

SPENT LIME EFFECTS ON YIELD, QUALITY, AND APHANOMYCES ROOT ROT OF SUGARBEET

Carol E. Windels¹, Jason R. Brantner¹, Albert L. Sims¹ and Carl A. Bradley²

Professor, Research Fellow, Associate Professor, and Assistant Professor, respectively

¹University of Minnesota, NW Research and Outreach Center, Crookston 56716

²North Dakota State University, Fargo (currently University of Illinois, Urbana)

Aphanomyces cochlioides (= *A. cochlioides*) is a serious economic pathogen and infests over 50% of acres planted to sugarbeet in the Red River Valley (RRV) and most acres in southern Minnesota. When soil is warm and wet, *A. cochlioides* causes damping-off of seedlings and root rot of older plants. Storage of diseased roots in piles contributes to additional losses. *A. cochlioides* persists in soil for years. Consequently, growing sugarbeet requires all available control options including early planting of resistant varieties treated with the fungicide Tachigaren and various cultural practices (e.g., cultivation and improved drainage) to avoid or lessen infections by *A. cochlioides*. However, when inoculum levels of the pathogen are high and soil is wet, implementation of these measures is inadequate for economic yields and fields often are abandoned or yield poorly. This chronic situation has generated interest in finding effective, alternative methods to control *A. cochlioides*.

The sugarbeet purification process results in the by-product "spent lime". Lime (calcium carbonate) precipitates impurities in sugarbeet juice. Purified juice is further processed into crystal sugar, but spent lime (14% less acid neutralizing power of fresh lime) contains impurities and becomes a sugarbeet industry by-product. Seven factories in the RRV and southern Minnesota generate 500,000 tons (dry weight) of spent lime annually and some has been stockpiled for 20 years. Literature on sugarbeet spent lime is limited and publications usually are in government and company documents. Most spent lime generated in Europe is applied to land as an amendment to increase soil pH and supply nutrients. In Great Britain, it is marketed and sold to conventional and organic growers as LimeX. In the late 1970s in the Salinas Valley of California, spent lime from a near-by sugarbeet processing factory was applied at 2 to 4.5 tons per acre in fields (pH less than 6.8) severely infested with the clubroot pathogen, *Plasmodiophora brassicae* (3). A single application gave "virtually complete control" of clubroot of crucifer crops for 2 to 3 years. In other areas of the world, various forms of lime (not spent lime) have been applied for over 200 years to control clubroot of crucifers, but results have been erratic. Little is known about how various forms of lime affect the clubroot pathogen.

Growers in southern Minnesota started applying spent lime (4 to 8 tons wet weight A⁻¹) to sugarbeet fields in the late 1990s to increase soil pH and reduce carryover of the soybean herbicides Pursuit and Raptor (1), which persist in soil and are toxic to sugarbeet. Spent lime increased sugarbeet yields in fields with and without herbicide carry-over (1) - and less *Aphanomyces* root rot was observed. Growers have continued to apply spent lime the year before planting sugarbeet (typically every 3 years). In the last couple of years, growers in the RRV also have been applying spent lime to their sugarbeet fields. In trials in the RRV, spent lime (3 and 10 tons wet weight A⁻¹) was applied in two *Aphanomyces*-infested fields (baseline pH values of 5.9 and 7.8) and within 1 year, there were significant reductions in *Aphanomyces*

root rot and increases in sucrose yields compared to the non-limed control (2). In 2003, a producer in Breckenridge, MN observed healthy sugarbeet roots in a 5-acre portion of a field where spent lime (20 to 25 tons wet weight A^{-1}) had been applied 7 years earlier - while the remainder of the field had poor stand, stunted growth, and severe *Aphanomyces* root rot.

The objectives in our 2006 field trials were to measure effects of spent lime applications made in October, 2003 and April, 2004 at Hillsboro, North Dakota and Breckenridge, Minnesota, respectively, on: (1.) yield, quality, and *Aphanomyces* root rot of sugarbeet and (2.) *Aphanomyces* soil index values (an indicator of activity and population levels of *A. cochlioides*).

