Identification, Symptomatology, and Epidemiology of Fire Blight on Le Conte Pear in the Nile Delta of Egypt

T. VAN DER ZWET, Research Plant Pathologist, USDA, Appalachian Fruit Research Station, Kearneysville, WV 25430

ABSTRACT

van der Zwet, T. 1986. Identification, symptomatology, and epidemiology of fire blight on Le Conte pear in the Nile Delta of Egypt. Plant Disease 70:230-234.

Outbreaks of fire blight caused by *Erwinia amylovora* on Le Conte pear, the first in Egypt since 1962, were associated with heavy rainfall during bloom in 1982, rainfall combined with wind storms during bloom in 1983, and one 2-day rain during bloom in 1984. The severe occurrence of the disease, expressed mainly as blossom blight, caused a loss of blossoms varying from 10 to 75% per tree. Of 24 bacterial isolates from blighted pear tissues tested for colony morphology, pathogenicity, and fatty-acid composition, 22 were *E. amylovora*. Two isolates were identified as *Pseudomonas syringae*. On Bartlett seedling shoots, Egyptian isolates of *E. amylovora* generally appeared more virulent than two standard American cultures used for comparison. However, fatty-acid profiles of the Egyptian isolates matched those of the standard cultures in the *E. amylovora* library.

Trees of the low chilling pear (*Pyrus communis* L.) cultivar Le Conte were first introduced into Egypt about 50 yr ago and were planted extensively after World War II. Today, about 4,000 ha of pears are grown in the lower Nile Delta region, 60% of which are located in Behera Governorate south of Alexandria between the Western Desert and the Rosetta branch of the Nile river (Fig. 1).

Bloom usually occurs between 15 March and 15 April. A second, late bloom occurs sometime between 20 September and 10 November. Rainfall comes between mid-November and early April, and the trees are irrigated weekly during the remainder of the growing season. Generally, irrigation starts 1 wk after the onset of bloom. Partial leaf drop usually occurs during the second half of December, and the trees pass through a dormant period of about 12 wk at minimum temperatures of about 5 C.

Fire blight (caused by *Erwinia amylovora* (Burr.) Winslow et al) was first reported from Egypt by El-Helaly et al (5) in 1964. The initial discovery occurred near Mamal El-Kezaz in the region between Alexandria and Damanhur (Fig. 1). After an intensive survey during 1966–1972, El-Goorani (4) reported that pear trees appeared free of fire blight.

During a survey in 1983, symptoms characteristic of both fire blight and blossom blast (caused by *Pseudomonas syringae* van Hall) were observed on pear and quince (*Cydonia oblonga* Mill.). In addition, apple (*Malus domestica* Borkh.) fruit were observed affected by a rot suspected to be caused by *Phytomonas melophthora* Allen & Riker (1) (=*Acetobacter pasteurianus* pv. *pomi* (Hansen) Beijerinck) (3). Therefore, all three organisms were considered in the identification of the principal causal agent. Disease occurrence in 1983 was reported (12).

MATERIALS AND METHODS

Isolations. Isolations from diseased apple, pear, and quince tissues were performed on nutrient-yeast-dextrose agar (NYDA) at the Plant Pathology Research Institute (PPRI) in Giza. Tissues were surface-disinfested with 1% sodium hypochlorite for 3 min and rinsed three times in sterile water for 3 min each. Small fragments of tissue at the border of

Accepted for publication 12 August 1985.

The publication costs of this article were defrayed in part by page charge payment. This article must therefore be hereby marked "*advertisement*" in accordance with 18 U.S.C. § 1734 solely to indicate this fact.

the necrotic area were placed on the medium. Plates were incubated at 26 C for 3 days. Resulting cultures and those obtained earlier by researchers of the University of Alexandria and PPRI were transferred to NYDA slants and shipped to the United States. All cultures were maintained on NYDA slants under mineral oil.

In the laboratory of the Appalachian Fruit Research Station (AFRS), diluted bacterial suspensions of the cultures were plated on Miller-Schroth (MS) medium (8). On this medium, colonies of *E. amylovora* show a characteristic orange hue with a red center, whereas pseudomonads and other bacteria appear blue or green. Usually, colonies of *E. amylovora* are smooth with entire translucent margins. Suspected *Pseudomonas* cultures were grown on King's medium B (10) and examined for fluorescence under ultraviolet radiation. Standard check cultures used for comparison were: *E. amylovora* isolates 273 from Cornell University and 330 isolated from pear at AFRS; *P. syringae* isolate 400 maintained at AFRS; and *P. melophthora*, received as *A. pasteurianus* pv. *pomi* from the National Collection of Plant Pathogenic Bacteria, Harpenden, England.

