Influence of Nitrogen and Calcium Fertilizer on Fire Blight Susceptibility of **Royal Gala Apple Trees**

Francisco Salcedo

Former Graduate Student FONAIAP-Monagas Venezuela

Frank B. Matta

Professor of Horticulture Department of Plant and Soil Sciences Mississippi State University

Frank Killebrew

Extension Plant Pathologist Department of Entomology and Plant Pathology Mississippi State University

James O. Garner, Jr.

Professor of Horticulture Department of Plant and Soil Sciences Mississippi State University

For more information, contact Frank Matta at (662) 325-7716. Bulletin 1093 was published by the Office of Agricultural Communications, a unit of the MSU Division of Agriculture, Forestry, and Veterinary Medicine. It was edited and designed by Robert A. Hearn, publications editor. The cover was designed by Betty Mac Wilson, graphic designer/illustrator. March, 2000

ABSTRACT

Effects of nitrogen fertilizers and calcium (CaCO₃ \cdot 2H₂O) on fire blight susceptibility of 'Royal Gala' apple trees were evaluated. Disease rating scores and infection ratios were used to determine fire blight susceptibility. Infection ratios were useful in determining treatment differences with respect to nitrogen source and rates. Disease rating scores did not show a treatment effect. However, the rates of nitrogen and lime might not have been sufficient to induce fire blight. The negative relationship between disease rating scores and infection ratios indicated that the evaluation methods were sound. Mineral leaf composition showed that all elements were present at adequate levels. There was no relationship between foliar elemental concentration and fire blight incidence.

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INTRODUCTION

Fire blight, caused by *Erwinia amylovora* [(Burr.) Winslow et al.], is undoubtedly the oldest, most serious, and most perplexing bacterial disease of pomaceous fruit trees (van der Zwet and Keil 1979). Fire blight development depends on interaction among the pathogen, host, and environment. Any cultural practice that overstimulates growth usually makes the plant or tree vulnerable to serious fire blight infection and can also affect the general productivity of trees (Aldwinckle and Beer 1979).

While rapid growth is desirable, it makes the apple tree more susceptible to fire blight, the major disease problem in apple production (van der Zwet and Keil 1979). Tree vigor has a major influence on the extent of fire blight damage. Once established, the distance the pathogen moves through the vascular system is in direct relationship to the rate of tree growth. Vigorously growing shoots are the most severely affected. Therefore, conditions that favor rapid shoot growth, such as high soil fertility and abundant soil moisture, increase the severity of damage to trees (Dreistadt et al. 1994). Shoot tissues with a high nitrogen content are often from vigorously growing shoots.

Nitrogen appeared to be a key element because most studies show that increasing nitrogen results in severe fire blight symptoms (Lemaire et al. 1990; Sugar et al. 1992). Tissues with high nitrogen levels are susceptible to fire blight (Hildebrand and Heinicke 1937; Thomas and Ark 1939). Lewis (1961) and Lewis and Kenworthy (1962) found that trees supplied with high nitrogen contained higher concentration of leaf nitrogen, which enhanced fire blight infection.

However, nitrogen deficiency also increased susceptibility as did deficiencies of calcium, copper, iron, magnesium, manganese, molybdenum, and zinc (Aldwinckle and Beer 1979). Lewis and Kenworthy also found that foliar nitrogen level was the same for the most susceptible and for the least susceptible trees. Overall, the role of nitrogen in fire blight susceptibility was not clear.

Calcium is also an important mineral nutrient that influences the resistance of host plants to bacterial pathogens (van der Zwet and Keil 1979). The highest resistance to fire blight was found in trees with a high foliar analysis of calcium (Lewis and Kerworthy 1962). It is well known that calcium has a major effect on cell wall structure and membrane integrity (Poovaiah and Leopold 1975; Poovaiah et al. 1988). Increased levels of calcium in host tissue -- present primarily as calcium-pectate in the middle lamella of cell walls -- could render tissue more tolerant to the action of cell-wall-degrading enzymes (Punja et al. 1985; Conway 1989; Fallahi et al. 1997), such as amylovorin, which is produced by *E. amylovora* (Godman et al. 1974). Calcium-induced resistance in apples affects several pathogens. Calcium deficiency is generally corrected by the application of four to eight foliar sprays of Ca(NO₃), at intervals throughout the growing season (Weis et al. 1980).

Understanding the nature and causes of tree nitrogen and calcium variability could assist in the development of management techniques to limit the number of highnitrogen/low-calcium trees and the development of sampling procedures to quantify the incidence of high-risk fruit accurately (Sugar et al. 1992).

