficult to determine an optimum N value from the figures. In both cases the values seem to cluster near 4.5% which may indicate that if more data were collected, the critical value would possibly have been within that range. The data also indicates that the critical value may be similar among the varieties. At the time of sampling the chance that applied N would increase yield is very low. Thus, tissue analysis may give an indication of the potential reduction in yield or protein production but it will not indicate the achievable yield or protein produced on a per acre basis.

Application and Use

Changes were made when the Fertilizer Guidelines for Wheat in Minnesota publication was updated in 2012. Some changes included a general framework of S guidelines for eroded low organic matter soils (less than 2.0% organic matter in the top six inches). We did not have any non-irrigated locations with an organic matter concentration 2.0% or less in the present study. We can only assume responses would be similar for irrigated and non-irrigated soils as long as the irrigation water is not providing available S to the plant which would lower the rate required. Currently, 25 lbs of S is suggested for wheat grown on sandy soil with low organic matter. If irrigated, our data suggests that wheat can benefit from S but the rate required is less, 10 lbs of S per acre should be sufficient for both irrigated and non-irrigated wheat. Currently 10-15 lbs of S applied using a sulfate-S source is suggested for wheat grown on soils with an organic matter concentration of 3.0% or less. The data currently supports this suggestion.

There may be some benefit in baking quality when S fertilizer is applied regardless of whether there is a yield or grain protein concentration response to S. Until protein premium/discounts reflect quality over the quantity there may be a limited impact to the bottom line of a wheat grower. Asparagine (an amino acid) data generated from the 2014 data was used to look at as a precursor of acrylamide formation upon baking. The concentration of asparagine decreased when S was applied even without a resulting increase in grain yield or protein concentration. Since grain S was typically increased when S was applied we can assume the production of cysteine and methionine (S containing amino acids) may be increasing when S is applied and may be offsetting the production of asparagine. Follow up research is needed to study the acrylamide issue, particularly looking at the impacts of S on asparagine and reducing sugars which are both important in the formation of acrylamide. Acrylamide is a known carcinogen that can be produced during baking at high temperatures.

Nitrogen response data for irrigated wheat indicates that 150 lbs of applied N split between an application after seeding and near jointing was sufficient for irrigated wheat. Some cautions on this data should be exercised as the amount of N in the irrigation water was not accounted for in that value and there was significant variation in the economic optimum N rate by year. Local knowledge should always be used when determining how much N should be supplied based on all N sources to the plant.

Our goal for comparing the varieties was to determine if tissue sampling could be used to determine responsiveness of varieties to N or S. Since there was no evidence that a variety by N or S interaction occurred, it is unlikely that the tissue data had much value in determining whether N or S would benefit one variety over another. One caution about this work is that since yield was only affected at one location it is hard to draw hard conclusions unless the effect can be replicated. More locations and one or two additional years of funding would greatly benefit this project to determine if similar effect can be replicated across sites and years. Overall, our data from this study and past research indicated that significant caution should be taken when using plant tissue samples for guiding fertilizer application as a specific critical level can be difficult to determine for use and late season tissue samples cannot be used during the year collected and have more value for subsequent years' management.

Materials and Methods

Small plot sulfur fertilization studies on non-irrigated soils will be established alongside two spring wheat variety trials. The locations were Crookston, Fergus Falls, and Staples. Staples was the only site where supplemental irrigation was supplied.

Six wheat varieties will be selected using the stability analysis conducted for spring wheat flag leaf tissue among varieties in 2011 and 2012. Variety selection will not be based on current planting trends or popularity. Two varieties will be selected that were considered in the high, average, and low response to sulfur categories and that vary in protein and yield potential. The varieties selected are Faller, Vantage, Select, Glenn, Mayville, and RB07. Sulfur rates used will be a non-fertilized control (0 lb S), 7.5, and 15 lbs S per acre. Sulfur was applied to the soil surface after planting. The source of sulfur was ammonium sulfate (21-0-0-24). Nitrogen was applied to balance the rate of nitrogen applied with the high rate of ammonium sulfate. Nitrogen, phosphorus, and potassium were kept at non-limiting rates according to current recommendations.

