SULFUR MANAGEMENT IN CORN AND SOYBEAN CROPPING SYSTEMS

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ABSTRACT

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Earlier planting dates in cooler soils, increased nutrient removal by higher yielding cultivars, and reductions in atmospheric sulfur (S) deposition suggest that S application may require reassessment in Michigan for optimal corn (*Zea mays* L.) and soybean (*Glycine max* L.) growth. Field experiments were conducted on corn to examine the effect of a range of S rates with increasing nitrogen (N) rates on grain yield. The results indicated S availability may be sufficient when optimal N rates are applied promoting sufficient root growth to exploit the soil profile and acquire sufficient S quantities for optimal plant growth, however, yield responses to S may vary with environmental conditions (i.e., soil organic matter, soil texture, soil phosphorus (P) levels, and crop rotation). Residual soil S, mineralization of soil organic matter, S deposition from rainfall, and possible sub-soil S accumulation should all be considered for potential S contributions to determine when S application is justified. A soybean field trial investigated S placement at 25 lbs S/acre while evaluating the presence of a starter fertilizer (20 lbs N and 50 lbs P_2O_5/acre) in a 2 × 2 inch band. Although starter fertilizer had no significant effect on soybean grain yield or quality, in both study years soybean canopy density and greenness were greater with starter fertilizer application prior to V4. No significant effect on grain yield from S placement occurred in either study year suggesting sufficient S was cycling in the soil profile for optimal soybean growth to occur. Evaluating residual soil S and organic matter levels at the time of planting may be necessary to determine if S application is justified for optimal corn and soybean growth.

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LITERATURE REVIEW

Introduction

The state recommendations for S application in Michigan corn and soybean production have not been updated in the last 30 years. As one of the 17 essential plant nutrients S has an important role in many internal plant functions. Since the last update of Michigan S recommendations more stringent clean air laws have been mandated, thus requiring power plants and industrial facilities to significantly reduce the levels of contaminants emitted through smoke stacks (i.e., sulfur dioxide). Deposition of sulfate and hydrogen sulfide through rain fall has traditionally supplied agricultural soils with sufficient S to meet plant requirements. In the early 1970s Wisconsin field studies demonstrated that alfalfa obtained optimal yields when receiving as much as 44% of necessary S from the atmosphere (Hoeft et al. 1972). However, the U.S. Environmental Protection Agency (EPA) reports that from 1980 to 2010 S emissions have been reduced by as much as 83% (National Atmospheric Deposition Program, 2012). Sulfur escape into the atmosphere has been reduced by flue gas scrubbing technology. When coal is combusted the sulfur that is naturally present will combine with oxygen forming SO_2. Before exhaust gas is released into the atmosphere it passes through a mixture of lime or limestone and water. The SO_2 will react with the lime mixture and is removed from the exhaust. Data collected from the southern half of Michigan has shown that S-deposition (wet weight) has been reduced from 30 kg ha^{-1} in 1980 to less than 10 kg ha^{-1} in 2010 (National Atmospheric Deposition Program, 2012).

Reductions in S deposition are not the sole cause of more frequent occurrences of S deficiency. Single super phosphate (SSP, 0-9-0-12S) was once commonly used as a phosphorus source since its development in the 1800s. However, comparable cost between single super phosphate and higher analysis P sources has caused growers to use triple super phosphate (TSP, 0-45-0) and more commonly mono-ammonium phosphate (MAP, 11-52-0) (Hagstrom, 1986). The change to high analysis sources has resulted in less incidental S application with fertilizer mixtures.

In addition to the declines in sulfur deposition and incidental S application a concurrent increase of corn yields has occurred requiring greater S quantities to reach optimal yields. In 1970 the national average corn yield was 4.5 Mg ha^{-1}, in 2011 it was 9.2 Mg ha^{-1} (Capehart, 2013). Similar increases in average soybean yield have occurred over the same period of time increasing from 1.7 Mg ha^{-1} (1974) to 2.7 Mg ha^{-1} (2009). The increase of corn and soybean yields results in higher rates of S removal in grain.

Sulfur in Plants

Within plants S is well known as a component of amino acids cysteine and methionine (Taiz and Zieger, 2010). Sulfate (SO_4), the plant available form of S, when taken up by the plant undergoes reduction before reacting with o-acetylserine (OAS) to form the amino acid cysteine. Nitrogen's dependence on S is associated with OAS. In the event that a plant should become S-deprived, OAS will accumulate causing transporters of S to be up-regulated (increased expression of a gene to create plant organelles) to increase S uptake (Smith et al., 1997). Conversely when N becomes limited and the production OAS decreases, however, there is not a consequential increase of N transporters (Reuveny et al., 1980). An S deficiency will primarily limit N assimilation to form cysteine, but will not hinder N uptake (Zhao et al., 1998).

Apart from amino acid production S is essential for the reduction of nitrate. When plants take up nitrate (NO_3) it is reduced to form organic N (ammonium, NH_4). The pathway to convert nitrate to ammonium is a two-step process. First, nitrate reductase will reduce nitrate to nitrite, a highly reactive form of N that can be toxic. Plants will immediately transport the nitrite from the cytosol to the chloroplasts in leaves and plastids in the roots. In these compartments nitrite is converted to ammonium via nitrite reductase. Sulfur is a component of nitrite reductase. Nitrite reductase is a single polypeptide (long continuous, un-branched, chain of amino acids linked by amide bonds) containing a heme (a non-protein chemical compound that is bound to a protein and is required for the protein's biological activity) and an iron-sulfur compound known as ferrodoxin (Fe_4S_4) (Taiz and Zieger, 2010). Without available S plants would be limited in their ability to reduce nitrate.

Soybeans and Sulfur

Cysteine and methionine have an important role in soybeans due to higher marketable contents in this crop, as soybean seed typically averages 40% protein and 20% oil (Krishnan, 2005). In the United States, a large percentage of the soybean produced is used as livestock feed to satisfy the high S requirement of poultry and swine diets (Chung and Baker, 1992; Waldroup and Hellwig, 1995). The use of genetic technologies has been investigated to promote more favorable traits and pathways that may potentially increase the S-containing amino acids of soybeans and improve protein content (Krishnan and Chronis, 2003; Krishnan, 2005). However, one of the constraints preventing the development of soybean cultivars with higher protein potential is the inverse relationship between seed protein and yield in both determinate and indeterminate cultivars (Wehrmann et al. 1987). Determinate variety traits may enhance the ability to overcome the inverse relationship to produce high-yielding soybean cultivars with seed protein concentrations in excess of 450 g kg^{-1} (Wilcox and Guodong, 1997).

Sulfur uptake and remobilization at seed set in soybeans can be up to 800 µg S $plant^{-1}$ d^{-1} (Sexton et al., 1998a). A large proportion (56 to 59%) of total plant S accumulates in late vegetative development and is remobilized shortly thereafter for seed fill with only 10 and 20% being supplied from the pods and leaves, respectively (Naeve and Shibles, 2005). A reduction in seed protein quality occurs by the production of the β-subunit from the β-conglycinin storage protein which is frequently encountered in S deficient environments due to the lack of S containing amino acids (Tierney et al., 1987). However, even in high S providing environments 10 to 20% of the seed storage proteins consisted of the β-subunit of β-conglycinin (Sexton et al., 1998b). The high percentage of low grain quality leads to the hypothesis that in high S-environments the production of high quality seed protein is limited by the reduction of sulfate and synthesis S-containing amino acids (Sexton et al., 1998b).

The timing of S availability may be crucial in protein development. The ratios of glycinin to β-conglycinin are the same for soybeans S deficient throughout vegetative growth stages and then received adequate S throughout reproductive stages as soybeans that received adequate S throughout the entire plant life cycle (Sexton et al., 1998c). A greater part of S assimilation occurs in the reproductive stages, thus the magnitude of S assimilation and synthesis of S-containing amino acids may be limited if the majority of S is acquired over a short period.

The contribution of mobilized N on soybean seed quality and yield was assayed and an N mobilization efficiency of 70% was achieved which illustrated that the availability of N in aboveground plant biomass was two to four times greater than what was required in mature seeds (Loberg et al., 1984; Vasilas et al., 1995; Naeve et al., 2008). Therefore, acquiring S may be a greater limiting factor than N to obtain greater soybean yields. Sulfur has also demonstrated effects on soybean root development and the plant-soil microbial relationship (Zhao et al., 2008). In a greenhouse study S applications of 30 and 60 mg kg^{-1} responded with an 8.6 – 25.2% and 10.9 – 33.2%, respectively, increase in soybean lateral root quantities. At 60 mg kg^{-1} the nodule count per plant was significantly higher than 30 mg kg^{-1} and no application of S (Zhao et al., 2008). A similar response accounted for a 60% increase in nodule activity when 45 kg S ha^{-1} was applied to kidney beans (*Phaseolus vulgaris* L.) (Janssen and Vitosh, 1973). The accumulation of free N compounds with marginal S availability suggests an inability to synthesize proteins resulting in a repression of nitrogen fixation.

Sulfur in Corn

Sulfur uptake and partitioning in corn (*Zea mays* L.) has recently been reassessed in a field trial using six commercial hybrids (Below et al., 2013). Total S uptake for 12.0 Mg ha^{-1} corn yield averaged 26 kg S ha^{-1} resulting in 15 kg S ha^{-1} removed with harvestable grain. Sulfur was determined to accumulate at 0.7 kg ha^{-1} d^{-1} and by R1 48% S-uptake was complete. Corn hybrids demonstrated more aggressive uptake following V10 and rate of uptake declined slightly at R1. In a previous field trial aerial accumulation of S reached 40 kg S ha^{-1} when corn grain yield was 16.3 Mg ha^{-1} (Karlen et al., 1988). At physiological maturity plants were separated by lower leaves, upper leaves, stalk and tassel, and ear and shank to demonstrate that S was partitioned within the respective plant parts at 11, 12, 13, and 64%. Minimal translocation from upper plant parts occurred during early grain fill and only 2 kg S ha^{-1} was provided from the lower leaves. The data suggested that if S sources are not adequate to meet aggressive S demands during the reproductive corn growth stages a topdress application should be considered to limit deficiency.

In the Atlantic Coastal Plains of the U.S., field research demonstrated that when residual soil S was equal to 1.5 mg kg^{-1} at planting the application of 22 and 44 kg S ha^{-1} imcreased corn yields 1 to 28% (Reneau, 1983). A critical N:S ratio of 16:1 suggested that the ratio may be a better measurement of S sufficiency due to variability in S concentration at different growth stages. Ammonium sulfate applied at 0, 34, 67, and 101 kg S ha^{-1} improved yield at only three of twelve locations (Kline et al., 1989). Differences were most prominent between 0 and 67 kg S ha^{-1} with yield increasing by 0.9 – 1.4 Mg ha^{-1}. The lack of yield response was attributed to high availability of S from other sources (e.g. rainfall, irrigation, and subsoil S accumulation). When S response has been investigated in corn production where plant stresses are present increasing corn yield is difficult to demonstrate (Chen et al., 2008). A four year trial was performed with two S rates (0 and 33 kg S ha^{-1}) applied in conjunction with seven N rates (0, 67, 100, 133, 167, 200, and 233 kg N ha^{-1}). Soil samples revealed that water soluble S was 3.8 mg kg^{-1} indicating that the site was likely to demonstrate a response based off the critical water-extractable soil S concentration of 5.6 mg kg^{-1} (Fox et al., 1964). Severe drought resulted in a average 2.6 Mg ha^{-1} grain yield with no S and a significant increase to 3.0 Mg ha^{-1} grain yield with S application. In 2003 effects from no S application demonstrated corn yields of 10.7 Mg ha^{-1} with a significant increase to 11.6 Mg ha $^{-1}$ with S application. Separating differences between S rates among all N rates demonstrated significant yield increases from S application occurring in 3 of 4 years at the 100 kg N ha^{-1} (2003) and 133 kg N ha^{-1}(2004 and 2005). There seemed to be few effects from S at low N rates of 0 to 100 kg N ha^{-1} and none at N rates higher than 200 kg N ha^{-1}.

Organic Sulfur

Organic forms of soil S are found in plant residues and humus. Within organic matter there exist one of two sources of S; carbon-bonded sulfur and organic sulfates (Freney, 1961). Sulfur is also commonly bound in organic matter as S esters and is converted to sulfate by sulfatase enzymes by the following reaction:

$$R\text{-}O\text{-}SO_3^- + H2O \xrightarrow{\textit{sulfatase}} R\text{-}OH + HSO_4^-$$

The measure of S in soil organic matter is closely correlated to N in an N:S ratio of about 8:1, varying from 5 to 13 across North America (Tabatabai, 1984). The strong relationship of N and S in plants may help explain why an 8:1 ratio remains constant for many cultivated regions. In a silty clay loam, continuous corn was grown for 11 years. At the end of each year plant residue was removed from each plot followed by treatment application rates of 2, 4, 8, and 16 Mg corn stover ha^{-1} yr^{-1} (stover nutrient analysis: 0.77, 0.17, and 0.09% N, S, and P respectively) with a standard 202 kg N ha^{-1} applied plus an additional 11.2 kg N ha^{-1} for each Mg of stover applied. After the 11 year trial the organic S in the soil had increased by 9% from the addition of 2 Mg residue ha^{-1} yr^{-1}, and 40% when 16 Mg residue ha^{-1} yr^{-1} was applied (Larson et al., 1972). The return of S from corn stover has led some scientists to hypothesize that agricultural systems may cycle S adequately to meet plant requirements.

Organic S can be added to the soil through the applications of animal manure. Sulfur concentration will vary depending on the manure form applied. Pig slurry ranges between 300 – 660 mg S kg $slurry^{-1}$ but solid manures may range between 0.69 – 1.4 kg S Mg^{-1} (Eriksen et al., 1995, Steineck et al., 1999). Discrepancies of S concentration in animal manures will vary based on animal diet. The determining factor in the availability of organic S sources is typically the carbon to sulfur ratio (C:S). A net mineralization or release of organic S results at a C:S ratio less than 200, whereas a net immobilization (conversion of inorganic nutrients to organic complexed nutrients) occurs at a C:S ratio greater than 400 (Freney, 1967). The change in plant available S that occurs between mineralization and immobilization ratios is either so small that it is negligible or there is no net change. The annual mineralization rate of organic S is estimated to be near 2% of total organic S as this is the annual mineralization rate of soil organic matter (McGrath and Zhao, 1995). However, S mineralization each year can be estimated, for example, a soil with 1% organic carbon in the top 20 cm of the soil profile with use of the following equation (McGrath and Zhao, 1995):

$$\frac{1 kg\ org\ C}{100\, kg\ soil} \times \frac{10^4 m^2}{ha} \times 0.2m\ depth \times bulk\ density \times \frac{1 kg\ org\ S}{100\, kg\ org\ C} \times \frac{2\, kg\ S\ \mathrm{min}}{100\, kg\ org\ S} = \frac{kg\, S\ \mathrm{min}}{ha}$$

This equation can be adjusted to calculate S mineralization based on more specific properties of any given soil.

