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Management of Frost Injury, Fire blight, and Fruit Russetting of Pear Using Biological and Cultural Methods.

Principal Investigator: Steven E. Lindow, Department of Plant and Microbial Biology, University of California, Berkeley, CA 94720-3102.
email - icelab@socrates.berkeley.edu.
Telephone (510) 642-4174. Fax (510) 642-4995.

Cooperators: Rachel Elkins, Cooperative Extension, Lake County, 883 Lakeport Blvd. Lakeport, CA 95453

Chuck Ingels, Cooperative Extension, Sacramento County, 4145 Branch Center Road, Sacramento, CA 95827-3898

ABSTRACT

The process of colonization of pear buds and flowers was monitored in 3 commercial pear orchards. Both the total number of indigenous bacteria in buds as well as the proportion of those bacteria that were "internal" to pear tissue varied greatly between orchards. Total indigenous bacterial populations associated with pear in the early spring in 2002 were generally low in number compared to that observed in previous years, with mean populations in individual buds ranging from about 100 to 10,000. Generally, a majority of the bacteria associated with buds were external, while the proportion that was internal to pear tissue increased in developing flower and tissues after bud break. The fraction of the total bacterial population associated with pear buds that was "internal" was generally less than 10% in all orchards, and highest populations were observed in Lake County orchards. Bacterial populations generally increased rapidly after bud enlargement. The applications of erradicant bactericides before bloom slightly enhanced the colonization of flowers with the biological control bacterium *Pseudomonas fluorescens* strain A506 early in the growing season compared to control trees to which the bacterium was applied without erradicant pre-treatment. More enhancement has been seen in other years when there were more indigenous bacteria to interfere with colonization. Application of strain A506 with the organo-silicon surfactant Breakthru resulted in a higher proportion of colonized flowers, especially early in the growing season than when the bacterium was applied in water alone. The proportion of flowers colonized with strain A506 throughout the spring generally increased with increasing concentrations of surfactant in which the bacteria were applied in a single application at the time of first bloom. Importantly, most flowers emerging from trees that were inoculated with strain A506 only a single time at "first bloom" with Blightban A506 in 0.2% or 0.5% Breakthru were as high as or higher throughout the main bloom, and into delayed bloom than that on trees that received weekly applications of the same amount of Blightban A506. Blightban A506 was also applied with high rates of surfactant in 2 large replicated trials in commercial orchards in 2002. No russetting of fruit was observed in any plot when surfactant was applied before substantial bloom had occurred. Russetting of fruit was observed only in special test plots in which trees were treated with surfactant at mid-bloom or later. These results suggest strongly that we should be able to greatly reduce the number of applications of the bacterium by applying it early in the season with a penetrating surfactant. In addition, by applying the bacterium only once early in the early spring before applications of Dithane and Terramycin and other pesticides are subsequently made to trees, we can avoid potential problems with compatibilities of the bacterium with these other pesticides.

Colonization of pear buds and flowers with indigenous bacteria in the early spring

Work during 2002 addressed important issues in the microbial ecology of pear trees that relate to the management of fruit russet, frost injury, and fire blight disease of pear. One major objective was to monitor populations of indigenous bacteria in pear buds and emerging florets during winter/early spring and relate them to weather parameters such as spring rainfall. An earlier analysis of data in which we had measured indigenous bacterial populations on the flowers of pear at the beginning of bloom revealed large variations in population size from year to year. In some years such as 1995 and 1998 there were large populations of bacteria of all types in flowers shortly after they open in the spring, while in other years flowers emerged nearly sterile and become colonized by bacteria that apparently immigrated to the open flower via the air or insect vectoring from nearby plant sources such as orchard cover crop and weed species. We presumed that winter rains allow colonization of the buds and/or the emerging florets as the buds begin to open. Importantly, we had found that the incidence of early season fire blight infection is generally inversely proportional to the population size of the indigenous bacteria in the emerging flowers on control trees (eg. there is less disease in years when there are a lot of indigenous bacteria - "natural biological control" of fire blight seems to be operating). We also noted that the colonization of flowers by antagonistic bacteria such as *Pseudomonas fluorescens* strain A506 is less efficient in years when there are large indigenous bacterial populations. Fortunately, the presence of large numbers of indigenous bacteria can apparently confer some degree of "natural biological control" in those years when A506 itself is inhibited in its growth on pear by the presence of indigenous bacteria. The presence of large numbers of indigenous bacteria on emerging pear flowers is also associated with a relatively high incidence of frost injury during mild frost events compared to orchards and years when indigenous bacteria populations were low. We therefore undertook a detailed temporal analysis of the processes that allow bacteria to develop in emerging pear flowers to better understand how to manage biological control agents of fire blight disease as well as frost injury and fruit russet and to better predict when indigenous bacteria will be sufficiently numerous to present a high hazard of fruit russet and frost injury. Beginning about 4 weeks before bud break in 2002 we monitored the process of colonization of pear buds and the flowers that emerged from these buds on a frequent basis to determine how rapidly bacterial populations changed and what weather factors were associated with the development of bacterial populations in buds and flowers. Bacterial populations were monitored on buds and flowers in two commercial pear orchards in Lake County, and one orchard in Sacramento County. In addition to measuring the total bacterial population on buds and flowers we also measured the "internal" populations. At each sampling time the 40 bud or flower samples for each orchard were divided into two separate pools of 20 samples each. Total bacterial populations were determined by macerating the bud or flower samples from one pool individually in a small amount of buffer and plating appropriate dilutions onto non-selective media. In contrast, "internal" populations were determined as before on the other 20 samples in a given pool after the buds or flowers were surface sterilized by treatment with 0.5% sodium hypochlorite.

Both the total number of indigenous bacteria in buds as well as the proportion of those bacteria that were "internal" to pear tissue varied somewhat between orchards (Figures 1-3). Total indigenous bacterial populations associated with pear in the early spring in 2002 was generally quite low, reflecting the relatively dry spring. Average total bacterial populations per bud ranged from about 100 to 10,000 cells in the various orchards. In most orchards there was little change in populations from February through March. (Figures 1-3). The fraction of the total

bacterial population associated with pear samples that was "internal" was generally about 10% in all of the orchards before bud break (Figures 1-3). Thus it seems that most bacteria on buds are not "inside" the buds and thus might be expected to be influenced strongly by both winter pesticide applications, as well as weather conditions. The dry weather in the spring of 2002 is consistent with the low populations of bacteria seen on buds and developing flowers; these populations are much lower (1000-fold) than those seen during wet springs such as1995 and 1998. The results of 2002 confirm our suspicions that bacterial population development in pear occurs rapidly during bud enlargement, and is not associated with large internal populations that developed during winter since the winter months.

In contrast to the relatively stable populations of bacteria that were observed on buds and "fingers" of pear in the early spring, more rapid bacterial multiplication was noted in some orchards upon flower opening (Figures 1-2). For example, total bacterial populations after mid-March on flowers emerging in a Sacramento County orchard increased over 100-fold within a few days (Figure 1). It was interesting to note that the increases in total bacterial populations were not associated with increases in "internal" populations, and thus the proportion of the total bacterial population that were within pear tissue decreased to less than 0.1% (Figure 1). The percentage of bacteria inside pear tissue thus seems to be somewhat variable from site to site within a year and between years; the factors affecting such variation are still unclear, but this phenomenon has importance for the control of frost and russet since such bacteria are important inoculum sources. This study is designed to be a relatively long-term study in which we will examine colonization of pear under a variety of weather conditions over a number of years so that the influence of weather conditions on bacterial populations can be better predicted. Predictions of indigenous bacterial populations before bloom will be useful in future predictions of the severity of fruit russet and of frost injury that are associated with these indigenous bacteria.

