Research Updates

Hot-water Dipping Eradicates Phylloxera from Grape Nursery Stock

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Additional index words.
Daktulosphaira vitifoliae, rootstock

Summary. **All life stages of grape phylloxera [*Daktulosphaira vitifoliae* (Fitch) (Homoptera: Phylloxeridae)] were eradicated with a hot-water treatment (dip) of 5 minutes at 43 °C (110 °F) to warm roots, followed by a 5-minute dip at 52 °C (125 °F). Neither grafted nor nongrafted dormant grape plants were damaged by the hot-water treatment.**

Grape phylloxera is the most serious insect pest of vineyards worldwide (Flaherty, 1992). This pest first was discovered in commercial vineyards in Oregon in 1990 (Connelly and Strik, 1991). There is no control for this pest other than replanting to vines grafted to resistant rootstocks. The easiest way to introduce grape phylloxera into a vineyard is by planting infested grafted or self-rooted nursery stock. If nurseries could eradicate phylloxera effectively without damaging planting stock, introducing phylloxera to new areas would be eliminated (Strik and Stonerod, 1995). Hot-water dipping has been used to control nematodes (Lear and Lider, 1959), crown gall (*Agrobacterium tumefaciens*) (Ophel et al., 1990), and certain life stages of phylloxera (Davidson and Nougaret, 1921; Flaherty, 1992) from nursery stock. However, depending on water temperature and length of treatment, there has been damage to grapevine cuttings (Wample et. al., 1991). Our objectives were to determine methods of dipping young nongrafted and grafted grapevines (nursery stock) to eradicate existing phylloxera populations without causing plant damage.

Materials and methods

Hot water dipping of phylloxera. In Jan. 1994 and 1995, phylloxera life stages (eggs, nymphs, nymph–adults, adults, and hibernants) (Flaherty, 1992) were dipped in hot water (52 °C; 125 °F) to determine percent survival. Phylloxera populations were maintained on 'Pinot Noir' (*Vitis vinifera L.*) root pieces in petri dishes in laboratory incubators (20 °C) before conducting hot-water dips. The root pieces were 3 to 5 mm in diameter and 5.0 to 7.5 cm long. All life stages were present in the populations living on the root pieces used for dipping. To obtain hibernants, young nymphs were forced into hibernation through depletion of food (no new root pieces) and by reduction of temperature (16 °C).

When populations of all life stages of phylloxera increased sufficiently, root pieces with populations were selected for hot-water dipping. A precount of each life stage was noted for each root piece. There were seven time lengths for dipping with three replicates each (total of 21 root pieces). The roots plus associated phylloxera populations were dipped for 0, 3, 5, 7, 9, 11, and 13 min at 52 °C in a hot-water bath.

Each root piece was placed on a large piece of filter paper (Whatman no. 1), folded and stapled in a teabag fashion. This package was covered by a second piece of filter paper to prevent tearing. The root piece was confined loosely within the filter paper so that the insects were not harmed, yet the they also could not escape when dipped in water. Three root pieces individually wrapped in filter paper then were bundled in cheesecloth with a piece of thread tied around the package to provide a handle for dipping.

After hot-water dipping, the outside covering of filter paper was removed and the paper inside was unfolded to expose the root piece. Root pieces and associated filter paper were placed in individual petri dishes in an incubator (20 °C). Every 7 d, the number of dead and live phylloxera in each life stage were counted. Percent survival was calculated for each treatment. Populations were maintained and checked for survival for 42 d.

In 1995, phylloxera populations were prepared and maintained as in 1994. However, based on results in 1994, a control, in which root pieces and associated phylloxera populations were dipped in water at room tempera-

Table 1. Number of nodes and percent budbreak of nongrafted vines dipped in 20 °C (68 °F) water for 10 min or hot water for 10 min [43 °C (110 °F) water for 5 min + 52 °C (125 °F) for 5 min]. The treatment effect was nonsignificant; means were averaged to show the significant rootstock effect.

Rootstock	Nodes (no.)	Budbreak (%)
5C	6.7	75
101-14	5.4	79
3309C	6.3	96
Freedom	6.1	78
Pinot noir	6.2	79
$LSD_{0.05}$	0.55	15.0

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ture (20 °C) for 5 min, was compared to a hot-water dip for 5 min at 52 °C. There were three root pieces per replicate and 10 replicates per treatment. Percent survival for each life stage was calculated every 7 d for 42 d.

HOT-WATER DIPPING OF PLANTS. In Winter 1994, 2-year-old dormant, self-rooted plants ('Pinot Noir' and the rootstocks 3309C, 5C, 101-14, and Freedom) growing in 1-gallon containers were used for the hot-water dipping study.

Just before dipping, plants were removed from pots, and roots were washed free of all soil. Roots were pruned to ≈15 cm long and shoots to six to seven buds. One plant of each cultivar was tied into a bundle (replicate) with 10 replicates per treatment.

