ADDITION OF SUPPLEMENTAL SPENT LIME TO PREVIOUSLY LIMED SOILS FOR CONTROL OF APHANOMYCES ROOT ROT ON SUGARBEET

Jason R. Brantner[1] and Ashok K. Chanda[2]

[1]Senior Research Fellow, [2]Assistant Professor and Extension Sugarbeet Pathologist
University of Minnesota, Department of Plant Pathology, St. Paul and Northwest Research and Outreach Center, Crookston

Aphanomyces cochlioides (= *A. cochlioides*) is an economic pathogen infesting over 90% of township sections planted to sugarbeet in the Red River Valley (RRV) and many fields in southern Minnesota. The pathogen is favored by warm and wet soil conditions and can cause damping-off, root rot, and chronic root rot of older plants. *Aphanomyces cochlioides* survives in soil for years, even when sugarbeet is not planted. Recommendations for growing sugarbeet in infested fields include early planting of partially resistant varieties treated with the fungicide Tachigaren and implementing cultural practices (e.g. cultivation and improved drainage) to avoid or reduce disease. When inoculum densities of the pathogen are high and soil is wet and warm, however, these measures are inadequate for economic yields - and fields may be abandoned or yield poorly. This chronic disease has generated interest in finding effective and alternative methods to supplement control of *A. cochlioides*.

The sugar purification process results in the by-product “spent lime”. Lime (calcium carbonate) precipitates impurities in sugarbeet juice. Purified juice is further processed into crystalized sugar, but spent lime (14% less acid neutralizing power than fresh lime) contains impurities and becomes a factory 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. Information on uses of 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”.

There is limited information in the literature on use of spent lime to reduce plant diseases. Campbell and Greathead (3) applied spent lime (2 to 4 tons A^{-1}) from a sugarbeet processing factory to fields (pH < 6.8) severely infested with the clubroot pathogen, *Plasmodiophora brassicae,* in the Salinas Valley, California. A single application gave “virtually complete control” of clubroot of crucifer crops grown repeatedly 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 inconsistent and little is known about how various forms of lime affect the pathogen. Sugarbeet growers in southern Minnesota apply spent lime the year before planting sugarbeet (typically every 3 years) and have observed less Aphanomyces root rot. In the RRV, reduction of *Aphanomyces* on sugarbeet by addition of spent lime to soil has been well documented, even eleven years after application (1,2,7).

OBJECTIVES

Our objectives were to evaluate the benefit of adding an additional 5 ton A^{-1} spent lime to a field trial where several rates of spent lime had been applied ten years earlier for: 1) effect on Aphanomyces diseases and 2) sugarbeet yield and quality.

MATERIALS AND METHODS

Establishment of field trials. Trials were originally established in a grower’s field near Breckenridge, MN (pH = 6.3) in April, 2004. At that time, the Breckenridge field plot area had a history of severe Aphanomyces root rot with a soil index value (SIV) of 98 (0 to 100 scale, 0 = no disease, 100 = potential for severe disease).

The site was divided into four, 1-acre experiments. Experiments included four rates of spent lime and a non-limed control in a randomized block design of four replicates (Fig. 1). Spent lime treatments were 0, 5, 10, 15 and 20 tons fresh weight A^{-1} (= 0, 2.7, 5.3, 8, and 10.6 tons dry weight, respectively); each plot was 33 x 60 ft. To allow lime treatments to stabilize in 2004, spring wheat was sown. Sugarbeet has been grown in one experiment each year

Experiment 1

5	0	10	20	15
10	5	20	15	0
10	15	20	0	5
0	5	10	15	20

Experiment 2

0	15	20	10	5
20	5	0	10	15
5	10	15	20	0
0	5	10	15	20

Experiment 3

10	15	0	5	20
15	5	20	0	10
20	10	15	0	5
0	5	10	15	20

Experiment 4

15	5	20	0	10
5	0	10	15	20
20	0	15	5	10
0	5	10	15	20

Breckenridge, MN (illustrated above)