Eradication of indigenous bacteria on pear in early spring to reduce fruit russet and enhance colonization of pear by biological control agents.

Since relatively large indigenous bacterial populations had been found on pear tissue at the time of first bloom in many previous years and since these large populations were associated with poor establishment of the biological control agent *Pseudomonas fluorescens* strain A506, we evaluated strategies to reduce such early season populations. Since the severity of fruit russet is proportional to the total population sizes of bacteria on pear tissues in the early spring (due to the contribution of IAA-produced by a subset of these bacterial populations), we were interested to know if we could reduce the population sizes of indigenous bacteria and thereby reduce the severity of fruit russet. *Pseudomonas fluorescens* strain A506, for which extensive field experimentation has been conducted over the last several years in California, has been registered as the product "Blightban A506" and commercial sales began in the 1996 growing season. We therefore conducted additional research to address issues relating to the best methods by which this bacterium can be established in pear trees to achieve biological control of frost injury, fire blight disease, and fruit russet, especially under conditions when pears might already harbor substantial populations of other bacteria in the early spring.

A field trial in which several early-season chemical eradicants were evaluated was established in a commercial pear orchard in Lake County. Both bleach (Sodium hypochlorite -

0.05%) as well as copper hydroxide (Kocide 101) was evaluated as bactericides to eradicate indigenous bacterial populations. These eradicants were applied with a high rate of this surfactant (0.5%) to determine if the bactericide could be made more accessible to the bacteria in and on pear tissues. Such eradicant treatments were made at first bloom (when the very first blossoms in an orchard had opened, but the vast majority were still closed). At about 10% bloom, weekly applications of *P. fluorescens* strain A506 (label rate of Blightban A506) were initiated. The application of eradicant bactericides increased the establishment of strain A506 early in the growing season (Figure 6). The proportion of flowers that were colonized by P. *fluorescens* strain A506 was higher at early sampling dates on trees receiving weekly applications of Blightban A506 on trees treated with eradicant bactericides than on trees without such earlier chemical treatments (Figure 6). In previous years we had noted a much larger effect of eradication of indigenous bacteria on enhancing colonization of strain A506. The relatively low indigenous bacterial populations on newly opened flowers in 2002 could not be substantially lowered with our eradicative treatments, hence making enhanced colonization rather modest. We also compared the colonization of pear by strain A506 when it was applied to trees in water without a surfactant and when it was applied in a solution of 0.05% Breakthru (Figure 5). Because the colonization of pear flowers was so efficient even without addition of Breakthru we did not see a consistent increase in the proportion of the flowers that were colonized by this bacterium when applied with the surfactant as we had previously seen. In the early spring we saw a higher proportion of flowers colonized, but this was not as apparent later, when nearly all flowers were colonized in either treatment. We had found that the bacterium is applied more uniformly to plant surfaces when mixed with this surfactant that has a very low surface tension. Presumably the surfactant allowed the bacterium to more fully colonize parts of the flowers and young fruit by allowing sprayed inoculum to move easily to all parts of the developing tissues. This also reflects a greater ability of the bacterium to prevent the growth of deleterious bacteria such as ice nucleation active bacteria capable of causing frost injury, of IAA-producing bacteria that cause fruit russet, and of *Erwinia amylovora*, the pathogen that causes fire blight disease.

The severity of fruit russet on trees receiving applications of Blightban A506 was often numerically lower than that of untreated trees in most cases, but because of variation in russet severity between trees often did not differ statistically (Table 1). The severity of fruit russet was higher on trees in which early season eradicant bactericides were applied in addition to weekly applications of Blightban as compared to where Blightban alone was applied (Table 1). We presume this was due to phytotoxicity of the copper compounds and bleach to the developing fruit; such phytotoxicity had not been previously seen, and we are not sure what aspects of the weather or flower development in 2001 predisposed them to phytotoxicity. It is also noteworthy that the application of a high rate of Breakthru (0.5%) at first bloom to this plot was not associated with fruit russet (Table 1). Thus this penetrating surfactant apparently does not harm pear tissues even at high rates when applied in the early spring to flowers that were not yet open. There was very little frost injury in the plot areas in 2002, so it was not possible to demonstrate frost control with strain A506 treatments as in previous years. While many A506 treatments significantly decreased frost injury, the levels of frost injury in all treatments was low (Table 1). The incidence of frost injury was reduced by over 50% by treatments of Blightban A506 (Table 1). The greatest frost control was associated with the application of strain A506 with 0.05% Breakthru, the treatment that achieved the highest population sizes of the biological control agent.

Establishment of *P. fluorescens* strain A506 in pear flowers by single early season applications with surfactants

Given what we had observed in previous years, that rapid colonization of flowers by indigenous bacteria could occur if flowers were inoculated with bacteria, we investigated approaches by which *P. fluorescens* strain A506 could be introduced into pear tissues before bloom so that flowers would be exposed to this antagonistic bacterium as soon as they opened. We evaluated the potential of introducing bacteria into pear tissues using relatively high rates of the penetrating surfactant Breakthru. Just as we had demonstrated in walnut that this surfactant could allow topically applied solutions to penetrate into tissues, we hypothesized that suspensions of *P. fluorescens* strain A506 could be made to enter pear tissues if applied with such surfactants. Organo-silicon surfactants such as Breakthru and related compounds have the unique ability to allow water solutions to penetrate into plant tissues through natural openings due to the low surface tension of such solutions. Normal sticker-spreaders do not have a sufficiently low surface tension to permit such penetration into plants. Laboratory tests had indicated that strain A506 was tolerant to over 3% Breakthru. Thus this bacterium was compatible with even high rates of surfactant. We inoculated pear trees at the time of the first bloom in an orchard with Blightban A506 with different concentrations of Breakthru and then measured the colonization of flowers by strain A506 after they emerged. Colonization of pear flowers from a single early season spray of strain A506 (at first bloom) led to high levels of colonization of pear flowers, largely irrespective of the amount of surfactant that was applied with the bacterium (Figure 4). Particularly early in the bloom, the fraction of flowers that were colonized by strain A506 was much higher when it was applied a single time with at least 0.1% Breakthru; very few flowers that were sprayed before they opened with the bacterium in water alone were colonized (Fig. 4). There was quite a bit of variability in our estimates of the fraction of flowers that were colonized, and we do not know why this was the case. In general, the fraction of flowers that were colonized with strain A506 were higher on trees receiving a single pre-bloom application of strain A506 in surfactant than on trees that were sprayed weekly with the bacterium in water alone. Thus the work of 2002 again demonstrated that a single early-season application of Blightban A506 is an effective method of applying the bacterium.

Because we had observed phytotoxicity on fruit in 2001 on trees treated with Blightban A506 at advanced stages of bloom (but not when applied before significant bloom had occurs) we established two trials in 2002 to determine the effect of application time of a mixture of 0.5% Breakthru and Blightban A506 on both the colonization efficiency of pear flowers, as well as on phytotoxicity. In a Lake County trial we found that an application of this mixture at a single time led to similarly higher levels of flower colonization by strain A506 irrespective of whether it was applied at first bloom or at full bloom (Fig. 7). Obviously, delaying application of the Blightban A506 + Breakthru mixture is not wise, since the purpose of its use is to preemptively colonize flowers with this antagonistic bacterium and thus such late application times leaves flowers unprotected from colonization by the fire blight pathogen as well as ice nucleation active bacteria that can cause frost injury. The severity of fruit russet increased substantially when the Blightban + Breakthru mixture was applied at any time after first bloom in this test (Table 5). Similar results were obtained in tests where this mixture was applied to advanced stages of bloom in Sacramento County (Table 4); very little fruit russet was seen when the Blightban + Breakthru mixture was applied at first bloom. These results thus show that application at the time of first bloom is thus probably an optimum time to apply the bacterium when used with a penetrating surfactant.