Hot-water treated plants were dipped for 5 min at 43 °C to preheat roots then in 52 °C water for 5 min. Preheating roots aides in maintaining a constant temperature (Goussard, 1977). The control plants were dipped in water at 20 °C for 10 min. All plants then were dipped in cold water (14 °C) for 2 min. To evaluate damage, plants were repotted and placed in a greenhouse. After budbreak, the numbers of nodes and primary, secondary, and tertiary shoots were counted to evaluate treatment effects.

In 1995, the response of bareroot grafted nursery vines to the hot-water dipping was evaluated using the same procedure as described for 1994. The rootstock–scion combinations used were 101-14–'Pinot Noir' clone Pommard, 3309C–'Chardonnay' clone Draper, and *Riparia gloire*–'Chardonnay' clone 76. One plant of each was bundled together; there were 10 replicates per dipping treatment.

After dipping, plants were set in flats of sawdust and placed in a screenhouse. To evaluate treatment effects, nodes and shoots were counted on each vine. The graft union of each plant then was sliced longitudinally and was evaluated for discoloration of pith and cambium tissues using a rating system from 1 to 4 (pith, 1 = cream or light green, 2 = tan, 3 = light brown, and 4 = dark brown or black; cambium, 1 = bright green, 2 = olive green, 3 = light brown, and 4 = cream. Data were subjected to analysis of variance, and means were compared using a protected least significant difference (SAS Institute, Cary, N.C.).

Results and discussion

GRAPE PHYLLOXERA. Data collected in 1994 on percent survival and amount of time to kill each life stage indicated that all life stages of phylloxera were killed with a 5-min dip in 52 °C water (data not shown). The 3-min dip was not totally effective at killing all individuals. About 21% of the eggs, 60% of the nymphs, and 15% of the adults survived for ≤7 d after dipping. At 14 d after dipping, insects subjected to a 3-min hot-water dip appeared to be dead. Davidson and Nougaret (1921) found a 1-min dip at 52 °C killed hibernants and eggs. Evaluation from 7 to 42 d after dipping for the 5-, 7-, 9-, 11-, and 13-min dips indicated all insects were dead. All stages in the control group were alive; eggs were hatching, nymphs and nymph-adults were molting, adults were laying eggs, and hibernants were becoming active.

The 1995 data confirmed that all life stages of phylloxera were killed when dipped in hot water (5 min at 43 °C + 5 min at 52 °C] and survived when dipped in 20 °C water for 10 min (data not shown). From 7 to 42 d after dipping, insects dipped in hot water were dead; insects in the control group were hatching, molting, reproducing, and breaking dormancy.

PLANTS. In 1994, data on percent budbreak of primary, secondary, and tertiary shoots showed that no plants displayed adverse effect from hot-water dipping ($P > 0.05$). However, there was a significant difference in percent budbreak among rootstocks (Table 1).

In 1995, there were no adverse effects of hot-water dipping on the graft union (based on a visual browning rating) or percent budbreak (Table 2). However, rootstock–scion plants did differ in overall ratings and percent budbreak (Table 2).

It is important to note that our objectives were to find treatments requiring the least amount of time in hot water required to kill all life stages of phylloxera and, thus, subject plants to the minimum possible contact with hot water. A hot-water dip of 5 min at 43 + 52 °C was sufficient to kill all stages of phylloxera and did not injure dormant plants.

Other experiments have indicated that bud injury from hot-water treatment is minimal at 52 and 54 °C with time lengths of 10, 20, or 30 min. Critical injury from dipping nonrooted cuttings appears to occur at 56 °C for 30 min. (Wample, 1993). In an experiment on using hot-water dips to eliminate crown gall, Wample (1991) found that percent budbreak after hot-

Table 2. Effect of hot-water dipping of grafted vines on scion budbreak and graft union damage in 1995.[z]

Scion–rootstock[y]	Dipping treatment[x]	Visual rating Pith[w]	Visual rating Cambium[w]	Shoot no./plant	Budbreak (%)[v]
PN–101-14	C	2.3	1.8	4.6	126
		2.1	1.7	4.9	133
	T	1.8	1.6	5.1	139
CH–3309C	C	2.7	2.4	2.3	79
		2.6	2.3	2.8	95
	T	2.4	2.2	3.3	111
CH–*R. gloire*	C	1.4	1.2	5.2	101
		1.4	1.3	4.9	99
	T	1.5	1.3	4.5	96
$LSD_{0.05}$[u]		0.51	0.42	0.91	26.2
ANOVA[t]					
Block		NS	NS	NS	NS
Dipping		NS	NS	NS	NS
Rootstock		***	***	***	**
D × P		NS	NS	NS	NS

[z] *Means for the main effect of rootstock–scion are underlined.*
[y] *PN = Pinot noir; CH = Chardonnay.*
[x] *C = control, dipped 10 min at 20 °C; T = treated, dipped 5 min at 43 °C + 5 min at 52 °C.*
[v] *Percentages >100 indicate presence of more than one shoot per node.*
[w] *Visual rating: pith, 1 = cream or green, 2 = tan, 3 = light brown, 4 = dark brown or black; cambium, 1 = bright green, 2 = olive green, 3 = light brown, 4 = cream.*
[u] *Least significant difference value is for main effect rootstock–scion combination as there was no dipping treatment effect.*
[t] *ANOVA = analysis of variance .*
NS, **, *** *Nonsignificant or significant at* $P < 0.01$ *or* 0.001, *respectively.*

water treatment at >54 °C depended on cultivar. Budbreak also depended on chilling requirements and storage of vines after dipping (Wample, 1993). Our results indicate that there was no critical plant injury at 52 °C, and a 5-min dip was successful for eradicating grape phylloxera.

