Virus-yellows Infected Sugarbeet Varieties: Effects of Harvest Dates and Nitrogen Fertilization

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Investigations in California have shown that the yellows disease of sugarbeet can cause up to 65% root yield loss, depending upon the yellows viruses involved, the virulence of the virus strains, time of infection and variety (1,2,3,6,9,10).² For a particular variety under similar virus conditions, root losses were consistent from test to test. However, the effect of yellows infection on sucrose percentage has remained inconclusive.

Yellows infection was also shown to influence purity. Bennett, Price, and McFarlane (3) found that under some conditions purity was slightly decreased with increases in sodium and potassium, but no change occurred for amino nitrogen. Goodban *et al.* (8) noted a slight decrease in processing quality and increased soluble nitrogen. Fife (7) demonstrated amino acid differences in infected and healthy plants.

Until recently, varieties with resistance to yellows were not available for comparison to determine the effect of yellows infection on root yield, sucrose percentage, or purity. From the breeding program at Salinas, California, several lines that show moderate resistance to yellows have been developed. Until lines with higher resistance are developed, these lines will be used as sources of resistance and as components in hybrid varieties. Currently, these lines with moderate resistance can provide the best control from losses by yellows. Because these lines will have to be grown under potential yellows infection, it is of interest to know how they perform in comparison to susceptible varieties.

The purposes of this experiment were to determine: (a) when sucrose losses occur in yellows infected lines; (b) how yellows affect a moderately resistant line in comparison with a more susceptible parental variety; (c) how yellows infection influences purity constituents; and (d) how the level of nitrogen fertility affects sucrose loss, resistance and purity factors for these populations. Nitrogen is known to influence sugar percentage, yield and quality characters of sugarbeet. Differences in nitrogen fertilization should help us to interpret the effects of yellows infection on sucrose loss and varietal resistance.

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² Numbers in parentheses refer to literature cited.

Methods and Materials

The primary data for this experiment were obtained at Salinas, California in 1967. Additional data were obtained from the 1967 yellows-resistance evaluation trial at Davis, California. The varieties used were open-pollinated US 75 (11) and moderately resistant selections 413 and 613. Line 413, a fifth successive yellowsresistance mass selection from US 75, was used in the Salinas test. Selection for resistance was based upon root size and freedom from yellowing in infected populations. Line 613, used in the Davis test, was selected two additional times for yellows resistance, but in several tests it appeared to be identical to 413. These selections will be called line 13.

The Salinas test was planted February 23, 1967. The Davis planting was made May 23 after aphid flights had ceased and the likelihood of natural infection was low. A combination of a virulent strain (strain 7) of beet yellows virus (BYV) and beet western yellows virus (BWYV) was used to inoculate the beet plants in approximately the 10-leaf stage, as previously described by Bennett *et al.* (3). The Salinas and Davis inoculations were made on May 2 and July 20, respectively. Entire plots were inoculated at Salinas because natural spread cannot be prevented (9). At Davis comparisons were made between inoculated and noninoculated subplots.

A split-block design with eight replications was used at Salinas. Each replication was divided into two nitrogen fertility treatments. Treatment 1 received 110 pounds of nitrogen, applied as ammonium sulfate in equal applications, at planting and on May 16. This treatment represented slightly less than our normal application to varietal trials for this planting date. Treatment 2 received the same applications as treatment 1 but in addition had 60 pounds applied to it on July 6. Treatment 1 was designed to become nitrogen deficient as the season progressed, whereas treatment 2 was designed to provide sufficient nitrogen throughout the growing season.

Within each split block at Salinas, the two varieties and eight harvest dates were completely randomized. At 2-week intervals, from July 25 to October 31, plots were harvested. Individual plots were single rows 32 ft long. On each side of the plots buffer rows were planted to prevent border effects due to differential dates of harvest. Each harvested plot was placed in two bags and cleaned weights were taken. The roots were rasped and the pulp frozen until four harvest dates could be run at the same time. Data were obtained on root yield, sucrose percentage,

amino nitrogen, sodium and potassium. Amino nitrogen (NH_2-N) , Na and K were determined from part of the lead-acetate filtrate used for the sucrose determination. Amino nitrogen (or noxious nitrogen) was determined in a spectrophotometer with Stanek-Pavlas copper reagent. A flame spectrophotometer was used for Na and K determinations.

