

Efficacy of precipitated calcium carbonate in managing fusarium root rot of field pea

Kishore Chittem · Mohamed F. R. Khan ·
Rubella S. Goswami

Received: 14 April 2016 / Accepted: 21 July 2016 / Published online: 5 August 2016
© Springer Science+Business Media Dordrecht 2016

Abstract Recently, root rots caused by *Fusarium* spp. have become a major constraint for field pea production in North Dakota. With the inadequate disease management options currently available, this study was initiated to determine whether precipitated calcium carbonate (PCC), a byproduct of the sugar processing industry, could be used for managing fusarium root rots of field pea. The efficiency of PCC in controlling *Fusarium* species associated with root rots was evaluated in vitro and under greenhouse, and field conditions. Effect of PCC on growth characteristics of six *Fusarium* spp. viz., *F. acuminatum*, *F. avenaceum*, *F. redolens*, *F. solani*, *F. graminearum*, and *F. oxysporum*, commonly associated with pea root rot was evaluated in vitro. Significant reduction in conidia production, conidial germination, and dry mycelial weight were recorded in presence of PCC in growth media. In the greenhouse, PCC efficacy was tested at varying rates (equivalent to 0, 2.2, 5.6, 11.2, and 22.5 t/ha of field application (corresponding to 0, 1, 2.5, 5 and 10 t/ac, respectively)) in pot culture experiments containing soil infested with *F. avenaceum* and *F. solani*, the two most aggressive and common *Fusarium* pathogens associated with field pea root rots in North Dakota. Significant reduction in root rot

disease severity was observed in both *F. avenaceum*, and *F. solani* infested pots at all rates of PCC application compared to the controls. Similar results were obtained from multi-location field experiments conducted in 2010 and 2011 with reduction in root rot severity following application of PCC. Results from this study demonstrated that PCC has the potential to serve as a strategy for Fusarium root rot management in field pea.

Keywords Precipitated calcium carbonate · Fusarium root rot · Field pea · *Fusarium avenaceum* · *Fusarium solani* f. sp. *pisi*

Introduction

North Dakota (ND) was the leading field pea (*Pisum sativum* L.) producer in USA accounting for more than 50 % of the acres planted until 2010. However, since 2010 the acres planted to field pea in ND has been on the decline (~35 % of the total US acres planted) (United States Department of Agriculture, National Agricultural Statistics Service [USDA-NASS]). Root rots, which have become a major constraint for field pea production in ND (Chittem et al. 2015), are considered as one of the important reasons for this decline. Recent surveys have indicated that *Fusarium* spp. including *F. avenaceum*, *F. culmorum*, *F. graminearum*, *F. oxysporum*, *F. redolens*, *F. solani* and *F. sporotrichioides* are the pathogens most frequently associated with pea root rots in ND (Gregoire and Bradley 2005; Mathew et al. 2008; Chittem et al. 2015). Among these, *F. avenaceum* was identified as the

K. Chittem · M. F. R. Khan
Department of Plant Pathology, North Dakota State University,
Fargo, ND 58102, USA

R. S. Goswami (✉)
Department of Agriculture and Natural Resources, Delaware State
University, Dover, DE 19901, USA
e-mail: rgoswami@desu.edu

most virulent and prevalent (Chittem et al. 2015) in causing pea root rots in ND. More recently, *Aphanomyces euteiches* was also reported to be associated with pea root rots in ND (Zitnick-Anderson and Pasche 2016). Currently there is a dearth of satisfactory methods to control root rot (Kraft and Pflieger 2001), seed treatments used are also limited in their efficacy (Samuel Markell, North Dakota State University, personal communication). The importance of this crop to ND and prevalence of the disease necessitates development of an integrated disease management program to reduce losses associated with fusarium root rot in dry peas.

Precipitated calcium carbonate (PCC) also known as spent lime or waste lime, a byproduct of sugar industry where lime (limestone) is used in the clarification process for its neutralizing ability. It is reported to be effective in reducing the severity of root rot in sugar beet and spinach caused by *A. cochlidioides* and *A. cladogamus*, respectively (Windels et al. 2005, and Ingemarsson 2004). Due to the soil borne nature of the root rot pathogens and similar pH requirements, we hypothesize that PCC could be effective for managing fusarium root rots of field pea as well. Further, spent lime also improves soil structure and tilth. Besides neutralizing soil pH, spent lime provides significant amounts of calcium (21 %), and other nutrients like nitrogen (0.6 %), phosphorous (0.7 %), potassium (0.05 %), and magnesium (1.1 %), and enriches the soil (Ingemarsson 2004). Increased or adequate soil calcium (Ca) levels has been shown to provide resistance against soil borne pathogens like *Rhizoctonia*, *Pythium*, *Sclerotinia*, *Botrytis* and *Fusarium* (Dordas 2008; Graham 1983). Ca is an important structural component of plant cell wall and plasma membrane, and is believed to play an important role in host resistance by affecting both their structure and function (Dordas 2008). Additionally, Ca plays an important role in responses of plants to biotic stresses via Ca signaling pathways (Zhang et al. 2014). PCC has been successfully used in soils with a pH up to 8 (Windels et al. 2007) and since the pH of most field pea growing soils in North Dakota ranges from 5 to 8 (Mathew 2006), it is believed that the use of PCC could be effective in reducing losses in this crop as well. Sims et al. (2007) reported that 500,000 tons dry weight of spent lime is produced by seven sugar factories in North Dakota and Minnesota that is available to growers free of cost. The advantages

of spreading PCC in root rot management would be two fold, providing a cheaper method for controlling the disease, and serving as an amendment that improves the chemical and physical properties of the soil.