The nitrogen trial consisted of only three of the varieties utilized in the S study (Faller, Mayville, and RB07) and six nitrogen rates (0, 60, 120, 180, 240, and 300 lbs of N per acre). Nitrogen was applied as urea (46-0-0) applied at two times with half of the nitrogen applied after seeding but before emergence and the remaining applied near jointing. Additional nutrients (P, K, and S) were applied as a pre-plant application. The varieties selected were done so based on previous flag leaf tissue data for N similar to selection characteristics outlined the preceding paragraph.

Soil samples were collected prior to fertilizer application at both sites at 0-6, 6-12, and 12-24" depths (1 composite sample was collected per site). 0-6" samples were analyzed for P, K, pH, and organic matter. All samples were either analyzed for nitrate-N or sulfate-S depending on the study. Flag leaf samples were collected from the S and N studies at anthesis by sampling 25-30 leaves. Leaves were dried, ground, and analyzed for total N or S depending on the study. Grain yield was measured for all plots adjusted to 13% moisture and a sub-sample of grain will be collected and analyzed for protein concentration (reported at 12% moisture) by NIR. Grain samples for the S study will be analyzed for total S.

Economic Benefit to a Typical 500 Acre Wheat Enterprise

Assuming a wheat price of $6 per bushel, the response at Staples would result in an additional $24 per acre in added crop value across the varieties. If S cost was $0.50 per lb of S, the rate needed to increase yield (15 lb per acre) would cost a grower $7.50 per acre resulting in a net profit of $16.50 per acre and would total $8,250 for a 500 acre operation. When S is deficient, application of S is typically highly profitable for hard red spring wheat.

Even with the low total cost associated with the rate of S needed to increase yield, if a site is not responsive a grower should highly consider using money intended for S for nitrogen especially in years where yield potential and protein discounts are greater. Even with increases in grain protein due to S, the concentration of grain protein was above 14% thus an increase would not results in any significant economic benefit to a grower unless a premium is being paid for protein. The overall increase in protein on average was only one to two fifths of a percent. Overall, S should not play a role in making decisions on fertilizer application for increasing grain protein concentration.

The economic benefit for the application of N is factored in to the data presented in Table 11. The values represented in Table 11 indicate the point at which a dollar invested in N will return a dollar in crop value. Rates applied beyond values will typically result in a negative return on average. These values are adjusted for the cost of N relative to the crop value. These data are meant to be an average value or a starting point and may need to be adjusted for other incidental sources of N or N loss in a given year. Only a single split application was used. Thus additional strategies using more splits may need to be research to determine if two application of N is sufficient of if the N should be split more times over the growing season.

Related Research

A S study was concluded in 2009 which was funded by the Minnesota Wheat Growers that studied the effect of S source, rate, and timing for wheat grown on soils with relatively high concentration of organic matter. This current study provided supporting data for the previous research but focuses on questions received following the previous study on whether we would expect response to S to be greater for varieties which are greater yielding than Glenn which was used in the previous research. We are also following up on information collected in a study funded in 2011 and 2012 that included a survey of flag leaf tissue nutrient concentration. The current research will determine if there is any value in tissue concentration data and whether tissue concentration can help predict variety responsiveness to a specific fertilizer.

A study funded by AFREC is currently using the grain samples collected in this study for analysis of amino acids. This analysis will be used with NIR data to develop curves to screen for a rough estimate of amino acid concentrations for un-ground and ground samples. Our emphasis is to allow for us to screen for some of the sulfur amino acids by a quick and cheap method. For someone whom wants exact values there still is a need to run chemical analysis on the grain.