To determine the nitrogen status, petioles were collected at each harvest date and petiole nitrates were determined by the method of Ulrich *et al.* (19).

The Salinas experiment was sprinkler-irrigated each week. Irrigation rates were governed to avoid as much movement of nitrogen between plots as possible.

The Davis data for US 75 and line 13 were derived from a variety trial, with blocks split for inoculated and noninoculated treatments. The trial was harvested on October 23. Data were obtained for yield, sucrose percentage, NH_2 -N, Na and K. Natural spread of yellows from inoculated to noninoculated plots was minimal.

Results and Discussion

The results of the Salinas and Davis tests are presented in Tables 1 to 4 and Figures 1 and 2. Table 1 gives the levels of significance obtained for varieties, dates of harvest and nitrogen treatments at Salinas, and varieties and infection treatments at Davis, for the characters studied. Figures 1 and 2 give the mean values within each date of harvest for the seven characters at Salinas. Table 2 gives the means within each variety, nitrogen, or infection treatment. Tables 3 and 4 give the means for the significant, first-order interactions.

Table 1.-Levels of significance obtained for main effects and interactions for varieties, dates of harvest, and nitrogen levels at Salinas and varieties and infections at Davis for seven characters.

Million Million	Root	0%	Gross	Petiole	1.11.5	Root	11.20
	yield	sucrose	sucrose	nitrate	NH2-N	Na	K
1. 1. 1. 1. 1.			Salinas	51.1		190 mar	100
Varieties	**						NS
Dates							••
Nitrogen		••	NS	••			NS
V×D	••	••				••	NS
VXN	NS	NS	NS	••			NS
DXN	NS		NS	••	NS	NS	NS
VXDXN	NS	NS	NS	••	NS	NS	NS
			Davis				
Varieties	••	NS	••				
Infection			••		(*)	NS	NS
VXI		•	••		(*)	NS	NS

(*) =Significance at the 10% level.

• = Significance at the 5% level.

•• = Significance at the 1% level.

Dates of Harvest

Duncan's multiple range test (0.05 level of probability) showed that the mean yield of beets over all varieties and treatments significantly increased every harvest from the first (9.1 T/A) to the sixth date (19.9 T/A) with the rate of increase being nearly linear (Figure 1). The sixth and seventh dates were not significantly different, but the eighth date showed a significant yield reduction from the seventh date.





Sucrose percentage also significantly increased for each date in a linear fashion through the fifth date from 12.2% to 14.6%. Between the fifth and sixth dates the decrease was significant to 13.6%. Increases for the remaining two harvests were slight. The highest sucrose percentage occurred in mid-September, with decreased percentages in late September and October.

The decrease between the fifth and sixth dates was probably not caused by increased NO₃ availability. Because sprinkler irrigation was used, a light rain (.18 inch) that occurred just before harvest could not have caused appreciable NO₃ leaching as suggested by Stout (16).

The growth curve for gross sucrose is the product of the curves for root weight and sucrose percentage. Therefore, the gross sucrose curve forms a more accurate indication of sucrose production than the curve of either of its components. Significant increases in gross sucrose occurred through the sixth date. However, after the sixth date, sucrose production leveled out. The eighth date showed a slight decrease from the seventh. The sixth, seventh, and eighth dates were not significantly different. Figure 1 shows that the mean gross sucrose curve does not have

the significant drop between the September 19 and October 3 dates as shown by the sucrose percentage curve. Instead, there appeared to be compensation by increased root yield.

It is of interest that reduced sucrose percentage and increased beet yield occurred during this part of the year, especially when one expects root growth to cease and sucrose percentage to increase. In the coastal valleys of California, summer temperatures may be lower than the temperatures of early fall. At the time sucrose percentage decreased, there was a warmer period which may have stimulated increased plant growth.