Gas liquid chromatography (GLC). GLC performed cooperatively at the University of Delaware involved analysis of the bacterial cellular fatty acids, resulting in a reproducible reference profile. Routine procedures for preparing bacterial samples for GLC analysis (6,7,9) were followed to produce chromatograms of the fatty acids of the saponified whole cells of the bacterial isolates. A Hewlett-Packard (HP) 9836 computer identified the chromatographic peaks and searched a library of stored fatty-acid profiles to permit identification of the bacterial cultures. This library is the basis for the commercially available brochure HP 5898A, *Microbial Identification System*. In 1984, a second GLC analysis was done on the isolates collected that year, including the standard cultures of *E. amylovora, P. syringae*, and *A. pasteurianus*.

Pathogenicity on pear fruit. Pathogenicity and virulence of all cultures were tested on immature Bartlett pear fruit collected in early June. Fruit were surface-disinfested and rinsed as described, and a small slice (10 mm in diameter) was removed with a flamed knife. A puncture was made in the exposed flesh with a sterile needle, and a loopful of bacteria was deposited aseptically in the hole. Standard cultures of *E. amylovora, P. syringae*, and *A. pasteurianus* were used as controls. Each isolate was tested in triplicate. Inoculated fruit were incubated in a moist chamber at 26 C, and degree of necrosis and oozing was examined after 1 and 2 wk. The test was repeated twice.

Pear tree inoculations. All isolates collected in 1984 and some from 1983 were tested for pathogenicity on succulent shoots of young Bartlett pear seedlings. Shoot tips were injected with 0.5 ml of a bacterial suspension (5×10^7 cells per milliliter), using a 26-gauge hypodermic needle. Four shoots (two per tree) were used per isolate, and the plants were kept in a humid room maintained at about 26 C. Uninoculated trees and trees injected with the standard cultures served as checks. Lengths of blighted portions of the shoots were measured 1 and 3 wk after inoculation. This test was repeated once.

These fruit and shoot pathogenicity studies were performed under strict containment at the U.S. Plant Disease Laboratory facilities at Ft. Detrick, MD, to prevent the possibility of releasing an extremely virulent isolate of *E. amylovora* into the eastern pome fruit industry. Upon completion, all cultures and plant material were destroyed by autoclaving.

RESULTS

Field observations. In 1983, trees bloomed considerably later (6–30 April) than usual. Also, rainfall continued through April into early May while some orchardists had already started irrigation, resulting in extra succulent growth of shoots. In Behera Governorate, fire blight was severe in most orchards visited. There was no loss of large branches or entire trees, but loss of blossoms per tree varied from 10 to 75%. Many trees showed 50–75% of blossoms blighted.

In 1984, trees started to bloom on about 12 March and continued until 27 April. In most orchards, first blight symptoms occurred about mid-April, and the disease became severe near the end of the month. Observations were extended to four other governorates, including Faiyum, an oasis 60 km south of Cairo (Fig. 1). Fire blight was moderate to severe throughout the northern part of Behera and very severe near the Mediterranean coast in Alexandria Governorate (Table 1). In this area, as many as 100% of the blossom clusters were blighted, apparently because of the nightly high humidity off the Mediterranean Sea. In 1984, the disease was also observed west of the town of Kafr El-Sheikh near the Rosetta branch of the Nile, apparently representing an eastward dissemination from Behera. Fire blight was not found in Gharbia (between the two Nile branches) and Faiyum governorates.

Throughout the pear orchards, the limb borer (*Zeuzera pyrina* L.) and bud weevil (*Hypothenomus asporicollis* Woll.) appeared to be serious insect pests. Damage by limb borers was most intense in trees older than 25 yr. This borer makes tunnels 4–5 mm in diameter throughout the tree, and from a distance, infested, dying limbs appear as if affected with fire blight. The bud weevil, an inconspicuous insect, was often found in buds and spurs of blighted shoots. Reflexed shoot tips were observed that appeared similar to the shepherd's crook symptom. However, these shoot tips and leaves were green and bud weevils were often found in young buds, some distance from the shoot tip. The possible relationship between fire blight and the two insects is under investigation. It is possible that either or both insects may disseminate the fire blight pathogen.