Previous reports suggest that PCC has been effective in reducing root rot and/or increasing yield in sugar beet grown in North Dakota and Minnesota (Windels et al. 2005). PCC when applied at 10 t/ac or 30 t/ac resulted in reduction of *Aphanomyces* soil index values up to 67 % in sugar beet (Windels et al. 2005). In greenhouse studies in Sweden, application of lime resulted in reduced root rot severity of sugar beet and caused increase in plant fresh weight (Ingemarsson 2004). Apart from these, lime has been directly used to control clubroot of cabbage in US (Campbell and Greathead 1989). Efficacy of lime in reducing root diseases has been documented in peas, tomato, potato, and red clover (Allmaras et al. 1987; Sonoda 1978; Smith and Gibbon 1976; Steiner and Alderman 2003). The objective of this study was to determine the efficacy of PCC as a strategy for managing root rot disease in field peas.

Materials and methods

Laboratory experiments

Effect of PCC on mycelial growth, conidia production, conidial germination and dry mycelial weight

Effect of PCC on growth characteristics of six *Fusarium* spp. - *F. avenaceum*, *F. acuminatum*, *F. solani*, *F. graminearum*, *F. redolens*, and *F. oxysporum* - commonly associated with field pea root rots in the region was tested in vitro. The isolates used in this study were obtained from pea roots (Chittem et al. 2015). Maximum radial growth, growth rate, conidia production, and conidia germination were studied on potato dextrose agar (PDA) media amended with PCC at concentrations equivalent to 2.2, 5.6, 11.2, and 22.5 t/ha (which corresponds to 1, 2.5, 5 and 10 t/ac, respectively) of field application. Effect of spent lime on fungal biomass reduction was evaluated using potato dextrose broth (PDB) amended with PCC. The amount of PCC to be added to PDA or PDB that corresponded to field application rates was determined considering that 1 ha of furrow slice (hfs) ploughed to a depth of 15 cm would weigh approximately 2,240,000 kg.

Amount of PCC to be added per liter of medium was calculated using the following formula :

$$= \frac{\text{Field application rate of PCC in mg}}{2,240,000}$$

For example, 2.25 t/ha (1 t/ac) will be, $\frac{2250,000,000}{2,240,000} = \sim 1000$ mg /Kg of soil.

Using the above mentioned hfs weight, 2.2, 5.6, 11.2, and 22.5 t/ha corresponded to approximately 1000, 2500, 5000, and 10,000 ppm respectively. Calculated amounts of PCC were added to PDA or PDB to give concentration equivalent to field application rates of PCC. PDA or PDB without any PCC added served as controls (0 t/ha).

Five mm plugs of actively growing cultures (5–7 days old) of each isolate were excised and transferred on to petri plates (100 mm \varnothing) containing PCC amended PDA and incubated in a growth chamber with a 14 h photo-period and day and night temperatures of 21° and 18 °C, respectively. Observations on radial growth were measured from 2 days after inoculation (DAI) to 6 DAI. Two perpendicular readings of colony diameters were made for each petri plate. Radial growth at 6 DAI was expressed as maximum radial growth, and growth rate was estimated from the observations from 2nd to 6th DAI using PROC REG procedure of SAS version 9.1. On sixth DAI five 5 mm diameter mycelial plugs were cut from each plate, transferred to test tubes containing 5 mL sterile distilled water, and shaken thoroughly. Aliquots from the tubes were transferred onto haemocytometer and number of conidia per mL was determined. From each plate, spores were counted twice. The experiment was a completely randomized design, with three replicates for each concentration, and was repeated two times.

Conidia were obtained for each isolate from fresh cultures grown on PDA. As the *F. graminearum* isolate used in this study failed to produce conidia on PDA, conidia for this isolate were obtained by growing the culture on Mung Bean Agar (MBA). Conidial concentration was adjusted to 1000 conidia/mL using a haemocytometer, 200 μ L of the conidial suspension was uniformly spread on petri dishes (65 mm \varnothing) and incubated at room temperature for 24 h. A total of 100 conidia per isolate were counted and the number of conidia germinated at each concentration was expressed as the percentage of germinating conidia. The experiment was laid out in a completely randomized design

with three replicates per concentration and was conducted two times.