Blightban A506 was also applied with high rates of surfactant in 2 large replicated trials in commercial pear orchards in 2002. The colonization of emerging flowers from early-season applications of bacteria and/or surfactant differed substantially in the different commercial orchards sprayed with airblast sprayers. At the Lake County orchard the fraction of flowers that were colonized by the biological control organism strain A506 were nearly as high on flowers treated a single time with Blightban A506 with 0.5% Breakthru as on trees treated weekly with Blightban A506 in water alone throughout the spring (Figure 8). Nearly all flowers that were tested throughout the spring were colonized by strain A506 irrespective of whether it had received only the single early application of the bacterium in surfactant or repeated applications throughout the spring (Fig. 8). In this trial we also evaluated the inclusion of iron chelate (1 lb/100 gal of Sequestrene 138) with the repeated Blightban applications (Fig. 8). The addition of iron was evaluated since studies from Oregon researchers had indicated that the efficacy of Blightban A506 for fire blight control could be increased in the presence of added iron by stimulating the bacterium to produce an antibiotic inhibitory to *Erwinia amylovora*. While we did not observe sufficient fire blight in our pear trials to evaluate the effect of iron addition on fire blight control, we did find that iron did not increase nor inhibit the colonization of flowers with strain A506. In a large-scale trial in Sacramento County, we also observed that addition of iron chelate to Blightban A506 sprays did not alter the colonization of flowers by strain A506 (Fig. 9). The application of iron chelate to trees did not alter the fruit russet of pear in either trial (Tables 2 and 3). As in the lake County trial, there was insufficient fire blight to evaluate the effectiveness of treatments on disease control. The possibilities if enhancing fire blight control with iron additions is intriguing and we will continue to evaluate it in trials in 2003.

While a high proportion of pear flowers that had been treated a single time with Blightban A506 + 0.5% Breakthru at first bloom that were sampled during the main bloom (late March/early April) on trees in a large-scale trial in Sacramento County were colonized by strain A506, the fraction of flowers that emerged later in the growing season that were colonized decreased (Fig. 9). When sampled in mid-bloom on March 26, the average population size of strain A506 on flowers treated a single time at first bloom with a mixture of Blightban A506 and Breakthru was higher than that on flowers treated with Blightban A506 weekly in water alone (Fig. 10); many flowers harbored over 3 million cells of strain A506 (Fig. 10). In contrast, a high percentage of flowers that had been sprayed weekly with Blightban A506 in water alone that were colonized by strain A506 remained high throughout most of the spring (Fig. 9). We believe that the reason that flowers throughout the spring were colonized by strain A506 from the single pre-bloom treatment in Lake County and not in Sacramento county is due to the relative timing of application of the bacteria. The bacteria were applied a single time in Sacramento County when the flower buds were relatively more closed compared to that in the Lake County plot. Since flower buds do not open simultaneously, and since the bacteria will gain entry into the more open flower buds more efficiently than to closed buds, it is likely that more of the flower buds were sufficiently open to allow colonization by strain A506 in the Lake County trial than at Sacramento County at the time of the single spray. Thus it is possible that if sprays are applied too early that reduced colonization could result. Our earlier work in 2000 had indicated that colonization of flowers from single applications to “fingers” was much less effective than to buds at “first bloom”. Apparently the stage of flower bud opening that allows flower colonization needs to be better defined. The best evidence is still that the best time to apply the single bacterial treatment with penetrating surfactants is after buds begin to open, but before many flowers appear (since open flowers appear to be at risk of phytotoxicity from the silicon surfactant). We will further explore

the effect of application time on flower colonization in 2003 to better define the "window of time" when Blightban can be both effectively and safely applied to trees.

Because we sampled individual pear flowers in our plots on several occasions both for the numbers of strain A506 (done by washing bacteria from the flowers and then dilution plating onto selective culture media) as well as a simpler assay where we detect the simple presence or absence of bacteria by a "flower rub" assay onto selective media, we could for the first time determine how estimates of colonization done by the different assays could be compared. The flower rub assay is much easier to employ that the flower wash assay, and has been adopted by many PCA's and farm advisors as a simple way to determine if flower colonization by strain A506 from Blightban A506 treatments has occurred. For example when sampled on March 26, average population sizes of strain A506 differed among the treatments applied to trees in the large-scale trial conducted in Sacramento County (Fig. 11). Highest populations of strain A506 (average = 10,000 cells/flower) were observed on flowers treated with Blightban A506 with penetrative surfactant, while lower populations (about 3,000 cells/flower) were recovered from flowers on trees treated weekly with Blightban A506 in water alone (Fig. 11). The incidence of colonization of these flowers was highly correlated with the numbers of cells recovered; while 84% of the flowers from the Blightban + Breakthru treatment had detectable A506 populations, only about 70% from the Blightban only treatment did (Fig. 11). Since one of the Lake County trials had 12 treatments we could compare the incidence of colonization with average A506 populations in flowers. A very striking correlation between increasing average A506 populations in flowers and increasing fraction of flowers with detectable A506 populations was observed (Fig. 12). By comparing Figure 10 with Figure 12 one can better understand why this would be found. Generally there is a wide range of populations of A506 on the same treated tree; some flowers might harbor as many as a million cells of A506 while others would be devoid of this bacterium (Fig. 10). Thus as the average number of bacteria in a group of flowers increases, the likelihood that a given flower has at least some strain A506 increases. The relationship shown in Figure 12 will therefore be very helpful in predicting how effective fire blight control will be given a measured proportion of flowers with at least some A506, since disease control increases with increasing population size of strain A506 in flowers.

An additional large-scale field trial to test the efficacy of early-season application of Blightban A506 done on Pink Lady apple supported the findings on flower colonization reported above for pear. This trial supported in part by the UC-IPM program, and done in cooperation with Brent Holtz of UC Cooperative Extension in Madera County was very similar in design to the two large pear trials described above. The percentage of flowers that were colonized by strain A506 on trees treated a single time at first bloom with Blightban A506 containing either 0.2% or 0.5% Breakthru were as high at all sampling times as that on trees treated weekly with Blightban A506 in water alone (Fig. 13). The populations of strain A506 on flowers treated with the Blightban A506 + Breakthru mixture were somewhat higher at a given date than that on flowers treated repeatedly with Blightban alone (Fig. 14). It is important to note, that when Blightban A506 was applied to trees a single time at first bloom but without Breakthru surfactant, that colonization of flowers was much less than when it was applied with Breakthru (Fig. 13). This points out the importance of the penetrating surfactant in making this strategy of biological control possible. While nearly all apple flowers sampled during the bloom period were colonized by strain A506 irrespective of whether Blightban A506 was applied frequently without surfactant or was applied only at first bloom with surfactant, those few flowers that emerged after mid-May were not frequently colonized. As will pear, the buds and those few flowers that emerge late in

the spring bloom period are somewhat different from main bloom flowers morphologically and were thus not sufficiently open at "first bloom" for strain A506 to penetrate into the flower bud when sprayed with Breakthru. Likewise, these flowers emerged after the last weekly sprays of Blightban in water alone were applied, and thus escaped inoculation. The application of Blightban A506 in Breakthru did not increase the severity of fruit russet on fruit compared to untreated trees (Table 6). Likewise, the application of iron chelate (as Sequestrene 138) did not affect the frequency of colonization of flowers with strain A506 (Fig. 13) nor did it affect fruit russet of apples at harvest (Table 6)

These results are very exciting in that they suggest that early season application of antagonistic bacteria may be a superior means of establishing these biological control organisms on trees. These results confirm that we should be able to greatly reduce the number of applications of the bacterium by applying it early in the presence of the penetrating surfactant. In addition, by applying the bacterium only once early in the early spring before applications of Dithane and Terramycin and other pesticides are subsequently made to trees, we can avoid potential problems with compatibilities of the bacterium with these other pesticides. Since strain A506 can be established on trees before these other pesticides need to be applied, and since we have already shown that the bacterium is quite tolerant of other pesticides such as Dithane and Terramycin if it has established on trees before these pesticides are applied, we can greatly reduce any possibility that they will interfere with the performance of strain A506 in biological control of frost, fire blight and fruit russet. Such an application strategy should thus also help increase the adoption of biological control strategies for fire blight and fruit russet since they will make it easier to integrate into existing management strategies. This approach will be a major focus of our work in the year 2003.

