Nitrogen and Phosphorus Fertilization of Soybean (*Glycine max* [L.] Merr.) in the Red River Valley Region of Manitoba, Canada.

By

Joseph Paul James Gervais

A Thesis Submitted to the Faculty of Graduate Studies of
The University of Manitoba
in Partial Fulfillment of the Requirements for the Degree of

MASTER OF SCIENCE

Department of Plant Science
University of Manitoba
Winnipeg

THE UNIVERSITY OF MANITOBA

FACULTY OF GRADUATE STUDIES

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Of

MASTER OF SCIENCE

ACKNOWLEDGEMENTS

My Committee:
Dr. Jane Froese
Dr. Rigas Karamanos
Dr. Tee Boon Goh
Mr. Bruce Brolley

Dr. Gary Crow for his help with the statistical analysis and his never-ending patience

Dr. Christian Willenborg for his invaluable help and encouragement

Dr. Martin Entz and Keith Bamford for the use of their seeder

Technicians and Summer Students:
Dave Audette, Anastasie Hacault, Pat Kenyon, Melissa Kiehn, Derek Lewis, Rebekah Nason, Don Poisson, Meagan Simpson, Shaan Tsai, and Stacey Waldron

Cooperators:
Byron Edel, Murray Froebe and Denis Cloutier

Funding for this project was provided by:

Viterra, Inc.

Manitoba Pulse Growers Association – Soybean Committee

Agrifood Research & Development Initiative (ARDI)
Project #04-568

Special thanks to my wife, Tabitha, for putting up with this seemingly never-ending project and to my son, Samuel, who never let me forget that he was there.

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ABSTRACT

Gervais, Joseph Paul James. M.Sc., The University of Manitoba, May, 2009. Nitrogen and Phosphorus Fertilization of Soybean (*Glycine max* [L.] Merr.) in the Red River Valley Region of Manitoba, Canada. Major Professor; Jane Froese.

Nitrogen (N) and phosphorus (P) fertilization of soybean was studied in two separate experiments in the Morris, St. Norbert and Homewood areas of the Red River valley in Manitoba from 2004 to 2006. All fields had a history of soybean production. In each experiment, six rates of fertilizer were applied using three application practices. For the nitrogen study: 0, 25, 50, 75, 100 and 125 kg N ha^{-1} were applied either as a single application at seeding with granular *Bradyrhizobium japonicum* inoculant, as a single application at seeding without *B. japonicum*, or as a split application with 25 kg N ha^{-1} applied at seeding with *B. japonicum* inoculant and the balance applied at R3 (early pod). For the phosphorus study: 0, 15, 30, 45, 60, and 75 kg P_2O_5 ha^{-1} was either seed-placed, banded 2.5 cm below the seed or seed-placed with *Penicillium bilaiae*-inoculated seed. Data was collected on emergence, nodulation, biomass, yield, seed weight, seed protein, seed oil, and seed phosphorus content. Nitrogen fertilization of soybean resulted in significant negative emergence, nodulation and seed oil content responses as well as a significant but small positive response of seed weight. Applying the N fertilizer as a split application resulted in a small positive response for seed protein content and a small negative response for seed oil content. Inoculation with *B. japonicum* was not significantly different from the non-inoculated practice. Overall, there was no significant effect found on soybean yield as a result of N fertilization regardless of the application practice used. For the phosphorus experiment, P fertilizer had almost no effect on soybean production. The P component of the mono-ammonium phosphate fertilizer did not negatively affect seedling emergence regardless of placement. Inoculation with *P. bilaiae* had almost no effect on soybean production. Only a mild response to P rate was observed for seed protein content (negative) and seed P content (positive). Overall, N fertilization of soybean in the Red River valley of Manitoba is not recommended and the use of expensive granular *Rhizobium* inoculants may not be necessary on land that has grown well-nodulated soybean crops in the past. Phosphorus fertilizer for soybean should only be applied to soils that are low in available phosphorus.

1. INTRODUCTION

Soybean (*Glycine max* (L.) Merr.) production in Manitoba has been steadily increasing over the last decade. In 1996, only 320 hectares of soybeans were grown in Manitoba. Ten years later, in 2006, Manitoba soybean production reached 142,000 hectares. In 2008, soybean production in Manitoba decreased slightly to 126,000 hectares but this overall explosion of soybean hectares over the past decade was made possible by the development of short season soybean cultivars suitable for production in the shorter growing season of Manitoba. The majority of soybean production occurs in the Red River valley region of the province.

Soils in the Red River valley of southern Manitoba are predominantly heavy clay soils with potential for high productivity (Ehrlich et al., 1953). These soils need to be properly managed and may have problems with excess water in some years. This area is suitable for the production of most major Canadian grain crops including: cereals, oilseeds, and pulses. Conventional tillage regimes tend to dominate the area as the excessive amounts of crop residue produced in most years prevents the shift to minimal or zero tillage. The average length of the frost-free period for the Red River valley is 105 to 125 days (Manitoba Agriculture, Food and Rural Initiatives, 2009a). The average growing degree days (GDD) above 10°C for the Red River valley ranges from 1000 closer to Winnipeg to 1150 at Morris and south to the Canada-US border (Manitoba Agriculture, Food and Rural Initiatives, 2009b). The average growing season precipitation ranges from 190 mm to 270 mm (Manitoba Agriculture, Food and Rural Initiatives, 2009c).

Another factor responsible for this sudden interest in soybean production by Manitoba farmers is the potential to lower their annual fertilizer bill by including this dinitrogen (N_2)-fixing crop in their rotation. Soybean, like other legumes, establishes a symbiotic relationship with the *Bradyrhizobium japonicum* bacteria. *Rhizobium*, in exchange for energy, transforms atmospheric dinitrogen into a form the plant can use. This allows soybean to meet its nitrogen requirement in soils that are low in available nitrogen. Soybean requires 180 to 225 kg N ha^{-1} to produce an average yield of 2300 kg ha^{-1} (Canadian Fertilizer Institute, 2001). There is debate as to how much of this N requirement is supplied by dinitrogen fixation and how much is supplied by available nitrogen in the soil. If soybean yield responds to an application of N fertilizer, then it is likely that dinitrogen fixation is not meeting the requirements of the crop. In which case, supplemental nitrogen fertilizer will be essential to achieve maximum yields.

Soybean requires 30 to 40 kg P_2O_5 ha^{-1} to produce a 2300 kg ha^{-1} seed yield (Canadian Fertilizer Institute, 2001). Concerns over the increasing concentrations of phosphorus appearing in Lake Winnipeg have led to new laws regulating the application of phosphorus fertilizer to farmland. Farmers must now be conscientious in their fertilizer applications so as not to increase soil P levels unnecessarily. In addition, the cost of phosphorus fertilizer has reached record levels in recent years furthering the demand for more economically diligent P application.

Rationale for the Current Study

Of the numerous studies available on the fertilization of soybean, the majority were conducted in either southern Ontario or the Corn Belt region of the USA. There is a very limited amount of research that has been conducted under Manitoba conditions. Both of the southern Ontario and U.S. Corn Belt regions are characterized by longer growing seasons and warmer temperatures. In addition, these studies were often conducted under row-crop conditions, irrigation, and/or simple two-crop rotations (corn-soybean). Soybean production in Manitoba is often solid-seeded, grown under dryland conditions, and often with more complex crop rotations. Also, there is little information on the inoculation of soybean with *Penicillium bilaiae* grown under Manitoba conditions. Finally, the majority of P fertilization of soybean studies have used triple super phosphate ($Ca(H_2PO_4)_2$) and other "only-P" fertilizers. Nitrogen-containing fertilizers like mono-ammonium phosphate (MAP; $NH_4H_2PO_4$) are more commonly used in Manitoba. All of these reasons make it necessary to reinvestigate the fundamental agronomic principles such as nitrogen and phosphorus fertilization of soybean.

Objectives

The objectives of the nitrogen study were to investigate the response of soybean to rate of N fertilizer, to determine the effectiveness of split applications of N fertilizer and to establish the need for application of *Rhizobium* inoculants to soybean crops grown on soil that has a history of soybean production.

The objectives of the phosphorus study were to examine the response of soybean to rate of P fertilizer, to establish a preferred rate and placement of P fertilizer, and to evaluate the impact of inoculation with *Penicillium bilaiae* on soybean production.

2. LITERATURE REVIEW

Nitrogen Fertilization of Soybean

Nitrogen Fertilizer Rate

Nitrogen Rate and Emergence

It is well known that nitrogenous fertilizer placed close to the seed at high rates can result in damage to germinating seedlings and subsequently poor emergence. Even if the fertilizer is not placed in close proximity to the seed, reduced plant stands can occur if too much nitrogen is applied. Welch et al. (1973) found broadcast and disked in applications of extremely high rates of N fertilizer (1440 and 1800 kg N ha^{-1}) resulted in reduced emergence and subsequent yield reductions in soybean.

Nitrogen Rate and Nodulation

There have been numerous studies revealing the negative effect of nitrogen fertilizer on the nodulation of soybeans. Nitrogen fertilizer has been shown to cause a decrease in nodule weight (Hardarson et al., 1984), and nodule size (Koutroubas et al., 1998; Buttery et al., 1988; Ham et al., 1975; Semu and Hume, 1979a; Starling et al., 1998; Hardarson et al., 1984; Taylor et al., 2005; Hesterman and Isleib, 1991; and Chen et al., 1992) or a reduction in the quantity of nodules (Ham et al., 1975; Semu and Hume, 1979a; Starling et al., 1998; Taylor et al., 2005; Hesterman and Isleib, 1991; Koutroubas et al., 1998; Beard and Hoover, 1971; La Favre and Eaglesham, 1987; Gibson and Harper, 1985; and Chen et al., 1992). These reductions have been reported to follow a linear trend (Koutroubas et al., 1998; Semu and Hume, 1979a; Beard and Hoover, 1971; and Chen et al., 1992).

Nitrogen fertilizer does not directly affect nodulation, but rather it is the resultant increase in soil nitrogen levels that adversely affects nodulation. Starling et al. (1998) and Herridge (1988) found that nodule numbers and mass decreased as soil nitrate (NO_3^-) concentrations increased. Herridge (1984) reported that high initial soil NO_3^- levels (30 ppm in top 0 - 30 cm soil) delayed initiation of the nodule, retarded nodule development and reduced the overall extent of nodulation.

Nitrogen-fixing legumes like soybean use all available sources of nitrogen for their growth and development and will preferentially utilize available soil nitrogen over fixing their own nitrogen. Increased soil nitrate levels have been reported to inhibit dinitrogen fixation in soybean (Herridge et al., 1984; Harper and Gibson, 1984; Gan et al., 2004). Such decreases in dinitrogen fixation have been reported to coincide with decreases in nodulation (Semu and Hume, 1979a; Ham et al., 1975; Gan et al., 2004). Goss et al. (2002) concluded increasing the rate of applied nitrogen fertilizer caused a subsequent decrease in nodulation as well as a decrease in the percent nitrogen derived from the atmosphere. In contrast, Allos and Bartholomew (1959) reported increasing nitrogen fertilizer rate resulted in an increase in legume growth and nitrogen uptake and in some cases this resulted in an increase in N_2 fixation. However, once the applied nitrogen rate exceeded the amount of nitrogen necessary for growth, there was a tendency for the fertilizer to substitute for dinitrogen fixation. Gan et al. (2004) and Gulden and Vessey (1998) found high concentrations of N reduced nodulation as well as dinitrogen fixation, yet at low concentrations of mineral N, nodulation and dinitrogen fixation were increased.

The soybean nodulation response to nitrogen may be influenced by soil moisture levels. Smith and Hume (1985) found nodule size was decreased by 50% in the plots where 200 kg N ha^{-1} was applied compared to unfertilized plots. However, when the nitrogen was applied to irrigated plots, the fertilizer had no effect on nodule size. Buttery et al. (1998) also found low soil moisture content resulted in reduced nodule weights in clay soil and reduced nodule numbers as well as nodule weights in sandy loam soil. Finally, Muldoon et al. (1980) reported a dry weather period during June and July of 1978 limited nodulation. Lyons and Earley (1952) also found the response of soybean to nitrogen fertilizer varied depending on soil moisture availability. In a hot, dry year an application of 100 lbs $acre^{-1}$ (112 kg ha^{-1}) of ammonium nitrate resulted in an 80 to 90% decrease in number of nodules, whereas in a wet year, applications of up to 1000 lbs $acre^{-1}$ (1120 kg ha^{-1}) resulted in only a 35% reduction in number of nodules. They concluded this effect was a result of soil moisture effects on nodulation and presumably dinitrogen fixation. The authors speculated that in dry years, nodulation was impaired enough so that the plants responded to nitrogen fertilizer whereas in wet years the nodules developed fully and were able to fix the nitrogen requirements of the plants. This is contrary to the idea that soybean will use soil nitrogen preferentially over fixed-nitrogen, due to the high energy cost of dinitrogen fixation. It is possible that the lack of response of nodulation to nitrogen fertilizer in wet years may be due to an increase in the amount of nitrogen lost as a result of leaching.

It may be possible to partially counteract the negative effect of nitrogen fertilizer on nodulation by increasing the rate of applied inoculant. Muldoon et al. (1980) found nodule number generally increased with greater numbers of applied *Rhizobium*. In 1988, Herridge and Brockwell (1988) reported the inhibition of soybean nodules due to high soil nitrate levels could be reduced by the application of higher rates of inoculant. At normal rates of inoculant (250 g peat culture inoculant kg^{-1} seed or approximately 500,000 rhizobia seed^{-1}) nitrogen fertilizer rates up to 300 kg N ha^{-1} as ammonium nitrate resulted in significant reductions in nodule numbers and size. However, when inoculant was applied at 100 and 1000 times the normal rate, the negative effect of nitrogen fertilizer was considerably reduced.

Despite the negative effect of nitrogen fertilizer on soybean nodulation, the *Rhizobium* population in the soil may not be affected by an increased nitrogen supply. In Ontario, Semu and Hume (1979b) reported decreases in number and mass of nodules due to the application of nitrogen fertilizer but found no effect of the fertilizer on the soil *Rhizobium* populations.

The negative effect of soil nitrogen on nodulation may be directly related to the length of time after inoculation when the plant is exposed to soil nitrogen. Malik et al. (1987) found an inhibitory effect of nitrate nitrogen on nodule formation in soybean. However, when the exposure of the roots to nitrate was delayed for 18 hours after inoculation, inhibition of nodules was reduced by a factor of 2.5. The researchers suggested nitrate has an inhibitory effect on an infection event that completes within 18 hours after inoculation and once completed, exposure to nitrate has little to no effect on nodulation. Gibson and Harper (1985) found when the initial nitrate concentration is allowed to "run-down" nodule formation increased as the nitrate concentration decreased.

They also found the addition of NO_3^- significantly delayed the initial nodulation of soybeans.

On the contrary, nitrogen fertilizer has in some cases, been found to enhance the nodulation of soybean. Tewari et al. (2004) found nodule dry weight, quantity, and size was increased by the deep-placement of three different forms of nitrogen fertilizer (urea, coated urea, and calcium cyanimide). Johnson and Hume (1972) reported the application of ammonium nitrate as well as a high rate of manure decreased nodulation, however, the addition of organic matter (ground corn cobs with or without manure) or the addition of manure alone, increased the weight per nodule. Hesterman and Isleib (1991) found a positive effect of nitrogen fertilizer on nodule size at one site-year out of four. The remaining site-years showed a negative response. Dadson and Acquaah (1984) reported significantly more nodules per plant when less than 40 kg N ha^{-1} was applied compared to the highest rate of 160 kg N ha^{-1}. Results such as these may indicate that nitrogen was the most limiting nutrient in these studies and therefore did not cause a reduction in nodulation as the plant needed to utilize all sources of nitrogen to meet its requirements.

Nitrogen Rate and Plant Height

Soybean plant height has been reported to be both significantly increased or unaffected by nitrogen fertilizer. Taylor et al. (2005) found no effect on plant height when nitrogen fertilizer was applied at the R1 stage (Table 2-1). Ham et al. (1975) reported plant height was either increased or unaffected by nitrogen fertilizer depending on location and year, whereas Starling et al. (1998) reported plant height at R1 was significantly increased by approximately 3 cm as a result of the application of 50 kg N ha^{-1} of starter nitrogen. Bharati et al. (1986) reported nitrogen fertilizer significantly increased soybean plant heights by approximately 3 cm and this height increase coincided with an increase in lodging.

Table 2-1. Stages of Soybean Development

Development Stage	Description
V1	Unfolded leaf at first node (unifoliate node)
V2	Unfolded leaf at second node
V3	Unfolded leaf at third node
Vn	Unfolded leaf at n node
R1	One flower at any node
R2	Flower at node immediately below the uppermost node of main stem with a completely unfolded leaf
R3	Pod 0.5 cm long at one of the four uppermost nodes of main stem with a completely unfolded leaf
R4	Pod 2 cm long at one of the four uppermost nodes of main stem with a completely unfolded leaf
R5	Beans beginning to develop at one of the four uppermost nodes of main stem with a completely unfolded leaf
R6	Pod containing full-sized green beans at one of the four uppermost nodes of main stem with a completely unfolded leaf
R7	Pods yellowing, 50% of leaves yellow. Physiological maturity.
R8	95% of pods brown. Harvest maturity.

Source: Fehr and Caviness, 1977 and Fehr et al., 1971

Nitrogen Rate and Biomass Production

Several researchers have found additions of nitrogen fertilizer significantly increased above ground dry matter production in soybean. Dadson and Acquaah (1984) reported the highest levels of dry matter production were associated with the highest rates of applied nitrogen (80 and 160 kg N ha^{-1}). In an experiment by Taylor et al. (2005), dry matter production at the R1 growth stage was increased by the application of nitrogen fertilizer at three different planting dates and Bhangoo and Albritton (1976) found soybean vegetative matter was increased with the application of nitrogen over the non-nitrogen treatments.

Other researchers such as Barker and Sawyer (2005), Schmitt et al. (2001), and Buttery et al. (1998) reported no effect of nitrogen fertilizer on the above ground dry matter production of soybean. Purcell and King (1996) hypothesized that the lack of response to nitrogen fertilization they observed in their irrigated treatments may be due to a slight decrease in plant population due to fertilizer injury when the fertilizer was applied at the V6 stage.

Nitrogen Rate and Yield

Overall, the response of soybean seed yield to nitrogen fertilizer tends to be quite variable and dependent on many other factors such as growing season temperatures, soil moisture, crop variety, soil nitrogen levels, strain of inoculant, timing of fertilizer application, and cropping history.

Researchers have reported a negative effect of nitrogen fertilizer on seed yield; however, the response tends to be inconsistent and dependent on year and location. Welch et al. (1973) did not find a consistent effect of nitrogen fertilizer on seed yield in Illinois. They found nitrogen applications up to 1800 kg N ha^{-1} reduced yield due to a reduction in plant populations resulting from the high fertilizer rate. However, in a second experiment, 1440 kg N ha^{-1} reduced emergence and yield in only one year out of two and actually increased yield in the second year. In Ontario, Semu and Hume (1979a) found nitrogen fertilizer rates up to 200 kg N ha^{-1} nitrogen fertilizer produced a mixed effect on soybean yield. At Ridgetown, they found a linear decrease in nodulation and N_2 fixation in response to nitrogen fertilizer but no overall yield effect leading them to believe that nitrogen fertilizer simply replaced dinitrogen fixation. At their Elora location, nitrogen fertilizer had a negative effect on yield presumably due to increased lodging with higher nitrogen rates. At Woodstock, they reported a significant positive effect for only one year out of two. The positive yield response was likely because the nodules were unable to supply the amount of nitrogen needed for the high yields produced that year, whereas, in 1977, yields were lower along with the nitrogen demand and the nodules apparently supplied enough nitrogen. Chen et al. (1992) found a response at only one out of three site years. Reese and Buss (1992) reported a positive effect on yield due to the application of starter nitrogen (28 kg N ha^{-1}) at only one of ten environments. The negative effect of nitrogen on yield has been reported to be a result of increased lodging (Cooper, 1971; Bharati et al., 1986) or stand reduction (Welch et al., 1973).

Crop residues may play an important role in determining whether or not there is a yield response to nitrogen fertilizer. Peterson and Varvel (1989) found soybean grain yields responded positively to nitrogen fertilizer application but only when the soybeans were grown after sorghum. When soybeans followed corn in rotation, nitrogen fertilizer did not increase soybean grain yields and actually caused a decline in seed yield at the highest fertilizer rate (68 kg N ha^{-1}). They suggest there were higher rates of nitrogen immobilization in the plots that followed sorghum compared to corn and that this nitrogen may have become available later in the growing season during the pod-fill stages thereby increasing yields. In contrast, Beard and Hoover (1971) however, found there was no significant difference in yield due to nitrogen fertilization when the barley straw from the previous crop was burned compared to when it was shredded.

Many researchers have found no response of soybean seed yield to external sources of nitrogen such as organic matter or nitrogen fertilizer. Fertilizer nitrogen has been shown to have no effect on seed yield (Koutroubas et al., 1998; Buttery et al., 1988; Criswell et al., 1976; Schmitt et al., 2001; Heatherly et al., 2003; Beard and Hoover, 1971; and Freeborn et al., 2001), and additions of organic matter did not affect seed yields either (Criswell et al., 1976).

Several researchers have discovered a positive response of soybean seed yield to nitrogen fertilization. However, these yield responses tended to be dependent on many different factors such as soil moisture (Lyons and Earley, 1952; Purcell and King, 1996; Ray et al., 2006b; Starling et al., 1998; and Bhangoo and Albritton, 1972), soil nitrate content (Taylor et al., 2005; Wood et al., 1993; Lamb et al., 1990; Stone et al., 1985; and Bhangoo and Albritton, 1976), soil texture (Hesterman and Isleib, 1991) and poor nodulation (Herridge, 1988; Johnson and Hume, 1972; Ham et al., 1975; and Johnson et al., 1975). Yield responses have been reported to follow a linear (Chen et al., 1992; Lamb et al., 1990; Semu and Hume, 1979a), or quadratic trend (Taylor et al., 2005) and may be due to an increase in seed number (Koutroubas et al., 1998; Ray et al., 2006b, Stone et al., 1985; Purcell and King, 1996; and Taylor et al., 2005; Starling et al., 1998) or seed size (Diebert et al., 1979; Ham et al., 1975; Dadson and Acquaah, 1984; Sorensen and Penas, 1978; Semu and Hume, 1979a).

Nodulation of soybean plants appears to be dependent on soil moisture levels. Researchers have found in dry years, plants tend to form fewer nodules and show positive yield responses to nitrogen fertilizer. In wetter years, however, yield responses to nitrogen fertilizer are small or do not occur. Older research by Lyons and Earley (1952) found in a dry year, applications of only 35 kg N ha^{-1} as ammonium nitrate resulted in increased yields. Whereas in a wet year, the application of up to 360 kg N ha^{-1} resulted in only a slight increase in yield. They attributed this effect to the result of soil moisture effects on nodulation and subsequently, dinitrogen fixation. Similar results have been obtained under irrigated soybean production. Purcell and King (1996) discovered no response of seed yield to nitrogen fertilizer under irrigated conditions but they did find a response under non-irrigated conditions. Starling et al. (1998) noted their greatest yield response due to nitrogen fertilizer application was at irrigated locations or at those which received rainfall within 24 hours of fertilization. Hesterman and Isleib (1991) noted N fertilizer applied at 120 lb/acre increased seed yields from 2.9 - 10.8 bu $acre^{-1}$ (430 – 1600 kg ha^{-1}) on loam soil but had no effect on clay soil presumably due to the difference in water holding capacity of the two different soil types.

Other researchers reported yield responded similarly under both irrigated and non-irrigated conditions. Ray et al. (2006b) studied the effects of high rates of nitrogen fertilizer (290 to 360 kg N ha^{-1} broadcast shortly after seeding as ammonium nitrate) compared to no fertilizer under irrigated and non-irrigated conditions. They found nitrogen fertilizer increased seed yield by 7.71% under irrigated conditions and by 15.53% under non-irrigated conditions and suggest this difference highlights the sensitivity of dinitrogen fixation to soil moisture levels.

Some researchers have reported the opposite; concluding yield increases resulting from nitrogen application are more pronounced under well-watered conditions. Starling et al. (1998) noted a 9% yield increase when 50 kg N ha^{-1} of starter nitrogen (as ammonium nitrate) was broadcast and incorporated before seeding at all of their locations, but the increase was greater at the irrigated locations and at the ones that received rainfall within 24 hours of nitrogen application. In contrast, Bhangoo and Albritton (1972) found an application of 112 kg N ha^{-1} (applied in three equally split applications) resulted in yield increases irrespective of the amount of moisture available during the growing season in each of three years.

Soybean grain yield response to nitrogen fertilizer has been found to be related to soil nitrate content. Taylor et al. (2005) reported positive seed yield responses from nitrogen fertilizer rates up to 75 kg N ha^{-1} on land with less than 8 kg NO_3-N ha^{-1} immediately before planting. Wood et al. (1993) reported a yield response to nitrogen fertilizer but only at the locations that had soil nitrate test values of less than 24 kg N ha^{-1} although the yield responses were not consistent. A study by Lamb et al. (1990) revealed a positive effect of nitrogen fertilizer on soybean yields but only on soils with an NO_3-N content of less than 90 kg N ha^{-1}. In these cases, the yield was still increasing when their highest nitrogen rate was applied (134 kg N ha^{-1}) indicating that maximum yields were not obtained on these soils. When the soil NO_3-N content was greater than 90 kg N ha^{-1} there was no response to added nitrogen fertilizer. Other research has found a positive yield response with residual soil nitrate levels up to 190 kg ha^{-1} (Stone et al., 1985). Bhangoo and Albritton (1976) concluded that all sources of nitrogen (soil, fertilizer and N_2-fixation) are necessary for optimum soybean yields.

Soybean crops that have inadequate nodulation appeared to have greater responses to nitrogen fertilization. Herridge and Brockwell (1988) reported yield increases in non-inoculated treatments when nitrogen fertilizer was added compared to almost no response in the inoculated treatments. Even though the application of ammonium nitrate increased seed yield over the control, Johnson and Hume (1972) found the application of 280 kg N ha^{-1} as NH_4NO_3 did not reach comparable yields to the 2856 kg ha^{-1} harvested from well-nodulated plants in the nearby area. As well, Ham et al. (1975) found a greater yield response to nitrogen fertilization in the non-nodulating isolines they tested in their experiment compared to the nodulating isolines. Similar results were obtained by Johnson et al. (1975) who discovered a seed yield response to nitrogen fertilizer in non-nodulating soybean and no response in nodulating soybean. In Ontario, Goss et al. (2002) reported no clear response in nodulating soybean but non-nodulating soybean did respond to increasing nitrogen fertilizer rate.

Inoculation with *Bradyrhizobium japonicum*

Inoculation and Plant Population

The application of *Rhizobium* inoculant has not been reported to have any negative effects on soybean germination and emergence (Muldoon et al., 1980). However, the form of inoculant has been reported to have a negative effect on plant populations but only indirectly. Semu and Hume (1979a) found populations were lower in the inoculated plots compared to the non-inoculated plots. They concluded this was an effect of the peat-based inoculant causing interference to seed flow through the seeding equipment, despite the fact it occurred both years at only one location out of three. At another location, they found lower plant populations (26.5 plants m^{-2}) in the non-inoculated treatment compared to the inoculated treatment (31.2 plants m^{-2}) but could offer no explanation.

Inoculation and Nodulation

Only Buttery et al. (1988) reported a negative effect of *Rhizobium* inoculation on nodulation of soybean. In the second year of their experiment, they found inoculated treatments had lower nodulation than the non-inoculated treatments. Although they were unable to explain this effect they speculated it was not directly related to nodulation itself.

Once *Rhizobium* populations have become established in a particular soil, responses to applications of *Rhizobium* inoculant are minimized. Semu and Hume (1979a) found a significant effect of inoculation on the number of nodules and nodule dry weight but only at a location that had not grown soybeans previously and therefore had never had inoculant applied to the soil. Hesterman and Isleib (1991) also found nodulation was increased by the application of inoculant at a location with no history of soybean production.

A lack of response to inoculant has been observed not only in soils that had inoculant applied in recent years but also in soils that last had inoculant applied decades earlier. In a study in Manitoba, McAndrew and Brolley (2003) found many well-developed nodules in check plots that had not received inoculant that year but had grown soybean approximately 15 and 29 years previously. They concluded that *Rhizobium* survive in Manitoba soils for long periods of time despite the lack of a host crop. Nelson et al. (1978) found total plant nodule weight was unaffected by soil or seed-applied inoculants despite having been 15 years since the last time soybean had been grown on the field. They concluded there is little justification for recommending the application of inoculant to soybean except in areas that had never grown soybean or where a previous crop was poorly nodulated. Hesterman and Isleib (1991), at one of their experiment locations, found the non-inoculated treatments did not consistently have fewer and smaller nodules compared to the inoculated plots. The researchers suspected naturalized *Rhizobium* were established at this location despite the fact the field had no history of soybean production. Pulver et al. (1985) reported Nigerian and Indonesian cultivars grown in Nigeria intermittently responded to inoculation with *B. japonicum* unlike cultivars that were developed in the U.S.A. for which inoculation with *B. japonicum* was essential in order to achieve maximum yields. This effect was attributed to the lack of compatibility between the native Nigerian *Rhizobium* spp. and the U.S. cultivars which required inoculation specifically with *Bradyrhizobium japonicum*.

Inoculation and Biomass

Inoculation with *Bradyrhizobium japonicum* has been reported to affect biomass production. In 1984, Dadson and Acquaah reported significantly greater biomass production per plant for inoculated soybeans compared to non-inoculated soybeans. They also found the inoculated plots treatments to have taller plants than the non-inoculated treatments. McAndrew and Brolley (2003), in a Manitoba study, also found well-nodulated plants to be taller. In contrast, Hesterman and Isleib (1991) found no effect of inoculant type or inoculant rate on soybean plant height; however, they did find nodules developing on non-inoculated plants indicating that there may have been naturalized *Rhizobium* present despite field histories that did not include inoculated soybeans.