MATERIALS AND METHODS

Establishment of field trials. Long-term trials were established at Hillsboro, ND (pH = 7.4) in October, 2003 and at Breckenridge, MN (pH = 6.5) in April, 2004. The Hillsboro site has a history of moderate *Aphanomyces* root rot and Breckenridge has severe root rot. Each site was divided into four, 1-acre experiments; each experiment included four rates of spent lime and an untreated control, replicated four times in a randomized block design. Treatments at Hillsboro were 0, 5, 10, 20 and 30 tons wet weight of spent lime A^{-1} (= 0, 3.3, 6.5, 13, and 19.5 tons dry weight A^{-1} , respectively) and at Breckenridge were 0, 5, 10, 15 and 20 tons wet weight A^{-1} (= 0, 2.7, 5.3, 8, and 10.6 tons dry weight A^{-1} , respectively). Each treatment plot measures 33 x 60 ft. The four experiments were established so sugarbeet could be sown in one experiment each year from 2005 to 2008; the three experiments not sown with sugarbeet in these years are sown by the grower-cooperator with the same crop as grown in the field. This approach allows evaluation of spent lime applications on sugarbeet and other crops in the rotation every season through 2008. To allow lime treatments to stabilize in 2004, corn 'DeKalb 3551RR' was sown across the four experiments at Hillsboro and wheat 'Grandin' was sown at Breckenridge. In 2005, sugarbeet was sown in one of the four experiments at each location for the first time; experiments not sown to sugarbeet at Hillsboro were fallowed (field was too wet to plant soybean) and at Breckenridge were planted to wheat 'Knudsen'. Previous results have been reported (9,10).

2006 Sugarbeet field trials. Sugarbeet was sown in one experiment (non-limed and limed plots, replicated four times) at Hillsboro on May 5 and at Breckenridge on May 9, 2006. Varieties Seedex Alpine (partially resistant to *Aphanomyces*) and Hillehog 2467RZ (susceptible and treated with 45 g of Tachigaren per unit of seed) were sown as subplots within lime treatment and control plots. Seed was sown every 2 inches in rows 60-feet long and 22- inches apart (six rows of each variety centered within each plot). A pre-plant application of the herbicide Nortron (3.75 lb a.i. A^{-1}) was incorporated into soil and the insecticide Counter 15G (12 lb product A^{-1}) was applied modified in-furrow at planting. After sugarbeet seedlings emerged, 10 feet of row was cut from the front and back of each plot, resulting in rows 40 feet long. Microrates of Progress + UpBeet + Stinger + Select + MSO (8.7 fl oz + 0.125-0.5 oz + 1.3 fl oz + 0-2 oz + 1.5% A^{-1} , respectively) were applied on May 29 and June 3, 12, and 20 at Hillsboro; Betanex (16 fl oz A^{-1}) was substituted for Progress on the last application date. The same microrate mix was applied at Breckenridge on May 29 and June 12 and 20, but rates varied slightly from those used at Hillsboro. Plants were hand-thinned to a 6-inch spacing on June 6 at Hillsboro and to a 4-inch spacing (because of considerable early-season *Aphanomyces* root rot) on June 9 at Breckenridge. Plots at both locations were cultivated on June 14. *Cercospora* leaf spot was controlled by

application of Eminent (13 oz A⁻¹) and Headline (9 fl oz A⁻¹) on August 21 and September 5, respectively, at Hillsboro and on July 27 and August 17, respectively, at Breckenridge (20 gpa at 100 psi). Alleys separating replicates were rototilled throughout the season.

Data were collected on seedling stand at 2 and 4 weeks after planting and shortly after thinning at both locations. Plots were harvested at Hillsboro on October 10 and Breckenridge on October 9 on the two middle rows of each variety per treatment. Ten roots were randomly selected and analyzed for yield and sucrose quality by the American Crystal Sugar Company Quality Laboratory, East Grand Forks, MN. On October 13, 20 roots were randomly selected from each subplot and rated for *Aphanomyces* root rot (0 – 7 scale, 0 = healthy and 7 = root completely rotted and foliage dead).

Aphanomyces soil index values (SIVs). Soil samples were collected from plots (including subplots where two sugarbeet varieties were grown in 2005) at Hillsboro on May 17 and 23, and Breckenridge on April 25 to 27 (total of 100 soil samples per location). Six soil cores (2.5-inch diameter x 6-inch depth) were collected randomly across each plot and combined. Soil samples were screened through 0.25-inch hardware cloth to remove debris and then stored in a walk-in cooler until assayed (usually within 1 month after collection).