This study had two main objectives: (1) to investigate the effects of nitrogen and calcium rates on fire blight susceptibility of 'Royal Gala;' and (2) to determine the correlation between foliar nutrient concentration and fire blight symptoms in 'Royal Gala.'

MATERIALS AND METHODS

This experiment was conducted at the Pontotoc Ridge-Flatwoods Branch Experiment Station (34°08' N, 89°00' E), a unit of the Mississippi Agricultural and Forestry Experiment Station, which is based at Mississippi State University. The experiment was initiated in 1995 and terminated December 1998. Seventy-two 1-year-old 'Royal Gala' apple trees were planted at a 20-foot by 6-foot spacing. The soil is an Atwoods silt loam, which has a fine-textured subsoil. The trees were trained to the modified central leader system. Dolomitic lime applied at 2 tons per acre pre-plant and phosphorus

 (P_2O_5) at 80 pounds per acre were deep plowed into the soil in the fall of 1994. Standard MSU Extension Service recommended cultural practices were followed. However, no streptomycin or copper sprays were applied for fire blight conexperimental trol. The design was a completely randomized design with four replications and six treatments. Two trees were included in each replication.

Soluble lime at the rate of 2 tons per acre was applied with each treatment. The treatments were ammonium nitrate (NH₄NO₃) at 0,

0.1, or 0.3 pound per tree; calcium nitrate $(Ca(NO_3)_2 \cdot 2H_2O)$ at 0.1 or 0.3 pound per tree; and a calcium nitrate foliar spray at 3 pounds per 100 gallons of water. Another treatment included 2 tons per acre of soluble lime but received no nitrogen; this treatment was compared with the control (no lime and no nitrogen). All fertilizer treatments were applied by hand and were spread under the tree canopy.

We used a scoring system and percentage infection ratio to determine natural fire blight susceptibility as influenced by the various treatments. The percentage infection ratio was calculated by dividing the number of infected shoots by the total number of shoots and multiplied by 100 according to LeLezer and Paulin (1984). The scoring system for rating fire blight severity was that of van der Zwet (1970). This rating system is based on the number of twigs infected, the age of the wood blighted, and the percentage of canopy blighted (Table 1). Trees are rated on a 1-10 scale: 1 indicates tree death due to fire blight, while 10 indicates no fire blight symptoms (Sloan et al. 1996). Natural fire blight infection was promoted by misting the trees with water using an over-canopy mist system. The system was turned on for 15 minutes at 9 a.m. and 3 p.m. from first pink to post-bloom stage.

Table 1. Scoring system for rating the severity of fire blight.

Blight infection				Percent	
Blight	Current season	2-year-old	3-year & older	Scaffold limbs	of tree
ratings 1	wood	wood	wood	or truck	blighted
10	none	_	_	_	0
9	few	—	—	—	1-3
8	several	few	—	—	4-6
7	many	several	few	upper 1/8	7-12
6	—	many	several	upper 1/4	13-25
5	—	—	many	upper	26-50
4	—	—	—	lower	51-75
3	—	—	—	lower 1/4	76-88
2	_		_	base	89-99
1				all	100

¹Rating system from van der Zwet et al. (1970). Score is based on combinations of number of infections, age of wood infected, and percent of tree blighted.

To determine leaf elemental nutrient content, 40 mature, healthy leaves from trees of each treatment were randomly taken. Leaf tissue analyses were performed by the Plant and Soil Science Laboratory at Mississippi State University. Trees were sampled twice per year, before harvest season in June and after harvest in September. Nitrogen, phosphorus, potassium, calcium, magnesium, iron, manganese, zinc, and copper were analyzed. All data were subjected to Analysis of Variance (ANOVA) using the Statistical Analysis System (SAS 1985). Means were separated using Fisher's protected least significant difference, (LSD) $P \le 0.05$. Correlations using Pearson Coefficient were calculated to determine the relationship between foliar elemental concentration and incidence of fire blight.

RESULTS AND DISCUSSION

Fire Blight Disease Rating

From 1996 through 1998, nitrogen fertilization did not affect fire blight disease susceptibility of 'Royal Gala' apple trees (Table 2). This response was not expected since previously nitrogen had been shown to increase fire blight infection (Hildebrand and Heinicke 1937; Thomas and Ark 1939; Lewis 1961; Lewis and Kenworthy 1962; Lemaire et al. 1990; Koseoglu et al. 1996; Stiles 1998). However, in this study the rate of nitrogen might not have been sufficient to contribute to fire blight susceptibility. Lewis (1961) reported that excess nitrogen increases fire blight susceptibility but did not include the prevalent feeling that a minimum amount of nitrogen would lessen susceptibility.

