

Project title: The effects of planting date, varietal susceptibility and residue management on severity of Fusarium wilt in lettuce

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Abstract

Fusarium wilt of lettuce, caused by *Fusarium oxysporum* f. sp. *lactucae*, is a serious disease affecting lettuce production in most major growing districts in California and Arizona. The disease is most problematic in warmer areas, which includes the San Joaquin Valley but also coastal locations in the vicinity of King City and points south. Our previous work has shown that although crisphead (= iceberg) cultivars differ in susceptibility to Fusarium wilt, all of them will succumb if disease pressure is high enough. Results obtained in 2013 showed some of the tested cultivars to be more resistant than observed in previous rounds of testing. To confirm that this was not due to reduced virulence of the isolate used in screening, additional tests were conducted using an isolate of the pathogen recovered from a severely diseased lettuce plant in 2014. The results were very similar to those obtained in 2013. Thus, it appears that higher levels of resistance may now be available in some iceberg varieties. Tests under field conditions will be conducted to further evaluate this possibility. If a variety is at least somewhat susceptible, the severity of Fusarium wilt will be determined in part by the temperatures that prevail during the growing season. Consequently, it may be possible to grow susceptible cultivars during the cooler part of the year. To test this possibility, we established a trial in a heavily infested field on the UC Davis campus. Seven cultivars that represented a range of susceptibilities to Fusarium wilt were planted on each of three different dates, separated by one week. Five weeks after planting most plants of the two most susceptible cultivars had died from the disease, whereas plants of the resistant leaf cultivar, Fossey, were either healthy or showed mild symptoms. There was no effect of planting date on these three cultivars. On the other hand, for two iceberg cultivars, disease was significantly more severe in the first planting than in the second planting. This differential effect was associated with higher temperatures during the second planting window. Inoculum levels are another important determinant of disease and consequently, minimizing the build-up of inoculum in soil is an essential component of disease management. During 2014 we completed an experiment designed to determine if drying crop residue on the soil surface prior to incorporation will influence pathogen survival. The results showed that after one year, there was not a significant difference in soil inoculum densities, between plots in which residue was incorporated immediately and those in which residue was dried before it was incorporated.

Objectives

1. Complete screening of leaf, romaine and crisphead cultivars for susceptibility to Fusarium wilt
2. Establish field trials to assess the effect of planting date and ambient temperatures on severity of Fusarium wilt
3. Determine the effect of drying crop residue prior to incorporation on the density of inoculum in soil

Procedures

Objective 1

Lettuce cultivars were tested for susceptibility to Fusarium wilt using a root-dip inoculation. Seedlings with one true leaf were up-rooted, the roots washed in water and dipped in a spore suspension of the pathogen. Inoculated plants were maintained in a greenhouse for 3-4 weeks, after which they were rated on a 1-4 scale: 1 = no symptoms, 2 = stunted and/or mild foliar symptoms, 3 = severely stunted and 4 = dead. Cultivars tested in 2014 were a subset of those tested in 2013. Seeds of each cultivar were kindly provided by Gowan seed.

Objective 2

Seven cultivars were planted on each of three dates (21 August, 28 August and 4 September) in a naturally infested field on the UC Davis Plant Pathology Department research farm. This included 'Fossey', a leaf type known to be resistant to Fusarium wilt, one romaine cultivar, 'Bondi', and five crisphead cultivars: 'Diamondback', 'Gabilan', 'Salute', 'Sidewinder' and 'Steamboat'. Entries were selected based on performance in past trials and suggestions from industry representatives. The field was divided into six equally sized blocks, and all planting dates of all cultivars were included in each block to minimize effects of heterogeneity of inoculum levels within the field. Accordingly there were six replications of each cultivar at each planting date. A replication consisted of ten plants that were transplanted into a bed with one foot spacing between plants. A stand count was taken one week after planting, and ratings for disease symptoms were conducted at three, four and five weeks after planting.

Objective 3

Infected lettuce plants were obtained from an experimental field on the Davis campus that is infested with the Fusarium wilt pathogen. Plants harvested from this field were transferred to microplots (2' x 2') and either incorporated immediately or spread over the soil surface and allowed to dry for eight days prior to incorporation. Each treatment (incorporated immediately or not) was represented by five plots. Thirteen days after incorporation of the dried residue, soil in all plots was mixed using a hand-held cultivator. At one, two, three, six, nine and 12 months after incorporation of the dried residue, soil cores were removed from each plot and assayed to

determine the state of decomposition of the residue and the density of inoculum. Inoculum density was estimated using soil dilution plating. A composite of multiple soil cores was taken from each replication and a sub-sample was suspended in water. Various dilutions of this suspension were spread over the surface of plates containing a *Fusarium*-selective medium. Colonies corresponding to the *Fusarium* wilt pathogen were enumerated seven days later. The experiment was conducted twice, with the first being initiated in August of 2013 and the second in October of 2013.