Soil samples were assayed in a controlled environment chamber to determine *Aphanomyces* soil index values (SIVs). This assay indicates potential for *Aphanomyces* diseases and populations of *A. cochlioides*. Twenty-five seed of sugarbeet ‘ACH 261’ were sown per pot (4 pots per soil sample) to “bait” *A. cochlioides* from soil. Pots were placed in a controlled environment chamber in a randomized block design at 70 ± 2 °F for 1 week for optimal emergence. Then temperatures were increased to 79 ± 2 °F (14 hour photoperiod) and soil was kept moist to favor infection and disease development. Stand counts were made twice weekly starting at emergence. Dying seedlings were removed at each stand count to prevent disease from spreading to adjacent plants. Four weeks after planting, surviving seedlings were rated for disease on a 0 to 3 scale (0 = healthy and 3 = stem and root brown, constricted, and plant dead). Disease ratings and numbers of dead seedlings during the 4-week assay were used to calculate an *Aphanomyces* SIV (0 to 100 scale, 0 = *Aphanomyces*-free and 100 = soil severely infested with *A. cochlioides*).

To determine soil pH, small quantities of soil from all plots collected in April and May, 2006 were oven-dried overnight at 86 °F and ground into powder with a mortar and pestle. A 5 gram quantity was removed and mixed with 5 ml of deionized water. After 10 minutes, a pH probe was inserted into the mixture, gently stirred for 3 seconds, and the pH was read (Accumet® pH Meter 15, Fisher Scientific).

Data analysis. Data were subjected to analysis of variance and if significant ($P = 0.05$), means were separated by Least Significant Difference (LSD). Regression analyses also were done to determine the rate of spent lime needed to maximize pounds of sucrose recovered per acre.

RESULTS

2006 Sugarbeet field trials. Hillsboro. Soil pH in non-limed plots averaged 7.1 (Table 1). All rates of spent lime significantly increased soil pH and there were small increases in pH values

Regression analysis revealed no significant relationship between amount of spent lime applied and yield of recoverable sucrose A⁻¹ (R² = 0.6782). In 2006, 5 tons wet weight of spent lime A⁻¹ was sufficient to significantly increase recovery of sucrose. Furthermore, plots treated with 5 and 30 tons of spent lime resulted in significantly more gross dollars A⁻¹ compared to the non-limed control; the other lime treatments had intermediate economic returns.

The sugarbeet variety Seedex Alpine (partially resistant to *A. cochlioides*) had significantly higher stands than the susceptible variety at 12 days after planting, but there were no significant differences in stand between varieties for the rest of the season (Table 1). Seedex Alpine yielded significantly lower percent sucrose and pounds of sucrose per ton compared to the susceptible variety but had significantly higher tons of roots, pounds of recoverable sucrose, and gross economic return compared to the *Aphanomyces*-susceptible variety.

Breckenridge. Soil pH in non-limed plots averaged 6.5 and all rates of spent lime increased soil pH (Table 2). Soil pH levels of samples collected in April, 2006 were slightly higher compared to measurements made in September, 2004 (9), 6 months after spent lime was applied.

Table 2. Breckenridge, MN: Soil pH, stands, root rot ratings, and harvest data of sugarbeet sown on May 9, 2006, 25 months after several rates of spent lime were applied in a field naturally infested with high inoculum densities of *Aphanomyces cochlioides*.

Main Treatments		Soil pH	No. plants/80-ft row (Days after planting) ^x			No. roots harvested/ 80 ft row	RRR 0-7 ^y	Yield (Ton/A)	Sucrose		Gross return (\$/A)	
Lime (Ton/A) ^v			13	28	Post-thinning				%	lb/T		lb recov./A
Wet wt.	Dry wt.											
0	0	6.53 a	253	242 b	133	76 a	4.7 a	14.3 a	15.2 a	270 a	3911 a	388 a
5	2.7	7.51 b	245	231 a	125	85 b	3.6 b	26.0 b	16.3 b	292 b	7550 b	812 b
10	5.3	7.61 b	252	245 bc	139	103 d	3.3 b	30.7 bc	16.2 b	289 b	8858 bc	942 b
15	8.0	7.78 c	243	254 c	145	106 d	3.3 b	31.5 c	16.4 b	291 b	9168 c	987 b
20	10.6	7.79 c	228	246 bc	134	96 c	3.3 b	30.5 bc	16.3 b	290 b	8849 bc	949 b
LSD (<i>P</i> = 0.05) ^z		0.16	NS	10	NS	6	0.5	5.1	0.6	14	1523	181
Variety ^w												
HM 2467RZ + 45 g Tach		-	230 a	243	137	93	3.7	22.8 a	16.2	288	6604 a	707 a
Seedex Alpine (0 Tach)		-	258 b	244	134	93	3.6	30.4 b	16.0	285	8731 b	924 b
LSD (<i>P</i> = 0.05) ^z			14	NS	NS	NS	NS	1.6	NS	NS	457	57

^v Spent lime was applied in April, 2004 in a randomized block design of four replicates per experiment (total of four experiments) and incorporated by cultivation. In 2004, the four experiments were sown with wheat; in 2005, one experiment was sown with sugarbeet and the other three experiments were sown with wheat. In 2006, one experiment was sown with two sugarbeet varieties and the other three experiments were sown with soybean. Each value in this portion of the table is averaged across both sugarbeet varieties sown in one experiment in 2006.