Conical flasks (250 mL) containing 50 mL of PDB amended with PCC were inoculated with 5 mm mycelial discs of actively growing cultures and incubated at room temperature on a rotary shaker at 120 rpm for 6 days. After 6 days the mycelium was harvested, dried in a hot air oven at 70 °C for 24 h and the dry mycelial weight was recorded. The experiment was a completely randomized design, each treatment was replicated three times and the experiment was conducted three times.

Greenhouse experiments

In greenhouse studies, efficacy of PCC in reducing root rot severity caused by *F. avenaceum*, and *F. solani* f. sp. *pisi*, the two pathogens most virulent, and commonly associated with root rot of field peas in ND was assessed. Three aggressive isolates of each pathogen species (Pea 41, FPS M60 and FA0601 of *F. avenaceum* (Chittem et al. 2015) and Fs 01.B1, F215, F54 of *F. solani* f. sp. *pisi* (Grünwald et al. 2003)) were used for this study. The inoculum was prepared as described in Chittem et al. 2015. The three isolates of each species were mixed together before inoculating the soil. The inoculum was mixed with sterilized (at 121 °C, 1.41 kg/cm² (20 psi) for 2 h on two consecutive days) field soil in 1 inoculum: 20 soil w/w ratio and filled into 15 cm deep, 25.4 x 12.7 cm rectangular pots (TO Plastics Inc. Clearwater, MN, USA). The pots were watered thoroughly and covered with a plastic wrap for 48 h to stabilize the inoculum (Sagar and Sugha 2004). The amount of PCC to be added in this study was calculated based on the amount available to each plant under field conditions based on the surface area. Different rates of PCC equivalent to 2.2, 5.6, 11.2, and 22.5 t/ha were applied to the pots containing soil inoculum mixture, mixed thoroughly to ensure uniformity, and the pots were left for 24 h before planting. Treatments with pots containing inoculum but no PCC and only autoclaved soil were used as controls. Each pot was planted with 15 seeds, and thinned to 10 seedlings after germination. DS Admiral, a known susceptible field pea cultivar to root rot was used for this study (Chittem et al. 2015). The plants were carefully removed from the pots 14 days after planting, and roots were washed with tap water to remove soil adhering to the surface. Washed roots were rated for root rot severity. Root rot severity was measured as the length of

lesions on tap root compared to whole root length, expressed as percentage. The experiment was set up as randomized complete block design with four replicates per treatment and the experiment was conducted twice. Soil pH before and after adding PCC was measured using a glass electrode (pH 510 series, Oakton Instruments, IL, USA) in 1:1 soil-water slurry (w/v).

Field experiments

Field studies were conducted in growers' fields with known history of root rot incidence. PCC was applied at the rates of 0 (untreated control), 11.2, 22.5 and 33.75 t/ha of wet weight [29 % moisture w/w] (corresponding to 0, 5, 10 and 15 t/ac) to the experimental plots. PCC was spread evenly and incorporated into the soil (to a depth of 10 cm) with a chisel plough in the fall of 2009. DS Admiral, a field pea cultivar known to be susceptible to root rot was used for planting. Each treatment was replicated four times. Stand counts were recorded 3 weeks after planting on the four inner rows. Root rot severity was assessed at the pre-flowering stage. Ten plants were collected from each plot from inner 6 m in the center of the 18 m long inner four rows by destructive sampling. Roots were washed under running tap water and assessed for incidence and severity of root rot. Root rot severity was measured as the percentage of root length covered by lesions (length of lesions/total root length X 100). Yield data was recorded at the end of crop season. The plots were 13.5 m wide x 18 m long, observations on stand counts, disease severity, and yield were recorded from the inner 3 m of the 18 m long rows. The experiment was laid out as randomized complete block design, conducted at Hickson, ND, and Morehead, MN and repeated over 2 years, in summer of 2010 and 2011. Soil pH of experimental sites was determined as described above.

Statistical analysis

Statistical analyses for the above studies were performed using SAS version 9.1 (SAS Institute Inc. 2002–2003). Homogeneity of variance between experiments was tested using Levene's test. Similar experiments were combined and analyzed using all replications using PROC ANOVA procedure. Mean separation was performed using Fisher's protected least significant difference (LSD) test.

Results

Laboratory experiments

Effect of PCC on mycelial growth, growth rate, conidia production, conidial germination, and fungal biomass

Test for homogeneity of variance between experiments were not significant, so all the three experiments were combined for analysis. The radial growth of only three of the six *Fusarium* species evaluated appeared to be affected significantly by the application of PCC. *F. graminearum* was affected the most, followed by *F. oxysporum* and *F. acuminatum*, in that order (Fig. 1a). Mycelial diameter of these three *Fusarium* spp. was significantly higher on PCC amended PDA plates compared to non-lime amended control plates. However, no significant differences in radial growth were observed between PCC amended plates at varying rates, indicating a lack of dose response. Though the differences in maximum mycelial growth were not statistically significant a similar trend was observed for the other three *Fusaria* viz., *F. avenaceum*, *F. redolens* and *F. solani* tested in this study. Additionally, it was observed that the growth of mycelia in the plates amended with PCC was less dense compared to that in plates without PCC for all *Fusarium* species included in this study.

