Evaluation of the MARYBLYT Computer Model for Predicting Blossom Blight on Apple in West Virginia and Maryland

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ABSTRACT

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The MARYBLYT computer model was evaluated for its accuracy in forecasting apple blossom infection by *Erwinia amylovora* and the subsequent appearance of fire blight symptoms. Temperature and rainfall data were collected and disease observations recorded in bearing orchards in West Virginia and Maryland during 1984–1993. Among the 13 primary infection events identified by the model at all sites in eight of the 10 yr, blossom blight symptoms appeared 10 times within ±1 day, twice within 2 days, and only once within 3 days of the MARYBLYT prediction. Only three times in 10 yr did MARYBLYT predict blossom infection without symptom development. In no instance did spurious symptoms appear that would indicate the model failed to identify an infection period. A blossom sampling procedure conducted during 5 yr (1985, 1987, 1988, 1990, and 1993) in which blossom blight occurred confirmed the presence of *E. amylovora* coincident with the model's threshold calculation of epiphytic infection potential. When blossoms were inoculated artificially by introducing a bacterial suspension (10^8 cfu/ml) into flower nectaries, blossom blight symptoms developed 0, 1–3, and >5 days prior to that predicted by the model in one, seven, and three trials, respectively. In 11 trials, an average of 57 degree days >12.7 C was accumulated between artificial inoculations and symptom appearance, which is consistent with the model's algorithm for symptom occurrence. The results of our field evaluations of MARYBLYT for predicting blossom infection and subsequent symptom development show that the model is accurate. Treatment decisions based on MARYBLYT can be expected to improve the level of control of this destructive disease.

Additional keywords: *Malus, Pyrus*

Fire blight in apples and pears, caused by the bacterium *Erwinia amylovora* (Burrill) Winslow et al, has been characterized as sporadic and highly destructive (17,21). Severe outbreaks of fire blight may occur in orchards with no history of the disease, and, similarly, little or no damage may occur in orchards following seasons with significant blight damage. Because of the erratic nature of fire blight, coupled with its destructive potential, many growers in the eastern United States routinely apply three or more sprays of streptomycin at regular intervals throughout the blossoming period. Such treatments sometimes fail to prevent serious losses and sometimes appear unnecessary when treated orchards are compared with nearby untreated orchards. In addition, there is a growing concern about the development of streptomycin-resistant strains of the pathogen where the antibiotic may be used excessively (9).

The earliest attempt to predict fire blight started in the early 1950s, when Mills (7) correlated the number of daily degree days above 15.6–21.1 C (60–70 F) with the occurrence of blossom blight in apple in New York. During a 5-yr period, Luepschen et al (5) observed that a minimum of 2 days with average temperatures exceeding 18.3 C (65 F) was required for severe blossom blight infections. In Illinois, Powell (8) developed a similar prediction method for blossom blight based on 18 C degree days (30 F degree days) above a base of 18.3 C coupled with light rain and/or high humidity. In California in the early 1970s, *E. amylovora* was detected in pear flowers when the daily mean temperature exceeded a linear temperature threshold of 16.7 C on 1 March to 14.4 C on 1 May (15,16). Later, Zoller and Sisevich (22) discovered that 10% of pear blossom samples in California contained *E. amylovora* when 200 C degree hours (DH) were reached and 40% when more than 336 C DH were accumulated; (1 C DH equals 1 degree above 18.3 C for 1 hr). During 1975–1985, *E. amylovora* has been recovered from blossoms of apples and pears throughout North America (1,2,14,15,19). In 1988, a detailed review of the above two California findings in relation to experimental prediction systems was published (20).

MARYBLYT, a computer model for fire blight prediction and management, was developed initially in 1987 (11,12). The model, which is based partly on the above-mentioned studies, identifies the conditions that are conducive to the development of four separate types of fire blight symptoms: blossom, canker, shoot, and trauma blights. During the blossom period, the model identifies dates with risk of infection, provides a qualitative assessment of risk (low, moderate, high, and infection), and predicts the date of the first visible symptoms. For blossom blight, the minimum conditions for infection by *E. amylovora* are: 1) blossoms open with stigmas and petals intact, 2) accumulation of at least 110 cumulative DHs (CDH) above 18.3 C from first open bloom, 3) a wetting event ≥0.25 mm of rain or a heavy dew or fog (sufficient to wet foliage) or a rain ≥2.5 mm the previous day, and 4) an average daily temperature of 15.6 C. Daily minimum and maximum temperatures are initially provided to the model when the phenological stage of the trees is green tip. The model predicts infection to occur when *E. amylovora* is present and all of the above minimum conditions develop in the sequence presented. The accumulation of 110 CDH above 18.3 C indicates the presence of epiphytic bacteria on the open blossom and equates to the threshold of a relative epiphytic infection potential (EIP) of 100. Blossom blight symptoms (BBS) are predicted to appear after passage of 57 cumulative degree days (CDD) above 12.7 C from the date of infection.