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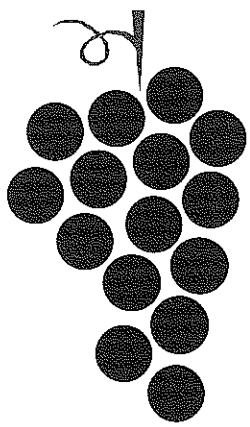

Fertilization Rate and Growth of 'Hamlin' Orange Trees Related to Preplant Leaf Nitrogen Levels in the Nursery

Laura Guazzelli, Frederick S. Davies, and James J. Ferguson

Additional index words. citrus, nitrate, nutrition, soil

Summary. **Our objectives were to determine the effects of leaf N concentration in citrus nursery trees on subsequent growth responses to fertilization for the first 2 years after planting and the impact of N fertilizer rate on soil NO_3-N concentration. 'Hamlin' orange [*Citrus sinensis* (L.) Osb.] trees on 'Swingle' citrumelo rootstock [*C. paradisi* Macf. x *P. trifoliata* (L.) Raf.] were purchased from commercial nurseries in Apr. 1992 (Expt. 1) and Jan. 1993 (Expt. 2) and were grown in the greenhouse at differing N rates. Five months later, trees for each experiment were separated into three groups (low, medium, and high) based on leaf N concentration and were planted in the field in Oct. 1992 (Expt. 1) or Apr. 1993 (Expt. 2). Trees were fertilized with granular material (8N–2.6P–6.6K–2Mg–0.2Mn–0.12Cu–0.27Zn–1.78Fe) with N at 0, 0.11, 0.17, 0.23, 0.28, or 0.34 kg/tree per year. Soil NO_3-N levels were determined at 0- to 15- and 16- to 30-cm depths for the 0.11-, 0.23-, and 0.34-kg rates over the first two seasons in Expt. 2. Preplant leaf N concentration in the nursery varied from 1.4% (Expt. 1) to 4.1% (Expt. 2) but had no effect on trunk diameter, height, shoot growth and number, or dry weight in year 1 (Expt. 1) or years 1 and 2 (Expt. 2) in the field. Similarly, fertilizer rate in the field had no effect on growth during year 1 in the field. However, trunk diameter increased with increasing N rate in year 2 and reached a maximum with N at 0.17 kg/tree per year but decreased at higher rates. Shoot number during the second growth flush in year 2 was much lower for nonfertilized vs. fertilized trees at all rates, which had similar shoot numbers. Nevertheless, leaf N concentrations increased during the season for trees with initially low levels, even for trees receiving low fertilizer rates. This suggests translocation of N from other organs to leaves. Soil NO_3-N levels were highest for the 0.34-kg rate and lowest at the 0.11-kg rate. Within 2 to 3 weeks of fertilizing, NO_3-N levels decreased rapidly in the root zone.**

Several young-tree fertilizer studies have been conducted on citrus in Florida. Optimum N rates per tree for the first year that trees are in the field in Florida were reported to be 56 (Rasmussen and Smith, 1961), 70 (Marler et al., 1987), 108 (Obreza and Rouse, 1993), and 230 g (Willis et al., 1990). Moreover, in several studies (Obreza and Rouse, 1993; Rasmussen and Smith, 1961), fertilization rate had no effect on tree growth during the first year in the field. Furthermore, the same lack of response to fertilizer rate occurred during the second year in studies by Rasmussen and Smith (1961) and Calvert (1969). In contrast, Obreza and Rouse (1993) found tree growth increased with increasing fertilization rate.

Differences in responses to fertilization by young citrus trees may result from several factors, including soil type (Obreza and Rouse, 1993), tree age and size (Calvert, 1969), rootstock (Wutscher, 1989), amount of stored reserves (Legaz et al., 1995), or type of nursery trees (bareroot or container grown) (Davies, unpublished). Some or all of these factors may account for the variation in fertilizer responses observed in field experiments. In our previous studies (Marler et al., 1987; Willis et al., 1990), leaf nutrient levels, particularly N, varied considerably among nursery trees before planting and appeared to be a possible reason for differences in fertilizer responses.

Concern over high soil NO_3-N in some areas of Florida also have caused growers to re-evaluate fertilization rates

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(McNeal et al., 1994; Willis et al., 1990). Soil NO_3-N levels >50 mg·kg^{-1} of soil have been measured below the root zone of young citrus trees (Willis et al., 1990). Therefore, monitoring NO_3-N levels has become an important part of any fertilizer rate study because overfertilization may lead to increased NO_3 levels in groundwater.