Results and Discussion

In 2013, 50 cultivars were tested using a root-dip inoculation procedure. Overall, disease severity ratings were somewhat lower than expected. Plants were inoculated with a known pathogenic isolate of *F. oxysporum* f. sp. *lactucae* that was maintained at low temperature since it was collected in 2002. To determine if lower disease severity in the 2013 trial was due to diminished virulence of the test isolate, we retested a subset of the cultivars using a new isolate of the pathogen. This isolate was recovered from a severely diseased lettuce plant in one of our field trials during 2014. Disease severity ratings in the 2014 inoculations were, on average, somewhat higher than what was obtained in 2013 (Table 1). However, based on a paired t-test, there was no significant difference in disease severity induced by the two isolates ($P = 0.412$). This suggests that cultivars with low ratings may have a level of resistance that would be useful under field conditions. Of particular interest is PRO 1583, which had average ratings of 1.60 and 1.10 in 2013 and 2014, respectively. Of the ten plants inoculated in the 2014 trial, eight were symptomless and two showed only mild stunting (rating = 1.5). Based on these results, PRO 1583 will be evaluated in a naturally infested field in 2015 to determine if it manifests resistance when subjected to high disease pressure.

Table 1. Cultivars tested for susceptibility to Fusarium wilt using a root-dip inoculation procedure.

Cultivar	Type	Rating	
		2013	2014
7101 A	Head	1.85	2.55
Black Belt	Head	2.15	1.50
Cheetah	Head	1.70	1.55
Corona	Head	2.20	2.95
EXP 1221	Head	1.85	2.30
Gabilan	Head	1.60	2.80
Keeper	Head	2.00	1.80
Navajo	Head	1.55	3.10
PRO 1555	Head	2.90	2.25
PRO 1565	Head	2.70	3.10
PRO 1569	Head	2.00	1.30
PRO 1583	Head	1.60	1.10
PRO 1587	Head	2.45	1.60
PRO 1617	Head	2.10	2.45
PRO 1618	Head	1.60	2.50
PRO 1619	Head	1.80	1.55
Regency 2.0	Head	2.45	2.60
Average		2.03	2.18

In the planting date experiment, differences in disease severity between cultivars were evident by three weeks after planting (Figure 1). Bondi was the most susceptible, with average ratings of 2.71 ± 0.11 , 3.00 ± 0.12 and 2.72 ± 0.10 for the first, second and third planting dates, respectively. Fossey was the least affected, with only mild stunting evident in a few plants in each of the three plantings. The remaining five cultivars were intermediate. There was no obvious difference between planting dates except for Steamboat, which was more severely affected in the third planting than in the other two (Figure 1). At four weeks after planting, disease was more severe for most cultivars across planting dates, with the exception of Fossey, which continued to show no more than mild stunting, and this in only a few plants. Bondi remained the most severely affected, with average severity ratings of 3.62 ± 0.10 , 3.52 ± 0.09 and 3.68 ± 0.05 for the first, second and third planting dates, respectively. Significant differences in disease severity between planting dates were evident for Gabilan, Salute and Sidewinder. In all cases, disease was most severe in the third planting (Figure 2).

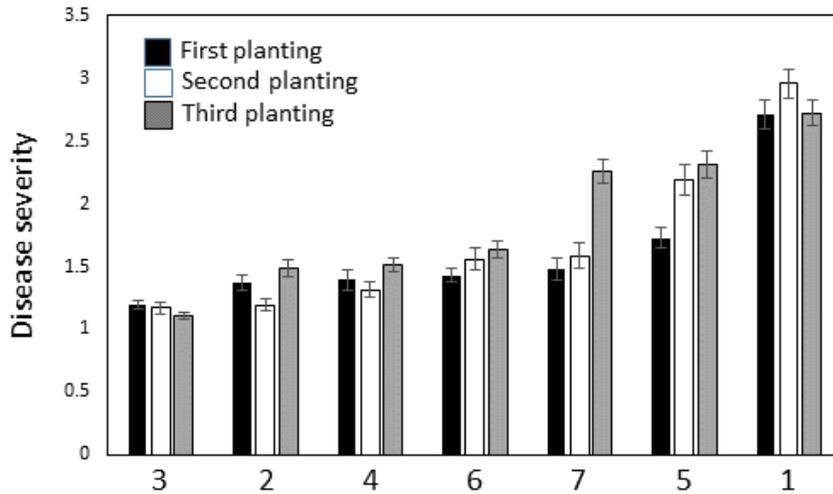


Figure 1. Disease severity in seven lettuce cultivars at three weeks after planting on either 21 August (first planting), 28 August (second planting) or 4 September (third planting). Numbers on the X axis correspond to cultivars as follows: 1 = Bondi, 2 = Diamondback, 3 = Fossey, 4 = Gabilan, 5 = Salute, 6 = Sidewinder and 7 = Steamboat. Error bars correspond to the standard error of the mean.