Elemental Sulfur

Synthetic fertilizers and soil amendments can also provide available S to plants. While many S sources come with additional nutrients elemental S (ES) is the exception. Elemental S is a widely accepted S source due to high analysis (99% S) and slow release properties (Germida and Janzen, 1993). However, for ES to become plant available it must undergo oxidation:

$$S^0 \rightarrow S_2O_3^{-2} \rightarrow S_4O_6^{-2} \rightarrow SO_4^{-2}$$

Elemental sulfur availability may be impacted by temperature, physical and chemical soil properties, amendment particle size, and management practices (Germida and Janzen, 1993).

Research has demonstrated temperature may affect the oxidation of ES. When applied to a sandy loam soil and incubated at 30°C, after 30 days ES had four times as much S oxidized than when incubated at 23°C (Li and Caldwell, 1966). Elemental sulfur has demonstrated oxidation 2 to 6 times faster at 30°C than at 5°C and was 2 to 4 times faster when temperatures were increased from 15°C to 30°C (Nor and Tabatabai, 1977). However, temperature effects on the rate of S oxidation vary by year and throughout the growing season making availability predictions difficult.

Soil properties including soil texture, soil micro-biota, soil pH, organic matter content, and soil moisture can lead to differences in oxidation. Some experimentation has reported no specific relationship between ES oxidation and soil texture (Rehm and Caldwell, 1969; McCaskill and Blair, 1987). However, other studies have found that S oxidation and sand content are directly related while S oxidation and clay content were inversely related (Janzen and Bettany, 1987).

To a limited extent S oxidation occurs by abiotic means, but is dominantly performed by soil micro-biota. The different microorganisms involved in S oxidation can be divided into three groups 1) chemolithotrophs, 2) photoautotrophs, and 3) heterotrophs (Germida and Janzen, 1993). As the overall heterotrophic microbial biomass increases the rate of S oxidation will also increase (Lawrence and Germida 1988). The rate of increase was observed among a range of soil clay content (8-75%), soil pH (5.5-7.8), and available organic C (9-35 mg kg^{-1}).

Particle size of ES can have an effect on S oxidation. Sulfur oxidation can be accelerated by increasing the surface area. Sulfur oxidation is a surficial process and soil microbes are limited to access only those atoms on the exterior of fertilizer particles. Spreading ES powder has demonstrated to be as effective as gypsum application in alfalfa trials in Nebraska (Fox et al., 1964). However, fine powders of ES are a potential explosive hazard when exposed to abrasion (Chien et al., 2011). Elemental S amendments with different particle sizes may have the potential to affect the efficiency and rate of S oxidation. If a higher surface area to mass ratio is ideal, then the manipulation of ES to produce thin plates may be preferable over spheres (Watkinson, 1989; Germida and Janzen, 1993).

Sulfate Fertilizers

Other S-containing sources such as ammonium sulfate (AS), ammonium thiosulfate (ATS), single super phosphate (SSP), and gypsum differ in their ability to provide S as each contains 24, 26, 12, and 18% S, respectively. Sulfur supplied through SSP is phosphate rock that has been acidulated with sulfuric acid (H_2SO_4) to form gypsum. This process further illustrates that there are only two main fertilizers sources that supply S in the plant available form SO_4, ammonium sulfate and calcium sulfate (gypsum). A negative aspect to using SO_4 based fertilizers includes leaching ability when excessive amounts of water are applied from precipitation or irrigation (Konopka et al., 1986).

Current conventional agriculture practices tend to use more high analysis fertilizers that contain high percentages of N, P_2O_5, and K_2O without other additives. One example would be the use of MAP in place of SSP. Single super phosphate supplies 11-12% S but only provides 16 to 20% P_2O_5, whereas MAP will provide 52% P_2O_5. Another example is the comparison between urea and AS. Ammonium sulfate is 21% N and 24% S while urea is 46% N with no significant S content. Historically low S recommendations have led growers to solely use high analysis fertilizers, resulting in minimal additions of S and greater opportunity for deficiencies.

A yield response to S has primarily been demonstrated in coarse textured low organic matter on soils that have no historic S application (Fox et al., 1964b, O'Leary and Rehm, 1990). Sulfur research has demonstrated that an early season S application, when S mineralization is limited, can encourage early season plant development in corn and influence yield (Rehm, 2005). Two S sources, ammonium sulfate (21-0-0-24) and ammonium thiosulfate (12-0-0-26), were placed either in a band 2.5 cm to the side and 5.0 cm below the seed at planting and with the seed. The two S sources were applied at 0, 6.7, 13.4, and 20.1 kg S ha^{-1}. Treatments that received thiosulfate demonstrated a reduction in emergence as the S rate increased when S was applied with the seed and in coarse textured soils. Early season S uptake also seemed to be diminished with thiosulfate when compared to ammonium sulfate. The increase in S uptake by using ammonium sulfate instead of thiosulfate ranged from 10 to 40%. No definite conclusion was given why greater S uptake occurred with ammonium sulfate, but oxidation of thiosulfate may have delayed sulfate availability. The data demonstrate that an early season S application may improve yield beyond no S application, however, caution should be taken when applying S with the seed in coarse textured soils especially when using thiosulfate.

Losses of Available Sulfur

Research has demonstrated that there may be differences in leaching ability among S soil amendments. In a pot study gypsum and three S containing N fertilizers (Ureas, Urea+ES powder, and AS) were tested in clay and loamy sand soil textures to determine S movement from the root zone (Dijksterhuis and Oenema, 1990). Ureas, which is a trademarked product (80% urea and 20% AS), demonstrated the greatest amount of leached S followed by AS and gypsum. The increased amount of S leached from pots treated with Ureas was believed to have occurred to due to the temporary pH increase caused by the hydrolysis of urea. Temporary pH increases may limit S adsorption and result in more S movement in solution.

Factors such as soil texture and soil moisture may also effect S lost through leaching. Coarse-textured soils with high sand content are more prone to S leaching due to high water pore velocity and limited exchange sites for S adsorption (Dick et al., 2008). The ability for water to move rapidly through soil pores allows solutes to be carried from the root zone to lower soil horizons where S is not easily recovered. Other conditions such as water logging of soil could potentially reduce sulfate and lead to volatilization of S (Banwart and Bremner, 1975). However, sulfur lost from the soil in gas form is typically less than 1 kg S ha^{-1} yr^{-1} for non-wetland soils (Noggle et al., 1986).

Applying Sulfur

When a corn field trial in the coastal plains of Virginia was performed, a positive yield response to S application occurred (Reneau, 1983). Sulfur was surface applied as calcium sulfate and incorporated in moderately-well to well-drained soils. Sulfur was applied at 0, 22, 44, and 66 kg S ha^{-1} with a standard rate of 224 kg N ha^{-1} pre-plant and incorporated with a rotary tiller. Grain yield results demonstrated that maximum yield was obtainable with 22 kg S ha^{-1}. Conversely, four S rates (0, 34, 67, and 101 kg S ha^{-1}) applied in a Delaware corn field trial in three increments did not demonstrate a significant yield response (Kline et al. 1989). However, no response may have been due to annual applications of poultry manure for 5 years previous to the study being performed.

A field study, investigating four N rates (56, 112, 168, and 224 kg N ha^{-1}) and four S rates (0, 22, 44, and 66 kg S ha^{-1}), was performed to determine N and S interaction effects on corn yield. Nitrogen was applied as 28 kg N ha^{-1} as ammonium nitrate at planting and the remainder at 50 cm corn while S was applied pre-plant broadcast as calcium sulfate (Rabuffetti and Kamprath, 1977). As the S rate increased from 0 to 66 kg S ha^{-1} when applying 56 kg N ha^{-1} yield decreased. When higher S rates (44 and 66 kg S ha^{-1}) were applied in conjunction with higher N rates (168 and 224 kg N ha^{-1}) significant yield increases occurred. Optimum yields for this experiment were obtained at 44 kg S ha^{-1} and 168 kg N ha^{-1}.

The effects of 0, 6.7, 13.4, and 20.1 kg S ha^{-1}on S uptake and yield were monitored using ammonium thiosulfate and ammonium sulfate placed near the seed in a 2.5 x 5 cm band or with the seed (Rehm, 2005). The study was conducted to determine the effect of rate, source, and placement of S fertilizers on corn yield. In the preceding autumn N, P, and K were added in a dry band application of 22, 9.9, and 37.2 kg ha^{-1}, respectively, 10 cm below the seed row. Additional N was side-dressed as urea to obtain optimal yields, total N applied ranged from 150 to 180 kg N ha^{-1} among the six sites. Ammonium thiosulfate caused yield reductions at two locations when higher rates (13.4 and 20.1 kg S ha^{-1}) were placed with the seed. The two locations that demonstrated reduced yield when ammonium thiosulfate was applied with the seed were characterized by a loamy fine sand texture, whereas other sites with loam, sandy loam, and silt loam didn't show significant reductions in corn yield. Both sources applied at 6.7 kg S ha^{-1} demonstrated an average yield increase of 11% from the check at five of the six locations. Sulfur source only demonstrated an effect on yield when placed with the seed in which case ammonium thiosulfate reduced yields in coarse textured soil, loamy sand. Additional S application research in soils with organic matter content of 20 g kg^{-1} or less have revealed that a rate of 13 kg S ha^{-1} in a 5 x 5 cm band or broadcast applying 28 kg S ha^{-1} may provide a sufficient quantity of S for corn (Rehm and Clapp 2008).

Residual Sulfur

Sulfur application to crops may be carried over into subsequent years providing some S to the second year rotation crop. In a pot study different soils were used to determine the effects of soil colloids, pH, and presence of phosphorus on sulfate adsorption in Cecil, White Store, and Nipe soils (Kamprath et al., 1956). In 1:1 type clay minerals, such as kaolinite, an increase in sulfate adsorption was demonstrated and was attributed to a larger quantity of free iron and aluminum oxides. Hydrated aluminum oxides have especially demonstrated a high capacity for adsorbing sulfate (Ensminger, 1954). Increased sulfate adsorption was demonstrated at pH less than 6 and was the highest at pH 4. The presence of phosphate also has a significant effect on sulfate adsorption. As the sulfate concentration remained constant at 5.4 meq $100g^{-1}$ and phosphate concentration was increased from 0 to 3.6 meq $100g^{-1}$ all adsorbed sulfate was displaced by phosphate. The displacement of S by P suggests that soils generally have a greater affinity for P than S. Thus high pH and high P containing soils may limit S retention whereas high concentrations of aluminum and iron oxides may increase S adsorption.

Soil Testing

As many as 20 different chemical extractants have been studied and tested to determine soil S including water, calcium or potassium phosphate, potassium chloride, and sodium bicarbonate (Dick et al., 2008). Soil S has most consistently been analyzed by extraction with calcium dihydrogen phosphate, $Ca(H_2PO_4)_2$ (Hoeft et al., 1973). The steady ability P has at displacing adsorbed SO_4 supports why calcium dihydrogen phosphate has been commonly used as an extractant (Ensminger and Freney, 1966). Many soil tests used today will extract sulfate with water in order to determine primarily plant available S.

Testing for Sulfur in Plants

The critical S level in corn plant tissue is between 1.2 - 2.0 g kg^{-1} (Jones, 1986). Tissue samples were collected from both the leaf opposite and below the ear at early silking and analyzed for total S and N:S ratio (Reneau, 1983). Values obtained from these samples demonstrated total tissue S was not as useful of a predictor as the N:S ratio for yield. When actual corn yield increases from each treatment were compared to tissue N:S a linear correlation was observed with R^2-value of 0.82.

Currently the only method for measuring plant S concentration is tissue sampling. However, because S use in plants is related to N, tools to assess N availability may be useful in assessing S sufficiency (Smith et al., 1997). Methods, such as plant tissue sampling, chlorophyll meters (measures light transmittance through plant leaves), and active sensing (use of sensor receptors to measure reflectance of near-infrared light), are all useful tools for predicting N status of plants. To better detect S deficiency recent research has demonstrated monitoring S in corn by using a Minolta SPAD-502 chlorophyll meter (Pagani and Echeverria, 2011). In order to prescribe general S recommendations by using chlorophyll meter readings (CMR) the data had to be normalized to reduce variability that is incurred by factors other than S availability. The normalization of readings can be performed by the establishment of reference areas within the field or area of interest eliminating variation in data that can be caused by environmental conditions, plant hybrids, or different growth stages (Blackmer and Schepers, 1995). By compensating for deficiencies with reference strips the CMR may appropriately demonstrate if S deficiencies exist. As much as 15% variance exists between CMR among plants making it necessary to collect samples from several plants to obtain more accurate measurements (Peterson et al., 1993).

Active sensing can be effective at determining N levels in plants and improve S management (Bausch and Diker, 2001). Active sensors function by using near-infrared light (NIR, ~770 nm). Near Infrared light has high reflectance and transmittance whereas visible light has low reflectance and transmittance (Gausman, 1974; Chappelle et al., 1992; Slaton et al., 2001). A comparison was performed between plant tissue sampling, chlorophyll meter reading, and active sensing (Bausch and Duke, 1996). The objective was to examine canopy reflectance on irrigated soils to determine N status, while simultaneously comparing the results from the three methods of canopy N status measurements (plant tissue sampling, chlorophyll meter reading, and active sensing). A 1:1 relationship between the reflectance index of remote sensing and the N sufficiency index was reported for CMR between V11 and R4 corn. An appropriate timing for active sensing use has been investigated to determine a growth stage when NDVI can be measured and yield predicted (Teal et al., 2006). Measuring NDVI at the eighth vegetative stage (V8) revealed a strong relationship with yield (R^2=0.77). All NDVI measurements collected before or after V8 showed weak exponential relationships.

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CHAPTER 2

EFFECTS OF SULFUR ON CORN YIELD AT DIFFERENT NITROGEN RATES

Abstract

Earlier planting dates in cooler soils, increased nutrient removal through greater yielding cultivars, and reductions in atmospheric sulfur (S) deposition suggest that S application may require reassessment for optimal corn (*Zea mays* L.) growth. In 2012 and 2013 a field study was initiated on a loam soil to evaluate corn response to S application by investigating nutrient uptake, grain yield, and agronomic efficiency (AE) at six rates of nitrogen (N) fertilizer. The study was arranged as a split-plot randomized complete block with four replications. Main plots consisted of three S rates (0, 23, and 45 kg S ha^{-1}) while sub-plots consisted of six N rates (0, 56, 112, 169, 225, and 281 kg N ha^{-1}). Corn tissue S concentrations were sufficient in one of two years, but did not accurately predict a yield response from S treatments. These data suggest that tissue tests may not be an adequate measurement for determining when a yield response to S application may occur. Significant yield differences were demonstrated between the 0, 23, and 45 kg S rates without N fertilizer in 2012 (6.7, 7.7, and 9.5 Mg ha^{-1}) and in 2013 (3.2, 5.7, and 3.5 Mg ha^{-1}), respectively. In 2013, a significant yield increase occurred from S application at 56 kg N ha^{-1}, but at N rates greater than 56 kg ha^{-1} S did no significantly impact yield. Averaged across N fertilizer rates there was no significant response to S application. These data suggest that soils with an organic matter content $\geq$ 28 g kg^{-1} and residual S $\geq$ 6-8 mg kg^{-1} are sufficient for optimal corn growth when optimal N rates ($>$ 56 kg N ha^{-1}) are applied.