Figure 2.—Mean values for each date of harvest over all treatments for petiole nitrate, amino nitrogen, sodium, and potassium at Salinas.

As shown by Figure 2, petiole-NO₃ concentration started at a relatively high level, rapidly decreased and remained low through the part of the season in which rapid growth occurred, and then rapidly increased again when growth essentially ceased. The values for the third, fourth and fifth dates were significantly different from the values for the other five dates. This pattern was probably produced by the relatively high availability of NO₃ at the beginning of the experiment followed by decreasing availability and continued rapid growth and use of NO₃ through mid-September. However, during the last 6 weeks, growth and use of nitrogen were apparently reduced and NO₅ increased in the petioles. Other studies have not shown this increase in nitrate concentration during the fall months (4,13,17).

The concentration of root NH_2 -N did not follow a curve similar to petiole-NO₃ over the 4 months that plots were harvested, but remained nearly constant at 680 to 730 ppm except for a significantly higher level at the second date and significantly lower levels at the first and third dates.

Sodium generally showed a decreasing concentration in the roots while rapid growth was occurring, but the concentration increased in the fall as growth slowed and stopped. Na concentration significantly decreased from 270 ppm for the first harvest to 180 ppm for the fourth. After the fifth harvest Na increased significantly for each harvest and by the eighth date was twice as high as the first and nearly three times as high as the fourth.

There was a significant decrease in K concentration from the first date to the eighth date of harvest. Except for a slight increase for the fifth harvest, concentrations decreased to the sixth harvest. The seventh and eighth dates showed slight increases that corresponded with the period of decreased growth rates for beet roots.

Variety and Infection Effects and Variety \times Infection Interactions

The US 75 variety and resistant selection 13 were significantly different for all characters except K at Salinas and sucrose percentage at Davis (Table 2). At Salinas, under uniform yellows infection, 13 was about a percentage point higher in sucrose and had 56% greater root weight to yield 68% more gross sucrose than US 75.

The surgery of the second s	Root	Schert (C)	Gross	Petiole	Root		
	yield T/A	Sucrose %	sucrose lbs/A	nitrate ppm	NH2-N ppm	Na ppm	K ppm
Land St. St.	N. Ares	Sa	linas	Line an		1.67	Charles to
Varieties							
US 75	12.6**1	13.1**	3340**	905**	815**	342**	2654
Line 13	19.7	14.1	5620	499	601	189	2784
Nitrogen							
110	15.9**	13.9**	4480	510**	639**	235**	2675
170	16.4	13.4	4480	894	778	297	2763
		I	Davis				
Varieties							
US 75	17.3**	12.4	4350**	A State	978**	786**	3193**
Line 13	19.8	12.7	5050	Same of	655	432	3495
Infection ·							
Check	22.3**	13.0**	5780**		762(*)	566	3341
BYV-BWYV	14.9	12.1	3620	12 Tr	871	652	3347

Table 2.-Mean values within variety or nitrogen treatments at Salinas and variety or infection treatments at Davis.

¹Indicates level of significance for each pair of means where (*), *, and ** are at the 10%, 5%, and 1% levels, respectively.

Yellows infection at Davis significantly reduced yield, sucrose percentage and gross sucrose as compared to noninoculated checks (Table 2). Significant variety \times infection interactions occurred for yield, sucrose percentage and gross sucrose (Tables 1,3), whereas NH₂-N interacted significantly only at the 10% level and Na and K showed nonsignificant differences.

It appeared that the variety difference in sucrose yield was primarily due to differences in resistance to yellows. At Davis US 75 and 13 were not significantly different for root yield or sucrose percentage when grown under noninoculated conditions (Table 3); but under inoculated conditions selection 13 was 0.8 percentage point higher in sucrose and had 38% greater root weight to yield 48% more gross sucrose than US 75. These data suggest that in selecting line 13 from US 75 for resistance to yellows, the production factors for gross sucrose were not appreciably changed under virus-free conditions, but that the increased performance under yellows is due to resistance factors that allow existing yield genes to function more efficiently.