Table 1. Severity of fruit russet and incidence of frost damage of pear treated at first bloom with *Pseudomonas fluorescens* strain A506 in different concentrations of a penetrating surfactant - Lakeport, 2002.

Treatment	Russet Severity (% of surface)	Frost Damage (% of fruit)
Control	0.31 b	1.8 % ab
Kocide + 0.5% Breakthru (1st bloom) +A506 weekly	2.12 a	0.0 % c
Bleach + 0.5% Breakthru (1st bloom) + A506 weekly	0.70 b	0.4 % bc
A506 weekly	0.54 b	0.6 % abc
A506 + 0.05% Breakthru weekly	0.59 b	0.6 % abc
A506 + FeEDDHA (1lb/100gal) weekly	0.47 b	1.3 % abc
A506 + 0.5% Breakthru (1st bloom only)	1.81 b	0.7 % abc
A506 + 0.2% Breakthru (1st bloom only)	0.42 b	1.9 % a
A506 + 0.1% Breakthru (1st bloom only)	0.71 b	2.9 % c
A506 + 0.05% Breakthru (1st bloom only)	0.34 b	0.5 % abc
A506 (1st bloom only)	0.36 b	0.6 % abc
Streptomycin + Terramycin weekly	0.39 b	0.3 % ab

Table 2. Severity of fruit russet and incidence of frost damage in pear in a large replicated trial in which *Pseudomonas fluorescens* strain A506 was applied to trees at bloom in different ways - Lake County, 2002.

Treatment	Russet Severity (% of surface)	Frost Damage (% of fruit)
Antibiotic program	0.62 b	1.7 % a
A506 + Breakthru at 1% bloom only + antibiotic program	2.58 a	0.6 % a
A506 weekly (20% bloom, full bloom, & petal fall)	0.60 b	0.3 % a
A506 weekly (full rate) combined with Fe chelate + antibiotic program	0.47 b	0.7 % a

Table 3. Severity of fruit russet to pear on trees treated at bloom with Blightban A506 with and without a penetrating surfactant - Sacramento County, 2002.

Treatment	Russet Severity (% of surface)
Antibiotics alone	0.72 b
Blightban A506 (1 bag/200 gal.) weekly in water	0.79 ab
Blightban A506 weekly in water + 1lb/100gal. Sequestrene 138	1.07 a
Blightban A506 once at first bloom in 0.5% Breakthru	0.98 ab

Table 4. Severity of fruit russet of pear on trees treated once at various times of bloom with a penetrating surfactant - Sacramento County, 2002.

Treatment	Russet Severity (% of surface)
5% bloom	2.22 c
20% bloom	6.04 ab
80% bloom	7.72 a
Full bloom	4.42 bc

Table 5. Severity of fruit russet and incidence of frost damage to pear at harvest from trees treated at different times at bloom with a silicon-based surfactant -Lake Co., 2002.

Treatment	Russet Severity (% of surface)	Frost Damage (% of fruit)
Breakthru (0.5%) 1st bloom	1.81 b	1.6 % a
Breakthru (0.5%) 20% bloom	8.90 a	0.0 % a
Breakthru (0.5%) 80% bloom	5.22 ab	0.6 % a
Breakthru (0.5%) full bloom	4.82 b	0.4 % a

Table 6. Severity of fruit russet to Pink Lady apples on trees treated at bloom with Blightban A506 with and without a penetrating surfactant - Chowchilla, 2002.

Treatment	Russet Severity (% of surface)
Antibiotics alone	0.47 a
Blightban A506 (1st bloom only)	0.54 a
Blightban A506 (weekly)	0.55 a
Blightban A506 (1st bloom only) + 0.2% Breakthru	0.47 a
Blightban A506 (1st bloom only) + 0.5% Breakthru	0.51 a
Blightban A506 (1st bloom, then weekly) + Sequestrene 138 (1 lb/100 gal)	0.41 a

Figure 1. Total bacterial populations (diamonds) and "internal" bacterial populations remaining in buds and clusters of pear that were surface sterilized with bleach (squares) that were collected from commercial Bartlett pear orchard "E" in Sacramento County in the spring of 2002. The vertical bars represent the standard error of the mean of log-transformed bacterial population sizes.

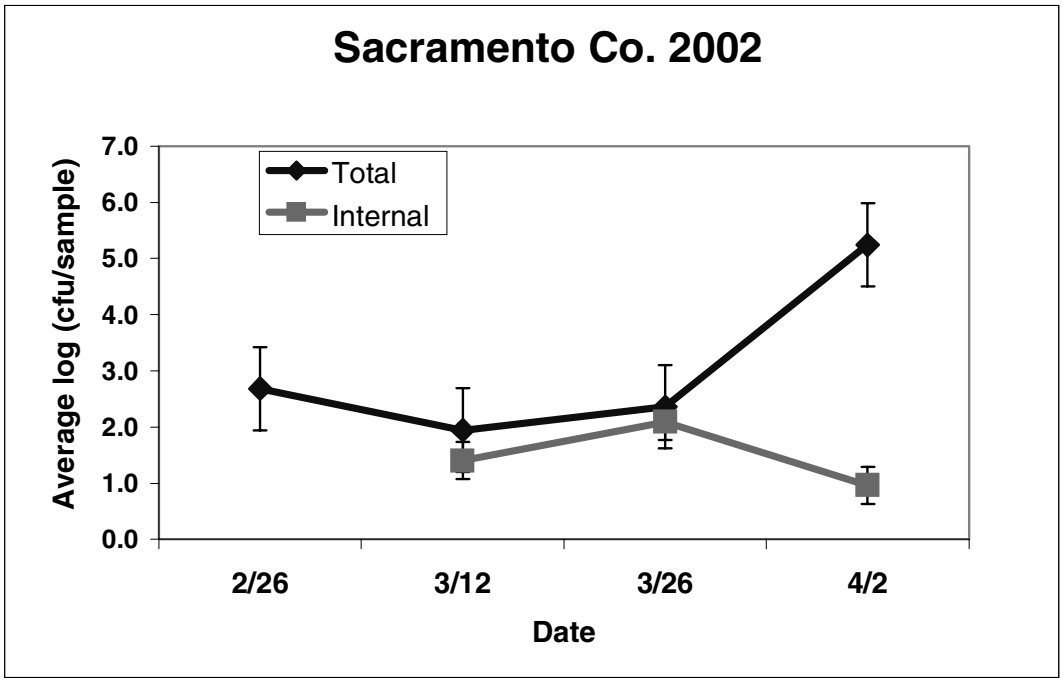


Figure 2. Total bacterial populations (diamonds) and "internal" bacterial populations remaining in buds and clusters of pear surface sterilized with bleach (squares) that were collected from a commercial Bartlett pear orchard in Upper Lake in the spring of 2002. The vertical bars represent the standard error of the mean of log-transformed bacterial population sizes.