Introduction

During the last 15 years S application has been found to increase corn grain yields throughout the upper Midwest Corn Belt (Chen et al., 2008; Kim et al., 2013). Data from southern Michigan has shown that S-deposition has been reduced from 30 kg SO_4 ha^{-1} in 1980 to less than 10 kg SO_4 ha^{-1} in 2010 (wet wt.) (National Atmospheric Deposition Program, 2012). Increased observations of S deficiency have been partially attributed to the increased use of high analysis fertilizers containing little or no S (Hagstrom, 1986; Chien et al., 2011). Corn yield has steadily increased over the last 30 years with the mean national corn yield increasing from 4.5 Mg ha^{-1} in 1970 to 9.2 Mg ha^{-1} in 2011 (Capehart, 2013). Increased corn yield at greater rates of N application creates a larger sink for plant nutrients and results in a greater demand for plant available S to achieve optimal corn yield (O'Leary and Rehm, 1990; Ciampitti and Vyn, 2013).

Current S removal in corn grain ranges from 1.0 – 1.2 g S kg grain^{-1} (Chen et al., 2008; Bender et al., 2013). Research has found that in high yielding corn environments ($\geq$ 12.0 Mg ha^{-1}) total vegetative S uptake at physiological maturity may range from 26 – 40 kg ha^{-1} within yields of 12.0 – 19.3 Mg ha^{-1} (Karlen et al., 1988; Bender et al., 2013). Positive yield responses to 33 kg S ha^{-1} have been observed in corn production with greater increases in yield occurring from S application at less than 133 kg N ha^{-1} (Chen et al., 2008). Sulfur fertilization (33 kg S ha^{-1}) with 100 kg N ha^{-1} as compared to 200 kg N ha^{-1} has resulted in similar grain yields when soil S concentrations were less than or equal to 8 mg kg^{-1} (Fox et al., 1964; Chen et al., 2008). More recent research has found that 28 kg S ha^{-1} applied with 157 kg N ha^{-1} increased grain yield 0.9 Mg ha^{-1} at one of four locations when S was pre-plant incorporated while mean yield increases of 1.3 Mg ha^{-1} were found at three of four locations with S application in a 5 × 5 cm band at planting as compared to treatments without S (Kim et al., 2013). However, literature during the last 20 years has reported yield responses with or without S application with little attention to multiple S rates or S response as influenced by N application.

To determine optimum N and S management programs in corn, research has investigated the use of N:S ratios, tissue sufficiency ranges at various growth stages, and partitioning of N and S in vegetative tissue and grain.   Nitrogen and S plant ratios have been considered optimum between 16 -25:1 with sufficient plant tissue S concentrations ranging from 2.1 – 4.0 g kg^{-1} at V6 and R1 (Reneau, 1983; Kline et al., 1989; Oenema and Postma, 2003; Chen et al., 2008). More recently, non-destructive methods such as SPAD chlorophyll meters and greenness indices have been used as S diagnostic tools to determine S sufficiency but detecting plant nutrient status with these instruments may be limited by environmental conditions (Rodriguez et al., 2005; Schlemmer et al., 2005). Although plant tissue levels may be critical to monitoring S availability early in the season before canopy closure prevents mid-season fertilizer applications, the use of SPAD chlorophyll meters has been suggested as an alternative tool for quicker assessment of S sufficiency in plant tissue (Pagani et al., 2009; Pagani and Echeverria, 2011). A range of S rates and higher degrees of soil S deficiency (soil organic matter (OM) $<$ 49 g kg^{-1}) have been suggested for further investigation in corn S management (Pagani et al., 2009).

Soil testing has previously been the standard indicator for recommending S applications (Hoeft et al., 1973). However, S recommendations based on SO_4 soil testing have not shown a good relationship with grain yield in Minnesota (Kim et al., 2013). Although soil testing may be the best method for determining soil S availability at the time of planting it does not predict critical soil S concentrations or grain yield response to applied S as growing conditions are not consistent from year to year (Pagani and Echeverria, 2011; Kim et al., 2013). The objective of this field study was to determine the influence of S and N rate on nutrient uptake, plant greenness grain yield, and agronomic efficiency (AE) of N fertilizer.

Materials and Methods

Field studies were conducted in 2012 and 2013 at the Michigan State University research agronomy farm in East Lansing, MI on a Capac loam (fine-loamy, mixed, active, mesic Aquic Glossudalfs). Soil characteristics over both study years included 7.0-7.3 pH, 45-51 mg kg^{-1} P (Bray-1 P), 124-127 mg kg^{-1} K, 1513-1669 mg kg^{-1} calcium (Ca), 6-8 mg kg^{-1} S (extracted with $Ca(H_2PO_4)_2$), 11-12 cmol$_c$ kg^{-1} cation exchange capacity (CEC), and 27-28 g kg^{-1} soil organic matter. Fields were previously cropped to corn in both study years and autumn chisel plowed after harvest followed by spring tillage using a soil finisher at a 10 cm depth.

Individual six row plots measured 4.5 m in width by 12.1 m in length. The experimental design was a split-plot randomized complete block with four replications. Individual replications contained 18 experimental units. Sulfur fertilizer rate was the main plot factor and was pre-plant incorporated to a depth of 10 cm using calcium sulfate at rates of 0, 23, and 45 kg S ha^{-1}. Main plots were split vertically to evaluate six N fertilizer rates (0, 56, 112, 169, 225, and 281 kg N ha^{-1}). Forty percent of the total N rate was pre-plant incorporated using urea (46-0-0) on the day of planting. Starter fertilizer was applied at planting in a 5 x 5 cm band with 23 kg P_2O_5 and 56 kg K_2O ha^{-1}. Sixty percent of the total N rate was sidedressed at V4 corn (31 May 2012 and 6 June 2013) using urea coated with a urease inhibitor [$CO(NH_2)_2$ + n-(n-butyl) thiophosphoric triamide] banded on the soil surface 10- to 15-cm to the side of the corn row (Koch Agronomic Services, LLC, Wichita, KS). The corn hybrid used over both study years was Dekalb DKC48-12 RIB (98 d relative maturity) (Monsanto Co., St. Louis, MO). Plots were planted on 11 May 2012 and 7 May 2013 to achieve a final plant population of 79,000 seeds ha^{-1}. Weed control consisted of acetochlor [2-chloro-N-ethoxymethyl-N-(2-ethyl-6-methylphenyl)acetamide], atrazine [2-chloro-4-(ethylamino)-6-(isopropylamino)], and glyphosate [N-(phosphonomethyl)glycine] applied to 5 cm weeds (30 May 2012 and 24 May 2013) followed by a second application of glyphosate [N-(phosphonomethyl)glycine] the first week of July in both study years. Environmental data were recorded throughout the growing season and obtained from the Michigan Automated Weather Network (http://www.agweather.geo.msu.edu/mawn/, Michigan State University, East Lansing, MI).

Chlorophyll meter readings were collected at V6 and R1 using a Minolta SPAD-502 chlorophyll meter (Spectrum Technologies Inc., Plainfield, IL). Twenty-five plants per plot were sampled and the average recorded. Chlorophyll measurements were collected from the youngest most fully developed leaf with a visible collar at V6 and from the R1 ear leaf half-way between the mid-rib and the leaf margin and midway between the stalk and the leaf tip (Blackmer and Schepers, 1995; Pagani et al., 2009). A relative greenness index (RGI) was calculated as:

$$RGI=GI_{field}/GI_{FT\ NS}$$

The GI_{field} is the index of greenness for treatments that may be deficient in N or S, and the $GI_{FT\ NS}$ is the index of greenness for treatments that have adequate N and S (45 kg S and 281 kg N ha^{-1}) (Pagani et al., 2009). Canopy normalized difference vegetation index (NDVI) was also collected at V6 using a Greenseeker® handheld red-band optical sensor (Trimble Navigation Ltd., Sunnyvale, CA). From row two or five, 60 to 80 NDVI readings were collected per plot and used to develop a relative greenness index for determining treatment effects on plant growth and color (Teal et al., 2006). Plant height measurements were collected from 20 plants per plot at V6 (height of leaf curve apex in youngest most fully developed leaf with an exposed collar) and R1 growth stages.

Plant tissue samples were collected for nutrient analysis at V6 (10 whole plants plot^{-1}), R1 (25 ear leaves plot^{-1}), and R6 (6 whole plants plot^{-1}). Tissue samples collected at V6 and R1 were dried at 60° C, ground to pass through a 1-mm mesh screen and analyzed for N, P, K, and S. Whole plant sampling at R6 had cobs and grain removed but husks returned to the sample. Vegetative fresh weights were collected, samples were pulverized with a gas powered flail grinder, homogenous sub-samples were collected, and sub-sample fresh weights were recorded. Whole plant sub-samples were analyzed for N using a micro-Kjeldahl digestion method and colorimetric analysis through a Lachat rapid flow injector autoanalyzer (Nelson and Sommers, 1973; Bremner 1996) (Lachat Instruments, Milwaukee, WI). Fifteen basal stalk samples were collected per plot to evaluate stalk nitrate concentration by removing stalk sections 15 cm above the soil surface and 20 cm in length (Binford et al., 1992; Fox et al., 2001). Stalk samples were dried at 60° C and ground to pass through a 1-mm mesh screen. A 10-g stalk sub-sample was added to 1 *M* KCl and shaken for 30 minutes at 3.33 hertz. Stalk extracts were filtered through Whatman #2 filter paper (12.5 cm diameter) and nitrate concentrations were determined with colorimetric analysis through Lachat rapid flow injector autoanalyzer (Huffman and Barbarick, 1981).

The center two rows of each plot were harvested with a Massey Ferguson 8XP small plot combine to determine grain yield, moisture, and test weight (Kincaid Equipment Manufacturing, Haven, KS). Final grain yields were corrected to 150 g kg^{-1} moisture. Agronomic efficiency (AE) of N was calculated to measure the effect of S rate on grain yield at different N rates. The AE was calculated by subtracting the grain yield of the untreated check from the yield of a treatment and dividing by the N rate of the treatment (Wortmann et al., 2011; Sorenson et al., 2012).

The data were subjected to analysis of variance using PROC MIXED in SAS (SAS Institute, Cary, NC). Normality of the residuals was evaluated by examination of normal probability and stem-and-leaf plots. Homogeneity of the variances was determined with Levene's test ($P \leq 0.05$). The data were determined to be significantly different by year ($P \leq 0.05$) and were analyzed individually. A nonlinear model was fit to yield data using PROC NLIN in SAS to determine differences in yield plateaus caused by S rate. When ANOVA generated a significant F-value ($P \leq 0.05$) means were separated using Fisher's least significant difference test.

Results and Discussion

Environmental Conditions

Mean 2012 growing season (May – Sept.) precipitation was 23 cm less than the 30-yr mean (Table 2.1, Fig. 2.1a). Monthly mean air temperatures (May – Aug.) during 2012 were 2° C greater than 2013 and 2° C above the 30-year mean for this time period (Table 2.1). Warmer air temperatures in combination with 50% less precipitation may have limited corn growth in 2012. Rainfall in 2013 was near normal May – Aug., but May and June precipitation resulted in five rainfall events greater than 2 cm, three of which occurred over a single 3 day period (Fig. 2.1b). Temperatures in 2013 were consistent with the 30-yr mean but were 2° C cooler in Aug. and 1° C warmer in Sept. than normal. Annual precipitation measurements indicate S deposition for southern Michigan is 0.13 kg SO_4 (wet wt.) cm rain^{-1} (National Atmospheric Deposition Program, 2012). When S concentrations and annual precipitation data were used to calculate S deposition, mean S deposition during the 2013 growing season would have been 5.4 kg S ha^{-1} (wet wt.) versus 2.7 kg S ha^{-1} in 2012 due to less precipitation.

Table 2.1. Growing season (May – Sept.) and 30-yr mean precipitation and temperature data for East Lansing, MI, 2012 – 2013.

	May	June	July	August	September	Total
Precipitation			cm			
2012	1.0	2.8	6.4	4.9	5.9	21.0
2013	7.6	15.4	6.8	9.7	2.1	41.6
30-yr ave.	9.1	9.1	7.6	9.1	9.1	44.0
Temperature			°C			
2012	17	21	23	21	16	
2013	16	19	21	19	18	
30-yr ave.	16	19	21	21	17	

Precipitation data was collected from Michigan Automated Weather Network (http://www.agweather.geo.msu.edu/mawn/). 30-yr averages come from the PRISM climate group (http://prism.oregonstate.edu/normals).

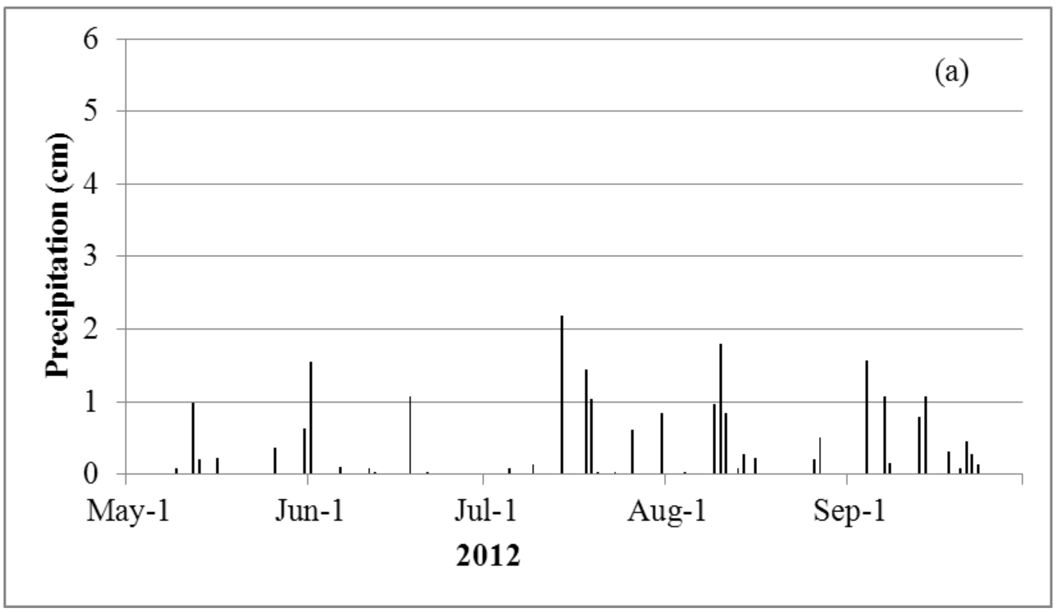
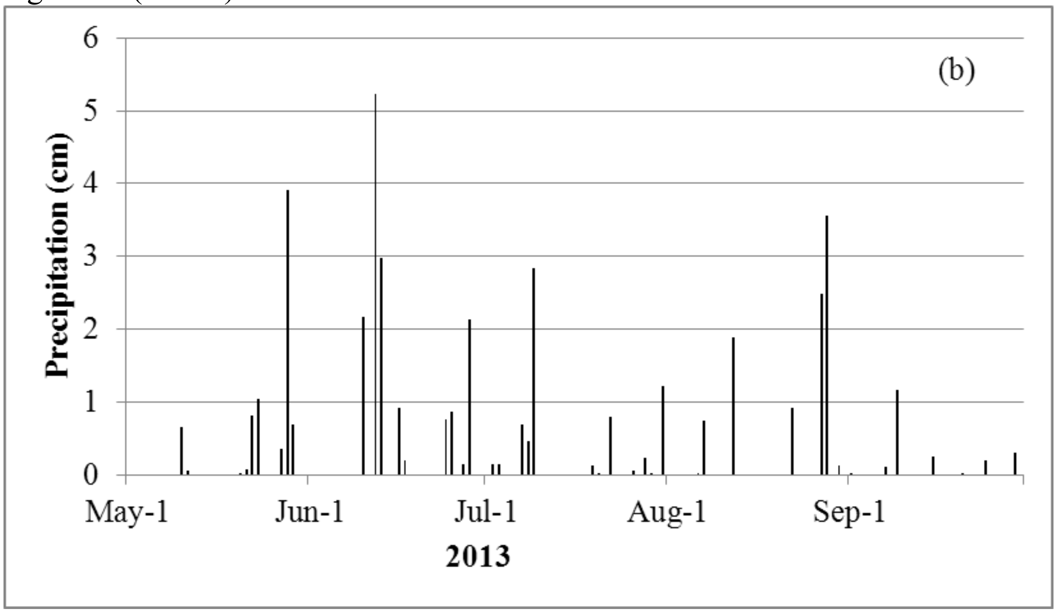


Figure 2.1. Daily precipitation May – Sept. for (a) 2012 and (b) 2013, East Lansing, MI.