In 1992, Jones (3) reported the MARYBLYT model to be more accurate in predicting blossom blight symptom appearance than infection periods on several apple cultivars in Michigan. The

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objective of our study was to evaluate the accuracy of the MARYBLYT model for predicting infection periods as well as the appearance of blossom blight symptoms on apple in West Virginia and Maryland. Validation of the model was based on the occurrence of natural infection during 1984–1993 and the use of artificial inoculations during 1987–1993.

MATERIALS AND METHODS

Version 4.0 (1992) of the MARYBLYT prediction system (13) was used to evaluate the 10-yr period under study: 5 yr (1987–1991) during which the earlier versions of the prediction system (4,11,12) were employed with grower cooperators in the field, 2 yr (1992–1993) with the version 4.0 system, and 3 yr (1984–1986) in which the system was applied retrospectively (a severe blossom blight epiphytotic occurred in 1985). In order to determine the accuracy of the model, the first symptoms were defined explicitly. The first visible symptoms of blossom blight on apple and pear are tiny ooze droplets behind the young ovary (Fig. 1A). Within 24 hr, these droplets are usually farther down on the peduncle and the ovary has become discolored (Fig. 1B). These early symptoms are difficult to spot, but blossom blight can quickly become a very important source of secondary inoculum when left as long as 48–72 hr after the first symptoms appear (Fig. 1C).

The incidence and severity of fire blight infection in West Virginia and Maryland were described qualitatively as none (no blight observed), light (occasional strikes on scattered trees, 1–6% of tree blighted), moderate (numerous strikes on many trees, 7–25% of tree blighted), and severe (heavy infection on most trees, 26–100% of tree blighted).