From 1996 through 1998, broadcast lime did not affect fire blight disease ratings (Table 3). Lemaire et al. (1990) presented contrasting results. Lewis (1961) and Stiles (1998) stated that applying high calcium levels to decrease tree shoot necrosis had a tendency to increase *E. amylovora* infection.

It is important to note that with the scoring system used, trees were scored 7 or 8, which indicated damage was generally confined to the upper one-eighth or outer perimeter of the tree. According to van der Zwet et al. (1970), a tree with a score of 8 may have several infections in the current season's wood or a few infections in 2-yearold wood with no more than 6% of the tree blighted. If blight reaches older wood, it stops without further penetration. Without additional infection, such a tree would be expected to recover with only minimal damage. No rating was lower than 7 on the basis of number of infections in 1-year-old wood alone. Such scoring suggests that blight epidemiological factors may be involved rather than inherent susceptibility to the disease.

Table 2. Effect of nitrogen rates on fire blight disease rating of 'Royal Gala' apple trees, 1996-1998.				
Nitrogen fertilizer	Disease rating ¹			
	1996	1997	1998	
NH₄NO₃ (g/tree)				
0.0	7.00 a ²	8.00 ab	8.25 a	
45.4	6.63 a	7.75 ab	8.33 a	
136.20	6.83 a	7.80 ab	8.20 a	
Ca(NO ₃) ₂ (g/tree)				
45.4	6.41 a	7.91 ab	8.18 a	
136.20	6.92 a	7.36 b	8.88 a	
Ca(NO ₃) ₂ (foliar) ³	7.09 a	8.09 a	8.63 a	
LSD _{0.05}	1.31	0.72	2.05	
¹ Rating from 1-10: 1 = tree death, 10 = no fire blight symptoms.				

²Mean comparison within columns by Fisher's Protected LSD at P = 0.05. Means with the same letter do not differ at the 5% probability level. ³Solution of Ca(NO₃)₂ at 3 pounds per 100 gallons.

Table 3. Effect of calcium in the form of lime fertilizer on fire blight disease rating of 'Royal Gala' apple trees, 1996-1998.

Calcium-lime		Disease rating ¹	
	1996	1997	1998
0 tons per acre	6.64 a ²	7.89 a	8.60 a
2 tons per acre	6.89 a	7.76 a	8.06 a
LSD _{0.05}	0.75	0.39	0.75

¹Rating from 1-10: 1= tree death, 10 = no fire blight symptoms. ²Mean comparison within columns by Fisher's Protected LSD at P = 0.05. Means with the same letter do not differ at the 5% probability level.

Table 4. Correlation between leaf elemental content,nitrogen and calcium fertilizer, infection ratio,and disease score in 'Royal Gala' apple trees.

Parameters		Disease score ¹	
	1996	1997	1998
Nitrogen Treatment	-0.03 ns	-0.04 ns	-0.04 ns
Calcium Treatment	-0.06 ns	-0.08 ns	-0.13 ns
Infection Ratio	-0.54 ***	-0.22 *	х
Calcium (Ca)	0.04 ns	0.07 ns	-0.32 *
Manganese (Mn)	-0.34 *	0.29 ns	-0.09 ns
Cooper (Cu)	-0.05	0.33 *	-0.10 ns

¹ns = nonsignificant; * = significant at 0.05 level; ** = significant at 0.01 level; *** = significant at 0.001 level; and x = no evaluation (trees were pruned).

Correlation analyses were conducted to determine the relationship between foliar nutrient levels of nitrogen and calcium fertilizer, infection ratio, and fire blight susceptibility. In 1996 and 1997, the disease rating score was negatively correlated with the infection ratio [Pearson coefficient was -0.54 and -0.22, respectively (Table 4)]. There was no correlation between nitrogen or calcium fertilization and fire blight disease score (Table 4). This response was not expected, since previously nitrogen had been shown to increase fire blight infection (Hildebrand and Heinicke 1937; Thomas and Ark 1939; Lewis 1961; Lewis and Kenworthy 1962; Lemaire et al. 1990; Koseoglu et al. 1996; Stiles 1998). In 1996, disease score was negatively correlated (r = -0.34) with manganese and positively correlated with the infection ratio (r = .54). In 1997, disease score was positively correlated with copper (r = 0.33) (Table 4). According to Huber et al. (1988), manganese interacts with nitrogen metabolism and is intimately involved in carbohydrate synthesis, photosynthesis, and the synthesis of phenols and other compounds associated with the defense of plants against pathogens. Reduction in some plant diseases by high rates of copper may result from a fungistatic effect (Huber 1980).