Figure 2.1(cont'd)



Nutrient Uptake Responses

Over both study years as N rate increased from 0 to 281 kg ha^{-1} vegetative N increased from 29 to 42 g kg $biomass^{-1}$ at V6 and 20 to 29 g kg $biomass^{-1}$ at R1 ($P \leq 0.01$) (data not shown). Sulfur rate had a direct relationship ($P \leq 0.05$) in 2012 with V6 total N uptake and in both study years with V6 S uptake (Table 2.2). In 2012 and 2013, 45 kg S ha^{-1} increased V6 S uptake by 8 and 45%, respectively ($P \leq 0.01$), but S concentrations were within the sufficiency range (0.21 – 0.40% S) in all treatments. Limited precipitation (< 3.8 cm prior to July 1) during 2012 may have reduced soil S leaching explaining the sufficient S tissue concentrations despite the absence of S fertilization. Data in 2013 suggest S uptake may not have been sufficient prior to V6 as tissue concentrations were low (≤ 2.0 g kg^{-1}) but not necessarily deficient where no S was applied (Table 2.2). May and June 2013 precipitation totals exceeded 20 cm which may have increased residual S leaching beyond developing corn roots despite 3 kg S ha^{-1} (wet weight) deposited from rainfall during the same period. Bender et al. (2013) found that corn required approximately 3 kg S ha^{-1} by V6 suggesting that S availability may be a greater concern in wet years when residual soil S is more readily leached and S-deposition simultaneously continues to decrease. In 2012, 23 and 45 kg S ha^{-1} increased V6 N concentration by 0.7 and 1.6 g kg^{-1}, respectively ($P \leq 0.05$), which concurs with Salvagiotti et al. (2009) who found that wheat N uptake increased by 26 to 28% when 30 kg S ha^{-1} was applied with 52 to 104 kg N ha^{-1}.

Ear leaf R1 N concentrations were not significantly increased ($P \leq 0.05$) by S in either study year (Table 2.2). In one of two study years R1 S concentration increased ($P \leq 0.05$) at the 45 kg S rate but both years resulted in low S concentrations ($\leq$ 1.8 g kg^{-1}) despite S fertilization. Oenema and Postma (2003) suggested that the critical S concentration for corn ear leaf at silking was 1.0 g kg^{-1}, yet S concentrations were considered low between 1.0 and 2.0 g kg^{-1}. Tissue S concentration from V6 to R1 decreased by 30 and 15% without S as compared to 28 and 40% reductions with S application in 2012 and 2013, respectively.

Table 2.2. Corn whole plant (V6) and ear leaf (R1) nutrient analysis as affected by sulfur rate. Nutrient concentrations were averaged across six nitrogen rates, East Lansing, MI, 2012 – 2013.

	V6		**R1**	
	N	**S**	**N**	**S**
		2012		
Sulfur (kg ha^{-1})		g kg^{-1}		
0	39.6 a	2.4 a	24.9 a	1.7 a†
23	40.3 ab	2.4 a	25.7 a	1.8 a
45	41.2 b	2.6 b	26.4 a	1.8 a
Significance (Pr>F)				
S	0.03	<0.01	0.36	0.44
N × S	1.00	0.54	0.49	0.85
		2013		
Sulfur (kg ha^{-1})		g kg^{-1}		
0	36.0 a	2.0 a	25.4 a	1.7 a
23	38.0 a	2.9 b	24.1 a	1.7 a
45	36.0 a	2.9 b	25.2 a	1.8 b
Significance (Pr>F)				
S	0.31	<0.01	0.28	0.04
N × S	0.21	0.17	0.07	0.27

†Means in the same column followed by the same letters for each year are not significantly different at $P \leq 0.05$.

A build-up of tissue S early in corn growth (e.g. prior to V6) followed by moderate declines (≥ 2.1 g kg^{-1} at V6 to 1.7 g kg^{-1} by R1) may increase opportunities for a positive yield response. Reneau (1983) indicated larger corn yield increases occurred when ear leaf S concentrations were below 1.7 g kg^{-1} at early silking. Few other studies have identified an optimum S concentration at R1 but have indicated an optimum S concentration range for corn as wide as 1.5 to 5.0 g kg^{-1} at silking (Pagani and Echeverria, 2011). Thus optimal S management may require S sufficiency concentrations to be maintained more critically during early corn growth (2.1 to 3.0 g kg^{-1} near V6) followed by S decreases to moderate concentrations (1.5 to 2.0 g kg^{-1}) near R1 due to growth dilution.

A 98% increase in 2013 precipitation as compared to 2012 may have caused a 35 to 90% decrease in stalk nitrate concentrations (Table 2.3). In 2012 stalk nitrate concentrations showed a direct relationship with S rate and were within the optimum range (250-2000 mg kg^{-1}), but the relationship was not significant (Binford et al., 1992; Fox et al., 2001). In 2013, stalk nitrate concentrations resulted in an N × S interaction ($P \leq 0.01$) (Table 2.4). Stalk nitrate concentration was 80 and 35% less as the S rate increased to 45 kg ha^{-1} at 225 and 281 kg N ha^{-1}, respectively, compared to similar N rates with no S. The data suggest that at 225 and 281 kg N ha^{-1}, N assimilation may have increased as S application increased. Kim et al. (2013) reported that 28 kg S ha^{-1} pre-plant incorporated increased N uptake and grain N by 6% at three of four and one of four locations, respectively. Despite 98% more precipitation in 2013 increasing vegetative N uptake by 12-156% from 2012, S application did not affect total vegetative N at physiological maturity (Table 2.5). These data suggest N uptake and partitioning may be more affected by soil moisture than actual S availability (Derby et al., 2005).

Table 2.3. Effects of nitrogen and sulfur rate on corn stalk nitrate concentrations at physiological maturity, East Lansing, MI, 2012 – 2013.

	2012	**2013**
Nitrogen (kg ha^{-1})	kg N ha^{-1}	
0	42 c	4 a†
56	240 c	3 a
112	529 c	44 a
169	1462 b	277 a
225	1971 b	849 b
281	2593 a	1690 c
Sulfur (kg ha^{-1})		
0	989 a	601 a
23	1042 a	560 a
45	1387 a	272 b
Significance (Pr>F)		
N	<0.01	0.01
S	0.57	<0.01
N × S	0.30	0.01

†Means followed by the same letters within a column are not significantly different at $P \leq 0.05$.

Table 2.4. Nitrogen by sulfur interaction on corn stalk nitrate concentrations at physiological maturity, East Lansing, MI, 2013.

	2013		
	kg S ha^{-1}		
	0	**23**	**45**
Nitrogen (kg ha^{-1})	mg kg^{-1}		
0	4 aA	3 aA	3 aA†
56	1 aA	1 aA	8 aA
112	18 aA	112 aA	1 aA
169	355 aA	285 abA	191 aA
225	1504 bB	736 bA	308 aA
281	1726 bB	2224 cB	1118 bA
Significance(Pr>F)			
N × S		0.01	

†Means in a column followed by the same lowercase letters are not significantly different at $P \leq 0.05$. Means in a row followed by the same uppercase letters are not significantly different at $P \leq 0.05$.

Table 2.5. Total vegetative nitrogen uptake at physiological maturity as affected by nitrogen and sulfur rate, East Lansing, MI, 2012 – 2013.

	2012	**2013**
Nitrogen (kg ha^{-1})	kg N ha^{-1}	
0	7.2 a	10.6 a†
56	12.6 a	14.1 ab
112	13.1 a	18.2 b
169	12.4 a	25.6 c
225	13.7 a	33.3 d
281	12.7 a	32.5 cd
Sulfur (kg ha^{-1})		
0	11.0 a	22.3 a
23	12.8 a	23.4 a
45	12.1 a	21.5 a
Significance (Pr>F)		
N	0.10	<0.01
S	0.81	0.51
N × S	0.85	0.25

†Means followed by the same letters within a column are not significantly different at $P \leq 0.05$.

Greenness Indices

Sulfur fertilization had no affect ($P \leq 0.05$) on SPAD data in either year (data not shown). Canopy greenness (i.e., SPAD and NDVI measurements) may have been affected by water availability in 2012 and overestimated chlorophyll content as reported by Rodriguez et al. (2005) and Schlemmer et al. (2005). Lack of water has been suggested to interfere with nutrient uptake making SPAD measurements difficult under water deficit conditions (Barraclough and Kyte, 2002; Pagani and Echeverria, 2011). Additionally where S is less mobile in the plant and deficiencies are more prevalent in the newest growth, greater characterization of S status may have been possible in both study years by measuring leaves in the upper canopy than the technique for assessing N status used by Blackmer and Schepers (1995) (Pagani and Echeverria, 2011).

In 2012 and 2013 relative NDVI at V6 was not affected by N or S application. An N × S interaction occurred ($P \leq 0.05$) in 2013 but not in 2012 (Table 2.6). As a plant defense mechanism to reduce plant temperatures under water-limiting conditions, reflectivity of near infrared light increases with water stress (Schlemmer et al., 2005).

Table 2.6. Effects of nitrogen and sulfur rate on V6 relative normalized difference vegetation index, East Lansing, MI, 2012 – 2013.

	2012	2013
Nitrogen (kg ha^{-1})	relative NDVI	
0	0.99 a	0.76 a
56	1.00 a	0.86 b
112	1.00 a	0.95 c
169	1.00 a	0.99 d
225	1.00 a	0.97 cd
281	1.00 a	1.00 d
Sulfur (kg ha^{-1})		
0	0.98 a	0.92 a
23	1.00 a	0.93 a
45	1.00 a	0.91 a
Significance (Pr>F)		
N	0.17	<0.01
S	0.35	0.55
N × S	0.53	<0.01

†Means followed by the same letters within a column are not significantly different at $P \leq 0.05$.

Plant response to drought conditions may explain why relative NDVI was 1 to 23% greater in 2012 as compared to 2013. Reductions in plant growth in 2012 from drought stress may have limited the ability of the Greenseeker® to determine differences in plant greenness caused by N or S. In 2013 45 kg S ha^{-1} with zero and 112 kg N ha^{-1} reduced relative NDVI by 12 and 5%, respectively, compared to the NDVI results obtained with the same N rates at 23 kg S ha^{-1} (Table 2.7).

Table 2.7. Nitrogen by sulfur interaction on V6 relative normalized difference vegetation index, East Lansing, MI, 2013.

	2013		
	kg S ha^{-1}		
	0	**23**	**45**
Nitrogen (kg ha^{-1})	relative NDVI		
0	0.80 aB	0.78 aB	0.69 aA
56	0.80 aA	0.88 bA	0.88 bA
112	1.00 bB	0.95 bcB	0.89 bA
169	1.00 bA	0.95 bcA	1.00 cA
225	0.93 bA	1.00 cA	0.95 bcA
281	0.98 bA	1.00 cA	1.00 cA
Significance(Pr>F)			
N × S		<0.01	

†Means in a column followed by the same lowercase letters are not significantly different at $P \leq 0.05$. Means in a row followed by the same uppercase letters are not significantly different at $P \leq 0.05$.

As S rate increased from 23 to 45 kg ha^{-1} lower relative NDVI values may explain yield reductions of 2.2 and 0.9 Mg ha^{-1} with zero and 112 kg N ha^{-1}, respectively. When 2013 relative NDVI was regressed against grain yield, zero S treatments resulted in an R^2 of 0.60. The R^2-values at 23 and 45 kg S rates increased to 0.78 and 0.79, respectively (data not shown), and may have been the cause of the 23% greater correlation between relative NDVI and grain yield. These data agree with Teal et al. (2006) who found that as much as 76% of grain yield variation was explained by NDVI. Improving the accuracy of determining N sufficiency by maintaining sufficient plant available S may allow growers to better predict yield variability in the field and make more informed decisions for in-season N applications (Raun et al., 2001; Samborski et al., 2009).

Corn Grain Yield

An N × S interaction ($P \leq 0.10$) on grain yield occurred in 2012 and 2013 (Table 2.8). Corn yields in 2013 with 23 and 45 kg S ha^{-1} at the 56 kg N ha^{-1} rate were similar to the 2012 grain yields at the zero N rate indicating that several large precipitation events in May and June of 2013 may have resulted in N losses near 45 – 56 kg N ha^{-1}.

Table 2.8. Interaction between nitrogen and sulfur application on corn grain yield, East Lansing, MI, 2012 – 2013.

	2012			**2013**		
	kg S ha^{-1}			kg S ha^{-1}		
	0	**23**	**45**	**0**	**23**	**45**
Nitrogen (kg ha^{-1})	grain yield (Mg ha^{-1})					
0	6.7 aA	7.7 aAB	9.5 aB	3.2 aA	5.7 aB	3.5 aA†
56	8.2 abA	8.5 abA	8.5 aA	6.8 bA	7.3 aAB	9.2 bB
112	8.8 abA	9.7 abA	10.1 aA	11.9 cA	11.7 bA	10.8 bcA
169	9.2 bA	9.4 abA	8.3 aA	12.9 cA	12.8 bcA	12.9 cdA
225	8.5 bA	10.6 bA	9.0 aA	13.2 cA	14.5 cdA	13.2 dA
281	9.0 bA	10.0 bA	9.6 aA	13.6 cA	15.1 dA	14.2 dA
Significance(Pr>F)						
	N	S	N × S	N	S	N × S
	<0.01	0.56	0.07	<0.01	0.37	0.10

†Means in a column for each year followed by the same lowercase letters are not significantly different at $P \leq 0.10$. Means in a row for each year followed by the same uppercase letters are not significantly different at $P \leq 0.10$.