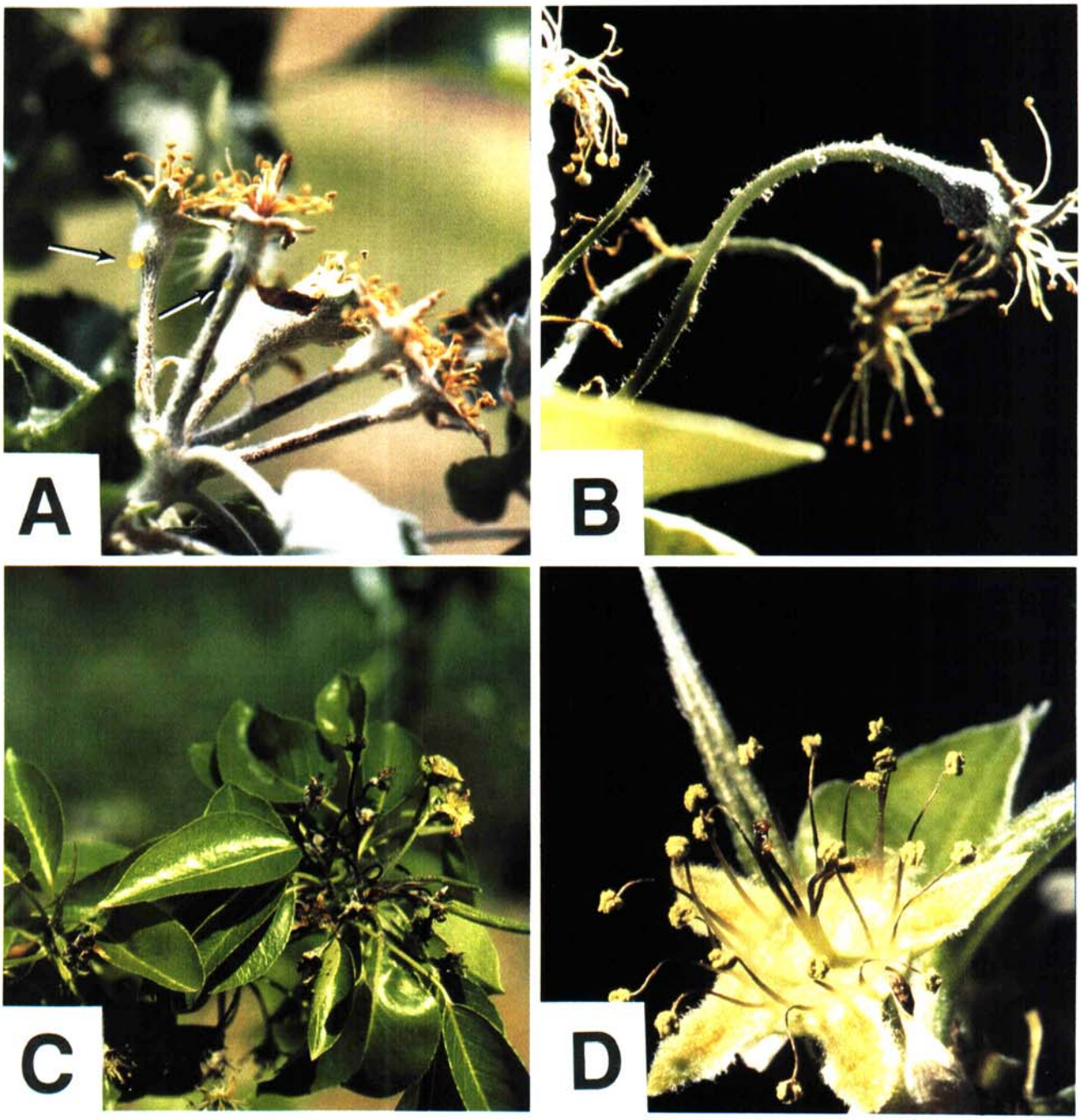

Fig. 1. Earliest visible symptoms of blossom blight on apple (A) and pear (B, C, and D) that were used to validate the MARYBLYT model: **(A)** Tiny ooze droplets (arrows) directly behind the young ovary at or within 6 hr of predicted expression of blossom blight symptoms; **(B)** multiple droplets farther down on the blossom peduncle within 24 hr of blossom blight symptoms, **(C)** numerous blackened peduncles and infection commencing into fruit spur, about 48–72 hr after blossom blight symptoms, and **(D)** infection and partial invasion of pistils after artificial infestation of stigma surface with *Erwinia amylovora*.

Table 1. Dates of blossom blight risk in apple orchards in Maryland and West Virginia, 1984–1993

Year	Location[y]	Apple cultivar	Dates of infection risk during bloom[z]							
			10 Apr.	15 Apr.	20 Apr.	25 Apr.	30 Apr.	5 May	10 May	15 May
1984	WMREC	Rome					MM	LMMMM	LMHHL	
	AFRS	Jonathan						MHMMM	MLHH*I*	
1985	WMREC	Rome	LL	HMMHH	HHHHH	*I I*HMM	H			
	AFRS	Jonathan	LL	MMMMH	H*I I I I*	HM				
1986	WMREC	Jonathan			MMML	LLHH*I*	H*I*M			
	AFRS	Jonathan		MMMMM	LHMML	LMM*I I*	MHHMM			
1987	WMREC	Jonathan			MHMM	LLLML				
	AFRS	Jonathan				MMLLL	MLLMM	MLM*I*M	HHH*I*	
1988	WMREC	Jonathan		MM	LLLMM	LLMMM	LLLMM	MMHM*I*	*I I*	
	AFRS	Jonathan			LLM	MLLLM	MLLLM	MMMHL	H*I*	
1989	WMREC	Jonathan			LLLL	LMMLM	*I I*M			
	AFRS	Jonathan			L	LMLHH	M*I*MML	MMM		
1990	WMREC	Gala	LL	MMLML	LMLLL	HH*I*HH	*I*HHHH	HHHHH	*I*H	
	AFRS	Jonathan	LL	MMLML	MMMMM	*I*HHHH	*I*			
1991	WMREC	Jonathan	LLM	MMHLM	MMMLL	LMH*I*H	*I I I*HH			
	AFRS	Jonathan	HMLMM	MMMML	MMMMM	MLMH*I*	HHHMM			
1992	WMREC	Jonathan			HH	*I*MLLL	ML*I*HM	MLLMM	MMH*I*H	H
	AFRS	Jonathan				HMMML	LML*I*H	MMLMM	MLML	
1993	WMREC	Jonathan				L	MHHHH	*I I I*HH	H	
	AFRS	Jonathan				MLLLL	HHHH*I*	*I I*HHH	HH	

[y] WMREC = Western Maryland Research and Education Center, Keedysville; AFRS = Appalachian Fruit Research Station, Bardane, West Virginia.
[z] L = low risk, M = moderate risk, H = high risk, *I* = infection.