Our objective was to determine if there is a carryover effect of greenhouse nutrition on subsequent growth and fertilization response of young citrus trees after planting in the field. We also were interested in learning if "loading" trees with N in the nursery would reduce fertilizer rates, costs, and soil NO_3-N levels in the field.

Materials and methods

EXPERIMENT 1. One hundred 'Hamlin' orange trees on 'Swingle' citrumelo rootstock were obtained from Revette Nurseries, Waverly, Fla., on 9 Mar. 1992. The trees were growing in 10 × 10 × 35-cm citripots in a commercial medium (1 peat moss : 1 perlite, v/v; limestone and superphosphate at 8.71 and 0.083 kg·m^{-3}) and averaged 50 cm in height. Trees were placed in a greenhouse, and composite leaf samples (consisting of 50 mature leaves) were collected randomly from the previous season's spring flush shoots of several trees. Leaf N concentration was determined using total Kjeldahl and an inductively coupled argon plasma spectrophotometer as described by Maurer and Davies (1993). Average leaf N concentration was 2.8% dry weight. The trees then were divided into groups of 33, 33, and 34 trees that received N at 0, 12, or 100 mg·L^{-1} per application weekly, respectively. Treatments were initiated on 27 Apr. 1992 and continued until trees were transplanted into the field in Oct. 1992. Liquid fertilizer was applied using a 2% fixed Dosatron injector (Dosatron International, Clearwater, Fla.). The irrigation system was run for 7 min with the last 2 min of irrigation used to flush the system. Water samples indicated that no fertilizer remained in the system after flushing. Trees were irrigated using one 3.8-L·h^{-1} dripper per container every 2 d to replenish water lost to transpiration and to return the medium to container capacity. No leaching of nutrients was observed at this irrigation rate and frequency and for this size tree.

Fully expanded 3- to 4-month-old tagged leaves were sampled from the middle of the shoot on 28 Aug. 1992 and were analyzed for N concentration as previously described. Trees then were grouped according to leaf N concentrations: low (1.4% to 1.6% N), medium (1.7% to 1.9% N), and high (2.0% to 2.2% N). Trees were selected for uniformity of height and trunk diameter and were divided into three groups of 18 trees each.

The field study consisted of three preplant leaf N concentrations and six postplant fertilizer rates arranged in a randomized complete-block design with three blocks and three individual tree replicates per treatment (54 trees total). Trees were planted at 2.8 × 6.1 m within and between rows, respectively, on 14 Oct. 1992 at the Fifield Farm, Gainesville, Fla. Granular fertilizer (8N–2.6P–6.6K–2Mg–0.2Mn–0.12Cu–0.27Zn–1.78Fe) was broadcast slightly outside the dripline with N at 0.056, 0.11, 0.17, 0.23, 0.28, and 0.34 kg/tree split into six applications per calendar year. Fertilizer was applied on 31 Oct. 1992 and 14 Feb., 27 Mar., 1 May, 12 June, 24 July, and 4 Sept. 1993. Soil type was Arredondo fine sand (loamy, siliceous, hyperthermic, Grossarenic Paleudults). Initial tree height, trunk diameter (10 to 15 cm above the bud union), and

Table 1. Shoot count for young 'Hamlin' orange trees related to flush, fertilizer rate, and preplant leaf N concentration (Expt. 2), 1994. Mean of four shoots per tree for three individual tree replicates per treatment.

	Shoot (no.)					
	Preplant leaf N (%)					
N (kg/	Low (3.1)		Medium (3.6)		High (3.8 to 4.1)	
tree/year)	Flush 1	Flush 2	Flush 1	Flush 2	Flush 1	Flush 2
0.0	152	18	117	3	91	0
0.11	133	25	118	7	115	15
0.17	131	7	141	19	114	12
0.23	146	7	110	8	98	24
0.28	102	1	98	16	66	30
0.34	57	4	128	4	103	1
Significance	*	*	*	*	*	*

*Significant at $P \leq 0.05$.

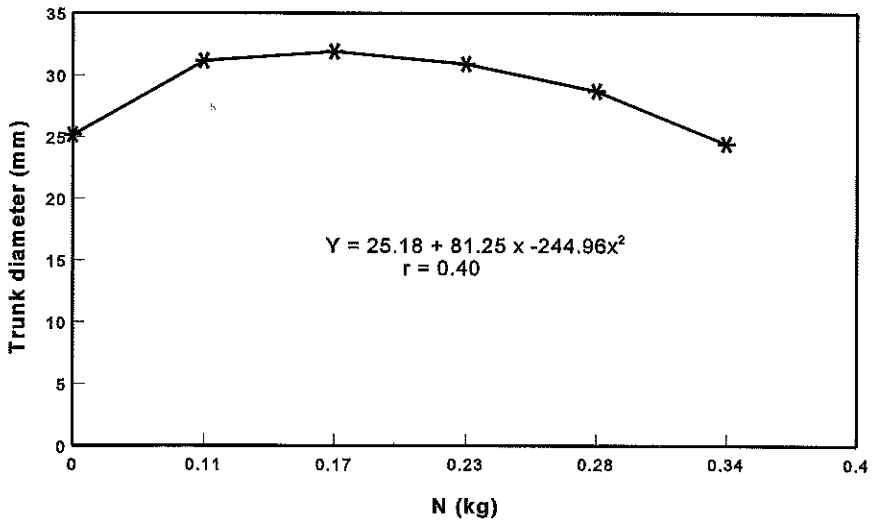

Fig. 1. Fertilizer rate effects on trunk diameter of 2-year-old 'Hamlin' orange trees at Gainesville, Fla., 1994. Each data point is the mean of nine single-tree replications derived by combining three replicates of three preplant leaf N concentrations.