Inoculation and Seed Yield

Inoculation of soybeans with *B. japonicum* has a tendency to increase seed yields in soils that have never grown inoculated soybean before. McAndrew and Brolley (2003) found soybean yields on new soybean land in Manitoba were maximized with the application of a commercial granular *B. japonicum* inoculant. Yields were increased from 1748 kg ha^{-1} in the non-inoculated treatment to 2622 kg ha^{-1} in the granular inoculant treatment. In Ontario, Semu and Hume (1979a) found a positive effect of inoculant on yield but only at the one location that had never grown soybean in the past. Although the seed yields of both the inoculated and non-inoculated treatments did not respond to nitrogen fertilizer (due to very high soil N levels), Koutroubas et al. (1998) found a positive response of inoculation on soybean seed yield. These results were obtained on land that had never grown soybean previously and had no native *B. japonicum* as evidenced by the lack of nodule formation on the non-inoculated treatments. Other researchers have also reported a similar positive effect of inoculation on soybean seed yields in areas that did not have indigenous *Rhizobium* populations (Muldoon et al., 1980; Dadson and Acquaah, 1984; Ciafardini and Barbieri, 1987).

Yield increases due to inoculation have even been reported on old soybean land. Buttery et al. (1988) found a yield increase with the application of *Rhizobium*, but only in one out of two years. The authors did not mention whether soybean had been grown previously, but the non-inoculated treatments were well nodulated suggesting soybean inoculant had been applied at some point in the past.

The application of inoculant does not always translate into a yield increase on new soybean land. In northwest Minnesota, Lamb et al. (1990) found a double rate of peat-based inoculant did not significantly improve yields over the non-inoculated treatments at 10 out of 12 site years despite the fact only one of the sites had grown soybeans previously. The non-inoculated treatments had little to no nodules, but the inoculated treatments, for the most part, only had nodules develop on the crown region of the root. They concluded the lack of response to inoculation was because not enough *Rhizobium* were applied despite applying double the recommended rate.

For the most part, soybean yields do not respond to the application of *Rhizobium* on land with a history of soybean production. Nelson et al. (1978) found no response of soybean yield to both seed and soil applied inoculants in "corn belt" soils of the U.S.A. where soybeans are grown on a regular basis. Muldoon et al. (1980) and Semu and Hume (1979a) found no yield response at locations that had grown soybeans previously. Buttery et al. (1988) did find a yield increase with the application of *Rhizobium*, but only in one out of two years.

Rate and Inoculation on Seed Quality

Increasing the nitrogen supply to soybean, either by nitrogen fertilizer application or by inoculation, has a tendency to increase the seed protein content and decrease seed oil content. Dadson and Acquaah (1984) found both nitrogen fertilizer and inoculation reduced the seed oil content of soybean. The highest seed oil contents occurred in non-inoculated treatments. In Ontario, Yin and Vyn (2005) also found a significant decrease (-4.2 g kg^{-1}) in seed oil concentration was associated with each 1000 kg ha^{-1} increase in yield. Sometimes, despite a reduction in seed oil content, the overall oil yield increases as a result of increased seed yield (Ham et al., 1975). Similar to nitrogen fertilizer, increasing the rate of applied inoculant may also result in a negative response on seed oil content. In another experiment in Ontario, Muldoon et al. (1980) found a decrease in oil content as a result of the application of both soil-applied and seed-applied inoculants. At the lowest rate of soil-applied inoculant (1/4× recommended rate) oil content was decreased by 30.0 g kg^{-1} and increasing rates of inoculant (1/4×, 1/2×, and 1× recommended rate) resulted in a linear decrease in oil content.

Several researchers have reported no effect of nitrogen fertilization on seed oil content. Schmitt et al. (2001), Starling et al. (1998), Taylor et al. (2005) and Reese and Buss (1992) all reported nitrogen fertilizer did not significantly affect soybean seed oil content despite some of these studies being on soils with low nitrate levels (Schmitt et al., 2001; Starling et al., 1998) or low organic matter levels (Reese and Buss, 1992).

Many researchers have reported a positive effect of nitrogen fertilizer on soybean protein content (Schmitt et al., 2001; Ham et al., 1975; Dadson and Acquaah, 1984; Brevedan et al., 1978; Bhangoo and Albritton, 1972) as well as a positive effect of inoculation on protein content (Dadson and Acquaah, 1984; Ciafardini and Barbieri, 1987; Muldoon et al., 1980).

Several studies have found no significant effect of nitrogen fertilizer on soybean seed protein content (Stone et al., 1985; Taylor et al., 2005; Nelson et al., 1978). Another study by Reese and Buss (1992) concluded the application of nitrogen fertilizer to have a negative effect on seed protein content. Ray et al. (2006a) also reported a negative effect of nitrogen fertilizer on seed protein content; however, they reported overall protein yields (kg protein ha^{-1}) increased due to a significant increase in seed yield.

Soil moisture level may also be a deciding factor in whether or not seed protein content responds to nitrogen fertilizer. In a dry year, Lyons and Earley (1952) found low rates of nitrogen fertilizer (35 kg N ha^{-1}) significantly increased the nitrogen content of soybean seed whereas in a wet year, applications of up to 360 kg N ha^{-1} had no effect. This result was likely a result of the effect of moisture on soybean nodulation and subsequently, dinitrogen fixation.

The seed protein response may be partially determined by soybean genotype. Starling et al. (1998) reported the nitrogen fertilizer effect on seed protein content varied with genotype in their study. Deibert et al. (1979) as well as Johnson et al. (1975) found nitrogen fertilizer increased soybean seed nitrogen content but only in non-nodulating isolines and not in nodulating isolines. However, Bhangoo and Albrittton (1976) reported seed protein content of both nodulating and non-nodulating soybean isolines responded positively to nitrogen fertilizer. This response was more pronounced in the non-nodulating isoline.

Timing of Fertilizer Application

It has been hypothesized that one could delay the onset of leaf senescence and thereby increase yield potential by ensuring an adequate supply of nitrogen is available during the latter stages of development. Seed nitrogen demand is suspected to be the cause of leaf senescence in soybean. Buttery (1986) reported a large portion of the nitrogen in the seed of soybean was translocated from the non-seed parts of the plant. Sinclair and de Wit (1976) using a simulation model, concluded the duration of the seed filling period could be extended by increasing the N supply from the roots, and thus decreasing the demand for redistributed N. Wesley et al. (1998) found the application of supplemental nitrogen at the R3 stage resulted in increased yields. However, since the leaf N concentrations were not affected they suggest the added nitrogen was translocated directly to the seed and was not stored in the leaf. Hayati et al. (1995) concluded seed nitrogen demand is not responsible for leaf senescence and that it is likely regulated by processes in the leaf itself. Egli et al. (1978) found increasing the nitrogen supply during the seed filling period did not affect maturity or the amount of redistributed nitrogen compared to the control. They concluded that it is not possible to prevent the redistribution of N from the non-seed parts of the plant simply by increasing the N supply to the roots and that N redistribution is not the sole cause of leaf senescence.

Timing of Fertilizer Application on Nodulation

The soybean stage of development as well as the rate of nitrogen fertilizer being applied may influence the extent of the post-seeding nitrogen fertilizer effect on nodulation. Gan et al. (2003) found a positive effect of nitrogen fertilizer (50 kg N ha^{-1}) on nodulation when it was applied at the V2 stage, whereas they found a negative effect when it was applied at the R1 or R3 stages. In contrast, Beard and Hoover (1971) reported nitrogen fertilizer rates of up to 112 kg N ha^{-1} applied at flowering (R1) did not affect nodulation but when the rate was increased to 168 kg N ha^{-1}, the number of nodules was decreased.

The presence of non-decomposed organic matter may also influence the severity of the effect of the nitrogen fertilizer on nodulation. Beard and Hoover (1971) noted in the plots where the previous crop's straw had been burned, the N fertilizer effect on nodulation was more severe than what was observed in the plots where the straw was shredded. When crop residues are left intact in the field, soil microbes temporarily immobilize soil nitrogen in their tissues as they consume the residues as a carbon source. When the crop residues are burned, the amount of carbon available to the microbes is reduced and therefore less N is immobilized and can remain available for the soybean plant to uptake.

Timing of Fertilizer Application on Biomass

The effectiveness of supplemental nitrogen applications appear to be dependent on the stage at which they are applied. However, it is not clear as to which stage is ideal. Researchers did not find a significant effect of N fertilizer applied at the R2 stage of development (Schmitt et al., 2001; Barker and Sawyer, 2005; Deibert et al., 1979) R3 stage (Gan et al., 2003) R4 stage (Schmitt et al., 2001), or the R5 stage (Gan et al., 2003; Schmitt et al., 2001). However, Gan et al. (2003) did find a significant positive effect when the N fertilizer was applied at either the V2 or R1 stages of development and Afza et al. (1987) found a positive response when it was applied at the R4 stage.

Timing of Fertilizer Application on Yield

Previous research has shown the application of nitrogen fertilizer at later stages of development can result in a positive effect on soybean seed yield. Gan et al. (2003) reported nitrogen applied at either the V2 or R1 development stages significantly increased soybean seed yield but when the nitrogen application was delayed until the R3 or R5 stages, there was no significant effect observed. Afza et al. (1987) found that additional soil or foliar-applied nitrogen or a combination of both during pod-filling stage (R5) resulted in a seed yield increase of 37% and 40% over starter nitrogen alone in two experiments.

Positive yield increases due to split nitrogen applications can vary from year to year. In the first year of an experiment conducted by Brevedan et al. (1978), the application of nitrogen fertilizer at both beginning bloom (R1) and end of bloom (R4 or R5) increased seed yield by 28% over the control, but single N applications at either plant stage had no effect. The opposite occurred in the second year when single applications of N at either the beginning or end of bloom significantly increased seed yield by approximately 32%, but applying at both stages did not, likely due to increased lodging.

The occurrence of a positive yield response to late-season nitrogen application could be directly related to the rate of applied N fertilizer as well as the plant-available nitrogen levels in the soil. Wesley et al. (1998) reported nitrogen fertilizer applied at the R3 to R4 stage significantly increased yields at six out of eight locations; however, increasing the rate from 20 to 40 lb N acre^{-1} (22 to 45 kg N ha^{-1}) resulted in little difference. Wood et al. (1993) observed both positive and negative responses to post-seeding nitrogen fertilizer applications (at R5 stage) at five out of seven locations. They noted the two locations where no response was observed had a much higher soil nitrate content at planting than the five locations were a response occurred.

There is some evidence that applying nitrogen fertilizer at later stages of soybean development does not affect seed yield. Several studies have found no significant effect of applying nitrogen fertilizer at the V6 development stage (Purcell and King, 1996), the R1 stage (Beard and Hoover, 1971; Welch et al., 1973), the R2 stage (Barker and Sawyer, 2005; Purcell and King, 1996), the R3 stage (Barker and Sawyer, 2005; Freeborn et al., 2001; Gutierrez-Boem et al., 2004), the R4 stage (Schmitt et al., 2001), the R5 stage (Welch et al., 1973; Gutierrez-Boem et al., 2004), and the R6 stage (Freeborn et al., 2001).

Timing of Fertilizer Application on Seed Quality

Nitrogen fertilizer applied late-season has been found to cause small increases in soybean seed protein content (Schmitt et al., 2001) or protein and oil content (Welsey et al., 1998). Other studies have concluded there was no effect of late-season nitrogen fertilizer on soybean seed protein (Welch et al., 1973; Barker and Sawyer, 2005; Wood et al., 1993; Deibert et al., 1979) or seed oil content (Schmitt et al., 2001). The increase in protein content amounted to a difference of 0.4 g kg^{-1} (Schmitt et al., 2001), whereas Wesley et al. (1998) reported a protein increase of 10.0 g kg^{-1} but only at four out of eight locations and Wood et al. (1993) found an increase at only one out of seven locations. The increase in seed oil content reported by Wesley et al. (1998) ranged from a 3.0 to 5.0

g kg^{-1} increase and was only observed at three out of eight locations. In all studies, the nitrogen fertilizer was applied between the R1 and R5 development stages.

Phosphorus Fertilization of Soybean

Fertilizer Placement

Phosphorus Rate and Placement Effects on Emergence

Phosphorus fertilizer when placed in the seed row can result in a reduction in emerging seedlings. In a Manitoba experiment, Bullen et al. (1980) observed a reduction in seedling emergence when 26.2 kg P ha^{-1} applied as triple super phosphate ($Ca(H_2PO_4)_2$; 0-46-0) was placed with the seed. Similar reductions in soybean emergence were reported by Bailey (1977) at 0-46-0 rates of 40 kg P_2O_5 ha^{-1} (17.4 kg P ha^{-1}) also in a Manitoba study. Clapp and Small (1970) reported a reduced plant stand from applications of liquid and granular fertilizers applied in the seed row at rates as low as 5 and 1.7 kg P ha^{-1}, but low rates of nitrogen and potassium fertilizer were applied at the same time as the P. In contrast, Lauzon and Miller (1997) found no detrimental effect of placing 6.5 kg P ha^{-1} with the seed. In Manitoba, the provincial government's agricultural department recommends that no more than 22.4 kg P ha^{-1} be placed with the seed if it is applied using high seed bed utilization (SBU). With low SBU, these rates may result in stand reductions (Manitoba Agriculture, Food and Rural Initiatives, 2007).

Phosphorus Placement Effects on Biomass

Phosphorus fertilizer placement has been found to affect soybean dry matter yields. Bullen et al. (1980) found banded phosphorus fertilizer produced significantly higher dry matter yields compared to broadcast. Hairston et al. (1990) found banded phosphorus resulted in significantly taller plants compared to broadcast, however, their results varied from location to location despite low initial soil test P levels. On the other hand, Bullen et al. (1980) also observed a reduction in dry matter yield in their seed-placed fertilizer treatment due to decreased plant populations.

Phosphorus Placement and Yield

The application of phosphorus fertilizer varies in its effects on soybean yields. Rehm (1986), Ham et al. (1973) reported higher yield increases per unit of fertilizer for broadcast P compared to banded P. In contrast, Hairston et al. (1990) reported higher yields were obtained with banded P compared to broadcast P and Bullen et al. (1980) found placing the P fertilizer 2.5 cm directly below the seed resulted in higher yields than seed-placed, broadcast or side-banding 2.5 cm to the side and 2.5 cm below the seed.

Soybean seed yield responses to phosphorus fertilizer placement tend to be infrequent and inconsistent. In two separate studies, Borges and Mallarino found no consistent effect of placement on seed yield (2003) and although the application of P fertilizer increased seed yields at low soil P testing locations (5 – 33 ppm Bray-P), there were no differences between fertilizer placements (2000). Haq and Mallarino (2005) found P fertilizer placement had no effect on seed yield in 35 trials over a period of seven years at low soil P testing sites (4 – 31 ppm Bray-P). Ham and Caldwell (1978) also reported no significant difference between several different P fertilizer placements on soybean seed yield in soil testing 7.5 ppm Olsen-P.

Phosphorus Placement and Seed Quality

Phosphorus fertilizer placement appears to have little to no effect on seed protein and oil contents. Ham et al. (1973) concluded P fertilizer placement did not affect seed protein or oil content in their study (soil test P: 3.5 – 35.5 ppm Bray). Haq and Mallarino

(2005) found a negative effect of fertilizer placement on soybean oil content at only one out of 35 trials and there was no effect on seed protein concentration. Soil test P ranged from 4 to 31 ppm (Bray).

Phosphorus Fertilizer Rate

Phosphorus Rate and Nodulation

As with other growth parameters, nodulation also tends to have a varying response to phosphorus fertilizer. Dadson and Acquaah (1984) found no significant effect of phosphorus fertilizer rate on the number of nodules per plant or nodule dry weight despite extremely low available soil P levels of 0.02 ppm. In contrast, Jones et al., (1977) found fertilizer P application increased the number and weight of nodules per plant on soil with 2 ppm Mehlich-P. In a greenhouse study, de Mooy and Pesek (1966) found a highly significant positive effect of P on nodule number, weight and leghemoglobin content. The former study was on high soil test P and the latter on low P soil, indicating soil P test levels may not be an accurate predictor of nodulation responses to applied P fertilizer.

Phosphorus Rate and Biomass

Researchers have shown phosphorus fertilizer can have an effect on soybean vegetative growth but the response depends on the soil test P values. In one case, fertilizer rates of 60 and 90 kg P ha^{-1} (138 and 207 kg P_2O_5 ha^{-1}) produced greater biomass than the control in very low P (0.02 ppm) testing soils (Dadson and Acquaah, 1984). In another study, Hairston et al. (1990) found 45 kg P ha^{-1} (104 kg P_2O_5 ha^{-1}) and 125 kg K ha^{-1} fertilizer resulted in taller plants but at only one out of three locations, despite all three sites being low in soil P (2.2 - 8.8 ppm). Finally, Bharati et al. (1986) reported a significant increase in soybean plant lodging due to P fertilization (74 and 111 kg P ha^{-1}) in high P testing soils (69 to 73 ppm Bray-P); however, no significant increase in plant height was observed.

Phosphorus Rate and Yield

Phosphorus fertilizer has been reported to provide a yield advantage to soybeans grown on low P testing soils. Ham and Caldwell (1978) reported a significant yield increase (+800 kg seed ha^{-1}) from the application of 35 kg P ha^{-1}. Aulakh et al. (2003) reported a significant yield increase of +1000 kg seed ha^{-1} with phosphorus fertilizer application rates up to 43.2 kg P ha^{-1}. Both studies were on soil that tested less than 10 ppm extractable P (Olsen). Sometimes the yield response varies as in the experiment of Dadson and Acquaah (1984) who obtained mixed results on a very low P testing soil (0.02 ppm). They observed a yield advantage of +500 kg seed ha^{-1} with the application of 90 kg P ha^{-1} over the 30 kg P ha^{-1} treatment, but not the 0 or 60 kg P ha^{-1} treatments. Haq and Mallarino (2005) reported a yield increase ranging from 170 to 1000 kg seed ha^{-1} from the addition of 14 kg P ha^{-1} at seven out of twelve locations (over a period of seven years) all of which had soil test P values between 4 and 8 ppm (Bray). However the remaining five locations, which also had low soil P values, did not show a response to phosphorus fertilizer.

Low phosphorus fertilizer application rates often result in a yield response but further increases in applied P seldom produce additional yield responses. Hairston et al. (1990) found a yield increase to P fertilization (15 kg P ha^{-1}) at two out of three locations over the control, but increasing the rate to 45 kg P ha^{-1} was no different than the 15 kg P ha^{-1} rate despite the low soil test P values of 2 – 9 ppm. However, there was only one location where the yield response was consistent and it was only an increase of 134 kg seed ha^{-1}. In addition, Borges and Mallarino (2003) found positive yield responses to P fertilizer when soil test P levels were less than 19 ppm (Bray). These yield responses were always achieved with the lowest rate of applied P (15 vs. 556 kg P ha^{-1}). Webb et al. (1992), in a long term study, found a response to phosphorus fertilizer only occurred with the addition of 11 kg P ha^{-1} when soil test P levels were <16 to 20 ppm (Bray). Any further P additions were not significant.

In some instances, soybeans grown on low P testing soils still do not respond to the addition of phosphorus fertilizer. Haq and Mallarino (2005) reported five out of twelve low soil P testing locations (4 to 7 ppm Bray-P) did not have a yield response to phosphorus fertilizer rates of up to 56 kg P ha^{-1}. As well, Bhangoo and Albritton (1972) found small but insignificant yield increases (0 to +360 kg seed ha^{-1}) as a result of P application (40 kg P ha^{-1}) despite soils being low in available P (5.6 ppm).

Large yield responses have also been reported to occur on high P testing soils. In the experiment of Jones and Lutz (1971), the addition of phosphorus fertilizer resulted in a significant yield increase of 1000 kg ha^{-1}. This yield response occurred with rates up to 48.8 kg P ha^{-1} on soils that tested high for P content (34 ppm Mehlich-P). Webb et al. (1992) reported a significant soybean seed yield increase from the addition of 11 kg P ha^{-1} to two soils with soil P test values of 18 and 42 ppm (Bray). In both experiments, however, further increases in P rate did not affect yield.

On the other hand, phosphorus fertilizer has been reported to have little to no effect on soybean seed yield when soil test phosphorus levels are in the medium to high range. Haq and Mallarino (2005) reported soybeans at 28 out of 35 locations, all of which had soil P test values ranging from 10 to 35 ppm (Bray), did not respond to the application of phosphorus fertilizer rates of up to 56 kg P ha^{-1}. Slaton et al. (2001) also reported a lack of response to 60 kg P ha^{-1} applied to soybeans grown on soils with soil test P values ranging from 8 to 25 ppm (Mehlich-III). Webb et al. (1992) found no effect of phosphorus fertilizer rates of 34 kg P ha^{-1} on soil with an initial soil P test (Bray) value of 75 ppm. Several other researchers have reported similar results when maximum phosphorus fertilizer rates ranging from 44 to 111 kg P ha^{-1} were applied to soybean on soils testing from 28 to 72 ppm (Bray) (Seguin and Zheng, 2006; Buah et al., 2000; Mallarino et al., 1991; Bharati et al., 1986).

The inconsistent responses to phosphorus fertilizer applications on low as well as high soil test P soils indicates there may be other limiting factors preventing soybean from utilizing fertilizer P. The inconsistencies also suggest the method for determining soil phosphorus content may not be accurately measuring the plant available P fraction of the soil.

Phosphorus Rate and Seed Quality

The application of phosphorus fertilizer to soybean has been reported to affect soybean seed oil and protein content. In some cases, both oil and protein were increased by phosphorus fertilizer (Ramalingaswamy and Nabasimham, 1977; Dadson and Acqaah, 1984). More often, seed protein content was increased while seed oil content was decreased (Haq and Mallarino, 2005; Jones and Lutz, 1971) but the opposite has also been reported (Haq and Mallarino, 2005). Significant increases in protein ranged from 5 to 21 g kg^{-1} (Ramalingaswamy and Nabasimham, 1977; Dadson and Acquaah, 1984; Haq and Mallarino, 2005; Jones and Lutz, 1971) and decreases ranged from 4 to 8 g kg^{-1} (Haq and Mallarino, 2005). Significant increases in oil ranged from 6 to 23 g kg^{-1} (Ramalingaswamy and Nabasimham, 1977; Dadson and Acquaah, 1984; Haq and Mallarino, 2005) and significant decreases ranged from 4 to 14 g kg^{-1} (Haq and Mallarino, 2005; Jones and Lutz, 1971).

In one case, a high P fertilization rate (90 kg P ha^{-1}) was required to cause a significant change in seed oil content (Dadson and Acquaah, 1984), in another (Ramalingaswamy and Nabasimham, 1977) only moderate application rates (40 kg P ha$^{-1)}$ were necessary. For seed protein content, Dadson and Acqaah (1984) reported their lowest rate of 30 kg P ha^{-1} was enough to increase seed protein content and an increase to 90 kg P ha^{-1} had no further effect. Haq and Mallarino, 2005 reported at the responsive sites in their study, the lowest rate of P fertilizer (14 kg P ha^{-1}) was enough to influence seed oil and protein contents, whereas the highest rate of P fertilizer (56 kg P ha^{-1}) further affected oil and protein contents at only three of those sites. Phosphorus fertilizer rate had no significant effect on soybean seed protein (Ham et al., 1973; Seguin and Zheng, 2006; Bhangoo and Albritton, 1972) or oil content (Ham et al., 1973; Seguin and Zheng, 2006).

The presence or lack of response on both high and low P testing soils indicates soil test P values may not be an accurate predictor of a seed oil and protein responses to P fertilization. Responses have been found on soils which tested from 0.02 ppm (Dadson and Acquaah, 1984) to 34 ppm Mehlich-P (Jones and Lutz, 1971). Haq and Mallarino (2005) found a response on seven sites ranging from 4 to 18 ppm (Bray), but failed to find a response at their remaining 28 sites which ranged from 4 to 31 ppm. Ham et al. (1973) reported no effect on soils ranging from 7 to 71 ppm (Bray). Seguin and Zheng (2006) found no effect on soil with P test values of up to 82 ppm while Bhangoo and Albritton, 1972 found no effect with soil P test values of 5.6 ppm.

The phosphorus content of soybean seed has been reported to be affected by phosphorus fertilization. Aulakh et al. (2003) applied up to 43.2 kg P ha^{-1} to soybean grown on low P testing soil (5 ppm Olsen-P) and found the P content of the seeds increased significantly from 1.9 to 4.6 g kg^{-1}.

Inoculation with *Penicillium bilaiae*

Penicillium bilaiae is a fungus that is able to solubilize unavailable forms of soil phosphorus, making them available for uptake by plant roots. Cunningham and Kuiack (1992) discovered that *P. bilaiae* produces oxalic and citric acid and in laboratory culture observations and believed these acids were responsible for the phosphate-solubilization ability of this organism. However, there may be other mechanisms involved. Asea et al. (1988) found 0.1 N HCL was unable to release as much phosphate from rock phosphate at the equivalent media pH levels as that released by *P. bilaiae.*

Several studies have concluded crops inoculated with *P. bilaiae* are able to utilize sources of P that are unavailable to non-inoculated crops. Kucey (1988) found crops inoculated with *P. bilaiae* responded to additions of rock phosphate, a plant-unavailable form of phosphorus. Using ^{32}P-labelled fertilizer, Chambers and Yeomans (1990) found that wheat inoculated with *P. bilaiae* showed increased soil P uptake as compared to non-inoculated wheat. In a greenhouse study, Asea et al. (1988) found wheat inoculated with *P. bilaiae* took up P from sources unavailable to un-inoculated plants as well as P from rock phosphate. Kucey (1988) reported increased dry matter and P uptake in wheat inoculated with *P. bilaiae* in both a greenhouse and field study. He also reported the addition of rock phosphate further increased wheat dry matter production. An increase in $NaHCO_3$-extractable P has been documented in soils that were inoculated with *P. bilaiae* both with and without added rock phosphate (Kucey, 1988).

P. bilaiae may also have an effect on the root architecture of plants. Gulden and Vessey (2000), in a greenhouse study, found an increase in the proportion of root containing root hairs (22%) and an increase in the mean root-hair length (33%). In a field study, Vessey and Heisinger (2001) found an increase in root length and dry weight as a result of inoculation with *P. bilaiae* without fertilizer. On the other hand, there was no effect of *P. bilaiae* at the second site in their study, despite both sites having responded to P fertilization.

Greenhouse studies showing responses to inoculation with *P. bilaiae* may not be representative of what will happen under field conditions. Kucey et al. (1989b) concluded the effect on P uptake by plants due to an increase in plant available P by P-solubilizing organisms will be enhanced since rooting volumes in greenhouses are usually restricted.

Penicillium bilaiae **and Emergence**

There have been no studies specifically examining the effect of *P. bilaiae* on soybean emergence. However, in a study of wheat, Kucey (1988) and Goos et al. (1994) found no significant effect of inoculation with *P. bilaiae* on emergence. Kucey (1988) also reported relatively high rates of *P. bilaiae* had no effect on wheat seed germination or seedling survival.

Penicillium bilaiae, **Dinitrogen Fixation and Nodulation**

The nodulation of field pea appears to have a variable response to inoculation with *P. bilaiae* but it is doubtful the fungus has any serious antagonistic effects on nodulation. In a greenhouse experiment, Downey and van Kessel (1990) reported a significant reduction in N_2 fixation after inoculating pea with *P. bilaiae.* They speculated the production of organic acids by *P. bilaiae* may have reduced the rhizosphere pH enough to partially inhibit nodulation since *Rhizobium* prefers a neutral or alkaline pH. Unfortunately, they did not measure nodulation in their study. In contrast, Gleddie (1993), in a growth chamber study, reported an increase in pea nodulation score when *P. bilaiae* or 10 kg P ha^{-1} as triple super phosphate was added to inoculated pea. Vessey and Heisinger (2001) found no significant effect of *P.bilaiae* inoculation on the nodulation of field pea. Rice et al. (1994) reported the co-culture of *R. meliloti* and *P. bilaiae* to produce a common delivery system for both *Rhizobium* and *P. bilaiae* inoculants is possible. Today, there are products marketed under the name of TagTeam™ (Philom Bios) which consist of both a *Rhizobium* spp. inoculant and *P. bilaiae* for dual inoculation of several western Canadian pulse crops.

Penicillium bilaiae and Biomass

Penicillium bilaiae has been reported to significantly increase dry matter production in greenhouse studies of wheat (*Triticum aestivum* L.) (Asea et al., 1988; Kucey, 1987; Kucey, 1988; Chambers and Yeomans, 1991), field pea (*Pisum sativum* L.) (Gleddie, 1993) and field beans (*Phaseolus vulgaris* L.) (Kucey, 1987). Field studies have also shown a positive effect of *P. bilaiae* on wheat dry matter (Kucey, 1987; Kucey, 1988), as well as lentil (*Lens culinaris* Medik.) and field pea vegetative growth at sites that responded to P fertilizer application (Gleddie, 1993). All of these experiments were conducted on soil with a soil test P of less than 8.8 ppm. Grant et al. (1999) reported flax (*Linum usitatissimum* L.) straw yield was significantly increased by inoculation with *P. bilaiae* despite soil P test values ranging from 12 to 29 ppm (Olsen).

In a study under controlled conditions, Downey and van Kessel (1990) reported a significant increase in dry matter production in pea as a result of inoculation with *P. bilaiae*. However, when field pea was inoculated with both *P. bilaiae* and *R. leguminosarum*, they observed a significant decrease in dry matter production as well as total plant nitrogen. They propose the organic acids formed by the fungi may have impeded the nodulation process and subsequently dinitrogen fixation.

Chambers and Yeomans (1991) observed no differences in flax dry matter production of *P. bilaiae*-treated pots compared to untreated control pots in a growth chamber study despite low soil test P levels (< 8.8 ppm Olsen-P). In an earlier study, Chambers and Yeomans (1990) examined the effect of *P. bilaiae* on wheat in both a growth chamber and a field study. They found no significant differences in dry matter yields in the growth chamber study (3 ppm Olsen-P), but in the field study (8.1 to 11.9 ppm Olsen-P) significant differences were observed in the early stages of growth but they disappeared at the later stages. Goos et al. (1994) found similar results for wheat in North Dakota despite a response to P fertilization. Gleddie et al. (1993) reported a similar increase in dry matter production at the early development stages for canola (*Brassica napus* L.) but eight weeks after germination, there was no longer a significant response; soil test P in this study ranged from 4 to 17 ppm.

Even when a location shows a P fertilizer response, there still may not be a response to *P. bilaiae*. Goos et al. (1994) reported P fertilization significantly increased wheat dry matter production at the two to four leaf stage at all four of their test locations, and at three locations for the six to seven leaf stage, whereas, inoculation with *P. bilaiae* significantly increased dry matter production only at one location, and only at the two to four leaf stage. Vessey and Heisinger (2001) reported a response to inoculation with *P. bilaiae* at only one out of two locations despite the presence of a P fertilizer response at both locations. Rice et al. (2000) found dual inoculation of alfalfa (*Medicago sativa* L.) with *Sinorhizobium meliloti* and *P. bilaiae* did not significantly increase hay dry matter yields compared to inoculation with *S. meliloti* alone. Unlike the other studies listed, there was lack of response to P fertilization in this study (8.8 - 16.6 ppm Olsen-P).

