FIVE-YEAR EFFECT OF A SINGLE FIELD APPLICATION OF VARIOUS RATES OF SPENT LIME ON APHANOMYCES, SUGARBEET AND ROTATON CROPS

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Aphanomyces cochlioides (= A. cochlioides) is a serious economic pathogen that 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, even when sugarbeet is not planted. Consequently, growing sugarbeet in infested fields requires all available control options including early planting of seed of partially resistant varieties treated with the fungicide Tachigaren and implementing various cultural practices (e.g. cultivation and improved drainage) to avoid or reduce infection by A. cochlioides. However, when inoculum densities of the pathogen are high and soil is wet, implementing 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 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". In the Salinas Valley of California, Campbell and Greathead (3) applied spent lime (2 to 4.5 tons A⁻¹) from a sugarbeet processing factory to fields (baseline pH less than 6.8) that were severely infested with the clubroot pathogen, *Plasmodiophora brassicae*. 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 erratic and little is known about how various forms of lime affect the pathogen.

Producers 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 thereby, 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 - and less Aphanomyces root rot was observed. Consequently, growers in southern Minnesota have continued to apply spent lime the year before planting sugarbeet (typically every 3 years).

To determine the effect of spent lime on reducing Aphanomyces root rot in the RRV, Bresnahan et al. (2) applied 3 and 10 tons wet weight A^{-1} in two *Aphanomyces*-infested fields (baseline pH values of 5.9 and 7.8). Within 1 year, Aphanomyces root rot was significantly reduced and sucrose yields increased compared to the non-limed control. 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 <u>7 years earlier</u>; the remainder of the field had poor stand, stunted growth, and severe Aphanomyces root rot. These promising results have sparked interest in soil-application of spent lime in the RRV and raised questions about rates and duration of applications.

OBJECTIVES

Objectives are two-fold based on experiments where single soil-applications of spent lime (at different rates) were made in October, 2003/April, 2004: 1.) determine Aphanomyces root rot, yield and quality of sugarbeet and to measure yield of rotation crops sown in 2008 (fifth growing season after application) and 2.) evaluate the overall effect of initial lime applications on sugarbeet and rotation crops over five growing seasons.

MATERIALS AND METHODS

Establishment of field trials. Experiments were established in growers' fields near Hillsboro, ND (pH = 7.02) in October, 2003 and Breckenridge, MN (pH = 6.3) in April, 2004. The Hillsboro site has a history of moderate Aphanomyces root rot and the soil index value (SIV) averaged 48 (0 to 100 scale, 0 = no disease, 100 = potential for severe disease). The Breckenridge site has a history of severe Aphanomyces root rot and the SIV averaged 98.

Each site was divided into four, 1-acre experiments. Each experiment included four rates of spent lime and an untreated control and was replicated four times in a randomized block design (Fig. 1). Treatments at Hillsboro were 0, 5, 10, 20 and 30 tons wet weight spent lime A^{-1} (= 0, 3.3, 6.5, 13 and 19.5 tons dry weight, 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, respectively); each plot measured 33 x 60 ft. 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. Sugarbeet was grown in one experiment each year from 2005 to 2008; the three other experiments were planted with the same crop grown in the field by the grower-cooperator from 2004-2008 (Fig. 1). Results have been reported for sugarbeet and rotation crops at both locations from 2005 to 2007 (16, 17, 18).

2008 Sugarbeet field trials. Sugarbeet was sown in one of the four experiments at Hillsboro on May 5 and Breckenridge on May 13. Varieties 'HM 3028RZ' (partially resistant to *Aphanomyces* + 45 g of Tachigaren per unit of seed) and 'Crystal R431' (susceptible, no Tachigaren) were sown as subplots in lime-treated and control plots. Seed was sown every 2.5 inches in rows 60-feet long and 22-inches apart (six rows of each variety centered within each plot). An application of the herbicide Nortron (3.75 lb a.i. A^{-1}) was incorporated into soil 2 to 4 days before planting. The insecticide Counter 15G (12 lb product A^{-1}) was applied modified in-furrow at planting. After sugarbeet seedlings emerged, 10 feet were cut from the front and back of each plot, resulting in rows 40 feet long. Microrates of Betamix + UpBeet + Stinger + SelectMax + MSO (8-10 fl oz + 0.125-0.25 oz + 1.3 fl oz + 3 fl oz + 1.5% A^{-1}, respectively) were applied at Hillsboro on June 21 and 30 and July 9 and at Breckenridge on June 18 and July 9. Plots were cultivated on July 2 at Hillsboro and June 23 at Breckenridge. Sugarbeet was hand-thinned on July 12 to a 6-inch spacing at Hillsboro and on July 15 to a 4-inch spacing (based on history of Aphanomyces root rot) at Breckenridge. At both locations, Cercospora leaf spot was controlled with one application of Headline (9 fl oz A⁻¹) on August 18 (20 gpa at 100 psi). Alleys separating replicates were rototilled throughout the season.

Stand counts were made at 5 to 7 weeks after planting and after thinning. Experiments were harvested at Hillsboro on October 1 and Breckenridge on September 30 (two middle rows of each variety per treatment). Twenty roots from each 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 Co. Quality Laboratory, East Grand Forks, MN.

2008 Rotation crop field trials. At Hillsboro, the three spent lime experiments not planted to sugarbeet were sown to wheat 'Howard' on April 25 by the grower-cooperator. Population densities were estimated on August 22 by counting numbers of tillers per square meter in each plot. Root rot caused by *Fusarium* species and *Bipolaris sorokineae* were evaluated on August 25 by randomly selecting 25 plants per plot and rating the subcrown internode on a 0 to 3 scale (0=clean, 3=>50% necrotic). Plots were harvested with a small plot combine (Wintersteiger Seedmuch, Dimmelstrasse, Germany) on August 26 by removing a 5 x 20 ft swath. Yields were adjusted to 13.5% moisture and calculated based on 60 lb per bushel.