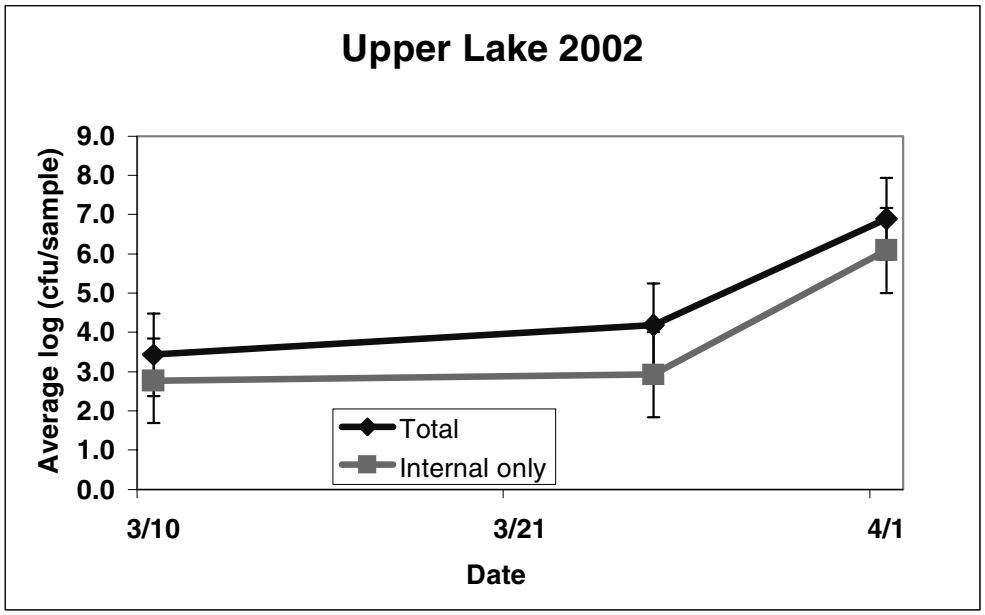


Figure 3. Total bacterial populations (diamonds) and "internal" bacterial populations remaining in buds and clusters of pear that were surface sterilized with bleach (squares) that were collected from a commercial Bartlett pear orchard in Scotts Valley in the spring of 2002. The vertical bars represent the standard error of the mean of log-transformed bacterial population sizes.

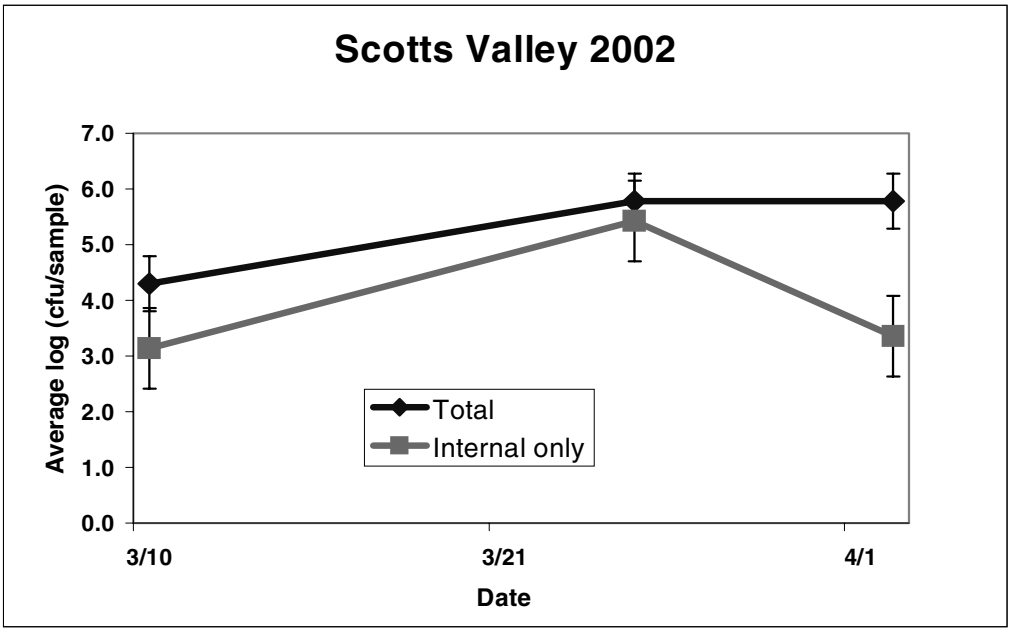


Figure 4. Percent of flowers colonized with *Pseudomonas fluorescens* strain A506 on Bartlett pear trees treated once only at the "first bloom" stage of growth [when only a few flowers were observed in an orchard] with a label rate of Blightban A506 in water alone (circles) or in 0.05% Breakthru (stars), 0.1% Breakthru (x's), 0.2% Breakthru (triangles) or 0.5% Breakthru (squares) compared with weekly applications of Blightban A506 in water (diamonds) in a Lake County plot in 2002. The vertical bars represent the standard error of the mean percent of flowers colonized.

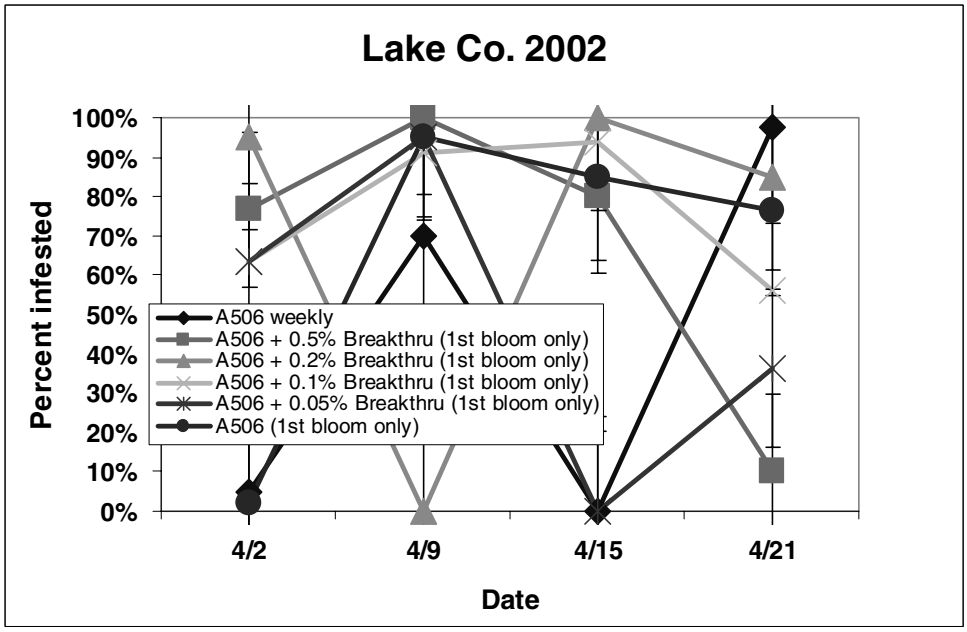


Figure 5. Percent of flowers colonized with *Pseudomonas fluorescens* strain A506 on Bartlett pear trees treated weekly with a label rate of Blightban A506 in water alone (diamonds), in 0.05% Breakthru (squares), or with 1 lb/100 gal Sequestrene 138 (FeEDDHA) (triangles). The vertical bars represent the standard error of the mean percent of colonized flowers.