***Penicillium bilaiae* and Yield**

There is some evidence to show inoculation with *P. bilaiae* can result in a reduction in seed yield. Grant et al. (1999) examined the response of flax to nitrogen and phosphorus fertilizers as well as inoculation with *P. bilaiae*. They found a lower seed yield in flax when it was inoculated with *P. bilaiae* compared to P fertilizer by itself (20 kg P ha^{-1}) in two site-years out of six. This negative effect of *P. bilaiae* was only apparent when they examined the P fertilizer and *P. bilaiae* treatments applied at the lowest nitrogen fertilizer rate (10 kg N ha^{-1}). Nonetheless, they were unable to offer an explanation for this effect. Overall, they concluded flax did not respond to inoculation with *P. bilaiae* in their study, most likely due to the relatively high soil P levels of their experimental sites.

Inoculation with *P. bilaiae* can increase yields of wheat (Kucey, 1988; Gleddie et al., 1991; Goos et al., 1994), lentil (Gleddie, 1993) and canola (Kucey, 1989a). In each of these experiments, soil test P levels were considered low (< 12 ppm). Gleddie et al. (1991) published results from a combined analysis of 55 wheat trials in western Canada. They found the addition of *P. bilaiae* with 0 or 10 kg P ha^{-1} resulted in significantly higher yields (+42 to 50 kg seed ha^{-1}) than P fertilizer alone, however *P. bilaiae* applied with higher rates of P fertilizer did not result in further yield increases. This effect was more apparent when they divided the trials into low P testing locations (< 9 ppm) and high P testing locations (> 9 ppm). The low P testing locations showed yield increases (+43 to 66 kg seed ha^{-1}) from inoculation with *P. bilaiae* with P fertilizer up to 30 kg P_2O_5 ha^{-1}. In a similar study of canola, Gleddie et al. (1993) concluded inoculation with *P. bilaiae* had no significant effect on seed yields at locations that did not respond to P fertilization. Overall, positive yield responses to inoculation with *P. bilaiae* tend to be limited to soils with low extractable P levels. However, the province of Manitoba recommended that if *P. bilaiae* is used on low P testing soils, P fertilizer rates should not be reduced, whereas on high P soils *P. bilaiae* may be substituted for P fertilizer (Manitoba Agriculture, Food and Rural Initiatives, 2007). Kucey (1988) concluded the yields of wheat plots that received both *P. bilaiae* and rock phosphate were equal to those obtained from the addition of the equivalent rate of P in the form of mono-ammonium phosphate on soil that had 4 ppm soil test P (Olsen). Inoculation with *P. bilaiae* increased wheat yields without applying P fertilizer by 66 kg ha^{-1} in North Dakota (Goos et al., 1994); soil test P was 8 - 12 ppm (Olsen). Gleddie et al. (1993) concluded the inoculation of canola with *P. bilaiae* with 10 kg P_2O_5 ha^{-1} produced similar yields to those achieved by the application of 20 kg P_2O_5 ha^{-1} alone.

Yield responses to *P. bilaiae* do not necessarily occur on soils that respond to P fertilization. Grant et al. (1999) found a yield response of flax to P fertilizer at one of their locations but they did not observe a similar yield response to *P. bilaiae* inoculation. Gleddie (1993) found inoculation with *P. bilaiae* had no significant effect on pea yields grown on soils that responded to P fertilization.

Low soil P test values may not be an effective tool to predict whether or not a yield response to *P. bilaiae* will occur. Chambers and Yeomans (1991) reported no yield response of flax to *P. bilaiae* despite low soil P test values of 5.3 to 8.8 ppm (Olsen). In an earlier study, Chambers and Yeomans (1990) concluded the inoculation of wheat with *P. bilaiae* in a growth chamber study did not significantly increase seed yield despite low soil P test levels of 3 ppm (Olsen). In their field study, at only one location (11.8 ppm soil test P) out of three did PB-50 (a commercial *P. bilaiae* inoculant) significantly increase yields and it was only when the PB-50 was applied with their highest P fertilizer rate (20 kg P_2O_5 ha^{-1}).

***Penicillium bilaiae* and Seed Quality**

There are no studies on the effect of *P. bilaiae* on soybean quality. However, in a field study at Plum Coulee, Manitoba, Kucey (1989a) found seed P content of canola was increased by the addition of *P. bilaiae* or MAP but not both. There was no affect of phosphate fertilizers or *P. bilaiae* inoculant on seed oil or protein content. At their Ellerslie, Alberta site, seed oil content was significantly increased by *P. bilaiae* when it was applied with 6.1 kg P ha^{-1} MAP or rock phosphate.

3. MATERIALS AND METHODS

Experimental Design

Environments

This study was conducted in Manitoba over a period of three years. In the preliminary year (2004), two experimental sites were established (Homewood and Morris). In the subsequent years (2005 and 2006) there were three experimental sites (Homewood, Morris and St. Norbert). The term *environment* is used to represent each combination of location and year. Each environment hosted two separate experiments: the first examined nitrogen fertility of soybeans and the second looked at phosphorus fertility. Both experiments evaluated the response of soybeans to increasing rates (R) of fertilizer when applied using three different practices (P).

Site Selection

Rhizobium has the ability to colonize and maintain populations in the soil to which they have been applied (McAndrew and Brolley, 2003; Nelson et al., 1978). As more and more Manitoba producers grow soybeans, *B. japonicum* becomes established in these areas and lands with no naturalized populations of *B. japonicum* disappear. Therefore, it was decided land that had grown inoculated soybeans at least once in the past would produce the most valuable results for Manitoba farmers. In addition, conducting the experiments on soil that had grown soybean would allow an evaluation of the need for further applications of *B. japonicum*. Soil associations and textures for each environment are listed in Table 3-1, legal land descriptions and coordinates are listed in Table 3-2, and cropping histories are listed in Table 3-3.

Table 3-1. Soil Associations and Texture for all Trial Locations

Location	Soil Association[z]	Texture
Homewood	Sperling	very fine sandy loam to silty clay
Morris	Red River – Emerson Transition	clay – silty loam to silty clay loam
St. Norbert	Red River	clay

[z] Ehrlich et al., 1953

Table 3-2. Legal Land Descriptions and Coordinates for Each Environment

Environment	Legal Land Description	Coordinates	
		Latitude	Longitude
Homewood, 2006	SW 29-6-3W	49°30'17.87"N	97°50'20.28"W
Homewood, 2005	SE 30-6-3W	49°30'11.60"N	97°51'10.85"W
Homewood, 2004	SE 29-6-3W	49°30'12.07"N	97°50'2.14"W
Morris, 2006	NE 16-4-1E	49°18'14.16"N	97°23'45.77"W
Morris, 2005	NW 15-4-1E	49°18'29.96"N	97°23'23.91"W
Morris, 2004	NE 16-4-1E	49°18'25.33"N	97°23'43.46"W
St. Norbert, 2006	River Lot 48 & 49 Parish St. Norbert	49°43'2.54"N	97°8'3.37"W
St. Norbert, 2005	River Lot 64 & 65 Parish St. Norbert	49°43'57.86"N	97°8'37.52"W

Table 3-3. Cropping History for all Environments

Environment	2006	2005	2004	2003	2002
Homewood, 2006	Soybean	Oat	Soybean	Soybean	
Homewood, 2005	—	Soybean	Oat	Beans	Wheat
Homewood, 2004	—	—	Soybean	Wheat	Sunflower
Morris, 2006	Soybean	Wheat	Soybean	Wheat	
Morris, 2005	—	Soybean	Wheat	Soybean	Wheat
Morris, 2004	—	—	Soybean	Soybean	Canary Seed
St. Norbert, 2006	Soybean	Barley	Soybean	Barley	Soybean
St. Norbert, 2005	—	Soybean	Barley	Canola	Barley

Soils were tested for fertility at each site prior to seeding with the exception of the 2004 locations. The soil was sampled at each environment and analyzed for nitrogen, phosphorus, potassium (K), sulphur (S), % organic matter (OM) content (2006 only) and pH by Agvise Labs, Northwood, North Dakota, USA or Bodycote Testing Group, Winnipeg, Manitoba. Testing methods used by each soil test laboratory are available in Appendix A and test results are summarized in Table 3-4.

Table 3-4. Soil Fertility Test Results for All Environments

Environment	Depth cm	pH	OM %	N kg ha^{-1}	Olsen P ppm	K ppm	S kg ha^{-1}
Homewood, 2004	0-15 15-60	—	—	—	—	—	—
Homewood, 2005†	0-15	7.6	—	12	21	483	11
	15-60	7.7		64	10	338	27
Homewood, 2006†	0-15	7.6	6.0	55	18	535	17
	15-60	8.1	3.4	26	5	332	15
Morris, 2004	0-15 15-60	—	—	—	—	—	—
Morris, 2005†	0-15	8.0	—	12	11	385	13
	15-60	8.3		40	6	304	74
Morris, 2006†	0-15	8.1	4.7	24	15	435	49
	15-60	8.4	3.4	22	5	273	46
St. Norbert, 2005†	0-15	7.1	—	19	26	543	36
	15-60	7.7		34	8	414	108
St. Norbert, 2006‡	0-15	7.3	7.5	76	32	>600	38
	15-60	7.7	5.5	102	9	498	68

† Soil sample processing conducted by Agvise Labs, Northwood, North Dakota, USA
‡ Soil sample processing conducted by Bodycote Testing Group, Winnipeg, Manitoba
— data not available

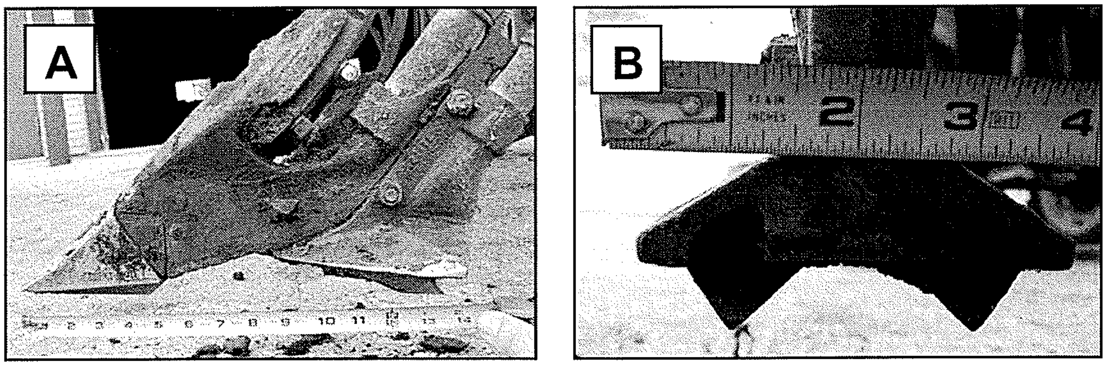
Trial Design

For this study, a Latin square split plot design was implemented with fertilizer rate (R) as the main plot and application practice (P) as the split plot. As such, each row and column of the Latin square contained six main plots. Main plots were split into three subplots for a total 18 plots per row or column and 108 plots per experimental trial. The order of the split plots within the main plots is identical in each column, whereas in each row, the order of split plots within the main plots has been randomized. Limitations of the seeding equipment prevented the randomization of the split plots between rows of a column. The seeder was not easily converted from one fertilizer application practice to another; therefore one complete practice had to be seeded before reconfiguring the seeder for the next practice (Appendix B).

Trial Establishment

In 2005 and 2006, an R-Tech JT-A10 "Jethro" seeder with double-shoot paired seed row openers was used. The paired seed row opener (Figure 3-1A) placed the seed in 10 rows approximately six centimeters wide with 20 cm seed row spacing. It also had the ability to apply fertilizer in a band approximately 2.5 cm below the paired seed row (Figure 3-1B). This type of opener is commonly used by producers in Manitoba for soybean production. All trials were seeded at a depth that would place the seed into moist soil, usually 2.5 to 3.5 cm below the soil surface.

The soybean cultivar OAC Prudence was used for all trials. Prudence is a common conventional soybean variety grown in Manitoba. The seed was not treated with a fungicide prior to seeding.



Double shoot paired seed row opener

Paired seed row openings

Figure 3-1. Double Shoot Paired Seed Row Openers on R-Tech J-10 Seeder

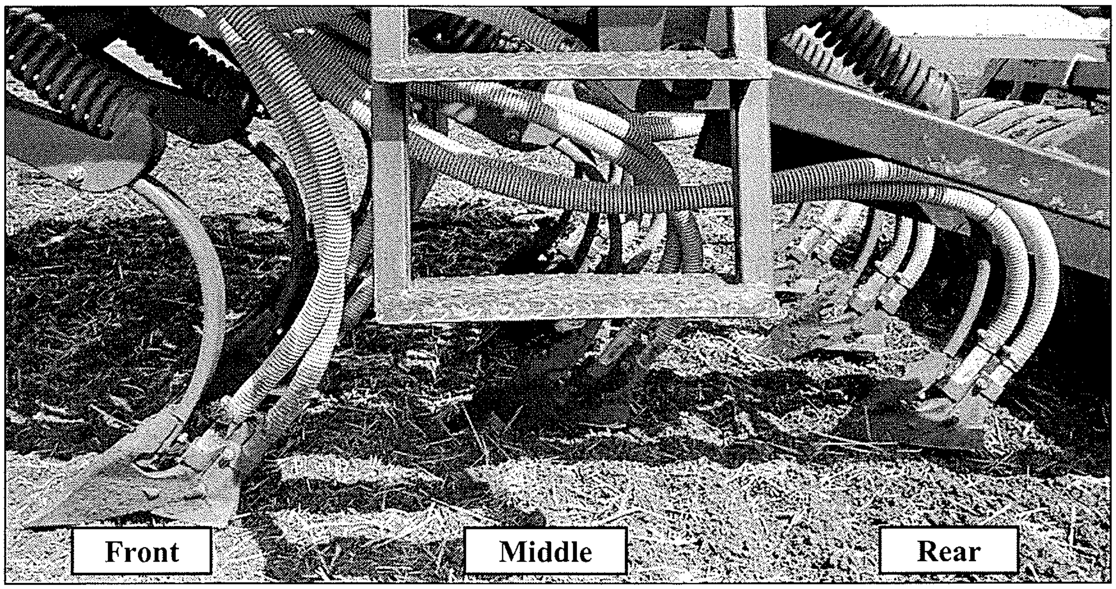


Figure 3-2. Front, Middle and Rear Rows of Openers on R-Tech J-10 Seeder

Treatments were calculated for a plot size of 2 m × 12 m. Midway through the growing season, the plot size was reduced to 2 m × 8 m by mowing 2 m off the front and back of each plot. The extra plot length ensured the seed, inoculant, and fertilizer were applied at the correct rates throughout the entire length of the plot as the seeder needed to travel approximately one metre before the seed/fertilizer/inoculant reached all openers. Seeder calibration settings are listed in Appendix C.

Nitrogen Study

Six rates of nitrogen fertilizer (0, 25, 50, 75, 100, 125 kg N ha^{-1}) were banded 2.5 cm below the seed row using each of three practices: Single-Non (single application of N fertilizer without *Rhizobium* inoculant), Single-Inoc (single application of N fertilizer with inoculant), or Split-Inoc (split application of N fertilizer with inoculant). Each practice had its own control where no treatment fertilizer was applied. Treatments are summarized in Table 3-5 (sample calculations can be found in Appendix D). Urea ($CO(NH_2)_2$; 46-0-0) was used for all treatment nitrogen with the exception of the Split-Inoc practice, which had the first 25 kg N ha^{-1} applied at seeding time as urea and the balance of the fertilizer applied as ammonium nitrate (AN; NH_4NO_3; 34-0-0) in mid-August (Table 3-5) at approximately the R3 (Table 2-1) development stage as a surface broadcast. AN was used for the broadcast application because it is less susceptible to volatilization losses compared to urea. In addition, at the onset of this study it was widely available and commonly used for mid-season broadcasting of N in Manitoba. By 2006, AN was no longer available to the average farmer, however it was still used in this experiment to maintain consistency.

Table 3-5. Soybean/Nitrogen Experiment Treatment Summary

	Practice (Split Plot)		**Rate (Main Plot)**			**Fertilizer**			
Tmt[†]	**Inoculant**	**Fertilizer Applications**	**Seeding**	**Split**	**Total**	**46-0-0 Rate**	**34-0-0 Rate**	**46-0-0 Applied**	**34-0-0 Applied**
			kg ha^{-1}			kg ha^{-1}		kg plot^{-1}[‡]	
1	No	Single	0	—	0	0	0	0	0
2	No	Single	25	—	25	54	0	0.130	0
3	No	Single	50	—	50	109	0	0.261	0
4	No	Single	75	—	75	163	0	0.391	0
5	No	Single	100	—	100	217	0	0.522	0
6	No	Single	125	—	125	272	0	0.652	0
7	Yes	Single	0	—	0	0	0	0	0
8	Yes	Single	25	—	25	54	0	0.130	0
9	Yes	Single	50	—	50	109	0	0.261	0
10	Yes	Single	75	—	75	163	0	0.391	0
11	Yes	Single	100	—	100	217	0	0.522	0
12	Yes	Single	125	—	125	272	0	0.652	0
13	Yes	Split	0	0	0	0	0	0	0
14	Yes	Split	25	0	25	54	0	0.130	0
15	Yes	Split	25	25	50	54	74	0.130	0.118
16	Yes	Split	25	50	75	54	147	0.130	0.235
17	Yes	Split	25	75	100	54	221	0.130	0.353
18	Yes	Split	25	100	125	54	294	0.130	0.471

[†] Tmt = Treatment

[‡] Plot = 24 m^2

Different rates of non-treatment fertilizer were applied in each year (Table 3-6) depending on soil test results. The seeding rate was lower than desired at all locations in 2005 as a result of improperly calibrating the seeder. This calibration misunderstanding resulted in approximately 25% less seed, inoculant and non-treatment fertilizer being applied in 2005 compared to 2006 (see Table 3-6).

Table 3-6. Seeding, Inoculant and Non-Treatment Fertilizer Application Rates for Soybean/Nitrogen Trials

Year	**Seeding Rate** *kg ha^{-1}*	 *100 Seed Weight (g)*	 *Seeds ha^{-1}*	**Inoculant (kg ha^{-1})**	**Non-Treatment Fertilizer N-P-K-S^z**
2004	unknown	unknown	475000	7.4	0-40-35-11.7
2005	70	16.1	414072	5.6	0-0-0-0
2006	115	19.6	557398	7.4	0-11-0-0

^{z} kg nutrient ha^{-1}

Phosphorus Study

Six rates of phosphorus fertilizer (0, 15, 30, 45, 60, and 75 kg P_2O_5 ha^{-1}; i.e., 0, 7, 13, 20, 26, and 33 kg P ha^{-1}) were applied at seeding using each of three practices: BBS (banded below the seed), SP (seed-placed), or SP+*P. bilaiae* (seed-placed with *Penicillium bilaiae* inoculated seed). Each practice had its own control where no fertilizer was applied. The complete treatment list is available in Table 3-7 and sample calculation can be found in Appendix D. Mono-ammonium phosphate was used as the phosphorus source for the treatments and since this fertilizer had a nitrogen component, treatment fertilizer was blended with urea to increase the N content of all treatments to the highest rate (approximately 16 kg N ha^{-1}). By doing this, any observed treatment effects should be a result of the phosphorus component in the fertilizer and not the nitrogen component. This is similar to what Rehm (1986) and Grant et al. (1999) did in their experiments to balance the N level of all treatments when they used MAP fertilizer as a P source. Vessey and Heisinger (2001) did not do this as they felt the amount of N applied was negligible (2.9 to 8.7 kg N ha^{-1}) and confirmed it with a lack of nodulation response to the added N.

Table 3-7. Soybean/Phosphorus Experiment Treatment Summary

	Practice (Split Plot)			**Fertilizer**			
Tmt	***P. bilaiae***	**Fertilizer Placement^z**	**P_2O_5 Rate (Main Plot)**	**11-52-0 Rate**	**46-0-0 Rate**	**11-52-0 Applied**	**46-0-0 Applied**
			— kg ha^{-1} —	kg ha^{-1}		kg plot$^{-1\,y}$	
1	No	SP	0	0	5.74	0	0.083
2	No	SP	15	28.85	4.59	0.069	0.066
3	No	SP	30	57.69	3.44	0.138	0.049
4	No	SP	45	86.54	2.29	0.208	0.033
5	No	SP	60	115.38	1.14	0.277	0.016
6	No	SP	75	144.23	0	0.346	0.000
7	No	BBS	0	0	5.74	0	0.083
8	No	BBS	15	28.85	4.59	0.069	0.066
9	No	BBS	30	57.69	3.44	0.138	0.049
10	No	BBS	45	86.54	2.29	0.208	0.033
11	No	BBS	60	115.38	1.14	0.277	0.016
12	No	BBS	75	144.23	0	0.346	0.000
13	Yes	SP	0	0	5.74	0	0.083
14	Yes	SP	15	28.85	4.59	0.069	0.066
15	Yes	SP	30	57.69	3.44	0.138	0.049
16	Yes	SP	45	86.54	2.29	0.208	0.033
17	Yes	SP	60	115.38	1.14	0.277	0.016
18	Yes	SP	75	144.23	0	0.346	0.000

Tmt = Treatment

^{z}Fertilizer Placement: SP = seed placed, BBS = banded below seed

^{y}Plot = 2 m × 12 m = 24 m^2

The phosphorus fertilizer was applied using one of two placements: banded below or placed with the seed. When the fertilizer was banded below the seed row, the seed and granular *Rhizobium* inoculant was directed through the rear shoot of the openers and the fertilizer was directed through the front shoot of the opener (Figure 3-1A). This placed the seed and inoculant together in a paired row at a depth of about 3.0 cm and the fertilizer in a band approximately 2.5 cm below the seed row for an overall depth of 5.5 cm. For the seed-placed fertilizer, in order to maximize the seed-fertilizer contact, the seed, granular inoculant and the fertilizer were all directed through the front shoot of the opener. This put the seed and fertilizer into a 2 cm wide row and with the 20 cm row spacing, this placement resulted in an SBU of 10% (Manitoba Agriculture, Food and Rural Initiatives, 2007). The depth of the openers was reduced by approximately 2.5 cm to account for the difference in depth from the rear shoot of the opener. This put the seed, inoculant and fertilizer at a similar soil depth as the other placement practice.

For the *Penicillium bilaiae* treatments, the commercial product Tagteam™ (Philom Bios) was used. Tagteam™ is a peat-based inoculant that includes both *P. bilaiae* as well as a *Rhizobium* inoculant. The peat-based formulation allowed for the seed to be treated in the field minimizing the time from application to seeding. The field pea version of this product was used in this experiment as it was only in 2007 that Tagteam™ was registered for use on soybeans in Western Canada. The pea rate that was used in this experiment was approximately 40% less than the soybean rate. The *Rhizobium* inoculant included as part of the field pea Tagteam™ is *Rhizobium leguminosarum* and is specific to field pea and will not inoculate soybeans. This allowed the experiment to isolate the effect of *P. bilaiae* without interference from the extra application of *Rhizobium* inoculant. For the remainder of this paper the name of the fungus, *Penicillium bilaiae* is used rather than the commercial product name TagTeam™. This was done as the purpose of the experiment was to evaluate the effect of *P. bilaiae* and not the product TagTeam™.

The *P. bilaiae* was applied to the seed in the field immediately before seeding. 15 kg of seed was placed in a mixing tub and approximately 250 ml of water was used to wet the seed so that the *P. bilaiae* peat inoculant would adhere to it, 24.3 g of *P. bilaiae* was then applied directly to the seed and mixed well to ensure even coating of all seed (sample calculation available in Appendix D). The SP+*P. bilaiae* practice was seeded last and the seeding equipment was rinsed with methyl hydrate in order to prevent contamination with the non-*P. bilaiae* treatments of the next trial.

Different rates of non-treatment fertilizer were applied in each year (Table 3-8) depending on soil test results. The seeding rate was lower than desired at all locations in 2005 as a result of improperly calibrating the seeder. This calibration misunderstanding resulted in approximately 25% less seed, inoculant and non-treatment fertilizer being applied in 2005 compared to 2006 (see Table 3-8).

Table 3-8. Seed, Inoculant and Non-Treatment Fertilizer Application Rates for Soybean/Phosphorus Trials

Year	**Seeding Rate**			**Inoculant ($kg\ ha^{-1}$)**	**Non-Tmt Fertilizer N-P-K-S**†	**Non-Tmt Fertilizer Placement**
	kg ha^{-1}	*100 Seed Wt. (g)*	*Seeds ha^{-1}*			
2004	N/D	N/D	475000	7.4	4.4-0-34-16.8	Broadcast
2005	70	16.1	414072	5.6	None	N/A
2006	115	19.6	557398	8	None	N/A

† kg nutrient ha^{-1}
N/D = not documented
N/A = not applicable

Sampling

Weather

Weather data was collected in the field using either a tipping bucket rain gauge with a Hobo data logger or from a Manitoba Agriculture, Food and Rural Initiatives weather station that was installed in the same field. When this was not possible, weather data was obtained from the nearest weather station available in Environment Canada's National Climate Data and Information Archive (Environment Canada, 2008). Table 3-9 lists the rainfall data sources used for each site year and Table 3-10 lists the sources of temperature data.

Table 3-9. Sources of Rainfall Data for All Environments

Environment	Time Period	Source of Rainfall Data
Homewood, 2004	Apr 1 – Oct 31	Environment Canada "Carman U of M CS"[z]
Homewood, 2005	Apr 1 – Oct 31	Environment Canada "Carman U of M CS"
Homewood, 2006	May 27 – Oct 31	In-field: Hobo data logger
	Apr 1 – May 26	Environment Canada "Carman U of M CS"
	30-year Average	Environment Canada "Elm Creek"[y]
Morris, 2004	Apr 1 – Oct 31	Environment Canada "Morris2"[x]
Morris, 2005	Apr 1 – Oct 31	Environment Canada "Morris2"
Morris, 2006	May 26 – Sep 25	In-field: Hobo data logger
	Apr 1 – May 25 & Sep 26 – Oct 31	Environment Canada "Morris2"
	30-year Average	Environment Canada "Morris2"
St. Norbert, 2005	Jun 22 – Oct 26	In-field: MAFRI[w] weather station
	Apr 1 – Jun 21 & Oct 27 – Oct 31	Environment Canada "Winnipeg Int'l Air"[v]
St. Norbert, 2006	June 9 – Oct 3	In-field: MAFRI weather station
	Apr 1 – Jun 8 & Oct 4 – Oct 31	Environment Canada "Winnipeg Int'l Air"
	30-year Average	Environment Canada "Winnipeg Int'l Air"

[z] Carman U of M CS weather station located approximately 10 km west

[y] Elm Creek 30-year average was used for Homewood trials (~20 km northwest)

[x] Morris2 weather station located approximately 3 km north

[w] Manitoba Agriculture, Food and Rural Initiatives

[v] Winnipeg Richardson International Airport located approximately 20 km north

Table 3-10. Sources of Temperature Data for All Environments

Environment	Time Period	Source of Temperature Data
Homewood, 2004	Apr 1 – Oct 31	Environment Canada "Carman U of M CS"[z]
Homewood, 2005	Apr 1 – Oct 31	Environment Canada "Carman U of M CS"[z]
Homewood, 2006	Apr 1 – Oct 31	Environment Canada "Carman U of M CS"[z]
	30-year Average	Environment Canada "Elm Creek"[y]
Morris, 2004	Apr 1 – Oct 31	Environment Canada "Emerson AUT"[x]
Morris, 2005	Apr 1 – Oct 31	Environment Canada "Emerson AUT"[x]
Morris, 2006	Apr 1 – Oct 31	Environment Canada "Emerson AUT"[x]
	30-year Average	Environment Canada "Emerson"[x]
St. Norbert, 2005	Apr 1 – Jun 22, Jul 26 – Aug 5 & Oct 27 – Oct 31	Environment Canada "Winnipeg Richardson Int'l A"[w]
	Jun 23 – Jul 25 & Aug 6 – Oct 25	In-field: MAFRI[v] weather station
St. Norbert, 2006	Apr 1 – Oct 31	Environment Canada "Winnipeg Richardson Int'l A"
	30-year Average	Environment Canada "Winnipeg Richardson Int'l A"

[z] Carman U of M CS weather station located approximately 10 km west
[y] Elm Creek 30-year average was used for Homewood trials (~20 km northwest)
[x] Emerson AUT and Emerson weather stations located approximately 40 km south
[w] Winnipeg Richardson International Airport located approximately 20 km north
[v] Manitoba Agriculture, Food and Rural Initiatives

Data Collection

At the V3 development stage a crop density measurement was conducted. The plants in one metre of four adjacent rows were counted. The area counted was equal to approximately 0.91 m^2. This was done twice in each plot and the mean of both measurements was used to determine the number of plants per hectare for each plot. In subsequent years (2005 and 2006), a 0.65 m diameter (0.33 m^2) circle was thrown randomly into each plot and the plants inside the circle were counted. This was done twice in each plot and the mean of the two measurements was used to calculate the crop density.

An evaluation of the root nodules was conducted between the R1 and R3 stages. Five whole plants were removed from each plot and placed in water to soften the soil.

The roots were then carefully washed and the nodules were visually assessed based on the key listed in Table 3-11. The same person performed the visual assessment of the roots for all environments. The scores for each of the four nodulation factors (quantity, size, colour, and distribution) were multiplied together to obtain an overall nodulation score. Similar nodule assessments were used by Corbin et al. (1977), Dean and Clark (1977), Rosas and Bliss (1984), Pulver et al. (1985), Herridge and Brockwell (1988), Hesterman and Isleib (1991), and Gleddie (1993). The nodulation assessment was conducted on the nitrogen trials in all three years and on the phosphorus trials in 2005 and 2006.