Sulfur application at the zero and 56 kg ha^{-1} N rates resulted in a positive yield response in both study years and in 2013, respectively. Data suggest that in the dry and wet field conditions during 2012 and 2013, respectively, soil S immobility from lack of mass flow or excessive mobility and potential S leaching may have occurred to cause a positive yield response from S application (Bohn et al., 1986; Chen et al., 2008). However, as N rates increased to 56 and 112 kg N ha^{-1} in 2012 and 2013, respectively, yield differences were moderated and demonstrated no significant affect from S application. At sub-optimal N rates (56 and 112 kg N ha^{-1} in 2012 and 2013, respectively) root growth may have been promoted sufficiently to increase plant access to naturally available S (i.e., residual soil S, mineralized S from soil OM) thus not resulting in a positive yield response from S application. As corn plants senesce older leaves at sub-optimal N rates, accelerated protein hydrolysis and greater S mobilization to the upper portion of the plant canopy may occur concealing S deficiency and further limiting yield response to S application (Loneragan et al., 1976; Marshner, 1995). Yield was not significantly increased from S application at N rates greater than 56 and 112 kg ha^{-1} in either 2012 or 2013, respectively. Greater N rates may increase S demand to satisfy physiological requirements as has been suggested by O'Leary and Rehm (1990) and Ciampitti and Vyn (2013). However, root growth may be sufficiently promoted from larger N rates ($\geq$ 56 kg N ha^{-1}) resulting in greater nutrient exploitation of the soil profile and sufficient S uptake for optimal corn growth and yield (Table 2.9) (Nibau et al., 2008).

Table 2.9. Corn R1 ear leaf nitrogen analysis, East Lansing, MI, 2012 – 2013.

	2012	**2013**
Nitrogen (kg ha^{-1})	g S kg^{-1}	
0	1.5 a	1.4 a†
56	1.6 a	1.5 a
112	1.8 b	1.8 b
169	1.8 b	1.8 b
225	1.8 b	1.9 b
281	1.9 b	1.9 b
Significance (Pr>F)		
N	<0.01	<0.01

†Means followed by the same letters within a column are not significantly different at $P \leq 0.05$.

In 2012, convergence criteria were not met to determine a nonlinear model for the 45 kg S rate and yield plateaus were not significantly different by S rate ($P >$ 0.10) at the zero and 23 kg ha^{-1} S applications (8.9 and 10.5 Mg ha^{-1}, respectively) (data not shown). Yield plateaus were not significantly different ($P >$ 0.10) in 2013 with 13.6, 15.8, and 13.7 Mg ha^{-1} representing yield maximums at zero, 23, and 45 kg S ha^{-1}, respectively. Nitrogen rate was regressed against yield at different S rates demonstrating no significant differences ($P >$ 0.10) among slopes caused by S in 2012 (0.38 and 0.49 at the 0 and 23kg S rates, respectively) and 2013 (2.07, 1.98, and 1.96 at the 0, 23, and 45 kg S rates, respectively).

Agronomic Efficiency of Nitrogen Fertilizer

In both study years agronomic efficiency (AE) decreased by 67 and 51%, respectively, as N rate increased from 56 to 281 kg N ha^{-1} ($P \leq 0.01$), but no significant main effects occurred from S application (Table 2.10). In 2012, grain yield increase per kg of N fertilizer applied ranged from 10.3 to 31.0 as N rate decreased from 281 to 56 kg ha^{-1}. Drought conditions during 2012 may have limited N movement through soil solution decreasing the AE of N fertilizer. In 2013, AE increased 162 to 284% as compared to 2012 suggesting soil moisture may have influenced AE to a larger extent than N rate over either study year (Derby et al., 2005).

Table 2.10. Main effects of nitrogen and sulfur on agronomic efficiency† of nitrogen, East Lansing, MI, 2012 – 2013.

	2012	**2013**
Nitrogen (kg ha^{-1})	— kg grain kg N^{-1} —	
56	31.0 b	81.4 c‡
112	25.7 ab	73.7 c
169	13.7 a	57.5 b
225	12.0 a	46.5 ab
281	10.3 a	39.6 a
Sulfur (kg ha^{-1})		
0	15.6 a	56.1 a
23	21.1 a	59.7 a
45	18.9 a	63.3 a
Significance (Pr > F)		
N	<0.01	<0.01
S	0.78	0.66
N × S	0.99	0.27

†Agronomic efficiency (AE) is calculated by subtracting yield of the control (no nitrogen or sulfur) from the yield of the treatment and dividing by the nitrogen rate of the treatment.
‡Values followed by the same letters within a column are not significantly different at $P \leq 0.05$.

Salvagiotti et al. (2009) indicated that S fertilization increased N use efficiency in wheat and proposed that S application promoted root growth to allow better exploitation of the soil profile and improve N recovery. Similar reports have been presented (Mandal et al., 2003), but S-activated root growth is more likely to occur under extreme S deficiency conditions (soil test ≤ 1 mg kg^{-1} and organic matter ≤ 10 g kg^{-1}) in coarse textured soil and not when S is adequately supplied (Nibau et al., 2008). Data in the current study suggest that increasing S availability will not significantly increase AE (Table 2.10). Sulfur is an essential nutrient in corn and directly affects N assimilation as a constituent of enzymes involved in N metabolism (i.e., nitrite reductase), but nutrient recovery as affected by root growth may be supported to a greater extent by localized N concentrations and not the presence of S fertilizer (Drew, 1975; Reuveny et al., 1980; Robinson, 1994; Salvagiotti et al., 2009).

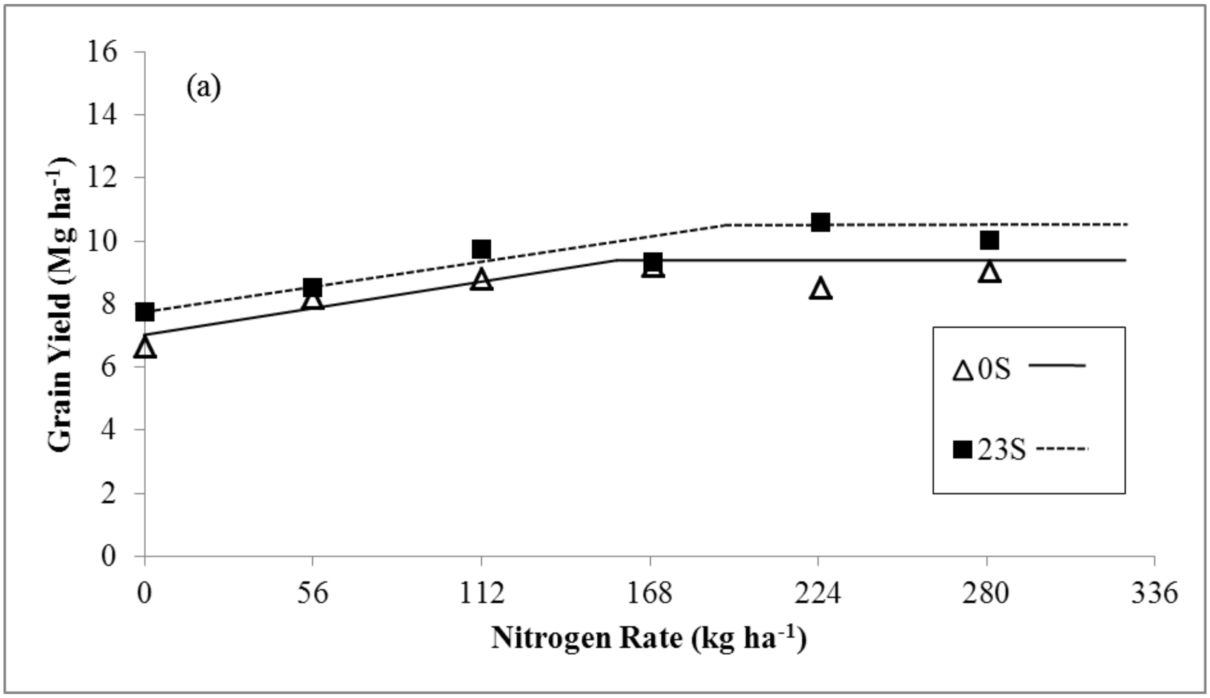
Conclusions

Tissue analyses did not accurately predict a yield response from S treatments, but greater correlation between yield and NDVI occurred with S fertilization. These data suggest greater accuracy for determining N sufficiency may occur by maintaining greater plant available S. However, no significant increase in grain yield resulted from S application despite reports of decreased S deposition and large nutrient requirements for modern corn cultivars. Soils that have traditionally been classified as non-responsive to S fertilization (i.e., organic matter ≥ 28 g kg^{-1} and soil S ≥ 6 – 8 mg kg^{-1}) resulted in a significant yield increase at sub-optimal N rates (≤ 56 kg N ha^{-1}) yet with N rates greater than 56 kg N ha^{-1} the data suggest sufficient root exploitation of the soil profile resulting in adequate S uptake for optimal corn growth and yield.

APPENDIX

Corn grain yield regressed against nitrogen rate in 2012 and 2013 separated by sulfur rate

Yield data from the East Lansing corn study were evaluated at each S rate in 2012 and 2013 to determine yield plateaus and rate of increase. PROC NLIN and PROC GLM statements in SAS were utilized to compare yield responses as affected by S rate. The data did not demonstrate a significant difference in yield plateau or slopes in either study year. In 2012, convergence criteria was not met to determine a nonlinear model for the 45 kg S rate and yield plateaus were not significantly different by S rate ($P >$ 0.10) at the zero and 23 kg ha^{-1} S rates (8.9 and 10.5 Mg ha^{-1}, respectively). Yield plateaus were not significantly different ($P >$ 0.10) in 2013 demonstrating 13.6, 15.8, and 13.7 Mg ha^{-1} as yield maximums at zero, 23, and 45 kg S ha^{-1}. Nitrogen rate was regressed against yield at different S rates demonstrating no significant differences ($P >$ 0.10) among slopes caused by S in 2012 (0.38 and 0.49 at the 0 and 23kg S rates, respectively) and 2013 (2.07, 1.98, and 1.96 at the 0, 23, and 45 kg S rates, respectively).



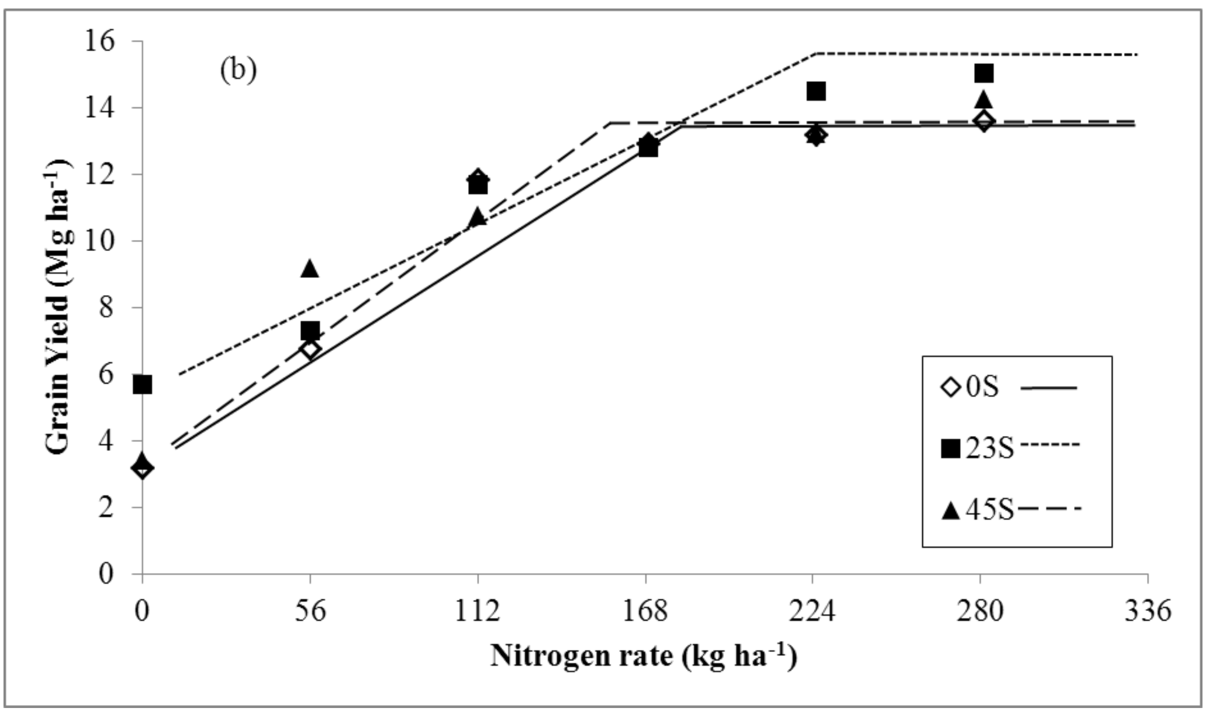


Figure A.1. Corn grain yield regressed against nitrogen rates for (a) 2012 and (b) 2013. Convergence criteria was not met for the 45 kg S rate in 2012 and not presented, East Lansing, MI.

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CHAPTER 3

SOYBEAN RESPONSE TO SULFUR PLACEMENT AND STARTER FERTILIZER

Abstract

Earlier planting dates, minimal rates of early season nutrient mineralization, increased nutrient removal, and reductions in atmospheric sulfur (S) deposition have increased concern regarding early season S availability for optimal soybean [*Glycine max* (L.) Merr.] growth. A field study was established to determine the effects of 25 lbs S/acre and a starter fertilizer of 20 lbs nitrogen (N) and 50 lbs phosphorus (P)/acre on soybean yield and quality. Main plots consisted of S placement (pre-plant incorporated, at planting in a 2 × 2 inch band, split, and no S) while sub-plots consisted of N and P starter fertilizer applied at planting (with or without in a 2 × 2 inch band). Data collection included SPAD chlorophyll measurements (V4, R5), petiole samples (R1), root nodule counts (R3), grain yield, quality, and yield components (pod producing nodes, pods/node, beans/pod, weight per 100 seeds). Starter fertilizer did not affect grain yield, however, root nodule numbers were not significantly reduced and soybean growth prior to V4 appeared to be positively impacted by starter N and P. Sulfur placement did not significantly affect grain yield in either study year. Sulfur application to soybeans may not affect yield when pre-plant soil tests demonstrate greater than 6 ppm S with soil organic matter (OM) near 3.0% or greater.

Introduction

Reports of sulfur (S) applications increasing crop yields have been described throughout the upper Midwest (Chen et al., 2005; Kaiser and Kim, 2013). Decreased atmospheric deposition of S, increased purity and usage of concentrated fertilizers, less incidental S in pesticides, and increased usage of minimum tillage systems that delay soil warming and allow accumulating crop residues to immobilize plant available S have all resulted in a decreased soil supply of S to crops (Hagstrom, 1986; Whitney, 1997; Rehm, 2005; Chien et al., 2011; National Atmospheric Deposition Program, 2012). Past research has indicated that S application is not necessary to achieve optimal soybean yield in Michigan but few recent data are available to substantiate this claim (Robertson et al., 1976).

