Supplement to Journal of Nematology 26(4S):606–611. 1994.

Effect of Lime on *Criconemella xenoplax* and Bacterial Canker in Two California Orchards[1]

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Abstract: In a peach orchard with an initial soil pH of 4.9, preplant application of 0, 13.2, 18.2, 27.3, or 54.2 kg lime/tree site altered soil pH (range after 1 year = 4.8–7.3) but did not affect numbers of *Criconemella xenoplax* or tree circumference. Liming also failed to reduce the incidence of bacterial canker, which affected 17% of the trees by the sixth year after planting. Four years after planting, numbers of *C. xenoplax* exceeded 400/100 cm^3 soil, regardless of treatment. Trees with higher densities of *C. xenoplax* had a higher incidence of canker. The nematophagous fungus *Hirsutella rhossiliensis* was not detected until the fourth year. Thereafter, the incidence of *H. rhossiliensis* and percentage *C. xenoplax* parasitized by *H. rhossiliensis* increased, but the increases lagged behind increases in numbers of nematodes. In an almond orchard with an initial soil pH of 4.6, preplant application of 0, 6.4, 12.8, or 25.0 kg lime/tree site altered soil pH (range after 1 year = 4.7–7.1). Numbers of *C. xenoplax* remained low (<20/100 cm^3 soil), whereas numbers of *Paratylenchus* sp. increased to high levels (>500/100 cm^3 soil), regardless of treatment. Low levels (<20/100 cm^3 soil) of *H. rhossiliensis*-parasitized *Paratylenchus* sp. were detected. No bacterial canker occurred, but tree circumference was greater after 6 years if soil pH was intermediate (6.0–7.0).
Key words: almond, biocontrol, biological control, *Criconemella xenoplax, Hirsutella rhossiliensis,* lime, nematode, *Paratylenchus* sp., peach, *Prunus dulcis, Prunus persica,* soil pH.

The ectoparasitic nematode *Criconemella xenoplax* predisposes stone fruit trees to bacterial canker in California (4,5,11,15) and to bacterial canker and cold injury in the southeastern United States (1,14,21, 22). From about 1965 to 1977, growers fumigated soil before and after planting to control the nematode and bacterial canker (18–22). In 1977, the registration for dibromochloropropane, the only fumigant used after planting, was suspended, and fumigation was limited to applications of 1,3-dichloropropene or methyl bromide before planting. The registration for methyl bromide, however, is currently under review, and 1,3-dichloropropene is no longer registered in California.

High densities of *C. xenoplax* and incidences of bacterial canker of peach (*Prunus persica*) and almond (*Prunus dulcis*) tend to occur in sandy soils (1,15), which often are acidic due to fertilization and leaching. Weaver and Wehunt (17) found that increasing soil pH with lime reduced the incidence of bacterial canker on peach seedlings grown in pots, and Wehunt and Weaver (18) reported that hydrated lime suppressed *C. xenoplax* in a peach orchard in Georgia. In contrast, a preliminary report (6) indicated that canker was not suppressed by lime in a California peach orchard, and numbers of *C. xenoplax* were not affected by soil pH in several studies (10,17,19).

Because growers have few methods for controlling bacterial canker, we reexamined the effect of liming on the incidence of this disease, numbers of *C. xenoplax*, and parasitism of *C. xenoplax* by the fungus *Hirsutella rhossiliensis* in a peach orchard and in an almond orchard in California.

Materials and Methods

Two orchards were selected in areas with a history of bacterial canker in Merced County, California. The peach or-

Received for publication 12 August 1993.
[1] Supported in part by grants from the California Cling Peach Advisory Board and the University of California Statewide IPM Program.
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We thank J. E. DeVay, H. English, B. F. Lownsbery, and E. I. Zehr for valuable comments and J. Braun for advice on statistical analysis.