Symptomatology. All symptoms observed on pear and quince were those of blossom blight (Fig. 2), all caused by

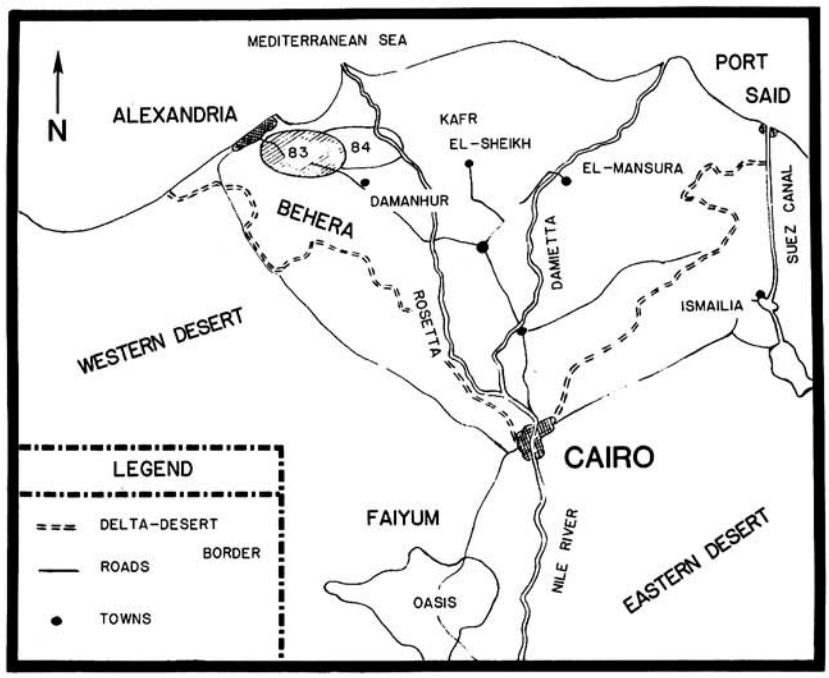

Fig. 1. Map of northeastern Egypt. Circled areas in Behera Governorate near Alexandria are those where fire blight occurred in 1983 and expanded in 1984.

Table 1. Survey of fire blight infection and limb borer infestation on pear, apple, and quince in various orchards throughout the Nile Delta and Faiyum Oasis regions of Egypt (May 1984)

Location		Orchard		Tree[a]			
County	Village	Name	Area[b]	Type	Age (yr)	Growth	Blight severity[c]
		Alexandria Governorate					
Alexandria	El-Mamura	Min. Agric.	10	Pear	10–20	Poor	4
		Behera Governorate					
Khorshid	Tawfikia	A. El-Gazar	25	Pear*	20	Good	2
			10	Pear	20	Good	3
Kafr El-Dawar	K. Abou-Kear	W. Hanafy	10	Pear	33	Medium	2
		A. Ghorbal	45	Pear*	30	Medium	2
		M. Abboud	25	Pear*	30	Medium	2
		S. El-Said	40	Pear*	30	Medium	2
		M. El-Shendidy	10	Pear*	20–45	Poor	2
		D. El-Shendidy	10	Apple	5	Medium	0
		A. Basuni	10	Pear	45	Poor	3
		F. Basuni	10	Pear	5	Good	2
		H. El-Ebrashy	46	Pear*	20	Medium	3
	Kafr El-Dawar	A. Aref	20	Pear*	12	Good	3
			1	Quince*	15	Good	3
Abo-Hummus	Berdan	G. Stino	33	Pear*	30	Good	1
			2	Apple		Good	0
		M. Stino	33	Pear	30	Good	2
		Kafr El-Sheikh Governorate					
Mutobas	El-Khalig	S. Shalaby	14	Pear	4	Good	3
Fuwa	Fuwa	M. El-Deeb	9	Pear	4–7	Good	0
Kafr El-Sheikh	El-Hamra	M. Abdel-Megid	2	Pear	4	Good	0
		Gharbia Governorate					
Zifta	M. Waseef	M. Mansour	46	Pear	28	Good	0
		I. El-Magraby	20	Pear	12	Good	0
		Faiyum Governorate					
Obshoway	Aboxah	H. Meibed	30	Pear	5–12	Good	0
			2	Pear	50	Good	0

*Tree types marked with an asterisk (*) are those from which bacterial isolates 750–754, 780–782, and 811–881 were obtained (Table 3).
[b]Area is in feddans: 1 feddan = about 0.4 ha.
[c]Severity rating: 0 = none, 1 = light (1–25%), 2 = moderate (26–50%), 3 = severe (51–75%), and 4 = very severe (76–100% of flowers blighted).