Table 3-11. Nodulation Assessment Scoring Key

Score	*Description*
Quantity(Q)	***Number of nodules found on root system (visual estimate)***
5	>50 nodules (lots of nodules everywhere)
4	35-50 nodules
3	Approx. 25-35 nodules (nodules everywhere)
2	10-25 nodules
1	<10 nodules
Size (S)	***Several nodules (not necessarily all) with diameter of***
5	Greater than 5mm
4	Approx. 4mm
3	Approx. 3mm
2	Approx. 2mm
1	Approx. 1mm
Colour (C)	***Internal colour of nodule (6-10 nodules plant^{-1})***
5	All pink
4	Some pink, some brown
3	All brown
2	Some brown, some green or white
1	All white or green
Distribution (D)	***Location of nodules on the root system***
3	Nodules found in crown region and on lateral roots
2	Nodules found in crown region only
1	Nodules found on lateral roots only
Overall Nodulation Score	$= Q \times S \times C \times D$

At the R3.5 stage, the remaining portions of the split application practice treatments were applied. In 2004, this was applied using a Hege cone fertilizer spreader that was pushed by hand through the plots. For 2005 and 2006, the split treatments were broadcast evenly by hand to the plots.

In 2005 and 2006, above ground crop dry matter was sampled at the R7 stage before substantial leaf drop had occurred. Above ground dry matter was measured by cutting the plants at approximately 2.5 cm above the soil surface from two 0.25 m^2 squares; one in the front half and one at the back half of the plot. The samples were placed into a drying oven one complete replicate at a time and dried to a constant weight at 65°C after which the samples were weighed.

In 2006, a crop height measurement was also collected at R7. One person held a two-metre long ruler in the centre of the plot while another determined the height of the crop. One height measurement was taken for each plot.

The soybeans were harvested at maturity. In 2004, a 60 cm brush mower was used to narrow the plots to the same width as the plot harvester. This was not done in subsequent years. A Hege plot harvester was used to harvest the soybeans. The seed was collected, placed into cloth bags and transported back to the lab where the seed bags were placed on a forced air drying bed for several weeks to bring all samples to the same moisture content. The seed was then cleaned by running through a Clipper Seed Cleaner (Model M2BC). All samples were then weighed and several sub-samples from random plots were placed in the drying oven and dried at 85 °C to a constant weight (approximately three days) to determine moisture content. The dry end weight was subtracted from the start weight to determine moisture content (Appendix D). Yields were then adjusted to 13% moisture content.

In 2004, 100 seed weight was determined by weighing a sample of 500 seeds from each plot and dividing by five. In 2005 and 2006, the weight of two samples of 200 seeds from each plot were averaged and divided by two to determine 100 seed weight. Other researchers have used a sample size of only 100 seeds to determine 100 seed weight (Starling et al., 1998; Dadson and Acquaah, 1984; Stone et al., 1985; and Freeborn et al., 2001; and Taylor et al., 2005). An electronic seed counter (Old Mill Company: Model 850-2) was used count the seed samples using the settings listed in Appendix C.

A 25 g sample was taken from each plot for quality analysis. Samples were analyzed for oil content using the NMR technique (ISO 5511, 1992). Next, the sample was ground in a coffee grinder and a 5 g sub-sample was used to determine nitrogen content using combustion nitrogen analysis (Williams et al. 1998) utilizing a LECO 528. For the phosphorus trials, another 25 g sub-sample was sent to Agvise Laboratories in North Dakota and analyzed for phosphorus content using either nitric perchloric acid digest test (2004) or nitric hydrogen peroxide digest test (2006) (Mills and Jones, 1996).

Data Analysis

The PROC MIXED procedure of SAS software (SAS Institute Inc., 2004) was used to analyze the data. Practice and rate were treated as fixed effects while environment, row, and column were treated as random effects. When the R × P interaction was not significant, contrasts were used to identify the shape (linear or quadratic) of the response to fertilizer rate averaged over all practices. When the R × P interaction was significant, contrasts were used to compare rate responses (linear or quadratic) of each practice to the others. When only the R effect was significant, all practices were combined and a single linear regression was calculated. When R and P were both significant, individual linear regressions were calculated for all practices. In an effort to eliminate the portion of the variation that was due to different environments, least squared means were converted to a percent of control basis before determining regression equations. When the R × P interaction was significant, linear regressions of all practices were superimposed onto a single interaction plot. Interaction plot regressions were not calculated on a percent of control basis. All linear regressions were computed using PROC REG (SAS Institute Inc., 2004). The pdmix800.sas macro (Saxton, 1998) was used to produce Tukey-Kramer least significant differences between practices if the P effect was found to be significant. Results were considered significant at $P \leq 0.05$ for all analyses.

4. RESULTS AND DISCUSSION

Growing Conditions

The first year of this study (2004) was characterized by an unusually high amount of rain falling during the month of May (Table 4-1) and cooler than normal temperatures (Table 4-2). Homewood and Morris received more than twice the 30-year average amount of rain for May. After the high initial spring rainfall amounts, Homewood received less than normal amounts for the remainder of the growing season which resulted in an overall rainfall amount that was only 6% higher than the 30-year average. Morris, on the other hand, received 39% more rainfall throughout the growing season in 2004 compared to the 30-year average; the majority falling in May, August and September. Temperatures were almost 2 °C cooler than normal for both locations.

In 2005, growing season rainfall at Homewood was similar to 2004 (Table 4-1). Morris, however, once again received substantially higher than normal rainfall amounts (+ 40%), but this time the rain fell mostly in June and resulted in an early summer flood of the Morris area. St. Norbert received normal rainfall amounts for 2005. Temperatures were approximately the same as the 30-year averages for all locations for the 2005 growing season (Table 4-2).

The 2006 growing season was a substantially dry for all locations (Table 4-1). Homewood, Morris and St. Norbert each received 30%, 48% and 62%, respectively, less rainfall than the 30-year averages for the growing season. Growing season temperatures were fairly close to the 30-year averages (Table 4-2).

Table 4-1. Growing Season Total Monthly Precipitation for All Environments

Environment	Growing Season Monthly Precipitation							
	Apr	*May*	*Jun*	*Jul*	*Aug*	*Sep*	*Oct*	*Total*
	mm							
30-Year Average (Elm Creek)^z	**33.4**	**51.5**	**81.0**	**71.1**	**70.0**	**56.7**	**30.5**	**394.2**
	Difference from 30-Year Average (mm)							
Homewood, 2004	-12.4	+65.1	-48.6	-20.9	+6.6	+30.3	+4.7	+24.8
Homewood, 2005	-14.4	+37.1	+58.8	+18.5	-46.4	-25.5	-3.7	+24.4
Homewood, 2006	+0.9	+13.6	-27.4	-56.5	-29.6	-3.3	-14.5	-116.8
30-Year Average (Morris)^y	**30.4**	**60.9**	**88.7**	**77.7**	**76.4**	**50.5**	**36.3**	**420.9**
	Difference from 30-Year Average (mm)							
Morris, 2004	+1.1	+96.9	-52.9	-27.8	+81.2	+51.3	n/a†	+149.3‡
Morris, 2005	-6	+32.9	+144.9	+1.5	-30	-5.7	+29.7	+167.3
Morris, 2006	-13.8	-23.7	-37.1	-64.7	-19.2	-17.3	-24.4	-200.2
30-Year Average (Winnipeg)^x	**31.9**	**58.0**	**89.5**	**70.6**	**75.1**	**51.9**	**31.0**	**408.0**
	Difference from 30-Year Average (mm)							
St. Norbert, 2005	+0.1	+8	+35.5	+14	-59.9	+6.7	-2	+2.4
St. Norbert, 2006	-15.4	-24.5	-78.1	-54.4	-33.3	-23.3	-22.5	-251.5

Data sources listed in Table 3-9.

† Data not available for this month.

‡ Difference is based on the April to September total of 384.6 mm.

[z] Elm Creek weather station located approximately 20 km northwest

[y] Morris2 weather station located approximately 3 km north

[x] Winnipeg Richardson International Airport located approximately 20 km north

Table 4-2. Growing Season Average Monthly Temperature for All Environments

Environment	**Average Monthly Temperature**							
	Apr	*May*	*Jun*	*Jul*	*Aug*	*Sep*	*Oct*	*Mean*
	°C							
30-Yr Avg. (Elm Creek)[z]	**4.2**	**12.5**	**16.9**	**19.4**	**18.2**	**12.3**	**5.5**	**12.7**
	Difference from 30-Year Average (°C)							
Homewood, 2004	0	-4.7	-2.3	-1.4	-4.2	+1.8	+0.5	-1.5
Homewood, 2005	+2.9	-2.3	+0.5	+0.3	-0.6	+2	+1.1	+0.6
Homewood, 2006	+4.3	-0.1	+1	+1	+0.6	+0.9	-1.7	+0.9
30-Yr Avg. (Emerson)[y]	**5.1**	**13.5**	**17.8**	**19.8**	**18.9**	**13.1**	**6.1**	**13.5**
	Difference from 30-Year Average (°C)							
Morris, 2004	-0.5	-4.7	-3.2	-1.6	-4.6	+2	+0.4	-1.8
Morris, 2005	+2.7	-2.9	+0.5	+1.1	-0.4	+2.3	+1.3	+0.6
Morris, 2006	-2.1	-0.4	+1.4	+1.5	+0.1	+0.3	-1.9	-0.2
30-Yr Avg. (Winnipeg)[x]	**4.0**	**12.0**	**17.0**	**19.5**	**18.5**	**12.3**	**5.3**	**12.7**
	Difference from 30-Year Average (°C)							
St. Norbert, 2005	+3.4	-2	+0.9	+0.9	-0.5	+2.2	+1.6	+0.9
St. Norbert, 2006	+5.4	+0	+1.2	+2	+1.3	+1.3	-1.8	+1.3

Data sources listed in Table 3-10.

[z] Elm Creek weather station located approximately 20 km northwest

[y] Emerson weather station located approximately 40 km south

[x] Winnipeg Richardson International Airport located approximately 20 km north

Data Collection – Nitrogen Fertilization of Soybean

The sampling of the nitrogen trials changed as more data parameters were added each year. Data collected from each environment, as well as the schedule of experiment events are summarized in Table 4-3 and Table 4-4, respectively. Appendix E contains the plot layouts, location maps and associated details. Of the eight environments, three were omitted from the data analysis due to either prohibitive weather conditions or human error; see Appendix G for more information.

Table 4-3. Data Sampled from Each Environment for Soybean-Nitrogen Experiment

Environment	Crop Density	Nodulation	Crop Biomass	Crop Height	Seed Yield	100 Seed Weight	Seed Protein	Seed Oil
Homewood, 2004	✓	✓	—	—	✓	✓	✓	✓
Morris, 2005	✓	✓	—	—	✓	✓	✓	✓
Homewood, 2006	✓	✓	✓	✓	✓	✓	✓	✓
Morris, 2006	✓	✓	✓	✓	✓	✓	✓	✓
St. Norbert, 2006	✓	✓	✓	✓	✓	✓	✓	✓

✓ = data collected
— = data not collected

Table 4-4. Schedule of Events for Soybean-Nitrogen Experiment

Environment	*Event*	*Growth Stage*[†]	*Date*
Homewood, 2004	Seeding	—	June 4
	Plant population	V3	July 16
	Herbicide application	Not documented	Not documented
	Nodule assessment	R3	August 12-13
	N split application	R3.5	August 19
	Harvest	R8	Not documented
Morris, 2005	Seeding	—	June 2
	Herbicide application	<V3	Not documented
	Flood	~V3	Late June – early July
	Plant population	V4	July 20
	Nodule assessment	R1	July 26
	N split application	R3.5	August 23
	Biomass	Not completed	—
	Harvest	R8	October 12
Homewood, 2006	Seeding	—	May 23
	Herbicide application	~V2	June 19
	Plant population	V3	June 27
	Nodule assessment	R2	July 21
	N split application	R4	August 3
	Biomass	R6.5	August 14
	Harvest	R8	September 27
Morris, 2006	Seeding	—	May 24
	Herbicide application	V2	June 12
	Plant population	V3	June 19
	Herbicide application	V6	July 4
	Nodule assessment	R2	July 20
	N split application	R5	August 3
	Biomass	R6.5	August 22
	Harvest	R8	September 25
St. Norbert, 2006	Seeding	—	May 22
	Herbicide application	<V3	June 14
	Plant population	V3	June 19
	Nodule assessment	R2	July 18
	N split application	R3.5	July 24
	Biomass	R7	August 15-16
	Harvest	R8	September 21

[†] Fehr and Caviness (1977) and Fehr et al. (1971)

Data Collection – Phosphorus Fertilization of Soybean

The sampling of the phosphorus trials changed as more data parameters were added each year. Data collected from each environment, as well as the schedule of experiment events are summarized in Table 4-5 and Table 4-6, respectively. Appendix F contains the plot layouts, location maps and associated details. Of the eight environments, three were omitted from the data analysis due to either prohibitive weather conditions or human error; see Appendix G for more information.

Table 4-5. Data Sampled from Each Environment for Soybean-Phosphorus Experiment

Environment	Crop Density	Nodulation	Crop Biomass	Crop Height	Seed Yield	100 Seed Weight	Seed Protein	Seed Oil	Seed Phosphorus
Homewood, 2004	✓	—	—	—	✓	✓	✓	✓	✓
Homewood, 2005	✓	✓	✓	—	✓	✓	✓	✓	—
Homewood, 2006	✓	✓	✓	✓	✓	✓	✓	✓	✓
Morris, 2006	✓	✓	✓	✓	✓	✓	✓	✓	✓
St. Norbert, 2006	✓	✓	✓	✓	✓	✓	✓	✓	✓

✓ = data collected
— = data not collected

Table 4-6. Schedule of Events for Soybean-Phosphorus Experiment

Environment	*Event*	*Growth Stage*[†]	*Date*
Homewood, 2004	Seeding	—	June 4
	Herbicide Application	Not documented	Not documented
	Plant population	V3	July 16
	Harvest	Not documented	Not documented
Homewood, 2005	Seeding	—	May 30
	Herbicide application	<V3	Not documented
	Plant population	V3	July 7
	Nodule assessment	R2	August 9
	Biomass	R7	September 8
	Harvest	R8	October 11
Homewood, 2006	Seeding	—	May 23
	Herbicide application	~V2	June 19
	Plant population	V3	June 27
	Nodule assessment	R4	July 27-28
	Biomass	R7	Aug 17
	Harvest	R8	Sept. 27
Morris, 2006	Seeding	—	May 24
	Herbicide application	~V2	June 12
	Plant population	V3	June 19
	Herbicide application	~V6	July 4
	Nodule assessment	R3	July 26
	Biomass	R7	Aug 23
	Harvest	R8	Sept. 25
St. Norbert, 2006	Seeding	—	May 22
	Herbicide application	<V3	June 14
	Plant population	V3	June 19
	Nodule assessment	R3	July 24
	Biomass	R7	Aug 16
	Harvest	R8	Sept. 21

[†] Fehr and Caviness (1977) and Fehr et al. (1971)

Results - Nitrogen Fertilization of Soybean

Emergence

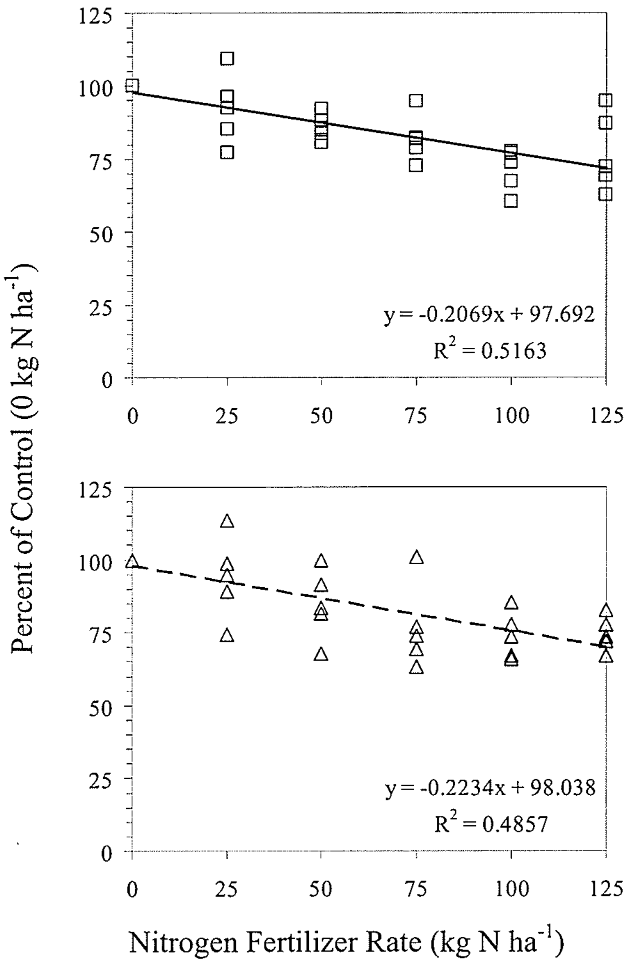
Both N fertilizer rate (R) and application practice (P) had significant effects on soybean emergence (Table 4-7). Linear contrasts revealed the Single-Inoc and Single-Non responses were significantly different from the Split-Inoc response. This is not unexpected as both Single-Inoc and Single-Non practices involved applying all of the fertilizer at seeding, which resulted in a negative effect on emergence, whereas the majority of the fertilizer in the Split-Inoc practice was not applied until R3.5. This, in turn, resulted in a significant R × P interaction. The negative responses of plant emergence to increasing fertilizer rate for both Single-Non and Single-Inoc are shown in Figure 4-1. A regression was not calculated for the Split-Inoc practice as only the 0 and 25 kg N ha^{-1} rates had been applied at the time of sampling. A significant ($Pr < 0.0001$) linear decrease in soybean density was observed for both the Single-Inoc and Single-Non practices (Table 4-7). For Single-Inoc and Single-Non, each 25 kg N ha^{-1} increment of fertilizer resulted in 5.2% and 5.6% fewer plants emerging, respectively. The R^2 values were moderate for both practices indicating the regressions only explained 52 and 49% of the variability in emergence populations for Single-Inoc and Single-Non practices, respectively.

The decrease in emergence observed in the current study was somewhat surprising given the placement of the nitrogen fertilizer and the clay soil texture. For all practices, the nitrogen fertilizer was banded 2.5 cm below the seed. Had the fertilizer been placed with the seed and/or if it had been a coarser-textured soil, it would likely have resulted in even greater seedling damage and reduced emergence.

Table 4-7. Type 3 Tests of Fixed Effects for Soybean-Nitrogen Experiment All Environments Combined

Effect	Emergence	Nodulation	Biomass	Height	Yield	100 Seed Weight	Protein	Oil
				Pr > F				
Rate (R)	**<0.0001**	**<0.0001**	0.1018	0.4629	0.8564	**0.0048**	**0.0138**	**0.0059**
Practice (P)	**<0.0001**	**<0.0001**	0.7930	0.4056	0.0532	0.2968	**0.0039**	**0.0088**
R x P	**0.0052**	**<0.0001**	0.1237	0.7515	**0.0003**	0.0612	0.1524	0.5759
R Linear	**<0.0001**	**<0.0001**	—	—	—	**0.0002**	**0.0006**	**0.0002**
R Quadratic	**0.0202**	**0.0188**	—	—	—	0.2041	0.2253	0.3191
				Linear Contrasts				
Single-Inoc vs. Single-Non	0.6668	0.8834	—	—	0.5946	—	—	—
Single-Inoc vs. Split-Inoc	**<0.0001**	**<0.0001**	—	—	**<0.0001**	—	—	—
Single-Non vs. Split-Inoc	**<0.0001**	**<0.0001**	—	—	**<0.0001**	—	—	—
				Quadratic Contrasts				
Single-Inoc vs. Single-Non	0.7738	0.3708	—	—	0.1884	—	—	—
Single-Inoc vs. Split-Inoc	0.9253	0.1414	—	—	0.5897	—	—	—
Single-Non vs. Split-Inoc	0.7000	**0.0168**	—	—	0.4245	—	—	—

Single-Non = Single application of N fertilizer, no inoculant applied
Single-Inoc = Single application of N fertilizer, inoculant applied
Split-Inoc = Split application of N fertilizer, inoculant applied



A. Soybean emergence response to nitrogen fertilizer applied at seeding with *Rhizobium* inoculant.
$n = 30$
$Pr \leq 0.0001$
Control mean = 412,465 plants ha^{-1}

B. Soybean emergence response to nitrogen fertilizer applied at seeding without *Rhizobium* inoculant.
$n = 30$
$Pr \leq 0.0001$
Control mean = 425,105 plants ha^{-1}

Figure 4-1. Inoculated and Non-Inoculated Soybean Emergence Responses to a Single Application of Nitrogen Fertilizer Expressed as a Percent of Control

Table 4-8. Effect of Nitrogen Fertilizer Application Practice on Soybean All Environments Combined

	Emergence	Nodulation	Biomass	Height	Yield	100 Seed Weight	Protein	Oil
Practice	plants ha^{-1}	score^{z}	kg ha^{-1}	cm	kg ha^{-1}	g 100 seed^{-1}	g kg^{-1}	g kg^{-1}
Single-Non	**361,616b**	**59.5b**	5,081a	83a	1,778a	18.54a	**410.0b**	**198.7a**
Single-Inoc	**350,190b**	**58.5b**	4,999a	82a	1,754a	18.54a	**410.5b**	**198.6a**
Split-Inoc	**398,165a**	**83.5a**	5,014a	82a	1,853a	18.73a	**416.4a**	**196.8b**
SE	58396	10.1	209	2.5	260	1.16	14.8	6.4

Means within column followed by a different letter are significantly different at $Pr \leq 0.05$.

[z]Nodulation score = visual rating of nodule size, quantity, colour and distribution

Single-Non = Single application of N fertilizer, no inoculant applied

Single-Inoc = Single application of N fertilizer, inoculant applied

Split-Inoc = Split application of N fertilizer, inoculant applied

Table 4-9. Overall Means for Soybean-Nitrogen Experiment for Each Environment

	Emergence	Nodulation	Biomass	Height	Yield	100 Seed Weight	Protein	Oil
Environment	plants ha^{-1}	Score	kg ha^{-1}	cm	kg ha^{-1}	g 100 seed^{-1}	g kg^{-1}	g kg^{-1}
Homewood, 2004	209,964	41.3	—	—	893	14.58	468.7	174.6
Homewood, 2006	475,627	52.3	4715	79	1,627	18.69	387.5	212.6
Morris, 2005	264,370	100.2	—	—	1,851	18.06	400.6	196.3
Morris, 2006	389,298	74.6	5381	87	2,300	21.15	412.6	201.7
St. Norbert, 2006	510,951	68.2	4992	81	2,299	20.54	392.1	205.1
Mean	370,042	67	5,029	82	1,794	19	412	198

— = data not collected for this parameter at this environment

The variability and subsequent intermediate R^2 values (near 0.5) can be at least partially explained by the sampling method. In 2004, each plant counted in the 0.91 m^2 sample area represented 11,000 plants ha^{-1}. In 2005 and 2006, each plant counted in the 0.33 m^2 circle represented 30,000 plants ha^{-1}. Therefore, the method used in 2004 was able to detect a difference in crop density of approximately 11,000 plants ha^{-1} whereas the method used in 2005 and 2006 could only detect a difference of 30,000 plants ha^{-1}. Therefore, any treatment differences of less than 30,000 plants ha^{-1} would not have been detected in 2005 and 2006.

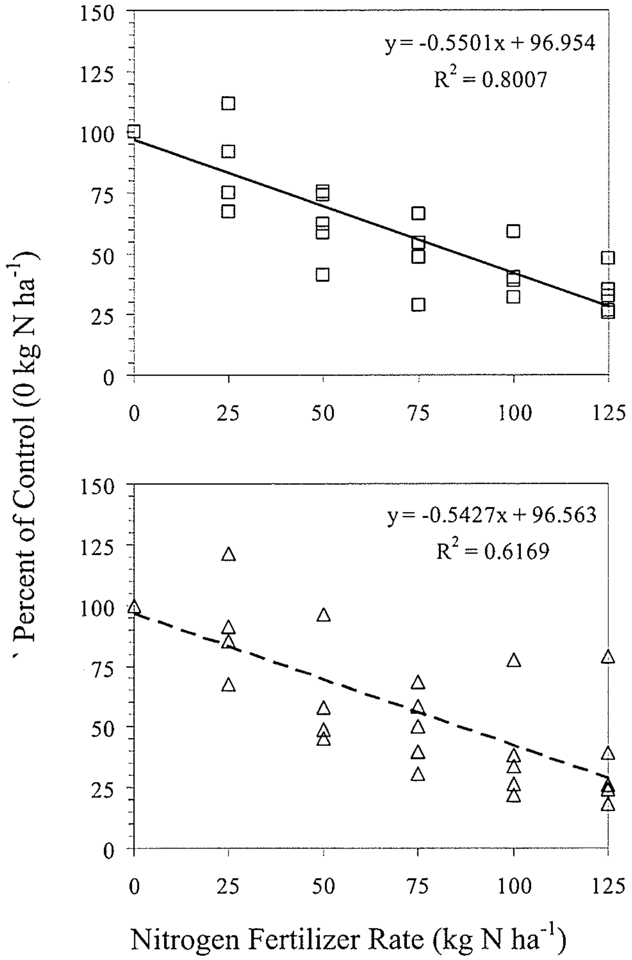
Populations ranged from 209,964 at Homewood in 2004 to 510,951 at St. Norbert in 2006 with an overall average of 370,042 plants ha^{-1} (Table 4-9). The recommended plant density for soybean in Manitoba is 400,000 plants ha^{-1} (Manitoba Agriculture, Food and Rural Initiatives, 2009). The wide range of plant populations is likely a result of the different seeding conditions (e.g., soil moisture, soil texture, soil temperature, seeding depth, rainfall, lack of seed treatment, etc.). Every effort was given to maintain a consistent seeding depth from year to year, but due to the differences in soil conditions, adjustments needed to be made to the seeding equipment and, therefore, slight changes in seeding depth likely occurred. Soil moisture is suspected to have been the largest source of error associated with seedling emergence in this study. Frequent and heavy rainfall early in May delayed seeding (St. Norbert, 2005) or forced seeding to be done in less than ideal conditions (Morris, 2004). Rain that fell after seeding left standing water in the trials (Morris, 2004; St. Norbert and Homewood, 2005; St. Norbert, 2006) or completely flooded the area (Morris, 2005). In addition, there were slight variations in seeding rate between years (Table 3-6).

In this study, applying relatively low rates of N fertilizer 2.5 cm below the seed row was enough to cause a negative effect on soybean emergence. This is in contrast to Welch et al. (1973) whose study indicated very high rates of N (1440 and 1800 kg N ha^{-1}) were needed to produce a response when the fertilizer was broadcast and disked into the soil.

Nodulation

Root nodulation was significantly affected by both nitrogen fertilizer rate as well as application practice (Table 4-7). The R × P interaction was also significant. The rate response showed a significant linear as well as quadratic trend and linear contrasts revealed the Split-Inoc practice was different from both of the single application practices (Single-Inoc and Single-Non). Quadratic contrasts revealed only the Single-Non practice was different from the Split-Inoc practice. Neither interaction is surprising as the fertilizer for the Split-Inoc practice was not applied until after the nodulation data was collected at the R1 stage and, as was the case with the emergence regressions, could therefore not influence nodulation. This negative effect of N on nodulation is similar to those reported by Koutroubas et al. (1998), Buttery et al. (1988), Ham et al. (1975), Semu and Hume (1979a), Starling et al. (1998), Hardarson et al. (1984), Taylor et al. (2005), Hesterman and Isleib (1991), Chen et al. (1992), Beard and Hoover (1971), La Favre and Eaglesham (1987), and Gibson and Harper (1985). Other researchers have also reported the nodulation response of soybean to nitrogen fertilizer followed a linear trend

(Koutroubas et al., 1998; Semu and Hume, 1979a; Beard and Hoover, 1971; and Chen et al., 1992).



A. Soybean nodulation response to nitrogen fertilizer applied at seeding with *Rhizobium* inoculant.
n = 30
$Pr \leq 0.0001$
Control mean = 91.5 score

B. Soybean nodulation response to nitrogen fertilizer applied at seeding without *Rhizobium* inoculant.
n = 30
$Pr \leq 0.0001$
Control mean = 93.3 score

Figure 4-2. Inoculated and Non-Inoculated Soybean Nodulation Responses to a Single Application of Nitrogen Fertilizer Expressed as a Percent of Control

Linear regressions were significant ($Pr \leq 0.0001$) for both Single-Inoc and Single-Non practices and nodulation was reduced by 13.8% and 13.6% for every 25 kg increment of fertilizer that was applied (Figure 4-2). The R^2 values were 80 and 62% for the Single-Inoc and Single-Non application practices, respectively, indicating a substantial portion of the variation in nodulation is explained by the regressions.

Inoculation had little effect on the nodulation of soybean in this study as can be seen in the regressions for both the inoculated and non-inoculated practices (Figure 4-2) as well as in the overall means for each practice (Table 4-8). Semu and Hume (1979a) reported inoculation did not affect nodule number or dry weight at locations where soybean had been grown previously. The slightly greater variability seen in the non-inoculated regression (Figure 4-2B) could be a result of strictly depending on naturalized *Rhizobium* to nodulate the plants. Aside from this difference, the lack of response to inoculation reveals the ability of *Rhizobium* applied in previous years to become established and thrive in high enough numbers to nodulate non-inoculated soybean plants. Nelson et al. (1978) reported similar findings in Indiana as did McAndrew and Brolley (2003) in a Manitoba study.