At Breckenridge, the three spent lime experiments not sown to sugarbeet were planted to corn 'ProSeed P92bt' on May 15 by the grower-cooperator. Plots were hand-harvested on October 29 by removing ears from 10 ft of the two center rows of each plot. Yields were adjusted to 15.5% moisture and calculated based on 56 lb per bushel.

Soil pH and Aphanomyces soil index values (SIVs) 2003-2008. Soil samples were collected from all plots at Hillsboro on October, 2003; July and October, 2004; June and October, 2005; June, 2006; May, 2007; and April 2008. They also were collected at Breckenridge in April and September, 2004; June and October, 2005; May, 2006; June, 2007; and May, 2008. For each sampling date and location, 80 soil samples were collected, except in the spring of 2006 and 2007, when 100 plots were sampled to include subplots sown to the partially resistant and susceptible sugarbeet varieties the previous year. Six soil cores (2.5-inch diameter x 6-inch depth) were collected

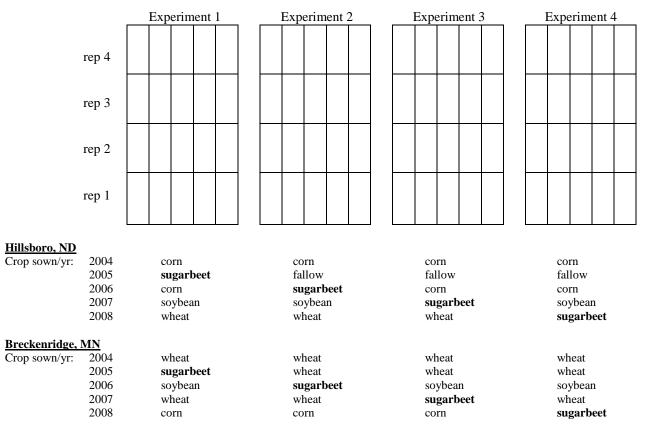


Fig. 1. Four experiments were established at Hillsboro, ND in October, 2003 and at Breckenridge, MN in April 2004. At Hillsboro, each experiment was treated with 0, 5, 10, 20 and 30 tons wet weight spent lime A⁻¹ and at Breckenridge with 0, 5, 10, 15 and 20 tons wet weight spent lime A⁻¹; experiments were arranged in a randomized block design and replicated four times. In 2004, all experiments were sown to corn at Hillsboro and wheat at Breckenridge to stabilize soil pH. In subsequent years, sugarbeet was planted in one of the experiments and other crops (noted above) were sown in the other three experiments.

randomly across each plot, combined, screened through 0.25-inch hardware cloth, and stored in a walk-in cooler until assayed (usually within 1 month after collection).

To determine soil pH, small quantities of soil were air-dried at least 24 hours 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). Soil pH was determined from samples collected at Hillsboro, as described above, on July, 2004; June, 2006; May, 2007; and April, 2008 and at Breckenridge on October 2004; May, 2006; June, 2007; and May 2008.

Bioassays to determine Aphanomyces soil index values (SIVs, which indicate potential for *Aphanomyces* activity and populations) were conducted by filling four (4 x 4 x 4-inch) plastic pots with soil from each sample. Then 25 seed of sugarbeet 'ACH 261' were sown per pot (to "bait" *A. cochlioides* from soil). Pots were placed in a controlled environment chamber in a randomized block design at 70 ± 2^{0} F for 1 week for optimal emergence. Temperatures then were increased to 79 ± 2^{0} F (14-hour photoperiod) and soil was kept moist to favor infection by *A. cochlioides*. Stand counts were made three times weekly beginning at emergence. Dying seedlings were removed at each stand count to prevent disease from spreading to adjacent plants. At 4 weeks after planting, surviving seedlings were rated for disease on a 0 to 3 scale (0 = healthy, 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*).

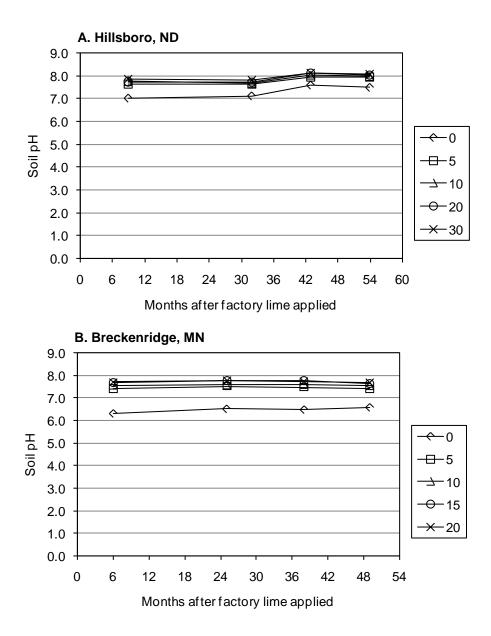
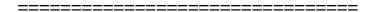


Fig. 2. Average soil pH values in plots at: A.) Hillsboro, ND treated with 0, 5, 10, 20, and 30 tons wet weight of spent lime A⁻¹ in October, 2003; soil samples were collected in July, 2004; June, 2006; May, 2007; and April, 2008 at 9, 32, 43, and 54onths after application and at: B.) Breckenridge, MN treated with 0, 5, 10, 15 and 20 tons wet weight of spent lime A⁻¹ in April, 2004; soil samples were collected in October, 2004; May, 2006; June, 2007; and May, 2008 at 6, 25, 38, and 49 months after application of spent lime, respectively. Each data point is an average of 16 plots.