Sulfur is particularly important in soybean production due to the high protein content of the seed (Sexton et al., 1998). By weight, soybean seed averages 40% protein and 20% oil (Krishnan, 2005). Sufficient available soil S at planting may be critical for optimal soybean growth as 59% of total S uptake is required by the end of soybean vegetative development (Naeve and Shibles, 2005). Traditionally soybeans in Michigan have been planted in mid-late May and early June. However recent research has demonstrated planting in late April to early May offers significant yield increases lending growers to adjust planting dates to optimize grain yield potential (Anderson and Vasilas, 1985; Pedersen and Lauer, 2004). As soybean growers in northerly latitudes attempt to maximize yield through earlier planting dates, S deficiency becomes a greater concern with cooler soil temperatures and where residual soil S levels are less than or equal to 8 ppm (Fox et al., 1964). Soil organic matter (OM) may contribute up to 3 lbs S/acre for every one percent soil OM (McGrath and Zhao, 1995). However, early planting dates may indirectly create a longer time period where soybeans are S-deficient as mineralization may be limited by cool soil temperatures (Roberts, 1985; Schoenau and Malhi, 2008). Earlier planted soybeans may require S fertilization to satisfy vegetative S requirements prior to soil organic S mineralization but yield response data to S application are limited.

Minimal soil microbial activity during early spring conditions may limit the extent of soybean nodulation and N_2 fixation (Lynch and Smith, 1993; Osborn and Riedall, 2006). A starter fertilizer application of N and P may increase early soybean growth and development minimizing yield loss potential in what is often difficult growing conditions. Broadcast application of greater than 100 lbs N/acre at planting or emergence has increased soybean yield 7% (Bly et al., 1998). Concerns are that N application is inconsistent at producing a positive yield response and may reduce N_2 fixation by decreasing nodule number (Salvagiotti et al., 2008). Phosphorus fertilization at planting may help maintain N_2 fixation while promoting earlier root development for increased S uptake (Cassman et al., 1980; Cadisch et al., 1993; Kaiser and Kim, 2013). Additional research is needed to investigate how early season N and P applications in the form of starter fertilizer influence S uptake, yield, and root nodulation. The objectives of this study were to 1) determine the effects of S placement on soybean growth, yield, and grain quality and 2) determine the effects of N and P starter fertilizer on early season nutrient uptake, yield, and root nodule formation.

Materials and Methods

Field studies were established in 2012 and 2013 on a Capac loam (fine-loamy, mixed, active, mesic Aquic Glossudalfs) at the Michigan State University research agronomy farm in East Lansing, Michigan. Field sites were previously cropped to corn (*Zea mays* L.) in both study years and autumn chisel plowed after previous crop harvest followed by spring tillage with a soil finisher to a 4 inch depth. Individual plots measured 15 feet in width by 50 feet in length and were six rows wide with 30 inch row spacing. Soil characteristics over both study years included 6.5-7.3 pH, 31-42 ppm P (Bray-1 P), 132-135 ppm K, 1934-1942 ppm Ca, 6-9 ppm S (extracted with $Ca(H_2PO_4)_2$), cation exchange capacity of 13 meq/100g, and 3.0% organic matter.

The study was designed as a split-plot randomized complete block with four replications. Sulfur placement was the main plot factor and consisted of four application intervals including 1) pre-plant broadcast and incorporated with a soil finisher to 4 inch depth, 2) at planting in a 2 × 2 inch band, 3) split with 15 lbs S/acre at planting in a 2 × 2 inch band and 10 lbs S/acre sidedressed on the soil surface in a band 4 inches to the side of the row at R1 (split), 4) and no sulfur. All S applications totaled 25 lbs S/acre. The S source for pre-plant application was calcium sulfate (0-0-0-16) and while the at-planting and split S applications were an ammonium sulfate based liquid fertilizer (0-0-0-17). Main plots were split vertically to evaluate soybean response to starter fertilizer (20 lbs N and 50 lbs P_2O_5/acre) applied at planting in a 2 × 2 inch band as di-ammonium phosphate (18-46-0). A maintenance application of 120 lbs K_2O/acre was applied pre-plant and incorporated. The soybean variety was Asgrow 2330 (relative maturity 2.3) in both study years (Monsanto Co., St. Louis, MO). Plots were planted on 21 May 2012 and 9 May 2013 to achieve a final plant population of 154,000 seeds/acre.

Plant height was assessed at V4 and R5 from 20 plants per plot (Osborne and Riedell, 2006). Chlorophyll meter readings were collected from 25 plants per plot using a Minolta SPAD-502 chlorophyll meter (Spectrum Technologies Inc., Plainfield, IL). Measurements were collected at V4 and R5 from the youngest most fully developed tri-foliate (Schlemmer et al., 2005). Petiole samples were taken at R1 collecting 20 petioles per plot selecting the youngest most fully developed trifoliate (Buckley and Wolkowski, 2012). Samples were dried at 150 °F, ground to pass through a 1-mm mesh screen, and analyzed for P and S.

Soybean roots were dug at R3 to assess nodulation (Zhao et al., 2008). A shovel was inserted to the soil 6 inch from the base of the soybean plant down to a 6 to 8 inch depth. Ten plants per plot were dug and soybean root balls were soaked and washed over a 2-mm mesh screen. Nodules that were removed during washing and retained in the 2-mm mesh screen were accounted for and averaged with the total number of nodules per plant.

Grain yield was collected from the center two rows of each plot using a small plot combine (Alamco, Nevada, IA) on 2 October 2012 and 1 October 2013. Grain yield was corrected to 13% moisture. Grain samples were used to determine grain oil and protein content with an NIR6500 using WinISI 2 version 1.5 software (Infrasoft international LLC., Eden Prairie, MN) (Osborne and Riedell, 2006). Ten plants per plot were collected at physiological maturity to count bean pod producing nodes, pods per node, beans per pod, and weight per 100 seeds.

Data were subjected to ANOVA using the PROC MIXED statement in SAS (SAS Institute). Normality of the residuals was evaluated by examination of normal probability and stem-and-leaf plots. Homogeneity of the variances was determined with Levene's test ($P \leq 0.10$). When ANOVA generated a significant F-value for a treatment ($P \leq 0.10$) means were compared by Fisher's least significant difference test (LSD). The data were determined to be significantly different by year ($P \leq 0.01$) and were analyzed separately.

Results and Discussion

Environmental Conditions

Warmer air temperatures in combination with 50% less precipitation may have limited plant growth in 2012 as compared to 2013 (Table 3.1, Fig. 3.1a). May through August 2013 rainfall was near or above normal but May and June precipitation resulted in three rainfall events greater than 1 inch, two of which occurred over a single 2 day period (Fig. 3.1b). Temperatures in 2013 were consistent with the 30-yr. mean but were 3°F cooler in Aug. and 2°F warmer than normal in Sept. Annual precipitation measurements indicated S deposition for southern Michigan was 0.57

lbs SO_4 (wet weight) per inch of rainfall (National Atmospheric Deposition Program, 2012). Sulfate concentrations and annual precipitation data were multiplied to calculate S deposition. The 2013 growing season received 9.4 lbs S/acre (wet weight) through atmospheric deposition while the 2012 growing season received 4.7 lbs S/acre due to less precipitation.

Table 3.1. Mean May – Sept. and 30-yr average precipitation and temperature data, East Lansing, MI, 2012 – 2013.

Month	**2012**	**2013**	**30-yr ave.**	**2012**	**2013**	**30-yr ave.**
	precip. (inch)			temperature (°F)		
May	0.39	3.00	3.60	63	60	61
June	1.12	6.06	3.60	69	67	66
July	2.51	2.66	3.00	74	70	70
Aug.	1.94	3.83	3.60	69	67	70
Sept.	2.32	0.82	3.60	61	65	63
Total	8.30	16.40	17.40			

Precipitation and temperature data were collected from Michigan Automated Weather Network (http://www.agweather.geo.msu.edu/mawn/). 30-yr averages come from the PRISM climate group (http://prism.oregonstate.edu/normals).

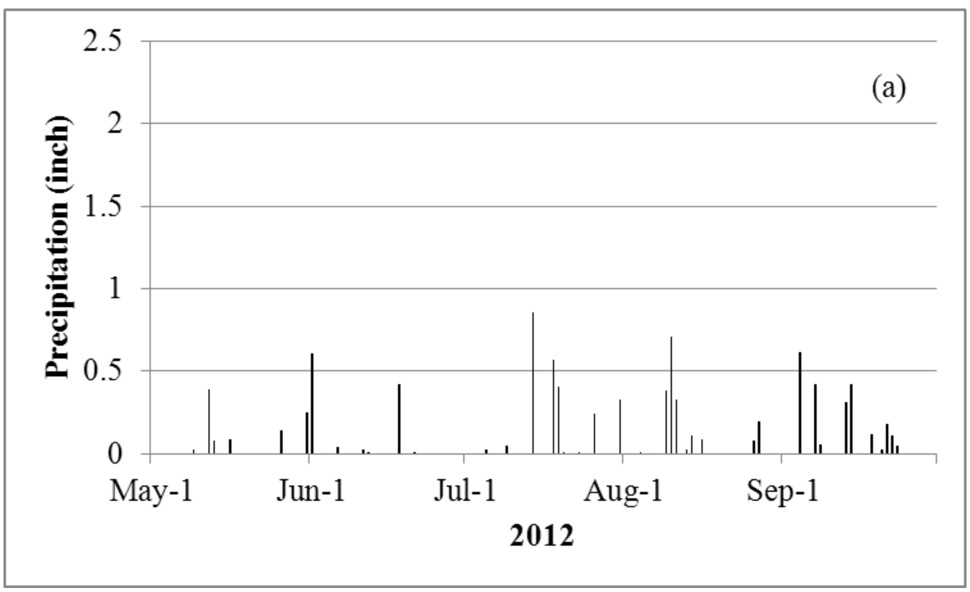
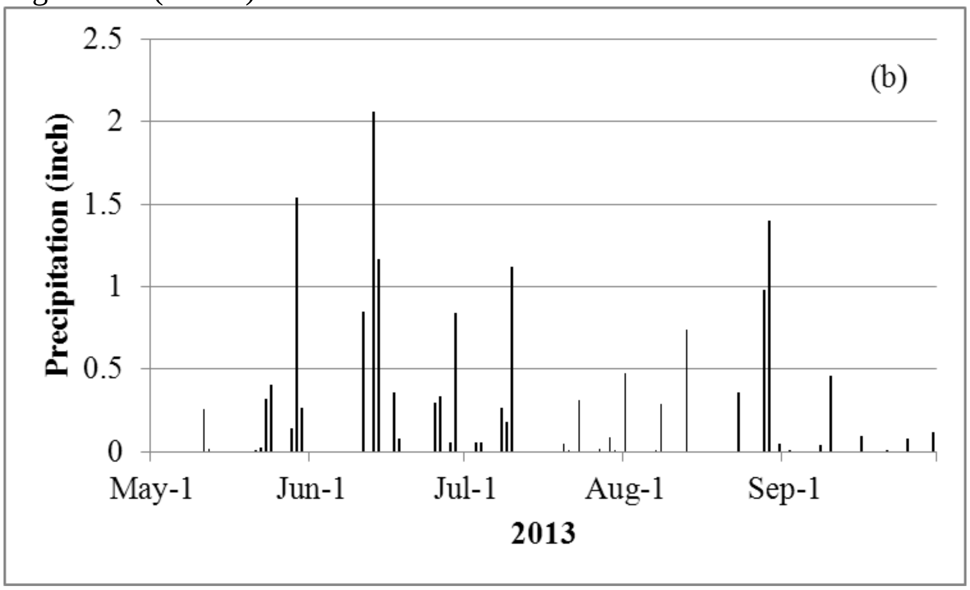


Figure 3.1. Daily precipitation May – Sept. for (a) 2012 and (b) 2013, East Lansing, MI.

Figure 3.1. (cont'd)



Plant Tissue Analyses

In 2012, soybean petiole S concentrations ranged from 0.24% without S to 0.27% with S application (Table 3.2). Developing soybean tissue acquires S (~25%) from the youngest most fully developed trifoliate (first trifoliate), and therefore maintaining first trifoliate S concentrations above 0.25% has been suggested as critical for S assimilation and optimum soybean growth (Sunarpi and Anderson, 1996). However, in 2012 without S application petiole concentrations were 0.24% S and grain yield was not significantly reduced. Oenema and Postma (2003) reported that percent S in the first trifoliate can be as low as 0.21% and remain sufficient for optimum soybean growth. In 2013, no treatment resulted in a petiole concentration less than 0.32% S. Applying S broadcast or at planting increased petiole S contents by an average of 5% across these treatments in 2013 and increased grain yield 3% but the differences were not significant. Data suggest that soybean first trifoliate S concentrations may be sufficient when residual soil S ranges from 6 – 9 ppm with 3.0% soil OM.

Table 3.2. Petiole R1 nutrient concentrations as affected by S placement and starter fertilization, East Lansing, MI, 2012 – 2013.

Fertilizer Treatment	2012	2013
S Placement	% S	
Pre-plant Incorporate	0.26	0.33
At Planting	0.25	0.34
Split	0.27	0.33
No Sulfur	0.24	0.32
Starter Fertilizer		
Starter	0.26	0.33
No Starter	0.25	0.33
LSD (0.10)		
S Placement	ns	ns
Starter	ns	ns
S Placement × Starter	ns	ns

Increased root branching and elongation from localized placement of N has suggested that early season S uptake may increase with starter fertilizer application (Nibau et al., 2008). However without starter fertilizer in either year of the current study, petiole S concentrations remained above 0.25% and increased by no more than 4% with starter fertilizer application. Kaiser and Kim (2013) found that starter N fertilizer did not increase percent S at V5 soybeans but did indicate that S uptake may be greater due to increased biomass from starter N fertilizer application. Early season biomass was not collected in the current study and may be a measurement to further elucidate S uptake patterns as affected by starter N fertilizer. Soybean petiole P concentrations were not deficient averaging 0.32 to 0.46% in 2012 and 2013, respectively, averaged across starter and no starter fertilizer treatments with no significant effects from starter fertilizer ($P >$ 0.10) (data not shown). The lack of a P response from starter fertilizer likely occurred due to pre-plant soil samples demonstrating P levels ranging from adequate to very high (31-42 ppm Bray-1 P) for optimal soybean growth (Lambert et al., 2006).

Root Nodule Assessment

In 2012 and 2013, R3 root nodule numbers were not significantly affected by starter fertilizer ($P > 0.10$) (Table 3.3). Research performed in growth chambers has indicated that moderate N rates (100 lbs/acre) tend to reduce nodule size while larger N rates (300 lbs/acre) seem to decrease the number of nodules (Harper and Cooper, 1971). Research has shown that N fertilizer inhibiting N_2 fixation has a local effect, which suggests N applied in a 2 × 2 inch band near the area of nodulation may decrease nodule number (Streeter, 1985; Takahashi et al., 1992). However the change in nodule number in both study years as affected by starter fertilizer was not significant, suggesting that 20 lbs N/acre may not be a large enough N rate to limit N_2 fixation despite application near the area of nodulation.