leaf count were recorded. In addition, visual evaluations were made weekly to determine time and distribution of each growth flush. Monthly trunk diameter and tree height measurements were made from 10 Apr. until 10 Sept. 1993. Ten 4- to 5-month-old spring-flush leaves were collected per tree on 23 Aug. for nutrient analysis (N–P–K–Ca–Mg–Fe–Mn–Zn–Cu) (Maurer and Davies, 1993). Trees were harvested above the soil line between 1 and 4 Oct. 1993. Shoot length was determined after harvest based on measurements of four shoots per growth flush per tree at harvest time. Shoot number per flush and fresh and dry weights also were determined for each tree.

All trees received the same amount of water using 90° 38-L·h^{-1} microsprinklers located 1 m northwest of the trunk (Marler and Davies, 1990). During the first 2 weeks after planting, trees were irrigated every 2 d. Trees then were irrigated at 30% soil water depletion (SWD) for 1.5 h as described by Marler and Davies (1990). Soil water deficit was determined using a neutron probe (model 4300; Troxler, Research Triangle Park, N.C.). Aluminum access tubes were placed 30 cm from the trunk with two tubes per block (six tubes total). Measurements of SWD were taken at a 30-cm depth twice weekly if no rain occurred. Fruit were removed from trees after the initial fruit set period.

Experiment 2. One hundred 'Hamlin' orange trees on Swingle citrumelo rootstock were obtained from Reed Brothers Nursery, Dundee, Fla., on 13 Jan. 1993 and were placed in the greenhouse. A composite leaf sample was taken from the trees on 19 Jan. to determine the initial leaf N concentration. The procedure and type of leaves sampled were the same as described in Expt. 1. Initial leaf N concentration averaged 4.6%. The plants were divided into groups of 33, 33, and 34 trees that received N at 0, 50, or 100 mg·L^{-1} per application weekly, respectively, beginning 21 Jan. 1993. The same liquid fertilizer formulation was used as in Expt. 1. Leaf samples were taken on 17 Apr. as described in Expt. 1 from every tree to determine N concentration. Trees then were separated into three groups based on the N concentration in the leaves: low (3.1%), medium (3.6%), and high (3.8% to 4.1%). Eighteen uniform trees from each N level (54 trees total) were selected and planted on 14 Apr. 1993 at the Fifield Farm. The same experimental and plot designs were used as in Expt. 1; soil series also was the same. Trees were planted at 6.1 m between rows and 3.7 m between trees. Fiberglass tree wraps were placed around tree trunks to prevent sprouting and herbicide injury.

Measurements of tree height, trunk diameter, and leaf count were taken monthly for two growing seasons from 14 Apr. 1993 until 10 Dec. 1994. Trunk diameters were measured 10 to 15 cm above the bud union. Shoot growth was measured by tagging three randomly selected shoots per tree at the beginning of the growth flush and measuring their final length. Granular fertilizer (same formula as Expt. 1) was applied four times yearly with N at 0, 0.11, 0.17, 0.23, 0.28, or 0.34 kg/tree on 18 May, 29 June, 10 Aug., and 21 Sept. 1993 and 17 Mar.,