Analysis of growth rate demonstrated a trend similar to maximum growth (Fig. 1a). *F. acuminatum*, *F. graminearum*, and *F. oxysporum*, showed an increase in growth rate with the addition of PCC, with *F. graminearum* being the one most affected. However, no significant change in the growth rate of the other *Fusarium* species was recorded in the lime amended plates compared to non-lime amended control (Fig. 1b).

Significant differences in conidia production were detected in all species where sporulation was observed, except for *F. redolens* (Fig. 1c). Number of conidia reduced with increasing rate of PCC. Conidia production was significantly reduced at all concentrations of lime tested i.e., 2.2, 5.6, 11.2 and 22.5 t/ha in *F. acuminatum*, *F. avenaceum*, and *F. oxysporum*. In case of *F. solani*, reduction in conidia production compared to the control was significant only at higher concentrations of 5.6, 11.2 and 22.5 t/ha. The reduction in conidial production compared to 0 t/ha ranged from 12 to 99 %. No conidial production was observed for *F. graminearum* in any of the treatments including the

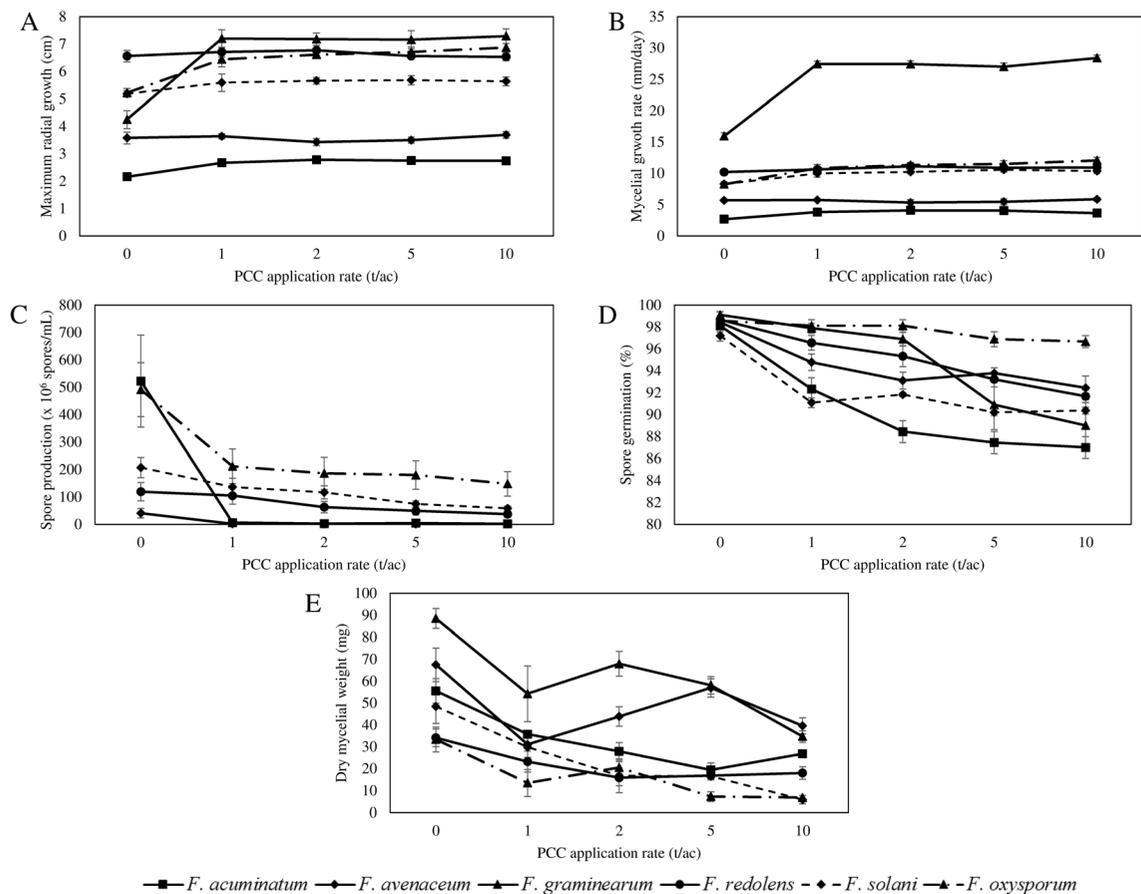


Fig. 1 Effect of precipitated calcium carbonate (PCC) on growth characteristics of *Fusarium* species commonly associated with field pea root rots in North Dakota; a. Maximum radial growth, b. Growth rate, c. Conidial production, d. Conidial germination, (*F. solani* on secondary axis), and e. mycelial dry weight. Each

mean represent average of three experiments for maximum radial growth, growth rate, spore production and dry mycelial weight and two experiments for spore production. Each experiment consisted of three replications. Error bars represent standard error of mean

control. Incubation of the plates for seven more days longer also did not result in any conidial production.