Data analysis. Sugarbeet data for 2008, the overall 2005-2008 summary for sugarbeet, and the 2004-2008 summary for rotation crops were analyzed for effect of lime rate by linear and quadratic contrasts for significance at P = 0.05 and 0.01.

RESULTS

2008 Sugarbeet field trials: *Hillsboro*. In May, 2008 (55 months after spent lime was applied), soil pH was 7.49 in the non-limed control and was significantly higher (near pH of 8) in limed plots, regardless of rate (Table1). Soil pH values tended to increase slightly in the limed and non-limed control plots since they were first measured in July, 2004 (9 months after spent lime was applied) but overall, remained similar every year (Fig. 2A).

 Table 1. Hillsboro, 2008: Soil pH and stands, root rot ratings, and harvest data of sugarbeet sown on May 5, 2008, 55 months after several rates of spent lime were applied in a field naturally infested with moderate inoculum densities of *Aphanomyces cochlioides*.

					ts/80-ft row er planting) ^x	No. roots Harvested/	RRR	Yield		Sucr	ose	Gross return
Main t	Main treatments		35	43	Post-thinning	80 ft row	$0-7^{Y}$	(Ton/A)	%	lb/T	lb recov./A	(\$/A)
Lime (T	`on/A) ^v											
Wet wt.	Dry wt.											
0	0	7.49	165	204	102	89	2.6	24.6	16.7	309	7595	967
5	3.3	7.96	184	208	98	90	2.0	26.0	16.8	309	8046	1027
10	6.5	7.97	175	203	96	88	2.1	25.8	16.4	301	7771	968
20	13.0	8.02	180	205	98	91	2.1	27.0	16.6	304	8194	1030
30	19.5	8.04	187	210	97	87	2.0	25.8	16.6	305	7866	991
	Linear ^Z	**	*	NS	NS	NS	**	NS	NS	NS	NS	NS
	Quadratic ^z	**	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Variety	N											
•	Crystal R431 (0 Tach)		183	205	96	84	2.2	26.0	16.7	306	7956	1007
HM 302 + 45 g		-	173	207	101	94	2.2	25.7	16.5	305	7833	986
	Contrast ^z		NS	NS	*	**	NS	NS	NS	NS	NS	NS

^v Spent lime was applied in October, 2003 in a randomized block design of four replicates per experiment (total of four experiments) and incorporated by cultivation. In 2008, sugarbeet was sown 55 months after spent lime had been applied; each value in this portion of the table is averaged across two sugarbeet varieties.

^w Sugarbeet varieties Crystal R431 (susceptible to *Aphanomyces*) and Hilleshog 3028RZ (partially resistant to *Aphanomyces* and treated with 45 g of Tachigaren [Tach] per unit of seed) were sown as subplots within each plot. Plots were harvested on October 1, 2008. Each value in this portion of the table is averaged across all lime treatments.

- ^X Plots were sown at 114,048 seeds per acre (seed every 2.5 inches in rows 22 inches apart) and hand-thinned to a 6-inch spacing on July 12. Post-thinning stand counts were made on July 29.
- ^Y RRR = Aphanomyces root rot rating, 0-7 scale (0 = roots healthy; 7 = root completely rotted and foliage dead).

^Z Orthogonal contrast, * = significant at P = 0.05, ** = significant at P = 0.01, NS = not significant.

There were no significant interactions between rate of lime and variety for any variable, so results are presented separately for these main effects (Table 1). Emergence was delayed because of dry soil. At 5 weeks after planting there was a significant linear effect of lime rate on stand but these differences disappeared about 1 week later and after thinning. By harvest, stands had declined about 10% and there were no significant differences among treatments. *Aphanomyces* caused some disease and root rot ratings (RRR) were significantly higher in the non-limed control (RRR = 2.6) compared to limed plots (average RRR = 2.1). A rating of 3 = 6 to 25% of the root scarred or rotted and a rating of 2 = superficial scarring on less than 5% of root surface. There were no significant differences among limed and non-limed control plots for root yield, percent sucrose, pounds of sucrose per ton, and for pounds of recoverable sucrose and gross return A⁻¹. There was a general trend for yield and pounds of recoverable sucrose and gross return A⁻¹ to be higher in limed plots than in the control.

The sugarbeet variety with partial resistance to *A. cochlioides* (HM 3028RZ) had significantly higher stands than the susceptible variety (Crystal R431) after thinning and at harvest (Table 1). There were no significant differences between the varieties for root yield, sucrose, and gross return.

 Table 2. Breckenridge, 2008:
 Soil pH, stands, root rot rating, and harvest data of sugarbeet sown on May 13, 2008, 49 months after several rates of spent lime were applied in a field naturally infested with high inoculum densities of *Aphanomyces cochlioides*.