The mean petiole-NO₃ concentration of line 13 was significantly lower than for US 75 at Salinas. Petioles were not sampled at Davis, and the influence of yellows was, therefore, not determined.

	Root		Gross	Root		Petiole	Root			
Variety	yield T/A	Sucrose %	sucrose lbs/A	NH2-N ppm	Na ppm	nitrate ppm	NH2-N ppm	Na ppm		
	BY	BYV-BWYV Noninoculated (Davis)					110 lbs N (Salinas)			
US 75	22.1	13.1	5780	870	700	614	716	304		
Line 13	22.4	12.9	5780	654	432	406	562	166		
	В	YV-BWYV	Inoculated	(Davis)		170 11	os N (Sali	inas)		
US 75	12.5	11.7	2920	1086	871	1196	915	381		
Line 13	17.3	12.5	4320	656	433	592	641	213		
LSD (.05)	2.1	.7	550	164	202	161	32	26		

Table 3.—Mean values within nitrogen or infection treatments for varieties at Salinas or Davis, respectively, for those characters that showed significant variety x infection or variety x nitrogen interactions.

At Salinas, 13 showed a significantly lower NH_2 -N concentration than US 75 (Table 2). At Davis, line 13 had nearly identical NH_2 -N levels under yellows-inoculated and noninoculated conditions while US 75 showed significantly different levels under the two infection treatments (Table 3). Both US 75 treatments were significantly higher than 13's concentrations.

At Salinas and Davis, the 13 selection had a significantly lower Na concentration than US 75 (Table 2). As with NH_2-N at Davis, 13 had nearly identical Na levels under inoculated and

noninoculated conditions; but US 75 did not show a significant difference between infection treatments, even though the inoculated treatment showed a mean increase of 171 ppm (Table 3).

US 75 and selection 13 differed significantly in K concentration at Davis but not at Salinas. Both K and Na concentrations were higher at Davis than Salinas, perhaps indicating greater availability of these elements at Davis. At the higher Davis concentrations, any real differences between varieties would probably be greater and might account for the significant difference. Unlike Na, however, 13 showed the higher K concentration.

Contrary to the sucrose yield factors, it appears that our selection procedure for yellows resistance has decreased NH_2 -N and Na and increased K in the resistant line. These two lines have genotypic differences for the levels of these constituents. How or why this occurred is not known, because selections were made without prior information on NH_2 -N, Na or K levels.

Whether these variety differences are specific for just the 13 selection from US 75, or whether they occur for all lines that show greater yellows resistance than their parental line, is yet to be determined. If decreased NH_2 -N and Na and increased K are linked with resistance to yellows, singly or together, they may serve as a selection criterion or at least as a supplement to our present scheme for obtaining yellows resistance. Russell (14) found that yellows tolerance in sugarbeet was associated with high K content and suggested a close genetic linkage between good tolerance and high K.

Nitrogen Effects

The two nitrogen levels caused highly significant differences in the measured characters except for gross sucrose and K (Table 2). As generally demonstrated with nitrogen treatments, the higher level of nitrogen caused the lower sucrose percentage and the higher root yield. However, in terms of gross sucrose produced, the two treatments produced identical amounts.

As would be expected, the high-nitrogen treatment caused significantly higher levels of petiole-NO₃ and NH₂-N than the low-nitrogen treatment. Sodium concentration was also significantly higher for the high-nitrogen treatment, but the K concentration was not significantly influenced by nitrogen. Similar results are reported in the literature for the influence of increasing nitrogen availability on purity characters (5,12,15,16).

Variety \times Nitrogen and Date \times Nitrogen Interactions

Because there was no significant variety \times nitrogen interaction for sucrose percentage, yield, gross sucrose or K, US 75 and 13 were apparently performing in a similar manner for

these attributes at the two nitrogen levels tested (Tables 1,3). The nitrogen levels did not indicate that the resistance of 13 for gross sucrose yield was partially dependent upon the nitrogen fertility.