Crop sown/yr:	2004	wheat	wheat	wheat	wheat
	2005	**sugarbeet**	wheat	wheat	wheat
	2006	soybean	**sugarbeet**	soybean	soybean
	2007	wheat	wheat	**sugarbeet**	wheat
	2008	corn	corn	corn	**sugarbeet**
	2009	**sugarbeet**	soybean	soybean	soybean
	2010	corn	**sugarbeet**	corn	corn
	2011	soybean	soybean	**sugarbeet**	soybean
	2012	wheat	wheat	wheat	**sugarbeet**
	2013	**sugarbeet**	soybean	soybean	soybean
	2014	corn	**sugarbeet**	soybean	corn
	2015	soybean	soybean	**sugarbeet**	soybean
	2016	corn	corn	**sugarbeet**	corn

Fig. 1. Four experiments were established at Breckenridge, MN in April 2004. Each experiment was treated with spent lime at 0, 5, 10, 15 and 20 tons fresh weight A^{-1}; experiments were arranged in a randomized complete block design with four replications (illustrated above). Sugarbeet plots are noted in **bold** the year when planted in an experiment; in 2015, sugarbeet test trials were planted in Experiment 3.

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from 2005 to 2016; the other experiments were planted with the same crop grown in the field by the grower-cooperator as noted in Fig. 1. In 2014, experiment 3 was planted to soybean. On October 31, 2014, original lime plots from the experiment 3 site in Fig. 1 were split in half and 5 ton A^{-1} fresh weight (3.5 tons dry weight) spent lime from the Minn-Dak Farmers' Cooperative sugar factory in Wahpeton, ND were spread in half of each plot. The lime was worked into the soil by a chisel plow on November 11. The experiment was planted to sugarbeet in 2015.

2016 Sugarbeet field trial. The same experiment site planted to sugarbeet in 2015 was planted to sugarbeet again in 2016. The Roundup Ready, Aphanomyces-susceptible sugarbeet variety 'BTS 82RR33' was sown on May 19. This was the fourth time Experiment 3 had been planted to sugarbeet since the original application of lime. The variety had a two-year Aphanomyces disease rating of 5.6 (1-9 scale) (5) and seed was treated with Kabina ST (14 g a.i./unit) but not with Tachigaren. Approval as an Aphanomyces specialty variety requires a 2-year rating of <= 4.4 (5). Seed was sown every 4.7 inches in rows 60-feet long and 22-inches apart (six rows of each variety centered within split plots). Twenty-foot alleys were cut later leaving 40-ft plots. Experiments followed standard fertility and production practices to obtain maximum sucrose yield and quality.

Stand counts were made 2, 4, 7, and 12 weeks after planting. Sugarbeet roots were harvested October 3 (two middle rows per subplot). Twenty roots per subplot were rated for Aphanomyces root rot (0 to 7 scale, 0=healthy and 7=root completely rotted and foliage dead). Ten roots were randomly selected and analyzed for yield and sucrose quality by the American Crystal Sugar Company Quality Laboratory, East Grand Forks, MN.

2016 Soil pH and Aphanomyces soil index values (SIVs). Soil samples were collected from subplots of supplemental lime and original lime in April, 2016. Five samples (6-inch depth) were collected randomly across each plot, combined, screened through 0.25-inch hardware cloth, and assayed for Aphanomyces soil index value. Subsamples were sent to Agvise Laboratories, Northwood, ND for pH, calcium, and other nutrient level determination.

Bioassays to determine Aphanomyces soil index values (SIVs, which indicate potential for *Aphanomyces* activity and populations under warm and wet conditions) were conducted by filling four (4 x 4 x 4-inch) plastic pots with soil from each sample. Then 25 seed of sugarbeet 'Crystal 985' were sown per pot to "bait" *A. cochlioides* from soil. Pots were placed in a growth chamber and arranged in a randomized block design at 70 $\pm$ 2 °F for 1 week. Temperature then was increased to 77 $\pm$ 2°F (14-hour photoperiod) and soil was kept moist to favor infection by *A. cochlioides*. Stand counts were made three times per week (beginning at emergence) and dying seedlings were removed to prevent disease spread, assayed in the laboratory, and examined microscopically to verify infection with *A. cochlioides*. At 4 weeks after planting, an Aphanomyces SIV was calculated (0 to 100 scale, 0 = *Aphanomyce*s-free and 100 = all seedlings dead and soil severely infested with *A. cochlioides*) for each pot.