			-	plants/80-ft row s after planting) ^X	No. roots Harvested/	RRR	Yield		Suci	rose	Gross return
Main treatments		Soil pH	49	Post-thinning	80 ft row	$0-7^{Y}$	(Ton/A)	%	lb/T	lb recov./A	(\$/A)
Lime (T	`on/A) ^v										
Wet wt.	Dry wt.	-									
0	0	6.58	155	89	78	3.8	21.0	14.5	263	5546	595
5	2.7	7.40	176	105	99	3.0	27.4	15.0	274	7514	844
10	5.3	7.55	176	106	98	3.0	27.7	14.7	265	7326	788
15	8.0	7.64	182	112	98	2.6	29.8	14.8	268	8000	876
20	10.6	7.68	184	105	99	2.9	29.7	14.6	264	7860	845
	Linear ^Z	**	NS	**	**	**	**	NS	NS	**	**
	Quadratic ^z	**	NS	**	**	**	**	NS	NS	**	*
Variety	W										
Crystal (0 Tac	Crystal R431		193	111	98	3.0	28.7	14.9	270	7759	856
HM 302 + 45 g	28RZ	-	156	96	90	3.1	25.5	14.6	263	6740	723
	Contrast ^z		**	**	**	NS	**	**	**	**	**

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 2008, sugarbeet was sown 49 months after spent lime had been applied; each value in this portion of the table is averaged across both sugarbeet varieties.

^W Sugarbeet varieties Crystal R431 (susceptible to *Aphanomyces*) and Hilleshog 3028RZ (partially resistant to *Aphanomyces* and treated with 45 g of Tachigaren [Tach] per unit of seed) were sown as subplots within each plot. Plots were harvested on September 30, 2008. Each value in this portion of the table is averaged across all lime treatments.

- ^X Plots were sown at 114,048 seeds per acre (seed every 2.5 inches in rows 22 inches apart) and hand-thinned to a 4-inch spacing on July 15. Post-thinning stand counts were made on July 29.
- ^Y RRR = Aphanomyces root rot rating, 0-7 scale (0 = roots healthy; 7 = root completely rotted and foliage dead).
- ^Z Orthogonal contrast, * = significant at P = 0.05, ** = significant at P = 0.01, NS = not significant.

Breckenridge. In 2008 (49 months after spent lime was applied), soil pH was 6.58 in non-limed plots and was significantly higher (average = 7.56) in limed plots, regardless of rate (Table 2). Soil pH values of samples collected in 2008 were slightly higher in all limed and non-limed plots compared to October, 2004 (5 months after lime was applied) but overall, remained stable from year-to-year.

There were no significant interactions between lime and sugarbeet variety for most variables measured, so results for these main factors are presented in Table 2. Cold weather in May and June slowed emergence and delayed onset of Aphanomyces seedling disease. By 49 days after planting, there were no differences in stand in plots treated with lime (all rates) and the non-limed control. Then, warm temperatures and rainfall favorable for Aphanomyces infections developed, so plots were thinned to a 4-inch spacing on July 15 in anticipation of stand losses. When stand counts were made 2 weeks later (noted as "post thinning stand" in Table 2), plant populations were significantly higher in all limed plots compared to the control. Aphanomyces root rot continued to reduce stand until harvest and number of harvestable roots was lowest in the non-limed control and equally higher in plots treated with lime. Aphanomyces root rot was significantly lower in the limed plots and averaged a rating of 2.9 compared to the non-lime significantly increased root yield and pounds of recoverable sucrose and gross revenue A⁻¹ compared to the control. There were no differences, however, among any treatments for percent sucrose and pounds of sugar per ton.

				Yield (bu/A) ^Y		
Lime (Te	on/A) ^X	2004	2005	2006	2007	2008
Wet weight	Dry weight	Corn	Fallow	Corn	Soybean	Wheat
0	0	144	_	132	48	71
5	3.3	140	-	132	50	73
10	6.5	138	-	138	49	75
20	13.0	134	-	152	47	75
30	19.5	136	-	148	47	73
Linear orthog	gonal contrast ^Z	**		NS	NS	NS
Juadratic orthog	gonal contrast ^Z	NS		NS	NS	NS

 Table 3.
 Hillsboro, 2004-2008: Yield of rotation crops from 2004 to 2008 after several rates of spent lime were applied in October, 2003 in a field naturally infested with moderate inoculum densities of *Aphanomyces cochlioides*.

^X Spent lime was applied in October, 2003 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 corn. From 2005 to 2008, one experiment was sown to sugarbeet and in the other three experiments, the grower-cooperator planted and maintained another crop following standard production practices. Experiments were left fallow in 2005 because the field was exceedingly wet. In 2006, rotation experiments were sown with corn 'DKC35-02 (RR2/YGCB)', 2007 with soybean 'Wensman 2090', and in 2008 with wheat 'Howard'.

^Y Corn yields were adjusted to 15.5% moisture and based on 56 pounds per bushel; soybean and wheat yields were adjusted to 13% moisture and based on 60 pounds per bushel.

^Z Orthogonal contrast, * = significant at P = 0.05, ** = significant at P = 0.01, NS = not significant.

		Yield (bu/A) ^Y						
Lime (To	$(on/A)^{X}$	2004	2005	2006	2007	2008		
Wet weight	Dry weight	Wheat	Wheat	Soybean	Wheat	Corn		
0	0	67	36	44	50	164		
5	2.7	66	44	49	47	173		
10	5.3	65	45	49	51	160		
15	8.0	62	46	48	48	169		
20	10.6	61	46	53	49	179		
Linear orthog	gonal contrast ^Z	**	**	P = 0.003	NS	NS		
Quadratic orthog	gonal contrast ^Z	NS	NS	LSD = 4	NS	NS		

 Table 4.
 Breckenridge, 2004-2008: Yield of rotation crops from 2004 to 2008 after several rates of spent lime were applied in a field naturally infested with high inoculum densities of *Aphanomyces cochlioides*.

