A STUDY OF METHODS FOR REDUCING BACTERIAL SOFT ROT IN WOUNDED FRESH MARKET TOMATOES

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Abstract. Post-inoculation treatments, including cupric ions, 500 ppm chlorine, and water heated to 140 F, had little effect on the development of soft rot lesions in wounded tomato fruit. Three pre-inoculation treatments, reducing initial inoculum, use of green rather than pink or ripe fruit, and use of decay tolerant cultivars, all significantly reduced soft rot development.

A need for reducing the potential amount of soft rot in mechanically harvested fresh market tomatoes has been partially answered by changes in the harvestor which have reduced the amount of fruit damage which occurs during harvest (4). However, even if the amount of damage in machine harvested fruit were reduced to that found in hand-picked fruit, soft rot could occassionally prevent successful marketing of machine-harvested tomatoes. A recent terminal market comparison of hand-harvested versus machine-harvested tomatoes revealed that 9-12% of the various fruit sizes of even the hand-harvested fruit were classified as "cut fruit" (6). Nineteen to 36% of the fruit harvested with a modified Hart-Carter machine were graded "cut fruit." Since rotted fruit pass through the machine, contamination of the wounds on tomatoes would be highly probable. If in the above study soft-rotted fruit were present on the plants before harvest, 19-36% of the various sizes of mechanically harvested fruit would very likely decay.

Early studies on bacterial soft rot of tomatoes established that wounds, growth cracks or stem scars were the only entry points for soft rotting bacteria (8, 13). Treatment of wash water in the harvest system through use of sodium hypochlorite has reduced bacterial soft rot (11). Eckert and Sommer (5), however, questioned the efficacy of chlorine compounds when soft rot bacteria were already present in wounds.

Immersion of tomato fruit in water heated to 140 F (60C) controlled buckeye rot caused by *Phytophthora parasitica* Dast. even after the organism had penetrated into the fruit (10). Soft rot of bell pepper fruit developing from inoculated stems was reduced by immersion in hot water baths (128 F for 1.5 min, 123 F for 2.5 min, or 133 F for 0.75 min) (7). Since *Erwinia carotovora* (L. R. Jones) Holland has a thermal death point at 51 C (124 F) (8), hot water treatment might successfully reduce the amount of decay developing from contaminated wounds in tomatoes.

Strangely enough pink and ripe fruit were reported to be more tolerant to soft rot than were green fruit (8, 9, 12, 13). Harvesting primarily pink or ripe fruit, therefore, might be another method for reducing the soft rot potential.

Varietal tolerance of tomato fruit to soft rot was reported by Bartz and Crill (1). Even though most fruit ultimately rotted, many were fully ripe for at least four days before rotting and probably would have been consumed before lesions appeared. Use of only those tomato lines reported to be highly tolerant to soft rot could be suggested as another method of reducing the soft rot potential in mechanical harvest systems.

The following is a report on the efficacy of several different methods of reducing the bacterial soft rot developing from contaminated wounds in fresh market tomato fruit.

Materials and Methods

Erwinia carotovora was grown in nutrient broth for ca. 24 hr before being centrifuged, resuspended in $\frac{1}{2}$ strength buffered saline (pH 7.0), and adjusted to designated populations as was described previously (1). The inoculation technique utilizing needles was also done as described previously except for the changes noted in certain tests.

In the chlorine (Cl) dip tests, either chlorine bleach (sodium hypochlorite) or calcium hypochlorite solutions served as sources of the hypochlorite ion. All solutions were adjusted to appropriate chlorine contents as determined by a Taylor Chemicals Inc. swimming pool tester. In most tests, pH of the chlorine solutions was not

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controlled, but the pH was buffered to 4 and 10 by a phosphate buffer in one series of tests.

Hot water treatment of wounded tomatoes was carried out in a commercial-sized formula warmer. Temperatures were monitored before, during and after the immersion of 10 tomatoes for 1 min. The volume of water was sufficient to prevent depression in bath temperature during the treatments.