Fig. 2. Typical blossom blight (*Erwinia amylovora*) in Le Conte pear extending downward into 1-yr-old shoot. Note papery bark (arrow), a characteristic symptom for blossom blast (*Pseudomonas syringae*) or fire blight infection in pear tissues with hybrid Oriental *Pyrus* parentage (cultivar Le Conte).

blossom infections. Especially on quince, the disease was more severe than ever reported. In 1983, blighted shoots 1–3 m long were observed, presumably resulting from more succulent shoot growth and extensive rainfall that year. Some cankers had papery bark (Fig. 2), a characteristic of blossom blast (*P. syringae*). Because the blossom blight phase of fire blight is indistinguishable in the field from that of blossom blast, the latter organism was considered as part of the overall problem. In some orchards (Ghorbal and El-Said), most symptoms were typical of blossom blast, whereas in others (El-Ebrashy, El-Gazar, and El-Shendidy), symptoms appeared characteristic of fire blight: gradual proximal extension of the canker, high incidence of shepherd's crooks, and numerous extensions of blighted tissues more than 1 m. The papery bark symptom has been observed in America on pear trees with hybrid Oriental *Pyrus* parentage, such as Le Conte.

Bacterial morphology and fatty-acid analysis. With the exception of five isolates, all bacterial cultures on NYDA from Alexandria, the PPRI, and those collected from pear and quince formed typical small, round, white glistening colonies characteristic of *E. amylovora*. Two isolates (780 and 782) were characteristic of *P. syringae*, and three isolates (750, 751, and 861) were mixed cultures containing *E. amylovora*. All isolates except 750, 751, 780, and 782 formed characteristic orange-red colonies on the selective MS medium. The first two isolates proved to be *E. amylovora*, whereas the last two were *P. syringae*. Of these, only isolate 782 on King's medium B showed characteristic fluorescence under ultraviolet light. *A. pasteurianus* was never isolated.

Two GLC analyses of the bacterial fatty acids also showed that all Egyptian isolates tested except 780 and 782 were *E. amylovora*. These two isolates were *P. syringae*. Table 2 shows the comparisons of the fatty-acid profiles of all isolates collected in 1983 and 1984 as well as those of the standard cultures of *E. amylovora*, *P. syringae*, and *A. pasteurianus*.

Pathogenicity tests. In fruit pathogenicity tests, all but five cultures from Alexandria, the PPRI, and those personally collected caused tissue necrosis accompanied by bacterial ooze in immature pear fruit. Three isolates (750, 751, and 754) were *E. amylovora*, whereas isolates 780 and 782 were *P. syringae*. The pathogenicity of five isolates from 1983 and eight from 1984 was reevaluated on fruit and shoots (Table 3). The *E. amylovora* isolate (754) from quince at first did not produce any oozing on pear fruit but was highly pathogenic on shoots. Isolate 512 was among the most virulent on fruit but not on shoots, whereas isolate 831 was very virulent on both tissues. With the

Table 2. Distribution and percent area of gas liquid chromatography fatty acid profiles for bacterial isolates collected in Egypt compared with standard cultures of *Erwinia amylovora*, *Pseudomonas syringae*, and *Acetobacter pasteurianus*

Fatty acid	*E. amylovora* (273, 330)	Egyptian bacterial isolates[a] 1–4	65–67	412–512	750–754	811–881	780–782	*P. syringae* (400)	*A. pasteurianus* (461, 462)
10:0 3OH	...	...	...	...	...	...	1.8	1.6	...
12:0	5.4	4.9	4.9	5.4	4.5	5.4	5.0	5.8	...
12:0 2OH	...	...	...	...	...	...	1.7	3.1	...
12:0 3OH	...	...	...	...	...	...	0.6	2.8	...
14:0	5.9	3.6	3.7	5.5	5.4	5.4	...	...	2.1
14:0 2OH	...	...	...	...	...	...		...	3.3
14:0 3OH	7.9	5.2	5.8	8.5	6.9	8.2	...	...	1.3
15:0	0.6	1.2	0.9	0.5	0.7	0.4	...	...	...
16:0	36.3	35.2	34.9	36.1	34.2	36.4	28.0	27.1	12.1
16:0 2OH	...	...	...	...	...	...	...	...	6.4
16:0 3OH	...	...	...	...	...	...	...	...	2.0
16:1 C9	23.0	28.2	25.1	21.2	20.2	20.7	26.7	36.1	...
17:0	0.6	3.1	2.8	1.2	1.3	1.0	...	...	...
17:0 cyc	11.0	4.6	7.7	10.2	9.7	11.1	2.0	4.5	...
18:0	0.5	0.5	0.7	0.7	0.4	0.6	0.8	1.0	3.7
18:1 tll	6.8	12.3	12.0	8.6	12.6	8.9	32.3	18.0	59.6
19:0 cyc	...	...	...	...	1.5	...	...	...	10.2