^x 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 'Knudsen'. From 2005 to 2008, one experiment was sown to sugarbeet and in the other three experiments, the grower-cooperator planted and maintained another crop following standard production practices. In 2005, rotation experiments were sown with wheat 'Knudsen', 2006 with soybean 'Pioneer 90M91', 2007 with wheat 'Agri-Pro Freyr', and in 2008 with corn ''ProSeed P92bt'.

Y Corn yields were adjusted to 15.5% moisture and based on 56 pounds per bushel; soybean and wheat yields were adjusted to 13% moisture and based on 60 pounds per bushel.

^Z Orthogonal contrast, * = significant at P = 0.05, ** = significant at P = 0.01, NS = not significant. In 2006, soybean yields were subjected to analysis of variance and if significant (P = 0.05), were separated by least significant difference (LSD).

There were no differences in Aphanomyces root rot between the partially resistant and susceptible varieties (Table 2). For all other variables measured (stand, root yield, sucrose, gross return), the susceptible variety (Crystal R431) significantly outperformed the partially resistant variety (HM 3028RZ).

2008 Rotation crop field trials. At Hillsboro, there were no significant differences among limed and non-limed plots for wheat plant populations, common root rot (data not shown) or yield (Table 3). At Breckenridge, application of spent lime had no significant effect on corn yield compared to the control (Table 4).

 Table 5. Hillsboro, ND (2005-2008): Average sugarbeet stands, root rot ratings, and harvest data from 2005 to 2008, the second to fifth growing season after several rates of spent lime initially were applied in October, 2003 in a field naturally infested with moderate inoculum densities of *Aphanomyces cochlioides*.

		No	. plants/80 ft.	row	RRR		Yield		Sucrose		
Main treatments		4 WAP ^w	Post- thinning	Harvested	0-7 ^x	LTM^{Y}	(Ton/A)	%	lb/T	lb recov./A	
Lime (To	on/A)										
Wet wt.	Dry wt.										
0	0	256	131	112	2.2	1.32	22.2	16.8	309	6827	
5	3.3	269	129	117	1.9	1.30	23.2	17.0	315	7333	
10	6.5	265	131	120	2.0	1.35	24.3	16.8	310	7490	
20	13.0	273	134	122	2.0	1.33	24.2	17.1	315	7562	
30	19.5	273	133	122	1.9	1.29	24.6	17.3	320	7896	
Lime line	ear ^z	**	NS	**	**	NS	**	*	*	**	
Lime qua	dratic ^Z	NS	NS	NS	NS	NS	NS	NS	NS	NS	

^w WAP = Weeks after planting.

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

 $^{\rm Y}$ LTM = Loss to molasses

^Z Orthogonal contrast, * = significant at P = 0.05, ** = significant at P = 0.01, NS = not significant.

 Table 6.
 Breckenridge, MN (2005-2008): Average sugarbeet stands, root rot ratings, and harvest data from 2005 to 2008, the second to fifth growing season after several rates of spent lime initially were applied in April, 2004 in a field naturally infested with high inoculum densities of *Aphanomyces cochlioides*.

		No. plants/80 ft. row			RRR		Yield		Sucrose		
Main treatments		4 WAP ^w	Post- thinning	Harvested	0-7 ^x	LTM ^Y	(Ton/A)	%	lb/T	lb recov./A	
Lime (To	on/A)										
Wet wt.	Dry wt.										
0	0	202	82	58	4.9	1.59	13.2	14.5	258	3459	
5	2.7	223	102	83	3.6	1.60	22.4	15.2	271	6120	
10	5.3	233	109	93	3.3	1.61	24.5	15.3	274	6730	
15	8.0	230	111	91	3.2	1.72	25.4	15.2	270	6898	
20	10.6	238	113	96	3.2	1.62	26.0	15.2	272	7076	
Lime line	ear ^z	**	**	**	**	NS	**	**	**	**	
Lime qua	dratic ^Z	*	**	**	**	NS	**	**	**	**	

^w WAP = Weeks after planting.

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

Y LTM = Loss to molasses

^Z Orthogonal contrast, * = significant at P = 0.05, ** = significant at P = 0.01, NS = not significant.

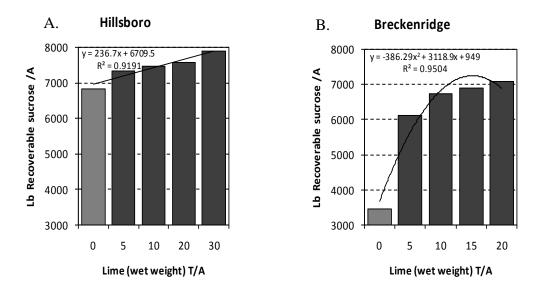


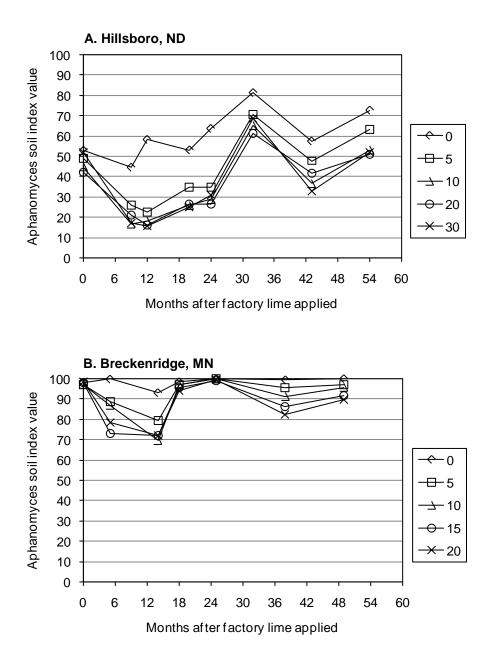
Fig 3. Yields of recoverable sugar averaged from 2005 to 2008 at **A.**) **Hillsboro**, **ND** after spent lime (0, 5, 10, 20, 30 tons wet weight A⁻¹) was applied in October, 2003 and **B.**) **Breckenridge**, **MN** after spent lime (0, 5, 10, 15, and 20 tons wet weight A⁻¹) was applied in April, 2004. The relationship between sugar yield and rate of lime is linear at Hillsboro and quadratic at Breckenridge.