Statistical analysis. Data were analyzed for effect of supplemental lime by analysis of variance and for effect of original lime rate and supplemental lime by original lime interactions using linear and quadratic contrasts for significance at P = 0.05, 0.01, and 0.001. Regression analyses were made for recoverable sucrose per acre and Aphanomyces root rot rating vs. original rate of lime. Correlation coefficients were determined for recoverable sucrose per acre and Aphanomyces root rot rating with soil calcium concentration.

RESULTS

2016 Sugarbeet field trial. There were no significant (P = 0.05) interactions between supplemental lime and rate of original lime for early season stand and all harvest parameters so results are presented for main effects of original lime and supplemental lime in Table 1. Averaged across both supplemental lime treatments, Aphanomyces root rot decreased with increasing original lime rates, while stand at 48 days after planting, number of harvested roots, root yield and quality all increased with increasing rate of original lime (Table 1). Averaged across all rates of original lime, addition of 5 ton supplemental lime A^{-1} resulted in significantly lower Aphanomyces root rot ratings and significantly higher early season stand, number of harvested roots, root yield and quality (Table 1).

Twelve years after application, original lime was still highly effective in reducing Aphanomyces root rot and increasing sugar beet yield. In plots without supplemental lime application, there was significant (P = 0.05) linear effect of original lime rate on all early season stand counts, Aphanomyces root rot ratings (Fig. 2B), and all harvest parameters (illustrated for recoverable sucrose A^{-1} in Fig. 2A).

2016 Soil pH, extractable calcium, and Aphanomyces soil index values (SIVs). In soil samples collected April, 2016, 12 years after original lime was applied and 18 months after additional lime was applied to half of each original lime plot, there was no significant original lime by supplemental lime interaction for Aphanomyces SIV. Aphanomyces SIVs were high in soil samples from all plots, but there was a significant (P = 0.05) linear response to original rate of lime (Table 1). Addition of 5 ton A^{-1} supplemental lime did not significantly reduced Aphanomyces SIV compared to those without supplemental lime (Table 1).

There was significant original lime by supplemental lime interaction for soil pH. Increase in soil pH by the addition of supplemental lime became less as original lime rate increased (data not shown). There was no significant original lime by supplemental lime interaction for soil extractable calcium. There was significant linear response to original lime rate so that soil calcium levels increased with increasing rates of original lime (Table 1). Supplemental lime also significantly increased soil extractable calcium across all original lime rates (Table 1). In soil from plots without supplemental lime, there was a significant linear effect of original lime rate on soil pH and extractable calcium 12 years after original lime was applied (data not shown). There was a strong negative correlation (r = -0.826) between soil extractable calcium and Aphanomyces root rot (Fig. 3A) and a strong positive correlation (r = 0.840) between soil extractable calcium and sugarbeet yield and quality (illustrated for recoverable sucrose A^{-1} in Fig. 3B).

Table 1. Soil pH, Aphanomyces soil index values, stands, root rot ratings, and harvest data for sugarbeet sown May 19, 2016, 12 years after several rates of spent lime (original lime) were applied and 19 months after 5 ton/A supplemental lime was applied in a field naturally infested with *Aphanomyces cochlioides.*

Main treatments	Soil pH[T]	Aph SIV (0-100)[U]	Soil extractable Ca[T]	Stand@ 48 DAP (plants/100 ft)[V]	No. root harvested/ 100 ft	Aph RRR (0-7)[W]	Yield (ton/A)	Sucrose %	Sucrose lb/ton	Sucrose lb/recov/A
Original lime (ton/A)[X]										
Wet wt. Dry wt.										
0 0	7.4	100	3593	76	63	5.3	15.7	12.4	211	3327
5 2.7	7.6	100	4073	105	93	3.9	21.0	12.4	215	4518
10 5.3	7.7	100	4388	117	108	3.3	21.8	12.6	220	4799
15 8.0	7.9	100	4747	154	142	1.6	24.5	13.1	231	5654
20 10.6	7.9	99	5268	167	152	1.2	25.0	13.1	234	5868
Linear[Y]	***	**	***	***	***	***	***	**	**	***
Quadratic[Y]	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Supplemental lime (ton/A)[Z]										
Wet wt. Dry wt.										
0 0	7.6	100	4132	115	101	3.5	20.3	12.6	220	4507
5 3.5	7.8	100	4696	133	122	2.6	22.9	12.8	224	5160
P-value[Y]	***	NS	***	**	**	**	**	NS	NS	**
Interaction										
Lime x Var (linear)[Y]	**	NS	NS	NS	NS	NS	NS	NS	NS	NS

T Soil pH and extractable calcium (Ca) values from Agvise Laboratories, Northwood, ND.