[a] Isolates 1–4 (1982) and 65–67 (1983) were collected by K. Abo-El-Dahab (University of Alexandria) from pear, 412–512 (1983) were collected by K. Y. Mickail (PPRI, Cairo) from pear, and 750–782 (1983) and 811–881 (1984) were collected by T. van der Zwet (USDA) from pear and quince in Egypt. Standard cultures used for comparison were *E. amylovora* isolates 273 (Cornell University) and 330 (AFRS), *P. syringae* isolate 400 (AFRS), and *A. pasteurianus* pv. *pomi* type cultures 461 and 462 (National Collection of Plant Pathogenic Bacteria).

exception of isolates 512 and 871, all Egyptian isolates blighted a higher percentage of the pear shoots 1 wk after inoculation than did the two standard isolates of *E. amylovora* from the United States. These differences were even more apparent 3 wk after inoculation.

Epidemiology. Average temperatures just before and during bloom usually vary from a minimum of 5 C to a maximum of 24 C. Before 1982, rainfall during March and April varied from about 1 mm in 1978 to 17 mm in 1980. In contrast, about 45 mm of precipitation fell in 1982, and nearly 20 mm fell in 1983. In addition, severe wind storms occurred during bloom in 1983. Only 5 mm of rain fell during the 1984 bloom period (7–9 April). In 1984, several trees in one orchard showed nearly all the blossoms blighted in the top half of the tree compared with nearly no blight in the lower half. The orchardist claimed that only the upper halves of these trees were in bloom during the rain of 7–9 April 1984, whereas the lower halves had set fruit.

In general, ooze droplets were rare and bacterial strands were never observed. Only small, dry ooze droplets were noted on blossom stems, which resulted in contaminated cultures. The only plant materials from which pathogens could be readily isolated were recently invaded quince and pear leaves (midveins) and their petioles (Fig. 3). No ooze streaking on the bark surface or shoot blight without blossom blight was observed.

Table 3. Comparisons of the degree of pathogenicity in pear fruit and shoots of 13 isolates of *Erwinia amylovora* and two isolates of *Pseudomonas syringae* collected in Egypt during 1983 and 1984 with standard isolates

Isolate[a]	Degree of fruit oozing after 2 wk	Shoots blighted after 3 wk (%)
	1983	
E. amylovora		
412	2	68
415	2	73
510	2	66
512	3	48
754	2	68
P. syringae		
780	0	0
782	1	0
	1984	
E. amylovora		
811	2	71
821	1	80
831	3	71
841	2	65
851	2	69
861	1	63
871	1	50
881	2	61
Standard isolates		
273	3	47
330	2	39
400	0	0
461	0	0
462	0	0

[a] Isolates: 412, 415, 510, 512, 754, 780, 782 (1983) and 811–881 (1984) are Egyptian isolates; control isolates: 273 and 330 (*E. amylovora*), 400 (*P. syringae*), and 461 and 462 (*A. pasteurianus*).
[b] Severity rating: 0 = none, 1 = light, 2 = moderate, and 3 = severe oozing of fruit.

Fig. 3. One-year-old blighted pear shoot with characteristic necrosis of petioles and leaf midrib. Note small cavities (arrows) in spurs where bud weevils (*Hypothenomus asporicollis*) were found.

DISCUSSION

It is possible that during 1964–1983, fire blight increased and spread near the center of its introduction but not to the extent that it was noticed by growers and researchers. Also, in some of these years, weather conditions probably were favorable for blight development. Therefore, it is likely that *E. amylovora* became widespread in a large area of Behera Governorate and that the delayed bloom (1983), together with the extended rainfall and wind storms (1982 and 1983), combined to initiate the epidemic of 1983 and the ensuing severe outbreak in 1984. Such an extended period of quiescence is not unusual and has been observed in North America (11). All orchards of young trees (no bloom) had no fire blight, whereas mature orchards near Damanhour and further south and east had very few or no blight symptoms.