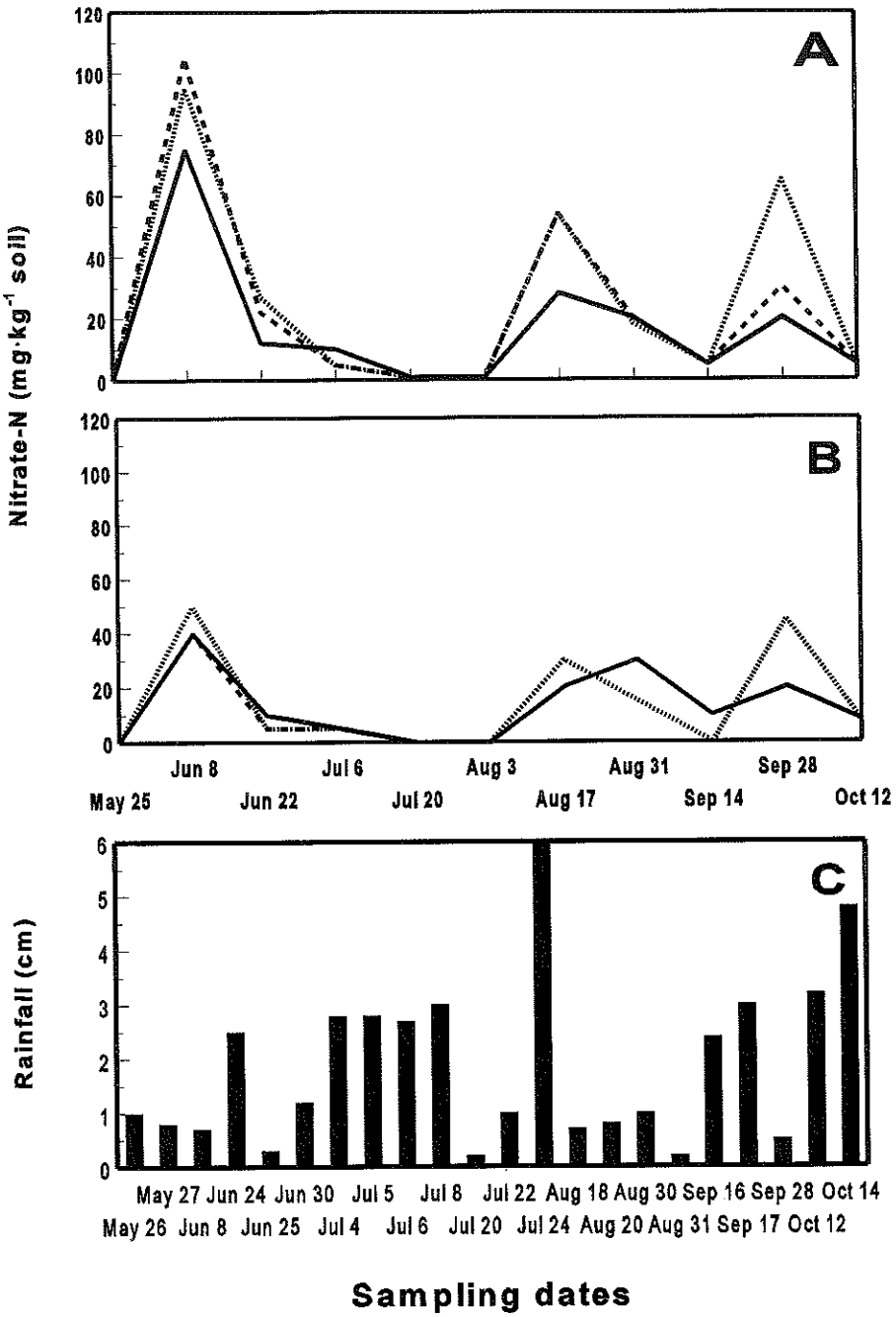

Fig. 2. Seasonal soil NO_3-N concentration at two depths under 1-year-old 'Hamlin' orange trees in response to three fertilizer rates in 1993. Each data point is the mean of four single-tree replications. (A) 0 to 15 cm; (B) 16 to 30 cm; (C) rainfall: N rates at 0.34 (= = =), 0.23 (- - -), and 11 (——) kg.

28 Apr., 9 June, and 21 July 1994. To test the effects of no fertilization on tree growth in the field, some trees in Expt. 2 were not fertilized (instead of being fertilized at the 0.056-kg rate). Ten 4- to 5-month-old spring-flush leaves were collected per tree on 23 Sept. 1994 for nutrient analysis as described in Expt. 1. Irrigation amount and timing was determined as described in Expt. 1.

SOIL ANALYSIS (EXPT. 2). Soil samples were taken to determine NO_3-N concentration for the 0.11-, 0.23-, and 0.34-kg N rates using four randomly selected single-tree replicates per treatment. Samples were taken at 0- to 15- and 16- to 30-cm depths with an auger (2.5 cm in diameter) from soil within a 30-cm radius of the tree. Holes left from sampling were refilled with soil. Soil samples were placed in paper bags and taken immediately to an oven and dried at 40 °C until they reached a constant weight. Forty ml of distilled water was added to a 20-g aliquot of soil. The solutions were shaken vigorously, allowed to stand for 4 h and filtered through Whatman no. 42 filter paper. Filtrates were analyzed for NO_3-N on an air-segmented spectrophotometer (rapid flow analyzer model RDF300; Alpken Corp., Silver Spring, Md). To follow the movement of nutrients over time, soil samples were collected 1, 3, and 5 weeks after fertilizer application. Samples were collected on 25 May; 8 and 22 June; 6 and 20 July; 3, 17, and 31 Aug.; 14 and 28 Sept.; and 12 Oct. 1993 and 24 Mar.; 7 and 21 Apr.; 5, 12, and 26 May; 16 and 30 June; 14 and 28 July; and 25 Aug. 1994.

Experiments 1 and 2 were analyzed separately using analysis of variance (ANOVA) and regression analysis by time, fertilizer rate, preplant leaf N concentration, and appropriate interactions.