Table 2. Accuracy of MARYBLYT predictions of first blossom blight in apple orchards in Maryland and West Virginia, 1984–1993

Year	Location[t]	Apple cultivar	MARYBLYT				Predicted incubation period[y] (days)	First blossom blight observed		Difference between BBS and observed blight (days)
			First infection		First symptoms					
			I date[u]	EIP CDH >18.3 C[v]	BBS date[w]	CDD >12.7 C[x]		Date	CDD >12.7 C	
1984	WMREC	Rome	...	...	...	...	...	NBO[z]	...	...
	AFRS	Jonathan	14 May	180	26 May	63	12	27 May	72	+1
1985	WMREC	Rome	25 Apr.	820	6 May	57	11	7 May	59	+1
	AFRS	Jonathan	21 Apr.	339	1 May	56	10	30 Apr.	50	−1
1986	WMREC	Jonathan	29 Apr.	230	11 May	58	12	13 May	65	+2
	AFRS	Jonathan	28 Apr.	145	8 May	56	10	9 May	62	+1
1987	WMREC	Jonathan	...	...	...	...	...	NBO	...	...
	AFRS	Jonathan	8 May	131	18 May	58	10	18 May	58	±0
1988	WMREC	Jonathan	9 May	121	19 May	58	10	19 May	58	±0
	AFRS	Jonathan	11 May	133	22 May	59	11	22 May	59	±0
1989	WMREC	Jonathan	30 Apr.	137	21 May	58	21	23 May	67	+2
	AFRS	Jonathan	1 May	149	22 May	57	21	NBO	...	...
1990	WMREC	Gala	27 Apr.	291	11 May	52	14	14 May	54	+3
	AFRS	Jonathan	25 Apr.	133	7 May	56	12	6 May	54	−1
1991	WMREC	Jonathan	28 Apr.	206	10 May	54	12	NBO	...	...
	AFRS	Jonathan	29 Apr.	182	12 May	52	13	NBO	...	...
1992	WMREC	Jonathan	25 Apr.	109	15 May	57	20	NBO	...	...
	AFRS	Jonathan	3 May	133	22 May	58	19	NBO	...	...
1993	WMREC	Jonathan	5 May	412	12 May	55	7	13 May	60	+1
	AFRS	Jonathan	4 May	368	11 May	58	7	11 May	58	±0

[t] WMREC = Western Maryland Research and Education Center, Keedysville; AFRS = Appalachian Fruit Research Station, Bardane, West Virginia.
[u] Predicted date of first fire blight infection.
[v] EIP = epiphytic infection potential (relative population of epiphytic *Erwinia amylovora*); CDH = cumulative degree hours (accumulation of 110 CDH > 18.3 estimates threshold population of bacteria required for infection [EIP = 100]).
[w] Predicted date of first blossom blight symptoms (BBS).
[x] CDD = cumulative degree days for interval between predicted date of first infection and predicted date of first blossom blight symptoms.
[y] Time between infection and predicted date of blossom blight symptoms.
[z] NBO = no blossom blight observed.

These assessments were based on the USDA blight scoring system (18) as recorded from our observations and reports from growers. For purposes of statistical tests, blight severity was recorded on a 0–3 scale (none to severe).

Orchard research sites. Orchards in which MARYBLYT was evaluated were located at the Appalachian Fruit Research Station (AFRS) in Bardane, West Virginia, the West Virginia University Experiment Farm (WVU) in Kearneysville (5 km west of AFRS), and the Western Maryland Research and Education Center (WMREC) in Keedysville (27 km northeast of AFRS).

At AFRS, all weather and research data were collected in blocks of the susceptible apple cultivars Jonathan and Rome Beauty. Tree age varied from 5 to 12 yr. Temperature and rainfall data were collected with a remote weather station (Automata, Grass Valley, CA), a hygrothermograph (Belfort Instrument Co., Baltimore, MD), and a standard minimum-maximum thermometer in a weather shelter.

To determine the presence of epiphytic bacteria, bulk samples of 100–200 blossoms were collected once or twice each week from a 1.5- to 4.0-ha orchard block and placed in a clear polyethylene bag (20). Tap water (0.5 ml per flower) was added to each bulk sample. The bags were shaken for 30 sec, and a 0.1-ml sample of wash water from each bag was spread on Miller-Schroth selective medium (6). Plates were then incubated at 26 C (79 F) for 48–72 hr, and the number of *E. amylovora* colonies was counted.