Summary: Effect of a single application of spent lime on sugarbeet (2005-2008). At each location, data were combined for sugarbeet grown in 2005 to 2008. The variable weather conditions among growing seasons not only affected severity of Aphanomyces root rot, but sugarbeet quality and yield. Also, different varieties were planted most years. Thus, this summary is limited to effects of lime (various rates) compared to the non-limed control.

Hillsboro. Overall, application of lime in October, 2003 significantly decreased Aphanomyces root rot compared to the control (Table 5). Plots treated with lime also had significant increases in seedling stand, number of harvested roots, root yield, percent sucrose, pounds of sucrose per ton and pounds of recoverable sugar A^{-1} compared to the non-limed control; values increased incrementally in a linear fashion with increasing rates of lime (Table 5). For instance, the linear relationship between rate of lime and pounds of recoverable sugar A^{-1} is illustrated in Fig. 3A ($R^2 = 0.9191$). Spent lime had no effect on loss to molasses (LTM, Table 5).

Breckenridge. Soil-application of lime in April, 2004 significantly decreased Aphanomyces root rot compared to the control (Table 6). Lime had no effect on LTM, but significantly increased seedling stand, number of harvested roots, root yield and sucrose yields compared no lime (Table 6). There were quadratic relationships between rate of lime and plant response for all factors (except LTM). For instance, with 5 tons of lime there was a steep increase in pounds of recoverable sucrose compared to the control and maximum yields occurred at 15 tons of lime A⁻¹ (Fig. 3B, $R^2 = 0.9504$). Even with dramatic increases in sucrose yields after soil-application of lime at Breckenridge (Fig. 3B), yields were not as high as in Hillsboro plots treated with comparable rates of lime (Fig. 3A).

Summary: Effect of a single application of spent line on rotation crops (2004-2008). In 2004 (the first growing season after application), there were significant linear decreases in yields of corn at Hillsboro and wheat at Breckenridge as plots were treated with increasing rates of spent line compared to the non-limed control (Tables 3 and 4, respectively). In subsequent years at Hillsboro, there were no significant effects of line on corn, soybean or wheat compared to the non-limed control (Table 3); in 2005, plots were fallow because excessive soil moisture delayed planting until late June. At Breckenridge in 2005, there was a positive, significant linear relationship between increasing rates of lime and wheat yields (Table 4). In 2006, all rates of lime significantly increased soybean yields compared to the control but in 2007 and 2008, there were no differences in wheat or corn yields, respectively, among limed and control plots.



Average Aphanomyces soil index values (SIVs) on a 0 to 100 scale (0 = no Aphanomyces, 100 = very high potential Fig. 4. for Aphanomyces disease if weather is wet and warm) in: A.) Hillsboro, ND plots treated with 0, 5, 10, 20 and 30 tons wet weight of spent lime A⁻¹ in October, 2003; soil samples were collected in October, 2004 ("baseline" before lime applied); July, 2004; October, 2004; June, 2005; October, 2005; June, 2006; May, 2007; and April, 2008 at 0, 9, 12, 20, 24, 32, 43 and 54 months after application, respectively, and at: B.) Breckenridge, MN treated with 0, 5, 10, 15 and 20 tons wet weight of spent lime A⁻¹ in April, 2004; soil samples were collected in April, 2004 ("baseline"); September, 2004; June, 2005, October, 2005; May, 2006; June, 2007; and May, 2008 at 0, 5, 14, 18, 25, 38 and 49 months after application, respectively. The SIVs were determined by adding soil of each sample to four (4 x 4 x 4-inch) plastic pots, planting with 25 seed of sugarbeet 'ACH 261' (to "bait" Aphanomyces cochlioides from soil), and placing in a controlled environment chamber at 70 \pm 2°F for 1 week for optimal emergence and then at 79 \pm 2°F (14-hour photoperiod) to favor infection by A. cochlioides. Stand counts were made three times weekly beginning at emergence and dying seedlings were removed to prevent disease spreading. At 4 weeks after planting, surviving seedlings were rated for disease with a 0 to 3 scale (0 = healthy, 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. Each data point is an average of at least 16 soil samples.

Summary: Effect of a single lime application on Aphanomyces soil index values (SIVs). The SIVs varied with date of sampling at both locations. Initially, SIVs decreased after treatment with spent lime for 2 years at Hillsboro (Fig. 4A) and 1 year at Breckenridge (Fig. 4B). Hillsboro plots treated with lime had lower SIVs than the non-limed control through 2008, but in 2006 and again in 2008, SIVs peaked for all limed and control plots and were higher than "baseline" SIVs (Fig. 4A). Breckenridge plots had equal and exceedingly high SIVs in limed and control plots in October, 2005 and May, 2006 (18 and 25 months after lime was applied, respectively) (Fig. 4B). Although SIVs were slightly lower in limed plots compared to the control in 2007 and 2008, overall SIVs have remained very high in all plots.