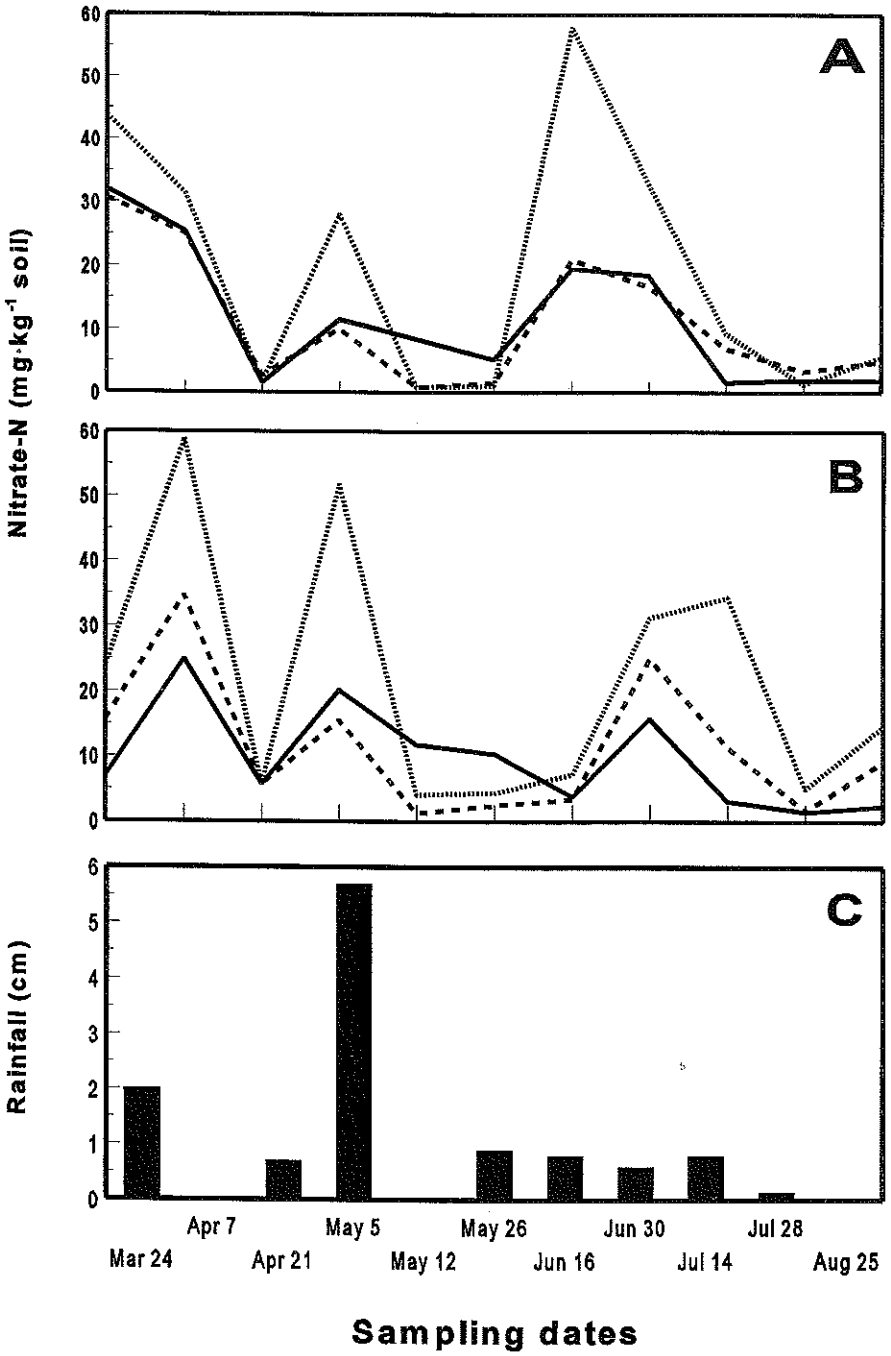

Fig. 3. Seasonal soil NO_3-N concentration at two depths under 2-year-old 'Hamlin' orange trees in response to three fertilizer rates in 1994. Each data point is the mean of four single-tree replications. (A) 0 to 15 cm; (B) 16 to 30 cm; (C) rainfall: N rates at 0.34 (= = =), 0.23 (- - -), and 11 (—) kg.

Results and Discussion

EXPERIMENT 1. There was no significant effect of preplant leaf N concentration on tree fresh weight, dry weight, flush count or length, or trunk diameter in 1993 (year 1) (data not shown). In addition, fertilization rate had an inconsistent effect on these growth characteristics, although there was a negative correlation ($y = 19.8 - 13.3x$, $r = -0.69$) for trunk diameter and N fertilizer rate for the intermediate preplant treatment (data not shown). There were no visual differences in tree vigor or appearance at the end of the first year.

Leaf N concentration in August was similar for all treatments (2.6% to 2.8%), except at the 0.056 rate (2.2%). Therefore, leaf N levels increased for trees with initially low N (1.4%) and increased or remained the same for those with initially high leaf N (2.2%).