Soybean Biomass and Height

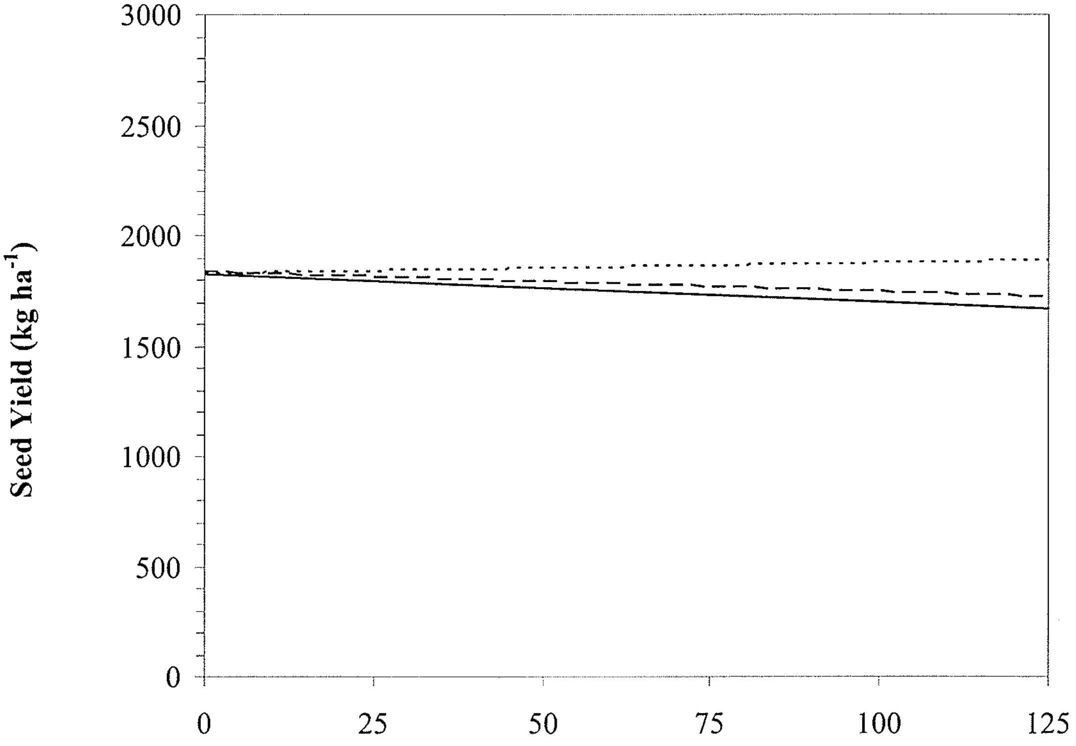
Neither nitrogen fertilizer rate nor application practice had a significant effect on soybean above ground dry matter or soybean plant height (Table 4-7). Other researchers have reported a similar lack of response of dry matter (Barker and Sawyer, 2005; Schmitt et al., 2001; and Buttery et al., 1998) as well as plant height (Taylor et al., 2005) to N fertilizer. But positive responses have been documented for biomass production (Dadson and Acquaah, 1984; Taylor et al., 2005; and Bhangoo and Albritton, 1976) and plant height (Ham et al. (1975), Starling et al. (1998) and Bharati et al. (1986).

The lack of response in this study suggests two implications: first, soybean is elastic, to a certain extent, in its ability to increase above ground biomass production to compensate for lower plant populations resulting from N fertilizer application. Similarly, Purcell and King (1996) hypothesized the lack of response to N fertilization in their study may have been due to a decrease in populations when the fertilizer was applied at the V6 stage. However, one might speculate that although the added nitrogen caused a reduction in emergence, it may also have provided the extra nitrogen required by the remaining plants to outgrow the thin plant stand. Second, that N fertilizer likely offsets the quantity of nitrogen that is fixed rather than supplements it. This coincides with the findings of Goss et al. (2002) who reported a decrease in the percent N derived from atmosphere with increasing rates of applied N fertilizer.

Seed Yield

Average yields for Risk Area 12 (Red River Valley) in Manitoba for 2004, 2005 and 2006 yields were 470, 1277 and 1546 kg ha^{-1}, respectively (Manitoba Agricultural Services Corporation, 2008). The average yields obtained in this study were all higher than the provincial averages for this area. This was likely a result of locating the experimental trials on the better parts of the fields. The rate of the nitrogen fertilizer application had no significant effect on soybean seed yield (Table 4-7). Application practice was significant at $Pr = 0.0532$ and the R x P interaction was strongly significant at $Pr = 0.0003$.

Linear contrasts revealed a significant interaction between the linear portions of the Split-Inoc curve and both of the Single-Inoc and Single-Non curves (Table 4-7). The linear interaction can be seen in Figure 4-3, however since Rate was not significant (Pr = 0.8564), the regression coefficients for the linear regressions for all three practices were very low. The significant interaction results from the lack of yield response of the Split practice, whereas there appears to be a slight negative yield response to the Single-Inoc and Single-Non practices.



——— **Single N Application, Inoculated**

y = -1.22x + 1826 $R^2 = 0.0086$ $Pr \leq 0.6260$ n = 30

— — — **Single N Application, Non-inoculated**

y = -0.95x + 1837 $R^2 = 0.0058$ $Pr \leq 0.6891$ n = 30

- - - - - **Split N Application, Inoculated**

y = 0.49x + 1824 $R^2 = 0.0018$ $Pr \leq 0.8257$ n = 30

Figure 4-3. Interaction of Seed Yield Responses to Nitrogen Fertilizer Applied Using Three Different Practices

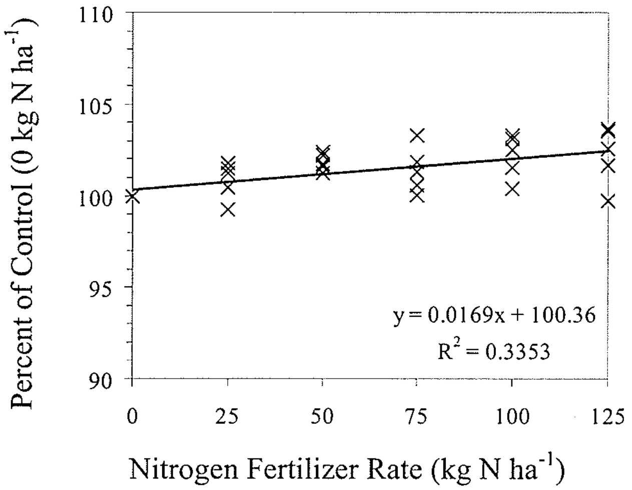
In this study, the split N fertilizer was applied by hand, and therefore no trampling of the crop occurred. In a commercial field of soybean, the split N fertilizer would be applied by large equipment, which would result in some trampling losses as the equipment moves across the field. These losses were not encountered in this study and therefore it is difficult to estimate how severe they might be. Nonetheless, this difference needs to be acknowledged.

Overall, nitrogen fertilizer, regardless of how it was applied, had little to no effect on soybean seed yield in this experiment. Other researchers have found similar results (Koutroubas et al., 1998; Buttery et al., 1988; Criswell et al., 1976; Schmitt et al., 2001; Heatherly et al., 2003; Beard and Hoover, 1971; and Freeborn et al., 2001). The lack of a yield response to N rate for the non-inoculated practice indicates that naturalized *Rhizobium* can be sufficient to provide the nitrogen requirements for yield. Previous research has shown that inadequately nodulated soybean tends to have a greater response to nitrogen fertilizer (Herridge and Brockwell, 1988; Johnson and Hume, 1972; Ham et al., 1975; Johnson et al., 1975; Goss et al., 2002).

When nitrogen fertilizer is applied to soybean, it tends to substitute for fixed nitrogen rather than supplement it. The lack of a yield response, but the presence of a negative nodulation response reveals that nitrogen was not limiting in this experiment and that as an increasing amount of N was applied, fewer nodules were required by the crop. This response hints at a reduction in N_2 fixation as decreases in nodulation have been reported to coincide with reductions in N_2 fixation (Semu and Hume, 1979a; Ham et al., 1975; Gan et al., 2004).

Seed Weight

Only N rate had a significant effect (*Pr* = 0.0048) on seed weight (Table 4-7). Since P was not significant, a linear regression was calculated for the response of seed weight to N fertilizer combined over all practices (Figure 4-4). This regression had a somewhat low regression coefficient ($R^2 = 0.34$) indicating the model explained only one third of the variation in seed weight. Each additional 25 kg ha^{-1} increment of N fertilizer resulted in a seed weight increase of 0.08 g per 100 seeds. Similarly, Taylor et al. (2005), Starling et al. (1998), Freeborn et al. (2001), and Stone et al. (1985) all reported no effect of N fertilizer on seed weight of soybean and Dadson and Acquaah, (1984) found an application of 160 kg N ha^{-1} was required to produce significantly larger seed size. Since seed weight increased slightly in response to N fertilizer but yield did not, it can be assumed that the quantity of seed produced decreased.

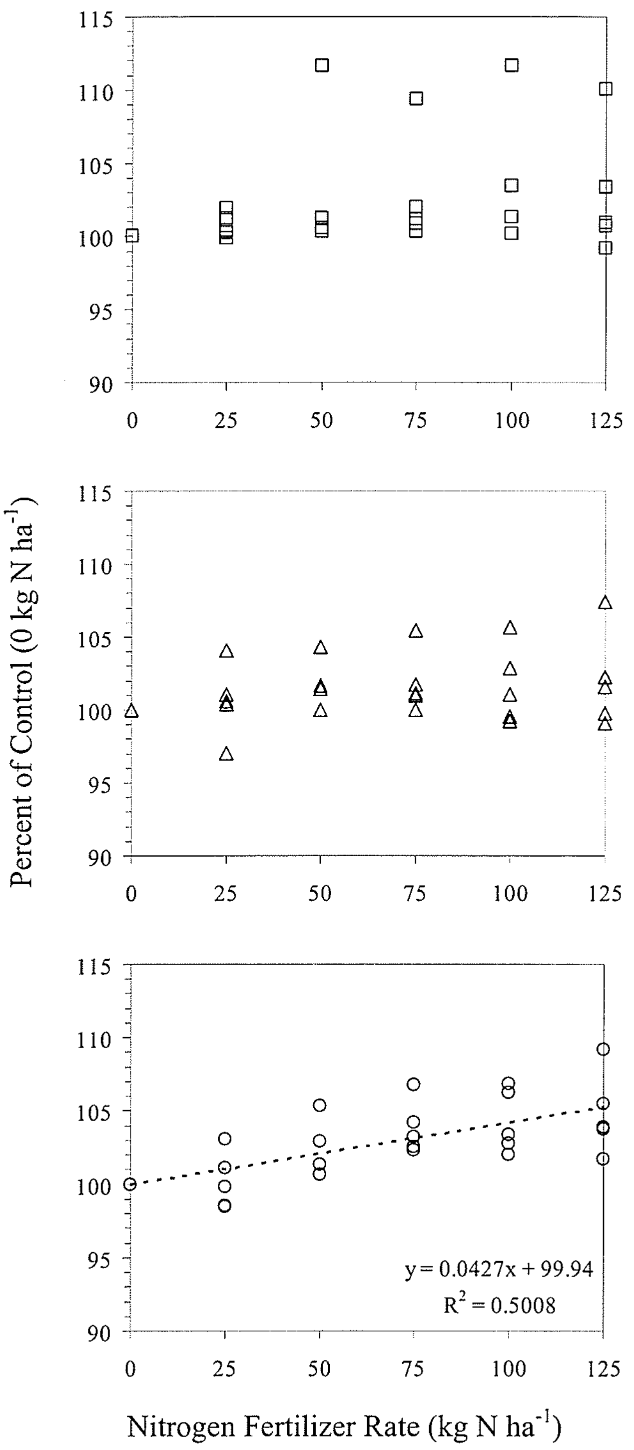


A. Soybean 100 seed weight response nitrogen fertilizer (all practices combined)
n = 30
Pr = 0.0008
Control mean = 18.35 g

Figure 4-4. Soybean 100 Seed Weight Response to Nitrogen Fertilizer Applied Using Three Different Application Practices Expressed as a Percent of Control

Seed Protein Content

Seed protein content was significantly affected by both nitrogen rate and application practice (Table 4-7). Contrasts revealed the rate response to be significantly linear (*Pr* = 0.0006) but not quadratic (*Pr* = 0.2253). The response to N fertilizer applied using the Split-Inoc practice had the only significant regression (Figure 4-5C). The Spring-Inoc and Spring-Non regressions were somewhat significant at *Pr* = 0.1024 and 0.1056, respectively.



A. Soybean seed protein response to nitrogen fertilizer applied at seeding with *Rhizobium* inoculant
$n = 30$
$Pr = 0.1024$
Control mean = 401.6 g kg^{-1}

B. Soybean seed protein response to nitrogen fertilizer applied at seeding without *Rhizobium* inoculant
$n = 30$
$Pr = 0.1056$
Control mean = 404.3 g kg^{-1}

C. Soybean seed protein response to nitrogen fertilizer applied as a split application (25 kg N ha^{-1} at seeding, balance at R6) with *Rhizobium* inoculant
$n = 30$
$Pr \leq 0.0001$
Control mean = 405.6 g kg^{-1}

Figure 4-5. Soybean Seed Protein Content Response to Nitrogen Fertilizer Applied Using Three Different Application Practices Expressed as a Percent of Control

When averaged over all rates, protein content of the Split-Inoc practice was significantly higher than Single-Inoc and Single-Non (Table 4-8). The lowest protein concentrations (387.5 g kg^{-1}) were obtained at Homewood in 2006 and the highest (468.7 g kg^{-1}) at Homewood in 2004 (Table 4-9).

There was no difference between the Single-Inoc and Single-Non practices. It appears that naturalized *B. japonicum* populations are sufficient to ensure an adequate N supply to the plant. This lack of response to inoculation is in contrast to the positive effects reported by Dadson and Acquaah (1984), Ciafardini and Barbieri (1987) and Muldoon et al. (1980).

Applying N fertilizer for the purpose of increasing seed protein content was, for the most part, unsuccessful in this experiment. These results are similar to those reported by Stone et al. (1985), Taylor et al. (2005), Nelson et al. (1978) but in contrast to the positive responses reported by Schmitt et al. (2001), Ham et al. (1975), Dadson and Acquaah (1984), Brevedan et al. (1978), Bhangoo and Albritton (1972) and the negative responses reported by Reese and Buss (1992), Purcell et al. (2004) and Ray et al. (2006a).

Protein levels responded significantly to N fertilizer only when it was applied as a split application. The highest rate of N fertilizer (125 kg N ha^{-1}) was required to produce a small (<22 g kg^{-1}) increase in protein content. Schmitt et al. (2001) reported an increase of 0.4 g kg^{-1} and Wesley et al. (1998) reported a protein increase of 10 g kg^{-1} (at only four out of eight sites) resulting from late-season applied N fertilizer. These results are in contrast to other studies that have reported no effect (Welch et al., 1973; Barker and Sawyer, 2005; Wood et al., 1993; Deibert et al., 1979). Overall, despite the significant increase in seed protein content resulting from a split application of N fertilizer, the cost of N fertilizer hardly warrants such a practice.

Seed Oil Content

Nitrogen fertilizer rate and application practice significantly influenced seed oil content (Table 4-7). Contrasts showed the rate response to be significantly linear. The regressions for Single-Inoc, Single-Non and Split-Inoc were significant at $Pr = 0.0508$, $Pr = 0.0195$ and $Pr = <0.0001$, respectively (Figure 4-6C). The regression coefficient for the Split-Inoc practice was 0.4342, indicating almost one third of the variability was explained by the regression. This negative effect of nitrogen fertilizer on soybean seed oil concentrations is similar to results reported by Dadson and Acquaah (1984), Ham et al. (1975) and Yin and Vyn (2005). Despite this statistically significant response to nitrogen fertilizer applied mid-season, the actual difference only decreased the seed oil content from 199.9 g kg^{-1} to 194.3 g kg^{-1} for the lowest (0 kg N ha^{-1}) to the highest (125 kg N ha^{-1}) rates, respectively; a difference of only 5.6 g kg^{-1}. Such a small decrease can hardly be described as biologically significant. Therefore, one could argue the results of the current study are not all that dissimilar to the results published by Welsey et al. (1998), Reese and Buss (1992), and Schmitt et al. (2001) who reported no effect of late-season applied nitrogen fertilizer on soybean seed oil content.

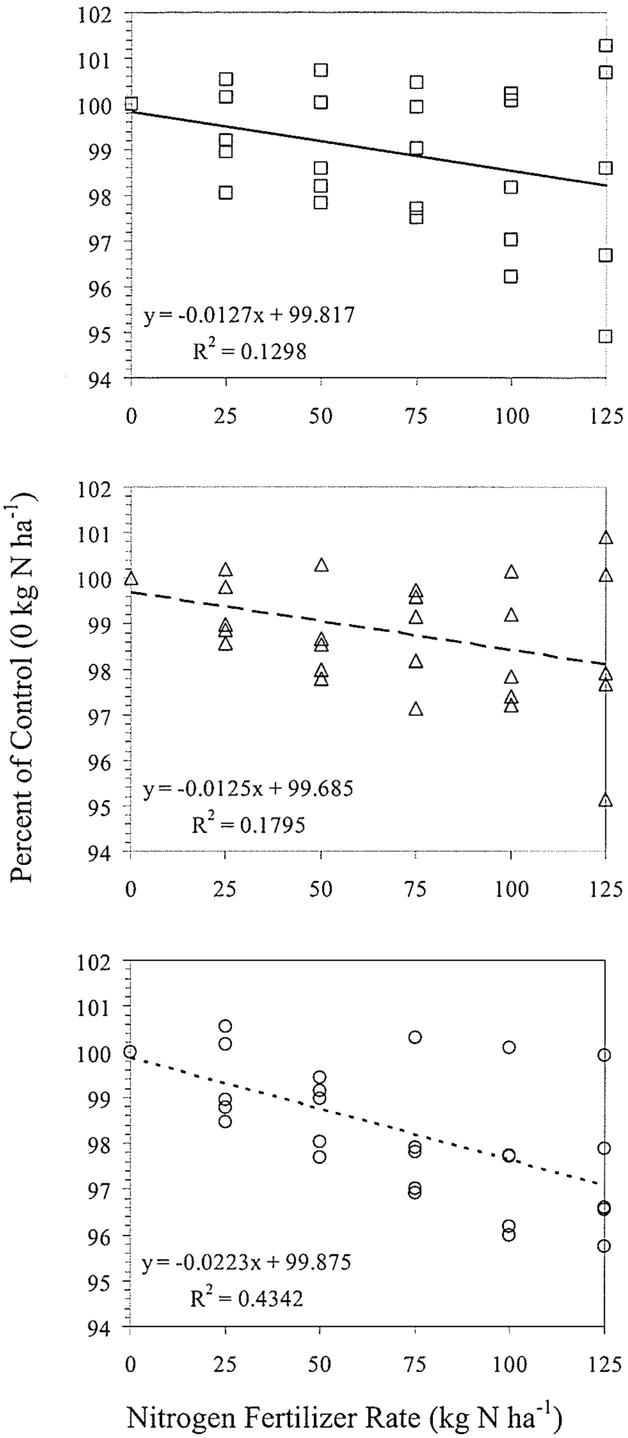
The regression coefficients for Spring-Inoc and Spring-Non were 0.1298 and 0.1795, respectively (Figure 4-6A and B). The low R^2 values for these practices are likely a result of the minimal differences in oil content between the highest and lowest rates of N fertilizer. This significant response to spring-applied fertilizer is in contrast to

Taylor et al. (2005), Schmitt et al. (2001), Reese and Buss (1992) and Starling et al. (1998) all of whom found no effect of nitrogen fertilizer on soybean seed oil content.

Some researchers have reported an overall increase in oil yield (kg oil ha^{-1}) due to an increase in seed yield, despite a reduction in seed oil concentrations (Ham et al., 1975) This was not the case in this study as nitrogen fertilizer had no effect on seed yield (Table 4-7).

When averaged over all rates, the Split-Inoc practice had significantly lower seed oil content than the Spring-Inoc and Spring-NonInoc practices (Table 4-8). However, the difference only amounted to 2.0 g kg^{-1} lower oil content. These results are not all that dissimilar to results published by Welsey et al. (1998) who reported a small increase (3.0 to 5.0 g kg^{-1}) in oil content as a result of late-season applied N fertilizer (but at only three out of eight locations), or Schmitt et al. (2001) who reported no effect of on seed oil content.

Inoculation with *B. japonicum* had no effect on seed oil content of soybean in this study. This is in contrast to Dadson and Acquaah (1984) and Muldoon et al. (1980) who reported a significant decrease in seed oil content as a result of inoculation.



A. Soybean seed oil content response to nitrogen fertilizer applied at seeding with *Rhizobium* inoculant
n = 30
Pr = 0.0508
Control mean = 200.5 g kg^{-1}

B. Seed oil content response to nitrogen fertilizer applied at seeding without *Rhizobium* inoculant
n = 30
Pr = 0.0195
Control mean = 200.9 g kg^{-1}

C. Seed oil content response to nitrogen fertilizer applied as a split application (25 kg N ha^{-1} at seeding, balance at R6) with *Rhizobium* inoculant
n = 30
Pr < 0.0001
Control mean = 199.9 g kg^{-1}

Figure 4-6. Soybean Seed Oil Content Response to Nitrogen Fertilizer Applied Using Three Different Application Practices Expressed as a Percent of Control

Summary

Soybean response to nitrogen fertilization varied, but overall, there was no obvious advantage to supplementing soybean with N fertilizer. Almost always when nitrogen rate had a significant effect on soybean it was a negative effect (emergence, nodulation, and seed oil content). Rarely was the effect positive (seed weight and seed protein content) and even then the response tended to be small and biologically insignificant. Of interest was the lack of a yield response despite the negative effect of N fertilizer on soybean emergence and nodulation. Such results hint at the crop's ability to compensate for thinner plant stands and its preference for soil nitrogen over fixed nitrogen. As well, there was no instance where inoculated soybean conferred an advantage over non-inoculated soybean. Applying N fertilizer as a split application did not affect seed yield but did result in small but significant changes in oil and protein content

Results - Phosphorus Fertilization of Soybean

Emergence

The rate of P fertilizer applied had a significant effect on emergence (Table 4-10) and application practice was significant at *Pr* = 0.0526. A linear regression revealed a significant positive influence of P fertilizer rate on emergence when it was applied using the SP+*P. bilaiae* practice (Figure 4-7). The regressions for the BBS and SP practices were not significant which resulted in a significant R × P interaction (Figure 4-8). Linear contrasts were only significant for BBS vs. SP+*P. bilaiae*; however, quadratic contrast were significant for both BBS vs. SP+*P. bilaiae* and SP vs. SP+*P. bilaiae* indicated an interaction at the quadratic portion of the responses. It is unclear why approximately 25% more plants emerged in the plots which received the highest P rate (75 kg P_2O_5 ha^{-1}) compared to the control (0 kg P_2O_5 ha^{-1}) in the SP+*P. bilaiae* practice (Figure 4-7).

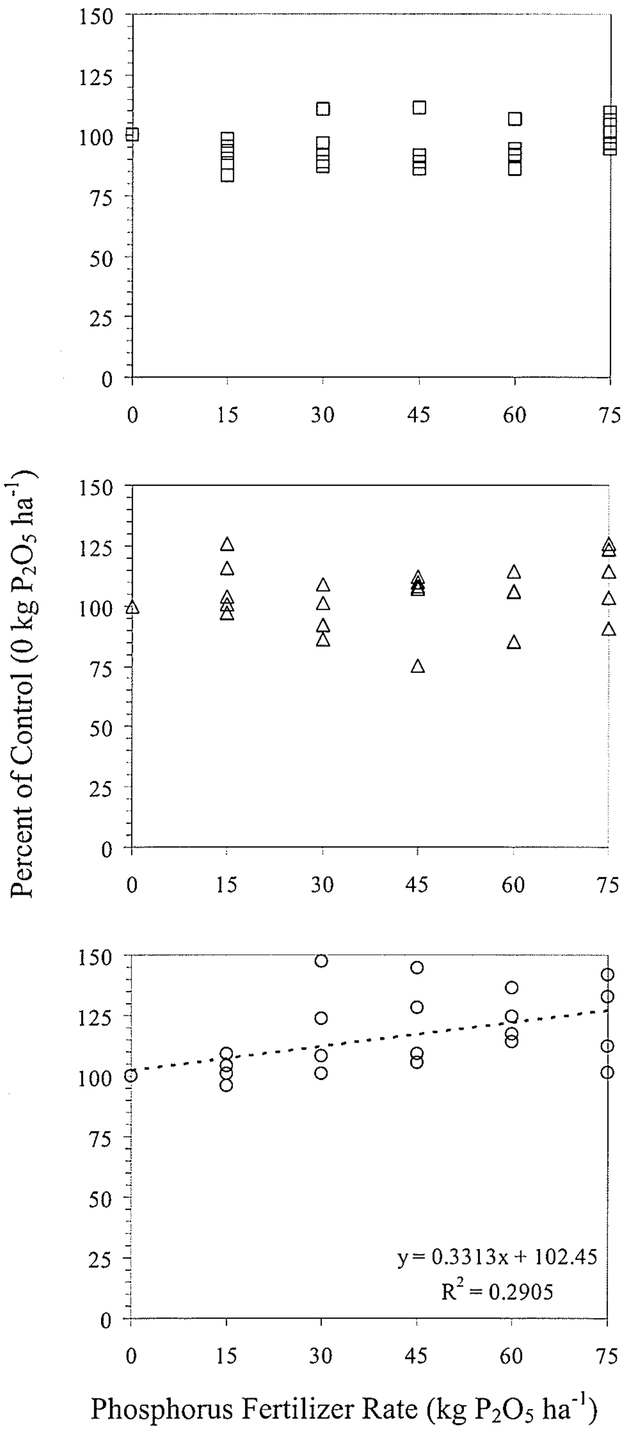
Table 4-10. Type 3 Tests of Fixed Effects for Soybean-Phosphorus Experiment All Environments Combined

Effect	Emergence	Nodulation	Biomass	Height	Yield	100 Seed Weight	Protein	Oil	Phosphorus
					Pr >F				
Rate (R)	**0.0377**	0.1443	0.3922	0.6713	0.6726	0.3710	0.2249	**0.0243**	**0.0016**
Practice (P)	0.0526	0.3853	0.3139	**0.0218**	0.6901	0.1396	0.9379	0.7219	0.3133
R x P	**0.0244**	0.2130	**0.0371**	0.2942	0.9216	0.7776	0.6752	0.2272	0.0843
R Linear	—	—	—	—	—	—	—	**0.0169**	**0.0004**
R Quadratic	—	—	—	—	—	—	—	0.3076	0.1963
					Linear Contrasts				
BBS vs. SP	0.3060	—	0.0718	—	—	—	—	—	—
BBS vs. SP+*P. bilaiae*	**0.0125**	—	0.1862	—	—	—	—	—	—
SP vs. SP+*P. bilaiae*	0.1359	—	0.7391	—	—	—	—	—	—
					Quadratic Contrasts				
BBS vs. SP	0.4975	—	0.4800	—	—	—	—	—	—
BBS vs. SP+*P. bilaiae*	**0.0059**	—	0.1122	—	—	—	—	—	—
SP vs. SP+*P. bilaiae*	**0.0373**	—	0.3466	—	—	—	—	—	—

BBS = Fertilizer banded below seed

SP = Fertilizer placed with seed

SP+*P. bilaiae* = Fertilizer placed with *Penicillium bilaiae*-inoculated seed

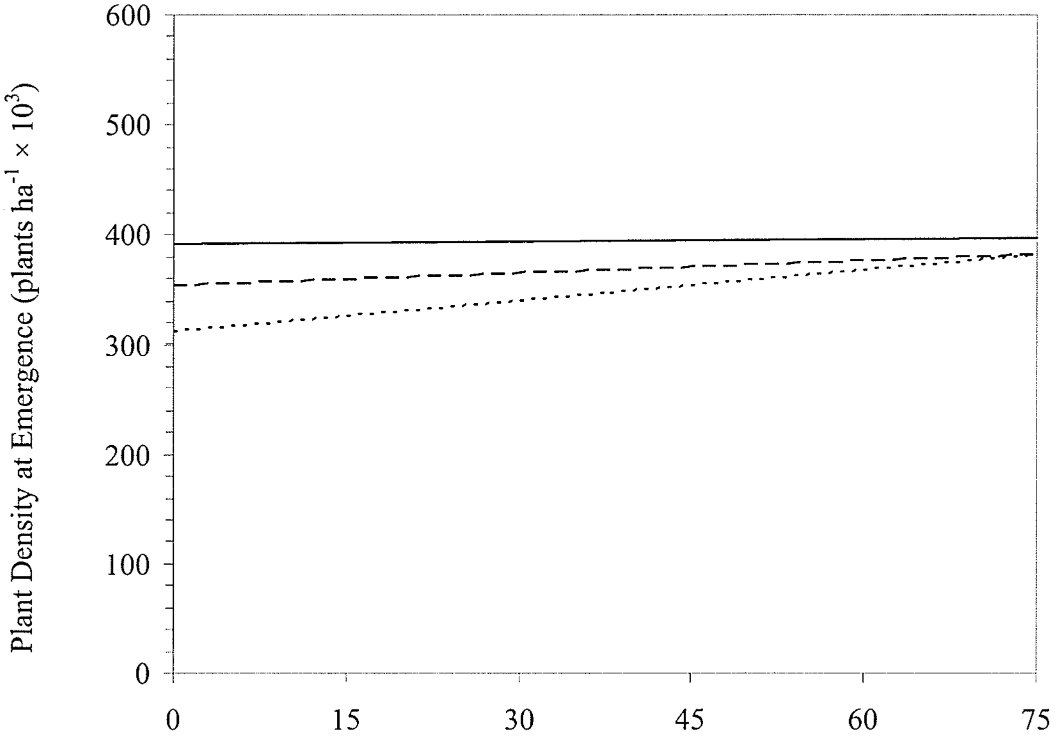


A. Soybean emergence response to phosphorus fertilizer banded below seed
n = 30
Pr = 0.6670
Control mean = 412,799 plants ha^{-1}

B. Soybean emergence response to seed-placed phosphorus fertilizer
n = 30
Pr = 0.2495
Control mean = 351,556 plants ha^{-1}

C. Soybean emergence response to seed-placed phosphorus fertilizer and inoculation with *P. bilaiae*
n = 24
Pr = 0.0066
Control mean = 304,169 plants ha^{-1}

Figure 4-7. Soybean Emergence Response to Phosphorus Fertilizer Applied Using Three Different Application Practices Expressed as a Percent of Control



——— **Banded below seed**

y = 81.34x + 391322 $R^2 = 0.0005$ $Pr \leq 0.9056$ n = 30

— — — **Seed placed**

y = 364.84x + 353275 $R^2 = 0.0116$ $Pr \leq 0.5717$ n = 30

- - - - - **Seed placed with *Penicillium bilaiae***

y = 940.24x + 311086 $R^2 = 0.2832$ $Pr \leq 0.0074$ n = 24

Figure 4-8. Interaction of Soybean Emergence Responses to Phosphorus Fertilizer Applied Using Three Different Practices

Table 4-11. Effect of Phosphorus Fertilizer Application Practice on Soybean All Environments Combined

	Emergence	Nodulation	Biomass	Height	Yield	100 Seed Weight	Protein	Oil	Phosphorus
Practice	plants ha^{-1}	score^{z}	kg ha^{-1}	cm	kg ha^{-1}	g 100 seed^{-1}	g kg^{-1}	g kg^{-1}	g kg^{-1}
BBS	**419494a**	91.64a	5433a	**81.9a**	2103a	18.80a	409.7a	197.7a	5.3a
SP	**395209ab**	99.78a	5502a	**80.4ab**	2127a	18.93a	409.2a	198.0a	5.2a
SP+*P. bilaiae*	**346411b**	101.02a	5715a	**79.4b**	2168a	18.83a	409.8a	197.7a	5.2a
SE	36514	6.98	414	1.8	301	1.30	12.7	7.1	0.5

Means within column followed by a different letter are significantly different at $Pr \leq 0.05$.