Green, pink and ripe fruit were selected on the basis of color development at the time of inoculation. Fruit were not considered ripe unless color development was almost complete. Breaking fruit were placed in the pink fruit group.

Five cultivars, 'Walter,' 'Floradel,' 'Tropic,' 'Florida MH-1,' 'Homestead-24,' and one breeding line suitable for mechanical harvest, (MH-9), were tested for innate tolerance to soft rot. These lines were all planted in adjacent rows as staked tomatoes in one plot at AREC-Bradenton during the Fall season of 1972.

In all tests, except where noted, fruit were stored at 70 F and ca. 90% relative humidity after inoculation. Fruit were removed when lesions first appeared in order to minimize secondary spread of the rot-inducing pathogens. Ten fruit comprised a replicate and data were expressed in terms of the average percentage of fruit having lesions at a given period after inoculation. Differences among treatment means were detected using the Duncan's New Multiple Range test. All percentage values were transformed by the arc sin square root of percentage transformation. One hundred percent values were treated as 100-¼n where n equalled the number of observations while ¼n equalled 0%.

Results

Wound depth and post-inoculation dip.—Different wound depths (1, 2 and 4 mm) were achieved by adjusting the length of straight pins protruding through the cork body of the standard wounding instrument. Inoculum concentration was standardized to 10^6 *E. carotovora* cells/ml of buffered saline. Within 1 hr after wounding the tomato fruit with needles dipped into the bacterial suspension, 10 fruit replicates were immersed for 1 min in either hot tap water (128 F), room temperature water (75-80 F), 250 ppm w/w Cu++ or 500 ppm w/w Cl from NaClO₂. The values reported were the average percentage of fruit decayed by 14 days after inoculation (Table 1).

Chlorine treatment reduced the subsequent amount of decay irrespective of wound depth as compared to the controls. The reduction was sig<u>Table 1</u>. Percentage of 'Walter' tomato fruit developing bacterial soft rot after 14 days at 70F following an immersion in various solutions.

Solution	1 <u>/</u> 1	$\frac{\text{Wound}}{271}$	1 depth 4 /2
H ₂ O 500 ppm Cl 250 ppm Cu 128F	22 a 10 b 32 a 48 a	58 a 32 a	67 a 43 b 63 a 83 a
<u>/1</u> Each value <u>/2</u> Each value <u>/3</u> Values not were statis level.	= avg of = avg of followed stically o	four-10 : three-10 by the sa hifferent	fruit reps. fruit reps. ame letter at the 5%

nificant (.05 level) for the shallow and deep wound depth treatments, but not for the intermediate one. Hot water treatment resulted in an increased amount of decay, but the differences were not significant. Cupric (Cu) ion treatments did not reduce decay, but caused phytoxicity at the wound sites. Deep wounds resulted in more decay than did the shallow wounds; the differences, however, were not significant.

Other tomato fruit were inoculated (2 mm wound depth) as above in a similar test and evaluated for decay 7 and 14 days after inoculation (Table 2). The post-inoculation dips included 500 ppm Cl buffered to pH's 4 and 10 with 0.1 M sodium phosphate buffers and tap water at room temperature (75-80 F), 120, 130 and 140 F. At seven days after inoculation, only the pH 10 chlorine solutions significantly reduced the amount of decay. By 14 days after inoculation, only the two chlorine treatments had markedly reduced

Table 2. Percentage of 'Walter' tomato fruit developing bacterial soft rot after 7 and 14 days at 70F following a 1 min post-inoculation immersion in various solutions.

Solution	Days i	n Storage
	7	14
H ₂ 0	23 1 a	57 a
500 ppm C1		
pH 4	17 a	40 a
pH 10	0 ь	23 a
Heated H ₂ O		
120F 2	20 a	50 a
130F	20 a	80 a
140F	7 ab	67 a

 $\frac{1}{1}$ Avg of three-10 fruit reps.