Temperature and rainfall data during pear bloom in Egypt were applied at the AFRS to the infection risk assessment system developed by Billing (2). In this system, the chance for fire blight development in the range of 24–30 C, combined with no rainfall, is medium. With any rainfall up to 2.5 mm, the risk is considered high. These infection risks are based mainly on the number of potential doublings of the population of the fire

blight pathogen. If in future years the bloom periods are totally devoid of rain, fire blight may pose a moderate threat to the Egyptian pear industry. On the other hand, if rains prevail during bloom or in areas with high humidity, the disease could remain a serious problem. Under Egyptian climatological conditions, in which no rain occurs between May and October and maximum temperatures remain in the mid-30s every day, the chance for extensive shoot, leaf, and fruit blight should be minimal. Without rainfall, a light secondary bloom during October and November may be conducive to infection. However, if such late bloom is extensive and rains occur, such conditions could play a major role in blight epidemiology. Monitoring pear flowers (8,13) in autumn and during spring bloom may clarify the bacterial life cycle and improve disease control.

ACKNOWLEDGMENTS

This work was supported by the Ministry of Agriculture and Food Security, Cairo, Egypt, in a cooperative agreement with the USDA. We wish to thank W. M. Dowler, USDA Plant Disease Laboratory, Ft. Detrick, MD, for assistance in providing containment facilities to perform pear fruit and shoot inoculation experiments, and M. Sasser, University of Delaware, Newark, for his assistance in identifying the Egyptian bacterial isolates through gas liquid chromatography analyses.

LITERATURE CITED

1. Allen, T. C., and Riker, A. J. 1932. A rot of apple fruit caused by *Phytomonas melophthora*, n. sp., following invasion by the apple maggot. Phytopathology 22:557-571.
2. Billing, E. 1980. Fire blight (*Erwinia amylovora*) and weather: A comparison of warning systems. Ann. Appl. Biol. 95:365-377.
3. Dhanvantari, B. N., Dye, D. W., and Young, J. M. 1978. *Pseudomonas pomi* Cole 1959 is a later subjective synonym of *Acetobacter pasteurianus* (Hansen 1879) Beijerinck 1898, and *Pseudomonas melophthora* Allen and Riker 1932 is a nomen dubium. Int. J. Syst. Bacteriol. 28:532-537.
4. El-Goorani, M. A. 1973. Current status of fire blight in Egypt. Plant Dis. Rep. 57:646.
5. El-Helaly, A. F., Abo-El-Dahab, M. K., and El-Goorani, M. A. 1964. The occurrence of the fire blight disease of pear in Egypt. Phytopathol. Mediterr. 3:156-163.
6. Maas, J. L., Finney, M. M., Civerolo, E. L., and Sasser, M. 1985. Association of an unusual strain of *Xanthomonas campestris* with apple. Phytopathology 75:438-445.
7. Miller, L. T., and Sasser, M. 1984. Fatty acid profiling of plant related bacteria. (Abstr.) Phytopathology 74:757.
8. Miller, T. D., and Schroth, M. N. 1972. Monitoring the epiphytic population of *Erwinia amylovora* on pear with a selective medium. Phytopathology 62:1175-1182.
9. Moss, C. W. 1981. Gas liquid chromatography as an analytical tool in microbiology. J. Chromatog. 203:337-347.
10. Sands, D. C., Schroth, M. N., and Hildebrand, D. C. 1980. *Pseudomonas*. Pages 36-44 in: Laboratory Guide for Identification of Plant Pathogenic Bacteria. N. W. Schaad, ed. Am. Phytopathol. Soc., St. Paul, MN.
11. van der Zwet, T., and Keil, H. L. 1979. Fire blight—a bacterial disease of rosaceous plants. U.S. Dep. Agric. Handb. 510. 200 pp.
12. van der Zwet, T., and Mickail, K. Y. 1983. Severe fire blight on low chilling pear in the Nile Delta. (Abstr.) Phytopathology 73:760.
13. van der Zwet, T., and Van Buskirk, P. D. 1984. Detection of endophytic and epiphytic *Erwinia amylovora* in various pear and apple tissues. Acta Hortic. 252:69-77.