chard soil (78% sand, 14% silt, and 8% clay; <1% organic matter; pH 4.9 in 0.01 M $CaCl_2$) had been fumigated with 1,3-dichloropropene in 1984 and planted in 1985. In 1986, 60 replant sites, representing 18% of the orchard, were identified. The trees in these sites had died, but the cause of death had not been determined. In January 1987, soil from each replant site (1.8 m^2 × 1.5 m deep) was removed with a backhoe, and lime (0, 13.2, 18.2, 27.3, or 54.2 kg/site) was mixed into the soil before it was returned to the hole. In a sixth treatment, lime (18.2 kg/site) was raked into the upper 5 cm of soil after the soil was returned to the hole. The lime contained 80% $CaCO_3$ and 2% $MgCO_3$. The quantity of lime applied per site was based on the SMP buffer method (13), and our goal was to achieve a range of soil pH values. There were 10 replications (one tree per replication) in a randomized block design. The tree sites were not fumigated after lime was applied. Peach trees ('Dr. Davis' on 'Nemaguard' rootstock) were planted in February 1987. Nemaguard is a good host for *C. xenoplax* (e.g., 14), and cultivars grafted onto Nemaguard rootstock are often susceptible to bacterial canker (1,15), but the relative susceptibility of peach and almond cultivars is not known (15).

The almond orchard location (82% sand, 15% silt, and 3% clay; <1% organic matter; pH 4.6 in 0.01 M $CaCl_2$) had been fumigated with 1,3-dichloropropene and planted with sweet potato (*Ipomoea batatas*) each year for three consecutive years (1982–84). Almond trees were planted in 1985. In 1986, 45 replant sites, representing 18% of the orchard, were identified. As in the peach orchard, the trees had died, but the cause of death had not been determined. In January 1987, the soil was removed, and lime (0, 6.4, 12.8, or 25.0 kg/site) was added as in the peach orchard. In a fifth treatment, lime (12.8 kg/site) was raked into the soil after the soil was returned to the hole. There were nine replications (one tree per replication) in a randomized block design. After the soil was returned to the hole, each site was fumigated with 454 g methyl bromide. Almond trees ('Nonpareil', 'Carmel', and 'Fritz' on 'Nemaguard' rootstock) were planted in March 1987.

Both orchards were maintained by the growers, who applied no lime or nematicide after planting. The peach orchard was drip-irrigated, and the almond orchard was flood-irrigated.

Soil samples were collected in April of 1987–1993 in the peach orchard and in April 1987–1992 in the almond orchard. Two soil cores (2 cm d by 100 cm deep) were collected from each tree, 33–50 cm from the trunk. The two cores were combined to make one sample, except that cores from sites treated with surface lime were collected at three depths (0–33, 34–66, and 67–100 cm) until discontinued after the fourth year. The samples were kept at 10 C for 24–48 hours before extraction. After mixing, a 500-cm^3 subsample was processed by elutriation (2) and centrifugation (9), and aliquots of the extracted sample were treated with NaOCl, rinsed, and spread onto water agar amended with streptomycin (8). We determined the numbers of *C. xenoplax* (parasitized or not parasitized by *H. rhossiliensis*) and other plant parasites per 100 cm^3 soil. Nematodes were not counted by stage of development, and numbers were not corrected for extraction efficiency. Soil pH was determined after the remaining sample was saturated with 0.01 M $CaCl_2$.

Trees were examined in April and May of each year, and instances of bacterial canker or other problems were noted. Trunk circumferences 20 cm above the soil surface were measured each October.

Trunk circumferences, transformed nematode counts ($\log_{10}[x + 1]$), and transformed percentages of nematodes parasitized by *H. rhossiliensis* (arcsin $x/100$) were regressed on level of lime, but regressions were not significant. Therefore, levels of lime were treated as class variables in an analysis of variance. The general linear models procedure (16) was used for both analyses. Finally, the Wilcoxon 2-sample test (16) was used to determine whether the incidence of canker was related to nematode density.

Results

Peach orchard: Addition of lime altered soil pH for the duration of the experiment in the peach orchard, although the differences between several treatments diminished with time (Fig. 1A). Surface application tended to alter the soil pH only at the surface; for example, pH values 1 year after surface application of lime and at depths of 0–33, 34–66, and 67–100 cm were 7.0, 5.3, and 5.5, respectively.

Numbers of *C. xenoplax* were low for the first 2 years but then increased exponentially (Fig. 1B). Parasitism by *H. rhossiliensis* was not detected until the fourth year, 1 year after numbers of *C. xenoplax* increased greatly. The percentage of trees with at least one *H. rhossiliensis*-parasitized *C. xenoplax* increased from 0% in 1989 to more than 40% in 1993 (Fig. 1C). The percentage of *C. xenoplax* parasitized by *H. rhossiliensis* increased from 1990–93, whether the percentage was based on data from all trees (data not shown) or from only those trees with at least one parasitized nematode over 6 years (Fig. 1C). Numbers of *C. xenoplax* and the numbers and percentages of *C. xenoplax* parasitized by *H. rhossiliensis* were not affected ($P > 0.05$) by levels of lime. Numbers of *Trichodorus* sp. (<50/100 cm^3 soil) and *Paratylenchus* sp. (usually <100/100 cm^3 soil) also were unaffected by lime.