12 Values not followed by the same letter were statistically significant at the 5% level. decay, whereas the hot water treatments resulted in an increased decay over the control treatment. None of the differences among the treatments was significant at 14 days after inoculation. In several other tests which are not reported here, immersion of wound inoculated fruit in Cl solutions did not reduce the amount of decay.

Concentration of initial inoculum versus rate of decay .- 'Floradel' mature green fruit were wound-inoculated using suspensions of E. carotovora at concentrations of 0, 10^2 , 10^4 , 10^6 and 10^8 bacteria/ml. At 3 days after inoculation, a reduction in the initial inoculum from 10⁸ to 10⁶ cells/ ml did reduce the amount of decay (Table 3); at 7 and 14 days, however, there was no marked reduction until the inoculum level was reduced to 10⁴ cells/ml. When the suspension was reduced to 10^2 cells/ml, there was no apparent increase in the rate of decay as compared to the control. Decay in the control was attributed to contamination by fungi and bacteria before and during storage, as many lesions developed at sites other than the inoculation sites.

Tolerance of fruit at different maturities.— Several experiments were designed to test the hypothesis that ripe and pink fruit were more tolerant to soft rot than were green fruit. Two of those comparisons were reported in Table 4. Green fruit were significantly (.01 level) more tolerant to soft rot than were pink or ripe fruit in both tests. In other comparisons which were not reported here, there was no test where green fruit decayed faster on the basis of appearance of lesions than did pink or ripe fruit. These reports contradict those reported earlier by other workers (7, 8, 11, 12).

One additional comparison of the tolerance to soft rot of ripe versus green tomato fruit conducted at both 70 F and 55 F storage temperatures also supported the above data (Table 5). There was no significant difference either at 8 or 12 days

<u>Table 3.</u> Percentage of stored 'Floradel' tomato fruit (70F) having lesions 3, 7 and 14 days after wound inoculations with different concentrations of <u>Erwinia carotovora</u>.

Conc	Days a	fter inco	culation
(cells/ml)	3	7	14
0	2 /1	4	<u> </u>
102	0	4	11
104	0	29	69
106 108	12	65	86
108	23	72	93

<u>1</u> Avg of three-15 fruit reps.

Table 4. Percentage of ripe, pink or green tomato fruit rotting after inoculation with <u>E</u>. carotovora during 7 days storage at 70F. $\frac{1}{2}$

	Test 1 <u>/</u> 2	Test 2 /3
Ripe	90 a	55 a
Pink	97 a	41 a
Green	23 b	27 Ъ

- $\frac{1}{1}$ Inoc with needles dipped in 10⁷ cells/ml.
- <u>/2</u> Avg of 'Homestead-24' and 'Walter' fruit
- combined; six-10 fruit reps.
- <u>1</u>³ Avg of four-10 fruit reps of 'Florida MH-1' fruit.

<u>14</u> Values not followed by the same letter were significantly different at the 99% level.

after inoculation between the two storage temperatures within the same maturity rating. However, ripe fruit were always more susceptible than green fruit at both storage temperatures and in some cases those differences were significant (.01 level).

Tolerance of different tomato cultivars to soft rot.-The test for tolerance to soft rot among tomato cultivars was similar to one reported earlier (1). Some additional common cultivars were tested. along with one breeding line potentially suitable for mechanical harvest (MH-9). 'Tropic' and 'Floradel,' two indeterminate cultivars, were highly suspectible to bacterial soft rot at all observation dates (Table 6). At 3 or 7 days after inoculation, the difference between 'Walter' and either 'Floradel' or 'Tropic' was significant (.05 level), indicating that a greater degree of susceptibility existed in common tomato cultivars than previously had been suggested in studies of the tolerance of 'Walter,' 'Homestead-24' and 'Florida MH-1' (1). Again, in this study, 'Florida MH-1' and 'Homestead-24' were the most rot-tolerant tomato lines.