^{z}Nodulation score = visual rating of nodule size, quantity, colour and distribution

BBS = Fertilizer banded below seed

SP = Fertilizer placed with seed

SP+*P. bilaiae* = Fertilizer placed with *Penicillium bilaiae*-inoculated seed

Table 4-12. Overall Means for Soybean-Phosphorus Experiment for Each Environment

	Emergence	Nodulation	Biomass	Height	Yield	100 Seed Weight	Protein	Oil	Phosphorus
Environment	plants ha^{-1}	score	kg ha^{-1}	cm	kg ha^{-1}	g 100 seed^{-1}	g kg^{-1}	g kg^{-1}	g kg^{-1}
Homewood, 2004	293554	—	—	—	1101	13.87	452.2	177.9	6.9
Homewood, 2005	274417	128.6	6254	—	2351	19.65	418.7	184.5	—
Homewood, 2006	452583	86.4	4633	80.0	1890	19.09	377.1	211.8	4.6
Morris, 2006	413786	106.5	6114	83.9	2820	21.24	405.2	203.2	4.9
St. Norbert, 2006	387807	99.7	5120	77.9	2486	20.41	394.4	211.8	4.7
Mean	364429	105.0	5530	81.0	2130	18.85	409.5	197.8	5.3

— = data not collected for this parameter at this environment

Contrary to the negative effect of P fertilizer on soybean emergence reported by Bullen et al. (1980) and Clapp and Small (1970), P fertilizer rate did not have a negative effect on emergence in this study. Mono-ammonium phosphate was used as the P source and the N component of this fertilizer was equalized so as P rate increased the amount of N applied was the same, including in the control treatment (0 kg P ha^{-1}). It is not certain as to whether or not the N component had an effect as there was no treatment in this study that did not have any N fertilizer applied. However, the control for the BBS practice had no P fertilizer applied and the N fertilizer was banded below the seed. This treatment had an average emergence of 412,799 plants ha^{-1}, which was significantly more than the SP practice at 351,556 plants ha^{-1} (Table 4-10). Both BBS and SP practices received 16 kg N ha^{-1}. The only difference between these two practices was the placement of the fertilizer. Since P rate had no significant negative effect on emergence and banding the fertilizer below the seed had higher emergence than seed placed fertilizer, one could logically conclude the placement of the N fertilizer was responsible for the reduced plant populations.

Table 4-13. Mean Emergence for Control Treatment (0 kg P_2O_5 ha^{-1}) of All Practices in Soybean/Phosphorus Experiment Averaged over All Environments

	Mean Emergence for Control Treatment (0 kg P ha^{-1})
Practice	plants ha^{-1}
BBS	**412,799a**
SP	**351,556b**
SP+*P. bilaiae*	**279,335c**
SE	41,173

Means followed by a different letter are significantly different at $Pr \leq 0.05$.
BBS = Fertilizer banded below seed
SP = Fertilizer placed with seed
SP+*P. bilaiae* = Fertilizer placed with *Penicillium bilaiae* inoculated seed

In spite of the significant positive response to P, overall lower levels of crop emergence were observed in the SP+P. bilaiae practice (Table 4-11). It was concluded the reduced plant populations were due to the negative effect of the peat-based inoculant that was used in this study on seed flow in the seeder. In Ontario, Semu and Hume (1979) had a similar problem with a peat-based *Rhizobium* inoculant. The peat-based inoculant coating on the seeds apparently slowed the flow of seed in the seeder resulting in a decreased seeding rate. In the present study, this was confirmed by completing a seeder calibration test comparing seeding rates between bare seed and seed inoculated with *P. bilaiae*. Seed was run through and collected from the seeder, weighed and subjected to an F-Test which revealed the *P. bilaiae*-treated seed had a significantly (*Pr* = 0.0002) lower seeding rate (Table 4-14). Both the standard deviation and the CV of the *P. bilaiae*-treated seed were about four times higher than the plain seed. This suggests an increase in variability in seeding rate which resulted from the addition of a peat-based inoculant. However, the difference in seeding rate only amounted to an approximate 3% lower seeding rate for the *P. bilaiae*-treated seed. The difference measured in the field was approximately 12% fewer plants in the SP+*P. bilaiae* practice compared to the SP practice (Table 4-11). It is not apparent as to why the difference in plant populations measured in the field was so much higher than the difference in seeding rate.

Table 4-14. Seeding Rate F-Test Between Plain Seed and *P. bilaiae*-Treated Seed

	Plain Seed	**_P. bilaiae_-Treated Seed**
n	10	10
Mean (g)	380.8	370.8
Standard Deviation	2.6	10.7
Coefficient of Variation	0.67	2.88
***Pr* > F**	**0.0002**	

Nodulation

Neither phosphorus fertilizer rate nor application practice had a significant effect on soybean nodulation (Table 4-10). These results are similar to results published by Dadson and Acquaah (1984) but in contrast to the positive effect of P fertilizer on nodulation reported by Jones et al., (1977) and de Mooy and Pesek (1966).

Neither fertilizer placement nor inoculation with *P. bilaiae* had a significant effect on nodulation (Table 4-11). These results were similar to what Vessey and Heisinger (2001) reported for field pea and Rice et al. (1994) reported for alfalfa. The reduction in rhizosphere pH suspected by Downey and van Kessel (1990) as a result of *P. bilaiae* inoculation either did not occur or was not enough to negatively affect nodulation in the current study. Since they only measured N_2 fixation and speculated there may have been a decrease in nodulation, it is possible the acidification of the rhizosphere may not have a negative effect on nodulation. Dinitrogen fixation was not measured in this study and therefore it is possible that N_2 fixation was decreased without there being a corresponding visual reduction in nodulation.

Crop Biomass

Above ground crop dry matter production did not respond to phosphorus fertilizer rate, placement or inoculation with *P. bilaiae* (Table 4-10). This is in contrast to the reduction in dry matter yields reported by Bullen et al. (1980) due a reduction in plant numbers resulting from seed-placed triple super phosphate fertilizer (0-46-0).

P. bilaiae did not influence soybean above ground dry matter yield. Gleddie (1993) reported field pea had a significant positive response to *P. bilaiae* inoculation at locations that responded to P fertilizer. Dry matter production did not respond to P fertilizer in the current study. It is possible that *P. bilaiae* may have had an effect on biomass production, but only at the earlier stages of growth. Chambers and Yeomans (1990) and Goos et al. (1994) reported such findings for wheat and Gleddie et al. (1993) reported the same for canola.

Crop Height

Phosphorus fertilizer rate did not have a significant effect on soybean plant height (Table 4-10). This result is similar to Bharati et al. (1986) who reported on high P soils (69 – 73 ppm Bray-P) there was no effect of P fertilizer on plant height. They did, however, report a significant increase in lodging with the addition of P fertilizer. Although lodging was not directly measured in the current study, it was not observed in the trials despite being grown on soils with relatively high soil P test values (Table 3-4).

Application practice had a significant effect (*Pr* = 0.0218) on soybean height. Soybean plants were significantly shorter in the SP+*P. bilaiae* practice compared to the BBS practice (Table 4-25), but the difference only amounted to 2.5 cm. The overall plant height in the SP practice was not significantly different from either of the BBS or SP+P. bilaiae practices at the $Pr \leq 0.05$ level. The reason for this effect is unclear, but it is suspected variability associated with sampling is responsible. It could also have been a result of crop density. When the density is taken into consideration, it becomes apparent that higher plant populations tend to have taller plants. It may be that this is a result of intra-specific competition as the BBS practice had the greatest emergence as well as the tallest plants and the SP+*P. bilaiae* practice had the lowest emergence and the shortest plants.

Seed Yield and 100 Seed Weight

Neither P fertilizer rate, nor application practice had a significant effect on soybean seed yield or 100 seed weight in this study (Table 4-10). The lack of yield response to P fertilization in this study is similar to results published by Lauzon and Miller (1997) in Ontario. Average yields for Risk Area 12 (Red River Valley) in Manitoba for 2004, 2005 and 2006 yields were 470, 1277 and 1546 kg ha^{-1}, respectively (Manitoba Agricultural Services Corporation, 2008). In this study, the average yields for 2004, 2005 and 2006 were 1101, 2351, and 2399 kg ha^{-1}, respectively. The higher yields obtained in this study compared to the provincial averages are likely a result of the experimental trials being grown on the better areas of the fields.

Seed Protein Content

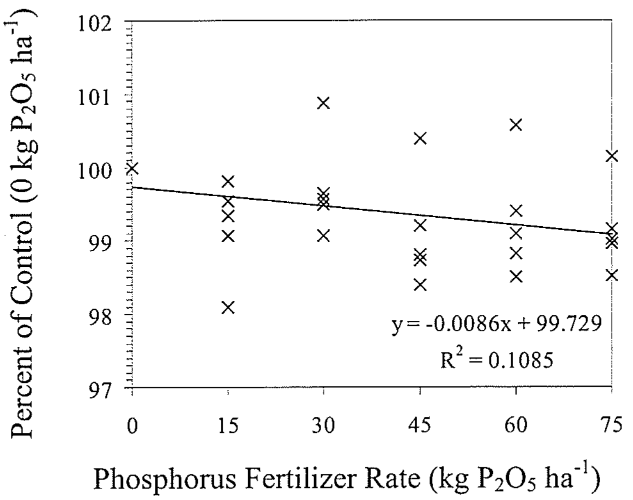
P fertilizer rate had no significant effect on soybean protein content (Table 4-10). These results are similar to those reported by Ham et al. (1973), Seguin and Zheng (2006), Bhangoo and Albritton (1972). This is in contrast to positive influences reported by Ramalingaswamy and Nabasimham (1977), Dadson and Acqaah (1984), Haq and Mallarino (2005), and Jones and Lutz (1971) or the negative influence reported by Haq and Mallarino (2005).

The placement of P fertilizer had no effect on soybean protein content in this study (Table 4-10). Similarly, Ham et al. (1973) and Haq and Mallarino (2005) found P fertilizer placement to have no effect on seed protein content.

P. bilaiae did not affect soybean seed protein concentrations. This finding is similar to what Kucey (1989a) reported for *P. bilaiae* inoculation of canola.

Seed Oil Content

Phosphate fertilizer rate had a significant effect on seed oil content (Table 4-10). Since practice as well as the R × P interaction were not significant, a single linear regression was used to describe the response of all practices combined (Figure 4-9). The regression coefficient was quite low ($R^2 = 0.1085$) and was significant at $Pr = 0.0755$. Increasing the P fertilizer rate from 0 kg P_2O_5 ha^{-1} to 75 kg P_2O_5 ha^{-1} decreased the oil content by 1.3 g kg^{-1}. Overall, there was little to no impact on seed oil concentrations as a result of P fertilization, regardless of placement or inoculation with P. bilaiae. This is in contrast to the significant P responses published by Ramalingaswamy and Nabasimham (1977), Dadson and Acqaah (1984), Haq and Mallarino (2005), Jones and Lutz (1971). The lack of response to P fertilizer placement is similar to research by Ham et al. (1973) and Haq and Mallarino (2005). Similarly, Kucey (1989a) found the addition of P fertilizers and *P. bilaiae* inoculant did not affect seed oil content of canola.

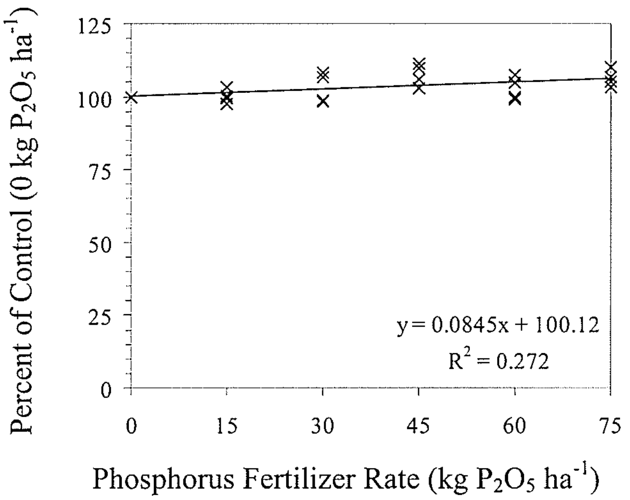


Soybean seed oil content response to phosphorus fertilizer (all practices combined)
n = 30
$Pr \leq 0.0755$
Control mean = 199.0 g kg^{-1}

Figure 4-9. Soybean Seed Oil Content Response to Phosphorus Fertilizer (All Practices Combined) Expressed as a Percent of Control

Seed Phosphorus Content

Phosphorus fertilizer rate had a significant effect on seed phosphorus content and the response was significantly linear (Table 4-10). A linear regression on the seed phosphorus content response (on a percent of control basis) was significant at $Pr = 0.009$ and had a regression coefficient of 0.272 (Figure 4-10). According to the regression equation, each 15 kg ha^{-1} increment of P_2O_5 translated into 1.27% higher seed phosphorus content compared to the control. With a control mean of 5.1 g kg^{-1} each 15 kg P_2O_5 ha^{-1} increased the P content by 0.065 g kg^{-1}. This is similar to results published by Aulakh et al. (2003) who also reported a positive effect of P fertilizer on the phosphorus content of soybean seed. However, they reported a significant seed phosphorus content increase of 1.9 to 4.6 g kg^{-1} from the application of rates up to 43.2 kg P ha^{-1} (100 kg P_2O_5 ha^{-1}). In contrast to the Aulakh et al. (2003) study, which was grown on a low P testing soil (5 ppm Olsen-P), the current study was conducted on medium to high P testing soils, ranging from 15 to 32 ppm (Table 3-4). This may partially explain the difference in the magnitude of response between the two studies. Contrary to the significant increase in seed P content of *P. bilaiae* inoculated canola reported by Kucey (1989a), *P. bilaiae* had no significant effect on seed phosphorus concentrations of soybean in this study.



Soybean seed phosphorus content response to phosphorus fertilizer (all practices combined)
n = 24
Pr = 0.009
Control mean = 5.1 g kg^{-1}

Figure 4-10. Soybean Seed Phosphorus Content Response to Phosphorus Fertilizer (All Practices Combined) Expressed as a Percent of Control

Summary

Phosphorus fertilizer had almost no effect on soybean production in this study. Emergence was not affected by the P component of the mono-ammonium phosphate fertilizer regardless of whether it was seed-placed or banded below the seed. Phosphorus fertilization of soybean in the high P-testing soils of the Red River valley of Manitoba did not improve yields or seed quality. *Penicillium bilaiae* inoculation did not offer any advantage over the non-inoculated treatments; likely a result of the medium to high P testing soils used in this study. This is in contrast to provincial recommendations which suggest, in high P soils, *P. bilaiae* may be substituted for P fertilizer (Manitoba Agriculture, Food and Rural Initiatives, 2007).

5. CONCLUSION

First of all, the soybean/nitrogen study demonstrated that biological dinitrogen fixation is more than adequate for supplying the nitrogen requirements of soybean grown in the Red River valley of Manitoba. Nitrogen fertilizer had an inhibitory effect on soybean nodulation, which may also indicate a reduction in N_2 fixation and therefore, given the high cost of nitrogen fertilizers in western Canada and the risk of crop injury, the application of N fertilizer to soybean should be avoided. Second, since inoculating soybean did not offer an advantage over not inoculating, expensive granular inoculants may not be necessary on land that has grown well-nodulated soybean crops in the past. Further research may be needed to confirm whether seed-applied inoculant is needed after a well-nodulated soybean crop had been grown in a previous season, as well as how many years may pass between applications of *Rhizobium* before a decrease in nodulation occurs. Results from this study indicate that naturalized *Rhizobium* may result in a greater variability in nodulation, but this variability does not appear to be detrimental to yield. Third, the timing of nitrogen fertilizer was not an effective strategy to supplement soybean with extra nitrogen. With ammonium nitrate no longer available as an agricultural fertilizer, the use of urea for the same purpose would be even less effective due to increased risk of volatilization losses. Fourth, if soybean is grown on a clean (relatively weed-free) field, it may be possible to reduce seeding rates since lower plant populations (300,000 plants ha^{-1}) did not yield significantly less than the recommended population level (400,000 plants ha^{-1}). At the very least, farmers may find they do not have to reseed a poorly germinated crop as soybean appears to have the ability to compensate for reduced plant stands.

Overall, phosphorus fertilization of soybean did not improve soybean seed yield or quality in the medium to high P soils of this study. The phosphorus component of mono-ammonium phosphate fertilizer did not appear to have a negative effect on soybean emergence in the heavy clay soils of this study. The relatively low quantity (approximately 16 kg N ha^{-1}) of nitrogen fertilizer that was applied in the seed row was sufficient to cause a reduction in plant emergence. Further research may be required to identify at what rate of N (as 11-52-0) applied with the seed is safe. Inoculation of soybean with *Penicillium bilaiae* had almost no significant effect on soybean production. This is likely due to the high soil test P levels. Further testing may be required on low P soils in Manitoba.

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APPENDIX A – SOIL TEST METHODS

Table A-1. Soil Test Methods for Agvise Labs and Bodycote Testing Group

	Agvise Labs[z]	**Bodycote Testing Group**[y]
	Testing Method	
N	Cadmium Reduction (Gelderman and Beegle, 1998)	Modified Kelowna Test (Ashworth and Mrazek, 1995) Continuous Flow Colorimetry (Carter, 1993)
P	Sodium Bicarbonate (Olsen) (Frank et al., 1998)	Modified Kelowna Test (Ashworth and Mrazek, 1995) Continuous Flow Colorimetry (American Public Health Association et al., 2005)
K	Ammonium Acetate (Warncke and Brown, 1998)	Modified Kelowna Test (Ashworth and Mrazek, 1995) Continuous Flow Colorimetry (Dieken et al., 1996)
S	KCL, Barium Chloride Turbidimetric (Combs et al., 1998)	Extractable SO_4 by 0.1 M $CaCl_2$ (McKeague, 1978) Inductively Coupled Plasma (American Public Health Association et al., 2005)
% OM	Loss on Ignition (Combs and Nathan, 1998)	Loss on Ignition (McKeague, 1978)
pH	1:1 Ratio, Soil:Water (Watson and Brown, 1998)	1:2 Ratio, Soil:Water (McKeague, 1978)

[z] Northwood, North Dakota, USA.
[y] Winnipeg, Manitoba, Canada

APPENDIX B – SEEDING METHOD

Figure D-1 shows a hypothetical seeding pattern similar to what was used in this experiment. The seeder starts at the lower left corner of the trial and seeds the first practice in six passes. After which, the seeder is reconfigured and the second practice is seeded which is then followed by the third practice. The entire trial is seeded with a total of 18 seeder passes.

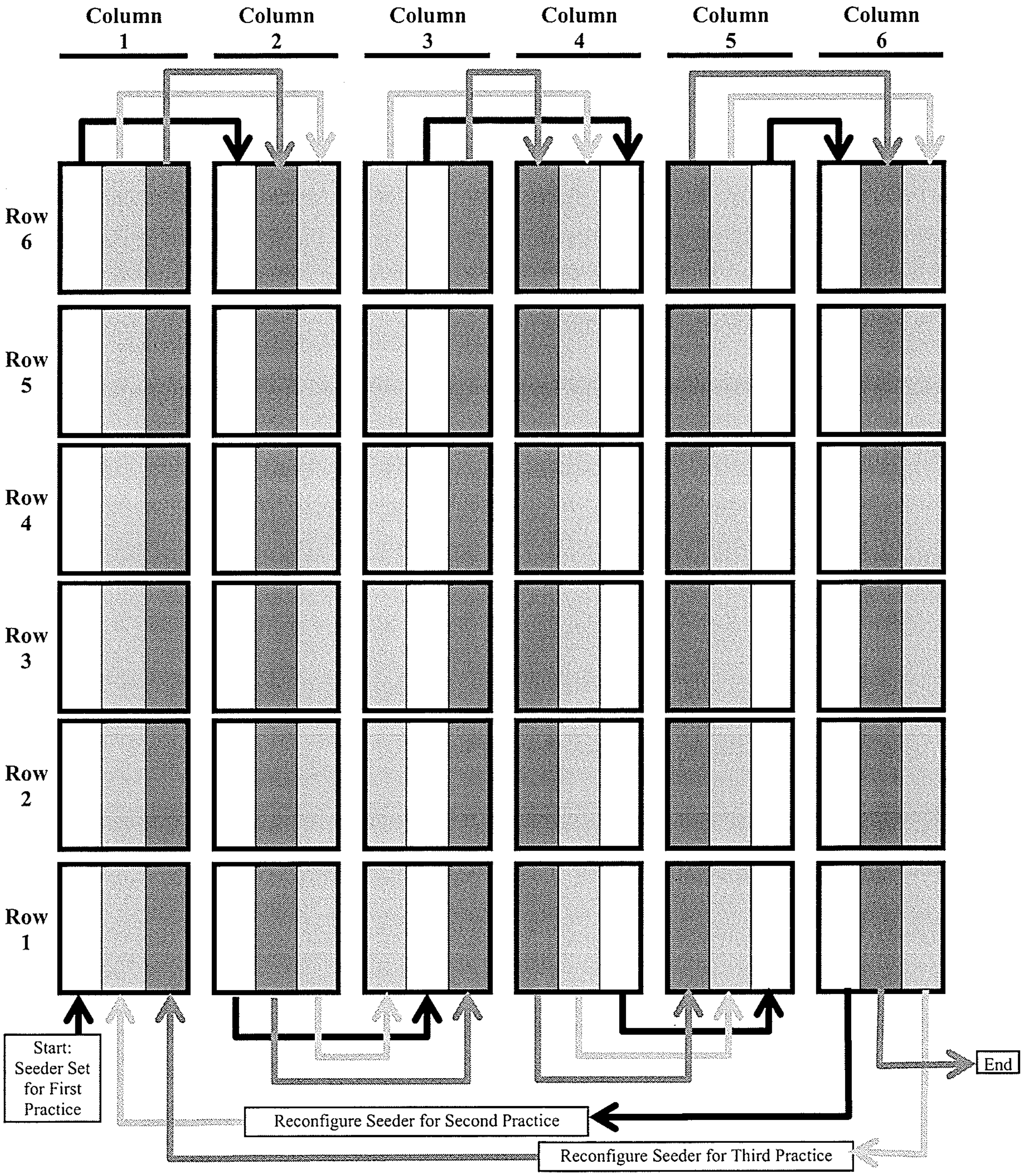


Figure D-1. Seeding Pattern for Latin Square Split Plot Demonstrating How Split Plots were Randomized and Seeded Within Each Main Plot

APPENDIX C – SEEDER CALIBRATION SETTINGS

Table C-1. Hoedrill Seeder Calibration Settings Used in 2004 Nitrogen Trials

Environment	— Box 1 —			— Box 2 —			— Box 3 —			—— S ——			— Cone —
	0-0-51-17			0-45-0			Seed			Inoculant			46-0-0
Homewood	A3	12	34	B4	19	27	A7	27	19	B1	19	27	Treatment
Morris	A3	12	34	B4	19	27	A7	27	19	B1	19	27	Treatment

Table C-2. R-Tech JT-A10 "Jethro" Seeder Calibration Settings Used in 2005 and 2006 Nitrogen Trials

Environment	Cone 0-max	Rear Bin —— Seed ——			Middle Bin — Inoculant —			Front Bin — Fertilizer —		
		0-max	*Cups*	*Flap*	*0-max*	*Cups*	*Flap*	*0-max*	*Cups*	*Flap*
Homewood, 2005	34.6	19	100%	oats	5	75%	min	—	—	—
St. Norbert, 2005	36.4	19	100%	oats	5	75%	min	—	—	—
Morris, 2005	35.1	19	100%	oats	5	75%	min	—	—	—
All locations, 2006	38.5	26	100%	3	6.5	75%	2	9	75%	2

Table C-3. Hoedrill Seeder Settings Used in 2004 Phosphorus Trials

Year	Site	— Box 1 —			— Box 2 —	— Box 3 —			— Box 4 —			- Cone -
		0-0-51-17			21-0-0-24	Seed			Inoculant			11-52-0
2004	Homewood	A3	12	34	N/D	A7	27	19	B1	19	27	N/A
2004	Morris	A3	12	34		A7	27	19	B1	19	27	

N/D: Not documented; N/A: Not applicable

Table C-4. Settings for R-Tech JT-A10 "Jethro" Seeder Used in 2005 and 2006 Phosphorus Trials

Year	Site	Cone 0-max	Rear Bin —— Seed ——			Middle Bin — Inoculant —			Front Bin — Fertilizer —		
			0-max	*Cups*	*Flap*	*0-max*	*Cups*	*Flap*	*0-max*	*Cups*	*Flap*
2005	Homewood	34.6	19	100%	oats	5	75%	min	Not used		
2005	St. Norbert	46.0	19	100%	oats	5	75%	min			
2006	Homewood	38.5	26	100%	3	6.5	75%	2			
2006	St. Norbert	38.5	26	100%	3	6.5	75%	2			
2006	Morris	38.5	26	100%	3	6.5	75%	2			

Table C-5. Settings for Electronic Seed Counter Model 850-2 (Old Mill Company)

Calibration Parameter	*Switch Setting*	*Dial Setting*
Auto Reset	Off	9
Feed Slow at	6	—
Function	Lot Size	—
Length Rejection	0	—
Sensitivity	Low	2.5

APPENDIX D – SAMPLE CALCULATIONS

Equation B-1. Plot Area

$$2\text{ m (W)} \times 12\text{ m (L)} = \frac{24\text{ m}^2}{\text{Plot}} \times \frac{1\text{ ha}}{10\,000\text{ m}^2} = \frac{0.0024\text{ ha}}{\text{plot}}$$

Equation B-2. Nitrogen Fertilizer Rate per Plot

A) Example fertilizer rate: 25 kg N ha^{-1} applied as urea (46-0-0)

$$\frac{25\text{ kg N}}{\text{ha}} \times \frac{1\text{ kg Urea}}{0.46\text{ kg N}} = \frac{54.3\text{ kg Urea}}{\text{ha}} \times \frac{0.0024\text{ ha}}{\text{plot}} \times \frac{1000\text{ g}}{\text{kg}} = \frac{130\text{ g Urea}}{\text{plot}}$$

B) Example fertilizer rate: 25 kg N ha^{-1} applied as ammonium nitrate (AN, 34-0-0)

$$\frac{25\text{ kg N}}{\text{ha}} \times \frac{1\text{ kg AN}}{0.34\text{ kg N}} = \frac{73.5\text{ kg AN}}{\text{ha}} \times \frac{0.0024\text{ ha}}{\text{plot}} \times \frac{1000\text{ g}}{\text{kg}} = \frac{176\text{ g AN}}{\text{plot}}$$

Equation B-3. Example Calculation of Phosphorus Fertilizer Rate per Plot

15 kg P_2O_5 ha^{-1} applied as mono-ammonium phosphate (11-52-0):

$$\frac{15\text{ kg }P_2O_5}{\text{Ha}} \times \frac{1\text{ kg MAP}}{0.52\text{ kg }P_2O_5} = \frac{28.8\text{ kg MAP}}{\text{ha}} \times \frac{0.0024\text{ ha}}{\text{plot}} \times \frac{1000\text{ g}}{\text{kg}} = \frac{69\text{ g MAP}}{\text{plot}}$$

Nitrogen fertilizer applied as part of 28.8 kg MAP ha^{-1}:

$$\frac{28.8\text{ kg MAP}}{\text{ha}} \times \frac{0.11\text{ kg N}}{1\text{ kg MAP}} = \frac{3.2\text{ kg N}}{\text{ha}} \times \frac{0.0024\text{ ha}}{\text{plot}} \times \frac{1000\text{ g}}{\text{kg}} = \frac{7.7\text{ g N}}{\text{plot}}$$

Highest P_2O_5 rate (75 kg P_2O_5 ha^{-1}) includes an N rate of:

$$\frac{75\text{ kg }P_2O_5}{\text{ha}} \times \frac{1\text{ kg MAP}}{0.52\text{ kg }P_2O_5} = \frac{144\text{ kg MAP}}{\text{ha}} \times \frac{0.11\text{ kg N}}{1\text{ kg MAP}} = \frac{15.9\text{ kg N}}{\text{ha}}$$

$$\frac{15.9\text{ kg N}}{\text{Ha}} \times \frac{0.0024\text{ ha}}{\text{plot}} \times \frac{1000\text{ g}}{\text{kg}} = \frac{38.1\text{ g N}}{\text{plot}}$$

Additional N requirement to equalize difference between highest (75 kg P_2O_5 ha^{-1}) and lowest MAP (15 kg P_2O_5 ha^{-1}) fertilizer rates:

$$\frac{38.1\text{ g N}}{\text{plot}} - \frac{7.7\text{ g N}}{\text{plot}} = \frac{30.4\text{ g N}}{\text{plot}} \times \frac{1\text{ g urea}}{0.46\text{ g N}} = \frac{66\text{ g Urea}}{\text{plot}}$$

Equation B-4. Total Area Seeded for *P. bilaiae* Practice

$$\frac{24\text{ m}^2}{\text{Plot}} \times \frac{6\text{ Plots}}{\text{Replicate}} \times \frac{6\text{ Replicates}}{\text{Trial}} \times \frac{1\text{ ha}}{10{,}000\text{ m}^2} = \quad 0.0864\text{ ha Trial}^{-1}$$

Equation B-5. Seed Required for *P. bilaiae* Practice

Seeding rate: 115 kg seed ha^{-1}

$$\frac{115\text{ kg Seed}}{\text{ha}} \times \frac{0.0864\text{ ha}}{\text{Trial}} = \quad 9.936\text{ kg Seed Trial}^{-1} \quad \text{or} \quad \sim 15\text{ kg Seed}$$

Equation B-6. Tagteam™ Required for *P. bilaiae* Practice

According to product label, one 2.2 kg bag of Tagteam™ treats 1360 kg of peas:

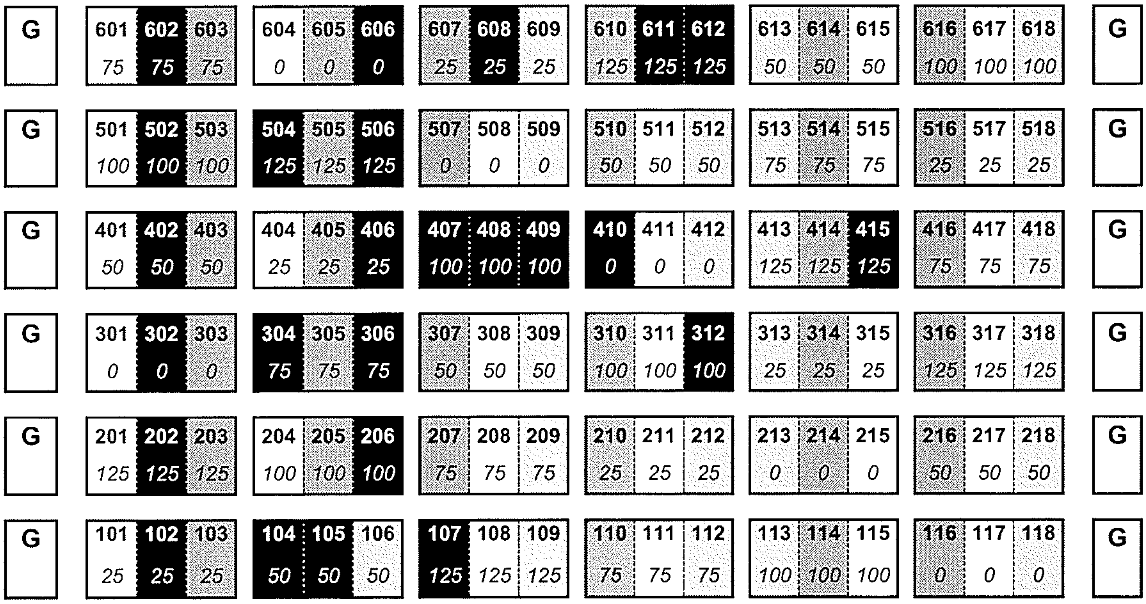
$$\frac{2.2\text{ kg bag}}{1360\text{ kg Seed}} = \frac{0.0016176\text{ kg}}{\text{kg Seed}} \times \frac{15\text{ kg}}{\text{Trial}} = \frac{0.0243\text{ kg Tagteam™}}{\text{Trial}}$$

Equation B-7. Determining Moisture Content of a Sample

$$\frac{\text{Sample Start Weight (g)} - \text{Sample End Weight (g)}}{\text{Sample Start Weight (g)}} \times 100 = \text{Moisture Content (\%)}$$

APPENDIX E – SOYBEAN/NITROGEN EXPERIMENT MAPS

2004 Homewood Soybean-Nitrogen Trial Location and Plot Maps



G	601 75	602 75	603 75	604 0	605 0	606 0	607 25	608 25	609 25	610 125	611 125	612 125	613 50	614 50	615 50	616 100	617 100	618 100	G
G	501 100	502 100	503 100	504 125	505 125	506 125	507 0	508 0	509 0	510 50	511 50	512 50	513 75	514 75	515 75	516 25	517 25	518 25	G
G	401 50	402 50	403 50	404 25	405 25	406 25	407 100	408 100	409 100	410 0	411 0	412 0	413 125	414 125	415 125	416 75	417 75	418 75	G
G	301 0	302 0	303 0	304 75	305 75	306 75	307 50	308 50	309 50	310 100	311 100	312 100	313 25	314 25	315 25	316 125	317 125	318 125	G
G	201 125	202 125	203 125	204 100	205 100	206 100	207 75	208 75	209 75	210 25	211 25	212 25	213 0	214 0	215 0	216 50	217 50	218 50	G
G	101 25	102 25	103 25	104 50	105 50	106 50	107 125	108 125	109 125	110 75	111 75	112 75	113 100	114 100	115 100	116 0	117 0	118 0	G

Legend:

Sample plot:

101
75

Plot Explanation:

Bold Top number is the plot number

Lower italicized number is fertilizer rate in kg of nutrient ha^{-1}

G

Guard/Border Plot

Background Shading Explanation:

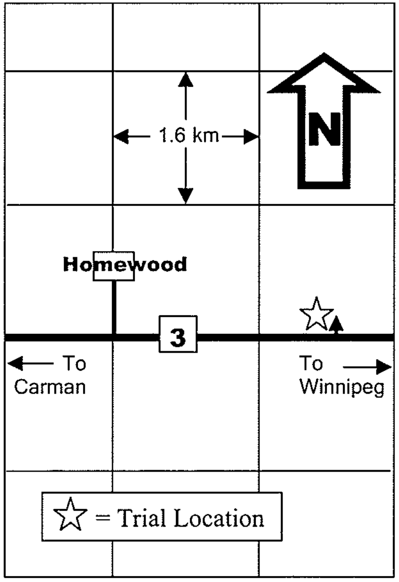
░ Single Application of N Fertilizer with Inoculant

▒ Split Application of N Fertilizer with Inoculant

□ Single Application of N Fertilizer with No Inoculant

■ Error Plot and/or Missing Data

Location Map:



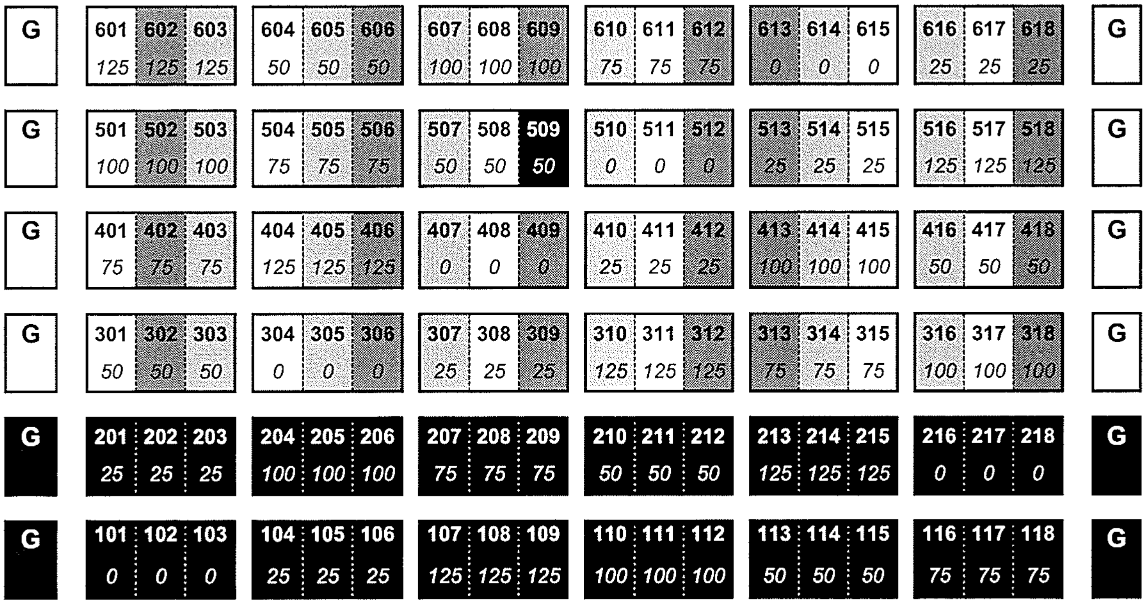
Notes:

Homewood, 2005 was seeded at a depth of approximately 2.5 – 3.0 cm. Faulty wiring on the distributor cone of the seeder resulted in seven plots being compromised and were, therefore, removed from the analysis.

During the process of trimming between the plots before harvest, difficulties were encountered in determining the outside rows. As a result, the number of rows for each plot varied and therefore, the number of harvested rows in each plot was recorded and yield data was adjusted accordingly.

An early fall frost prevented the crop from reaching full maturity. Plots were harvested with a significant amount of immature green seed, which was removed from the sample before determining yield and quality.

2005 Morris Soybean-Nitrogen Trial Location and Plot Maps



□ G	□ 601 125	▓ 602 125	░ 603 125	□ 604 50	░ 605 50	▓ 606 50	░ 607 100	□ 608 100	▓ 609 100	░ 610 75	□ 611 75	▓ 612 75	▓ 613 0	░ 614 0	□ 615 0	░ 616 25	□ 617 25	▓ 618 25	□ G
□ G	□ 501 100	▓ 502 100	░ 503 100	□ 504 75	░ 505 75	▓ 506 75	░ 507 50	□ 508 50	■ 509 50	░ 510 0	□ 511 0	▓ 512 0	▓ 513 25	░ 514 25	□ 515 25	░ 516 125	□ 517 125	▓ 518 125	□ G
□ G	□ 401 75	▓ 402 75	░ 403 75	□ 404 125	░ 405 125	▓ 406 125	░ 407 0	□ 408 0	▓ 409 0	░ 410 25	□ 411 25	▓ 412 25	▓ 413 100	░ 414 100	□ 415 100	░ 416 50	□ 417 50	▓ 418 50	□ G
□ G	□ 301 50	▓ 302 50	░ 303 50	□ 304 0	░ 305 0	▓ 306 0	░ 307 25	□ 308 25	▓ 309 25	░ 310 125	□ 311 125	▓ 312 125	▓ 313 75	░ 314 75	□ 315 75	░ 316 100	□ 317 100	▓ 318 100	□ G
■ G	■ 201 25	■ 202 25	■ 203 25	■ 204 100	■ 205 100	■ 206 100	■ 207 75	■ 208 75	■ 209 75	■ 210 50	■ 211 50	■ 212 50	■ 213 125	■ 214 125	■ 215 125	■ 216 0	■ 217 0	■ 218 0	■ G
■ G	■ 101 0	■ 102 0	■ 103 0	■ 104 25	■ 105 25	■ 106 25	■ 107 125	■ 108 125	■ 109 125	■ 110 100	■ 111 100	■ 112 100	■ 113 50	■ 114 50	■ 115 50	■ 116 75	■ 117 75	■ 118 75	■ G

Legend:

Sample plot:

Plot Explanation:

101
75

Bold Top number is the actual plot number
Lower italicized number is fertilizer rate in kg of nutrient ha^{-1}

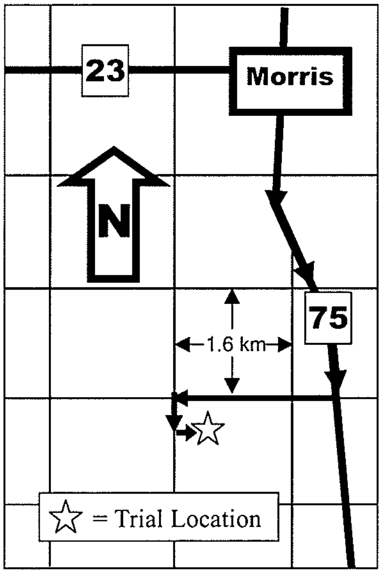
G

Guard/Border Plot

Background Shading Explanation:

▓ Single Application of N Fertilizer with Inoculant
░ Split Application of N Fertilizer with Inoculant
□ Single Application of N Fertilizer with No Inoculant
■ Error Plot and/or Missing Data

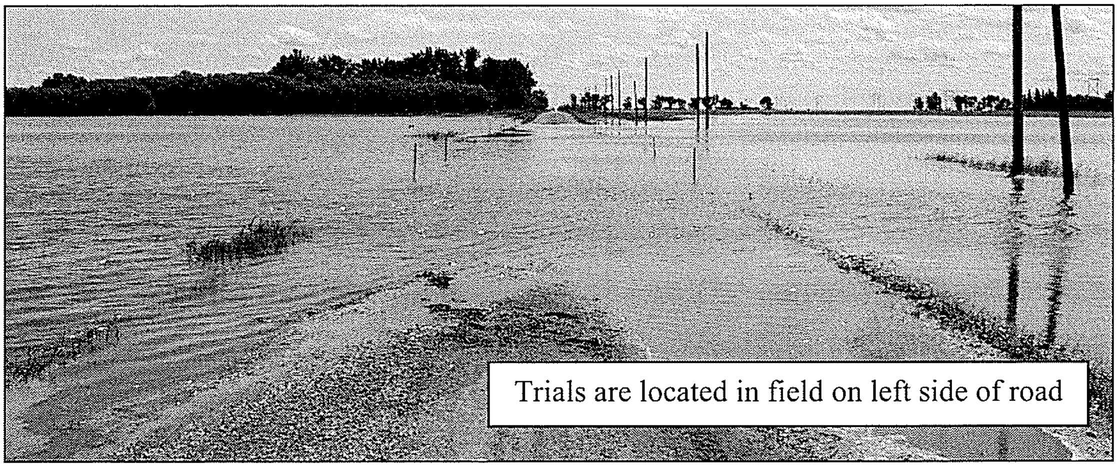
Trial Location:



Notes:

Shortly after seeding the Morris experiment, heavy rainfall resulted in a severe flood of the area. When the water receded approximately two weeks later, four replicates of the nitrogen trial were salvaged as the trial was located on a slight slope. Only the surviving four replicates were included in the data analysis. This trial was seeded at a depth of 2.0 – 2.5 cm and was later sprayed with Odyssey™ (imazamox and imazethapyr) at 42 g ha^{-1} by the cooperator. Biomass sampling was not completed because the desired plant stage was missed.

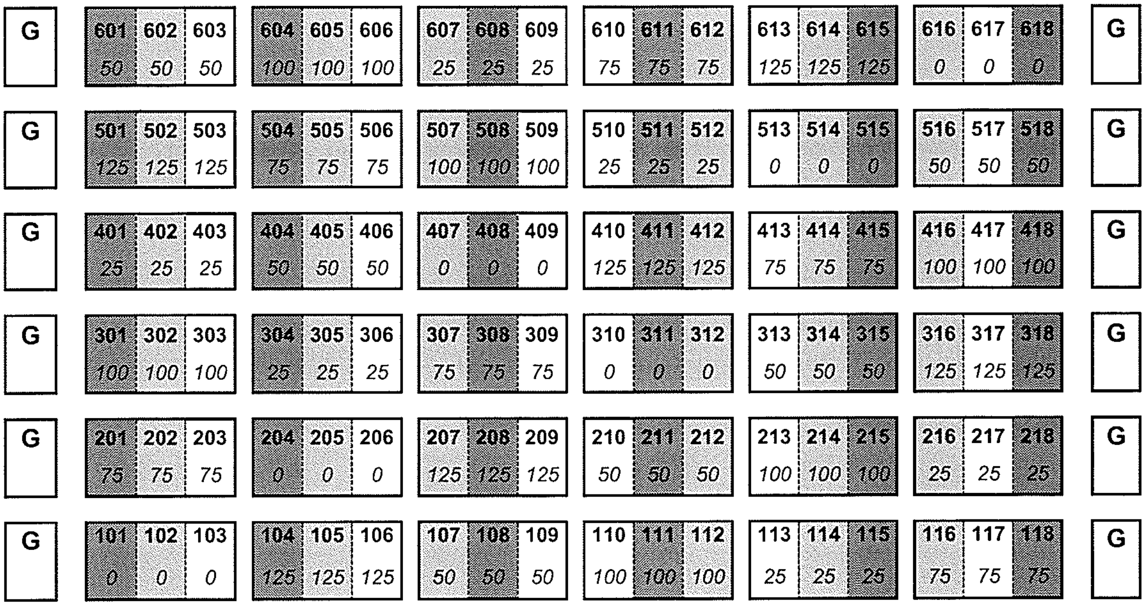
Morris Experimental Trials under Water (July 5, 2005)



Trials are located in field on left side of road

Road to Morris Experimental Trials under Water (July 5, 2005)

2006 Homewood Soybean-Nitrogen Trial Location and Plot Maps



G	**601** *50*	**602** *50*	**603** *50*	**604** *100*	**605** *100*	**606** *100*	**607** *25*	**608** *25*	**609** *25*	**610** *75*	**611** *75*	**612** *75*	**613** *125*	**614** *125*	**615** *125*	**616** *0*	**617** *0*	**618** *0*	G
G	**501** *125*	**502** *125*	**503** *125*	**504** *75*	**505** *75*	**506** *75*	**507** *100*	**508** *100*	**509** *100*	**510** *25*	**511** *25*	**512** *25*	**513** *0*	**514** *0*	**515** *0*	**516** *50*	**517** *50*	**518** *50*	G
G	**401** *25*	**402** *25*	**403** *25*	**404** *50*	**405** *50*	**406** *50*	**407** *0*	**408** *0*	**409** *0*	**410** *125*	**411** *125*	**412** *125*	**413** *75*	**414** *75*	**415** *75*	**416** *100*	**417** *100*	**418** *100*	G
G	**301** *100*	**302** *100*	**303** *100*	**304** *25*	**305** *25*	**306** *25*	**307** *75*	**308** *75*	**309** *75*	**310** *0*	**311** *0*	**312** *0*	**313** *50*	**314** *50*	**315** *50*	**316** *125*	**317** *125*	**318** *125*	G
G	**201** *75*	**202** *75*	**203** *75*	**204** *0*	**205** *0*	**206** *0*	**207** *125*	**208** *125*	**209** *125*	**210** *50*	**211** *50*	**212** *50*	**213** *100*	**214** *100*	**215** *100*	**216** *25*	**217** *25*	**218** *25*	G
G	**101** *0*	**102** *0*	**103** *0*	**104** *125*	**105** *125*	**106** *125*	**107** *50*	**108** *50*	**109** *50*	**110** *100*	**111** *100*	**112** *100*	**113** *25*	**114** *25*	**115** *25*	**116** *75*	**117** *75*	**118** *75*	G

Legend:

Sample plot: *Plot Explanation:*

101
75

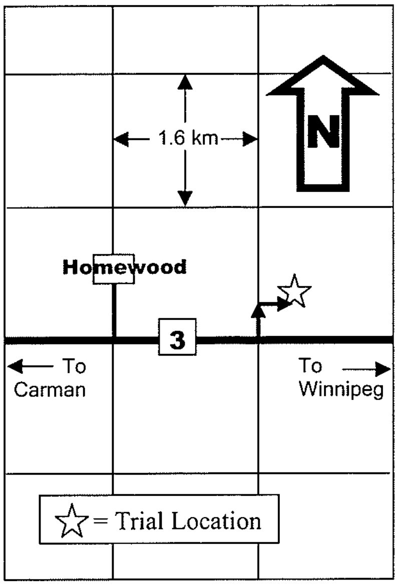
Bold Top number is the actual plot number
Lower italicized number is fertilizer rate in kg of nutrient ha^{-1}

G — Guard/Border Plot

Background Shading Explanation:

▓ Single Application of N Fertilizer with Inoculant
▒ Split Application of N Fertilizer with Inoculant
☐ Single Application of N Fertilizer with No Inoculant

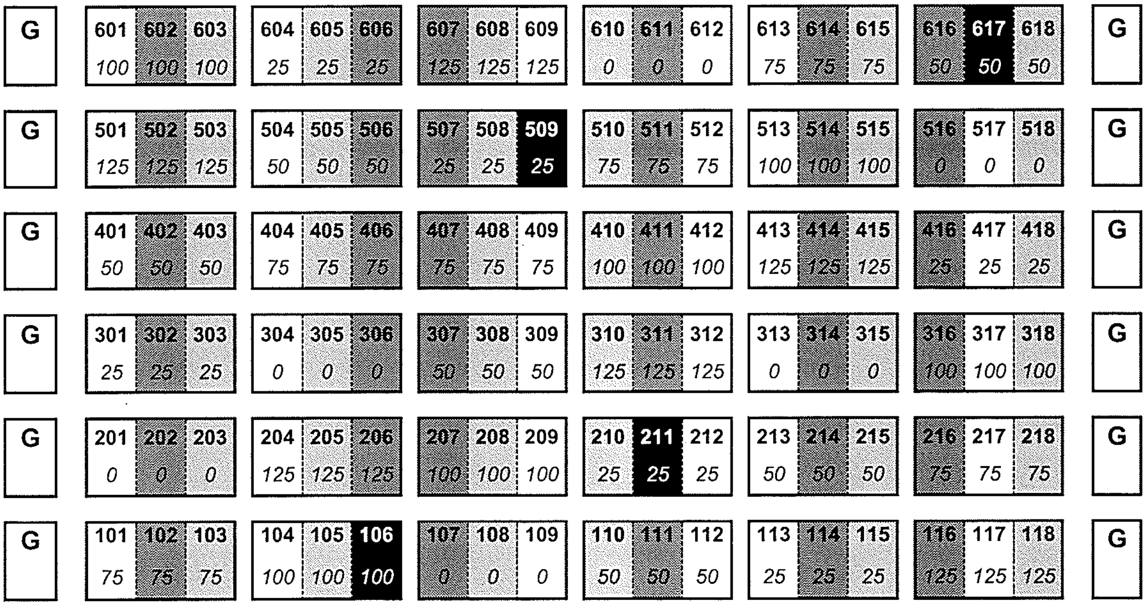
Trial Location:



Notes:

The Homewood site was seeded on May 23 at a depth of approximately 2.5 – 3.0 cm. Weed pressure was low and the field was sprayed with Pursuit™ and Poast Ultra™ at recommended rates of 210 ml ha^{-1} and 1111.5 ml ha^{-1}, respectively, by the cooperator on June 19.

2006 Morris Soybean-Nitrogen Trial Location and Plot Maps



G	□ **601** *100*	▓ **602** *100*	░ **603** *100*	□ **604** *25*	░ **605** *25*	▓ **606** *25*	▓ **607** *125*	░ **608** *125*	□ **609** *125*	░ **610** *0*	▓ **611** *0*	□ **612** *0*	□ **613** *75*	▓ **614** *75*	░ **615** *75*	▓ **616** *50*	■ **617** *50*	░ **618** *50*	G
G	□ **501** *125*	▓ **502** *125*	░ **503** *125*	□ **504** *50*	░ **505** *50*	▓ **506** *50*	▓ **507** *25*	░ **508** *25*	■ **509** *25*	░ **510** *75*	▓ **511** *75*	□ **512** *75*	□ **513** *100*	▓ **514** *100*	░ **515** *100*	▓ **516** *0*	□ **517** *0*	░ **518** *0*	G
G	□ **401** *50*	▓ **402** *50*	░ **403** *50*	□ **404** *75*	░ **405** *75*	▓ **406** *75*	▓ **407** *75*	░ **408** *75*	□ **409** *75*	░ **410** *100*	▓ **411** *100*	□ **412** *100*	□ **413** *125*	▓ **414** *125*	░ **415** *125*	▓ **416** *25*	□ **417** *25*	░ **418** *25*	G
G	□ **301** *25*	▓ **302** *25*	░ **303** *25*	□ **304** *0*	░ **305** *0*	▓ **306** *0*	▓ **307** *50*	░ **308** *50*	□ **309** *50*	░ **310** *125*	▓ **311** *125*	□ **312** *125*	□ **313** *0*	▓ **314** *0*	░ **315** *0*	▓ **316** *100*	□ **317** *100*	░ **318** *100*	G
G	□ **201** *0*	▓ **202** *0*	░ **203** *0*	□ **204** *125*	░ **205** *125*	▓ **206** *125*	▓ **207** *100*	░ **208** *100*	□ **209** *100*	░ **210** *25*	■ **211** *25*	□ **212** *25*	□ **213** *50*	▓ **214** *50*	░ **215** *50*	▓ **216** *75*	□ **217** *75*	░ **218** *75*	G
G	□ **101** *75*	▓ **102** *75*	░ **103** *75*	□ **104** *100*	░ **105** *100*	■ **106** *100*	▓ **107** *0*	░ **108** *0*	□ **109** *0*	░ **110** *50*	▓ **111** *50*	□ **112** *50*	□ **113** *25*	▓ **114** *25*	░ **115** *25*	▓ **116** *125*	□ **117** *125*	░ **118** *125*	G

Legend:

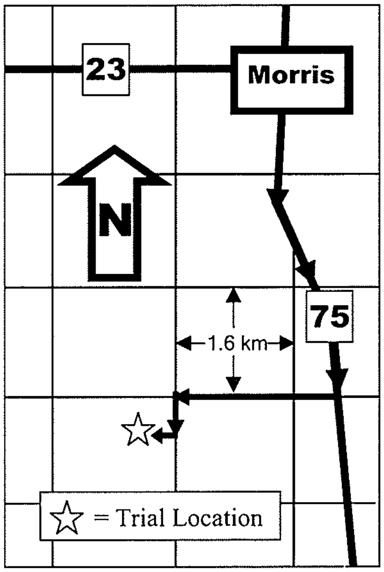
Sample plot: *Plot Explanation:*

101 *75*	**Bold Top number is the actual plot number** *Lower italicized number is fertilizer rate in kg of nutrient* ha^{-1}
G	Guard/Border Plot

Background Shading Explanation:

▓	Single Application of N Fertilizer with Inoculant
░	Split Application of N Fertilizer with Inoculant
□	Single Application of N Fertilizer with No Inoculant
■	Error Plot and/or Missing Data

Trial Location:

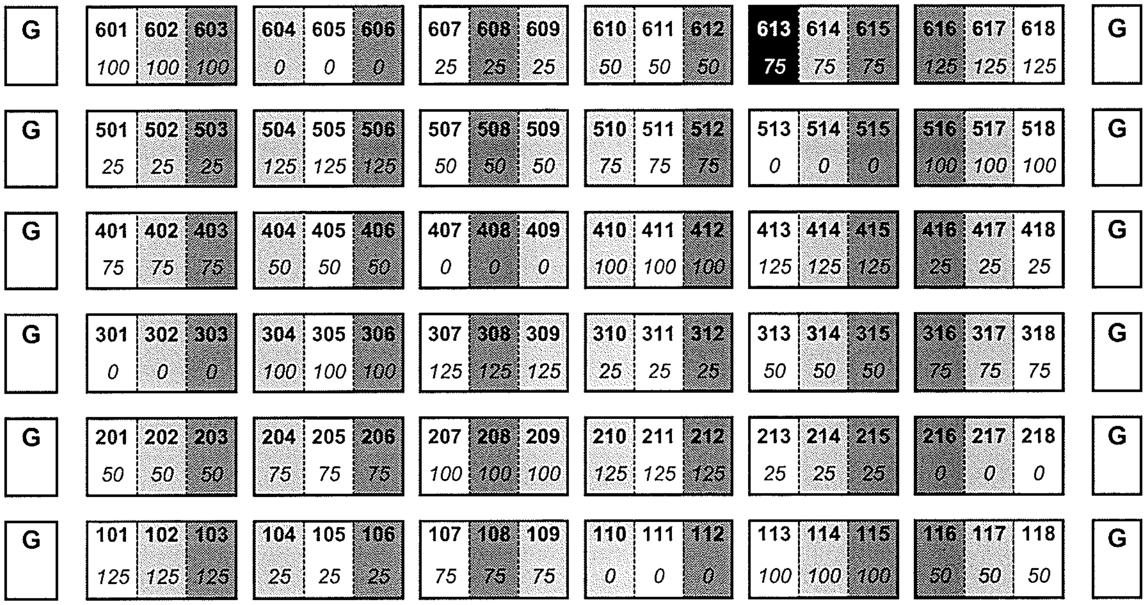


Notes:

Morris was seeded on May 24 at a depth of 2.5 – 3.0 cm. A problem occurred with the seeder cone which resulted in erratic feeding and stalling. Plots 106, 211, 509, and 617 were compromised and were omitted from the data analysis. As well, treatments 1, 13 and 7 were applied to plots 304, 305, and 306 rather than the intended plots of 404, 405, and 406. To fix the problem, treatments 4, 16 and 10 (originally intended for 304, 305 and 306) were applied to 404, 405 and 406.

Broadleaf weed pressure was high at this location, with extremely dense populations of smartweed (*Polygonum lapathifolium* L.), Canada thistle (*Cirsium arvense* (L.) Scop.), biennial wormwood (*Artemisia biennis* Willd.), wild mustard (*Brassica kaber* (D.C.) L.C. Wheeler) and wild buckwheat (*Polygonum convolvulus* L.). Herbicide was applied twice, once on June 12 with Odyssey™ at 42 g ha^{-1} and a second time on July 4 with Basagran™ (bentazon), Reflex™ (fomesafen) and Agral 90™ (non-ionic adjuvant), which were applied at the recommended rates of 1753 ml ha^{-1}, 580 ml ha^{-1}, and 1000 ml per 1000 l of solution, respectively.