Conidial germination was also affected by the presence of PCC in the growth media. The germination percentage of conidia of *F. graminearum*, and *F. oxysporum* was reduced only at higher concentrations of 11.2, and 22 t/ha, but in the other species studied, conidial germination was significantly reduced at all concentrations i.e., 2.2, 5.6, 11.2 and 22.5 t/ha (Fig. 1d) compared to the control plates. Percentage reduction in conidial germination compared to 0 t/ha ranged between 1 and 41 %.

Fungal biomass production measured in terms of dry mycelial weight, for all the isolates of *Fusarium* species studied in this experiment was found to be significantly lower in the presence of lime as compared to the control (Fig. 1e). The reduction in biomass as relative to the rate

of PCC applied however, varied between species. The reduction in fungal biomass production compared to 0 t/ha ranged between 16 and 88 %.

Greenhouse experiments

Levene's test for homogeneity of variance showed that the two experiments can be combined, so data from the two experiments were combined for analysis to determine the effect of lime in reducing root rot severity. The experiments showed significant reduction in root rot disease severity, in both *F. avenaceum*, and *F. solani* treated pots at all the rates of lime application (2.2 to 22.5 t/ha) compared to the non-inoculated control (Table 1). Inoculated pots with no lime applied always had recorded highest root rot severity (17.66 % and 16.47 % for *F. avenaceum* and *F. solani* respectively).

Table 1 Effect of precipitated calcium carbonate (PCC) on root rot disease severity caused by *Fusarium avenaceum* and *F. solani* f. sp. *pisi* in greenhouse trials

PCC rate (t/ha)	pH	Mean root rot severity (%) ^a	
		<i>F. avenaceum</i>	<i>F. solani</i> f. sp. <i>pisi</i>
0	6.86	17.66a*	16.47a
2.2	7.42	11.37b	11.92b
5.6	7.48	8.32bc	10.82b
11.2	7.57	6.09 cd	7.72bc
22.5	7.61	3.91d	5.37c
Non-inoculated control	6.86	0.04e	0.04d

* means with the same letter are not statistically different ($\alpha=0.05$)

^a mean root rot severity was calculated as the percentage of root length covered by lesions 14 days after planting. Means are average of eight replications. Each replication consisted of ten seedlings

For both pathogens, disease severity decreased in response to lime application.

Field experiments

Test of homogeneity of variance was significant between the four field experiments (2 years, two locations), so the experiments were analyzed separately. Significant differences in root rot severity were recorded between the treatments in 3 of the 4 field trials conducted over 2 years (Table 2). Lime applied at higher rates always had less root rot severity as compared to the control. In 2010, at the Hickson site, lime application at all the rates i.e., 11.2, 22.5, 33.7 t/ha (5, 10, and 15 t/ac, respectively) showed significantly lower amounts of disease compared to no lime application. However, at the Moorhead site, lime application only at the rates of 22.5 and 33.7 t/ha showed significant reduction in root rot severity compared to the control. In 2010, the trials were affected by high weed pressure at both locations and no significant differences in stand counts (6 to 7 plants/m) or yield (445 to 579 g/m at Hickson) were observed with the application lime at either of the two sites. In 2011, statistically significant differences in disease severity were observed only at Hickson site. Disease severity in the plot with no PCC application was significantly higher compared to the lime applied plots. In Moorhead, the differences in disease severity were not significant (Table 2). The trials at both locations

Table 2 Effect of Precipitated calcium carbonate (PCC) on root rot severity of field pea in field trials conducted in 2010 and 2011 at Hickson, ND and Moorhead, MN

PCC rate (t/ha)	Root rot severity (%) ^a			
	Hickson		Moorhead	
	2010	2011	2010	2011
0	15.55a*	17.61a	12.22a	10.04
11.2	10.41b	11.69b	9.01ab	7.61
22.5	6.99c	7.15c	6.17bc	6.56
33.75	4.51d	5.76c	3.71c	4.88

* means followed by the same letter are not statistically different ($\alpha=0.05$) according to Fisher's least significant difference test

^a mean root rot severity was calculated as the percentage of root length covered by lesions measured at pre-flowering stage. Each mean represents the average of four replications. Each replication constituted of ten roots

were affected by heavy rain and water logging in 2011. No significant differences between treatments were observed for stand counts (4 to 5 plants/m and 7 to 8 plants/m at Hickson and Moorhead, respectively) and yield (99 to 147 g/m and 185 to 235 g/m at Hickson and Moorhead, respectively).