All experimental trees in the peach orchard appeared healthy for the first five growing seasons, and trunk circumference was unaffected ($P > 0.05$) by lime. In the spring of the sixth year, however, the emerging leaves on 10 of the 60 trees wilted, despite adequate soil moisture. Severe cambial discoloration and streaking were observed when bark was removed from main scaffold limbs near the trunk; the exposed bark and cambium had a sour odor. Bark and cambial tissue above and below these cankers appeared healthy, as did the roots and crowns of affected trees. Based on these symptoms, we concluded

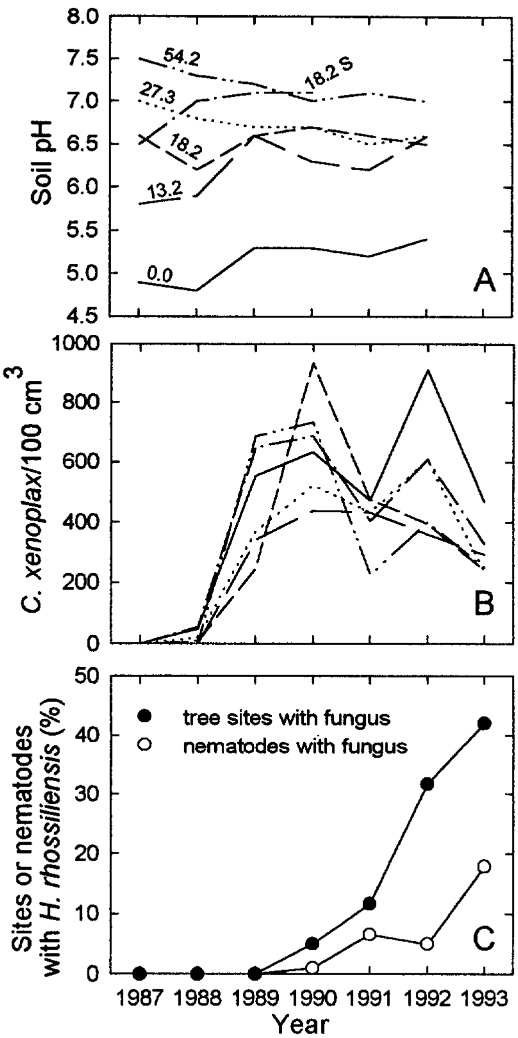

FIG. 1. Effect of lime applications on soil pH, numbers of *Criconemella xenoplax*, and incidence of *Hirsutella rhossiliensis*-parasitized *C. xenoplax* in a peach orchard. A) Soil pH. Numbers indicate the quantity of lime applied (kg per tree site), and S indicates a surface application (lime was mixed into the soil in the other treatments). B) Population fluctuation of *C. xenoplax*; see labels in Fig. 1A. Values for nematode density in the surface application are based on combined data from the three sampling depths. C) Percentage of tree sites with *H. rhossiliensis*-parasitized *C. xenoplax* and percentage of *C. xenoplax* parasitized by the fungus (excluding data from trees that had no parasitized nematodes over 6 years). For A and B, values are means of 10 replicate trees.

that the disease was bacterial canker. Affected trees tended to be clustered in the center of the orchard. The grower removed seven of these trees and removed two to three affected scaffolds from the remaining three trees in the summer of 1992.

Trees with bacterial canker in 1992 were associated with large numbers (averaged over 1989–92) of *C. xenoplax* (Fig. 2), and the incidence of bacterial canker was related ($P = 0.005$) to the numbers of *C. xenoplax*. Among the 10 affected trees, four had received 18.2 kg of lime applied to the soil surface; two received 13.2 kg, one received 18.2 kg, two received 27.3 kg, and one received 54.2 kg of lime incorporated into the soil.

Almond orchard: As in the peach orchard, application of lime affected soil pH values, and differences persisted for the duration of the experiment (Fig. 3A). Surface application tended to alter the soil pH only at the surface; for example, pH values 1 year after surface application of lime and at depths of 0–33, 34–66, and 67–100 cm were 6.0, 5.4, and 4.5, respectively.