2006 St. Norbert Soybean-Nitrogen Trial Location and Plot Maps



G	□ **601** *100*	░ **602** *100*	▓ **603** *100*	░ **604** *0*	□ **605** *0*	▓ **606** *0*	□ **607** *25*	▓ **608** *25*	░ **609** *25*	░ **610** *50*	□ **611** *50*	▓ **612** *50*	■ **613** *75*	░ **614** *75*	▓ **615** *75*	▓ **616** *125*	░ **617** *125*	□ **618** *125*	G
G	□ **501** *25*	░ **502** *25*	▓ **503** *25*	░ **504** *125*	□ **505** *125*	▓ **506** *125*	□ **507** *50*	▓ **508** *50*	░ **509** *50*	░ **510** *75*	□ **511** *75*	▓ **512** *75*	□ **513** *0*	░ **514** *0*	▓ **515** *0*	▓ **516** *100*	░ **517** *100*	□ **518** *100*	G
G	□ **401** *75*	░ **402** *75*	▓ **403** *75*	░ **404** *50*	□ **405** *50*	▓ **406** *50*	□ **407** *0*	▓ **408** *0*	░ **409** *0*	░ **410** *100*	□ **411** *100*	▓ **412** *100*	□ **413** *125*	░ **414** *125*	▓ **415** *125*	▓ **416** *25*	░ **417** *25*	□ **418** *25*	G
G	□ **301** *0*	░ **302** *0*	▓ **303** *0*	░ **304** *100*	□ **305** *100*	▓ **306** *100*	□ **307** *125*	▓ **308** *125*	░ **309** *125*	░ **310** *25*	□ **311** *25*	▓ **312** *25*	□ **313** *50*	░ **314** *50*	▓ **315** *50*	▓ **316** *75*	░ **317** *75*	□ **318** *75*	G
G	□ **201** *50*	░ **202** *50*	▓ **203** *50*	░ **204** *75*	□ **205** *75*	▓ **206** *75*	□ **207** *100*	▓ **208** *100*	░ **209** *100*	░ **210** *125*	□ **211** *125*	▓ **212** *125*	□ **213** *25*	░ **214** *25*	▓ **215** *25*	▓ **216** *0*	░ **217** *0*	□ **218** *0*	G
G	□ **101** *125*	░ **102** *125*	▓ **103** *125*	░ **104** *25*	□ **105** *25*	▓ **106** *25*	□ **107** *75*	▓ **108** *75*	░ **109** *75*	░ **110** *0*	□ **111** *0*	▓ **112** *0*	□ **113** *100*	░ **114** *100*	▓ **115** *100*	▓ **116** *50*	░ **117** *50*	□ **118** *50*	G

Legend:

Sample plot:

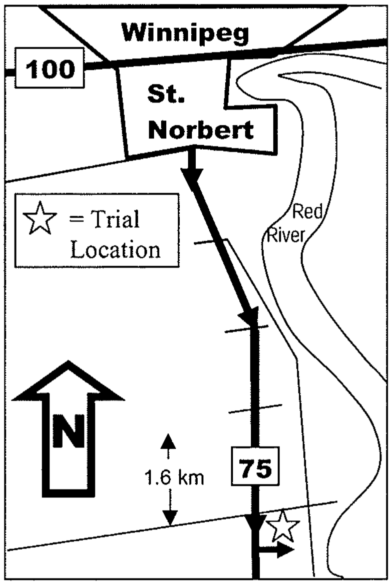
Plot Explanation:

101 *75*	**Bold Top number is the actual plot number** *Lower italicized number is fertilizer rate in kg of nutrient ha^{-1}*
G	Guard/Border Plot

Background Shading Explanation:

▓	Single Application of N Fertilizer with Inoculant
░	Split Application of N Fertilizer with Inoculant
□	Single Application of N Fertilizer with No Inoculant
■	Error Plot and/or Missing Data

Trial Location:



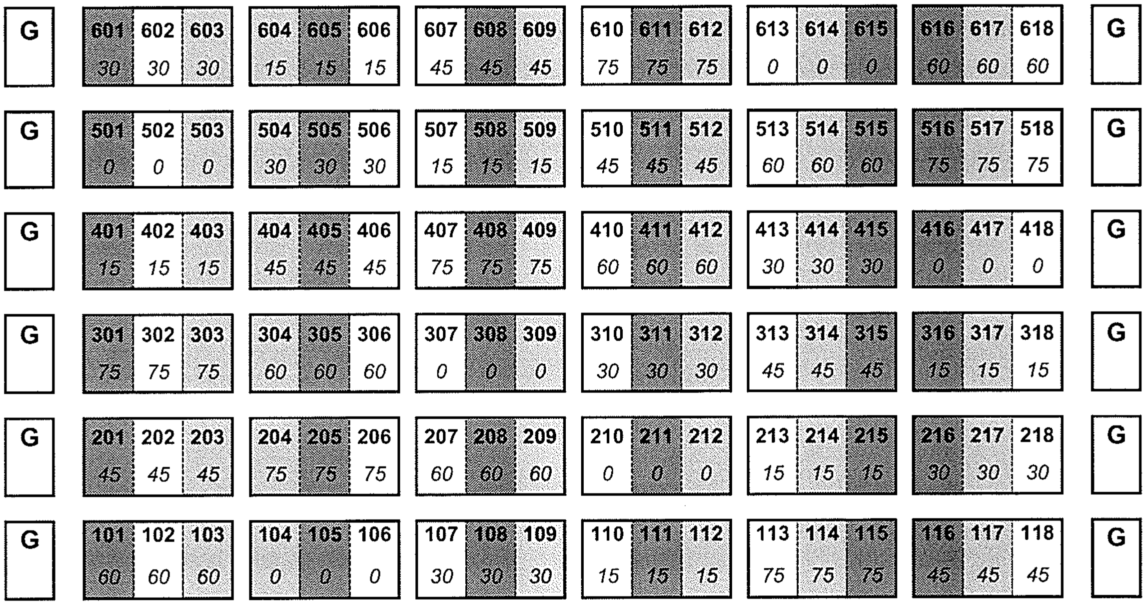
Notes:

This trial was seeded at a depth of 2.5 – 3.0 cm. Plot 613 was tripped late and therefore the fertilizer treatment was not applied to the entire plot, this plot was removed from the analysis.

On June 14, an application of Odyssey™ at 42 g ha^{-1} was made to control green foxtail (*Setaria viridis* (L.) Beauv.). Uneven crop growth was observed at this location; probably due to cultivation passes made by the cooperator during the previous fall as the variability seemed to follow an east-west pattern.

APPENDIX F – SOYBEAN/PHOSPHORUS EXPERIMENT MAPS

2004 Homewood Soybean-Phosphorus Trial Location and Plot Maps



G	601 30	602 30	603 30	604 15	605 15	606 15	607 45	608 45	609 45	610 75	611 75	612 75	613 0	614 0	615 0	616 60	617 60	618 60	G
G	501 0	502 0	503 0	504 30	505 30	506 30	507 15	508 15	509 15	510 45	511 45	512 45	513 60	514 60	515 60	516 75	517 75	518 75	G
G	401 15	402 15	403 15	404 45	405 45	406 45	407 75	408 75	409 75	410 60	411 60	412 60	413 30	414 30	415 30	416 0	417 0	418 0	G
G	301 75	302 75	303 75	304 60	305 60	306 60	307 0	308 0	309 0	310 30	311 30	312 30	313 45	314 45	315 45	316 15	317 15	318 15	G
G	201 45	202 45	203 45	204 75	205 75	206 75	207 60	208 60	209 60	210 0	211 0	212 0	213 15	214 15	215 15	216 30	217 30	218 30	G
G	101 60	102 60	103 60	104 0	105 0	106 0	107 30	108 30	109 30	110 15	111 15	112 15	113 75	114 75	115 75	116 45	117 45	118 45	G

Legend:

Sample plot:

Plot Explanation:

101
75

Bold Top number is the plot number
Lower italicized number is fertilizer rate in kg of P_2O_5 ha^{-1}

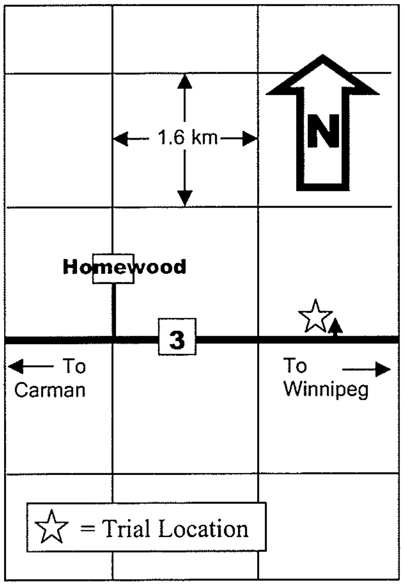
G

Guard / border plot

Background Shading Explanation:

░ Seed-placed fertilizer with *P. bilaiae*
▓ Fertilizer banded below seed
□ Seed-placed fertilizer

Trial Location:

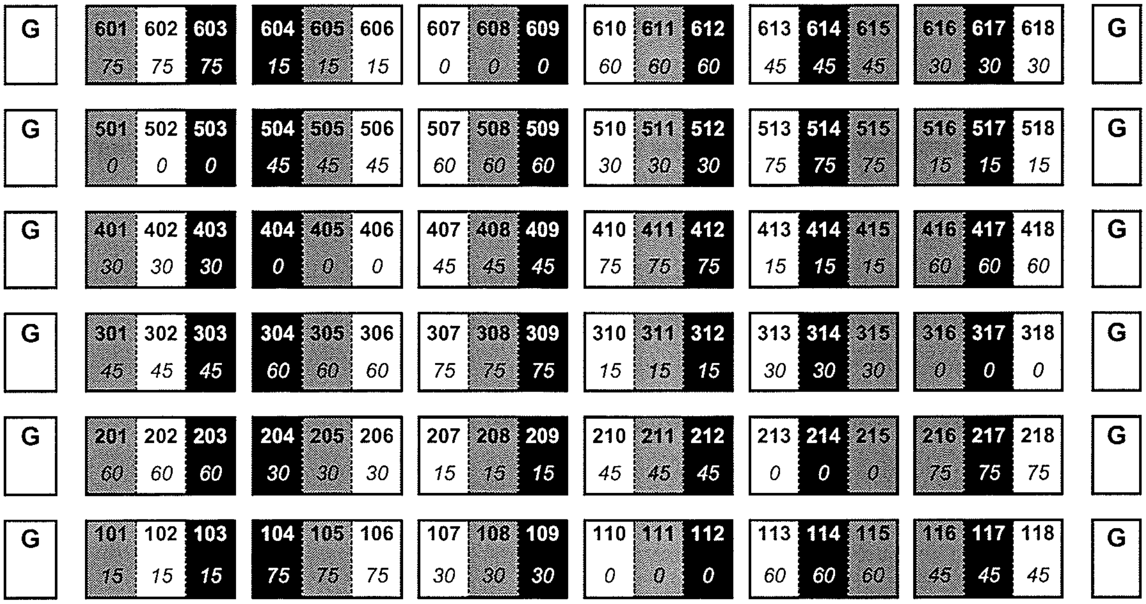


Notes:

This site-year was seeded on June 4 at a depth of 2.5 – 3.0 cm. Plants started emerging by June 17. A nodule assessment was not done on the phosphorus trial at Homewood in 2004 as this data parameter was not part of this experiment until 2005.

Narrowing the plots for harvest left some of the plots with more rows than others. The number of rows harvested was recorded and yields were adjusted accordingly.

2005 Homewood Soybean-Phosphorus Trial Location and Plot Maps

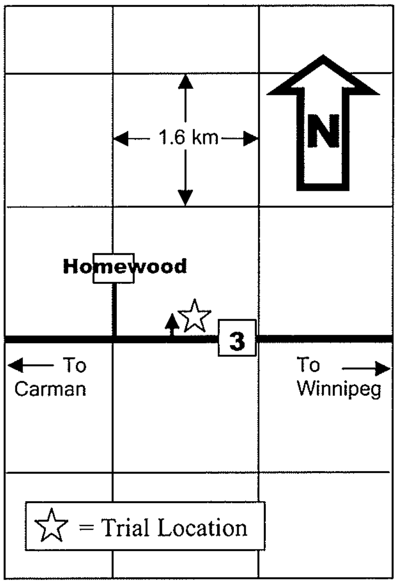


G	▨ 601 *75*	□ 602 *75*	■ 603 *75*	■ 604 *15*	▨ 605 *15*	□ 606 *15*	□ 607 *0*	▨ 608 *0*	■ 609 *0*	□ 610 *60*	▨ 611 *60*	■ 612 *60*	□ 613 *45*	■ 614 *45*	▨ 615 *45*	▨ 616 *30*	■ 617 *30*	□ 618 *30*	G
G	▨ 501 *0*	□ 502 *0*	■ 503 *0*	■ 504 *45*	▨ 505 *45*	□ 506 *45*	□ 507 *60*	▨ 508 *60*	■ 509 *60*	□ 510 *30*	▨ 511 *30*	■ 512 *30*	□ 513 *75*	■ 514 *75*	▨ 515 *75*	▨ 516 *15*	■ 517 *15*	□ 518 *15*	G
G	▨ 401 *30*	□ 402 *30*	■ 403 *30*	■ 404 *0*	▨ 405 *0*	□ 406 *0*	□ 407 *45*	▨ 408 *45*	■ 409 *45*	□ 410 *75*	▨ 411 *75*	■ 412 *75*	□ 413 *15*	■ 414 *15*	▨ 415 *15*	▨ 416 *60*	■ 417 *60*	□ 418 *60*	G
G	▨ 301 *45*	□ 302 *45*	■ 303 *45*	■ 304 *60*	▨ 305 *60*	□ 306 *60*	□ 307 *75*	▨ 308 *75*	■ 309 *75*	□ 310 *15*	▨ 311 *15*	■ 312 *15*	□ 313 *30*	■ 314 *30*	▨ 315 *30*	▨ 316 *0*	■ 317 *0*	□ 318 *0*	G
G	▨ 201 *60*	□ 202 *60*	■ 203 *60*	■ 204 *30*	▨ 205 *30*	□ 206 *30*	□ 207 *15*	▨ 208 *15*	■ 209 *15*	□ 210 *45*	▨ 211 *45*	■ 212 *45*	□ 213 *0*	■ 214 *0*	▨ 215 *0*	▨ 216 *75*	■ 217 *75*	□ 218 *75*	G
G	▨ 101 *15*	□ 102 *15*	■ 103 *15*	■ 104 *75*	▨ 105 *75*	□ 106 *75*	□ 107 *30*	▨ 108 *30*	■ 109 *30*	□ 110 *0*	▨ 111 *0*	■ 112 *0*	□ 113 *60*	■ 114 *60*	▨ 115 *60*	▨ 116 *45*	■ 117 *45*	□ 118 *45*	G

Legend:

Sample plot:	***Plot Explanation:***
101 *75*	**Bold Top number is the plot number** *Lower italicized number is fertilizer rate in kg of $P_2O_5\ ha^{-1}$*
G	Guard / border plot
	Background Shading Explanation:
▨	Fertilizer banded below seed
□	Seed-placed fertilizer
■	Error Plot and/or Missing Data

Trial Location:

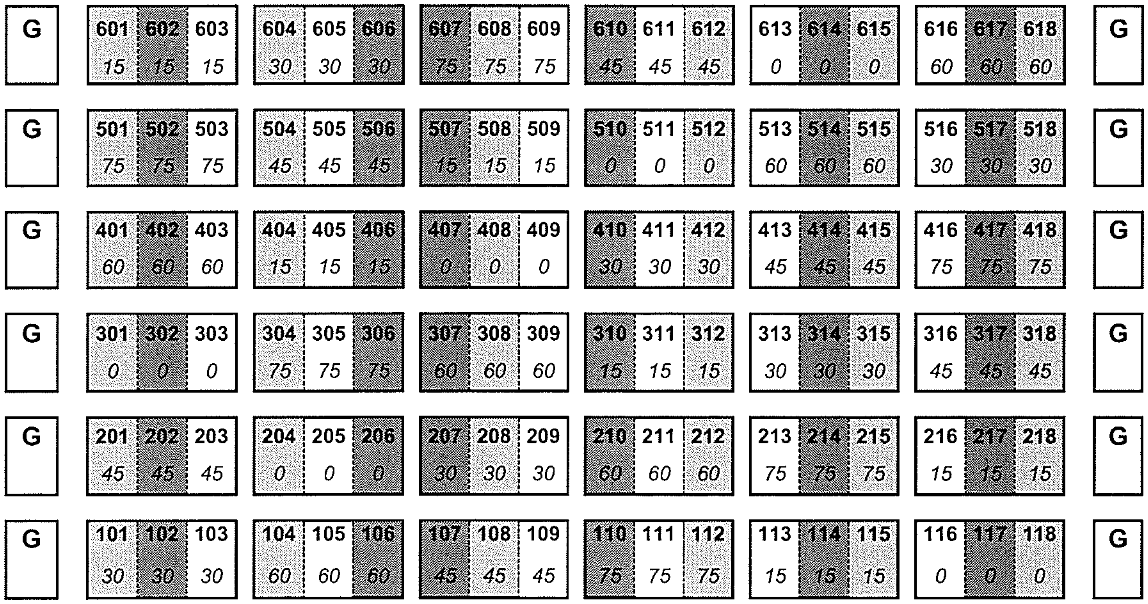


Notes:

Adequate time was not allowed for the *P. bilaiae*-treated seed to dry before it was placed in the seeder. During seeding, the slightly moist seed bridged over the seed cups and caused a reduction in seeding rate; it was estimated less than 25% of the seed was applied. For this reason the SP+*P. bilaiae* practice was removed from the data analysis. This trial was seeded at a depth of 2.5 – 3.0 cm.

The trial was sprayed with Pursuit™ and Poast Ultra™ at recommended rates of 210 ml ha^{-1} and 1111.5 ml ha^{-1}, respectively, by the cooperator before V3 as directed by the product label.

2006 Homewood Soybean-Phosphorus Trial Location and Plot Maps



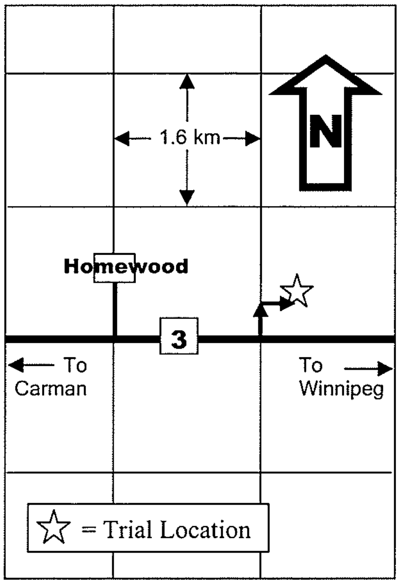
G	▒ **601** *15*	▓ **602** *15*	☐ **603** *15*	▒ **604** *30*	☐ **605** *30*	▓ **606** *30*	▓ **607** *75*	▒ **608** *75*	☐ **609** *75*	▓ **610** *45*	☐ **611** *45*	▒ **612** *45*	☐ **613** *0*	▓ **614** *0*	▒ **615** *0*	☐ **616** *60*	▓ **617** *60*	▒ **618** *60*	G
G	▒ **501** *75*	▓ **502** *75*	☐ **503** *75*	▒ **504** *45*	☐ **505** *45*	▓ **506** *45*	▓ **507** *15*	▒ **508** *15*	☐ **509** *15*	▓ **510** *0*	☐ **511** *0*	▒ **512** *0*	☐ **513** *60*	▓ **514** *60*	▒ **515** *60*	☐ **516** *30*	▓ **517** *30*	▒ **518** *30*	G
G	▒ **401** *60*	▓ **402** *60*	☐ **403** *60*	▒ **404** *15*	☐ **405** *15*	▓ **406** *15*	▓ **407** *0*	▒ **408** *0*	☐ **409** *0*	▓ **410** *30*	☐ **411** *30*	▒ **412** *30*	☐ **413** *45*	▓ **414** *45*	▒ **415** *45*	☐ **416** *75*	▓ **417** *75*	▒ **418** *75*	G
G	▒ **301** *0*	▓ **302** *0*	☐ **303** *0*	▒ **304** *75*	☐ **305** *75*	▓ **306** *75*	▓ **307** *60*	▒ **308** *60*	☐ **309** *60*	▓ **310** *15*	☐ **311** *15*	▒ **312** *15*	☐ **313** *30*	▓ **314** *30*	▒ **315** *30*	☐ **316** *45*	▓ **317** *45*	▒ **318** *45*	G
G	▒ **201** *45*	▓ **202** *45*	☐ **203** *45*	▒ **204** *0*	☐ **205** *0*	▓ **206** *0*	▓ **207** *30*	▒ **208** *30*	☐ **209** *30*	▓ **210** *60*	☐ **211** *60*	▒ **212** *60*	☐ **213** *75*	▓ **214** *75*	▒ **215** *75*	☐ **216** *15*	▓ **217** *15*	▒ **218** *15*	G
G	▒ **101** *30*	▓ **102** *30*	☐ **103** *30*	▒ **104** *60*	☐ **105** *60*	▓ **106** *60*	▓ **107** *45*	▒ **108** *45*	☐ **109** *45*	▓ **110** *75*	☐ **111** *75*	▒ **112** *75*	☐ **113** *15*	▓ **114** *15*	▒ **115** *15*	☐ **116** *0*	▓ **117** *0*	▒ **118** *0*	G

Legend:

Sample plot:

101 75	***Plot Explanation:*** **Bold Top number is the plot number** *Lower italicized number is fertilizer rate in kg of P_2O_5 ha^{-1}*
G	Guard / border plot
	Background Shading Explanation:
▒	Seed-placed fertilizer with *P. bilaiae*
▓	Fertilizer banded below seed
☐	Seed-placed fertilizer

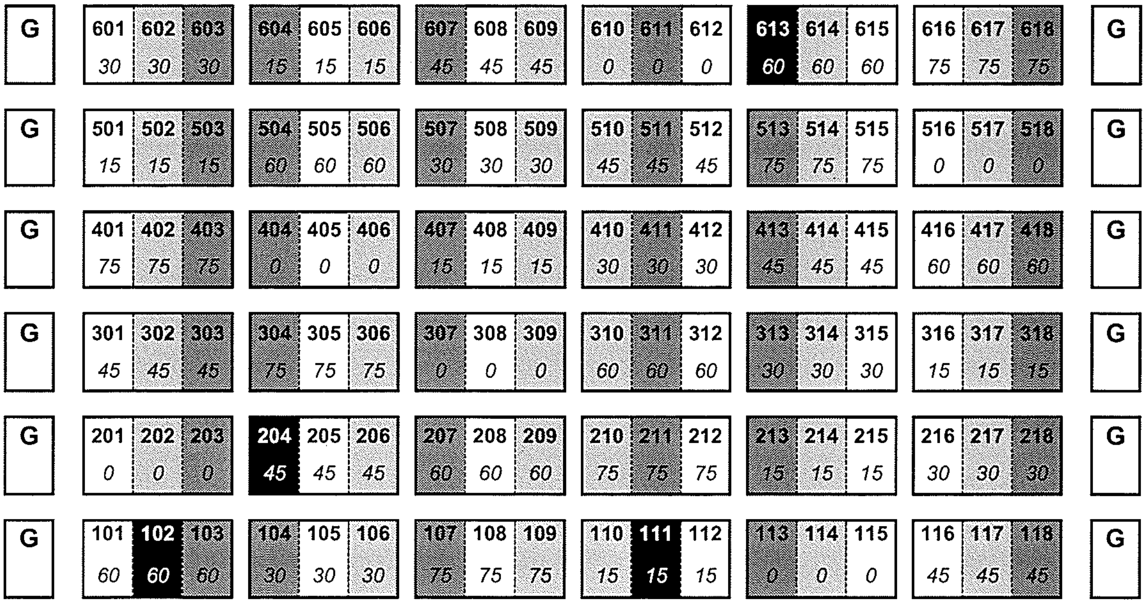
Trial Location:



Notes:

The banded fertilizer treatments were seeded approximately 1.5 cm deeper than the seed-placed treatments as a result of a seeder calibration error. Seed-placed treatments were seeded at a depth of 2.5 – 3.0 cm. This location was seeded on May 23. On June 19, the producer cooperator sprayed the trials with Pursuit™ and Poast Ultra™ at the recommended rates of 210 ml ha^{-1} and 1111.5 ml ha^{-1}, respectively.

2006 Morris Soybean-Phosphorus Trial Location and Plot Maps



G	601 30	602 30	603 30	604 15	605 15	606 15	607 45	608 45	609 45	610 0	611 0	612 0	613 60	614 60	615 60	616 75	617 75	618 75	G
G	501 15	502 15	503 15	504 60	505 60	506 60	507 30	508 30	509 30	510 45	511 45	512 45	513 75	514 75	515 75	516 0	517 0	518 0	G
G	401 75	402 75	403 75	404 0	405 0	406 0	407 15	408 15	409 15	410 30	411 30	412 30	413 45	414 45	415 45	416 60	417 60	418 60	G
G	301 45	302 45	303 45	304 75	305 75	306 75	307 0	308 0	309 0	310 60	311 60	312 60	313 30	314 30	315 30	316 15	317 15	318 15	G
G	201 0	202 0	203 0	204 45	205 45	206 45	207 60	208 60	209 60	210 75	211 75	212 75	213 15	214 15	215 15	216 30	217 30	218 30	G
G	101 60	102 60	103 60	104 30	105 30	106 30	107 75	108 75	109 75	110 15	111 15	112 15	113 0	114 0	115 0	116 45	117 45	118 45	G

Legend:

Sample plot:

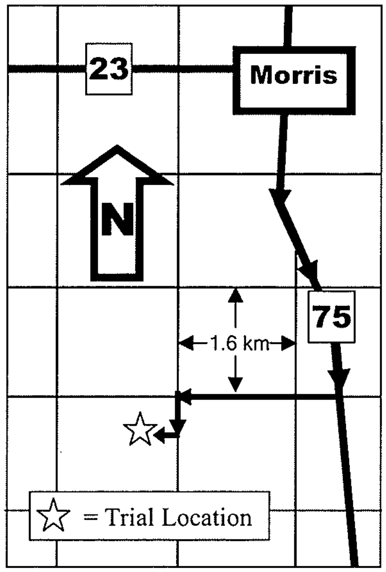
Plot Explanation:

101 75	**Bold Top number is the plot number** *Lower italicized number is fertilizer rate in kg of* $P_2O_5\ ha^{-1}$
G	Guard / border plot

Background Shading Explanation:

░	Seed-placed fertilizer with *P. bilaiae*
▓	Fertilizer banded below seed
□	Seed-placed fertilizer
■	Error Plot and/or Missing Data

Trial Location:

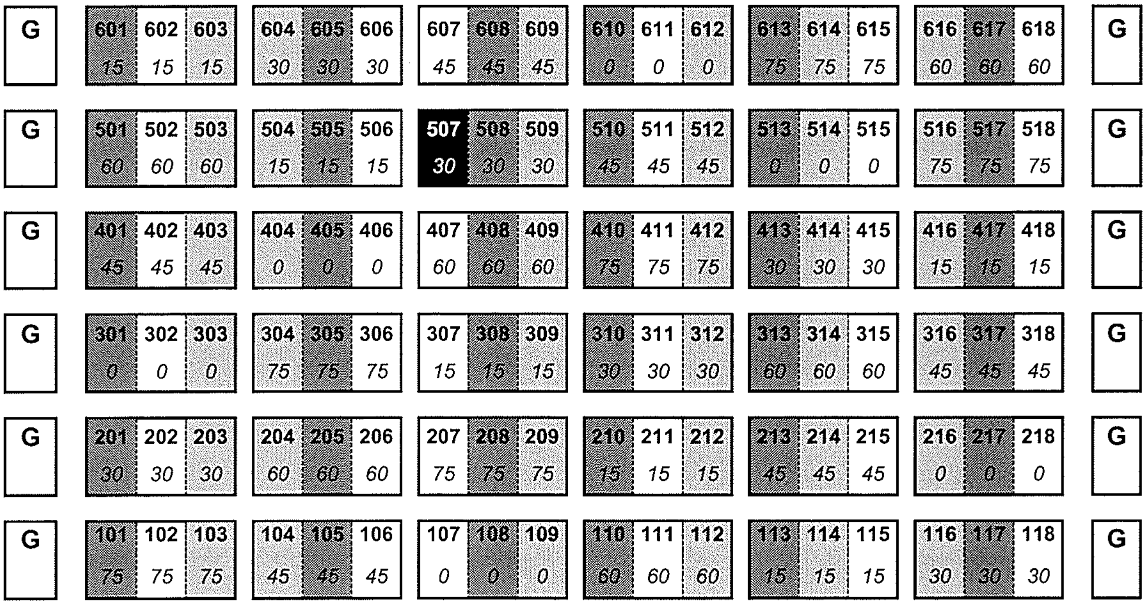


Notes:

Mechanical failure of the seeder cone resulted in plots 102, 111, 204, and 613 being compromised and as such, were discarded from the experiment. This trial was seeded at a depth of 2.5 – 3.0 cm.

The weed spectrum for this trial was similar to that of the nitrogen trial at this location. The site was sprayed for weed control on June 12 as well as on July 4 at the same time using the same rates and products as the nitrogen trial at this location.

2006 St. Norbert Soybean-Phosphorus Trial Location and Plot Maps



G	601 15	602 15	603 15	604 30	605 30	606 30	607 45	608 45	609 45	610 0	611 0	612 0	613 75	614 75	615 75	616 60	617 60	618 60	G
G	501 60	502 60	503 60	504 15	505 15	506 15	507 30	508 30	509 30	510 45	511 45	512 45	513 0	514 0	515 0	516 75	517 75	518 75	G
G	401 45	402 45	403 45	404 0	405 0	406 0	407 60	408 60	409 60	410 75	411 75	412 75	413 30	414 30	415 30	416 15	417 15	418 15	G
G	301 0	302 0	303 0	304 75	305 75	306 75	307 15	308 15	309 15	310 30	311 30	312 30	313 60	314 60	315 60	316 45	317 45	318 45	G
G	201 30	202 30	203 30	204 60	205 60	206 60	207 75	208 75	209 75	210 15	211 15	212 15	213 45	214 45	215 45	216 0	217 0	218 0	G
G	101 75	102 75	103 75	104 45	105 45	106 45	107 0	108 0	109 0	110 60	111 60	112 60	113 15	114 15	115 15	116 30	117 30	118 30	G

Legend:

Sample plot:

101 *75*

Plot Explanation:

Bold Top number is the plot number

Lower italicized number is fertilizer rate in kg of $P_2O_5\ ha^{-1}$

G

Guard / border plot

Background Shading Explanation:

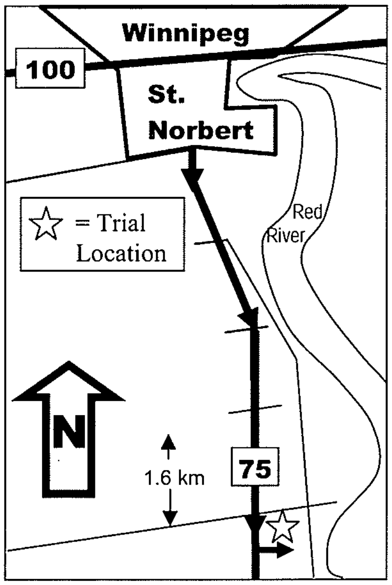
▒ Seed-placed fertilizer with *P. bilaiae*

▓ Fertilizer banded below seed

☐ Seed-placed fertilizer

■ Error Plot and/or Missing Data

Trial Location:



Notes:

While seeding, plot 507 was compromised from a cone failure and was removed from the analysis. This trial was seeded at a depth of 2.5 – 3.0 cm.

On June 14, the site was sprayed with Odyssey at the recommended rate of 42.7 g ha^{-1} to control green foxtail. There was some unevenness in growth noted in this experiment likely caused by a field operation (such as tillage) that occurred the previous fall.

APPENDIX G – DISCARDED ENVIRONMENTS

Morris, 2004 – Nitrogen and Phosphorus Experiments

Seeding of the Morris experimental site was delayed by steady rains until early June. This trial was seeded when the soil was still quite wet since further delays would not allow a long enough growing season for the soybeans to mature. The clay soil of the Morris area tended to stick to the packer wheels of the seeder, essentially rolling up the top layer of soil (including the seed and fertilizer) onto the packer wheels. This resulted in uneven germination and questionable fertilizer application. As a result, this environment was omitted from the data analysis.

Homewood, 2005 - Nitrogen Experiment

A human error compromised the trial at this location.

Morris, 2005 - Phosphorus Experiment

This environment was seeded on June 2 but shortly after, heavy rains left this trial under water for approximately two weeks. After the water drained, it was discovered the entire trial had been destroyed.

St. Norbert, 2005 - Nitrogen and Phosphorus Experiments

Overall, very poor emergence rates and low plant populations were observed at this location, likely a result of heavy rains occurring both before and after seeding as well as the lack of a seed treatment. Populations ranged from 30,350 to 257,977 plants ha^{-1} with an average of 126,257 and 140,510 plants ha^{-1} for the N and P experiments, respectively. Due to the extremely variable and low plant populations, this environment was not included in the data analysis.