Recommended Future Research

Further research work is needed to determine the potential health benefits that S fertilization may play in beyond potential for increase in grain yield and protein concentration. Research connecting the application of S with a decrease in acrylamide is needed for hard red spring wheat. The asparagine data presented in this report shows that there may be an impact of S in reducing asparagine but reducing sugar concentrations are needed to give a full picture of the potential for acrylamide formation. In addition, measuring acrylamide directly on bread baked with wheat would be beneficial to regress acrylamide content with asparagine and reducing sugars to determine if the relationship between the two is similar in hard red spring wheat as for other crops.

Publication

Kaiser, D.E., and J. Wiersma. 2016. Variation in response to sulfur among hard red spring wheat varieties. Agron. Abs. CD-ROM. ASA-CSSA-SSSA. Madison, WI.

Appendix

Table 1. Trial location, planting information, and monthly total precipitation for spring wheat S rate studies.

					Monthly Total Precipation		
Location	County	Soil Type	Soil Texture	Seeding Date	May	June	July
					-----------inches--------		
Crookston	Polk	Wheatville	Sandy Loam	15-Apr	6.7	6.8	4.6
Fergus Falls	Otter Tail	Formdale	Clay Loam	13-Apr	1.0	1.6	7.2
Staples	Wadena	Verndale	Sandy Loam	13-Apr	1.7	1.9	10.3

Table 2. Spring soil test averages across replication for Spring wheat S trials.

	Soil Test (0-6:) ł				
Location	P	K	SOM	pH	Sulfate-S ‡
	---ppm----		---%---	-	--lb/ac--
Crookston	5	117	4.1	8.2	208 §
Fergus Falls	21	191	4.8	6.3	28
Staples	16	69	1.8	6.8	16

ł P, Bray-P1 phosphorus; K, ammonium acetate potassium; SOM, soil organic matter; pH, soil ph ‡ 0-2 foot soil sulfate-S § Sample was collected from 0-1'

Table 3. Summary of statistical signficance of main effects of variety (V), sulfur rates (S), and their interaction (VxS) for spring wheat grain yield, protein concentration, and total protein produced per acre.

	Grain Yield ł			Grain Protein ł			Protein Yield ł		
Location	V	S	VxS	V	S	VxS	V	S	VxS
Crookston	<0.001	0.81	0.66	<0.001	0.63	0.60	0.10	0.02	<0.001
Fergus Falls	0.04	0.04	0.33	0.03	0.63	0.71	0.75	0.01	0.02
Staples	<0.001	<0.01	0.41	<0.001	<0.001	0.53	0.76	0.50	0.57

Table 4. Summary of hard spring wheat garin yield, grain protein concentration, and protein production per acre for individual varieties at Crookston (CR), Fergus Falls (FF), and Staples (ST) Minnesota during 2015. Average values were calculated for variety and sulfur main effects by and across locations (AVG).

	Grain Yield ł				Grain Protein ł				Protein Yield ł			
Variety	CR	FF	ST	AVG	CR	FF	ST	AVG	CR	FF	ST	AVG
	---Bushels/ac (@13%)-----				-----%(@12%)------				------pounds/ac (13%)-------			
Faller	94.4a	83.7b	40.5d	73.0bc	14.2e	15.9b	18.3e	16.1e	804	797b	444c	682d
Glenn	86.0b	83.3b	43.9c	71.1cd	15.5b	16.5a	19.5b	17.2b	799	822b	513b	711bc
Mayville	86.6b	89.2a	45.5bc	73.6b	15.2c	16.1ab	18.7c	16.7c	788	861a	510b	720ab
RB07	95.6a	84.2b	51.6a	77.1a	14.5d	15.8b	18.4de	16.2de	834	803b	572a	737a
Select	86.0b	83.5b	47.7b	72.4bc	14.8d	15.8b	18.5cd	16.4d	764	788b	530b	694cd
Vantage	83.2b	815b	44.7c	69.8d	15.9a	16.5a	19.8a	17.4a	795	805b	530b	710bc
S Rate (lb/ac)												
0	89	832b	46.5a	73	15.0	16.2	18.3b	16.5b	799	807b	510b	705
7.5	88	828b	47.4a	73	15.0	16.0	19.0a	16.7ab	790	792b	840a	708
15	89	86.7a	43.1b	73	15.0	16.1	19.2a	16.8a	802	839a	500b	714

ł within columns for each main effect, numbers followed by the same letter are not statistically significant at P≤0.05 probibility level.