Discussion

In this study we evaluated the effectiveness of PCC or spent lime, a byproduct from the sugar beet industry as a management strategy for controlling fusarium root rot of field peas. Effectiveness of spent lime in controlling aphanomyces root rot in sugar beet and spinach has been demonstrated under field and greenhouse conditions (Windels et al. 2010; Ingemarsson 2004). However, no information is available on the influence of PCC on plant pathogenic *Fusarium* spp. under in vitro conditions based on current literature. Therefore, we initiated a study to evaluate the effect of PCC on mycelial growth, conidia production, conidial germination, and fungal biomass production. Based on our laboratory experiments, except for *F. acuminatum*, *F. graminearum* and *F. oxysporum*, no significant differences in either radial growth or growth rate was observed in other *Fusarium* species including, *F. avenaceum*, *F. solani*, *F. graminearum*, and *F. redolens*. However, visible differences in the mycelial density were observed. The mycelium in the lime amended plates was always

sparse, i.e. less dense compared to the control plates, similar to the kind of growth that can be observed when the cultures were grown on water agar, and a tendency towards aerial growth appeared to be favored. These visible differences in mycelial density were further quantified in terms of dry mycelial weight or fungal biomass using potato dextrose broth amended with PCC. Application of lime at all the rates resulted in significant reduction in fungal biomass compared to PCC non-amended flasks. More than 50 % reduction in dry mycelial weight was observed for *F. acuminatum*, *F. graminearum*, *F. solani* and *F. oxysporum*.

In this study we also observed significant reduction in conidia production and conidial germination in presence of spent lime in the media. Allmaras et al. (1987), in a field study reported a 37 % reduction in propagule density of *F. solani* f. sp. *solani* over 3 years with a single application of lime. In a laboratory based study, Ulfig (2006) observed that application of lime to increase the pH to 12 resulted in elimination of keratinophilic *F. solani* from the sludge. Their study was focused on keratinolytic and keratinophilic fungi, which are of less significance to agriculture, and reported that addition of lime to sludge resulted in either complete elimination or significant reduction in keratinolytic and keratinophilic fungi. In greenhouse trials, addition of PCC to pots resulted in a slight increase of pH (6.86, 7.42, 7.48, 7.57, and 7.61 for 0, 2.2, 5.6, 11.2 and 22.5 t/ha, respectively). Our data from greenhouse trials showed that root rot severity was significantly lower in PCC treated pots at all the rates of application compared to the control pots for both the pathogens. Disease severity decreased with increasing rates of lime application. Disease severity was lowest at 22.5 t/ha for *F. avenaceum* inoculated pots (3.91 %, Table 1), however, there were no significant differences between 11.2 and 22.5 t/ha treatments for *F. solani* f. sp. *pisi*. Studies on efficacy of spent lime in controlling aphanomyces root rot (Windels et al. 2005, 2007, 2008, 2009, 2010) have demonstrated a decrease in *Aphanomyces* disease severity index values. Similar findings with the application of slaked lime and factory lime was also reported by Ingemarsson (2004). Efficacy of liming has been successfully demonstrated in either reducing propagule density or control of soil borne diseases in many crops including field peas, tomato, potato, red clover, and canola (Allmaras et al. 1987; Sonoda 1978., and Smith and Gibbon 1976; Woltz, et al. 1992; Steiner, and Alderman. 2003; Brauer et al.

2002; Arshad et al. 1997; Hubbell et al. 2001). However, there are contrary reports where liming has resulted in increase of disease severity in field pea and red clover (Lyndon Porter, unpublished data; Steiner and Alderman 2003). PCC application, apart from resulting in suppression of the disease, has also been shown to result in enhancing the yield and quality of sugar beet (Giles and Cattanaach 2004; Cattanaach et al. 2008; Windels et al. 2009). We have observed similar reduction in disease severity in our field experiments. However, we believe that our results were insufficient for establishing the effect on stand count and yield. Stand counts were affected by high weed pressure in 2010 and water logging in 2011. In 2010 yield data could not be collected due to heavy rains and water logged conditions.

PCC adsorbs various micro nutrients during the clarification process, and is considered enriched with nutrients compared to the commercial lime (Ingemarsson 2004; Sims et al. 2010). It is known to supply phosphorus to sugar beet, and response of sugar beet to PCC application was similar to phosphorus fertilizer application. (Sims et al. 2007; Salisbury and Hills 1987). In addition to supplying phosphorus, PCC also provides Ca, Mg, Na, and K (Sims et al. 2010; Ingemarsson 2004). Based on our results, and other published information about PCC efficacy and contrasting reports, we hypothesize that the effect of PCC in reducing root rot severity involves more than just alteration (increase) of soil pH. Several effects of pH in suppression of disease have been reported including direct effect on pathogen growth, alteration of the soil conditions to make them favorable for the plant, and antagonistic microbial growth, or change in the nutrient availability (Crowley and Alvey 2002). Suppression of fusarium wilt is thought to be the result of increased competition for carbon (C) and iron (Fe) in the soil by non-pathogenic *Fusarium* and other microbial antagonists (Alabouvette 1999). Allmaras et al. (1987) proposed that the reduction in propagule density of *F. solani* in lime applied field pea plots may be due to increased exchangeable Ca, which might have improved resistance to pathogen attack, and impaired saprophytic survival ability of the pathogen or by favoring the growth of microbial antagonists. The pH of the experimental sites at both locations was slightly alkaline (7.5 at Hickson and 7.8 at Moorhead), and we did not see any noticeable increase in soil pH with application of lime. Therefore, we believe that the reduction in root rot disease severity under field