Table 3.3. Soybean R3 root nodule counts as affected by S placement and starter fertilization, East Lansing, MI, 2012 – 2013.

Fertilizer Treatment	**2012**	**2013**
S Placement	Nodules per Plant	
Pre-plant Incorporate	23	24
At Planting	23	24
Split	23	22
No Sulfur	23	24
Starter Fertilizer		
Starter	22	24
No Starter	24	23
LSD (0.10)		
S Placement	ns	ns
Starter	ns	ns
S Placement × Starter	ns	ns

Root nodulation at R3 averaged 23 and 24 nodules per plant in 2012 and 2013, respectively, but was not significantly affected by S placement ($P > 0.10$). Scherer and Lange (1996) found in a laboratory study that with an S rate of 36 lbs /acre N accumulation from N_2 fixation in Broad bean (*Vicia Faba* L.) was not limited with the presence of N fertilizer at 250 lbs N/acre. Research by Zhao et al. (2008) further reported that elemental S promoted nodule formation when higher rates of S (60 and 120 lbs S/acre) were basal applied. Although no difference in nodule number resulted from S application in the current study, S may be beneficial when greater rates of N fertilizer are applied (> 20 lbs N/acre) and maintaining a normal rate of N_2 fixation is a concern.

Plant Greenness

Sulfur at planting increased the 2012 relative SPAD chlorophyll measurement by 2% at V4 ($P \leq 0.10$), but S did not affect SPAD measurements during other growth stages or in 2013 as all readings remained above 0.95 (data not shown). Hitsuda et al. (2004) found that chlorophyll meter readings did not accurately predict the S status of wheat or soybean which may help explain the minimal variation of the data from the current study. Starter fertilizer application did not affect soybean greenness in either study year as relative SPAD measurements remained greater than 0.95. However, visual observations made one week prior to V4 in 2012 and 2013 noted that starter fertilizer resulted in greener and denser plant canopies than plots not receiving starter N and P applications. Osborne and Riedell (2006) found a similar response as 20 lbs N/acre applied at planting significantly increased V4 soybean biomass by 10%. Despite the inability of SPAD to document the visual observations in the current study, starter fertilizer appeared to positively impact early-season soybean growth and may have a greater impact on earlier planted soybeans. Future research determining soybean response to starter N and P fertilizer may be improved by collecting vegetative biomass as compared to SPAD chlorophyll measurements.

Grain Yield, Quality, and Components

Grain yield was not significantly affected from S placement or starter fertilizer in 2012 or 2013 (*P* > 0.10) (Table 3.4). Pre-plant soil samples demonstrated residual S concentrations ranging from 6 – 9 ppm while mineralization of the 3% soil organic matter may have contributed up to 9 lbs S/acre as has been suggested by McGrath and Zhao (1995) and Schoenu and Malhi (2008). Soil data from the current study suggest that 21 – 27 lbs. S/acre were available for soybean uptake during the growing season. Previous research has indicated that soybean requires 6.5 lbs S/acre to produce 15 bu (FAO Sulphur Network, 1992; Lantmann and Castro, 2004). Data from the current study suggest that mean yields of 42 and 64 bu/acre in 2012 and 2013, respectively, required 18 – 28 lbs S/acre. Pre-plant soil tests indicate that sufficient S was available at the time of planting and may explain why no significant response to S application was observed in either study year.

Table 3.4. Soybean grain yield and moisture as affected by S placement and starter fertilization, East Lansing, MI, 2012 – 2013.

Fertilizer Treatment	2012	2013	2012	2013
S Placement	bu/acre		%	
Pre-Plant Incorporate	45	66	10.9	13.4
At Planting	36	64	10.8	13.3
Split	42	61	10.8	13.4
No Sulfur	42	63	11.0	13.4
Starter Fertilizer				
Starter	42	63	10.9	13.3
No Starter	41	64	10.8	13.4
LSD (0.10)				
S Placement	ns	ns	ns	ns
Starter	ns	ns	ns	ns
S Placement × Starter	ns	ns	ns	ns

Main effects from starter fertilizer were not significant in 2012 or 2013 (*P* > 0.10) (Table 3.4). A positive yield response in soybeans from starter N fertilizer has been demonstrated in about one-half of the published studies yet the magnitude of the response does not seem to vary by different rates of N application (i.e., 0-45, 45-90, or > 90 lbs N/acre) (Salvagiotti et al., 2008). Osborne and Riedell (2006) found that soybean yield did not respond to starter N fertilizer when drought conditions occurred, but yield increased 5% in two of three years with 15 lbs N/acre compared to the untreated check. Timing of N fertilizer may have a more critical role in increasing yield as soybeans receiving N (20 – 290 lbs N/acre) have increased yield 8 – 12% when N application occurred during the reproductive stages, most particularly after R3 (Wesley et al., 1998; Ray et al., 2005). Salvagiotti et al. (2008) further indicated that N supply from N_2 fixation is insufficient in high yielding environments (> 70 bu/acre) due to greater N requirements from increased sink size (i.e., greater biomass and pod set in higher yielding soybeans). Data in the current study did not demonstrate a significant yield increase but more consistent yield increases from N fertilizer may occur as different placements of N fertilizer are utilized (i.e., deep banded ($\geq$ 8 inch) or top dressed during reproductive stages) (Gan et al., 2002).

Neither S placement nor starter fertilizer affected the number of pod producing nodes (12-13 and 15-17 in 2012 and 2013, respectively), pods per node (2 pods/node, 2012-2013), beans per pod (3 beans/pod, 2012-2013), or 100 seed weight (0.56-0.60 oz./100 seeds, 2012-2013) ($P >$ 0.10; data not shown). No differences in seed oil occurred from starter fertilizer or S placement in either study year, but percent protein was significantly increased by S broadcasted or in a 2 × 2 inch band at planting ($P \leq$ 0.10) in 2013 (Table 3.5). Applying the total 25 lbs S/acre before or at planting increased protein 1% and may indicate that sufficient S availability at planting is critical for optimal soybean grain protein production. However Wilson (2004) reported that soybean grain protein ranges from 34 to 57% with an average of 42%. These data suggest that despite an increase in grain protein from S application in the current study the results are below average and may not be significant considering the wide range of soybean grain protein content. Oil and protein were 10 and 12% greater, respectively, from 2012 to 2013 suggesting the 98% increase in precipitation may have been a greater controlling factor for soybean seed quality than the fertilizer treatments. Results from the current study support previous research indicating that grain N (protein) and oil concentration were decreased in drought conditions and may not be significantly affected by N or S fertilization (Sexton et al., 1998; Osborne and Riedell, 2006).

Table 3.5. Soybean grain oil and protein as affected by S placement and starter fertilization at East Lansing, MI in 2012 and 2013.

	Oil		**Protein**	
Fertilizer Treatment	**2012**	**2013**	**2012**	**2013**
S Placement		%		
Pre-plant Incorporate	20	22	36	41
At Planting	20	22	36	41
Split	20	22	36	40
No Sulfur	20	22	36	40
Starter Fertilizer				
Starter	20	22	36	40
No Starter	20	22	36	40
LSD (0.10)				
S Placement	ns	ns	ns	0.44
Starter	ns	ns	ns	ns
S Placement × Starter	ns	ns	ns	ns

Conclusions

Grain yield did not respond to starter N and P fertilizer in 2012 or 2013. Although visually starter fertilizer appeared to improve plant greenness and canopy density in both study years, soybean SPAD chlorophyll measurements were no less than 95% of maximum with or without N and P fertilizer. Nitrogen fertilizer may increase early soybean growth when used at 20 lbs

N/acre and be a small enough rate to not significantly reduce root nodule formation. Soybean grain yield did not significantly respond to S application in either study year compared to the untreated check demonstrating that residual soil S and mineralized S from soil organic matter were sufficient for optimal plant growth. Alternative options for maintaining sufficient soil S may include fertilization in alternate rotation years as positive yield responses from S have been more consistent in corn (Rehm, 2005; Kim et al., 2013). Depending upon S application rate, sufficient residual soil S may be available in the year following corn application for optimal soybean growth and development. Prior to S application, consideration of all potentially available S sources (i.e., soil OM, residual soil S, and S-deposition) may be critical in determining whether S application is warranted.

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CHAPTER 4

CORN RESPONSE TO SULFUR IN HIGH PHOSPHORUS SOILS

Abstract

Sulfur (S) fertilization has traditionally been recommended for Michigan corn (*Zea mays* L.) production on coarse textured low organic matter soils, but S recommendations may vary by crop rotation, sub-soil characteristics, and soil phosphorus (P) concentrations. A two year field study was initiated on a loamy sand to determine corn yield response to S fertilization by investigating nutrient uptake, nitrogen (N):S ratios, and grain yield as affected by N fertilizer rate. Main plots consisted of three S rates (0, 20, and 40 lbs S/acre) while sub-plots consisted of six N rates (0, 50, 100, 150, 200, and 250 lbs N/acre). Tissue analyses indicated S availability was optimal to excessive resulting in R1 N:S ratios ranging from 13 – 10:1 as S rate increased from 0 to 40 lbs/acre, respectively. Reduced N:S ratios from increased S rates were associated significant 22 and 10% yield losses at the zero and 250 lb N rates, respectively, in one of two years. Sulfur availability at 12 – 24 inches beneath the soil surface suggested potential for S accumulation (i.e., pH < 6 and presence of metal oxides) and may need to be considered when the upper soil profile (0-8 inches) suggests S may be limiting. Inorganic S accumulation within corn plants was suggested by low N:S ratios which may have caused assimilation competition between sulfate and nitrate (i.e., sulfite and nitrite reductase) which may explain the negative yield response from S application in one of two study years.

Introduction

Positive corn yield responses to S fertilization have become more frequent in the upper Midwest Corn Belt. Yield increases from 14 – 42 bu/acre with applications of 25 – 30 lbs S/acre have been recorded suggesting that S deficiencies may be more prevalent than 10 – 15 years ago (Chen et al., 2008; Kim et al., 2013). Past research in Michigan has suggested that S application on corn was not necessary due to sufficiently available S from soil organic matter (OM), S contributions through atmospheric deposition, or a combination of the two (Robertson et al., 1976). However, the U.S. Environmental Protection Agency (EPA) has reported that between 1980 and 2010 S emissions decreased by 83% reducing annual sulfate deposition in Michigan from 25 lbs S/acre to less than 8 lbs S/acre (wet weight), respectively (National Trends Sulfur Dioxide Levels, 2011). Improved higher-yielding corn germplasm that produces 200 – 250 bu/acre and removes 16 – 20 lbs S/acre with grain harvest in conjunction with decreased S deposition may create greater opportunities for S deficiency when soil S is not replenished (Bender et al., 2013).

A large percentage of Michigan's corn acreage is grown on soils that cannot provide or retain sufficient S in the root zone due to a coarse soil texture (i.e., loamy sand and sandy loam), low percent clay (<25%), soil OM less than 2.0%, and pH greater than 6 (Warncke et al., 2009). Sulfate adsorption is negligible at soil pH's greater than 6 and is also susceptible to leaching (Bettany and Steward, 1983; Curtin and Syers, 1990). A portion of Michigan corn production is in rotation with potatoes (*Solanum tuberosum* L.) which require high soil phosphorus (P) levels (75-150 ppm) and may further limit S adsorption due to greater adsorption ability of phosphate to soil clay particles and iron (Fe) and aluminum (Al) oxides (Schoenau and Malhi, 2008). In addition, S applications are made in the year of potato production and are exposed to irrigation which may increase soil S mobility and lead to greater S leaching and accumulation where sub-soils indicate greater S adsorption opportunities (i.e., pH > 6 and presence of metal oxides or clay particles) (Harward et al., 1962; Fox et al., 1971; Bohn et al., 1986). Determining an optimal S rate in soils susceptible to S leaching may be critical to maintaining sufficient plant available S in the root zone. The objective of this study was to determine the corn response to S fertilization on a coarse textured, low organic matter soil with high soil P.

Materials and Methods

Field experiments were conducted in 2012 and 2013 at the Michigan State University Montcalm research center on a Montcalm (coarse-loamy, mixed, semi-active, frigid Alfic Haplorthods) and McBride (coarse-loamy, mixed, semi-active, frigid Alfic Fragiorthods) loamy sand. Soil analysis over both study years included 6.1 – 6.2 pH, 151 – 160 ppm P (Bray-1 P), 161 – 194 ppm potassium (K), 567 – 599 ppm calcium (Ca) , 8 – 9 ppm sulfate (SO_4, extracted with $Ca(H_2PO_4)_2$), 7 meq/100g cation exchange capacity (CEC), and 1.5 – 1.6% soil OM. Field sites were previously cropped to corn in both study years and autumn chisel plowed, followed by spring tillage using a soil finisher (4 inch depth). Individual plots measured 15 feet in width by 40 feet in length (six rows) with 30 inch row spacing.

The experimental design was a split-plot randomized complete block with four replications. Sulfur rate was the main plot factor and was pre-plant incorporated as calcium sulfate at 0, 20, and 40 lbs S/acre. Main plots were split vertically and evaluated with six N rates (0, 50, 100, 150, 200, and 250 lbs N/acre), totaling 18 treatments per replication. Forty percent of the N rate was pre-plant incorporated as urea (46-0-0) the day of planting. Starter fertilizer was applied at planting in a 2 × 2 inch band at 20 lbs P_2O_5 and 50 lbs K_2O/acre. The remaining sixty percent of the N rate was sidedressed at V6 using urea with a urease inhibitor [$CO(NH_2)_2$ + n-(n-butyl) thiophosphoric triamide] (Koch Agronomic Services, LLC, Wichita, KS). Sidedress applications were made on the soil surface by banding the N fertilizer 4 – 6 inches to the side of the corn row. The corn hybrid used for both study years was Dekalb DKC48-12RIB (98 day relative maturity) (Monsanto Co., St. Louis, MO). Plots were planted on 27 Apr. 2012 and 6 May 2013 to achieve a final plant population of 32,000 plants/acre. Pest control followed standard management programs used in the region. Mean annual rainfall for this location was 32.4 inches with S deposited annually at a rate of approximately 0.27 lbs SO_4 per inch of rainfall resulting in 8 – 10 lbs. (wet weight) of sulfate deposition per acre (National Atmospheric Deposition Program, 2012). In the event that precipitation was limited during the growing season additional irrigation was added through a center-pivot (Table 4.1).

Table 4.1. Mean May – Sept. precipitation and 30-yr mean with irrigation, Entrican, MI, 2012 – 2013.