Fire Blight Infection Ratio

In 1996, calcium nitrate at 0.10 pound per tree increased the fire blight infection ratio compared with the control. The remaining treatments had no effect. In 1997, calcium nitrate at 0.30 pound per tree increased the fire blight infection ratio compared with the control. The remaining treatments had no effect (Table 5). Broadcasted lime (2 tons per acre) did not influence fire blight infection ratio (Table 6). Using this evaluation system, it was observed that the use of calcium nitrate as a source of nitrogen increased fire blight susceptibility in 'Royal Gala' apple trees. Previous studies found that trees supplied with a complete nutrient solution except nitrogen

 Table 5. Effect of nitrogen source and rates on fire blight disease infection ratio of 'Royal Gala' apple trees, 1996 and 1997.

Nitrogen source	Infection ratio (%) ¹			
	1996	1997		
NH₄NO₃ (lb/tree)				
0.0	25.83 b ²	7.80 b		
0.10	30.25 ab	11.50 ab		
0.30	24.25 b	13.70 ab		
$Ca(NO_3)_2$ (lb/tree)				
0.10	38.55 a	9.27 ab		
0.30	27.72 ab	18.00 a		
Ca(NO ₃) ₂ (foliar) ³	22.18 b	10.27 ab		
LSD _{0.05}	11.92	8.81		
¹ Ratio between number of infected shoots and the number of shoots on a tree. ² Mean comparison within columns by Fisher's Protected LSD at P = 0.05. Means with the same letter do not differ at the 5% probability level. ³ Solution of 1,326 grams of Ca(NO ₃) ₂ per 100 gallons of water.				

failed to develop blight when inoculated with *E. amylovora*, whereas those supplied with a similar solution containing calcium nitrate $[Ca(NO_3)_2]$ were susceptible (Nightingale 1932). The effect of cations, like calcium, on nitrate uptake may counter the negative charges on root cell walls so that nitrate ions may migrate more closely to the plasmalemma and its uptake sites. Consequently, tree vigor increased after fertilization (Boynton 1966).

The infection ratios and disease rating scores are closely related for trees that received the 0.10-pound calcium nitrate treatment in 1996 and the 0.30-pound calcium nitrate treatment in 1997. In 1996, trees treated with 0.10 pound of calcium nitrate had a 38.6% infec-

tion ratio and a disease rating score of 5 (26-50% tree blighted). In 1997, trees treated with 0.30 pound of calcium nitrate had an 18% infection ratio and a disease rating score of 6 (13-25% tree blighted).

As previously noted, disease rating scores showed no differences ($P \le 0.05$) among fertilizer treatments. However, the infection ratio evaluation system showed some differences. These results suggested that the use of quantitative evaluations could be more accurate when only a few damaged shoots are observed in the orchard. Moreover, correlation analysis showed similarities between the evaluation systems. The scoring system proposed by van der Zwet et al. (1970) can be used for this type of experiment with greater precision. Results of this research suggested that infection ratio was useful in determining treatment differences with respect to nitrogen. Disease score did not show a

treatment effect, which indicates this rating system might be more restrictive. However, the rates of nitrogen and lime might not have been sufficient to induce fire blight susceptibility. Due to the negative relationship between fire blight disease score and infection ratio, we concluded that the evaluation methods were sound. Future research should be directed at applying calcium as a foliar spray and including higher rates of nitrogen. Moreover, to further define the relationship between host-plant nutrition and disease resistance, it will be necessary to carry out further and more detailed investigations on plant nutrition.

Table 6. Effect of calcium in the form of lime fertilizer on fire blight disease infection ratio of 'Royal Gala' apple trees, 1996-1997.

Calcium-lime	Infection ratio (%) ¹	
	1996	1997
0 tons/acre	28.74 a ²	12.65 a
2 tons/acre	27.18 a	15.33 a
LSD _{0.05}	6.88	2.65

¹Ratio between number of infected shoots and the number of shoots on a tree. ²Mean comparison within columns by Fisher's Protected LSD at P = 0.05. Means with the same letter do not differ at the 5% probability level.

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