U Aphanomyces SIV = soil index value (determined in a 4-week greenhouse assay of soil); 0-100 scale where 0 = soil *Aphanomyces*-free, 100 = all seedlings dead by 4 weeks after planting and soil severely infested with *Aphanomyces*.

V DAP = days after planting; plots were sown at 61,400 seed/A (seed every 4.65 inches in rows 22 inches apart).

W RRR = Aphanomyces root rot rating, 0-7 scale (0 = roots healthy; 7 = root completely rotted and foliage dead).

X Original 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 2016, sugarbeet was sown on May 19, 12 years after original spent lime had been applied; each value in this portion of the table is averaged across two supplemental lime treatments (0 or 5 ton/A). Plots were harvested on October 3.

Y * = significant at $P = 0.05$, ** = significant at $P = 0.01$, *** = significant at P=0.001, NS = not significant.

Z On October 31, 2014, plots were split and supplemental lime (5 ton/A) was applied to half of each original lime plot. Each value in this portion of the table is averaged across all original lime treatments.

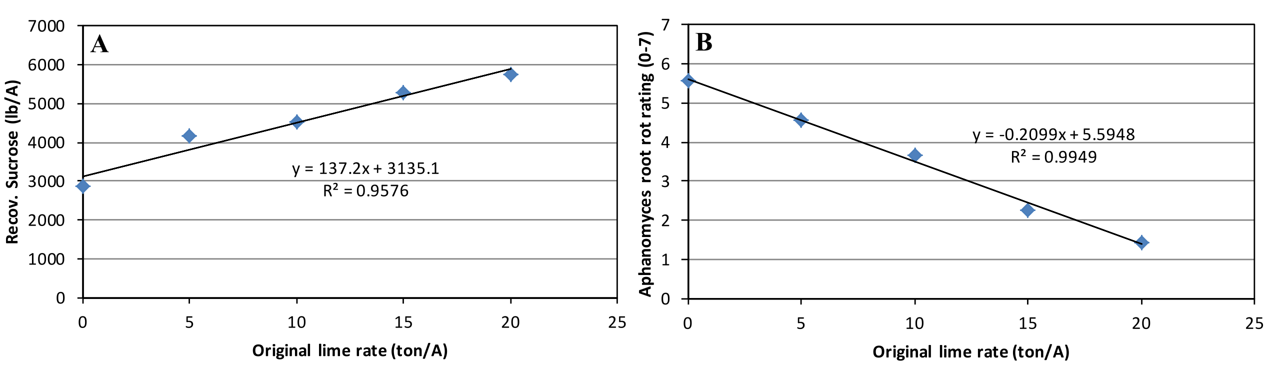

Fig. 2. Regression of **A)** recoverable sucrose and **B)** Aphanomyces root rot rating vs. original rate of lime per acre (with no supplemental lime) in a grower's field near Breckenridge, MN, naturally infested with *Aphanomyces cochlioides*. Plots were amended with original lime in April, 2004. Data is from sugarbeet sown May 19, 2016 (12 years after spent lime application) and harvested October 3, 2016.