Significant variety \times nitrogen interactions did occur for petiole-NO₃, NH₂-N and Na. While selection 13 showed only a mean increase of 186 ppm for petiole-NO₃ from the low to high nitrogen treatments, US 75 had a mean increase of 582 pp. Similarly both NH₂-N and Na increased more in US 75 than in 13 from the low to high nitrogen treatments.

Although the selected nitrogen treatments caused these significant responses for the two varieties, it appeared that greater differential rates of application would have given additional information on the influences of nitrogen. According to Ulrich (18), the critical level between deficient and sufficient levels of nitrogen is 1000 ppm NO_a-N in the petioles. The only treatment mean above the critical level for sufficient nitrogen is US 75 at 170 pounds nitrogen per acre whereas 13 at this level was in the deficiency range at 592 ppm.

The only significant interactions for date \times nitrogen occurred for sucrose percentage and petiole-NO₃ (Table 1). This suggested that the nitrogen level does not cause differential responses for the other characters, or that the nitrogen treatments were not different enough to detect response differences.

Variety \times Date Interactions

Significant variety \times date interactions occurred for beet yield, sucrose percentage, gross sucrose, petiole-NO_a, NH₂-N and Na (Tables 1,4). These interactions indicated that the rate of change for the specific character being measured from date to date was not the same for both varieties. There was no interaction for K.

Under uniform yellows infection at Salinas, 13 and US 75 showed a differential rate of root growth with 13 growing proportionately faster than US 75. Increased growth occurred to the seventh harvest date for both varieties, then decreased for the eighth date. For this decrease in weight only US 75 showed a significant loss.

In the case of sucrose percentage, line 13 increased in sucrose concentration proportionately faster than US 75. At the first harvest there was less than 5% difference, but this difference consistently increased until there was greater than 11% difference at the end of the experiment.

The environmental influences that caused fluctuations in sucrose percentage from date to date appeared to be nearly equal

		Root		Gross	Petiole	Root	
Da ha	te of rvest	yield* T∕A	Sucrose %	sucrose lbs/A	nitrate ppm	NH2-N ppm	Na ppm
				US	75		
۱.	July 25	7.34ª	11.89 ^a	1740ª	1282 ⁿ	781ahe	3534
2.	Aug. 8	9.715	12.455	2420^{6}	$1127^{\rm b}$	959°	288^{he}
3.	Aug. 22	10.96°	13.17°	2890°	493a	725ª	248 ⁿ¹
4.	Sept. 4	12.75ª	13.81^{d}	3520^{a}	527a	825 rd	225ª
ŏ.	Sept. 19	14.20°	14.12°	4020°	410ª	810 ^{bed}	228ª
6.	Oct. 3	15.401=	13.07°	4020°	1130 ^b	8634	316 cd
7.	Oct. 17	15.64*	13.32°	4170^{*}	1090 ^b	810 ^{hed}	433°
8.	Oct. 31	14.86"	13.30%	3960"	1180^{5}	751 ^{ab}	647%
1.	July 25	10.82ª	12.41ª	2680ª	611hrd	514a	186 ^{ab}
2.	Aug. 8	14.82 ^b	12.955	38405	479abed	651°	177 ^{ab}
3.	Aug. 22	16.94°	13.941	4730°	271 ⁿ ¹	543ab	152ª
4.	Sept. 4	20.84 ^d	14.58^{d}	6080^{d}	245 a	616°	137ª
5.	Sept. 19	21.28ª	15.11*	6430e	411abe	624 °	137*
6.	Oct. 3	24.35''	14.24 ^{ed}	6930 r	486a bed	602 ^{bc}	166 ^{ab}
7.	Oct. 17	24.42	14.54 ^d	70907	707 ed	646°	216^{h}
8.	Oct. 31	24.07°	14.95*	72001	786 ^d	616°	343°

Table 4.—Mean values within variety treatments for dates of harvest for those characters that showed significant variety x date interactions.