The model's EIP index was compared with the recovery of *E. amylovora* from blossoms in 1985, 1987, 1988, 1990, and 1993, and the first BBS prediction of the season was compared with the appearance of natural symptoms and of those from artificial inoculations during 1987–1993. A nonparametric sign test (10) was used to test the null hypothesis that each difference between observed and predicted fire blight had a median value of 0.

At WVU, the MARYBLYT model was evaluated during 1990–1993 in a mini-block (six × six trees) of Rome Beauty apples on M.111 rootstock established in 1981. Temperature and rainfall data were collected with a hygrothermograph and manual rain gauge. In 1990, the trees were randomly assigned to receive one of three treatments (three single-tree replicates) just prior to the first predicted blossom blight infection period: 1) streptomycin (Agri-Strep 17WP, 100 mg/L), 2) water, and 3) nontreated control. The streptomycin and water only treatments were applied to visible runoff with a handgun sprayer. The number of fire blight strikes per tree were counted approximately 1 wk after MARYBLYT predicted BBS. Presence of *E. amylovora* in blossoms was confirmed by isolation on nutrient-yeast-glucose agar (NYGA). Proof of pathogenicity was made by inoculating immature pear fruit. In all 3 yr, symptom development was monitored by visual examination of blossoms at regular intervals and was compared with that predicted by the MARYBLYT model. In addition, during 1990–1993, copies of the MARYBLYT program were distributed to two cooperating fruit growers and two other growers provided daily weather and phenological data by telephone during the prebloom and bloom periods.

At WMREC, fire blight observations were made in a block of 8-yr-old Gala apple trees in 1990 and on 9- to 10-yr-old Rome trees during 1984–1985. The remaining observations (1986–1989, 1991–1993) were collected in grower orchards on 11- to 18-yr-old Rome and Jonathan trees in Smithburg, Maryland, and Gala trees in Thurmont, Maryland. Temperature and rainfall data were collected with a hygrothermograph and manual rain gauge.

For all locations combined, the Spearman nonparametric correlation coefficient (r_s) was determined for the relationship between disease severity and EIP at first infection and between severity and the number of consecutive high risk plus infection risk days.

Blossom inoculations. Artificial blossom inoculations were used in a series of tests at AFRS to validate the MARYBLYT model. During seven bloom seasons (1987–1993), individual flowers were inoculated with 50 µl of 10^8 cfu/ml of *E. amylovora* applied through a micropipette dispenser into blossom nectaries. Cultures of virulent strains of the bacterium were maintained on NYGA slants. Specific concentrations of bacterial cells were prepared by suspending cells in 0.1 M phosphate buffer (pH 6.5), and cell densities were determined with a previously calibrated photoelectric colorimeter (Klett-Summerson, Inc., New York). Dilution platings were performed on NYGA plates to verify numbers of bacterial cells in specific concentrations of inoculum. Colony-forming units were counted after 72 hr. Data on differences between observed and predicted blossom blight were analyzed with the nonparametric sign test (10).

During 1987–1990, additional inoculation experiments were performed with various dosages of *E. amylovora*: 10^2, 10^3, 10^4, 10^5, and 10^8 cfu/ml and 10, 25, and 50 µl of inoculum suspension per blossom. To avoid the influence of natural inoculum, unopened Jonathan apple blossoms were inoculated with three separate strains of *E. amylovora* at three concentrations in phosphate buffer as well as with a mixture of all three strains. Controls were injected with buffer only. In addition, pear blossoms were forced from dormant buds in a growth chamber maintained at 21 C (70 F) and 40% RH. When 100 blooms were fully open, their stigmas were infested with *E. amylovora*, carefully applied with a toothpick from a 24-hr culture on NYGA. Data on percent blighted blossoms and clusters were subjected to analysis of variance, and means were separated with single degree of freedom orthogonal contrast comparisons. For the inoculum concentration experiment, data were analyzed with regression techniques to determine optimum fit to a linear or polynomial equation.