Table 5. Effect of variety and sulfur rate on asparagine content in the wheat grain collected from 2014 at Crookston, Kimbal and Staples.

	Grain Yield ǂ			
Variety	**CR**	**K**	**ST**	**AVG**
	-----umol/gram (@14%)------------			
Faller	3.5c	3.9c	8.6b	5.2c
Glenn	3.3c	3.1d	5.1c	3.5d
Mayville	4.6b	5.8a	10.4a	6.6ab
RB07	3.7c	5.2b	10.1a	6.3b
Select	5.4a	5.2b	10.3a	6.9a
Vantage	3.1c	3.7c	8.1b	4.9c
S Rate (lb/ac)				
0	3.7	4.7a	9.7a	6.0a
7.5	4.0	4.4b	8.6b	5.5b
15	4.0	4.3b	8.0b	5.3b

ǂ within columns for each main effect, numbers followed by the same letter are not statistically significant at P<0.05 probablily level.

Table 6. Summary of statistics for the analysis of grain yield and protein concentration across years for each given environment.

	Grain Yield			Grain Protein Conc.		
Environment	Variety	Sulfur	VxS	Variety	Sulfur	VxS
	---------------------P>F---------------------					
Crookston	<0.001	ns	ns	<0.001	ns	ns
Kimball/FF	<0.01	ns	ns	<0.001	ns	ns
Staples	<0.001	0.01	ns	<0.001	<0.001	0.05ǂ
All	<0.001	0.03	ns	<0.001	<0.01	ns

ǂ Grain protein was not increased by S for Fall and RB07.

Table 7. Summary of Grain Yield and Protein Data collected at Staples from 2014-2016

S Rate	Yield	Protein
lb S/ac	**-bu/ac-**	**-----%-----**
0	63.6b	17.7b
7.5	67.6a	18.1a
15	67.0a	18.3a

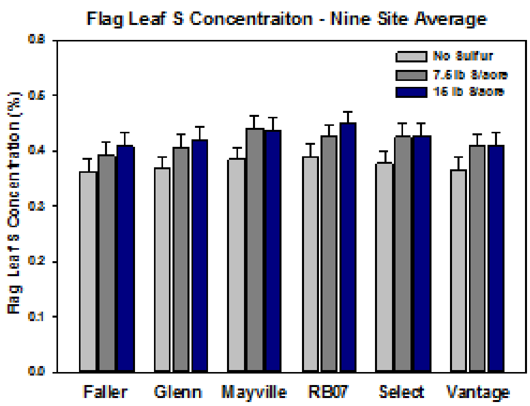

Figure 1. Flag leaf S concentration summarized by variety across six locations from 2014-2016

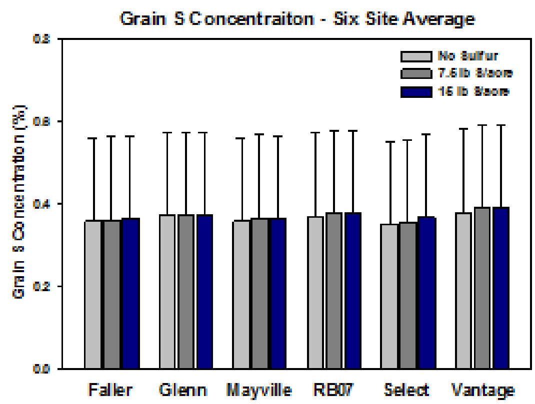

Figure 2. Grain S concentration summarized by variety across six locations from 2014-2015