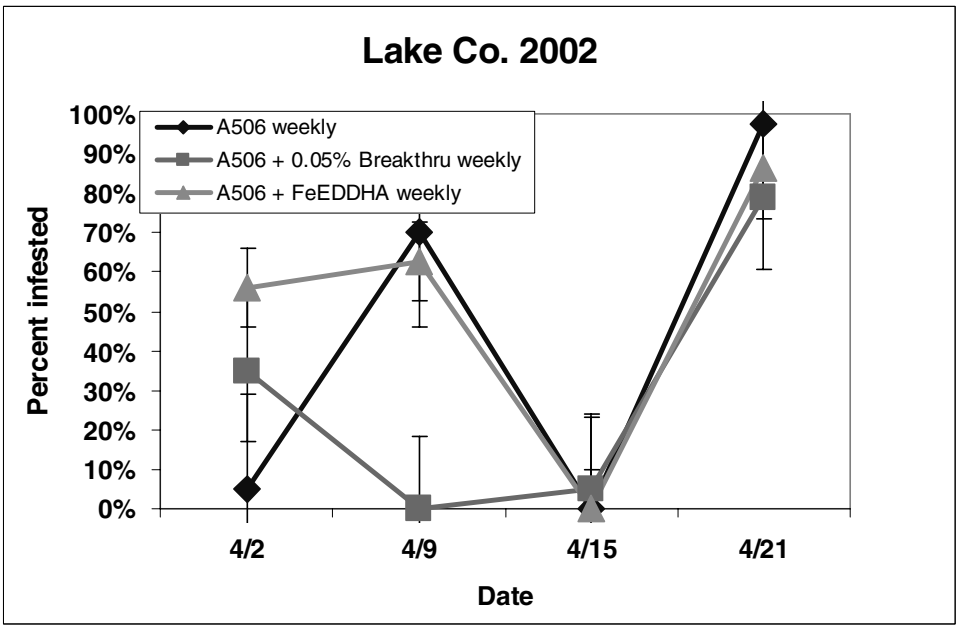


Figure 6. Percent of flowers colonzed with *Pseudomonas fluorescens* strain A506 on Bartlett pear trees treated weekly starting at first bloom with Blightban A506 in water to trees receiving no previous treatment (x's) or to trees treated at first bloom with Kocide 101 in 0.5% Breakthru (triangles) or to trees treated at first bloom with 0.5% sodium hypochlorite in 0.5% Breakthru (x's). The vertical bars represent the standard error of the mean percent colonized flowers.

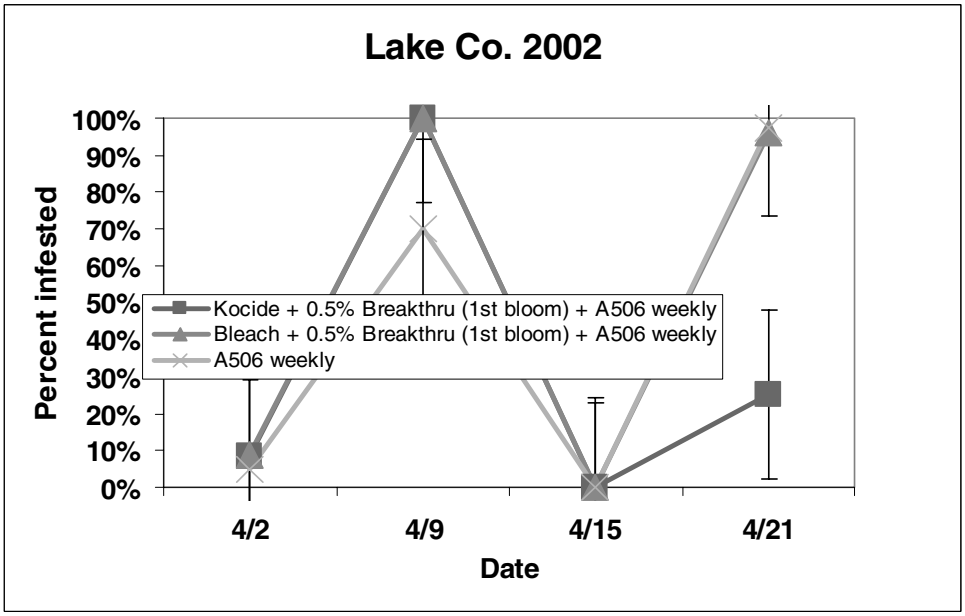


Figure 7. Percentage of flowers on trees treated with Blightban A506 and 0.5% Breakthru that were colonized with Pseudomonas fluorescens strain A506 when sprayed at first bloom (diamonds), 20% bloom (squares), 80% bloom (triangles), or full bloom (x's). The vertical bars represent the standard error of the mean percentage of flowers colonized.

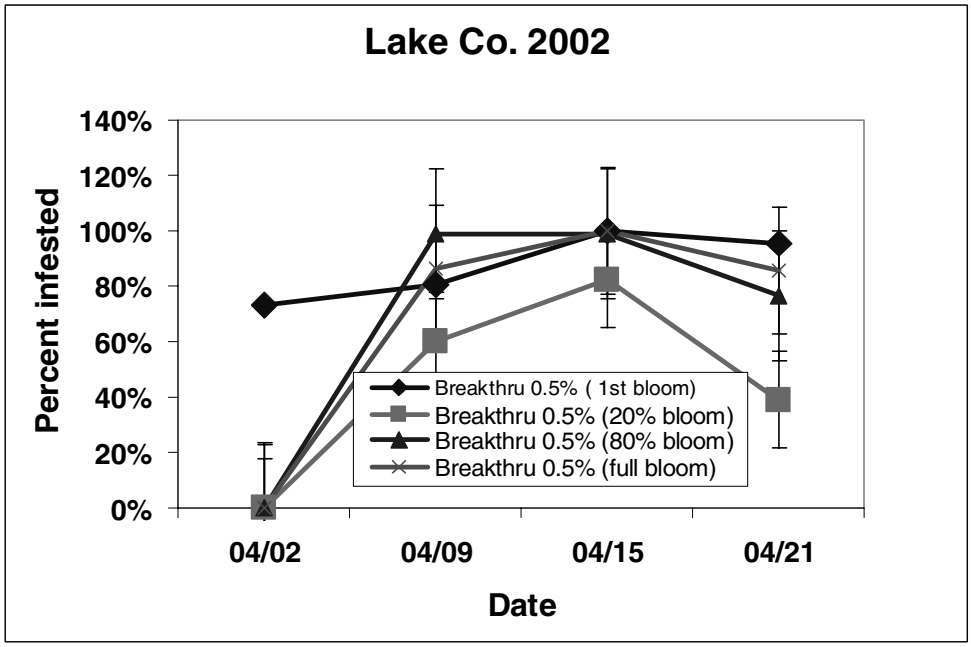


Figure 8. The percent of flowers of Bartlett pear from a commercial orchard in Upper Lake that harbored *Pseudomonas fluorescens* strain A506 as measured with a "flower rub" assay at different times during the growing season. Trees received either a normal antibiotic spray program selected by the cooperating grower (diamonds), were treated weekly with Blightban A506 in water alone (triangles), were treated a single time at first bloom with Blightban A506 in 0.5% Breakthru (x's), or were treated weekly with a mixture of Blightban A506 and 1 lb/100 gal Sequestrene 138 in water (squares).