Table 3. Comparison of earliest appearance of fire blight symptoms on Jonathan apple blossoms artificially inoculated with 10^8 cfu/ml of *Erwinia amylovora* and predicted date of blossom blight symptoms (BBS) by MARYBLYT during 1987–1993

Year	Date of inoculation	Date of first symptoms	Incubation period (days)	Date of predicted BBS[y]	Difference (days)	CDD >12.7[z]
1987	1 May	11 May	10	14 May	−3	57
	7 May	15 May	8	17 May	−2	55
1988	5 May	16 May	11	17 May	−1	54
	13 May	22 May	9	22 May	±0	53
1989	28 Apr.	14 May	16	21 May	−7	61
1990	24 Apr.	3 May	9	4 May	−1	57
1991	18 Apr.	2 May	14	7 May	−5	57
	28 Apr.	9 May	11	12 May	−3	58
1992	30 Apr.	7 May	7	18 May	−11	57
1993	2 May	8 May	6	10 May	−2	61
	4 May	9 May	5	11 May	−2	56
Mean			9.6		−3.4	57

[y] Calculated as if infection occurred on date of inoculation.
[z] Cumulative degree days for interval between predicted date of first infection and predicted date of first blossom blight symptoms.

RESULTS

Orchard research sites. The 10-yr average bloom periods in western Maryland and eastern West Virginia ranged from 10–15 April to 10–15 May, with a general peak between 20 April and 5 May (Table 1). The total bloom period for Jonathan ranged from 9 days in Maryland (1987) to 25 days in West Virginia (1991).

At AFRS in West Virginia, blossom blight was severe in 2 yr (1985 and 1993), moderate in 1 yr (1990), light in 4 yr (1984, 1986, 1987, and 1988), and absent

in 3 yr (1989, 1991, and 1992). In the 7 yr when blossom blight occurred, the MARYBLYT model predicted the appearance of first visible symptoms 10 times within ±1 day (Table 2). In 1989, 1991, and 1992, however, the model predicted blossom blight but none occurred. EIP values for first infection were 149, 182, and 133 at AFRS in 1989, 1991, and 1992, respectively, and 206 and 109 at WMREC in 1991 and 1992, respectively. At AFRS, the EIP reached only 242 by petal fall in 1991 and a maximum EIP total of only 170 during the entire bloom season of 1992. The general occurrence and severity of blossom blight positively correlated with the EIP at first infection risk dates; in general, EIP below 300 resulted in light to no blossom blight, whereas EIP above 300 resulted in moderate to severe blossom blight (1985 and 1993) ($r_s = 0.61$, $P \leq 0.007$). This correlation agrees with the observation by Zoller and Sisevich (22) that 40% of pear blossoms contained *E. amylovora* bacteria at 336 CDH.

The number of consecutive high infection risk dates had a positive correlation with disease severity ($r_s = 0.65$, $P \leq 0.002$), with seven or more consecutive high risk plus infection risk days resulting in severe blossom blight (with one exception at WMREC in 1991).

The average number of CDD > 12.7 C between 18 first predicted infection dates and predicted BBS dates calculated in the MARYBLYT program was 56.8 and the average number for 13 actual blossom blight observations was 59.7. The difference between predicted BBS dates and observed blight dates averaged 1.8 days. Blossom blight symptoms were never observed more than 1 day before the predicted BBS date. The nonparametric sign test was not significant (chi-square = 1.23, df = 1), indicating that MARYBLYT accurately predicted the occurrence of blossom blight symptoms.

In the experiments and observations at WVU, the first natural infection period on Rome Beauty blossoms in 1990 resulted in a mean of 8.6 blossom infections per tree, compared with fewer than 1.0 infection per tree when streptomycin was applied the day prior to natural infection (significantly different at $P \leq 0.05$). When environmental conditions (except for presence of moisture) and the EIP (at 306) were favorable for blossom blight infection, the application of water with a handgun sprayer initiated blossom blight and resulted in a mean of 5.3 infections per tree. However, this degree of blight was not significantly different from the level of natural infection.

In 1990, MARYBLYT predicted occurrence of BBS to within 24 hr of blight appearance. The presence of *E. amylovora* was confirmed by isolation. As at AFRS, the model predicted two infection risk dates (in 1991 and 1992) when blossom blight symptoms did not develop. Generally, when symptoms were not detected after recorded infection dates, the EIP level was at or just above the minimum value, reflecting a low inoculum (infection) potential.

At WMREC, blossom blight was moderate to severe in 5 yr (1985, 1986, 1990, 1992, and 1993) and light or absent in 5 yr (1984, 1987, 1988, 1989, and 1991). Generally, bloom periods coincided closely with those at AFRS, except in 1987. Comparison of the blossom blight occurrences for AFRS and WMREC in 1985, 1987, and 1989 added validity to the MARYBLYT system. Blossom blight was severe at both locations in 1985 but light in West Virginia in 1987 and light in Maryland in 1989.