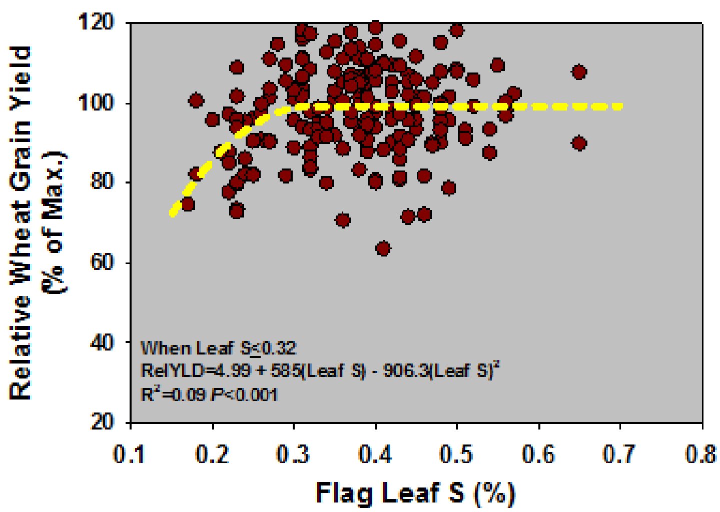

Figure 3. Prediction of relative wheat grain yield using flag leaf S concentration

Table 8. Spring soil test averages across replications for Spring wheat N trial

	Soil Test (0-6") ǂ				
Location	**P**	**K**	**SOM**	**pH**	**Nitrate-N ‡**
	----ppm----		**----%---**		**--lb/ac--**
Staples	14	80	1.5	7.3	24

ǂ P. Bray-P1 phosphorus; K, ammonium acetate potassium; SOM, soil organic matter; pH, soil pH.
‡ 0 to 2 foot soil nitrate-N

Table 9. Summary of statistical significance of main effects of variety (V), nitrogen rate (N), and their interaction (VxN) for spring wheat grain yield, protein concentration, and total protein produced per acre during 2016.

	Grain Yield ƚ			Grain Protein ƚ			Protein Yield ƚ		
Location	V	N	VxN	V	N	VxN	V	N	VxN
	---*P>F*---								
Staples	0.05	<0.001	0.15	<0.001	<0.001	<0.01	0.02	<0.001	0.17

Table 10. Summary of hard red spring wheat grain yield, grain protein concentration, and protein production per acre for individual varieties at Staples Minnesota from 2014-2016. Average values were calculated for data across six nitrogen rates and across years (AVG).

	Grain Yield ƚ				Grain Protein ƚ				Protein Yield ƚ			
	2014	**2015**	**2016**	**AVG**	**2014**	**2015**	**2016**	**AVG**	**2014**	**2015**	**2016**	**AVG**
	-----bushels/ac (@13%)--------				------------% (@ 12%)-----------				-------pounds/ac (@13%)---------			
Faller	69a	62a	36b	56	16.7b	14.0b	16.4c	15.7c	699a	543b	363b	534
Mayville	60c	63a	36b	53	17.8a	14.7a	17.6a	16.7a	646b	577a	387b	538
RB07	63b	57b	41a	54	17.7a	14.7a	16.9b	16.4b	667b	527b	424a	539

ƚ within column, numbers followed by the same letter are not statistically significant at *P*≤0.05 probability level.

Table 11. Summary of economic optimum nitrogen rates using the maximum return to N model for irrigated HRSW at Staples, MN in 2014-2016 assuming no discounts.