DISCUSSION

Single soil applications of spent lime are having long-term, beneficial effects on sugarbeet, based on results from 2005 through 2008 (second through fifth growing season after application). *Aphanomyces* has been especially active on sugarbeet at Breckenridge in each of the four growing seasons (2005-2008) and spent lime applied in April, 2004 has consistently provided <u>dramatic</u> reductions in root rot and increased sugarbeet yields (16, 17, 18). The Hillsboro site has moderate potential for Aphanomyces root rot, but this disease has been negligible every season except 2008, when the pathogen was measurably active; spent lime has always increased yields compared to the non-limed control (16, 17, 18) and sometimes, has statistically improved yields (16, 17). It is unknown why spent lime reduces Aphanomyces root rot and/or increases sugarbeet yields. The amendment contains nitrogen, phosphorus, potassium, and other inorganic and organic nutrients (8) obtained from the sucrose extraction process. These nutrients may alter the soil and the rhizosphere (area around roots of intense microbial activity stimulated by root exudates) environments. Various types of amendments reduce some soilborne diseases (4, 7, 11, 14) because they create complex interactions among biological, chemical, and physical factors in the soil. These interactions alter nutrient uptake by plants, improve physical condition of soil (e.g. improving water drainage, which results in less root disease), increase beneficial microorganisms in the soil and rhizosphere, and induce plant resistance.

Application of spent lime at both sites increased soil pH within a few months (15), and these values continued to remain relatively stable through 2008. *Aphanomyces cochlioides* causes severe root rot of sugarbeet over a range of soil pH values from 5.5 to 8, so benefits of spent lime treatments are more complicated than increasing soil pH. More likely, the effect of increased soil pH after application of spent lime involves changes in availability of micronutrients to the root and/or favors increases of beneficial microorganisms in the rhizosphere.

Despite the dramatic reduction of Aphanomyces diseases at Breckenridge and negligible reductions at Hillsboro, effects on Aphanomyces SIVs have been minor from year-to-year in lime-treated plots. This suggests population densities of *A. cochlioides* are <u>not</u> reduced (oospores are long-term survival/overwintering spores). One hypothesis to explain why spent lime does not consistently reduce Aphanomyces SIVs, yet reduces Aphanomyces root rot in the field is that calcium ions may interfere with production of sporangia (structures originating from germinating oospores that produce infective zoospores when soil is warm and wet). Spent lime is a rich source of calcium from the sucrose extraction process (9). Soil tests indicate that additional calcium is unnecessary for crop production in most fields in the RRV and southern Minnesota (8, 9). Calcium ions are important for species of *Pythium* and *Phytophthora* (oospore-forming pathogens related to *Aphanomyces*) in production of sporangia and in motility of zoospores and their ability infect susceptible plants (5, 12). High levels of extracellular calcium, however, interfere with these functions and are associated with suppression of diseases caused by *Pythium* and *Phytophthora*. Overall effects of calcium ions are poorly understood but are reported to affect zoospores, increase soil microbe populations and soil fungistasis, and enhance plant resistance to several pathogens (7, 11, 12).

Our research continues to explore underlying roles of spent lime in suppression of Aphanomyces root rot. In assays at Breckenridge and Hillsboro, the sugarbeet rhizosphere proved to be an abundant source of streptomyces antagonistic to *A. cochlioides*, especially when soil was treated with 10 tons of spent lime A^{-1} compared to the non-limed control (17). Streptomyces often produce antibiotics that are antifungal in activity. Fluorescent pseudomonads were abundant in the sugarbeet rhizosphere in 2006 (18), but only a low proportion of the population was antagonistic to *A. cochlioides*. On the other hand, the laboratory assay favored antibiotic-producing bacteria. Some bacteria are biologically active in other ways, e.g., altering availability of soil nutrients, competing with pathogens for nutrients, or stimulating plant resistance (4, 13, 14).

Constituents in spent lime also may directly affect *A. cochlioides*. In our preliminary studies, soil extracts from field plots treated with 20 tons of spent lime A^{-1} directly prevented production of sporangia of *A. cochlioides*. It is unknown if this response was caused by excess calcium ions or other nutrients in spent lime. Water controls (adjusted to pH values corresponding to diluted extracts of spent lime) produced abundant zoosporangia, which released motile zoospore inoculum (*unpublished*).

Sugarbeet varieties sown in 2005 to 2008, varied from year-to-year because of availability and changing production practices (e.g., resistance to rhizomania) but always included an Aphanomyces-susceptible and partially resistant variety (treated with Tachigaren). Under conditions of severe Aphanomyces root rot, optimal yields were obtained with the partially resistant variety in limed plots (18). In 2008 at Breckenridge, however, both varieties had the same Aphanomyces root rot index at harvest and the susceptible variety out-yielded the partially resistant variety (Table 2). At Hillsboro, similar trends were noted, although there were no significant differences between varieties for any yield variables (Table 1). These results likely occurred in 2008 because Aphanomyces root rot was less severe than in previous years and the susceptible variety had a greater yield potential than the partially resistant variety.

To date, a single soil-application of spent lime in our experiments at Hillsboro and Breckenridge (2004-2008) had no negative effect on rotation crops (spring wheat, corn, and soybean) except in 2004, the first growing season after application. Sometimes, crop yields were significantly higher in plots treated with spent lime compared to the nonlimed control. Smith et al. (10) reported an increase in soybean yields at some rates of spent lime. Giles and Cattanach (6) reported variable effects of spent lime applications on wheat, with yields increasing or decreasing compared to the non-limed control. It is unknown why variable responses occur on rotation crops but may be associated with low rates of lime, inadequate time for soil to stabilize after application, production practices, as well as differences in soil types and associated soil characteristics.