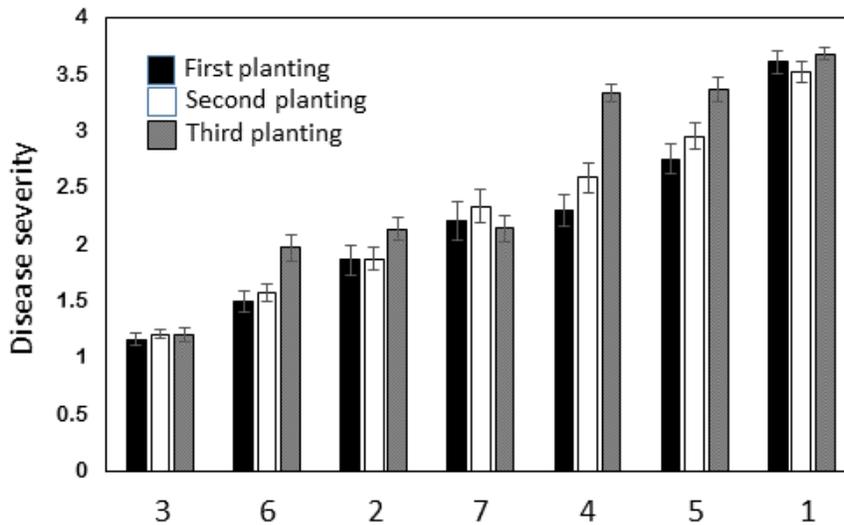


Figure 2. Disease severity in seven lettuce cultivars at four weeks after planting on either 21 August (first planting), 28 August (second planting) or 4 September (third planting). Numbers on the X axis correspond to cultivars as follows: 1 = Bondi, 2 = Diamondback, 3 = Fossey, 4 = Gabilan, 5 = Salute, 6 = Sidewinder and 7 = Steamboat. Error bars correspond to the standard error of the mean.

By week five, mortality was extensive in Bondi, Gabilan and Salute. Fossey remained the least affected, although some plants showed definite stunting, and average severity was as high as 1.78 ± 0.15 (second planting date). Planting date effects were clearly evident for Diamondback and

Steamboat, with disease being significantly more severe in the first planting than either of the other two (Figure 3).

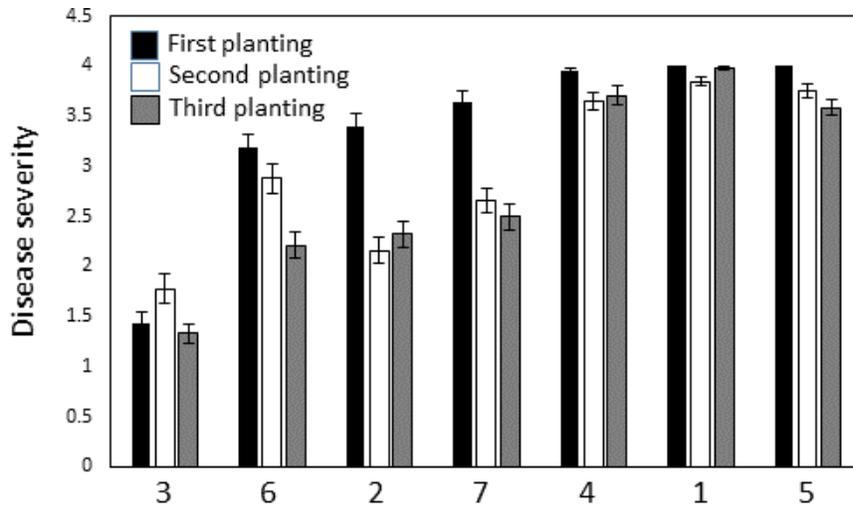


Figure 3. Disease severity in seven lettuce cultivars at five weeks after planting on either 21 August (first planting), 28 August (second planting) or 4 September (third planting). Numbers on the X axis correspond to cultivars as follows: 1 = Bondi, 2 = Diamondback, 3 = Fossey, 4 = Gabilan, 5 = Salute, 6 = Sidewinder and 7 = Steamboat. Error bars correspond to the standard error of the mean.

It is noteworthy that the planting date effect at five weeks was the reverse of what was observed at three and four weeks. This is most likely due to the pattern of temperature differences between planting dates over the course of the experiment. Fusarium wilt tends to be more severe under warm conditions and during the first week after planting, ambient temperature was higher in the second than in the first planting, whereas beyond three weeks, this pattern was reversed (Figure 4). Thus, our findings indicate that temperature differences of the magnitude that occurred in 2014 are sufficient to influence development of Fusarium wilt in susceptible cultivars.