Criconemella xenoplax was not detected in the almond orchard until the fifth year, and mean densities never exceeded 19/100 cm^3 soil (Fig. 3B). *Trichodorus* sp. and *Paratylenchus* sp. were present in the almond orchard. Whereas numbers of *Trichodorus* sp. seldom exceeded 50/100 cm^3 soil, those of *Paratylenchus* sp. frequently exceeded 500/100 cm^3 soil in the fourth and fifth years after planting (Fig. 3B). *Paratylenchus* sp. parasitized by *H. rhossiliensis* were observed occasionally (<20/100 cm^3 soil).

No bacterial canker was observed, and almond trees remained healthy. Trunk circumferences in 1991, the last year in which trunks were measured, were not affected by block or cultivar but were higher ($P < 0.05$) when intermediate levels of lime were incorporated into the soil (Fig. 3C).

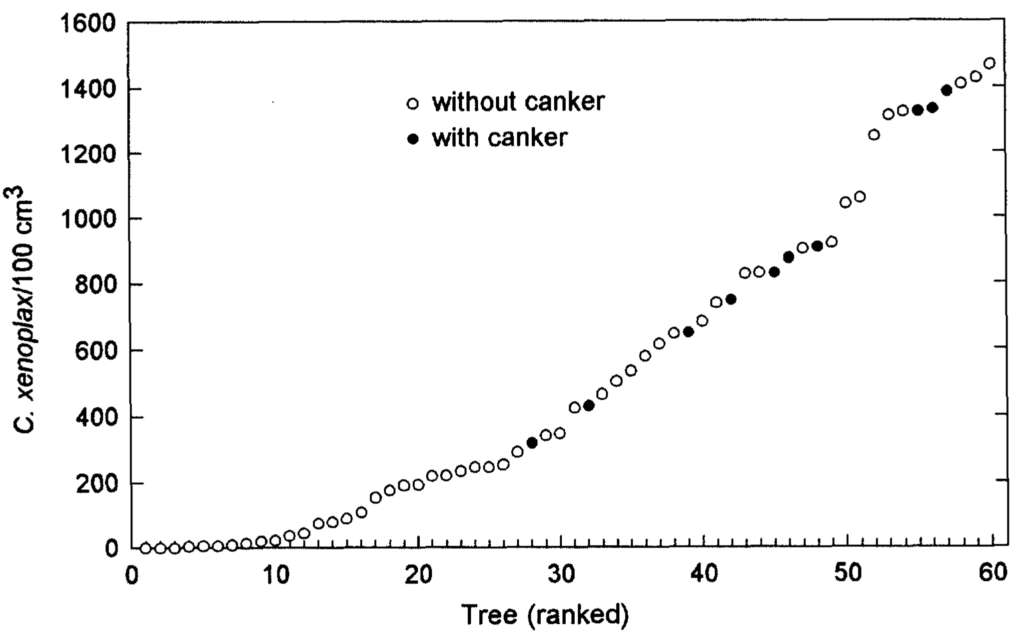

FIG. 2. Relationship between numbers of *Criconemella xenoplax* and incidence of bacterial canker in a peach orchard. The *y* axis indicates density of *C. xenoplax* averaged over 4 years (1989–1992), omitting data from 1987 and 1988, when nematode counts were low in all plots, and data from 1993, 1 year after the severe incidence of canker. For trees that received surface lime, nematode numbers were averaged across depth. On the *x* axis, the 60 trees in the experiment are ranked from low to high with respect to nematode density. Apparently healthy trees and those with canker are represented by hollow and filled circles, respectively. The relationship between nematode density and incidence of canker was significant ($P = 0.005$) according to the Wilcoxon 2-sample test.