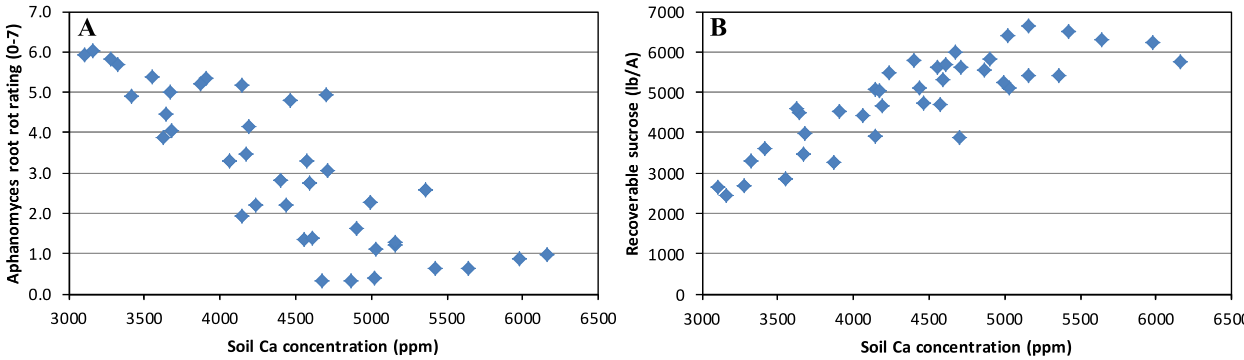

Fig. 3. Scatter plots of **A)** Aphanomyces root rot rating and **B)** recoverable sucrose and soil extractable calcium (Ca) concentration for 40 plots in a grower's field near Breckenridge, MN, naturally infested with *Aphanomyces cochlioides*. Plots were amended with original lime in April, 2004 and supplemental lime on October 31, 2014. Data is from sugarbeet sown May 19, 2016 (12 years after original lime and 19 months after supplemental lime application) and harvested October 3, 2016. Correlation coefficients for Aphanomyces root rot rating and recoverable sucrose with soil calcium concentration were -0.826 and 0.840, respectively, both significant at P = 0.01.

DISCUSSION

Growing sugarbeet in back-to-back seasons on site 3 of the experiment, late planting (May 19), and adequate early-season plus excessive mid- to late-season rainfall resulted in severe Aphanomyces disease pressure. Stand in plots with no original or supplemental lime application declined from 177 plants per 100 feet of row on June 2 to just 75 plants per 100 feet of row on July 6 (a 58% reduction), indicating high disease pressure (data not shown). Rainfall at the Wahpeton North Dakota Agricultural Weather Network station in July, August, and September was 4.85, 3.88, and 3.74 inches, respectively, higher than normal. As a result, Aphanomyces was active for much of the season and yields were severely impacted across all treatments. Even under this severe disease pressure, original and supplemental lime significantly reduced Aphanomyces root rot and increased sugarbeet stands, yield, and quality (Table 1).

In 2015, results could not justify application of supplemental lime to soils where lime had previously been applied at 10 ton A^{-1} or higher. In contrast, results from 2016 suggest a benefit from application of supplemental lime, regardless of previous lime application rate. The 2016 results are likely influenced by the severe disease pressure and back-to-back sugarbeet crop described above. Analysis of soils from sugarbeet fields in Sweden showed negative correlation between Aphanomyces severity index and soil calcium levels (6). Similarly, addition of calcium to soils reduced Aphanomyces root rot of pea in Sweden (4). At the Breckenridge field trial site, the strong positive correlation between soil extractable calcium and sugar beet yield and quality and negative correlation between soil extractable calcium and Aphanomyces root rot corroborate results from Sweden and suggest that building soil calcium levels through addition of lime may be an effective strategy for reducing Aphanomyces in Minnesota and North Dakota.

Use of the highly susceptible variety without Tachigaren seed treatment in this trial was intended to increase the likelihood of seeing differences from lime treatments. Data from previous trials over several years, with various levels of disease severity, utilizing both a resistant variety treated with Tachigaren and a susceptible variety without Tachigaren indicate better results with a resistant variety treated with Tachigaren. It is also worth noting that application of lime cannot be expected to protect a sugarbeet variety with a low level of resistance to Aphanomyces, especially if the conditions are highly favorable for disease development.

SUMMARY AND CONCLUSIONS

1. Across all rates of original lime, supplemental lime applied at 5 ton A^{-1} significantly decreased Aphanomyces root rot and increased sugarbeet stand, yield, and quality.
2. Both original and supplemental lime applications significantly increased soil extractable calcium, which was negatively correlated with Aphanomyces root rot and positively correlated with sugar yield.
3. Twelve years after application, original spent lime rates with no supplemental lime significantly reduced Aphanomyces root rot and increased sugarbeet stand, yield, and quality.
4. Building soil calcium levels by application of spent lime may be an effective strategy for reducing Aphanomyces in sugarbeet, but data is not available on how much soil calcium is needed versus too much.

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