conditions may have involved multiple factors such as improved root growth and thus resistance to pathogen attack by supplying nutrients, and by favoring the growth of antagonistic microbes in rhizosphere. Furthermore, addition of lime to soil reduces soil bulk density, i.e. soil becomes less compact, improves soil aeration, and prevents soil crusting (Webster, and Nyborg. 1986). *Fusarium* root rot in field pea is favored by soil compaction and agronomic practices like zero tillage (Tu 1994; Fernandez et al. 2008). Thus we believe that PCC in addition to bringing favorable changes in soil chemical and biological properties, also alters physical properties that are favorable for plant growth and unfavorable for pathogen growth. Our results indicate that PCC can be a potential inexpensive strategy for management of this disease particularly in areas where it is easily available.

Acknowledgments Part of this material is based upon work that is supported by the National Institute of Food and Agriculture, U.S. Department of Agriculture, under award number 2009-38640-19953 through the North Central Region SARE program under subaward number GNC09-106. USDA is an equal opportunity employer and service provider. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

References

- Alabouvette, C. (1999). *Fusarium* wilt suppressive soils: an example of disease-suppressive soils. *Australasian Plant Pathology*, 28, 57–64.
- Allmaras, R. R., Kraft, J. M., & Pikul, J. L. (1987). Lime and gypsum effects on pea-root-pathogen inoculum and related factors in a wheat-peas rotation. *Agronomy Journal*, 79, 439–445.
- Arshad, M. A., Woods, D. L., Turkington, T. K., & Gill, K. S. (1997). Canola root rot and yield response to liming and tillage. *Agronomy Journal*, 89, 17–22.
- Brauer, D., Belesky, D., & Ritchey, D. (2002). Effects of lime and calcium on root development and nodulation of clovers. *Crop Science*, 42, 1640–1646.
- Campbell, R.N., & Greathead, A.S. (1989). Control of clubroot of crucifers by liming. Soil-borne plant pathogens: Management of diseases with Macro- and Micro nutrients. St. Paul, MN: American Phytopathological Society, pp. 90–101.
- Cattanach, N. R., Overstreet, L. F., & Giles, J. F. (2008). Effect of spent lime on sugar production and crop yields following sugarbeet. *2007 Sugarbeet Research and Extension Reports*, 38, 108–109.
- Chittam, K., Mathew, F. M., Gregoire, M., Lamppa, R. S., Chang, Y. W., Markell, S. G., Bradley, C. A., Barasubiye, T., & Goswami, R. S. (2015). Identification and characterization of *Fusarium* spp. Associated with root rots of field pea in North Dakota. *European Journal Plant Pathology*, 143, 641–649.
- Crowley, D. E., & Alvey, S. A. (2002). Regulation of microbial processes by soil pH. In Z. Rengel (Ed.), *Handbook of plant growth: pH as master variable* (pp. 351–382). New York: Marcel Dekker, Inc.
- Dordas, C. (2008). Role of nutrients in controlling plant diseases in sustainable agriculture. A review. *Agronomy for Sustainable Development*, 28, 33–46.
- Fernandez, M. R., Huber, D., Basnyat, P., & Zentner, R. P. (2008). Impact of agronomic practices on populations of *Fusarium* and other fungi in cereal and noncereal crop residues on the Canadian prairies. *Soil and Tillage Research*, 100, 60–71.
- Giles, J. F., & Cattanach, N. R. (2004). Effect of spent lime on sugar production and crops following sugarbeet. *2003 Sugarbeet Research and Extension Reports*, 35, 100–104.
- Graham, R. D. (1983). Effects of nutrient stress on susceptibility of plants to disease with particular reference to the trace elements. *Advances in Botanical Research*, 10, 221–276.
- Gregoire, M., & Bradley, C. (2005). Survey of root rot diseases affecting dry pea in North Dakota. *Phytopathology*, 95, S36 (Abstract).
- Grünwald, N. J., Coffman, V. A., & Kraft, J. M. (2003). Sources of partial resistance to *Fusarium* root rot in the *Pisum* core collection. *Plant Disease*, 87, 1197–1200.
- Hubbell, L. A., Christenson, D. R., & List, R. R. (2001). Applying different rates of lime to alkaline soils and the effects on corn, navy beans and sugar beets. American Society of Sugar Beet Technologists. Proceedings of the 31st biennial meeting, 31, 86–91.
- Ingemarsson, A. (2004). Effects of lime and organic amendments on soil borne pathogens, especially *Aphanomyces* spp. of sugarbeet and spinach. M. S. thesis. Swedish University of Agricultural Sciences.
- Kraft, J. M., & Pflieger, F. L. (2001). *Compendium of Pea Diseases and Pests* (2nd ed.). St. Paul: The American Phytopathological Society.
- Mathew, F. M. (2006). Effect of pH on performance of *Coniothyrium minitans* Campbell isolates retrieved from highly alkaline soils. M. S. thesis. North Dakota State Univ., Fargo. Pp. 30.
- Mathew, F., Barasubiye, T., Markell, S. G., & Goswami, R. S. (2008). Detection and identification of *Fusarium* species in field pea roots. *Phytopathology*, 95, S100.
- Sagar, V., & Sugha, S. K. (2004). Role of soil amendment and repeated cropping in the management of pea root rot. *Tropical Science*, 44, 1–5.
- Sailsbery, R., & Hills, F. J. (1987). Waste lime supplies phosphorus to sugarbeet. *California Agriculture*, 41, 17–18.
- Sims, A. L., Windels, C. E., & Bradley, C. A. (2010). Content and potential availability of selected nutrients in field-applied sugar beet factory lime. *Communications in Soil Science and Plant Analysis*, 41, 438–453.
- Sims, A. L., Windels, C. E., & Bradley, C. A. (2007). Field applications of sugar factory spent lime: effects on soil phosphorus. American Society of Sugarbeet Technology, Proceedings of the 35th biennial meeting, March 1–3, 2007, pp 77–79.
- Smith, A. H., & Gibbon, A. W. (1976). Sugar factory waste lime and potato scab. *British Sugarbeet Review*, 44, 26.
- Sonoda, R. M. (1978). Effect of fungicide, lime and fertilizer regimes on *Fusarium* crown rot of tomato. Fort Pierce ARC