Because of continuing, positive effects of lime applications made in 2003 (Hillsboro) and 2004 (Breckenridge) on sugarbeet in 2008, the trials will continue in 2009 (with sugarbeet sown in plots last planted to beets in 2005). Experiments at Hillsboro and Breckenridge have soil pH values atypical of most fields in the RRV (7.5 and 6.5, respectively). Consequently, we would like to establish long-term lime trials in fields with a pH of about 8 and a history of severe Aphanomyces root rot.

SUMMARY AND CONCLUSIONS

- 1. Application of spent lime at both sites increased soil pH within a few months, and these values remained stable and relatively unchanged in 2008 (baseline pH is 7.0 at Hillsboro and 6.3 at Breckenridge).
- 2. Initial soil applications of spent lime continued to have long-term effects in reducing Aphanomyces root rot and/or increasing sugarbeet yields in 2008, the fifth growing season after the amendment was applied.
- 3. Spent lime had no adverse effects on rotation crops, except the first season after application, when yields of wheat and corn were reduced.
- 4. Populations of *A. cochlioides*, as measured by Aphanomyces Soil Index Values (SIVs) in the greenhouse, were not significantly reduced by application of spent lime.

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LITERATURE CITED

- 1. Bresnahan, G.A., A.G. Dexter, and W.C. Koskinen. 1999. The effect of soil pH on sugarbeet yield and herbicide degradation. Sugarbeet Res. Ext. Rept. 29:82-88.
- 2. Bresnahan, G.A., A.G. Dexter, C.E. Windels, J.R. Brantner, and J.L. Luecke. 2003. The effect of spent lime on sugarbeet yield and *Aphanomyces cochlioides* suppression. Sugarbeet Res. Ext. Rept. 33:273-276.
- Campbell, R.N., and A.S. Greathead. 1989. Control of clubroot of crucifers by liming. Pages 90-101 in: Soilborne Plant Pathogens: Management of Diseases with Macro- and Micronutrients. APS Press, Am. Phytopathological Soc., St. Paul, Minnesota. 217 pp.
- 4. Cook, R.J., and K.F. Baker. 1988. The Nature and Practice of Biological Control of Plant Pathogens. The American Phytopathological Society. 539 pp.
- 5. Deacon, J. 2006. The Microbial World: Fungal Zoospores. http://helios.bto.ed.ac.uk/bto/microbes/zoospore.htm.
- 6. Giles, J.F., and N.R. Cattanach. 2005. Effect of spent lime on sugarbeet production and crops following sugarbeet. 2004b Sugarbeet Res. Ext. Rept. 35:100-104.
- Ko, W.H. and C.W. Kao. 1989. Evidence for the role of calcium in reducing root disease incited by *Pythium* spp. Pages 205-217 *in*: Soilborne Plant Pathogens: Management of Diseases with Macro- and Micronutrients. A.W. Englehard (ed.). APS Press, Am. Phytopathological Soc., St. Paul, MN 217pp.
- 8. Sims, A.L., and K. Hoff. 2006. Sugarbeet factory lime: It's effects on soil chemical properties. 2005 Sugarbeet Res.Ext. Rept. 36:91-93.
- 9. Sims, A.L., C.E. Windels, and C. Bradley. 2006. Levels of specific nutrients in sugar beet factory spent lime and their impact on crop yield and soil indices. 2005 Sugarbeet Res. Ext. Rept. 36:95-104.
- 10. Smith, L.J., T.E. Cymbaluk, and J.D. Nielsen. 2006. Spent lime rate effects on sugarbeet yield and quality (2004), wheat and soybean (2005). 2005 Sugarbeet Res. Ext. Rept. 36:105-112.
- 11. Sun, S. and J. Huang. 1985. Formulated soil amendment for controlling Fusarium wilt and other soilbornediseases. Plant Dis. 69:917-920.
- 12. von Broembsen, S.L. and J.W. Deacon. 1997. Calcium interference with zoospore biology and infectivity of *Phytophthora parasitica* in nutrient irrigation solutions. Phytopathology 87:522-528.
- 13. van Loon, L.C., P.A.H.M. Baker, and C.M.J. Pieterse. 1998. Systemic resistance induced by rhizosphere bacteria. Annu. Rev. Phytopathol. 36:453-483.
- 14. Weller, D.M., J.M. Raaijmakers, B.B. McSpadden Gardener, and L.S. Thomashow. 2002. Microbial populations responsible for specific soil suppressiveness to plant pathogens. Annu. Rev. Phytopathol. 40:309-348.
- 15. Windels, C.E., A.L. Sims, J.R. Brantner, and C. Bradley. 2005. Reclamation and fertilization of *Aphanomyces*-infested sugarbeet fields amended with industrial spent lime. 2004b Sugarbeet Res. Ext. Rept. 35:218-223.
- 16. Windels, C.E., A.L. Sims, J.R. Brantner, and C.A. Bradley. 2006. Spent lime effects on Aphanomyces, soil microorganisms, and sugarbeet. 2005 Sugarbeet Res. Ext. Rept. 36:250-261.
- 17. Windels, C.E., A.L. Sims, J.R. Brantner, and C.A. Bradley. 2007. Spent lime effects on sugarbeet, root rot, microorganisms, and rotation crops. 2006 Sugarbeet Res. Ext. Rept. 37:208-219.
- Windels, C.E., J.R. Brantner, A.L. Sims, and C.A. Bradley. 2008. Long-term effects of a single application of spent lime on sugarbeet, Aphanomyces root rot, rotation crops, and antagonistic microorganisms. 2007 Sugarbeet Res. Ext. Rept. 38:251-262.