	Ratio of Price N: Price per bushel of spring wheat					
Location	**0.00**	**0.05**	**0.10**	**0.15**	**0.20**	**0.25**
	--------------lb N/acre----------------------					
2014	164	148	130	113	95	78
2015	238	205	171	137	103	70
2016	98	88	77	67	56	46
Average	179	154	131	109	86	64

Table 12. Summary of statistical significance of main effects of variety (V), nitrogen rate (N), and their interaction (VxN) for spring wheat grain yield, pretoin concentration, total protein produced per acre, and flag leaf N concentration across years (2014-2016).

	Grain Yield ƚ			Grain Protein ƚ			Protein Yield ƚ			Flag Leaf %N ƚ		
Location	**V**	**N**	**VxN**	**V**	**N**	**VxN**	**V**	**N**	**VxN**	**V**	**N**	**VxN**
	--P>F--											
Staples	0.26	<0.001	0.18	<0.001	<0.001	<0.01	0.94	<0.001	0.17	<0.001	<0.001	0.14

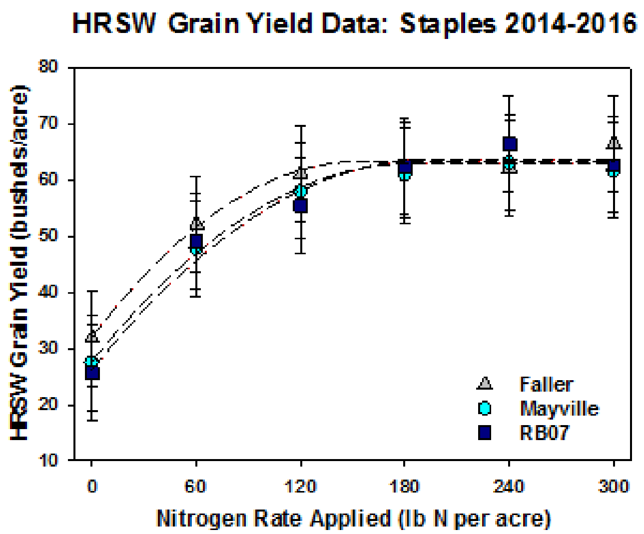

Figure 4. Effect of nitrogen on grain yield of three hard red spring wheat varieties grown under irrigation.

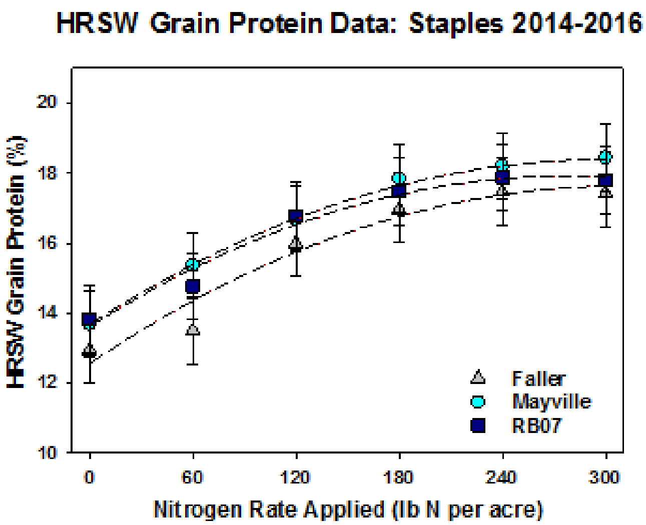

Figure 5. Effect of nitrogen on grain protein concentration of three hard red spring wheat varieties grown under irrigation.