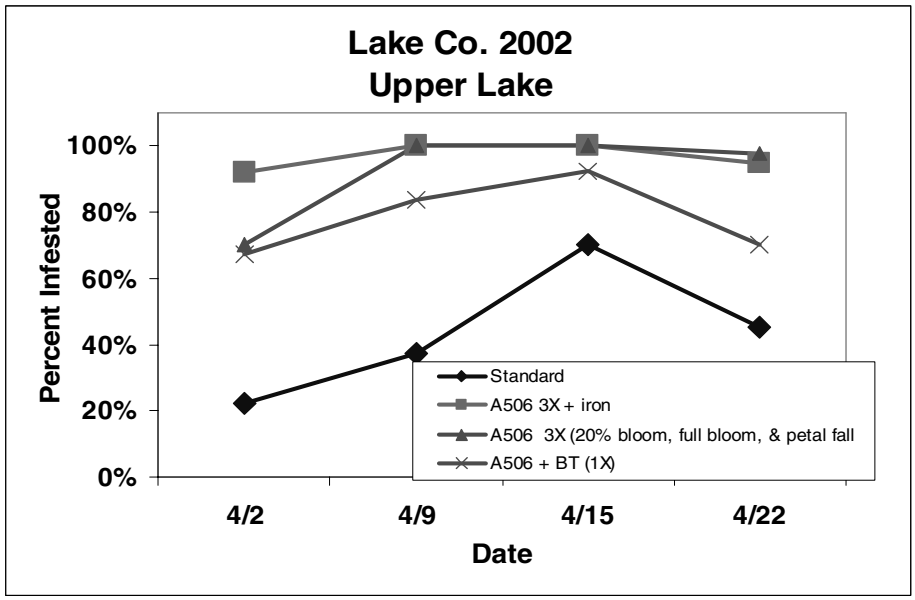


Figure 9. The percent of flowers of Bartlett pear from a commercial orchard in Sacramento County that harbored *Pseudomonas fluorescens* strain A506 as measured with a "flower rub" assay at different times during the growing season. Trees received either a normal antibiotic spray program selected by the cooperating grower (diamonds), were treated weekly with Blightban A506 in water alone (squares), were treated a single time at first bloom with Blightban A506 in 0.5% Breakthru (x's), or were treated weekly with a mixture of Blightban A506 and 1 lb/100 gal Sequestrene 138 in water (triangles).

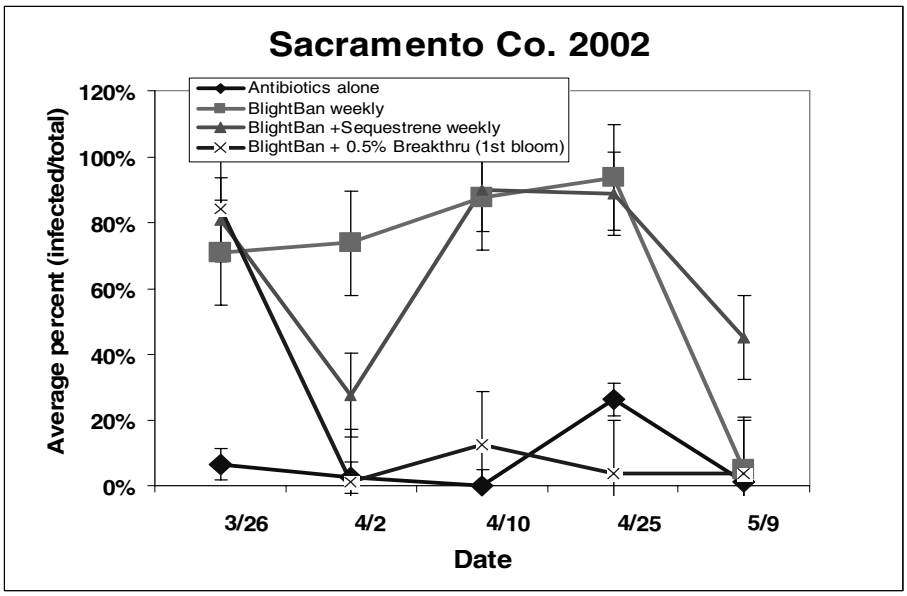
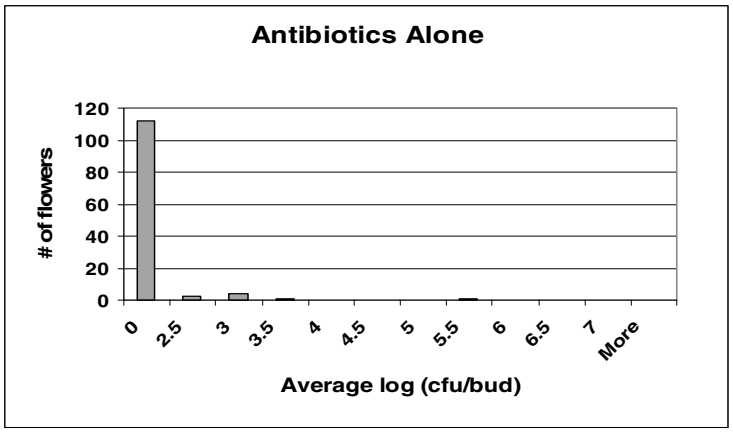
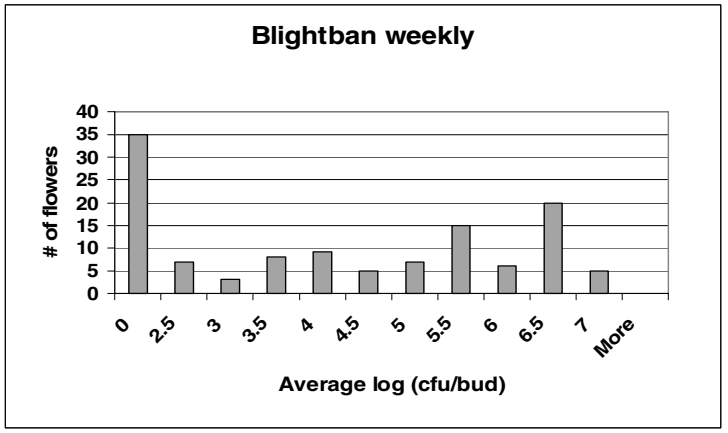
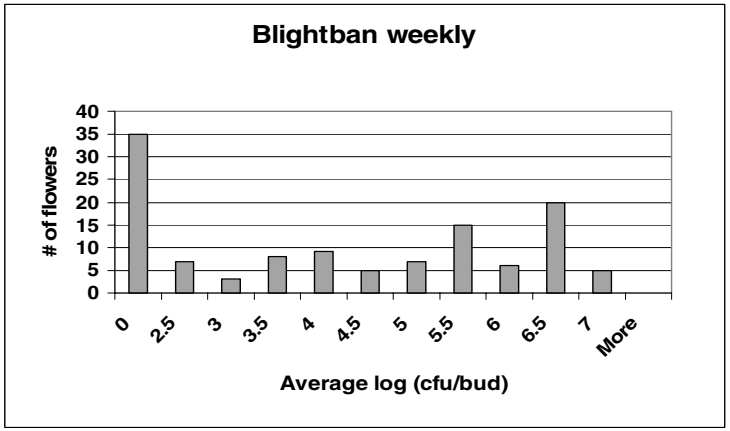


Figure 10. The distribution of the numbers of cells of *Pseudomonas fluorescens* strain A506 recovered from flowers of Bartlett pear from a commercial orchard in Sacramento County from trees that had received either a normal antibiotic spray program selected by the cooperating grower (upper left) or were treated weekly with Blightban A506 in water alone (upper right), were treated a single time at first bloom with Blightban A506 in 0.5% Breakthru (lower left), or were treated weekly with a mixture of Blightban A506 and 1 lb/100 gal Sequestrene 138 in water (lower right).