* Means within varieties followed by the same letter are not significantly different (0.05 level of probability) by Duncan's multiple range test.

for both varieties. For example, the reduction between the fifth and sixth dates affected both varieties. However, whereas 13 nearly regained the high sucrose level of the fifth date, US 75 remained stationary with nearly no resumption of increasing sucrose percentage.

Because both sucrose percentage and root weight showed significant interactions, gross sucrose would be expected to, and does. Again 13 increased faster in total sucrose than US 75. For 13 there was an increase in gross sucrose for every progressive date of harvest. US 75 essentially stopped increasing after the fifth harvest and showed a slight decline for the eighth harvest. For US 75 there was no significant difference for harvest dates five through eight. For 13 the sixth through eighth dates differed nonsignificantly but sucrose yield showed a continued increase.

These variety \times date of harvest data indicated that the reduction of sucrose yield for the more susceptible US 75 occurred continuously following virus infection and that both sucrose percentage and beet yield were affected. Furthermore, as a consequence of its greater susceptibility, US 75 was unable to sustain continued sucrose increase as the environment became less conducive to sugarbeet growth after the middle of September, whereas moderately resistant 13 maintained some sucrose production.

For petiole-NO₃ US 75 and 13 showed a significant variety \times date interaction at the 5% level (Tables 1,4). US 75 showed much higher concentrations for the first two and last three dates of harvest, but there was little difference for the third, fourth, and fifth dates.

US 75 and 13 showed a significant interaction for NH_2 -N concentration through the course of the season. At every date of harvest US 75 had a higher concentration than 13, and although the curves for each variety were broken and changes from date to date were inconsistent, the trend was for US 75 to have decreasing and 13 to have increasing concentrations.

The interaction for Na resulted primarily from the first and last three dates of harvest. For the last three dates US 75 increased in Na concentration proportionately faster than 13. For the second through fifth dates there was proportionately little difference with US 75 continuing to show higher concentrations.

Under yellows conditions these data for 1967 indicated that harvesting after mid-September resulted in only slight yield increases. However, these increases were associated with rapidly deteriorating quality since several melassigenic components rapidly increased in concentration as the growth rate had decreased or stopped. This was particularly true with susceptible US 75.

Summary

Field tests at Salinas and Davis, California, in 1967 compared the effects of yellows infection on the moderately resistant selection 13 and its more susceptible parental sugarbeet variety, US 75. A combination of beet yellows virus and beet western yellows virus was used to inoculate the tests. At Davis inoculated and noninoculated plots were compared. At Salinas the uniformly infected test was grown under two nitrogen levels and harvested at eight dates with 2-week intervals starting July 25 and ending October 31. Root yield, sucrose percentage, and NH₂-N, Na and K concentrations were measured for all treatments. Petiole-NO₃ levels were measured at Salinas.

Under infected conditions at Salinas, root yield and sucrose percentage increased faster in the 13 line than in the US 75 variety. Line 13 increased in gross sucrose through October 31 whereas US 75 showed no increase after September 19. Decreases in the Na, K and petiole-NO₂ concentrations corresponded with the period of rapid growth. However, these constituents increased in concentration as the growth rate decreased in September and October. Amino nitrogen showed little change through the course of the season. When free from yellows, US 75 and selection 13 were not different for root yield, sucrose percentage, or gross sucrose but were significantly different for Na, NH_2 -N and K. The Na and NH_2 -N concentrations were higher and the K concentration lower for US 75.

When infected, US 75 and selection 13 were significantly different for root yield and sucrose percentage with selection 13 yielding 68% more gross sucrose. Infection with yellows caused no significant change in Na, NH_{a} -N or K levels in selection 13 but caused increased concentration of these impurities in US 75. Resistance in line 13 to yellows was not due to selection for increased vigor or yielding ability but to selection for resistance factors.

The high nitrogen treatment caused increased root yields, decreased sucrose percentages, and increased petiole- NO_8 , Na and NH_2 -N concentrations, but did not influence K significantly. The nitrogen treatments caused greater changes in US 75 than in moderately resistant 13. The nitrogen level did not appear to influence the resistance of line 13.

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