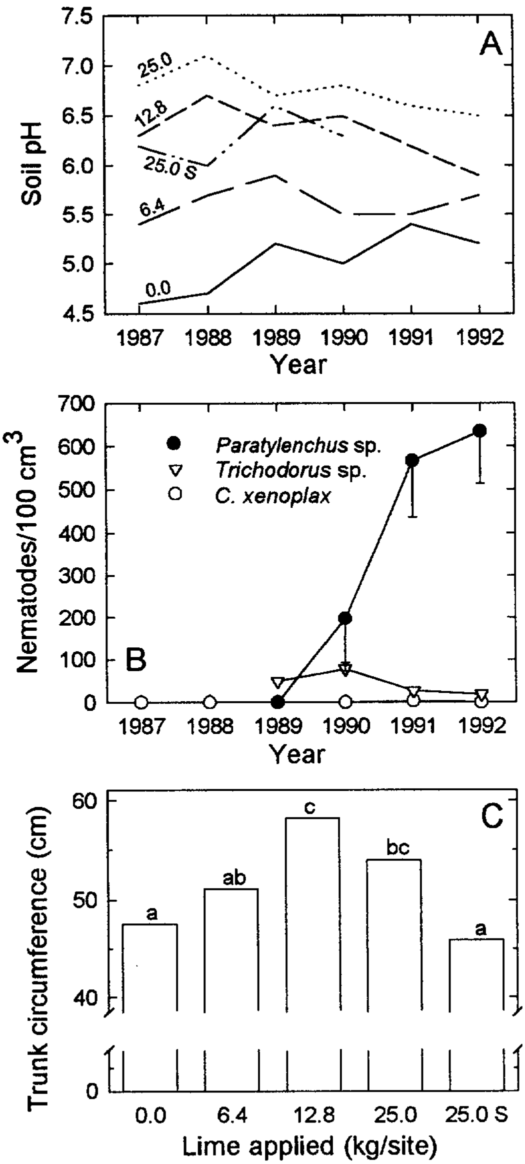

FIG. 3. Effect of lime application on soil pH, numbers of *Criconemella xenoplax* and *Paratylenchus* sp., and trunk circumference in an almond orchard. A) Soil pH. Numbers indicate the quantity of lime applied (kg per tree site), and S indicates a surface application (lime was mixed into the soil in the other treatments). B) Population fluctuations of *C. xenoplax* and *Paratylenchus* sp. averaged over all treatments. Vertical lines indicate 1 SE. C) Circumference of trunks. Values with a similar letter do not differ ($P > 0.05$) according to Duncan's multiple-range test. Values are means of 10, 45, and 10 replicate trees for A, B, and C, respectively.

Surface application of lime tended to decrease rather than increase trunk circumference (Fig. 3C).

Discussion

Our test orchards were located in a region with a history of bacterial canker; thus we expected canker to occur at both orchards. Because growers and farm advisors had told us that trees 2–5 years old were most susceptible to canker, we became concerned that our experiment would not address canker control at all when no canker was observed through the fifth year. At that time, the manager of the peach orchard told us that he was pleased with the apparent health of his trees and that perhaps drip irrigation was the key to suppressing bacterial canker. Without any signal detectable by us, severe canker occurred in the sixth year. The sudden appearance of symptoms and rapid decline of apparently healthy trees has been described before (1).

Canker might have been more severe or may have occurred sooner had the soil profile not been disrupted to 1.5 m before planting (7,15) and had the orchard site not been fumigated. Although proper soil pH is important for tree growth (e.g., 3) and may suppress bacterial canker (1,17, 18), liming with $CaCO_3$ did not suppress *C. xenoplax* or canker in this or other (6,19) studies.

The incidence of canker in the peach orchard was related to *C. xenoplax* density (Fig. 2), even though some trees with high levels of *C. xenoplax* remained canker free. Bacterial canker is a complex disease (15), and we do not understand it sufficiently to predict when it will occur, but our data confirm other reports (5,11,14,18–22) that indicate an important role for *C. xenoplax*.

Hirsutella rhossiliensis was present in the peach orchard, but fungal parasitism appeared to lag behind increases in nematode numbers. If the fungus did suppress nematode population density, the suppression did not maintain *C. xenoplax* at low levels.

We expected high numbers of *C. xenoplax* in the almond orchard because the soil is sandy and the orchard is surrounded by others with high numbers of *C. xenoplax*.

But the 3 years of sweet potato plus four fumigations that preceded establishment of the orchard may have reduced *C. xenoplax* to numbers so low that recovery was slow. Competition between *Paratylenchus* sp. and *C. xenoplax* also may be involved; McKenry and Kretch (12) reported that low numbers of *C. xenoplax* were correlated with high numbers of *Paratylenchus* sp. The absence of bacterial canker in the almond orchard may reflect the low levels of *C. xenoplax* persisting throughout the test period.

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