	Precipitation			**Irrigation**	
	2012	**2013**	**30-yr ave.**	**2012**	**2013**
	inch			inch	
May	0.8	4.5	3.2	0	0
June	2.1	2.3	3.4	2.0	0
July	5.8	1.4	2.9	2.8	3.5
Aug.	5.2	4.1	3.3	1.6	2.6
Sept.	0.3	1.3	3.2	0	0
Total	14.2	13.6	16.0	6.4	6.1

Precipitation data were collected from Michigan Automated Weather Network (http://www.agweather.geo.msu.edu/mawn/).
http://www.weather.com/weather/wxclimatology/monthly/graph/48858

Chlorophyll meter readings were collected at V6 and R1 using a Minolta SPAD-502 chlorophyll meter (Spectrum Technologies Inc., Plainfield, IL). Twenty-five plants per plot were sampled and the average recorded. Chlorophyll measurements were collected from the youngest most fully developed leaf with a visible collar at V6 and from the ear leaf during R1 half way between the mid-rib and the leaf margin and midway between the stalk and the tip of the leaf (Blackmer and Schepers, 1995; Pagani et al., 2009). A relative greenness index (RGI) was calculated as:

$$RGI=GI_{field}/GI_{FT\ NS}$$

The relative greenness index GI_{field} is the index of greenness for treatments that may be deficient in N or S, and the $GI_{FT\ NS}$ is the index of greenness for the treatment that had adequate N and S (45 kg S and 281 kg N ha^{-1}) (Pagani et al., 2009). Canopy normalized difference vegetation index (NDVI) was also collected using a Greenseeker® handheld red-band optical sensor at V6 (Trimble Navigation Ltd., Sunnyvale, CA) (Teal et al., 2006). From row two or five, 60 to 80 NDVI readings were collected per plot and a relative greenness index was developed to determine treatment effects on overall plant growth and color. Plant height measurements were collected from 20 plants per plot at V6 and R1.

Plant tissue samples were collected for nutrient analysis at V6 (10 whole plants/plot), R1 (25 ear leaves/plot), and R6 (6 whole plants/plot). Tissue samples collected at V6 and R1 were dried at 150 °F, ground to pass through a 1-mm mesh screen, and analyzed for N, P, K, and S. Whole plant sampling at R6 had cobs and grain removed but husks returned to the sample. Vegetative tissue fresh weights were collected, samples were pulverized with a gas powered flail grinder, homogeneous sub-samples were collected, and fresh weights were recorded. Whole plant sub-samples were analyzed for N using a micro-Kjeldahl digestion method and colorimetric analysis through a Lachat rapid flow injector auto-analyzer (Lachat Intruments, Milwaukee, WI) (Nelson and Sommers, 1973; Bremner 1996). Fifteen basal stalk samples were collected at R6 to evaluate stalk nitrate concentrations. Stalk sections were cut 6 inches above the soil surface and were 8 inches in length (Binford et al., 1990; Fox et al., 2001). Stalk samples were dried at 150° F and ground to pass through a 0.04 inch mesh screen. A 0.35 oz. stalk sub-sample was added to 1.7 fl. oz. of 1 *M* KCl and shaken for 30 minutes at 200 rpm. Stalk extract was filtered through Whatman #2 filter paper (5 inch diameter), and nitrate concentrations were determined with colorimetric analysis through Lachat rapid flow injector autoanalyzer (Lachat Instruments, Milwaukee, WI) (Huffman and Barbarick, 1981). The center two rows of each plot were harvested on 9 October 2012 and 29 October 2013 with a Massey Ferguson 8XP small plot combine to determine grain yield, moisture, and test weight. Final grain yield was corrected to 15% moisture.

Data were subjected to ANOVA using the PROC MIXED statement in SAS (SAS Institute, Cary, NC). Normality of the residuals was evaluated by examination of normal probability and stem-and-leaf plots. Homogeneity of the variances was evaluated with Levene's test ($P \leq 0.10$). When ANOVA generated a significant F-value for a treatment ($P \leq 0.10$) the means were compared by Fisher's least significant difference test (LSD). Due to a greater understanding of corn response to N application, the emphasis of this paper will be on the main effects of S and N × S interactions.

Results and Discussion

Corn Growth Response and Nutrient Uptake

Plant height and total biomass at V6 and R1 corn were not affected by S application and did not result in a significant N × S interaction ($P > 0.10$) (data not shown). Sulfur application rate did not influence N concentration at V6 or R1 tissue sampling (Table 4.2). Sulfur uptake was directly related ($P \leq 0.10$) with S rate at V6 in 2013 and at R1 in both study years as greater S rates resulted in increased S uptake. Increased S uptake occurring from S fertilization may be considered luxury consumption as tissue concentrations were within the sufficiency range (0.21 to 0.40%) without S application (Oenema and Postma, 2003). Data suggest that V6 corn plants had adequate S available from residual soil S early in the season to meet requirements for optimal growth. Bender et al. (2013) found that nutrient uptake in corn requires approximately 2.7 lbs S/acre prior to V6.

Table 4.2. Corn V6 whole plant and R1 ear leaf N and S concentrations average across N rates as affected by S rate, Entrican, MI, 2012 – 2013.

	V6		**R1**	
	N	**S**	**N**	**S**
		2012		
S rate (lbs/acre)		%		
0	3.61	0.25	3.59	0.22
20	3.54	0.25	3.48	0.30
40	3.66	0.26	3.59	0.35
LSD (0.10)	ns	ns	ns	<0.01
		2013		
S rate (lbs/acre)		%		
0	2.88	0.21	3.18	0.25
20	2.88	0.23	3.21	0.28
40	2.87	0.25	3.00	0.30
LSD (0.10)	ns	0.02	ns	0.03

Pre-plant soil tests in the current study indicated 16 lbs residual S/acre were available at planting both study years indicating there was sufficient S to satisfy early corn growth requirements. Reneau (1983) reported that larger yield increases may be expected when S fertilization is managed to obtain ear leaf S concentrations below 0.17 % at R1, but no clear reason was given as to why this was the case. However in the current study no S treatment resulted in R1 concentrations below 0.21% in 2012 or 2013. Excessive S uptake may have occurred from fertilization but even without S fertilizer ear leaf concentrations were greater than 0.21% suggesting that other sources may have contributed to S availability and prevented growth dilution of S in R1 corn.

Stalk nitrate concentrations ranged from 0 to 4887 ppm in 2012 and 0 to 2605 ppm in 2013 (Table 4.3). Past literature has shown that optimal stalk nitrate concentrations range between 250 to 2000 ppm (Binford et al., 1990; Fox et al., 2001). In both study years a significant N × S interaction ($P \leq 0.10$) occurred affecting stalk nitrate concentrations. Mean stalk nitrates across N and S treatments were 72% lower in 2013 as compared to 2012 resulting in sub-optimal concentrations for many of the treatments. Lower stalk nitrate concentrations may explain the 4 – 11% yield reduction observed in 2013 as compared to 2012. However, 20 lbs S/acre may have reduced stalk nitrate accumulation at the supra-optimal 250 lb N rate as nitrate concentrations were reduced by 2945 and 1505 ppm in 2012 and 2013, respectively. Kim et al. (2013) reported that S increased N uptake at one of four locations and increased grain N accumulation at two of four locations, suggesting that in the current study S may have improved N assimilation at 250 lbs N/acre resulting in lower stalk nitrate accumulation.

Table 4.3. Stalk nitrate concentration as influenced by N and S applications, Entrican, MI, 2012 – 2013.

	2012			2013		
	lbs S/acre			lbs S/acre		
	0	20	40	0	20	40
N rate (lbs/acre)	ppm					
0	0	0	0	9	0	0
50	1	0	0	1	0	0
100	16	4	95	2	62	3
150	1609	1661	966	114	247	230
200	3228	3198	4410	621	939	371
250	4887	1942	3520	2605	1100	928
LSD (0.10)		1107			592	

Grain Yield Response to Sulfur

Averaged grain yield was 145 – 245 and 139 – 229 bu/acre in 2012 and 2013, respectively. In 2012, S was not necessary to achieve optimal corn yields as neither the 20 lbs. nor the 40 lbs. S rates resulted in significant yield differences ($P >$ 0.10) from the zero S rate (Table 4.4).

Table 4.4. Grain yield and moisture as affected by S rate, Entrican, MI, 2012 – 2013.

	2012		2013	
	Yield	Moisture	Yield	Moisture
S rate (lbs/acre)	bu/acre	%	bu/acre	%
0	213	18.0	202	22.1
20	208	17.9	194	22.2
40	218	18.1	196	22.1
LSD (0.10)	6.5	ns	ns	ns

In 2013 no significant main effects from S occurred, but an N × S interaction was present ($P \leq$ 0.10). Without N application, grain yield decreased 29 and 46 bu/acre at 20 and 40 lbs S/acre, respectively, indicating a negative yield effect from S application (Table 4.5). Additionally, at 250 lbs N/acre there was a significant 12 and 8% yield loss with the application of 20 and 40 lbs S/acre, respectively.

Table 4.5. Corn grain yield response to N × S interaction, Entrican, MI, 2013.

	2013		
	lbs S/acre		
	0	**20**	**40**
N rate (lbs/acre)	grain yield (bu/acre)		
0	163	136	118
50	182	169	173
100	201	199	203
150	213	221	229
200	221	234	231
250	239	211	220
LSD (0.10)		18	

These data suggest that S may have been provided by other sources (i.e., residual soil S or OM mineralization) and was sufficient for optimal corn growth at low and high N rates. Nitrogen and S do not compete for nutrient absorption into corn plants, however, reduced yield from high application rates of N or S has been suggested as being caused by competition or interference between nitrite and sulfite reductase in N and S assimilation pathways, respectively. Both nitrite and sulfite reductase require the same electron donor, ferrodoxin, for reduction processes and both enzymes' substrates (NO_2 and SO_3) are chemically similar (Hoefgen and Hesse, 2008). Rabuffetti and Kamprath (1977) found that at 50 lbs N/acre S additions negatively affected corn grain yield, whereas at 150 or 200 lbs N/acre S fertilizer increased yield. An optimum N:S ratio (16:1) may be necessary for a positive yield response to occur from S application as suggested by Reneau (1983). Similar to the current study results, Weil and Mughogho (2000) indicated yield reductions when N:S ratios were too low ($\leq$ 8:1) due to excessive S availability. Calculated N:S ratios from R1 ear leaf concentrations in the current study demonstrated that when averaged across N rates 40 lbs S/acre resulted in N:S ratios that were 15 and 23% lower in 2012 and 2013, respectively, than the zero S rate (Table 4.6).

Table 4.6. Main effects of S on R1 N:S ratios, Entrican, MI, 2012 – 2013.

	2012	**2013**
S rate (lbs/acre)	R1 N:S	
0	13	13
20	12	12
40	11	10
LSD (0.10)	0.5	1.0

Sulfur application decreased N:S ratios below 13:1 in both study years and may have been due to inorganic S accumulation within the plant as was suggested by Reneau (1983), which was directly related with the negative yield responses in 2013. Sulfur fertilization may have increased mid-season (R1) S concentrations to excessive levels but other S sources (i.e., precipitation, soil OM, etc.) may have increased S availability beyond S provided from treatments (Kline et al., 1989). Lower N:S ratios than those calculated in the current study may have resulted following R1 considering only 48% of S uptake has occurred by this growth stage as compared to N where 65% of uptake is complete (Bender et al., 2013).

Factors Affecting Sulfur Response

Compared to the zero S treatment, decreased N:S ratios at R1 suggest that other sources may have contributed to plant available S apart from fertilizer treatments. Based on precipitation data in 2012 and 2013, S deposition was calculated to be approximately 4.2 and 4.0 lbs SO_4/acre (wet weight), respectively, during the growing season (Table 4.1) (National Atmospheric Deposition Program, 2012). Residual soil sulfate levels were 8 and 9 ppm providing approximately 16 and

18 lbs S/acre at planting in 2012 and 2013, respectively. Pre-plant soil tests also demonstrated OM content of 1.6 and 1.5% in 2012 and 2013, respectively, which may have contributed approximately 3 lbs S/acre annually for every one percent of soil OM (Roberts, 1985; McGrath and Zhao, 1995; Schoenau and Malhi, 2008). When examined in total, residual soil S, mineralized S from soil OM, and S deposition may have provided up to 24 and 25 lbs S/acre during the 2012 and 2013 growing seasons, respectively. Soil contributions in addition to atmospheric S deposition may have provided 92 to 96% of the suggested corn S requirement for 190 bu/acre corn yield (Bender et al., 2013). Soil and environmental data suggest that S application increased S uptake and may have caused N and S competition between assimilation pathways (i.e., nitrite and sulfite reductase) which may explain why yield was significantly reduced when S fertilizer was applied at the zero and 250 lbs. N rates in 2013.

Additionally, pre-plant soil sampling demonstrated high soil P levels (> 150 ppm) in both study years due to the high P requirement of other crops in rotation (i.e., potatoes). Phosphate has greater adsorption potential than S in the soil and at the current P levels the anion exchange sites (AEC; positively charged clay surface or minerals where negatively charged compounds may bind and release) may have been saturated in the upper soil profile (0-8 inch) (Rajan and Fox, 1975). Additionally growers apply 50 – 100 lbs S/acre in the year of potato production, which may have resulted in S leaching in previous years especially with high rainfall and irrigation at the current study field site (Warncke et al., 2009; Bohn et al., 1986). The soil profile in the current study had two characteristics that may have increased sub-soil S availability to developing corn roots. At the 9 to 15 inch soil depth, an accumulation of iron (Fe) and aluminum (Al) oxides has been described which may have offered considerable potential for sulfate adsorption at soil pH's less than 6 (Harward et al., 1962; Fox et al., 1971; OSD, 2013).

Sub-soil horizons indicated acid pH ranging from 5.1 to 6.0, which may have caused S accumulation (Gebhardt and Colemnan, 1974; Schoenau and Malhi, 2008). Low N:S ratios at R1 may have been due to corn root access to sub-soil S mid-season and luxury consumption post V6. These hypotheses are supported by a previous report indicating that increased percent clay, amorphous Fe and Al, and AEC at depths of 16 to 24 inches may limit the potential for positive corn yield responses from S fertilization (Kline et al., 1989).

Conclusions

The field site selected for this study demonstrated what has traditionally been considered to be optimum S response conditions in Michigan: coarse textured soil (loamy sand), low organic matter (< 2.0%), and no manure application in the previous 5 years. However, S fertilization did not seem to be necessary to obtain optimal yield in 2012 and reduced yield by an average 36 and 23 bu/acre at the zero and 250 lb N rates, respectively, in 2013. High soil P levels (> 150 ppm) in the upper 8 inches of the soil profile may have limited S adsorption and led to leaching in previous years. Sub-soil characteristics that promote S adsorption (e.g., pH < 6 and presence of Fe and Al oxides) suggest that S may have accumulated 9 to 15 inches beneath the soil surface possibly providing sufficient S for optimal corn growth as roots developed during the growing season. These assumptions were supported by R1 ear leaf concentrations that demonstrated 15 and 23% reductions in N:S ratios as S rate increased from zero to 40 lbs S/acre in 2012 and 2013, respectively, which may indicate inorganic S accumulation within corn plants and competition between N and S assimilation pathways (i.e., nitrite and sulfite reductase). These data serve as a reminder that careful assessment of all available S sources should be considered in corn fertilizer programs in lieu of recommendations based solely on soil OM and residual soil S. Planting corn in coarse textured soils (e.g., sandy loam and loamy sand) with large quantities of soil P (> 150 ppm) may suggest a high potential for S leaching. Soil sampling 1 – 2 feet beneath the soil surface to determine if sub-soil S accumulation has occurred may be necessary to adequately determine all S contributions that may be available to a developing corn plant.

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