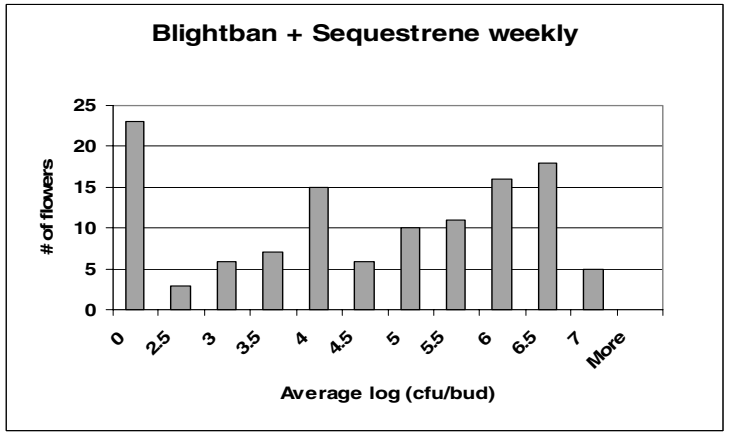


Figure 11. Relationship between the numbers of cells of *Pseudomonas fluorescens* strain A506 recovered from flowers of Bartlett pear from a commercial orchard in Sacramento County from trees that had received either a normal antibiotic spray program selected by the cooperating grower, were treated weekly with Blightban A506 in water alone, were treated a single time at first bloom with Blightban A506 in 0.5% Breakthru, or were treated weekly with a mixture of Blightban A506 and 1 lb/100 gal Sequestrene 138 in water, and the percentage of flowers sampled at this same time that harbored *Pseudomonas fluorescens* strain A506 as measured with a "flower rub" assay.

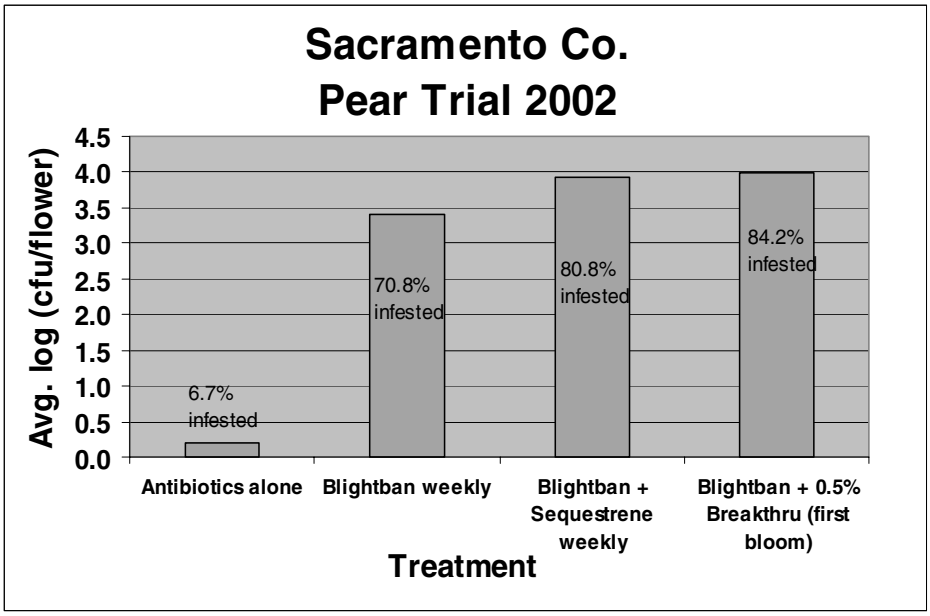


Figure 12. Relationship between the numbers of cells of *Pseudomonas fluorescens* strain A506 recovered from flowers of Bartlett pear from a commercial orchard in Lake County that had received various Blightban treatments and the percentage of flowers sampled at this same time from each treatment that harbored *Pseudomonas fluorescens* strain A506 as measured with a "flower rub" assay

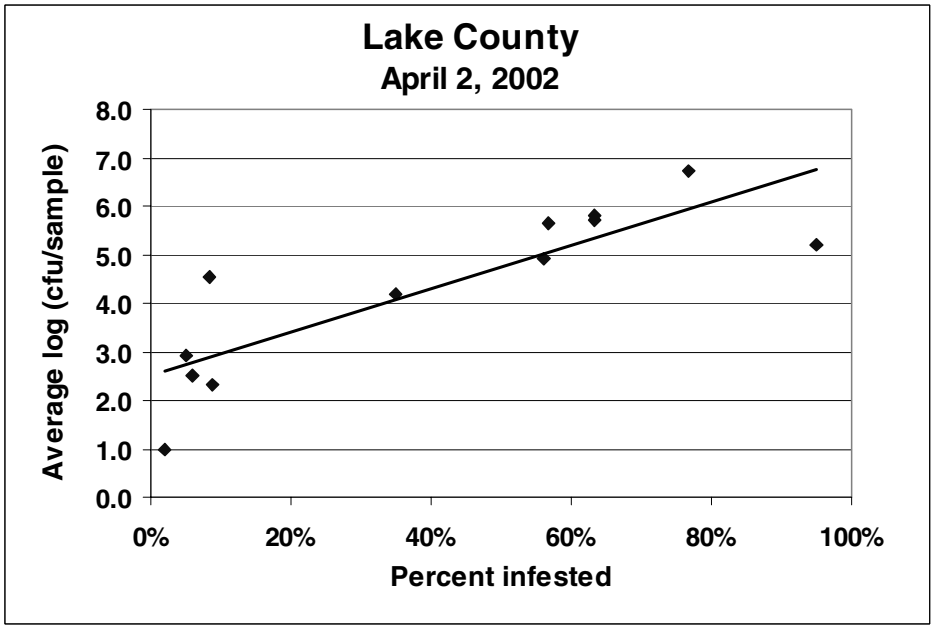


Figure13. The percent of flowers of Pink Lady Apple from a commercial orchard near Chowchilla that harbored *Pseudomonas fluorescens* strain A506 as measured with a "flower rub" assay at different times during the growing season. Trees were either untreated (circles), were treated weekly with Blightban A506 in water alone (x's), were treated a single time at first bloom with Blightban A506 in 0.5% Breakthru triangles), were treated a single time at first bloom with Blightban A506 in 0.2% Breakthru (squares), were treated a single time at first bloom with Blightban A506 without surfactant (diamonds), or were treated weekly with a mixture of Blightban A506 and 1 lb/100 gal Sequestrene 138 in water (stars).

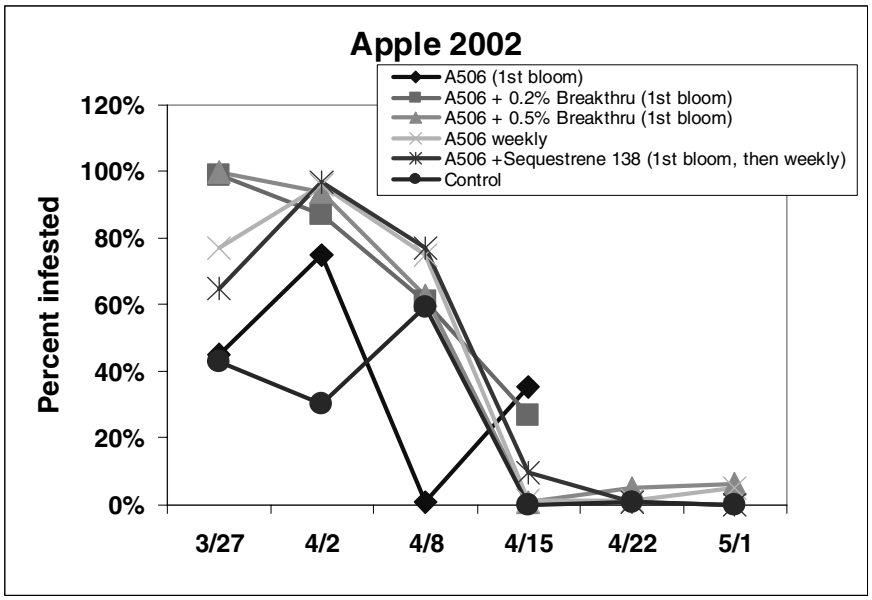


Figure 14. The distribution of the numbers of cells of *Pseudomonas fluorescens* strain A506 recovered from flowers of Pink Lady Apple from a commercial orchard near Chowchilla from trees receiving different Blightban A506 treatments.