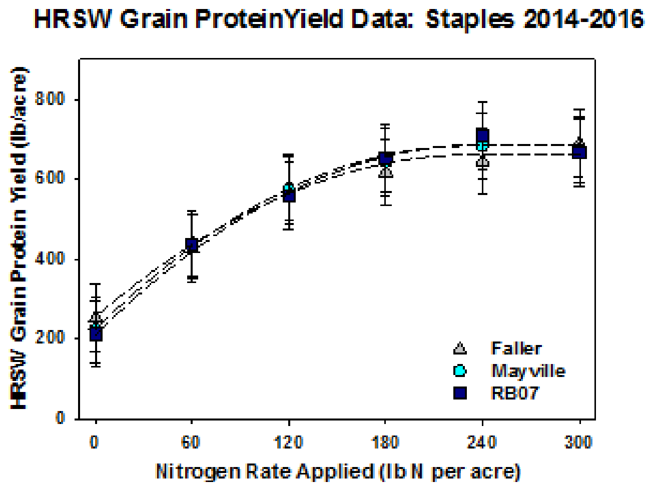

Figure 6. Effect of nitrogen on grain protein yield of three hard red spring wheat varieties grown under irrigation.

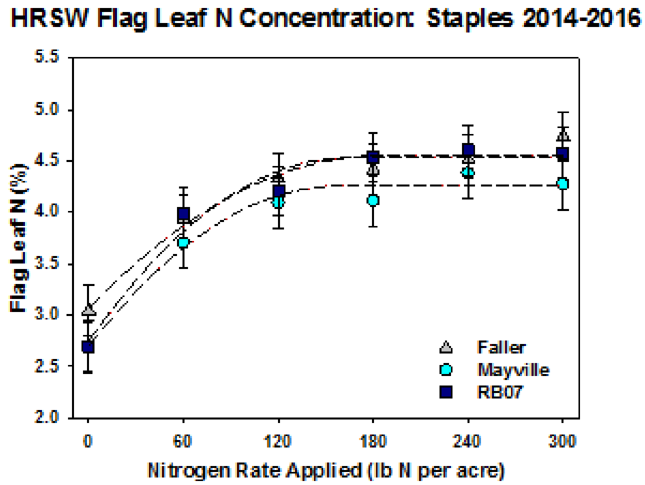

Figure 7. Effect of nitrogen on flag leaf nitrogen concentration at anthesis of three hard red spring wheat varieties grown under irrigation.

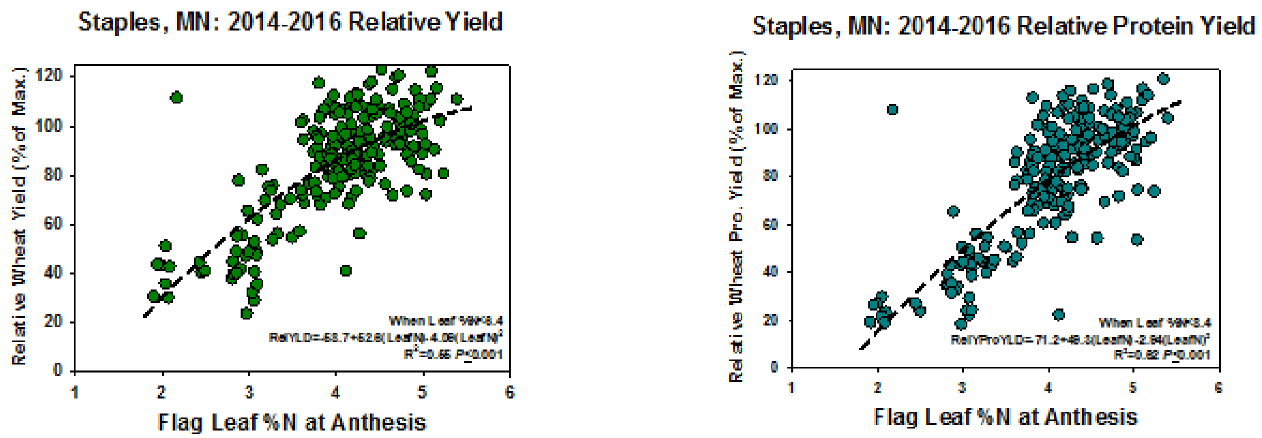

Figure 8. Relationship between flag leaf tissue N concentration for samples collected at anthesis and relative yield and relative protein yield (both expressed as % of maximum) across three wheat varieties for an irrigated soil at Staples, MN.