Blossom inoculations. The incubation periods (the time between inoculation and first visible symptoms) with artificial inoculations were compared with the predicted BBS dates, calculated as if infection occurred on the date of inoculation. Of the 11 periods, one showed no difference, seven varied 1–3 days, and three varied 5–11 days (Table 3). The latter three occurred under conditions of extended cool weather (12.8–18.0 C) and coincided with an absence of natural blossom blight. The sign test indicated a significant difference between observed and predicted blossom blight (chi-square = 8.1, df = 1, $P \leq 0.005$). The average CDD > 12.7, calculated from the date of inoculation to date of first observed symptoms, was 57, which is consistent with the model's algorithm for BBS.

When stigma surfaces of 100 forced pear blossoms were carefully infested and kept at 21 C and low humidity (40%), 68% became infected, but infection never spread down to the base of the pistils (Fig. 1D). In 1987, more apple blossoms and clusters blighted after inoculation with 10^8 cfu/ml than after inoculation with 10^5 or 10^2 cfu/ml; the latter two inoculum dosages also were significantly different ($P \leq 0.0001$) (Table 4). Differences were observed among strains, with strain 1112 causing more blighted blossoms than strain 260 or the mixture and more blighted clusters than either strain 259 or 260 or the mixture. Strain 260 also caused fewer blighted clusters than strain 259 or the mixture. The incubation period for this experiment averaged 10.8 days.

Analysis of variance showed a significant effect ($P \leq 0.0001$) for inoculum concentration in individual Jonathan blossoms inoculated in 1989 with 10^5, 10^4, 10^3, or 10^2 cfu/ml of a mixture of three strains of *E. amylovora*. Only concentrations at 10^5 cfu/ml caused significantly greater blossom and cluster infections than the buffer control. The level of natural infection in blossoms and clusters did not differ significantly from the buffer control or inoculations with 10^2 cfu/ml. No significant relationships were noted for the effects of inoculum droplet size (50, 25, and 10 μl) and the interaction between inoculum concentration and droplet size. Inoculum concentrations below 1×10^5 cfu did not cause infection greater than that with buffer alone. When regression analysis was performed on the relationship between inoculum concentration and percent infected blossoms and clusters, linear regression did not reveal a significant relationship between inoculum concentration and percent infection for either blossoms or clusters. However, a polynomial regression equation of the form $Y = b_0 + b_1 I + b_2 I^2 + b_3 I^3$, where Y = percent infection and I = inoculum concentration (in colony-forming units) was significant for the relationship between inoculum concentration and blossom infection: $Y = 8.84 + 0.0134^* I - 1.33 \times 10^{-6^*} I^2 + 1.19 \times 10^{-11^*} I^3$, with all parameter estimates significant at $P \leq 0.01$ and $R^2 = 0.33$ ($P \leq 0.0001$). Parameter estimates for percent cluster infection were not significant ($P \leq 0.05$) (*data not shown*).

DISCUSSION

The MARYBLYT model was quite accurate in predicting the appearance of

Table 4. Percentage of blighted blossoms and blossom clusters on Jonathan apple after artificial inoculation with three concentrations of three strains, alone or mixed, of *Erwinia amylovora* in West Virginia, 1987

	Percent blighted blossoms[x]			Percent blighted clusters[y]		
Strain	1×10^8 cfu/ml	1×10^5 cfu/ml	1×10^2 cfu/ml	1×10^8 cfu/ml	1×10^5 cfu/ml	1×10^2 cfu/ml
AFRS 259 (NY)	61.5 ab[z]	20.0 a	12.5 a	82.5 ab	52.5 b	7.5 ab
AFRS 260 (NY)	46.5 b	18.0 ab	2.5 b	15.0 c	22.5 bc	0.0 b
AFRS 1112 (Ont.)	79.5 a	30.0 a	5.0 b	100.0 a	72.5 a	17.5 a
Mixture	55.0 b	18.5 a	1.5 b	77.5 b	45.0 ab	20.0 a
Natural infection	2.0 c	2.0 bc	2.0 b	7.5 c	7.5 c	7.5 ab
Control (buffer)	0.0 c	0.0 c	0.0 b	0.0 c	0.0 c	0.0 b

[x] Based on inoculation on 1 May (first symptoms on 12 May) of 200 blossoms (five blooms in each of 10 blossom clusters) with 50 μl of bacterial suspension in a micropipette; each value is the mean of four replications.