^w Sugarbeet varieties Hilleshog 2467 RZ (susceptible to *Aphanomyces* and treated with 45 g of Tachigaren [Tach] per unit of seed) and Seedex Alpine (partially resistant to *Aphanomyces*) were sown as subplots within each spent lime treatment plot. Plots were harvested on October 9, 2006. Each value in this portion of the table is averaged across all lime treatments.

^x Plots were sown at 142,560 seeds per acre (seed every 2 inches in row 22 inches apart) and hand-thinned to a 4-inch spacing on June 9 (34 days after planting). Post-thinning stand counts were made on June 12 (3 days after thinning).

^y RRR = *Aphanomyces* root rot rating, 0–7 scale (0 = roots healthy; 7 = root completely rotted and foliage dead).

^z LSD = Least significant difference, *P* = 0.05; for each column, values followed by the same letter are not significantly different; NS = not significantly different.

There were no significant interactions between rate of lime and sugarbeet variety for nearly all data collected at Breckenridge, so results are presented separately for these main effects (Table 2). Within 2 weeks after planting, weather was too dry for *A. cochlioides* to infect seedlings and there were no differences in stands among limed and non-limed control plots. Rainfall from about mid May through mid June resulted in considerable activity of *A. cochlioides*. At 28 days after planting, there were significant differences in stand among treatments. Stands were statistically lower in plots treated with 5 tons of lime compared to the other limed plots and control; stands were highest in plots treated with 15 tons of spent lime A⁻¹ (Table 2). These results are explained by a significant interaction ($P = 0.016$) in stand in non-limed control plots, which was significantly higher for the *Aphanomyces*-susceptible variety HM 2467RZ treated with 45 g of Tachigaren than for the *Aphanomyces*-resistant variety Seedex Alpine with no Tachigaren (data not shown). This interaction did not occur in limed plots (data not shown). Thus, the benefit of sowing Tachigaren-treated seed of a susceptible variety in non-limed plots was so effective, it obscured the positive effect of spent lime on maintaining seedling stands of both varieties.

Sugarbeet stands were the same across all plots after thinning but considerable stand loss occurred over the rest of the season (Table 2). At harvest, all rates of spent lime resulted in significantly higher stands than the non-limed control. Among spent lime treatments, stands were significantly highest and equal in plots treated with 10 and 15 tons of lime, lowest at 5 tons, and intermediate at 20 tons. In the non-limed control, *Aphanomyces* root rot ratings averaged 4.7 (= 50 to 75% of the root surface was constricted, rotted, and/or scarred) and were significantly higher than in limed plots which averaged a rating of 3.4 (= 25% of root surface was affected by disease). Among lime treatments, there were no significant differences in *Aphanomyces* root rot ratings but the 5 ton rate resulted in somewhat more root rot than the higher rates of spent lime.

Sugarbeet root yields were significantly higher for all rates of spent lime compared to the control; among lime treatments, yields were significantly higher in the 15 ton plots compared to 5 tons and were intermediate for 10 and 20 tons A⁻¹ (Table 2). All rates of spent lime resulted in significant and equal increases in percent sucrose, pounds of sucrose per ton, and gross return compared to the control. Although all rates of lime significantly increased pounds of recoverable sucrose A⁻¹ compared to the non-limed control, there were differences among lime treatments. A significantly higher amount of sucrose was recovered from plots treated with 15 tons of spent lime compared to 5 tons; amounts were intermediate in the 10 and 20 ton A⁻¹ plots. Regression analysis confirmed significantly highest recoverable sucrose in plots treated with 15 tons wet weight of lime A⁻¹ ($R^2 = 0.9785$).

The *Aphanomyces*-resistant variety (Seedex Alpine) resulted in significantly higher stands at 13 days after planting than the susceptible variety (Hilleshog 2467RZ) but there were no differences in stand or *Aphanomyces* root rot between the two varieties for the rest of the season (Table 2). Yet, Seedex Alpine resulted in significantly higher tons of roots, pounds of recoverable sucrose, and gross return A⁻¹ than the susceptible variety.