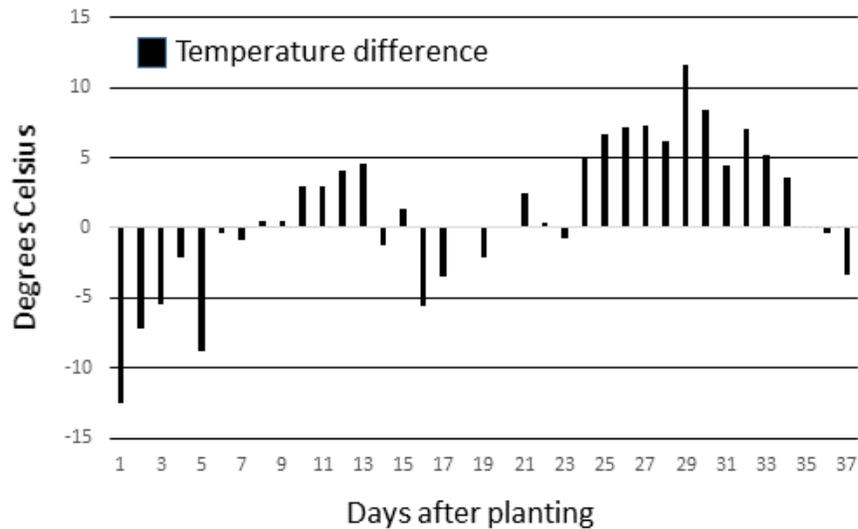


Figure 4. The difference in the daily high temperature between the first and second planting dates.

The observed differences in disease severity most likely understate the potential importance of planting date. That is because of the very high inoculum levels present in our experimental plot. Whereas *Fusarium wilt* can be damaging in commercial fields with as little as 100 propagules per gram of soil, the field in which our experiment was conducted has > 500 propagules per gram of soil throughout the plot. The beneficial effect of lower temperature may be greater where inoculum levels are closer to what is commonly recorded for commercial fields.

Residue treatment experiments were conducted in soil that was initially free of the pathogen. In the first experiment, inoculum levels at one month after incorporation were 34% lower, on average, in plots in which residue was allowed to dry on the soil surface prior to incorporation. However, by the last sampling date, in August of 2014, there was very little difference between the two treatments (Figure 5).

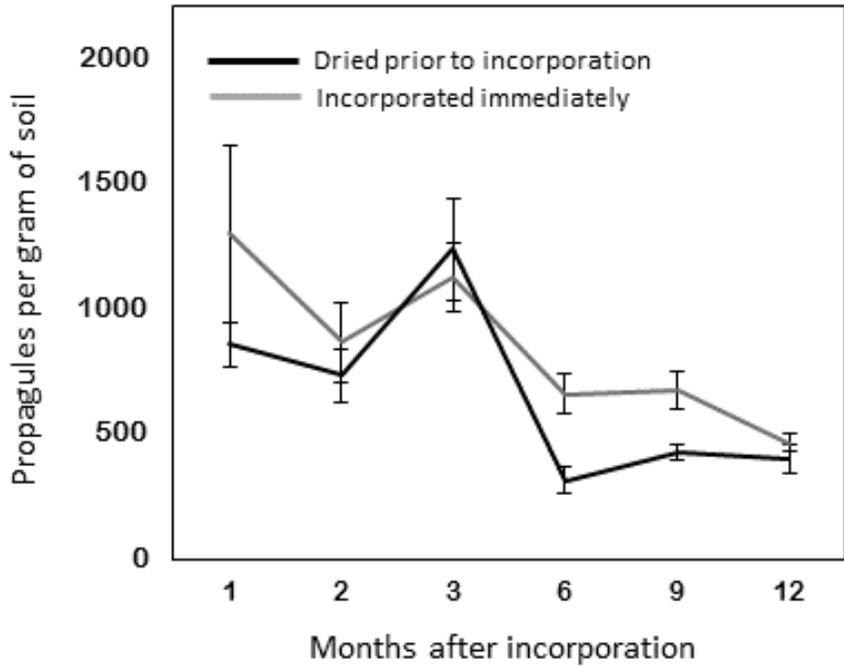


Figure 5. The density of inoculum of *Fusarium oxysporum* f. sp. *lactucae* in soil at intervals after incorporation of infested crop residue. Residue was either incorporated immediately or after drying on the soil surface.

The experiment described above was repeated, with an initiation date in October of 2013. The results were overall somewhat more variable than in the first experiment but there was likewise not a significant difference in final inoculum levels between the two treatments.