[y] Based on examination on 20 May of 40 blossom clusters with infection advanced into woody tissue showing necrosis in nearest leaves; each value is the mean of four replications.

[z] Means followed by different letters in columns are significantly different ($P \leq 0.05$) according to single degree of freedom orthogonal contrast comparisons.

natural blossom blight symptoms at one location in West Virginia and one in Maryland. Our results confirm those of Jones (3) on apple in Michigan. Actual blossom blight symptoms were usually observed 1–2 days after they were predicted to occur. In the years in which blossom blight symptoms were predicted, early symptoms were visible when orchards were visited a few days before or after the predicted date, indicating that infections had occurred about as predicted (Table 2). MARYBLYT predicted the possibility of blight at AFRS in 1989, 1991, and 1992 and at WVU and WMREC in 1991 and 1992, but no symptoms were observed. Either weather conditions were unfavorable for blight development or we did not examine enough trees to detect it at very low levels.

With artificial inoculations, 10 μl of 10^3 cfu/ml and 50 μl of 10^2 cfu/ml resulted in blossom infections of 4.0 and 1.5%, respectively. Blossom infections of 9.0–15.0% resulting from inoculation with 10–50 μl of buffer only seem to indicate that epiphytic *E. amylovora* was present in the blossoms. This confirms reports by numerous investigators (1,2,14,15,19). The MARYBLYT model could not be validated with the artificial inoculations and consistently overestimated the time required for the first visible symptoms to appear. One explanation may be that the time required for bacterial multiplication is not accounted for with artificial inoculation, resulting in visible symptoms before the predicted date. This suggests that caution should be employed in developing and validating disease models based on artificial inoculations.

Breaking down the various types of fire blight into categories (13) on the basis of time and place of infection has contributed significantly to our understanding of the disease, especially the blossom blight phase. One correlation that seems to have the greatest bearing on the severity of fire blight from year to year is the epiphytic infection (inoculum) potential. An EIP of 100 (= 110 CDH > 18.3 C) is the threshold MARYBLYT uses to predict blossom infections and is based directly on the report of Zoller and Sisevich (22) for colonization of 3–5% of pear flowers in California. The amount of the disease that is evident from year to year seems to be directly correlated with the EIP and the number of days in which conditions are favorable for infection. The higher the EIP and the longer conditions remain favorable, the greater the chances for infection to continue. The severity of fire blight also increases under these conditions (22).

Some features of MARYBLYT may need additional clarification to make the model useful for individuals not familiar with the disease and its prediction (4,13). One requirement for the prediction of blossom blight is that blossoms must be open with petals intact. Thus, this stage starts when the very first blossom opens. Also, the observation of the time of petal fall is critical, because the model stops the blossom blight prediction phase as soon as petal fall is entered in the phenology column. Therefore, this stage should not be entered until the very last blossom has dropped its petals. Also, when late or secondary ("rattail") bloom occurs, potentially serious blossom infections may be missed. In 1993 at AFRS, "late" blossoms were observed on Empire apple, and these initiated fire blight rather than the normal bloom about 2 wk earlier. Growers using the model will need to make a judgment about extending the blossom phase by delaying the entry of petal fall when secondary bloom remains present. The current MARYBLYT version 4.1 provides an audible warning to consider this fact.

Judgment is required also when entering certain wetness situations. In the MARYBLYT program, wetness is defined as the actual presence of wet foliage and not just wet grass or ground cover on the orchard floor. The contributions of dew and fog of various durations and intensities to infection may require some subjective judgment if these situations are to be entered correctly. The same is true in using the trauma blight feature of MARYBLYT after injury to trees by frost, hail, or windstorms. While frost is defined by the minimum temperature of −2.2 C (≤28 F), hail and wind must be severe enough to cause damage to blossoms or foliage and, hence, may be judged somewhat subjectively.

The MARYBLYT computer model has proved to be an effective tool for growers, extension workers, and researchers. The most important aspect of the program is the ability to predict outbreaks of fire blight based on tree phenology, pathogen development, and weather conditions. The number of antibiotic sprays (approximately 28) that would have been applied in 10 years using the MARYBLYT prediction system would have been approximately one-half the number applied routinely without a prediction system. In addition, eliminating unnecessary sprays would reduce the selection pressure for strains of *E. amylovora* resistant to streptomycin.

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