- Research Report RL 1978–1. Florida University Agricultural Research Center.
- Steiner, J. J., & Alderman, S. C. (2003). Red clover seed production. VI. Effect and economics of soil pH adjusted by lime application. *Crop Science*, *43*, 624–630.
- Tu, J. C. (1994). Effects of soil compaction, temperature, and moisture on the development of the Fusarium root rot complex of pea in southern Ontario. *Phytoprotection*, *75*, 125–131.
- Ulfing, K. (2006). Sludge liming decreases the growth of keratinolytic and keratinophilic fungi. *Polish Journal of Environmental Studies*, *15*, 341–346.
- Webster, G. R., & Nyborg, M. (1986). Effects of tillage and amendments on yields and selected soil properties of two solonchic soils. *Canadian Journal of Soil Science*, *66*, 455–470.
- Windels, C. E., Sims, A. L., Bartner, J. R., & Bradley, C. A. (2005). Reclamation and fertilization of *Aphanomyces* infested sugarbeet fields amended with industrial waste lime. *2004 Sugarbeet Research and Extension Reports*, *35*, 218–224.
- Windels, C. E., Bradley, C. A., Sims, A. L., & Brantner, J. R. (2007). Spent lime effects on yield, quality, and *Aphanomyces* root rot of sugarbeet. *2006 Sugarbeet Research and Extension Reports*, *37*, 208–219.
- Windels, C. E., Bradley, C. A., Sims, A. L. & Brantner, J. R. (2008). Long-Term Effects of A Single Application of Spent Lime on Sugarbeet, *Aphanomyces* Root Rot, Rotation Crops, and Antagonistic Microorganisms. *2007 Sugarbeet Research and Extension Reports*, *38*, 251–262.
- Windels, C. E., Bradley, C. A., Sims, A. L. & Brantner, J. R. (2009). Five-Year Effect of a Single Field Application of Various Rates of Spent Lime on *Aphanomyces*, Sugarbeet, and Rotation Crops. *2008 Sugarbeet Research and Extension Reports*, *39*, 237–249.
- Windels, C. E., Bradley, C. A., Sims, A. L. & Brantner, J. R. (2010). A Single Field Application of Spent Lime Continues to Reduce *Aphanomyces* Root Rot and Increases Sugarbeet Yields During Sixth Growing Season. *2009 Sugarbeet Research and Extension Reports*, *40*, 248–255.
- Woltz, S. S., Scott, J. W., & Jones, J. P. (1992). Sodium chloride, nitrogen source, and lime influence Fusarium crown rot severity in tomato. *HortScience*, *27*, 1087–1088.
- Zhang, L., Du, L., & Poovaiah, B. W. (2014). Calcium signaling and biotic responses in plants. *Plant Signaling and Behaviour*, *9*(11), e973818.
- Zitnick-Anderson, K., & Pasche, J. (2016). First report of *Aphanomyces* root rot caused by *Aphanomyces euteiches* on field pea in North Dakota. *Plant Disease*, *100*, 522.