Table 5. Percentage of ripe and green 'Florida $\overline{\text{MH-1}}$ ' tomatoes rotting after inoculation with E. carotovora during 8 and 12 days storage at 55 and 70F.

	Days aft	ter inoculation
	8	12
Ripe		
70F	25 a	45 a
55F	13 a	38 a 4
Green		
70F	ОЪ	8 Ъ
55F	5 b	5 b `
		i i i i i i i i i i i i i i i i i i i

 $\frac{1}{1}$ Inoc with needles dipped into 10⁶ cells/ml.

 $\frac{1}{2}$ Average of four-10 fruit reps. 73 Values not followed by the same

³ Values not followed by the same letter were significantly different at the .01 level.

-15.

<u>Table 6.</u> Percentage of fruit of 5 cultivars and 1 breeding line of tomatoes rotting 3, 7 and 15 days after inoculation with 10^7 E. <u>carotovora</u> cells/ml.

	Days after inoculation		
	3	7	15
Tropic	2 <u>1 /</u> 1 a <u>/</u> 2	47 a	89 a
Floradel	14 ab	36 ab	91 a
Walter	8 bc	27 с	89 a
Florida MH-1	2 c	9 d	68 c
Homes tead-24	1 c	3 d	84 ab
MH-9	16 ab	29 bc	79 bc

<u>1</u> Average of ten-10 fruit reps.

 $\frac{1}{2}$ Differences among values not followed by the same letter are statistically different.

Discussion

The problem of preventing soft rot bacteria present in wounds from initiating lesions must be answered before mechanical harvest systems can operate successfully in fields with high populations of soft rotting bacteria. Fields with such high inoculum levels have often been noted in Florida during periods of wet, warm weather.

None of the post-inoculation treatments of tomato fruit tested by us has consistently reduced soft rot developing from contaminated wounds. Three pre-inoculation procedures, however, did successfully reduce the rate of appearance of lesions. Lower initial inoculum did, as expected, result in the most consistent reduction in the amount of decay. Inoculum reduction can be achieved through proper sanitation. Bleach has been used in field washers to achieve inoculum reduction (11), but hypochlorite solutions did not consistently reduce decay if applied to fruit after wounds had been contaminated. Thus, if chlorine compounds were used in mechanical harvest systems to achieve sanitation, all wound situations such as fruit separation areas should be treated in order to reduce the number of contaminated wounds.

Reductions in the amount of decay at a given inoculum level can be achieved through use of decay tolerant cultivars. 'Florida MH-1' and 'Homestead-24,' previously reported to be highly tolerant to bacterial soft rot (1), were the most tolerant lines in this study. Again, they were significantly more tolerant than 'Walter.' However, 'Walter' appeared to possess some tolerance to soft rot as compared to 'Tropic' and 'Floradel.' Burgis (3) has reported that 'Tropic' was more susceptible to

chilling damage than was 'Walter.' This suggests that decay tolerant tomato lines might be selected on the basis of tolerance to chilling. That is not always the case, however, as is indicated by the performance of 'Walter' and 'Homestead' in tests of tolerance to chilling (3) as compared to their performance in tests of tolerance to soft rot (1).

Several reports suggested that pink and ripe tomato fruit were more tolerant to soft rot than were green fruit (8, 9, 12, 13). Our results, however, were contrary to those earlier reports, which were based on either larger wounds or much heavier inoculum levels or increase in size of the decayed area. The wound inoculation technique and inoculum levels employed in this study were designed to approximate natural conditions. Thus, with small wounds, which would not be noted during packout procedures, and with realistic inoculum levels, ripe and pink tomato fruit were more susceptible than were green fruit. Since ripe fruit were also reported more susceptible to sour rot (2), the mechanical harvest of advanced maturity fruit would appear to invite more postharvest decay than would be encountered after mechanical harvest of primarily mature green fruit.

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