Aphanomyces soil index values (SIVs). Hillsboro. For soil samples collected in May, 2006, *Aphanomyces* SIVs varied depending on 2005 crop history (data not shown). For instance, 2006 SIVs were very high (= 95) in plots sown to sugarbeet in 2005. On the other hand, 2006, SIVs in

the remaining limed and non-limed experiments (fallowed in 2005) were the same and averaged 61. Overall, *Aphanomyces* SIVs were higher in 2006 than in 2004. In 2004, 9 months after spent lime was applied, the *Aphanomyces* SIV in the non-limed control was 45 and across limed plots averaged 20 (9).

Breckenridge. *Aphanomyces* SIVs were extremely high and averaged nearly 100 (data not shown) for soil samples collected in April, 2006, regardless of 2005 cropping history (sugarbeet or wheat). In 2004, 5 months after spent lime was applied, *Aphanomyces* SIVs in the non-limed control averaged 100 and across limed plots averaged 82 (9).

DISCUSSION

Application of spent lime, two growing seasons before planting sugarbeet in 2006, significantly increased sucrose yields and economic returns at both locations, despite no *Aphanomyces* disease pressure at Hillsboro and severe *Aphanomyces* root rot at Breckenridge. Similar results were reported in 2005, one growing season after spent lime was applied (10). Although soil index values (SIVs) at both locations indicated high potential for disease in 2006, soil moisture was low at Hillsboro, so *A. cochlioides* was inactive. On the other hand, *A. cochlioides* was active early in the growing season at Breckenridge and wet soil conditions occurred intermittently until harvest. In 2006, the *Aphanomyces*-resistant variety was superior to the susceptible variety for most harvest data measured at both locations, a trend that also was observed in 2005 (10). This illustrates the excellent yield potential of an *Aphanomyces*-resistant variety grown in the absence, or presence, of disease pressure.

The pH of lime-amended plots increased compared to non-limed controls at both locations, although pH has not changed at Hillsboro since 2004 and at Breckenridge has increased only slightly since 2004 (9). Severe *Aphanomyces* root rot, however, occurs naturally in fields over a wide range of pH values (5 to 8) in Minnesota and North Dakota. Improved production of sugarbeet (2,10) by soil-application of spent lime may be attributed to increases in soil pH, which alters availability of micronutrients to the root and/or favors increases of beneficial microorganisms in the rhizosphere (4,6,7,8). Spent lime also contains nitrogen, phosphorus, potassium, and other inorganic and organic nutrients (5) that directly fertilize crops. Additionally, spent lime alters physical properties of the soil, e.g., improving water drainage, which results in less *Aphanomyces* root rot.

Aphanomyces SIVs were surprisingly high in June, 2006 in all limed and non-limed plots at both locations, despite SIVs dropping within a few months after spent lime was applied in 2004. In 2005, SIVs remained low in limed plots - except where sugarbeet was sown, where they returned to pre-limed levels (10). It is unknown why sugarbeet (and in 2006, soil fallowed or planted to wheat in 2005) negated earlier suppression of *Aphanomyces* SIVs. Perhaps lime suppresses germination of oospores (survival spores that produce infective zoospores) of *A. cochlioides* and this inhibition is overcome when crop roots release exudates into soil (including rotation crops which are non-hosts of *A. cochlioides*). This theory, however, does not explain why planting sugarbeet and rotation crops in limed soil returned SIVs to pre-limed levels at Breckenridge and to higher than pre-limed levels at Hillsboro, yet yields of sugarbeet increased at both locations. *Aphanomyces* SIVs in fields also may vary over time because of changing environmental conditions and their effects on survival structures of *A. cochlioides*.

In summary, application of spent lime two growing seasons before planting sugarbeet in 2006 significantly increased recoverable sucrose and economic return at two locations, despite no *Aphanomyces* disease pressure at Hillsboro and severe *Aphanomyces* root rot at Breckenridge. When *A. cochlioides* was inactive, sucrose yields significantly improved with a lime application of 5 tons wet weight A⁻¹ (= 3.3 tons dry weight) or higher compared to the non-limed control. When *A. cochlioides* was active increasing rates of lime tended to decrease root rot and increased sugarbeet yields; 15 tons wet weight (= 8 ton dry weight) spent lime A⁻¹ was optimal; 10 tons wet weight gave better results than 5 ton wet weight A⁻¹. Within months after spent lime was applied, *Aphanomyces* soil index values (SIVs) decreased compared to non-limed controls. Two growing seasons later, SIVs in all plots (limed and non-limed) increased to pre-limed levels or higher in plots sown to sugarbeet as well as rotation crops.

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