EXPERIMENT 2. As in Expt. 1, no significant effect of preplant N concentration was observed on tree trunk diameter, height, shoot length, or shoot number in 1993 (data not shown) or 1994 (Table 1). Similarly, fertilizer rate in the field had no consistent effect on trunk diameter, tree height, or shoot count or length in 1993. However, in 1994 fertilizer rate had a significant quadratic effect on trunk diameter for all three preplant N

levels (Fig. 1). Data were combined for the three preplant N concentrations for each fertilizer rate. Trunk diameter was greatest with N at 0.11 to 0.17 kg and least at the 0 and 0.34 kg. Similarly, the number of second growth flushes generally was less at the 0- and 0.54-kg rates than at the intermediate rates (Table 1). Six of nine (includes all three preplant N treatments combined) nonfertilized trees had no second growth flush. Therefore, it was not until the end of the second season that the effects of lack of fertilization became apparent in this soil series. These optimum rates are in agreement with those determined by Obreza and Rouse (1993) in southwestern Florida and are slightly higher than those determined by Marler et al. (1987) in a different soil type in northern Florida. These data also demonstrate the disadvantages of using high fertilization rates that decrease growth and increase fertilization costs.

In September, leaf N concentration averaged 3.4% and was similar for all fertilization rates, except for the nonfertilized treatment, which averaged 2.8%. Leaf N concentrations for young trees are higher than those found in mature trees, and these values are similar to those reported by Willis et al. (1990) and Maurer and Davies (1993). Preplant leaf N concentration increased during the season for the low preplant treatment (3.1%), remained about the same for the intermediate preplant level (3.6%), or decreased for the high preplant level (3.8% to 4.1%). These data and those from Expt. 1 suggest that trees with low initial leaf N may increase N uptake and those with high initial leaf N decrease uptake as suggested by Syvertsen and Smith (1996). Alternatively, Legaz et al. (1995) found that N was translocated from old leaves, trunks, and roots to new organs, which could account for similarities in leaf N concentrations.

Soil NO_3-N (Expt. 2). In 1993, soil NO_3-N levels were highest for the 0.34-kg N rate, intermediate for the 0.23-kg N rate, and lowest for the 0.11-kg N rate (Fig. 2a). The highest rate had peak levels within 1 week of fertilization of 50 to 100 mg·kg^{-1} at the 15-cm depth. The 0.23- and 0.11-kg N rates had peak NO_3-N levels of 25 to 75 mg·kg^{-1}. Nevertheless, NO_3-N levels decreased to 10 mg·kg^{-1} within 2 weeks of fertilization for all three rates. A similar pattern occurred at the 16- to 30-cm depth, but NO_3-N levels were lower than at the 15-cm depth (Fig. 2B).

Nitrate-N levels were low at both depths following heavy rainfall periods in June and July (Fig. 2C). In fact, no peak occurred following the 12 June fertilizer application, strongly suggesting leaching to below the root zone.

Soil NO_3-N patterns in 1994 were similar to those in 1993 (Fig. 3). The highest rate produced the greatest soil NO_3-N levels, followed sequentially by the other rates (Fig. 3a and b) for both depths. Again, soil NO_3-N levels decreased rapidly within 2 to 3 weeks of fertilization. Heavy rainfall from June to 22 July 1993 (21.0 cm) and 5 May to 10 June 1994 (13.5 cm) reduced soil NO_3-N levels considerably.

These soil NO_3-N data are in agreement with similar studies by Willis et al. (1990) and support using reduced N rates for fertilizing young trees in Florida. They also suggest relatively rapid uptake or losses of soil NO_3-N within the root zone. Syvertsen and Smith (1996) found that seedling citrus trees took up 27% to 51% of applied N within 7 d of application, which accounts for a portion of the rapid decrease in NO_3-N within 2 weeks of application. Soil NO_3-N levels also decrease due to volatilization, leaching, and denitrification (McNeal et al., 1994). McNeal et al. (1994) observed large pulses of NO_3-N in groundwater following fertilization of mature citrus trees on similar soils. Nitrate-N dissipated more slowly in their study, probably because they measured levels in ground water beneath the root zone rather than in soil samples within the shallow root zone of a young citrus tree. Nitrate-N concentrations in the soil are not necessarily correlated with NO_3 in the groundwater. There are no established limits for soil NO_3-N as there are for NO_3 in drinking water.

Our hypothesis that preplant leaf N concentration in the nursery affects subsequent growth responses to fertilizer in the field was not supported in either experiment using a wide range of preplant N levels (1.4% to 4.6%). Citrus trees apparently adjust leaf N concentrations to relatively stable levels either by increasing uptake (Syvertsen and Smith, 1996) or reallocating N reserves from trunk, roots, and old leaves (Legaz et al., 1995). Data strongly suggest that fertilization rates for young citrus trees in Florida can be reduced as currently recommended (Tucker et al., 1995) to reduce NO_3-N in the types of soils tested. Nevertheless, NO_3-N levels decrease rapidly following fertilization and use of recommended rates of N at 0.11 to 0.17 kg/tree yearly for 2-year-old trees will not produce excessively high NO_3-N levels in the soils tested. Frequent applications of fertilizer at lower rates will decrease NO_3-N in the soil further (Willis et al., 1990).

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